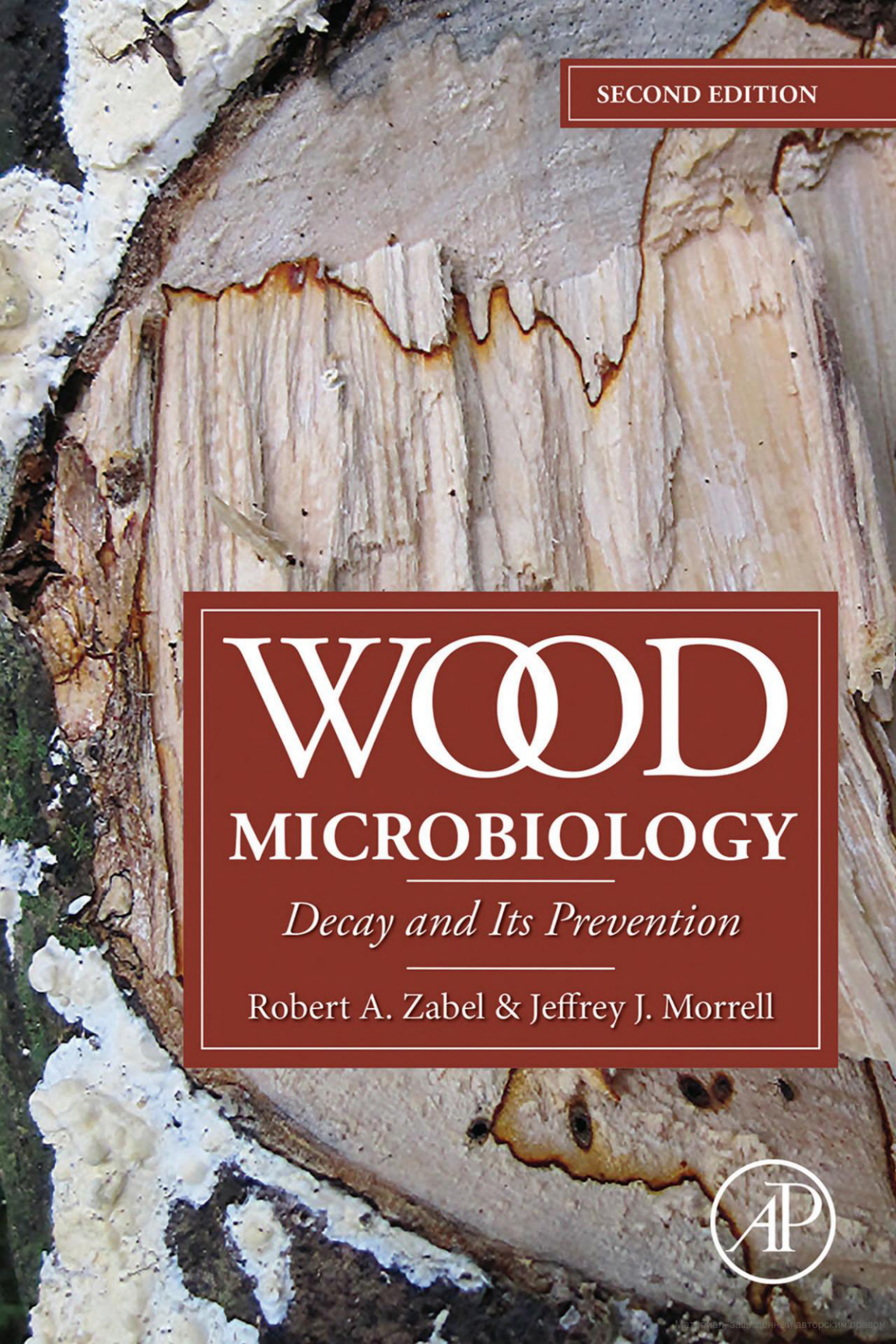

SECOND EDITION
WOOD
MICROBIOLOGY
Decay and Its Prevention
Robert A. Zabel & Jeffrey J. Morrell

Wood Microbiology

WOOD MICROBIOLOGY

Decay and Its Prevention

Second Edition

ROBERT A. ZABEL
SUNY College of Environmental Science and Forestry, Syracuse, NY, United States

JEFFREY J. MORRELL
National Centre for Timber Durability and Design Life, University of the Sunshine Coast, Brisbane, QLD, Australia

Figure Courtesy of Cover:

SERI ROBINSON
Wood Science and Engineering College of Forestry, Oregon State University, Corvallis, OR, United States

ACADEMIC PRESS
An imprint of Elsevier

Academic Press is an imprint of Elsevier
125 London Wall, London EC2Y 5AS, United Kingdom
525 B Street, Suite 1650, San Diego, CA 92101, United States
50 Hampshire Street, 5th Floor, Cambridge, MA 02139, United States
The Boulevard, Langford Lane, Kidlington, Oxford OX5 1GB, United Kingdom

Notices
Knowledge and best practice in this field are constantly changing. As new research and experience broaden our understanding, changes in research methods, professional practices, or medical treatment may become necessary.

Practitioners and researchers must always rely on their own experience and knowledge in evaluating and using any information, methods, compounds, or experiments described herein. In using such information or methods they should be mindful of their own safety and the safety of others, including parties for whom they have a professional responsibility.

To the fullest extent of the law, neither the Publisher nor the authors, contributors, or editors, assume any liability for any injury and/or damage to persons or property as a matter of products liability, negligence or otherwise, or from any use or operation of any methods, products, instructions, or ideas contained in the material herein.

British Library Cataloguing-in-Publication Data
A catalogue record for this book is available from the British Library

Library of Congress Cataloging-in-Publication Data
A catalog record for this book is available from the Library of Congress

ISBN: 978-0-12-819465-2

Publisher: Charlotte Cockle
Acquisitions Editor: Anna Valutkevich
Editorial Project Manager: Sara Valentino
Production Project Manager: Sreejith Viswanathan
Cover Designer: Vicky Pearson

Typeset by MPS Limited, Chennai, India

Dedication

Dedicated to the late Theodore C. Scheffer and his students and colleagues at the Forest Products Laboratory at Madison whose research achievements over the past 75 years have laid the foundation for the principles and practices for the microbiology of wood.

Contents

Foreword

It has been almost 30 years since the first edition of *Wood Microbiology* was published. Since that time, applied microbiology has blossomed as a discipline and we have seen amazing technological advances in a range of fields related to timber durability. Unfortunately, Bob Zabel did not live to see these advances, as he passed away at the age of 81 in 1998. I have no doubt that he would have embraced these advances.

Bob was one of that amazing generation that went off to fight in World War II then came back and quietly built careers. He rose through the ranks to become Vice President at the SUNY College of Environmental Science and Forestry, then had an epiphany and stepped down to return to teaching. Countless students, including this author, benefited from that decision and were privileged to take his Wood Microbiology class on the ESF main campus, as well as his co-taught Forest Pathology class at the ESF Summer Station at Cranberry Lake.

The original edition of this text was an expansion of class notes; this revision attempts to update areas/topics as needed while still keeping the flavor of the original edition. This means that it tends to still include a great deal of historical information to set the context for the current status of the field. It is difficult to see where we will go if we do not understand what has gone before us.

I am indebted to my spouse, Patricia, for her patience as this edition slowly evolved. I also gratefully acknowledge the efforts of Dr. Barry Goodell, University of Massachusetts, as he tried to bring me into the 21st Century for taxonomy and to Dr. Seri Robinson, Oregon State University, for providing the new cover art.

Finally, I extend my deep appreciation to the Zabel family who has agreed that all proceeds from the book will go to fund undergraduate scholarships at SUNY ESF.

We hope you enjoy the new edition.

Preface

We believe a need still exists for a textbook on wood decay caused by fungi and related biologic deteriorations to collect and summarize the rapidly expanding literature and experience on this topic in a single source. A need also exists to relate this information to the basic principles of biology.

It is our hope that this introductory text on the principles of decay and discoloration processes in wood and related topics will facilitate wiser use of wood resources and stimulate speculation and research in the area. We also hope it stimulates a fascination with fungi and their unique capabilities and important roles in the biologic world.

The textbook is based on a series of lectures on wood decay, its prevention, and control presented to junior and senior level students who were majoring in forest biology or wood products at SUNY-ESF and Oregon State University. It has been updated to capture changes since the first edition.

Emphasis in the textbook is placed on the major fungal agents that damage wood during the growth, harvesting, storage, conversion, and use of wood for a range of major purposes. The characteristics and appearances, causes (etiology), detection, effects on various use properties and prevention or control are stressed.

Chapters 1 and 2 trace the origin and history of wood microbiology, discuss the major types of wood deterioration, and relate wood decay to the broader subject of biodeterioration. Chapters 3–5 review the general characteristics, types and classification, growth needs, and metabolism of fungi as the major cause of wood decays and discolorations. An emphasis is placed on relating the growth needs of fungi to decay control principles. Chapter 6 summarizes key features of the structural aspects of wood and wood moisture relationships that are central to fungal survival and growth in wood and the subsequent development of damaging decays and discolorations.

The emphasis in Chapters 7–11 is on the basic anatomical, physical, and chemical aspects of wood decay. Chapters 7–10 cover the basic aspects of wood decay including types, appearances, evidences, the anatomical and ultrastructural features of decay, decay effects on various physical and strength properties of wood, and the chemical aspects of decay.

Chapter 11 reviews the ways fungi colonize wood and their interactions during decay development. It is a prelude to Part 3 which considers the major decay problems.

In Part 3, Chapters 12–15 review the principal decays and discolorations which may develop in wood in standing stems, during harvest, storage, conversion, and various major uses. Special emphasis is placed on the decay problems in buildings and utility poles. Chapter 16 discusses the methodologies and approaches to decay detection. Chapter 17 reviews mildew problems on wood-based coatings and related industrial problems caused by fungi and bacteria. Chapters 18–19 review the principles of decay control and prevention by uses of naturally durable woods and wood preservatives. The characteristics and special uses of the major wood preservatives are discussed along with environmental concerns and restraints. The difficulties and approaches to developing new wood preservatives are discussed. In Part 4, a final chapter speculates on future decay prevention approaches involving biological controls, new wood treatments and developing more durable woods. An emphasis is placed on the role of biotechnology and future wood uses and modifications. Research trends and future career opportunities in wood microbiology are discussed.

The textbook is designed specifically for use in a two or three credit hour one semester course. It assumes that the student has some background in general biology and organic chemistry.

The book was also designed to serve as a useful information source to wood processors, engineers, architects and other professionals who are engaged in the practical aspects of wood use and need to know more about the principal biodeterioration problems of a major wood use, why decay occurs, as well as how to recognize and prevent or minimize its development. It is also intended to provide some background on wood biomodifications for those interested in applying fungi for useful purposes in this period of expanding biotechnology.

Summaries at the end of each chapter list what we believe are the key essentials. The suggested further readings and the literature cited will lead interested readers to more detailed coverages of each topic.

Writing a textbook in the midst of an information explosion, new biologic insights, and mounting environmental concerns is challenging. Rapid advances in the related fields of mycology, physiology, genetics, electron microscopy, and biochemistry have required constant substantial revisions of ideas. These rapid changes and flux of ideas about valid

information on wood decay place added important on generalization and synthesis of the current literature for students.

Special thanks are extended to our many colleagues and associates for their encouragement, wise counsel, and often assistance in reviews and improvements in the chapters. Particularly helpful to the senior author at SUNY-CESF were Dr. James Worrall, Dr. Paul Manion, Dr. Chun K. Wang, and Dr. David Griffin. Colleagues who reviewed the original chapters were Dr. John Simeone (Chapter 2 - Wood deterioration agents), Dr. Chun Wang (Chapter 3 - The characteristics and classification of fungi and bacteria), Dr. David Griffin (Chapter 4 - Factors affecting the growth and survival of fungi in wood (fungal ecology)), Dr. James Nakas (Chapter 5 - Fungal metabolism in relation to wood decay), Dr. Wilfred C6te (Chapter 6 -The decay setting: some structural, chemical, and moisture features of wood in relation to decay development), Dr. James Worrall (Chapter 7 - General features, recognition, and anatomical aspects of wood decay), Dr. Tor Timmell (Chapter 8 - Chemical changes in wood caused by decay fungi), Dr. Robert Hanna (Chapter 9 - Ultrastructural features of wood decay), and Dr. Paul Manion (Chapter 12 - Decays originating in the stems of living trees).

The authors also acknowledge the helpful comments provided by Jerrold E. Winandy, U.S. Forest Products laboratory (Chapter 10 - Changes in the strength and physical properties of wood caused by decay fungi), Dr. Wayne Wilcox, University of California, Berkeley (Chapter 15 - Decay problems associated with some major uses of wood products), Dr. Theodore C. Scheffer, Senior Research Associate Emeritus, Oregon State University (Chapter 14 - Wood molds, stains and discolorations; Chapter 18 - Natural decay resistance (wood durability)), and Professor Robert D. Graham, Oregon State University (Chapter 19 - Chemical protection of wood (wood preservation)). Their collective efforts significantly improved these Chapters and are greatly appreciated.

Special thanks are due also to the many students in this course for their interest and innumerable sharp and penetrating questions that placed many issues in clearer perspective.

As with any document, it is expected that some portions will become outdated as new information becomes available. It is our hope that this text stimulates others, much in the same way that our mentors encouraged and stimulated our interests in this field.

CHAPTER ONE

Introduction to wood microbiology

Contents

This textbook focuses primarily on the damaging decays and discolorations that fungi may cause to wood under some use-conditions. A major emphasis is placed on recognition, causes, conditions favoring development, effects on various wood use properties, and prevention or control of these serious wood defects.

This introductory chapter begins with a review of the unique qualities of wood for a wide range of uses and the benefits of its production. This is done to put the negative effects that may occur in some wood uses into proper perspective. It is not too early to emphasize that most wood defect problems can be avoided or greatly minimized when wood us properly used or handled.

Other agents that may degrade or destroy wood are briefly discussed because their damage can be confused with decay. Wood decay losses at various stages of production and use are reviewed to justify the level of control effort and suggest research priorities. The historical aspects of major wood decay understandings are traced to establish its relation to other related fields, and probable emergence as a specialized facet of

Wood Microbiology.
DOI: https://doi.org/10.1016/B978-0-12-819465-2.00001-2

microbiology. The fungal cause of decay resulted in much of the early information on decay appearing in phytopathological or mycological journals. Unfortunately, this early information on decay prevention and controls failed to reach engineering and architectural sources where the design of wood structures was determined. Later, we will see that design of structures in terms of water shedding or retention can be crucial to effective decay prevention. The introductory chapter ends with some basic concepts, and the definitions of some terms that will be used throughout the book.

Wood - a remarkable material

Wood is a remarkable material of great value and importance in the world economy. It is used extensively as a structural material, fuel, or industrial raw material in many parts of the world. It is estimated that wood accounts for one-fourth of the value of the major industrial materials in the United States (National Research Council, 1990). As a renewable natural resource, it is available in large quantities at relatively low costs. An estimated one-third of the landed area of the world is in forests. As a land crop, wood supplies can be increased in both volume and quality by wise forest management practices. Wood production in the forest ecosystem is often associated with many other forest values and amenities such as soil development and enrichment, wildlife resources, moderating and extending water run-off, providing superb recreational settings, reducing atmospheric pollution, and landscape aesthetics. More recently, forests have also been examined for their possible roles as carbon sinks for sequestration of atmospheric carbon dioxide to slow the rate of global climate change

Wood is also unique in a plant evolutionary sense since it was the vertical development of perennial vascular tissue (wood) that led to the aerial development of land plants. The vertical stem or trunk of the tree consists of elongated cells with unusual strength, flexibility, and durability both at macroscopic and ultrastructural levels. These properties permit stems to bear heavy crown loads and to withstand high horizontal stress from wind and ice loads. The long-term selection for these properties has led to many unique properties of wood.

Wood value and uses

As a structural material, wood has high strength per unit weight, and is easily shaped and fastened. It is a convenient energy source and a major inexpensive source of cellulose and its many derivatives for the chemical industry. The color patterns and textures of woods are often pleasing, leading to uses for many decorative purposes. Wood is available in a wide range of textures, colors, densities, and chemical compositions supporting a wide range of important uses such as construction timbers and lumber, decorative paneling, plywood, piling and wharves, railroad ties, poles and posts, packaging and crates, paper and paper products, cellulose derivatives, charcoal, and thousands of specialized miscellaneous uses ranging from pipe bowls to fiddle heads. It is important to note that, despite its use in a variety of structural applications, over half of the wood consumed each year on the planet is burned for either cooking or energy production.

Potential uses of wood

Wood looms even more significantly as a valuable raw material in the future as expanding world populations place increasing stress on natural land ecosystems as sources for food, fiber, and energy. Modern intensive forestry practices have the potential to substantially increase both the yields and quality of wood. Trees are efficient radiant energy transducers in many regions and their biomass can be converted to alcohol, as alternative combustion engine energy sources. This process is already well underway for production of aviation fuels. The selective decay actions of fungi and related fermentation activities may permit the utilization of wood as cheap sources of animal feed or protein. As a material, wood is readily biodegradable under certain conditions and returns natural substances to ecosystem cycling. It has low energy requirements for conversion into various products as compared with other structural materials. As relative raw materials costs and availability change, wood has the potential to replace petroleum as a base for the production of a wide range of industrial chemicals and polymers and perhaps energy itself. Although not considered as often, the simple use of wood in a structure effectively

sequesters this carbon for the life of the structure and potentially beyond further enhancing the potential influence of timber on atmospheric carbon dioxide levels. Use of wood in properly designed structures is an important aspect of this process.

Wood disadvantages

Wood has some serious disadvantages that limit its usefulness for some purposes, including:

1. Wood is primarily biodegraded by the action of fungi and, under the proper conditions, these fungi may decay and weaken or discolor the wood to the point where replacement is required. Other biological agents also attack wood. Termites are a serious threat to untreated wood in many tropical regions. Marine invertebrates chew tunnels in wood in various salt water uses and cause serious damage. These other bioagents that can degrade or destroy wood are discussed in Chapter 2. In many cases, their actions can be controlled or minimized by judicious use of treated or naturally durable wood.
2. Wood combusts at low kindling temperatures and, in certain size configurations and conditions, burns readily. Chemical treatments and wood design can reduce the combustion hazard. One advantage of wood is that it burns at a predictable rate and rarely fails catastrophically.
3. Wood is dimensionally unstable at moisture contents below the fiber saturation point (fsp) and swells as it wets and shrinks as it dried. This problem is compounded because the largest dimensional changes occur in the tangential plane. Differential shrinkage often leads to deep check formation in the radial plane in round stock such as poles or piling. Chemical treatments may reduce the dimensional changes, but these treatments are expensive.
4. Wood, as a natural product, displays considerable variability in its appearance, chemical composition, and physical properties. Some differences are the result of species or growth conditions and are accounted for in specifications. High safety factors in critical design uses of wood are used to minimize this disadvantage. Conversely, the

variability in color and texture of wood provides its beauty and aesthetic appeal for many home uses.

5. Wood has a large bulk per unit weight for fuel, pulping and chemical uses

In summary of this brief discussion of wood disadvantages, it is stressed that effective wood handling, properly designed and maintained structures, chemical treatments, and proper use of standards and specifications, can dramatically minimize these disadvantages. In a long-term environmental setting, the biodegradability of wood may also minimize accumulation of solid wastes created when less degradable materials such as plastics are used.

Decay losses and future wood needs

Accurate estimates of decay losses are useful to justify controls and serve as research incentives. Decay losses are difficult to quantify because of the multiplicity of wood uses under a wide range of environmental conditions. Experienced guesses are that 10% of the annual timber cut in the United States is used to replace wood that decayed in service, much of it primarily from improper use and care. Added to this base raw material cost would be, in many cases, the added costs representing processing, fabricating, finishing, merchandising, and assembly or replacement operations. The substantial labor costs incurred by replacement and, in some cases, the inconvenience of interrupted services would have to be added. Another subtle loss source may be eventual wood replacement by more expensive, less environmentally friendly, and less satisfactory materials.

Large additional supplies of wood are required to meet burgeoning population needs beyond the next century. A substantial first step in meeting future timber needs may be simply to handle and use wood more effectively, thereby drastically reducing decay losses. In a related sense, forest pathologists and entomologists recognize that control of tree diseases and forest insect pest problems also can dramatically increase future wood supplies.

Reducing decay losses

Properly used wood is an amazingly durable organic material. Only a few specialized microorganisms, primarily the higher fungi, have solved

the biochemical riddle of its rapid digestion. Experts agree that much is already known about effective and economical control of decay in most wood uses. The central control problem is that much of this information is fragmented and not readily available or generally known by wood processors, designers, merchandisers, and users. Most architecture and engineering students learn relatively little about wood compared to steel and concrete, yet many go on to careers designing with wood. Until recently, wood has been available at low cost, and the ease of replacement has reduced the incentive to conserve. Furthermore, the central information of wood microbiology lies across many disciplines and is often inaccessible at both academic and trade levels. The internet provides a wealth of information on wood durability, but much of it is anecdotal and, in some cases, wildly inaccurate.

Wood pathology vs wood microbiology

The subject dealing with decays and stains in wood products has generally been titled as either wood pathology or products pathology. Traditionally, wood decay has been considered as a specialized facet of forest pathology (which is a subject area of phytopathology) and most forest pathology textbooks have included a chapter on wood products decays and lumber stains. This was logical and natural since forest pathology initially emphasized stem decay problems and the causal fungi. This approach reflected, in part, the early emphasis of foresters on forest protection and wood harvesting and the phytopathologist's concern with fungi.

Mounting afforestation problems and epidemic diseases and shifts in phytopathology, focused interest in forest pathology more on the disease process itself (the adverse reaction of living organisms to disease agents) and began the separation of wood biodeterioration problems from tree disease problems.

Actually, the term wood pathology is a misnomer. The subject is not a pathology at all since it deals primarily with the deterioration of non-living materials caused by biotic and abiotic agents. Products pathology, as another term, suffers from the same "non-living" dilemma. Products pathology is more correctly a subject matter in phytopathology that involves the diseases of stored fruits, vegetables, and grains that are still

living entities. A more logical setting for fungal related wood defect problems is "applied industrial microbiology" which studies the microorganisms that adversely affect properties or the appearance of food products, textiles, leathers, organic materials, paper, or wood. This field also includes matters of substrate biomodification and fermentation by microorganisms.

We might see the emerging discipline of wood microbiology in clearer perspective by listing some closely related or overlapping fields where information on wood decays and stains and their causal agents may appear.

1. Phytopathology deals with the understanding and control of plant disease, although there are some similarities between decay and tissue disintegration diseases.
2. Forest Pathology deals with tree diseases. Some decays and stains originate in the living stem and continue as problems in the wood product; some tree pathogens are also wood saprogens.
3. Mycology is the study of all aspects of fungi. Most wood saprobes are fungi, although a few bacteria are also important.
4. Microbiology includes the study of all small organisms including fungi and bacteria. It includes the diseases and deteriorations they cause and all facets of their use (biotechnology).
5. Wood Preservation is an engineering subject dealing with the physical and chemical aspects of protecting wood from fire, insects, and fungi.

The reality is that disciplinary boundaries are in constant flux and shift as need and opportunity arises. Wood Microbiology integrates the microbial aspects of wood deterioration and wood biomodification from all these areas. The field should focus information on wood biodeterioration in a recognized discipline and minimize the problem of widely dispersed information sources.

Wood defect literature now appears in wood technology and microbiology journals rather than traditional phytopathological or forestry journals, reflecting a trend to place data where it can be most directly applied.

Historical perspectives of wood pathology

Historical awareness of a subject is important because it clearly displays the ever-evolving nature of knowledge and the occasional

integration of the pieces into great unifying concepts. History also demonstrates the danger of dogma and the need for constant questioning and probing in the search for better explanations of events.

In this section, the term "wood pathology" will be used for historical reasons. Wood pathology had its origin in forest pathology, where pathologists were interested in the nature of wood decay in tree stems, building rots, and wood storage problems.

Concern about wood decay and pragmatic methods to reduce this damage long preceded any recorded understanding of the cause and nature of decay. The high value of the biblical "Cedars of Lebanon" was due to their natural durability, important in shipbuilding and temple construction. The early Greeks knew that vertical bearing beams should rest on stone and not in direct contact with soil. Pliny, the Roman historian, recorded the susceptibility of sapwood to decay, listed durable woods, and reported that soaking wood in cedar oil reduced decay. In 1832, more than 40 years prior to discovering the true fungal nature of decay, the first successful wood preservation process, Kyanizing, was introduced in Europe using mercuric chloride. At about this time in Germany (1833), Theodor Hartig first recorded the microscopic appearance of fungal hyphae in decayed wood. Microscopic forms of life were then assumed to arise spontaneously from decomposition products. In 1863, Schlact reported on the effects and microscopic features of hyphae on a tropical wood and nearly 100 years later, it was recognized that his descriptions and the drawings were typical of a new type of decay called soft rot. He also assumed the hyphae were the result and not the cause of the cell wall decomposition.

Forest pathology originated at the time of the settling of the great spontaneous generation vs "life from life" controversy of the mid-19th century. Shortly after the classic researches of Tyndall and Pasteur had destroyed the concept of spontaneous generation and established the clear role of microorganisms in causing fermentations, the stage was set in 1874 for Robert Hartig's clear resolution of the causal relationship between the presence of hyphae and subsequent decay in wood. His early insights into the nature of decay were remarkable. He clearly connected the external fruiting body to the internal hyphae, and the hyphae and their growth on the cell walls to decay. He established later that some decays were specific for a kind of fungus and subsequently attributed these differences to enzymes. His extensive publications on wood decay and tree diseases along with his cadre of students and research associates led to rapid

developments in forest pathology and wood pathology in Europe and the United States.

Two decades later, in 1899 in the United States, the Federal Government established the Mississippi Valley Laboratory at St. Louis, Missouri for reconnaissance and research on forest disease problems. Dr. Herman von Schrenk was the initial appointee and later Director of a small group. He was extremely productive and within a few years published a series of pioneering papers on the stem decays of timber species, wood decay in buildings, blue stain, preservative evaluations, and wood durability. Associated later with him were G. Hedgcock, P. Spaulding, and Catherine Rumbold. Hedgcock published a classic study on chromogenic fungi that discolor wood (1906). Spaulding contributed studies on the culturing of decay fungi and slash decomposition. Catherine Rumbald studied blue stain problems and insect roles as vectors.

The expansion of the railroads and the building program associated with the rapid western expansion of the country at the beginning of the 20th century led to the use of many local, non-durable woods for construction as the supplies of durable oaks, chestnut, cypress, and the cedars dwindled or were not easily available. This also was a period of increased harvesting of southern and western pines and serious sapstain problems as processing became year-round and resulted in summer air seasoning. Serious decay and discoloration problems led to concerns at the national level. One outcome was the formation of the American Wood Preservers' Association in 1904 for standardization of specifications for wood preservatives and treatments. Another was that the research of von Schrenk's group shifted increasingly to study decay of structural timbers or railroad ties and began research on sapstain control and wood preservation. There was also growing concern about the threat of chestnut blight and other introduced tree pathogens.

A decision was made at the federal level to separate the forest and wood pathology programs. In 1907, the Mississippi Valley Laboratory (MVL) was discontinued and the U.S. Forest Service's Forest Products Laboratory (FPL) at Madison, Wisconsin was assigned research responsibilities for the wood pathology program. Thus, it became the first titled program of wood pathology in the United States and continues to play a leadership role in wood pathology and wood preservation matters. The initial FPL research responsibilities included studies of the causes and controls of decay and stains in wood products, mycology, the physiology of wood products fungi, and wood preservatives. The forest pathology

program was transferred to the Bureau of Plant Industry as the Office of Investigations in Forest Pathology. In 1953, it was also assigned administratively to the U.S. Forest Service.

From its beginning in 1899 in the United States, the field of wood pathology expanded steadily. Principal researchers were at the Forest Products Laboratory, as well as some forestry colleges, military organizations responsible for supplies, the chemical industry, and allied fields such as botany and wood technology.

Many major contributions shaped the development of wood microbiology to the present period. In 1931, Hubert published the first American textbook on forest pathology, which contained a special section titled Wood Pathology and reviewed the principal research contributions of this early period. Some detail is presented here to show the rapid progress and significant accomplishments of the initial handful of dedicated researchers who defined the future course of the field. The major research topics with selected contributions were as follows:

(a) Decay effects on wood properties such as strength (Colley, 1921, cited in Hubert, 1931) and heat conductivity (Hubert, 1924, cited in Hubert, 1931);

(b) Chemical properties of decayed wood (Hawley and Wise, 1926, cited in Hubert, 1931) and the use of decayed pulpwood for paper (Rue, 1924);

(c) Wood durability was attributed to soluble extractives (Hawley et al., 1924, cited in Hubert, 1931) and extensive durability tests on local timbers were reported (Humphrey, 1916, cited in Hubert, 1931; Schmitz and Daniels, 1921, cited in Hubert, 1931);

(d) Relationships between various moisture content levels in wood and decay development (Snell, 1921, cited in Hubert, 1931) and sapstain (Colley and Rumbold, 1930, cited in Hubert, 1931);

(e) Decay in buildings and lumber caused by *Meruliporia incrassata* (Humphrey, 1923, cited in Hubert, 1931);

(f) Decay problems and associated causal fungi in ties, mine timbers, poles, and piling (Humphrey, 1917, 1920, 1923, cited in Hubert, 1931);

(g) Taxonomic studies of sapstain fungi (Hedgcock, 1906 and Rumbold, 1929, cited in Hubert, 1931) and their control (Hubert, 1929, cited in Hubert, 1931); and the evaluation of preservatives (Humphrey and Flemming, 1915, cited in Hubert, 1931; Richards, 1923, cited in Hubert, 1931).

In 1940, Scheffer and Lindgren completed a detailed study on the causes of lumber stains and their control. The application of their recommendations did much to minimize the serious sapstain problems and helped foster the development of a number of sapstain control companies. In 1946, Cartwright and Findlay published their classic "Decay of Timber and Its Prevention" which presented a worldwide summary of wood pathology information. The book was revised in 1958 and still serves as an important reference source for the field.

Davidson et al. (1942) and Nobles (1948) published cultural keys that greatly facilitated cultural identification of decay fungi isolated from timber and wood products prior to the advent of genetic sequencing technologies.

In 1954, Savory established clearly that a new and significant type of decay termed "soft rot" was caused by some Ascomycetes dispelling the near century old dogma that only Basidiomycetes caused decay. Contributions toward understanding this rot type were made by Corbett (1965), Duncan (1960), and Levy (1978). Nilsson (1973) established that substantial numbers of microfungi may cause soft rots.

In 1961, Cowling reported on the strikingly differential effects of white and brown rot fungi on a range of physical and chemical properties of sweetgum sapwood. Particularly significant were insights on the nature of the enzyme systems involved based on ultrastructural dimensional restraints.

In 1965, Duncan and Lombard provided extensive information that was collected at the FPL over a 50 year period on the major decay fungi associated with various wood products and wood uses, nationwide.

Verrall (1966) contributed to the understanding of decay problems in buildings in relation to various water sources and later (1965) demonstrated the effectiveness of dip or brush-on preservative treatments for some above ground wood uses.

Insights on the anatomical and ultrastructural features of decay were provided by Wilcox (1970) and Liese (1970). Koenigs (1972) postulated an intriguing non-enzymatic process to explain how brown rot drastically alter cell wall components in crystalline zones inaccessible to enzymes early in the decay process. The production of free radicals has since been shown to be an important first step in the decay process for many organisms. In a decade of study, Reese (1977) and associates provided insight on the nature and mode of action of the "cellulase" enzyme complex and opened the door to commercial wood fermentation possibilities. Ericksson

(1978) clarified identities, roles, and sequences of the cellulose degrading enzymes for a white rot fungus. Blanchette et al. (1987) provided new information on the nature of decay by white rot fungi with the scanning electron microscope.

In long term comprehensive studies on decay origins and organism sequences in stem decays, Shigo (1967), Shigo and Marx (1977) demonstrated a compartmentalizing protective system in living stems that may have major implications in subsequent wood product treatments and use.

Treatments with agricultural fumigants were developed by Graham and Corden (1980) for controlling decay in poles and piling in service. Butcher (1970) and Rayner and Todd (1979) have provided new insights on the sequences and many interactions of fungi in the decay process, invalidating the old concept of one fungus -one decay. Nilsson and Holt (1982), and Nilsson and Singh (1983) have shown that bacteria can cause soft rot cavities and unusual tunnels in the secondary cell wall.

Alternatives to the protection of wood by loading the cell cavities with potent toxicants are being explored increasingly as a result of growing environmental concerns. Richard (1974) reported on the use of antagonistic fungi to control decay in utility poles although subsequent studies found these agents to incompletely effective. Preston (1986) reported on new wood preservative compounds and proposed accelerated testing procedures. These tests, coupled with changes in regulatory issues, led to the nearly complete replacement of chromated copper arsenate with other copper systems. Rowell and Ellis (1979) have reported on chemical ways to bulk or modify wood and enhance its decay resistance. Continued development of these technologies has led to production of acetylated wood, furfurylated wood and thermal modification as alternative treatments. These treatments have gained markets, especially in Europe.

Kirk and Farrell (1987) identified the first enzymes in the "lignase" system with its significant delignifying and biopulping possibilities in this emerging biotechnology period. These results led to a cascade of research to better understand lignin degradation.

Another indication of the rapidly growing interest in wood as a valuable renewable resource and its decay problems was the appearance of three books. Loewus and Runeckles (1977) and Higuchi (1985) edited books covering the biosynthesis and degradation of wood. Liese (1975) edited a book on the interactions of fungi and bacteria in decay development and the enzymatic mechanisms of the decay process. Nicholas (1973) edited a book on wood deterioration and its prevention that has

become the most widely used book in the field. A textbook on the microbial and enzymatic degradation of wood and wood components was prepared by Eriksson et al. (1990). Several comprehensive textbooks stressing the ecological aspects of wood-inhabiting microorganisms during decay development have been prepared by Rayner and Todd (1979) and Rayner and Boddy (1988). Several American Chemical Society books have followed up on these publications. At the same time the first edition of this book appeared, Eaton and Hale (1993) developed a similar treatise with more of a European flavor.

The historical highlights of achievements in wood microbiology indicate an initial primary concern with understanding and controlling fungal damage to wood. Current trends suggest that wood microbiology will also concern itself increasingly with the beneficial uses of fungi to enhance wood properties or transform it into new products.

Concepts and terminology in wood microbiology

An important part of the preparation for any profession is to learn the specific vocabulary or language of that field. Terms developed to denote specific concepts, structures, or events help to simplify the complexities of the biological world, making differences, similarities and various interrelationships more easily understood. It must be remembered that these concepts and definitions are arbitrary and only their usefulness justifies their existence. In some cases, concepts cannot be precisely defined so as to include all related phenomena. Many times a series of events occur in an intergrading series or a continuum without easy separations. In other cases, authorities simply disagree on definitions and their conflicting views appear in the literature. The following terms described below will be used in this book and an understanding of their meaning will be helpful to understand the wood microbiology literature. The terms are contrasted with their counterparts in forest pathology where applicable.

First, a clear distinction between deterioration and disease is necessary because of the forest pathology origin and current wood pathology designation of what we prefer to call now wood microbiology.

Deterioration is the destructive change in the properties of a nonliving material caused by a wide range of chemical, physical, or biotic agents. It contrasts directly with disease. Textiles, paper products, wood, plastics, coatings are examples of wood-related organic materials that may

be degraded or destroyed by biotic agents. The tendering of fabrics by the ultra violet portion of sunlight is an example of a physical deterioration agent. Corrosion of the steel beams in a bridge by salt is an example of chemical deterioration.

Disease (plant) is a sustained abnormal physiological process or processes of a plant or its parts that may threaten the life of the plant or its parts or reduce its economic value. It is emphasized that disease involves reactions and changes (symptoms as responses) in the stressed living organisms whereas deterioration concerns only changes in non-living organic materials. Biotic or abiotic agents may instigate the changes in either case.

Biodeterioration (biodegradation) is a subset of deterioration. It is a negative term and can be defined as any undesirable change in the properties of a non-living material caused by the living activities of organisms. The agents involved are many and varied including bacteria, algae, fungi, invertebrates, and vertebrates such as rodents and birds. The major processes involved are assimilation, including invasion and digestion of organic materials such as wood or textiles, mechanical damage by the rasping activities of marine borers or insects, the corrosion of metals by the chemical activities of bacteria, and functional impairments such as the fouling of ship hulls by growth and accumulation of barnacles.

Two major types of wood biodeterioration that are the central focus of this textbook are decay and discoloration (stain). Decay reflects changes in the chemical and physical properties of wood caused primarily by the enzymatic activities of microorganisms. Discoloration is defined as changes in the normal color of wood resulting from growth of fungi in and on surface of the wood or chemical changes in cells or cell contents. Molds are a subset of stains that primarily discolor through the presence of spores on the wood surface.

Other examples of materials that may be degraded or destroyed by the activities of fungi and bacteria are electrical and optical equipment, stored foods, leathers, plastics, petroleum products, textiles, and pharmaceutical products.

Biomodification is the positive term used for the biotic processes involved in the breakdown or conversion of organic or waste materials into innocuous or useful products by microorganisms. Topics of interest include the bioconversion of trash, garbage, toxic chemical wastes, pesticides, and sewage sludges into useful products or harmless wastes. A major chemical industry utilizes fungi or bacteria to produce a wide range of chemicals and pharmaceuticals from organic materials. There has been

great interest in the biological delignification and conversion of wood wastes into palatable animal fodders, alcohol for fuels, or yeast production as protein and vitamin sources.

Fungi and bacteria are important destructive agents in both disease and biodeterioration. Bacteria are unicellular **procaryotes** that reproduce by fission. Fungi are filamentous **eucaryotes** without chlorophyll that digest various carbon compounds externally. A single filament of a fungus is known as a **hypha (hyphae-plural**). Collectively a mass of hyphae are commonly called **mycelium**. Fungi reproduce primarily by spores that are one to several celled units of reproduction. They are liberated from the mycelium and each cell is capable of reproducing the fungus.

Biodeterioration is recognized by changes in the appearance or properties of the organic material or the physical presence of the causal agent. Disease is recognized by evidences termed as signs or symptoms. Signs are the actual physical presence of the disease-causing organism on the diseased plant. Symptoms are the physiological response or reactions of the host to the presence of the disease-causing agent or organism (e.g. tissue swelling, resinosis, dwarfing).

A **saprogen** is an organism that secures its food from dead materials or carbon sources. A parasite is an organism dependent part or all of the time on another organism of a different taxonomic species and deriving all or part of its food from this living organism. Substratum is a non-living organic material that serves as food to a living organism. The host is a living organism serving as a source of food to a parasite. A pathogen is an organism capable of causing a disease. Generally, pathogens are parasites, but toxin producers can be exceptions.

As we proceed through the remaining chapters, it should become clear that deterioration involves a complex array of interacting processes involving microorganisms, insects, and the environment acting on the non-living, and, therefore, non-responsive wood substrate. This is an important concept since the wood cannot react to protect itself and has major implications for wood usage.

Summary

1. Wood has numerous beneficial properties

2. The active study of wood deterioration dates to the late 1800s and has gradually progressed from a simple quantification of the losses to more basic studies of the nature of decay
3. The term wood microbiology may be useful for describing the field since it cuts across a variety of disciplines related to the decay process.

References

Blanchette, R.A., Otjen, L., Carlson, M.C., 1987. Lignin distribution in cell walls of birch wood decayed by white rot Basidiomycetes. Phytopathology 77 (5), 684–690.
Butcher, J., 1970. Techniques for the analysis of fungal floras in wood. Mater. U. Org. 6, 209–232.
Corbett, N.H., 1965. Micro-morphological studies of the degradation of lignified cell walls by ascomycetes and fungi imperfecti. J. Inst. Wood Sci. 14, 18–29.
Cowling, E.B., 1961. Comparative Biochemistry of the Decay of Sweetgum Sapwood by White-Rot and Brown-Rot Fungi. U.S. Department Agriculture, Technical Bulletin No. 1258, Madison, Wisconsin.
Davidson, R.W., Campbell, W.A., Vaughn, D.B., 1942. Fungi Causing Decay of Living Oaks in Eastern United States and Their Cultural Identification. U.S.D.A. Technical Bulletin No. 785, Washington, D.C.
Duncan, C.G., 1960. Wood-attacking Capacities and Physiology of Soft Rot Fungi. U.S. D.A. Forest Products Laboratory Report 2173, Madison, Wisconsin.
Eaton, R.A., Hale, M.D.C., 1993. Wood: Decay, Pests and Protection. Chapman & Hall, London, U.K, 546 p.
Ericksson, K.E.L., 1978. Enzyme mechanism involved in cellulose hydrolysis by the white rot fungus, *Sporotrichium pulverulentum*. Biotechnol. Bioeng. 20, 317–332.
Eriksson, K.E.L., Blanchette, R.A., Ander, P., 1990. Microbial and Enzymatic Degradation of Wood and Wood Components. Springer-Verlag, New York, p. 407.
Graham, R.D., Corden, M.E., 1980. Controlling Biological Deterioration of Wood With Volatile Chemicals. Electrical Power Research Institute EL-1480, Palo Alto, Calif.
Higuchi, T. (Ed.), 1985. Biosynthesis and Biodegradation of Wood Components. Academic Press, Inc, New York, N.Y, 679 pp.
Koenigs, J.W., 1972. Effects of hydrogen peroxide on cellulose and its susceptibility of cellulase. Mater. Org. 7, 133–147.
Levy, C.R., 1978. Soft rot. Proc. Am. Wood Preserv. Assoc. 74, 145–163.
Liese, W., 1970. Ultrastructural aspects of woody tissue disintegration. Ann. Rev. Phytopathol. 8, 231–258.
Liese, W. (Ed.), 1975. Biological Transformation of Wood by Microorganisms. Springer-Verlag, New York, N.Y, 203 pp.
Loewus, F.A., Runeckles, V.C. (Eds.), 1977. The Structure, Biosynthesis, and Degradation of Wood. Plenum Press, New York, 527 pp.
National Research Council (Ed.), 1990. Forestry Research: A Mandate for Change. National Academy Press, Washington, DC.
Nilsson, T., 1973. Studies on Wood Degradation and Cellulolytic Activity of Microfungi. Studia Forestalia Suecica, Stockholm, Sweden, p. 104.
Nobles, M.K., 1948. Studies in forest pathology VI. Identification of cultures of wood rotting fungi. Can. J. Bot. 26, 281–431.
Rayner, A.D.M., Boddy, L., 1988. Fungal Decomposition of Wood. Its Biology and Ecology. John Wiley & Sons, New York, p. 587.
Rayner, A.D.M., Todd, N.K., 1979. Population and community structure and dynamics of fungi in decaying wood. Adv. Bot. Res. 7, 380–420.

Reese, B.T., 1977. Degradation of polymeric carbohydrates by microbial enzymes. In: Loewus, F.A., Runeckles, J.C. (Eds.), Recent Advances in Phytochemistry: The Structure, Biosynthesis and Degradation of Wood. Plenum Press, NY, pp. 311–367.
Rowell, R.M., Ellis, W.D., 1979. Chemical modification of wood. Reaction of methyl isocyanate with southern pine. Wood Sci. 12 (1), 52–58.
Savory, J.G., 1954. Breakdown of timber by Ascomycetes and fungi imperfecti. Ann. Appl. Biol. 41, 336–347.
Scheffer, T.C., Lindgren, R.M., 1940. Stains of Sapwood Products and Their Control. U. S.D.A., Technical Bulletin No. 714, Washington, D.C.
Shigo, A.L., 1967. Successions of organisms in discoloration and decay of wood. Int. Rev. Forestry Res. 2, 237–299.
Shigo, A.L., Marx, H., 1977. CODIT (Compartmentalization of Decay in Trees). U.S.D. A., Information Bulletin No. 405, Washington, D.C, 73 pp.
Verrall, A.F., 1965. Preserving Wood by Brush, Dip, and Short Soak Methods. U.S.D.A. Forest Service, Technical Bulletin 1334, Washington, D.C.
Verrall, A.F., 1966. Building Decay Associated With Rain Seepage. U.S.D.A. Forest Service Technical Bulletin 1356, Washington, D.C.
Wilcox, W.W., 1970. Anatomical changes in wood cell walls attacked by fungi and bacteria. Botanical Rev. 36 (1), 1–28.

Sources of historical information on early developments in forest pathology, wood pathology, and wood preservation

Graham, R., 1973. History of wood preservation. In: Nicholas, Darrel. (Ed.), Wood Deterioration and Its Prevention by Preservative Treatments. Vol. 1 Degradation and Protection of Wood. Syracuse University Press, Syracuse, New York, pp. 1–30.
Hartley, C., 1950. The division of forest pathology. U.S. Dept. Agr., Plant Disease Reptr. Suppl. 195, 445–462 (detailed coverage of the early history of the Division of Forest Pathology and detailed review of principal research achievements).
Hubert, E.B. 1931. Outline of Forest Pathology. McGraw Hill, New York. 534 pp. (A brief historical treatment of forest pathology and wood pathology including the early developments in Europe).
Liese, W., 1967. History of wood pathology. Wood N.V., Leiden, Netherlands. Sci. Technol. 1, 169–173.
Merrill, W., Lambert, D.H., Liese, W., 1975. Robert Hartig 1839–1901. Phytopathological Classics No. 12. American Phytopathological Society, St. Paul, Minn, The authors' introduction to their English translation of Hartig, R. 1874. Wichtige Krankheiten der Waldbaume.
Van Groenou, H.B., Rischen, H.W.L., Van Den Berge, J., 1951. Wood Preservation During the Last 50 Years. A.W. Sitjthoff's Hitgeversn, aatschappij, Amsterdam, The Netherlands.

Additional sources for mycological, Phytopathological, and forestry terminology and definitions

Agrios, G.N., 1988. Plant Pathology, third ed. Academic Press, New York, p. 803 (a useful glossary).
Hawksworth, D.L., Sutton, B.C., Ainsworth, G.C., 1983. Ainsworth & Bisby's Dictionary of the Fungi, seventh ed. Commonwealth Mycological Institute, Kew, Surrey.
Link, G.K.K., 1933. Etiological phytopathology. Phytopathology 23, 843–862. Society of American Foresters. 1971. Terminology of Forest Science: Technology Practice and Products. Society of American Foresters, Washington, D.C.

Whetzel, H.H., 1929. The terminology of phytopathology. Int. Cong. Plant Sci. Proc. 2, 1204–1215.

Sources for additional information on wood uses and treatments

Anon, 2010. Wood Handbook: Wood as an Engineering Material. Forest Products Laboratory, Forest Service, U.S. Dept. Agric., Madison, WI.

Core, H.A., Cote, W.A., Day, A.C., 1979. Wood Structure and Identification, second ed. Syracuse University Press, Syracuse, N. Y.

Fleischer, H.O., 1976. Future role of wood as a material. Am. Inst. Chem. Eng. Symp. Ser. 71 (146), 1–3.

Goldman, I.S., 1975. Potential for converting wood into plastics. Science Sept (12), 847–852.

Haygreen, J.G., Bowyer, J.L., 1989. Forest Products and Wood Science: An Introduction. Iowa State University Press, Ames, Iowa, 500 p.

Kollmann, F.F.P., Cote, W.A., 1968. Principles of Wood Science and Technology, vol. 1. Springer-Verlag, New York.

Nicholas, D.D., 1973. Wood Deterioration and Its Prevention by Preservative Treatments. Vol. 1. Degradation and Protection of Wood. Vol. II. Preservatives & Preservative Systems. Syracuse University Press, Syracuse, New York.

Panshin, A.J., deZeeuw, C., 1980. Textbook of Wood Technology: Structure, Identification, Uses, and Properties of the Commercial Woods of the United States, fourth ed. McGraw-Hill Inc, New York, NY, 722 p.

President's Advisory Panel on Timber and the Environment (PAPTE), 1977. Report of the President's Advisory Panel on Timber and the Environment. Gov't Printing Office, Washington, D.C.

CHAPTER TWO

Wood deterioration agents

Contents

Though wood is readily decomposed and recycled in the forest ecosystem by various biotic and abiotic agents, it is a very durable organic material when properly used and maintained. Evidence of this durability in the northeastern United States lies in the sound condition of many wooden homes after several hundred years of use. Longer, effective uses of wood are known for churches in Norway and ornate temples in Japan. Samples of wood found in the tombs of the Pharaohs are reported to show no signs of deterioration.

Yet, under some conditions of exposure or use—often when it becomes wet—wood is rapidly decomposed by biotic (living agents) or abiotic (chemical or physical agents) acting alone or in combinations.

Three major types of biotic agents are responsible for most wood destruction. Fungi, and to a lesser extent, bacteria, are believed to cause the majority of wood losses. Insects such as termites and beetles also cause major deterioration losses, particularly in tropical regions. Marine borers cause substantial damage to wood submerged in salt or brackish waters in temperate and tropical zones.

Combustion and weathering are the principal abiotic types of wood destruction. Combinations of biotic and abiotic agents are often involved

Wood Microbiology.
DOI: https://doi.org/10.1016/B978-0-12-819465-2.00002-4

in the wood decomposition process in natural environments. For example, termites, carpenter ants, beetles, or wasps may invade wood prior to and in the early stages of fungal decay and accelerate the destruction. Insect adults and larvae may introduce decay and stain fungi to the wood and, in some cases, utilize them as a food source. Presumed symbiotic associations have developed between some wood inhabiting insects and fungi. Wood weathering appears to result from the combined action of photo-degradation, chemical oxidants, and fungi.

While this textbook focuses primarily on wood decay and discoloration damage caused by fungi, it is important to be aware of the other types of damage. Some may be confused with types or stages of decay. Others may be associated with decay development or occur independently on the same wood product. For example, mechanical damage to loading platforms or docks and chemical spills on floors may superficially resemble late stages of certain decays that are fibrous in texture. Advanced weathering damage on a utility pole surface may superficially resemble soft rot. Prolonged exposures of wood to high temperatures, such as in dry kilns, produces surface discolorations resembling some biotic stains. Enzymatic stains that develop in seasoning lumber can be confused with sapstain caused by fungi. Termite or carpenter ant damage may superficially resemble the final stages of decay where the annual rings separate.

The purpose of this chapter is to present an overview of the major deterioration agents other than fungi that, under some conditions of exposure or use, may damage wood. Emphasis will be placed on defect appearances, causal agents, factors affecting development, wood property changes, prevention or controls, and any relationship to fungal defects. Since coverage of these non-fungal wood deterioration problems is not a prime purpose of this presentation, major reliance will be placed on references and recent review articles.

The major agents and types of wood decomposition are grouped under abiotic and biotic categories and listed as follows:

Abiotic damage

1. Weathering—primarily photodegradation by ultra-violet light and oxidation.
2. Thermal decomposition—distillation or burning.

   a. Low temperature exposure (below 200 °C).
   b. High temperature exposures in absence of oxygen or pyrolysis (above 200 °C).
   c. Combustion (above 275 °C).
3. Chemical decomposition—hydrolysis and oxidation.
   a. Exposure to strong acids.
   b. Exposure to strong bases.
   c. Exposures to strong oxidizing agents and some organic solvents.
4. Mechanical wear—breakage and erosion of surface fragments.

Biotic damage

5. Animal attack—mechanical disruption.
   a. Borings and surface rasping by marine borers.
   b. Tunneling and excavation by insects (termites, boring beetles, and Hymenoptera such as carpenter ants) and marine borers (shipworms, pholads, and gribbles).
6. Decays and discolorations—penetration and digestion.
   a. Cell wall etching and tunneling by bacteria.
   b. Surface molding by fungi.
   c. Sapwood staining by fungi.
   d. Decays by fungi (soft rots, brown rots, and white rots).

Wood weathering

Weathered wood often has a familiar gray color that many find aesthetically pleasing. As a result, weathered wood is highly valued for decorative and interior wall paneling due to its pleasing color and texture. These colors are caused by chemical and physical disintegration near the surface. This damage has no appreciable effect on strength and is primarily a disfigurement problem. Freshly surfaced wood begins to change color after a few weeks of outdoor exposure, although the chemical reactions associated with this damage begin within 24 hours of exposure. Light woods darken, slowly initially, to a brown. Dark woods first bleach and then also become brown. After several years of outdoor exposure, the wood surface gradually develops an attractive gray sheen and roughened texture.

The process of photodegradation is also called actinic degradation which describes solar radiation leading to degradation of a material. The major events associated with weathering are photochemical damage (short-wave and long-wave UV) to wood cell wall constituents, oxidation of the breakdown products, leaching of the soluble decomposition products, and related mechanical damage of surface elements from the constant swelling and shrinkage of the wood associated with surface wetting and drying.

The initial color change to brown occurs as the energy in the sunlight is released into the wood. This energy leads to the formation of free radicals that initially affect lignin and extractives. Continued creation of free radicals leads to further decomposition of the structural carbohydrates and oxidation of phenolic moieties. The origins, identities, and characteristics of these free radicals have been reported by Hon et al. (1980). Surface leaching then removes the soluble decomposition products, exposing the more photo-resistant structural carbohydrates that are also photo-chemically degraded and oxidized by decomposition products and atmospheric agents. Xylans are decomposed and leached more readily than cellulose or glucan-rich hemicelluloses. The residual cellulose and the surface growth of pigmented fungi such as *Aureobasidium pullulans* form the gray color. The weathered outer shell that develops greatly reduces the weathering rate and protects the surface wood from further photochemical damage. However, continual wetting and drying of the weathered surface with its concomitant swelling and shrinking leads to surface checking, localized mechanical failures, and slow exfoliation of the weathered surface. These weathering losses are negligible over the service life of most large wood assemblies; however, they may become significant for long-term uses of wood in implements, thin plywood surfaces, and some round stock such as insulator pins on utility poles. There is considerable variation in weathering rates that reflect differences in geographic location, test method, and wood species. Jamison (1937) reported that weathering of western white pine dowels (5 cm in diameter by 45.7 cm long) suspended above ground outdoors in Idaho for 10 years in full sunlight, partial shade, or dense shade caused weight losses of 7.9, 4.8, or 2.3%, respectively. Smaller dowels (1.25 cm in diameter) lost 16.4% of their weight after a seven-year exposure in full sunlight. Weathering has been estimated to remove 6–7 mm of the outer wood surface per century in temperate zones (Browne, 1960; Feist, 1977; Kuhne et al.,

1972) and 1 mm per century for wood exposed in northern climes. Rates will tend to be faster in more extreme UV exposures.

Microscopic examinations of weathered wood indicate that substantial micro-checking occurs between cells and that wood substance is lost around bordered pits on radial walls. Only remnants of the ray cells remain after prolonged UV exposure (Minuitti, 1967). Micro-checking and splits between cells suggests wall embrittlement by photo-degradation which probably facilitates the surface fragment exfoliation with moisture changes. Earlywood fibers are damaged more readily than latewood fibers, accounting for the rough, corrugated surface of severely weathered wood.

Coatings or films that absorb or reflect the damaging UV portion of light and reduce surface moisture changes are the conventional methods of preventing weathering in outdoor wood exposures. Water-repellent treatments also reduce moisture fluctuations in some outdoor wood uses. Wood treatments with chemicals such as chromic acid reduce weathering and are reported to double the service life of latex and oil-based paints (Feist and Ellis, 1978). Hexavalent chromium, however, is also a potential carcinogen and its use is severely restricted. A comprehensive review of wood weathering and an extensive historic bibliography on the topic was assembled by Feist (1982).

Virtually all carbon-based materials experience actinic degradation including cellulose-based fabrics and even many of the synthetic polymers such as high-density polyethylene or polypropylene used in wood/plastic composites. A number of additives are incorporated into these materials to slow this process. In some cases, the goal is to render a material more susceptible to photodegradation such as disposable paper products to facilitate biodegradation and minimize ecological concerns in landfills. The yellowing and embrittlement of paper from aging is a serious problem in libraries that results from the oxidation of residual lignin moieties and is related again to the more general phenomenon of wood weathering. Although it has little practical effect on wood properties, disfigurement of wood surfaced via weathering remains one of the major problems with wood in exposed applications and is a major cause of early replacement.

Wood thermal decomposition

The thermal decomposition of wood, as with most carbon compounds, occurs readily at elevated temperatures. Slow changes begin around 100 °C and can include color changes, serious strength losses, reductions

in hygroscopicity, weight losses, and the evolution of gases such as CO, CO_2, CH_4, and water vapor. The changes are time dependent and increase rapidly at higher temperatures. Combustion, with the emission of light and heat, occurs at temperatures around 275 °C.

Low temperature processes (below 200 °C): Low temperature effects on wood are particularly important because significant strength losses occur within this range. Wood thermal decomposition begins at 100 °C. Over prolonged exposures, wood turns brown, the surface becomes brittle, and slow losses in weight and strength occur. This temperature effect can be readily observed on blocks left in a hot-air oven (104 °C) for a few weeks or lumber that is over-dried in a dry kiln. The brownish color and surface brashness of the wood resemble the early stages of some fungal decays (brown rots). The uniformity of discoloration and the lack of fungal structures readily separates heat damage from early decay.

MacLean (1951) determined wood weight losses associated with various temperatures and periods of exposure, averaged for 11 commercial species, as follows:

Exposure period	Temperature (°C)	Weight loss-percent
1 yr	93	2.7
470 d	121	26.8
400 h	149	14.8
102 h	167	21.4

Strength is rapidly reduced by exposures to elevated temperatures (Fig. 2.1). No smoke or wood glowing is produced when wood is exposed to temperatures below 200 °C, but the polymers begin to decompose releasing CO_2 and water vapor.

High-temperature processes (above 200 °C): Most wood used on this planet is burned for heating and cooking.

In combustion, the wood is rapidly decomposed at temperatures above 200 °C in the presence of oxygen, and the flammable gases CH_4 and CO are released. At temperatures around 275 °C, the gases ignite (the kindling temperature) and, thereafter, the heat released accelerates the burning or decomposition process. While wood burns, one attractive feature of timber is that it burns at a steady rate. Thus, providing ample time for evacuation of a structure.

Pyrolysis (heating in absence of O_2) is a well-known wood distillation process during which the flammable gases evolved are CH_4 and CO along

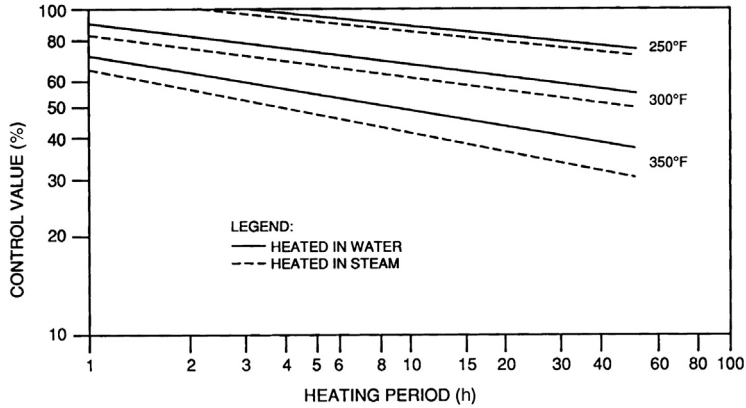


Figure 2.1 Comparison of average values of modulus of rupture (%) for Douglas-fir and Sitka spruce specimens heated in water and in steam at 250°F, 300°F, and 350°F. *From MacLean, J.D. 1954. Effect of heating in water on the strength properties of wood. Proc. Am. Wood Preserv. Assoc. 50, 253–280.*

with many other compounds such as acetic acid, methanol, formic acid, furfural, phenols, and cresols. The acids cause the eye sting of smoke, and the furfural products contribute to the characteristic odor of wood smoke. The remaining final product is charcoal (carbon), which has many industrial uses. There is periodic interest in pyrolysis of wood for bioenergy as well as the production of biofuels. This interest is typically driven by increased costs for traditional fossil fuel sources

The order of wood cell wall component breakdown with increasing high temperatures is hemicelluloses, cellulose, and lignin. Hemicelluloses are least heat stable and decompose within a temperature range of 225–325 °C. Lignin decomposes over a temperature range of 250–500 °C, while cellulose decomposition begins at higher temperatures and in a more limited range (325–375 °C) (Shafizadeh and Chin, 1977). The texture of carbonized wood at late stages of decomposition resembles a brown cubical rot, and it is of interest to note also that lignin, as in some decays, is the last wood component to be completely consumed.

Fire-retardant chemicals: Effective retardant chemicals include ammonium phosphates, guanyl urea phosphate, ammonium sulfate, borax, urea, and zinc chloride. Most fire retardant formulations have been empirically determined, and their protective mechanisms remain poorly understood. Many fire retardants actually accelerate the initial charring of the wood, creating a layer of char that limits further access to oxygen. The more acidic fire retardants can actually initiate wood degradation at

lower temperatures and there were very active research programs in the 1990s to develop non-acidic systems for timber.

A reference book has been assembled on the Chemistry and Uses of Fire Retardants (Lyons, 1970), and there are several comprehensive reviews of the degradation and protection of wood from thermal attack (Goldstein, 1973; LeVan, 1984; White and Dietenberger, 2010).

Chemical decomposition of wood

As a structural material, wood displays considerable resistance to attack by most chemicals. For this reason, wood is often used to construct storage vats, tanks, cooling towers, or structures where contact with caustic chemicals may occur by condensation, aerosols, or splashing. For example, evidence of considerable wood deterioration was reported in several Kraft pulp mills due to prolonged wood exposure to weak acids or bases at elevated temperatures and high humidities (Barton, 1982). As a major chemical raw material for the paper and cellulose derivative industries, a wealth of information has been developed on the reactions of wood and its constituents to produce many chemicals. This information forms the basis of a wide range of industrial processes. Information on these topics is available in wood chemistry, cellulose chemistry, and papermaking textbooks. This section reviews only those cases where wood contact with caustic chemicals in various structural uses causes degradation that may be confused with some types of decay or the resulting damage has close analogies to the decay process.

Smith (1980) developed a list of timber species recommended for use in various corrosive environments such as containers for acids, exposure to acid fumes, or containers for mild corrosive liquids.

Coniferous woods are generally more resistant to corrosive chemical attack than most hardwoods and less permeable wood species tend to degrade more slowly. Chemically resistant woods are those species that are high in alpha cellulose, lignin, and low in xylans.

Acids degrade primarily the carbohydrates in wood, and the high resistance of lignin to strong acids is the basis for its analytical determination by solubilizing wood carbohydrates with 72% H_2SO_4. The resulting, filtered insoluble residue is defined as Klason lignin. Acids hydrolyze the β

(1−4) glycosidic linkages in cellulose and hemicellulose resulting in drastic reductions in tensile strength. The wood in early decomposition stages turns brown and becomes brittle or brash. The depolymerization mechanisms and early reductions in some strength properties are analogous to brown rot degradation.

Alkalis attack wood more severely at equivalent concentrations and time temperature conditions than acids. Alkalis dissolve the hemicelluloses and modify the lignin to form soluble lignin-alkali complexes. The cellulose is essentially unmodified. Many wood pulping processes employ alkaline reactions of this type.

High concentrations of strongly alkaline chemicals cause the wood to become fibrous and bleached in a manner similar to wood attacked by some white rots. The wood swells and sharp strength reductions occur.

Selected data from Wangaard (1966) illustrates the differences between the effects of a strong acid and strong base at several concentrations and temperatures on the strength of a conifer and a hardwood (Table 2.1).

Wood exposed to a series of alcohols, acetone, and benzene can experience decreased swelling and strength loss as molecular weight and structural complexity of these organic compounds increases (Erickson and Reese, 1940). Treatment of wood with ammonia temporarily causes large reductions in bending resistance and permits the wood to be bent at sharp angles into a variety of shapes without breaking.

The treatment of wood with some salts is reported to increase crushing resistance while treatment with acid salts such as Na_2CrO_3 reduces strength (Ross, 1956). The treatment of wood with salt-type preservatives such as the oxides or acid salts of copper, chromium, and arsenic (CCA) does not appear to seriously affect strength (Thompson, 1982) unless the wood is subsequently dried at high temperatures (Barnes and Winandy, 1986).

Table 2.1 The effects of temperature and several concentrations of HCl and NaOH on wood strength (modulus of rupture).

Species	Modulus of rupture (as a percent of control)							
	2% HCl		10% HCl		2% NaOH		10% NaOH	
	20 °C	50 °C	20 °C	50 °C	20 °C	50 °C	20 °C	50 °C
Douglas-fir	91	85	76	57	56	40	39	28
White oak	70	51	39	30	26	22	20	15

Data selected from Wangaard, F.F., 1966. Resistance of wood to chemical degradation. For. Prod. J. 16 (2), 53−64.

The prolonged contact of wood with iron causes localized embrittlement and loss in tensile strength (Baker, 1974). There are reports that wood decomposition from iron reduces nail-holding properties in cases where the fastener was initially driven into green lumber. The use of galvanized fasteners or dry lumber when feasible minimizes the problem. As iron oxidizes (rusts) to form ferric hydroxide, it catalyzes the oxidation and depolymerization of cellulose into oxycellulose. It is interesting to note that one theory on brown rot decay mechanisms proposed a similar catalytic role for iron to initiate cellulose decomposition (Koenigs, 1974). A comprehensive study of the various effects of iron on wood properties has been reported by Marion and Wissing (1960).

Mechanical wear

The mechanical wear of wood is a minor source of wood deterioration and involves forces that rupture and detach small portions of the surface wood. This damage is important only in a few cases of special wood uses where surface friction or rupturing is severe, such as stair treads, baffles in cooling towers, factory floors around heavy machinery, and spikes and plate contacts in railroad ties. Wind-driven sand particles can cause considerable mechanical damage to poles, posts, and unpainted wood in desert regions and along beaches. Another example of mechanical damage is seen frequently on loading docks or platforms. Heavy loads with sharp corners abrade and split the surface wood. Over time, this damage develops a fibrous texture similar to some late decay stages. Preventative methods include the selection of woods with high surface hardness, edge grain alignment of wood in severe friction zones, and, in some cases, the protection of high damage zones with metal plates or use of polymer-hardened wood.

Insect damage to wood

Insects, 1ike fungi, are also major agents in the biodeterioration of wood. Insects use wood for habitation and as a food source. The wood is chewed into small fragments for both purposes. Collections of the chewed fragments and/or fecal material, known as frass, are a useful indicator of hidden insect damage and can sometimes be used to identify the specific organism involved. Insect damage to wood generally consists of discrete tunnels, surface channels, or chewed zones and, in most cases, can be distinguished readily from the fungal-associated stains and decays. Insects are common vectors of stain and decay fungi, and insect and fungal damage

often develop under the same conditions and are associated in many wood uses. Hence, a review of the major types of insect damage, their characteristics, prevention, and/or control is necessary.

Insects are the largest class in the Arthropoda and are characterized by segmented bodies, jointed legs (six), and a hard, chitinous exoskeleton. Of the six insect orders that cause wood damage, the Isoptera, Coleoptera, and Hymenoptera are the most important. Insects are reported to cause many billions of dollars in damage to wood structures annually (Coulson and Lund, 1973; Baker, 1972; Furniss and Carolina, 1977). In fact, subterranean termites cause over 5 billion dollars in damage in the United States per year (Ebeling, 1968). In many cases, insect infestations are also associated with fungal decay that further aggravates the damage (Amburgey, 1979).

Types of damage: Types of insect damage are classified according to the causal species for scientific purposes and the type of damage, the wood product, or time of attack for practical purposes. Wood can be damaged in the living tree, the freshly fallen log, the sawn or round products in storage, or in the final product during service. Considerable insect damage takes place in weakened or freshly fallen trees and stored logs, but the effects show up later in the finished wood product. Common terms for the many types of damage are powder posting, pith flecks, birdseye, pitch pockets, pin holing, honeycombing, and grub holes.

Insect life cycle: Insects typically have life cycles that begin with an egg and progress through several immature molts, climaxing with a fully functional adult that mates and produces eggs to complete the life cycle (Fig. 2.2). Two types of development are: complete metamorphosis in which the insect passes through egg, larval, pupal, and adult stages and incomplete metamorphosis consisting of egg, nymph, and adult stages.

The stage at which insects attack wood varies, but generally the Coleoptera damage wood during the larval stages, while the Isoptera and Hymenoptera cause their damage in the nymph and adult stages.

Isoptera (termites)

Isoptera: The wood-attacking termites are members of the order Isoptera and live in large social colonies consisting of castes that differ in appearance and function (Krishna and Weesner, 1969; Ebeling,

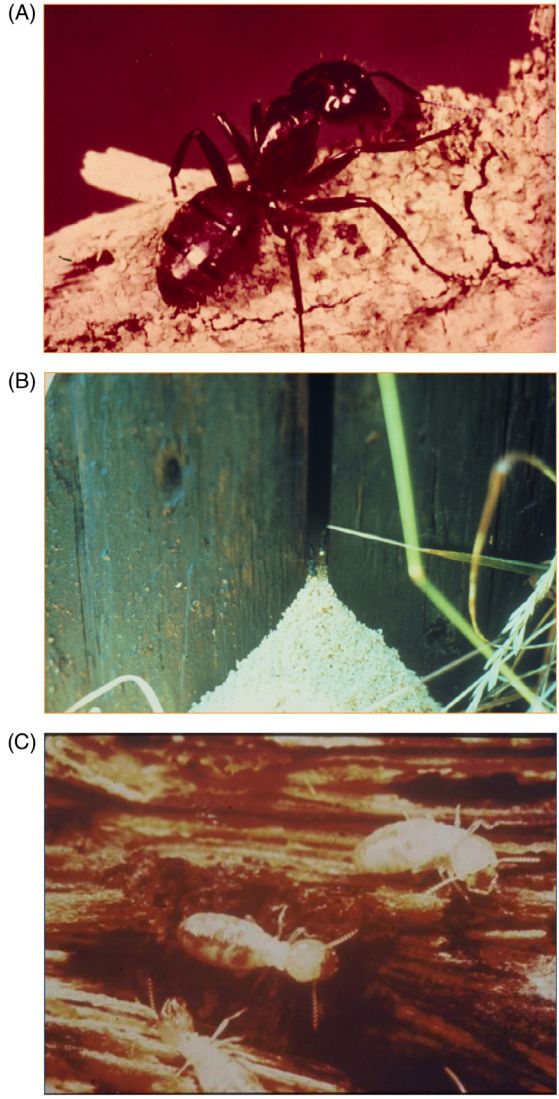


Figure 2.2 Termite and carpenter ant damage is often confused, although the differences are striking. (A) Carpenter ant workers with deep constrictions between body segments. (B) Workers excavate tunnels and remove the excess wood. (C) Termite workers without noticeable constrictions between body segments in timber that has a dirty appearance. *Courtesy Dr. John Simeone.*

1968; Beal et al., 1983). All members in a given colony are descendants of one original pair. Termites are often confused with ants and are commonly called "white ants." Termites differ in many ways from ants including morphology, food sources, and environmental requirements.

There are over 2000 termite species located worldwide. These species are generally confined to regions where the average annual temperature exceeds 10 °C (50°F). This zone lies approximately between the latitudes 50° South and 50° North although these zones are expanding as climates become warmer (Fig. 2.3). Some termites may also extend their ranges further north or south of these zones in heated, man-made structures (Esenther, 1969). The colony usually begins when a male and female reproductive mate and the female begins to lay eggs. Over time, the now "queen", lays eggs that develop into workers that forage for food and feed the queen or, eventually, soldiers that guard the colony. A single Formosan subterranean termite colony can approach 7 million workers, giving termites the potential to cause massive damage once they enter a structure.

Termites have specific environmental requirements including a food source (generally wood), oxygen, and adequate moisture levels. Termites generally occupy wood internally and are negatively phototropic. They seem to require higher than ambient levels of carbon dioxide. The wood is chewed and digested in the hindgut by enzymes released from associated symbiotic protozoa and/or bacteria. These protozoa are not present in the newly hatched nymph, but are transferred by exchange of body secretions and by consuming dead or dying members of the colony (Moore, 1979). Termites utilize primarily the cellulose in the wood, and the fecal pellets contain high levels of lignin. It is speculated that more advanced termite species may produce cellulase themselves or utilize, in some cases, the cellulase enzymes released in wood by decay fungi. Some termites appear to be attracted to the chemicals produced by some decay

Figure 2.3 Map showing the northern limits for distribution of termites in the United States. (A) Subterranean and (B) drywood termites. *USDA Forest Products Laboratory.*

fungi (Esenther et al., 1961). Fungal termite attractants could be useful in termite detection and control procedures.

Termites vary in the amounts of water required to establish successful colonies. Drywood termites, so-called because of their ability to attack dry wood (<13% water content), obtain their water needs from the wood and are highly efficient in their uses of water. Dampwood and subterranean termites require more water and invade wood that is constantly moist and usually in ground contact. Some termite species also construct earthen tubes that connect wood above the ground with the soil. The humid air in these tubes contacts the wood above the ground and increases its moisture content. Termites also uses these tubes to carry moistened soil to the wood above, making it easier to attack.

Termite groups: Of the six major termite families, only the dampwood, subterranean, and drywood termite families are important in the United States (Table 2.2). Of these three groups the subterranean termites have the widest distribution and cause the most damage.

The majority of subterranean termites of economic importance in the United States are species in the genus *Reticulitermes*. As their name suggests, subterranean termites build their nests in the soil, although they can survive in extremely wet wood not in ground contact. These termites infest woody debris in soil and invade wood structures through direct soil contact. Subterranean termites also build earthen tubes over masonry or concrete foundations to reach wood above the ground (Fig. 2.4). Initially the termite workers chew and digest the less dense springwood, leaving the summerwood unless they are forced to utilize this wood later (Behr et al., 1972). As they chew and tunnel through the wood, termite workers deposit small amounts of soil and fecal matter into the wood, giving the damaged wood a characteristic "dirty" appearance. The most common subterranean termite in North America is *Reticulitermes flavipes*, but there are a number of other species in this genus found across the country. A well-established colony can approach one million workers. In addition to

Table 2.2 Families of termites that can attack wood.

Family	Common name	Type of damage
Rhinotermitidae	Subterranean termites	Honeycomb damp wood
Kalotermitidae	Drywood termites	Honeycomb dry wood
Termitidae	Subterranean, mound builders	Honeycomb cellulose
Mastotermitidae	–	Honeycomb damp/dry wood
Hodotermitidae	Harvester termites	Plant consumers

Figure 2.4 Stain and tunnels (pinhole galleries) damage in soft maple lumber caused by attack of ambrosia beetle (*Trypodendron lineatum* Oliver) after the tree was cut. Most ambrosia beetles invade dying trees, slash, or green roundwood. Ambrosia beetle attack is sometimes confused with decay if the tunnels are not obvious.

Figure 2.5 Carpenter ant invasion and internal damage in a treated Douglas-fir utility pole is indicated by the accumulating pile of fresh frass (arrow) at the base of the pole.

R. flavipes, the Formosan termite, *Coptotermes formosanus*, was introduced into the United States from Asia. It is prevalent in most of the Hawaiian Islands as well as the Gulf Coast and very southern tip of California. Formosan termites are characterized by a rapid feeding rate, large colony size, and apparent tolerance of many commonly used wood preservatives. Formosan termite nests can be distinguished from subterranean termites by the presence of an extremely durable secretion called "cardboard" (Fig. 2.5). A major effort is underway to reduce the spread of this insect from the few areas where it has become established. Fortunately, its range appears to be limited to warmer climates.

Dampwood termites are confined to the Pacific Northwest, the Pacific Southwest, and southern Florida. These species are characterized by their need for very wet wood. Wood invaded by these termites is generally in ground contact, but reproductive swarmers can also infest very wet wood in solid lumber stacks or wood subjected to continuous wetting. These insects are often associated with wood decay due to the higher wood moisture levels. As with subterranean termites, dampwood termites remove the softer springwood first and also preferentially remove the weaker, decayed wood. Wood damaged by dampwood termites can be recognized by the appearance of the frass, which is squeezed into pellets to recover moisture.

Drywood termites do not require soil contact or high wood moisture content to invade wood. As a result, these insects can attack wood in roofs, rafters, and other building zones not normally considered to be susceptible to termite attack. Fortunately, drywood termites are confined to the Pacific Southwest and cause damage only in a few geographic areas. Drywood termite colonies are often transported in wood items such as furniture to areas far outside their natural range. While they can survive in these protected environments, they generally do not spread because the climatic conditions are not suitable for new colony establishment.

Wood damaged by drywood termites differs from the others in that the galleries overlap the springwood/summerwood boundaries. Drywood termites produce distinctive barrel-shaped fecal pellets that are pushed out of the gallery through holes termed "kickholes". These holes are immediately resealed, but the deposition of the fecal pellets outside the wood is an excellent indicator of termite attack.

Termite prevention: Entomologists have long sought effective preventive methods for limiting termite damage (Moore, 1979; Snyder, 1969). Much of this effort has been exerted to control subterranean termites since these insects cause most of the insect-related wood damage. Termite damage can be reduced by several simple construction practices including removing wood debris around structures, properly sealing cracks in cement floors or foundations, filling the top layer of hollow concrete blocks with cement or capping with solid so-called "termite blocks" to prevent termite invasion, and using pressure-treated wood when wood is in soil contact (DeGroot, 1982; Moore, 1979; Beal et al., 1983). Regular inspections of crawl spaces or foundations in structures are recommended to detect infestations before substantial damage occurs. Termite attack can also be prevented by the use of chemical soil drenches around the base of a wood

structure. Chemicals formerly used for this purpose have included Aldrin (0.5%), Chlordane (1.0%), Dieldrin (0.5%), or Heptachlor (0.5%). Chloropyrifos has also been used with some success (Moore, 1979) as have some synthetic pyrethroids, but changing perceptions about the use of chemicals have encouraged alternative protection methods. Changes in insecticides permitted at the federal and state levels require that users check with local Extension Service offices to determine current recommended insecticides for termite control. A number of non-chemical control methods have been developed for limited the risk of termite attack. These generally involve physical barriers in the ground underneath a structure. Two common approaches are to install a stainless steel screen underneath the foundation with holes that are too small for termites to penetrate. Alternatively, finely ground granite or volcanic rocks are placed between the soil and the concrete foundation to act as a barrier to upward termite tunneling. In both cases, the barrier must be complete or else the workers will eventually locate gaps in the protective layer.

Dampwood termites are controlled by the procedures used for the subterranean termite; however, prevention can be more easily affected by removing wood from direct soil contact or by removing moisture sources.

Drywood termite attack is more difficult to control since the wood need not be wet or in ground contact for damage to occur. Infestations can be limited by screening around vents or crawl spaces, removing infested wood, and by the use of fluoridated silica aerogel dusts (Wagner, 1965).

Termite control: Once a termite colony has been detected in a structure, many of the same techniques used for prevention can be used to eliminate the infestation. In addition, application of emulsified insecticides under pressure through holes drilled in the wood can speed up colony demise; however, care must be taken to avoid over-application of pesticides. A more recently developed approach to termite control involves establishing baiting stations around a house. Once termites feed on the untreated bait in the station, wood treated with a system pesticide is substituted. The workers feed on these stakes and carry the pesticide back to the colony where it spreads through the colony, eventually causing colony failure.

External application of these chemicals has little effect since termites rarely venture out of the wood. Tenting the structure followed by fumigation with methyl bromide or sulfuryl fluoride has been successful with drywood and formosan termite infestations (Moore, 1979), but these chemicals have short residual times in the wood and will not prevent

reinfestation. Methyl bromide is also under considerable regulatory pressure due to its contributions to the depletion of the ozone layer.

Coleoptera (beetles)

Beetles are members of the order Coleoptera, the largest order of insects containing nearly 40% of the known species. There are nine families of Coleoptera that cause wood damage; most species attack only living trees, logs in storage or seasoning lumber (Table 2.3), but they are important because the defects they cause appear later in the final wood product and may be confused with active wood infestations. Most beetle damage is caused by insects in their larval stages. The wood species attacked and the conditions necessary for attack vary widely with insect species.

Pre-harvest beetles: A number of beetles in the families Brentidae, Lymexylidae, Scolytidae, and the Platypodidae attack standing and freshly cut trees. These insects normally do not cause damage to seasoned wood, although they may continue to damage wood as it initially seasons. The first two families cause extensive damage to hardwood logs that are not removed promptly from the woods. The Scolytidae tends to mine in between the bark and the xylem, where they cut off the flow of water and nutrients to the living tree. Mortality typically occurs when large numbers of beetles attack weakened trees. These beetles typically carry a sapstain fungus into the wood and the growth of these fungi into the sapwood sharply reduces value (Fig. 2.6). The Platypodidae also carry stain fungi into the wood, but the adults of these beetles tend to tunnel more

Table 2.3 Families of wood-destroying Coleoptera.

Family	Common name	Damage	Product type
Anobiidae	Death watch beetle	Powder posting	Furniture, structures
Bostrichidae	Powder-post beetle	Powder Post	Hardwood lumber
Brentidae	Timber worms	Tunneling	Hardwood logs
Buprestidae	Flat-headed borers	Tunneling	Lumber and product
Cerambycidae	Round-headed borers	Tunneling	Trees and products
Lyctidae	Powder-post beetle	Powder posting	Hardwoods
Lymexylidae	Timber worms	Pinholes	Hardwood logs
Platypodidae	Flat-footed beetles	Pinholes, stain	Logs
Scolytidae	Bark beetle	Pinholes, stain	Trees or green log

Figure 2.6 Marine borers cause severe damage to wood in salt-water exposures in some regions. (A) Cross section of a Douglas-fir piling showing numerous tunnels causes by *Toredo* sp. (B) Mouth parts of a shipworm removed for the wood. (C) Piling form the intertidal zone showing the hour-glass shape produced by *Limnoria* attack.

deeply into the wood, leading to more widespread discoloration. Prompt removal of the bark and drying the wood can limit damage by these beetles. Where rapid processing is not feasible, the use of ponding or continuous spraying makes the wood too wet far insect development.

This problem is particularly acute with the Ambrosia beetles that invade freshly felled logs, forming small tunnels that may penetrate deep into the sapwood and later become associated with sapstain. Ambrosia beetles inoculate the tunnel walls with the spores of fungi carried in special structures termed mycangia. They obtain nourishment from the growth of the fungi that produce a shallow, gray stain in the tunnel wall. These fungi can be particularly troublesome when freshly felled trees are stored under moist conditions, but they can also attack freshly sawn wood

where the damage is often mistaken for powderpost beetle attack. The wet condition of the wood clearly eliminates powderpost beetle attack. Ambrosia beetles have even been observed boring into wood that has been treated with copper-based waterborne preservatives because the adults do not feed on the wood and are, therefore, not affected by the treatment. Some ambrosia beetles attack living trees. The Columbia timber beetle (*Corthylus columbianus*) commonly attacks living soft maple, oak, and sycamore trees in the eastern United States. The unique stain pattern on logs ends or board surfaces that have experienced with successive beetle attacks is often confused with early decay. Ambrosia beetle attack can be limited by ponding, rapid processing, and spraying the logs with insecticides (Fisher et al., 1954; Gray and Borden, 1985; Mclean, 1985). Chemical treatment is generally not an attractive option because of the costs as well as the potential for runoff and contamination. Controlling bark beetle attack is discussed in greater detail in the later chapter on sapstains.

Post-harvest beetles: Wood that is cut, milled, and dried is still susceptible to attack by members of the Anobiidae, Bostrichidae, Lyctidae, Cerambycidae, and Buprestidae. The former three groups are collectively called the powder-post beetles because of the flour-like frass that the larvae leave in their tunnels (Moore and Koehler, 1980). Powder-post beetles are reported to be the most important wood products attacking beetles.

Anobiidae: The powder-post beetles in this family are also called the Death-watch or furniture beetles. The name derives from the tapping sounds adult beetles make with their heads as a mating signal. This sound is most easily heard in the walls of a quiet room such as that occupied by a person sitting with an ill person, hence the name origin.

There are numerous species of Anobiidae, but *Anobium punctatum* (an introduced pest), the common furniture beetle (*Xyletinus peltatus*), and (*Hemicoelus carinatus*) cause considerable wood damage. The Anobiidae are common in southern pine building timbers in the southeastern and hardwood structures in the northeastern United States. Powder-post beetles attack primarily sapwood, although heartwood is not completely immune to attack. The Anobiidae digest cellulose in the wood cell wall with the assistance of yeast cells in their digestive tract. In general, beetles in this family can attack wood at moisture contents of 15% and above, although the optimum moisture levels for development are at moisture contents greater than 30% or in decayed wood (Moore, 1979). Damaged wood has

numerous small tunnels packed with frass and small exit holes on the wood surface.

Bostrichidae. -These beetles are called the false powder-post beetles and commonly occur in dying twigs and branches of many hardwoods. They cause significant damage in hardwood lumber. These beetles attack hardwood sapwood, utilizing the starches present in the ray cells. Wood damaged by these beetles has numerous small tunnels filled with tightly packed frass.

Lyctidae: Lyctids, the true powder-post beetles, are reported to be the most important destructive agents of hardwoods (Williams, 1985). The adult beetles attack hardwood sapwood to obtain the free sugars in the ray cells. Lyctids infest wood at moisture contents ranging from 8 to 32% with the greatest activity between 10 and 20% (Christian, 1941; Moore, 1979). Unfortunately, many seasoned wood products fall within this moisture range. Wood damaged by Lyctids is filled with small tunnels loosely packed with frass. Adult exit bore holes may be evident in older infestations and frass will fall from these holes when the wood is jarred. Lyctid infestations can be prevented by sealing the surface to prevent access to the vessel elements or by application of boron solutions, but sealing will not affect existing infestations. Existing infestations are generally controlled by heating the core of the wood above 56 C for at least 30 minutes.

Cerambycidae: The round-headed borers comprise one of the largest beetle families, and many are associated with damage to trees or wood products. These beetles have long antennae that give rise to their also being called long-horned beetles.

In general, these beetles infest green or partially seasoned lumber when the bark remains attached. The adult female lays her eggs on the bark surface, the larvae hatch and tunnel inward where they typically mine the area between the bark and the xylem. After a period of time, they typically mine further inward where they continue to attack the wood until they obtain sufficient nutrition to pupate and emerge from the wood. Long-horn beetle damage consists of round to oval tunnels that are tightly packed with frass throughout the wood. Often these galleries are exposed in sawn material after the damage has occurred. Long horned beetles can be common in fire killed logs that are not rapidly process after the fire. For example, the new house borer (*Arhopalus productus*) lays its eggs on the bark of freshly killed softwoods. The larvae bore inward and tunnel for 1–2 years before emerging. Failure to rapidly

remove and process fire-killed trees can result in these beetles surviving processing and continuing to develop in the finished lumber if the wood is not kiln dried. The adults often emerge one to two years afterwards and can even bore through flooring and drywall.

The Asian longhorn beetle (*Anoplophora glabripennis*) was accidently introduced from Asia on solid wood packing materials. It attacks and can kill a wide range of hardwood species including maple, willows, and elms. This species is an excellent example of the damage that can be inflicted when a species is introduced into a new habitat where it has few natural enemies and a wealth of susceptible hosts.

While most long-horned beetles do not reinfest timber, several species including the old-house borer (*Hylotrupes bajulus*) can repeatedly reinfest the same wood and cause serious structural damage. This species is believed to be an imported pest that primarily attacks seasoned coniferous wood, but can also invade unseasoned wood. The old house borer is a serious problem in buildings in coastal regions. Generally, larvae develop more rapidly in wood between 15 and 25% moisture content, but will survive for long periods at lower moisture levels. The amount of damage associated with the larval stage is confined to galleries loosely packed with frass in a few isolated boards; however, more severe damage occasionally results when reinfestation occurs in areas with moisture problems.

Long-horned borer attack is often an indicator of delayed processing that has allowed the beetle larvae to tunnel deeply into the wood before processing.

Buprestidae: Buprestids, also called flat-headed borers, are also found in wood and wood products where they tunnel beneath the bark, in twigs, and in the heartwood or sapwood of freshly cut logs. As with the long-horned borers, the females lay their eggs on the bark surface then the larvae tunnel into the xylem.

Many buprestids attach living trees and their damage becomes evident on the finished product. One example is the western redcedar borer (*Trachykele blondeli*) attacks the heartwood of living western redcedar trees. It does not kill the tree, but the damage is exposed when lumber is sawn. A far more dangerous buprestid is the emerald ash borer (*Agrilus planipennis*) which is an invasive species from Southeast Asia that attacks and kills ash trees. It has spread into large areas of the U.S. Midwest and threatens the continued presence of ash as a forest tree.

Most buprestids do not attack seasoned wood, although some, such as the golden buprestid (*Buprestis aurulenta*) can survive for long periods in

dry wood. Many buprestids are metallic colored, and members of this group are also called the metallic wood borers. Larvae of these species are distinguished by their flattened appearance near the mouth, and the tunnels they bore are tightly packed with frass. Following pupation the adults chew their way out of the wood, leaving a characteristic D-shaped exit hole on the surface. Generally the amount of damage associated with buprestid attack is minimal; however, heavy infestations of the golden buprestid can cause significant structural damage in log structures and utility poles. Although reinfestation is not reported, the high levels of damage in isolated structures suggest that reinfestation does occasionally occur. As with the long-horned borers, buprestids can survive through sawing and continue to develop in the finished product if the wood is not kiln dried.

Hymenoptera (bees and ants)

While termites and beetles cause the majority of insect-related wood damage, several members of the Hymenoptera including the Siricidae, Apidae, and Formicidae also significantly damage wood.

Siricidae: The Siricidae or horntail wasps attack trees that are stressed and declining as well as fire-damaged or freshly-harvested timber. The female penetrates the bark and wood with a long ovipositor and lays eggs along with a deposit of fungal spores. As the larvae grow, they depend on the fungus mycelium for food. Typically, a larva chews a 25–75-cm-long, C-shaped tunnel over a 2–3-year period. After pupating, the adult emerges through a large circular hole. Generally, the damage associated with these insects is minimal, but the size of the exit holes and the occasional disturbing appearance of a large and noisy adult indoors can cause concern (Morgan, 1968). These insects can be particularly abundant in lumber sawn from fire-killed timber.

Anthophoridae: The Anthophoridae or carpenter bees belong to the genus Xylocarpa and construct their nests in the wood. Carpenter bees resemble bumblebees in appearance and do not use wood as a food source. They excavate a series of 12-mm-diameter tunnels along the grain for 10–15 cm to deposit their eggs along with nectar and pollen to provide nourishment for the developing larvae.

Generally carpenter bees attack uncoated soft wood such as cedar or pine, but weathered wood of almost any species can be attacked. Since

they do not use the wood for nourishment, they are also able to attack wood treated with most waterborne wood preservatives. Carpenter bees will reinfest the same wood and can cause substantial wood damage if infestations go undetected for several years.

Formicidae: The wood-attacking carpenter ants, belonging to the genus *Camponotus*, are social insects that have queens, winged males, and workers of varying sizes in a given colony (Simeone, 1954; Furniss, 1944). Carpenter ants are often confused with termites. Carpenter ants have constrictions between individual body segments, and the winged adults have two pairs of different equal sized wings. Termites have two pairs of equal-sized wings, and their body segments are not constricted. Carpenter ants remove the wood to construct galleries to raise their young, but their food resources come from outside the nest. Termites, of course, also use wood as a food source.

Carpenter ants occur throughout the United States, but are most important in the Pacific Northwest and the Northeast, where lengthy infestations can result in considerable damage to houses. Ant infestations in the home are also a nuisance due to the large numbers of foraging insects. In the northeastern United States, carpenter ants commonly tunnel into the untreated heartwood zones of cedar and Douglas-fir transmission poles. Studies on their biology and control in utility poles have been reported by Hansen and Klotz (2005). Of the native carpenter ants, the black carpenter ant, *Camponotus pennsylvanicus* Degeer, has been studied most extensively in the eastern U.S. while the *C. novaboracensis* and *C. vicinus* have been more heavily studied in the Pacific Northwest (Mankowski and Morrell, 2000; Hansen and Klotz, 2005). Carpenter ants are scavengers, and common food sources include aphid secretions and insects. In structural infestations, carpenter ants often search our sugars, proteins and water. Winged reproductives emerge and swarm in the late spring and early summer. After mating, the males die and the females search for a suitable site. In general, females will search for moist wood or other materials. Simeone (1954) has shown that successful colonies of *C. pennsylvanicus* could only be established in wood above 15% moisture content, while Mankowski and Morrell (2000) showed that *C. modoc* Wheeler and *C. vicinus* Mayr were able to establish colonies in wood at much lower moisture levels. Although partially decayed wood in structures is selected often for nesting sites (Moore, 1979), there is no consistent association of colonies with decayed wood. Colonies develop slowly at first, but increase rapidly after the first year, ultimately approaching

2–3 thousand individuals when the winged reproductives are produced. Colonies are quite mobile and there is often a main nest site such as a log or stump, with several satellite nests in the surrounding area.

Wood damaged by carpenter ants has numerous clean tunnels primarily in the springwood (Fig. 2.2). Carpenter ants need not necessarily tunnel into only wood; they will also bore into other soft material. They are particularly fond of foam insulation, likely because of the ease of tunnel, but possibly also because it provides a more stable temperature environment. Generally, the nest can be detected by the presence of piles of frass and insect fragments below the entrances to the infested wood (Fig. 2.2). Carpenter ants do not cause significant wood strength losses unless the colony is left undisturbed for long periods. Infestations by carpenter ants can be limited by keeping the wood dry, using pressure-treated wood in high hazard areas, and eliminating wood debris from around structures (Furniss, 1944). Carpenter ant control is usually effected using regular application of barrier sprays designed to repel and, therefore, exclude workers from a house. Local extension agents should be contacted to determine insecticides currently recommended for ant management.

Marine borer damage

Marine borer is the collective term used for the many invertebrates that burrow into and damage wood exposed to ocean or brackish waters. The two major phyla involved are the Mollusca (mollusks) and Crustacea (crustaceans). These animals primarily chew and burrow into wood for protection although there is emerging evidence that some species also feed on wood. It has been estimated that marine borers cause losses in marine structures of approximately 500 million dollars annually but this figure is both old and a crude estimate of the potential economic impacts of these organisms (United States Navy, 1965). They were clearly more important in the era of wooden sailing vessels, but they remain important because of the volume of timber using in infrastructure such s piers and wharfs. It is also important to note that marine borers are important recyclers of wood in marine environments and their droppings help feed the deeper ocean community.

While marine borer damage to shipping has been a long-term problem, recorded historically as early as 350 BCE by Theophrastus

(Turner, 1959), very little is known about the biology of the organisms, and certain stages in the life cycles of these invertebrate animals are still poorly understood. As a result, our ability to develop effective prevention and control methods, based upon a thorough knowledge of vulnerable points in the marine borer life cycles, has lagged far behind the methods available for other pest problems. In general, we continue to depend on a limited number of highly toxic, broad spectrum-chemicals to protect wood in marine environments.

The wood-attacking marine borers are separated into 3 major groups (shipworms, pholads, and isopods that include both *Limnoria* and *Sphaeroma*) based upon anatomy, physiology, and the nature of wood attack (Morrell et al., 1984; Helsing, 1979). The shipworms and pholads are mollusks, while *Limnoria* and *Sphaeroma* are crustaceans.

Shipworms: The shipworms, or Teridinidae, are important in temperate waters where they begin life as microscopic, free-swimming larvae that filter feed for several hours to a few weeks before they settle on a wood surface (Quayle, 1959). Once settled, the larvae bore into the wood using a pair of tiny, chitinous shells located near the head and become trapped for the remainder of their lives. This inability to move was exploited by captains when they sailed their shipworm-infested wooden vessels into tidal "freshwater" rivers for several months to eliminate the shipworm infestations. Once shipworm larvae bore into the wood, they undergo rapid changes in morphology. The two shells near the head calcify and develop teeth, and the body begins to elongate into the worm-like adult. As the shipworm grows, it burrows further into the wood, depositing a fine layer of calcium carbonate on the surface of the tunnel. This layer is seen readily when the wood is x-rayed and can be used to detect shipworm infestations. Although shipworms grow to lengths of 0.3–1.5 m, the only external sign of infestation is a small hole (<3 mm in diameter) through which the animal exposes a pair of feather-like siphons that filter food and exchange oxygen and waste products with the surrounding water. At the slightest sign of danger, the shipworm withdraws the siphons and covers the hole with a hardened pallet, making surface detection extremely difficult. This pallet resists desiccation and allows shipworms to survive out of water for up to 10 days. Shipworms were formerly considered to only use food for shelter but emerging evidence indicates that they have a collection of symbiotic organisms in their gut that digest cellulose.

The two major shipworm genera are *Teredo* and *Bankia*; however, these genera have different salinity and temperature requirements and

generally do not occur together. For example, *Teredo navalis* is found in warmer waters, and a reduced salinity tolerance permits it to survive far upstream in many estuaries. This tolerance to low salinity resulted in losses totaling 25 million dollars in the San Francisco Bay area during a period of low stream flow in the mid-1920s and stimulated interest on marine borer control (Hill and Kofoid, 1927).

Bankia setacea is also found in a variety of West Coast temperate waters, but cannot tolerate low salinities. Unlike *T. navalis*, *B. setacea* adults are much larger, eventually reaching 1.5−1.8 m in length. Although they can cause tremendous destruction, they are short-lived with life cycles lasting only 1−2 years.

While adults of both species are destructive to essentially all species of untreated wood, their attack can be prevented by using wood, pressure treated with creosote or inorganic salts. Attack can also be prevented by encasing the submerged wood in plastic, metal, or concrete barriers (Steiger and Horeczko, 1982). In early sea-faring days, shipbuilders followed a similar practice by covering the hull with copper nails or sheathing.

Pholads: Pholads are wood-and rock-burrowing mollusks that, like shipworms, begin life as small, free-swimming larvae. Once these organisms settle on the wood, they burrow near the surface and remain entirely within their shells, which enlarge as they grow. Pholad damage weakens the wood surface to the point where waves can erode the damaged wood and expose new wood to attack. The most common wood-boring pholad, *Martesia striata*, grows to be 5−6.25 cm long by 2.5 cm wide and feeds through a hole 0.25 cm in diameter. Although they are small in size, heavy pholad attack can completely destroy untreated piling within one year. Pholads are a serious problem warmer waters such as those in Hawaii and portions of southern Florida. They are generally more prevalent in tropical waters. Pholad attack can be prevented by the use of barriers or wood pressure treated with creosote. Frequently a second marine borer that is resistant to creosote is associated with pholads. In these cases, the wood must be protected with both creosote and an inorganic arsenical in a process known as dual treatment.

Isopods: Unlike shipworms and pholads, the wood-boring isopods are small crustaceans that move freely about the wood surface. The *Limnoria* species, also known as gribbles, are by far the most destructive of these isopods and attack the wood surface from the mudline to the high tide line (Kalnins, 1976; Lane, 1959; Ray, 1959). Most of this attack is concentrated

in the more highly oxygenated tidal zone where the waves wear the weakened wood away to eventually produce hour-glass shaped piling. Gribbles chew small tunnels which penetrate only a short distance into the wood (<2.5 cm). The *Limnoria* swim freely in and out of the burrow, and the wood may be colonized by thousands of individuals.

Limnoria are small, averaging 3–6 mm in length, and their taxonomy is quite complex (Menzies, 1959). There are 20 species of *Limnoria*, but only *L. lignorum*, *L. quadripunctata*, and *L. tripunctata* are of importance along the coasta1 United States. The latter species is the most important due to its ability to attack creosoted wood in warm water ports (Kalnins, 1976). The ability of this species to tolerate high levels of creosote has long perplexed scientists. This phenomenon has been attributed to selective detoxification, which renders the creosote harmless or to the presence of symbiotic flora in the midgut of the animal that degrades creosote (Geyer, 1982; Ray, 1959). There is normally a dense microflora on the surface of submerged wood (Kohlmeyer and Kohlmeyer, 1979; Barghoorn and Linder, 1944; Boyle and Mitchell, 1984; Kirchman and Mitchell, 1983; Cundell and Mitchell, 1977; Meyers and Reynolds, 1957), and it appears to provide some nutrition to the *Limnoria*.

Limnoria attack can be prevented by the use of barriers or by pressure treating the wood with creosote where *L. tripunctata* resistance is absent or also with an inorganic arsenical where it is present (American Wood Protection Association, 2017).

In addition to wood attack by these three groups, a number of other minor wood-boring organisms attack wood in marine environments. *Sphaeroma terebrans* is a mobile, warm water crustacean that normally burrows in mangrove roots, but has a remarkable tolerance of inorganic arsenical salt-treated wood. A shipworm *Xylophaga* sp. has been found to attack wood at depths of up to 2000 m and has caused concern where wood supports are used at great depths. This species probably plays an important role in recycling nutrients into a generally barren environment.

While we know a great deal about the marine wood borers, there is still much more to be learned before we can develop control methods based on knowledge of the biology of the target organism. Such information will become increasingly important as environmental concerns are brought to bear on the broad-spectrum pesticides currently used to protect wood in marine environments. Reductions in pollution in marine waters in harbors will also favor increased borer damage and there is already evidence of that occurring in New York harbor.

Wood decays and discolorations caused by fungi

The decays and discolorations in wood caused by fungi, and to a lesser extent bacteria, are major sources of loss both in timber production and wood use. While termite, ant and beetle damage is more easily detecting, fungal decay accounts for far more damage. Understanding the nature of these agents and identifying prevention or control strategies are major purposes of this treatise.

Decays and discolorations differ sharply in cause and nature from the other wood deterioration agents discussed above. Microorganisms, the casual agents, are unique organisms that have evolved systems to penetrate, invade, externally digest, and absorb soluble constituents from complex substrates such as wood. Fungi and bacteria in ecosystems have important decomposer roles and the steady release of carbon dioxide and other elements are critical to plant photosynthesis and, thereby, continuing life. In this role, a limited number of specialized fungi are the major decomposers of wood.

Bacteria grow primarily on wood surfaces and are carried into wood by microfauna, fungi, and water menisci during wetting and drying. Bacterial damage to wood is often judged to be minor but they can have more substantial effects at localized zones of the wood. Bacteria cause localized wall etchings and tunnels or cavities in the cell walls. When wood is stored under water or kept wet, bacteria may destroy parenchyma cells and pit membranes causing substantial increases in wood permeability. These changes cause the logs to become water soaked and sink.

Molds are fungi that grow into extremely wet wood by utilizing available proteins, lipids and simple carbon compounds stored in the wood parenchyma. Fungal hyphae and spores on the wood surface produce colors such as black, gray, green, purples, and red; "mold-like" odors; and in some cases the huge numbers of associated spores present potential problems as allergens (Robbins and Morrell, 2017). Mold spores can normally be removed by brushing or planning, but their presence causes major wood quality losses and the hyphae can continue to attack the interior.

Stains are degrading discolorations caused by fungi that invade the sapwood of many commercial woods during log storage or lumber seasoning. The stain fungi primarily invade parenchymatous tissues in the sapwood, and the discolorations result from the masses of pigmented hyphae in the wood cells. Though staining fungi cause little damage to the prosenchyma

cells in wood, several properties in addition to color, such as toughness and permeability, may be adversely affected. Damage by stain fungi is much more difficult to remove.

Decays are the major type of damage to wood in use and are caused by a limited group of fungi that have evolved to utilize one or more the three primary polymers in wood. Decay essentially is the result of wood digestion by fungi. The slow, progressive digestion of the wood causes a continuum of changes in appearance and physical and chemical properties. Only a limited group of fungi possess the enzymatic capability of digesting wood. Various groups of fungi attack the wood cell wall constituents in different ways and sequences that result in several types of decay.

Soft rots are caused by microfungi that selectively attack the S-2 portion of the cell wall. High wood moisture contents and direct soil exposures seem to favor soft rot development. Brown rots are caused by a group of fungi that attack primarily the carbohydrates in the cell wall. White rots are caused by a group of fungi that attack all of the wood polymers in the cell wall. The white and brown fungi are primarily in the Class Basidiomycotina, while soft rot fungi are primarily Ascomycotina although there are exceptions in both cases. All decays, in the final stages, result in drastic changes in strength and other use properties or total destruction.

The decays and discolorations in wood, briefly described above, are the central theme of this textbook and are discussed in detail in subsequent chapters. The characteristic appearances of most decays and discolorations and the presence of characteristic hyphae or microscopic features in the wood permit their separation from all other types of wood deterioration

Summary

The major types of wood damage, the causal agents, and descriptions are summarized in Table 2.4.

1. Fungi and insects are the major biotic agents responsible for wood destruction in use and recycling in terrestrial ecosystems. Fungi cause white, brown, or soft rot degradation that result in the loss or significant reduction in many wood use properties. Fungi also cause surface

Table 2.4 The major types of wood damage and their descriptions.

Type of damage	Causal agent(s)	General descriptions	Prevention or control
Weathering	Ultraviolet light, oxidation, swelling and shrinkage, leaching, and fungi	Unprotected surfaces develop a gray color and roughened texture.	Ultraviolet light resistant coatings.
Thermal decomposition	High temperature	<200 °C uniform surface brittleness. >200 °C charcoal in absence of oxygen; combustion around 275 °C.	Fire retardant chemicals.
Chemical decomposition	Caustic chemicals	With acids wood turns brown, chars, and becomes brittle; with bases wood bleaches and defibrillates.	Chemically resistant woods.
Mechanical damage	Mechanical forces rupturing surface tissues	Selective surface erosion in heavy friction zones.	High specific gravity woods, edge grain, or chemically hardened woods.
Insect damage	Termites	Localized honeycomb cavities, wood soiled and filled with frass.	Insecticides or keep wood dry.
	Borers	Tunnels, cavities, pinholes.	
	Ants	Localized honeycomb cavities wood channels clean.	
Marine borer damage	Shipworm	Interior tunnels with lime-coated walls.	Protective surface barriers or use wood preservatives.
	Pholads	Large interior tunnels—near surface.	
	Bribble	Surface tunneling in tidal zone.	
Decay	Fungi	White fibrous pockets or punky texture. Brown fibrous	Keep wood dry or use wood preservatives.

(*Continued*)

Table 2.4 (Continued)

Type of damage	Causal agent(s)	General descriptions	Prevention or control
		pockets or cubical checking patters. Soft surface embrittlement and exfoliation in small fragments.	
Molds	Fungil	Colored spores or mycelium on the wood surface.	Dry wood or use protective chemicals.
Stains	Fungi	Sapwood discolored gray, black, brown, blue and intensified in ray parenchyma.	Dry wood or use protective chemicals.
Bay cell and cell wall damage	Bacteria	Soft surfaces, ray cells destroyed, microscopic tunnels in cell walls.	Keep wood dry of use wood preservatives.

molding and sapwood discolorations that develop during seasoning or when the wood becomes too wet.

2. Termites, beetles, and hymenopterans are the principal types of insects that damage wood in use. Termites chew the wood, forming large cavities for nests that have a honeycomb pattern. Subterranean termites build their nests in soil and require wood at high moisture contents. They are the most economically important insects in the United States. Beetles and ants chew tunnels, channels, or cavities in wood and many attack wood in the tree, stored log, or when in the green condition.
3. Insect and fungal damage in wood are often associated because they have similar environmental requisites or a particularly fungus is required for development of an insect. Two basic prevention or control measures for both agents in many wood uses are to keep the wood dry or to treat the wood with protective chemicals.
4. Marine borers cause great damage to unprotected wood used in marine environments. Shipworms, pholads, and *Limnoria* are the major marine degradation agents. They may riddle the wood interior with tunnels or chew the surface in the intertidal zone. Principal prevention methods are installation of physical barriers around the wood or the use of wood treated with protective chemicals.

5. Combustion and weathering are the principal abiotic types of wood destruction. Fire retardants and structural design can reduce the fire hazard in many wood uses. Weathering is the surface destruction of wood by the combined action of ultraviolet radiation, oxidation, leaching, and mechanical forces. This process results in severe aesthetic losses in some wood uses and can be controlled by maintaining protective coatings on the exposed surfaces.
6. Caustic chemicals and mechanical forces degrade and damage wood in some special uses or situations, and their presence must be considered when wood is used in industrial applications.

References

Amburgey, T.L., 1979. Review and checklist of the literature on interactions between wood-inhabiting fungi and subterranean termites. 1960–1978. Sociobiology 4 (2), 279–296.

American Wood Protection Association, 2017. Book of Standards. American Wood Protection Association, Birmingham, Alabama.

Baker, A.J., 1974. Degradation of Wood by Products of Metal Corrosion. USDA Forest Service, FPL Res. Paper No. FPL 229.

Baker, W.L., 1972. Eastern Forest Insects. USDA Miscellaneous Publication 1175, Washington, DC, 462 p.

Barghoorn, E.S., Linder, D.H., 1944. Marine fungi: their taxonomy and biology. Farlowia 1, 395–467.

Barnes, H.M., Winandy, J.E., 1986. Effect of seasoning and preservative on properties of wood. Proc. Am. Wood Preserv. Assoc. 82, 95–105.

Barton, G.M., 1982. A study of wood deterioration in two British Columbia Kraft pulp mills. Chapt. 10. In: Meyer, R.W., Kellogg, R.M. (Eds.), Structural Uses of Wood in Adverse Environments. Soc. of Wood Science and Tech., Van Nostrand Reinhold Co., NY, pp. 142–148. ill.

Beal, R.H., Maulin, J.K., Jones, S.C., 1983. Subterranean Termites: Their Prevention and Control in Buildings. USDA Forest Service Home and Garden Bull. 64, Washington, DC.

Behr, L.A., Behr, C.T., Wilson, L.F., 1972. Influence of wood hardness on feeding by the eastern subterranean termite, *Reticulitermes flavipes* (Isoptera: Rhinotermidae). Ann. Ent. Soc. Am. 65, 457–460.

Boyle, P.J., Mitchell, R., 1984. The microbial ecology of crustacean wood borers. In: Costlow, J.D., Tipper, R.C. (Eds.), Marine Biodeterioration: An Interdisciplinary Study. Naval Institute Press, Annapolis, MD, pp. 17–23.

Browne, F.L., 1960. Wood siding left to weather naturally. S. Lumberm. 201 (2513), 141–143.

Christian, M.B., 1941. Biology of the powder-post beetle *Lyctus planicollus* Le Conte and *Lyctus parallelopipedus* (Melsh). Part II. La. Conserv. Rev 10 (1), 40–42.

Coulson, R.N., Lund, A.E., 1973. The degradation of wood by insects. In: Nicholas, D. O. (Ed.), Wood Deterioration and its Prevention by Preservative Treatment. Syracuse University Press, Syracuse, NY.

Cundell, A.M., Mitchell, R., 1977. Microbial succession on a wooden surface exposed to the sea. Int. Biodeterior. Bull. 13, 67–73.

DeGroot, R.C., 1982. Alternatives to termiticides in building protection. Termiticides in Building Protection. National Institute of Building Sciences, Washington, DC, pp. 91–94.
Ebeling, W., 1968. Termites: Identification, Biology, and Control of Termites Attacking Buildings. California Agric. Experiment Stat. Extension Service, Manual 38.
Erickson, H.D., Reese, L.W., 1940. The effect of several chemicals on the swelling and crushing strength of wood. J. Agric. Res. 60, 593–603.
Esenther, G.R., 1969. Termites in wisconsin. Ann. Entomol. Soc. Am. 62 (6), 1274–1284.
Esenther, G.R., Allen, T.C., Casida, J.E., Shenfelt, R.D., 1961. Termite attractant from fungus-infected wood. Science 134, 50.
Feist, W.C., 1977. Finishing wood for exterior applications—paints, stains, and pretreatments. Chapt. 19. In: Ed, Goldstein, I.S. (Ed.), Wood Technology: Chemical Aspects. American Chern. Soc., Washington, DC, pp. 294–300.
Feist, W.C., 1982. Weathering of wood in structural uses. Chapt. 12. In: Meyer, R.W., Kellog, R.M. (Eds.), Structural Uses of Wood in Adverse Environments. Van Nostrand Reinhold Co, NY, pp. 156–176.
Feist, W.C., Ellis, W.O., 1978. Fixation of hexavalent chromium on wood surfaces. Wood Sci. 11 (2), 76–81.
Fisher, R.C., Thompson, G.H., Webb, W.E., 1954. Ambrosia beetles in forest and sawmill: their biology, economic importance, and control. For. Abstr. 15 (1), 3–15.
Furniss, R.L., 1944. Carpenter Ant Control in Oregon. Oregon Agric. Experiment Stat. Circular 158, Corvallis, OR, 12 p.
Furniss, R.L., Carolina, V.M., 1977. Western Forest Insects. USDA Forest Service Miscellaneous Publication 1339, Washington, DC.
Geyer, H., 1982. The influence of wood-inhabiting marine fungi on food selection, feeding, and reproduction of *Limnoria tripunctata* Menzies (Crustacea: Isopoda). Int. J. Wood Preserv. 2 (2), 77–89.
Goldstein, I.S., 1973. Degradation and protection of wood from thermal attack. In: Nicholas, D.O. (Ed.), Wood Deterioration and its Prevention by Preservative Treatment. Vol. 1, Degradation and Protection of Wood. Syracuse University Press, Syracuse, NY, pp. 307–399.
Gray, D.R., Borden, J.H., 1985. Ambrosia beetle attack on logs before and after processing through a dryland sorting area. For. Chron. 61, 229–302.
Hon, D.N.-S., Ifju, G., Feist, W.C., 1980. Characteristics of free radicals in wood. Wood Fiber 12 (2), 121–130.
Hansen, L.D., Klotz, J.H., 2005. Carpenter Ants of the United States and Canada. Cornell University Press.
Helsing, G.G., 1979. Recognizing and Controlling Marine Borers. Bull. SG-49. Extension Marine Advisory Program. Oregon State University, Corvallis, OR.
Hill, C.L., Kofoid, C.A., 1927. Marine Borers and Their Relation to Marine Construction on the Pacific Coast. Final Report. San Francisco Bay Piling Committee. University of California Press, San Francisco, CA.
Jamison, G.M., 1937. Loss of weight of wood due to weathering. J. For. 35, 460–462.
Kalnins, M.A., 1976. Characterization of the attack on wood by marine borer *Limnoria tripunctata* (Menzies). Proc. Am. Wood Pres. Assoc. 72, 1–13.
Kirchman, D., Mitchell, R., 1983. Biochemical interactions between microorganisms and marine fouling invertebrates. In: Oxley, T.A., Barry, S. (Eds.), Biodeterioration, vol. 5. J. Wiley and Sons Ltd., NY.
Koenigs, J.W., 1974. Hydrogen peroxide and iron: a proposed system for decomposition of wood by brown rot Basidiomycetes. Wood Fiber 6 (1), 66–80.

Kohlmeyer, J., Kohlmeyer, E., 1979. Marine Mycology: The Higher Fungi. Academic Press, NY, 690 pp.
Krishna, K., Weesner, F.M. (Eds.), 1969. Biology of Termites. Academic Press, NY.
Kuhne, H., Leukens, U., Sell, J., Walchi, O., 1972. Outdoor weathering tests on wood and exterior wood finishes. Eidgenossische Materialprufungs-Und Versuchsanstalt (EMPA) Dubendorf, Ber. 198, 1–51.
Lane, C.L., 1959. The general histology and nutrition of *Limnoria*. In: Ray, D.L. (Ed.), Marine Boring and Fouling Organisms. University of Washington Press, Seattle, WA.
LeVan, S.L., 1984. Chemistry of fire retardancy. In: Rowell, R. (Ed.), Chemistry of Solid Wood, Advances in Chemistry Series 307. American Chemical Society, Washington D.C.
Lyons, J.W., 1970. The Chemistry and Uses of Fire Retardants. John Wiley and Sons, NY.
MacLean, J.D., 1951. Rate of disintegration of wood under different heating conditions. Proc. Amer. Wood Pres. Assoc. 47, 155–168.
MacLean, J.D., 1954. Effect of heating in water on the strength properties of wood. Proc. Am. Wood Preserv. Assoc. 50, 253–280.
Marion, J.E., Wissing, A., 1960. The chemical and mechanical deterioration of wood in contact with iron. Part IV. Prevention of deterioration. Svensk. Papperstid 63, 174–183.
Mclean, J.A., 1985. Ambrosia beetles: a multi-million dollar degrade problem of sawlogs in coastal British Columbia. For. Chron. 61 (4), 295–298.
Menzies, R.J., 1959. The identification and distribution of the species of *Limnoria*. In: Ray, D.L. (Ed.), Marine Boring and Fouling Organisms. University of Washington Press, Seattle, WA.
Meyers, S.P., Reynolds, O.S., 1957. Incidence of marine fungi in relation to wood borers attack. Science 126, 969.
Minuitti, V.P., 1967. Microscopic Observations of Ultraviolet Irradiated and Weather Softwood Surfaces and Clear Coatings. USDA, for. Sere Res. Paper FPL 74. For. Prod. Lab, Madison, WI.
Moore, H.B., 1979. Wood-inhabiting Insects in Houses: Their Identification, Biology, Prevention, and Control. U.S. Department of Housing and Urban Development, Washington, DC.
Moore, W.S., Koehler, C.S., 1980. Powder-Post Beetles and Their Control. Leaflet 21017. Division of Agric. Sciences, University of California, Berkeley, CA, 7 p.
Morgan, F.D., 1968. Bionomics of siricidae. Ann. Rev. Entomol. 13, 239–256.
Morrell, J.J., Helsing, G.G., Graham, R.D., 1984. Marine Wood Maintenance Manual: A Guide for Proper Use of Douglas-fir in Marine Exposures. Research Bull. 48. Forest Res. Lab., Oregon State University, Corvallis, OR.
Quayle, D.B., 1959. The early development of *Bankia setacea* Tryon. In: Ray, D.L. (Ed.), Marine Boring and Fouling Organisms. University of Washington Press, Seattle, WA.
Ray, D.L., 1959. Nutritional physiology of *Limnoria*. In: Ray, D.L. (Ed.), Marine Boring and Fouling Organisms. University of Washington Press, Seattle, WA.
Ross, J.D., 1956. Chemical resistance of western woods. For. Prod. J. 6 (6), 34–37.
Shafizadeh, F., Chin, P.P.S., 1977. Thermal deterioration of wood. Chapt. 5. In: Goldstein, I.S. (Ed.), Wood Technology: Chemical Aspects. American Chem. Soc., Washington, DC, pp. 57–81.
Simeone, J.B., 1954. Carpenter Ants and Their Control. Bulletin 39. State University of New York College of Forestry, Syracuse, NY, 19 p.
Smith, C.A., 1980. Wood for corrosive environments. Timber Trade J. 312 (5404), 23, 31.

Snyder, T.E., 1969. Control of Non Subterranean Termites. USDA Farmers Bull. 2018, Washington, DC, 16 p.
Steiger, F., Horeczko, G., 1982. The protection of timber piling from marine borer attack by application of plastic barriers. Int. J. Wood Pres. 2 (3), 127–129.
Thompson, W.S., 1982. Adverse environments and related design considerations—chemical effects. Chapt. 8. In: Meyer, R.W., Kellogg, R.M. (Eds.), Structural Uses of Wood in Adverse Environments. Soc. of Wood Science and Tech., Van Nostrand Reinhold Co., NY, pp. 117–129.
Turner, R.D., 1959. The status of systematic work in the Teredinidae. In: Ray, D.L. (Ed.), Marine Boring and Fouling Organisms. University of Washington Press, Seattle, WA.
United States Navy, 1965. Marine Biology Operational Handbook: Inspection, Repair, and Preservation of Waterfront Structures. NAVDOCKS MO-311. Bureau of Yards and Docks, Washington, DC.
Wangaard, F.F., 1966. Resistance of wood to chemical degradation. For. Prod. J. 16 (2), 53–64.
White, R.H., Dietenberger, M.A., 2010. Fire safety. USDA Wood Handbook: Wood as an Engineering Material. USDA Forest Products Laboratory General Technical Report 112, Madison, WI, 463 p.
Williams, L.H., 1985. Integrated Protection Against Lyctid Beetle Infestations. Part I. The Basis for Developing Beetle Preventative Measures for Use by Hardwood Industries. USDA Southern Forest Experiment Stat., New Orleans, LA.

Further reading

Graham, R.D., Wilson, M.M., Oteng-Amoako, A., 1976. Wood-metal Corrosion: An Annotated Survey. Research Bulletin 21. Oregon State University, Forest Research Laboratory, Corvallis, OR, 34 p.

CHAPTER THREE

The characteristics and classification of fungi and bacteria

Contents

Wood Microbiology.
DOI: https://doi.org/10.1016/B978-0-12-819465-2.00003-6

Fungi and bacteria are the principal microorganisms that invade wood during its growth, processing, storage, or use, causing decay or other property changes.

An understanding of the decay process and its prevention or control depends, in part, on understanding the features and capabilities of these decay agents. This chapter emphasizes the unique nature of fungi and their relationship to the other major life forms. It reviews fungal structures, growth patterns, life cycles, reproductive modes, and variable features of fungi, placing emphasis on the wood-inhabitors. Classification systems are presented to facilitate the taxonomic placement and recognition of some of the major wood-inhabiting microorganisms. Since bacteria cause only minor damage to structural wood in most environments, they are only briefly covered.

Fungi in relation to other life forms

Prior to the invention of the microscope, all life forms were grouped into the Plant and Animal Kingdoms. The existence of small life forms was unknown. Visible fungal structures were considered to be excrescences of dying or dead plants. Late in the 17th century, the development of the microscope and startling reports of Anton van Leeuwenhoek on the "wee animalcules" opened a window to the hidden world of small life forms (the resolving limit of the unaided eye is approximately 0.1 mm). The study of natural materials revealed a microcosmos, teeming with prodigious numbers and diverse kinds of unicellular and other small life forms. The term microorganism was introduced and microbiology began. At first, it was believed that the fungal "threads" observed in decayed wood and other small organisms seen in organic materials originated spontaneously or were again excrescences of dying or dead materials.

Nearly a century later, the causative role of microorganisms in disease and decay was established by Louis Pasteur, Robert Koch and others. It was soon recognized that many of the microorganisms involved in fermentation and decay were neither plants nor animals and required a new

category. The term Protista was proposed for the unicellular or small life forms with unspecialized tissues.

Until a few decades ago, the three Kingdom classification considered fungi (despite their lack of chlorophyll) to be simple plants because of the similarity of some groups to the algae. Fungi and algae were grouped in the Thallophyta (thallus plants - tissues not specialized into roots, stems or leaves) and separated by the heterotrophic nature of fungi and photosynthetic nature of algae.

The development of the electron microscope in the 1950, expanded the dimensions of the microscopic realm a thousand-fold and led to the startling discovery of two very different cell types - prokaryotic and eukaryotic. Prokaryotic cells are small, primitive bacteria, exhibit no mitosis or cytoplasmic streaming, and have no membrane bound organelles or an organized nucleus. Eukaryotic cells are more structurally complex. These cells divide by mitosis, exhibit cytoplasmic streaming, and have organized nuclei with double membranes, mitochondria, and plastids. As we have delved further into the origin of cells, we now know that some organelles such as the mitochondria have bacterial origins (Margulis, 1981). Additional evidence clearly established the distinct nature of fungi compared to plants, with differences in cell-wall composition, heterotrophy, the external mode of digestion, and the cytochrome C system (Lindenmayer, 1965). Based on these characteristics and others, Whittaker (1969) proposed a new classification grouping living organisms into five kingdoms and placed Fungi in a separate Kingdom. Organisms were divided into three domains: Archea (Archaebacteria), Eubacteria, and Eukaryote. Cavalier-Smith further segregated this classification into the Kingdoms: Plantae, Animalia, Protista, Fungi, Eubacteria, and Archaebacteria.

Eubacteria: prokaryotic cells (bacteria)

Archaebacteria: prokaryotic cells in extreme environments

Protista: Unicellular or closely related organisms with eukaryotic cells (i.e. protozoa along with single-celled and colonial algae)

Fungi: Filamentous eukaryotic cells, generally multicellular, heterotrophic, and with external digestion (i.e. Glomeromycota, Neocallimastigomycota, Blastocladiomycota, Microsporidia, Chytridiomycota, Ascomycota, Basidiomycota). These groups will be discussed in more detail later in this chapter.

Plantae: Walled eukaryotic cells, multicellular and highly differentiated, and autotrophic (photosynthetic) i.e. higher algae, liverworts, mosses, ferns, and seed plants

Animalia: Wall-less eukaryotic cells, multicellular and highly differentiated, heterotrophic, with ingestion and internal digestion (i.e. invertebrates, vertebrates)

It is important to note that these classifications are still very much in flux as new organisms are discovered, genomes are sorted, and we probe into the very nature of what constitutes life.

Fungi are eukaryotes, with those that we are concerned with generally composed of filamentous cells, often multicellular, heterotrophic, and functioning through external digestion. The fungi, as a higher life form, are speculated to have evolved from the Protista along with the separate animal and plant lines. Currently there are a diverse range of ideas on the groupings of organisms in the Protista and their evolutionary relationships to the other kingdoms.

Eubacteria are single-celled prokaryotes that lack membrane bound organelles and reproduce through mitosis. Although of less importance from the standpoint of structural wood degradation, they cause significant degradation of lignified woody residues in soils as they slowly digest woody biomass, typically after fungal systems have removed much of the carbohydrate fraction. Eubacteria (bacteria) thus have a very important niche role in carbon cycling of woody biomass, and they are generally accepted to be among the oldest organisms on the planet.

Bacteria

Bacteria are unicellular Prokaryotes, although in some forms such as the actinobacteria, chains of cells have a filamentous form. These organisms represent the oldest, simplest life forms, and consist of extremely small cells averaging only a few microns in length. Common cell shapes are round (cocci), cylindrical (rods), club-shaped (indeterminant rods), and helical (spirilla). The cell wall consists of *peptidoglycan* (a polymer of *N*-acetylglucosamine, *N*-acetylmuramic acid, and several amino acids). Bacteria reproduce by transverse binary fission, although budding occurs in one bacterial group. Under ideal growth conditions, cells may divide every 20 minutes and accumulate in large numbers on suitable substrates or surfaces such as the rhizosphere. Their prodigious reproductive rates allow them to rapidly occupy exposed surfaces.

Along with their tremendous reproductive potential, some bacteria are very resistant to environmental extremes. For example, temperatures of

121 °C for 15 minutes are required to sterilize media in the laboratory to kill some bacterial contaminants with some bacterial spores requiring even greater temperatures/times for elimination. Masses of bacteria appear as small viscous colonies in media cultures and may be confused with yeasts. Many bacteria appear to be adapted for growth on surfaces and rapidly exploit a wide range of energy sources. Some are motile, but most are not and require air, water, surface contacts or animal vectors for dispersal.

Wood-inhabiting bacteria are heterotrophs. Some, such as the gliding bacteria (*Cytophaga* spp.) are important cellulose decomposers, while others, including the cylindrical bacteria (rods), are associated with wood discoloration (*Clostridium* sp.) or invade and damage parenchyma cells and pit membranes during storage (*Bacillus polymyxa*). Many bacteria are associated with wood-degrading fungi, but their exact roles are uncertain. Some bacteria are *lithotrophs* obtaining energy by oxidation of various inorganic compounds such as iron or sulfur (see reducing bacteria in Chapter 6). A few bacteria are photosynthetic and utilize H_2S as an electron source. Many bacteria that aid in the digestion of wood in the environment have formed symbiotic relationships with animals such as termites and shipworms, and these relationships are touched on later in this chapter.

Bacteria are ubiquitous and estimates for the total number of species have been estimated to be several billion. Many species have never been isolated; we know them because we can sequence their DNA, while others are commonly isolated on a variety of materials. The roles of bacteria are still being determined, but we are beginning to understand that they play important positive and negative roles in animal, plant and even fungal health. Many bacteria are human, animal, or plant pathogens and important in medicine, agriculture, or the chemical industry. Although bacteria are ubiquitous in wood, and we know that they can be quite destructive over long periods of time in certain environments, there is still much more we need to understand about their roles in wood degradation processes as both free-living and symbiotic organisms.

Fungi

Fungi play three major roles in the ecosystem. Some fungi are pathogens and attack living plants or animals to cause diseases. Other fungi are mutualistic *symbionts* and have developed beneficial associations with

other organisms (e.g. mycorrhiza, lichens). Most fungi are *saprobes* and the principal agents in terrestrial ecosystems that decay plant debris to release carbon dioxide that sustains photosynthesis in green plants. It is in this latter role that fungi are the major decay agents of wood.

Most fungi that attack wood are multicellular eukaryotes. All are heterotrophs and utilize carbon compounds as an energy source. The fungal body (thallus) of wood-degrading fungi consists of a series of small, interconnected tube-like cells called *hyphae*. The key to understanding the unique fungal *"hyphal system"* is to remember that it has adapted to penetrate, externally digest, absorb, and metabolize a wide range of organic materials (e.g. plant materials, wood). Masses of hyphae are termed collectively "*mycelium*." Fungi reproduce primarily by spores formed by fragmentation or abstriction of sections of specialized hyphae.

Macroscopic appearances of fungi

Most of the hyphal systems of fungi that cause disease or decay are internal to the host or substrate, and not readily visible to the unaided eye. At times, however, mycelial masses or structures are visible externally to the unaided eye and may be useful for decay or disease detection (Fig. 3.1). Web-like masses of hyphae known as "*mycelial fans*" often grow on the surface of decaying wood in the ground or in environments with high humidity, and are a useful indicator of the presence of internal decay. *Rhizomorphs* are thick, cable-like masses of hyphae that can transport water, moisten wood, and facilitate its invasion. They are characteristic of several important building decay fungi. *Mold* is a general term describing the visual appearances of colored-spore masses that may grow on the surface of wet wood. The term *mildew* describes the appearance of black pigmented fungi that develop on surfaces such as painted wood in humid environments. Mold and mildew are often used interchangeably.

Some fungi form hardened cushions of mycelium known as stroma on plant or wood surfaces. Asexual or sexual fruiting structures may develop on or within the stroma. The most common visible form of many of the higher fungi is the *fruiting body or sporophore* bearing the sexual spores (e.g. mushrooms, bracket fungi, puffballs, perithecia, etc.). The form of these structures and the type and manner in which the spores are formed are important criteria used with one method for classifying fungi and will be discussed later in this chapter.

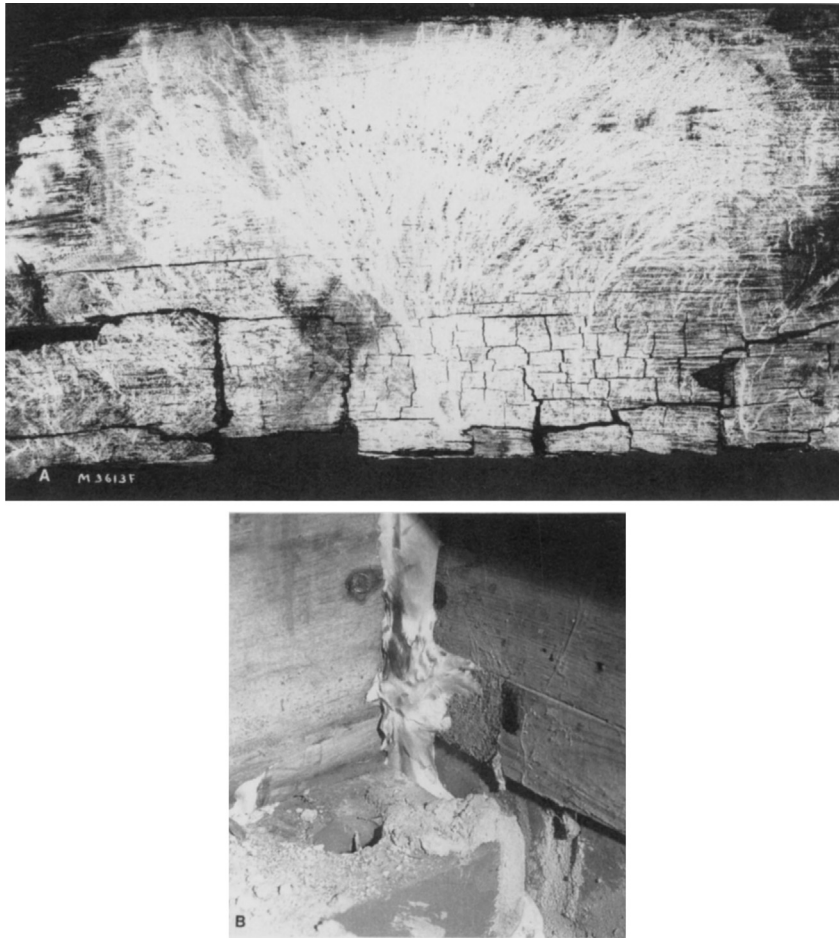


Figure 3.1 Common structures of wood-attacking fungi that are visible to the unaided eye include: (A) Mycelial fans of *Meruliporia incrassata* on surface of a decaying board. (B) Rhizomorphs of *M. incrassata* conducting water from a damp basement corner to the floor joist above, and Photos A and B courtesy of the U.S. Forest Products Laboratory, Madison, WI.

Estimates of the number of fungal species vary widely. Until recently, it was estimated that there were 350,000 species of fungi; however, recent studies using high throughput sequencing suggest that the number is much higher, perhaps approaching 1–5 million species. These numbers are certain to change as methods improve and our definitions of what constitute a species are refined.

Fungi range in size from unicellular groups such as the yeasts to some wood-decay fungi whose individual sporophore may weigh 45 kg. The fungal thallus of some root-inhabiting fungi can be massive and occupy several hectares of land.

Microscopic features of fungi

Hyphae are the basic cellular unit of filamentous fungal structures. Individual hyphae are small and, with few exceptions, can be seen only after considerable magnification. Individual hypha range from as small as 0.5 to up to 20 μm, with most ranging from 2 to 10 μm in diameter. Typical hyphal features in higher fungi seen with the ordinary light microscope include cell walls, cross walls or septa, vacuoles, various inclusions such as fat globules and crystals, and occasionally, nuclei. Most fungal nuclei are very small and special stains are often required for observation. Hyphal cells may be uninucleate or multinucleate, but many decay fungi have binucleate cells. This latter stage is referred to as the dikaryon stage if the nuclei are different genetically, arise from fusion of two hyphae and do not fuse. This nuclear condition is unique for fungi.

Septa or cross walls are present in the higher fungi (Ascomycota, Basidiomycota and fungi that have not yet been classified generally known as Deuteromycotina) that are the focus of this chapter. Septa or cross walls in the higher forms of fungi probably function as cell wall strengthening devices, but they may also function to maintain turgor pressure or as a protective mechanisms in the event that cells are damaged. Hyphal systems that are multinucleate are termed coenocytic.

Many septa are perforated, permitting organelles and some nuclei to migrate from cell to cell. Many Ascomycota have simple, one to several perforate septa. *Septa* with large swollen pore margins called *dolipore septa* characterize many members of the Basidiomycota (Fig. 3.2). These septa often have an associated amorphous material that appears to plug the pore. The dolipore septum allows nuclear migration in monokaryons. Dolipore septa can be visualized at higher magnifications (600 ×) and their presence can help to confirm that an isolate is a member of the Basidiomycota.

Hyphal growth occurs primarily by apical extension. Vacuoles usually develop a few cells behind the hyphal tip, in a zone known as the spitzenkörper in the higher fungi, and the resulting turgor pressure within

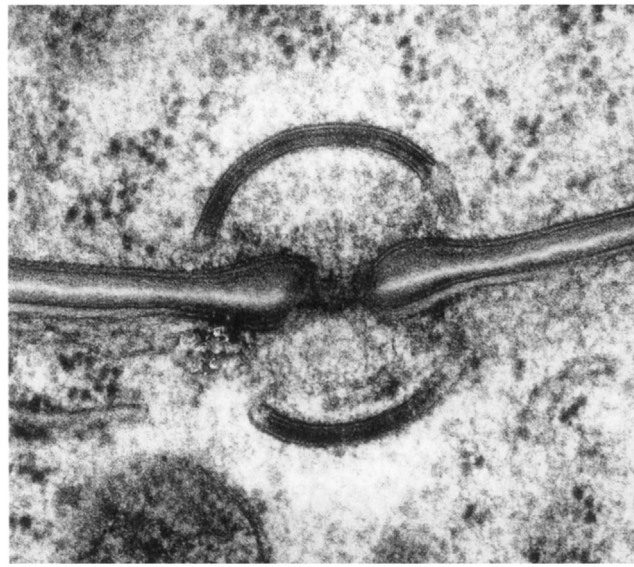


Figure 3.2 Transmission electron micrograph of a dolipore septum from the mycelium of *Auricularia auricula-judae* which is characteristic of many wood inhabiting Basidiomycota. X97,500. Prepared by freeze substitution and courtesy Lu and McLaughlin (1991) and reprinted by permission of the New York Botanical Garden. *From Lü, H. and McLaughlin, D.J. (1991). Ultrastructure of the septal pore apparatus and early septum initiation in Auricularia auricula-judae, Mycologia, 83 (3), 322–334.*

the cell protoplasts is the presumed driving force extending the plastic tip. Hyphal branching is common and usually begins early behind the developing hyphal tips. The fusion of adjacent hyphae of the same genotype by *anastomosis* is also common, resulting in a complex network of interconnected cells. Cytoplasmic streaming moves material steadily towards the tips, leaving empty or vacuolated hyphae behind. This process helps conserve scarce nutrients such as nitrogen and places these materials at the actively growing hyphae tip where they can be used for cell wall synthesis or enzyme production.

In some Basidiomycota, special hyphal structures called *clamp connections* develop in the dikaryotic hyphae during septa formation. In these cases, a branch develops near the forming septum in the hyphal apex. The two nuclei in the cell simultaneously divide mitotically and migrate separately in opposite directions along the hyphae. As the nuclei migrate, the branching point curves backward and anastomoses or fuses with cell wall just behind the developing septum. This results in the apical cell and the cell penultimate to it each containing two nuclei genetically identical to

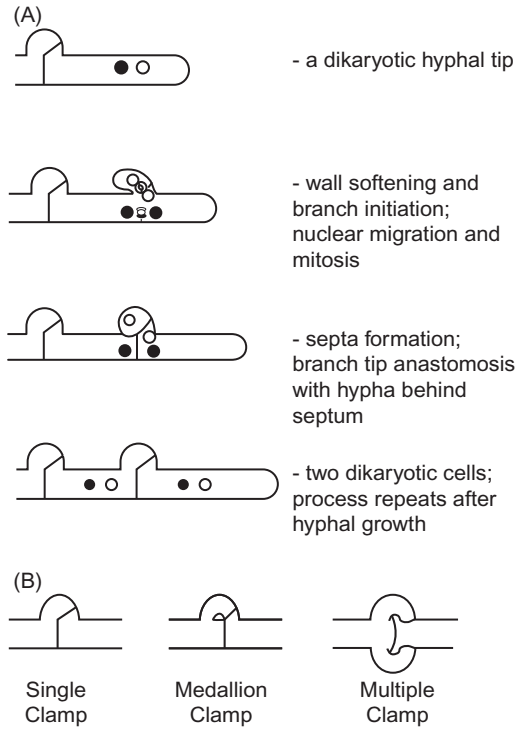


Figure 3.3 Hyphal structures known as clamp connections are present in many wood decay fungi in the Basidiomycota and traditionally were used as diagnostic microscopic features. (A) Development stages of clamp connections and (B) Examples of several types of clamp connections. Drawings courtesy of Dr. J. Worrall.

the original pair (Fig. 3.3). Clamp connections may be single or multiple and may occur at each septa, consistently, randomly or infrequently depending on the fungal species. Clamp presence, type, shape and size are important characters in cultural keys for the identifying of decay fungi (Fig. 3.3).

Hyphal wall structure

Fungi, like plants, possess firm cell walls that provide the rigidity needed for the formation of large fungal structures such as shelving sporophores or rhizomorphs as well as functions such as the forcible discharge of spores or penetration of plant cell walls. Hyphal walls also serve as important storage reserves for some fungi. Structurally, fungal cell walls consist of an inner network of microfibrils embedded in an amorphous matrix that also

forms the outer layers or often lamellae of the wall. Chemically, the walls consist of 80–90% polysaccharides with the remainder composed of proteins and lipids. Chitin, glucans, and in a few cases primarily in the higher fungi, chitosan forms the microfibrils that serve as the skeletal framework of the walls. Chitin is the principal skeletal material and is present in the inner walls of most septate fungi. The chitinous nature of the cell wall is one of the fundamental differences separating most fungi from plants, although some fungi have other polymers in their cell walls (Gow et al., 2016).

The roles of the proteins and lipids in the walls vary. Some may be cell-wall bound enzymes while others may function in structural integrity while still others may be important in recognition phenomena.

Fungal cell walls are surprisingly complex, both chemically and structurally. Photomicrographs of the various cell wall layers, after successive enzymatic treatments, were assembled by Becket et al. (1974) for the representatives of the major fungal groups. Bartnicki-Garcia (1968) suggested that differences in chemical composition of the cell wall were closely related to taxonomic position. The fungal cell wall is now viewed as a far more complex organelle that serves to protect the fungus, but also functions in surface recognition (Gow et al., 2016). The wall foundation consists of fibrous and gel-like carbohydrate polymers that act as a scaffold for proteins and other components. The cell wall is layered with the outer layer containing the greatest variety of components. Structural formulas for some common cell-wall constituents are presented in Fig. 3.4.

Fungal cell walls are readily modified by self-digestion (autolysis) and translocation of wall metabolites to growing hyphal apices. This activity may be an important means of nitrogen conservation. In addition, hyphal fusion, hyphal branching, clamp formation, some spore formation, and spore germination reflect situations where localized zones of the wall are enzymatically softened and disassembled.

Since the hyphae of most disease and decay causing fungi are internal to the host or substrate, it is often difficult to detect or determine the degree of invasion. Procedures have been developed to estimate the fungal biomass in decayed wood by acid hydrolysis of the decayed material and glucosamine detection and quantification (Swift, 1973). Chitin content can be determined directly from the glucosamine residues; however, chitin assays are difficult to perform and not entirely appropriate for quantifying fungal biomass for some species (Wu and Stahmann, 1975). When chitin assays are used, standards must be developed to relate the chitin

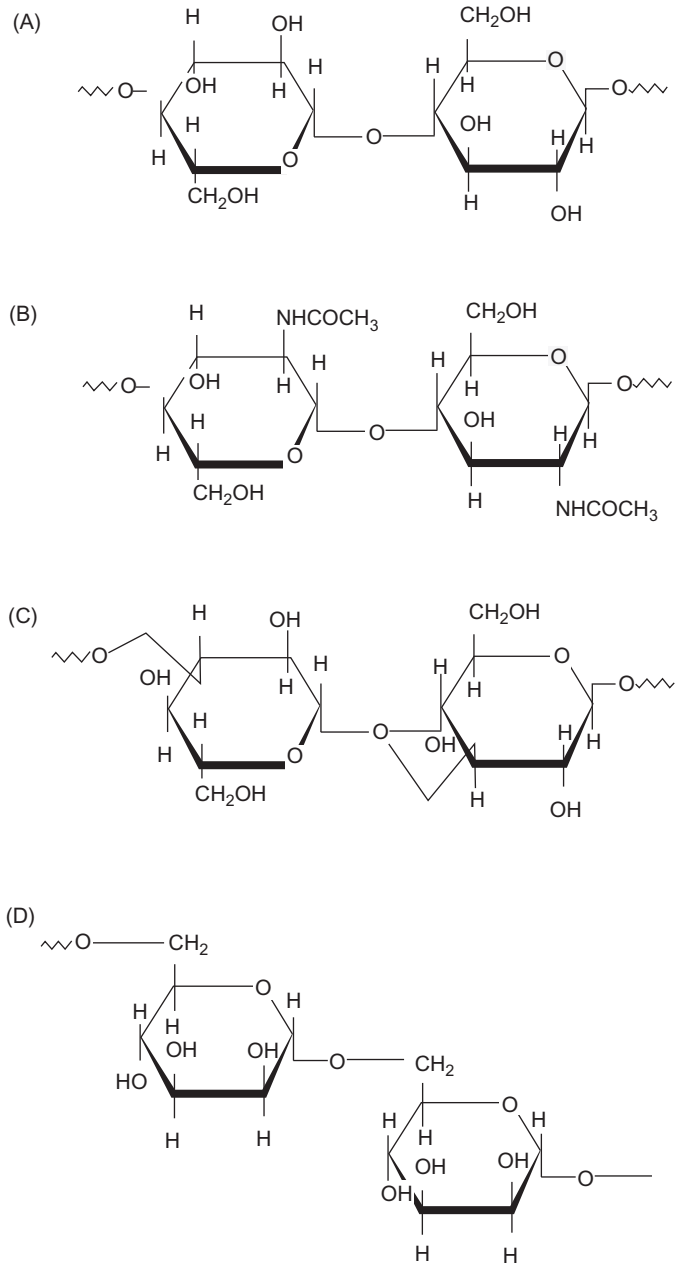


Figure 3.4 Chemical structures of portions of repeating units of several common polysaccharides found in fungal cell walls. (A) Cellulose, (B) Chitin (deacetylation yields chitosan), (C) Glucan, and (D) Mannan.

content to the biomass for the fungus tested. Chitin content of hyphae can vary substantially with the nutritional status of the fungus. Ergosterol is required for cell wall synthesis and has also been used to estimate the fungal massin a variety of substrates (Stahl and Parkin, 1996; Bjurman, 1994; De Saad et al., 2003; Montgomery et al., 2000; Parsi and Gorecki, 2006), but this requires developing background information on the substrate and the organisms present. As with chitin, care must be taken because ergosterol content can vary among fungal species and even with the condition of the fungus. Thus, using chitin or ergosterol as indirect measures of fungal biomass within a substrate can be difficult and prone to error.

Hyphae may also be visualized through various cell wall stains and plant-based lectins. These procedures make it easier to observe fungi in the wood, but it is still exceedingly difficult to accurately determine the volume of hyphae in the wood or the amount of damage they cause on a microscopic scale.

Fungal ultrastructure

The electron microscope, atomic force microscopy and newer high-resolution light microscopes and other techniques have revealed a wealth of information on the fine structure of both wood and fungal ultrastructure. The close relationship between structure and function has done much to clarify the major metabolic events occurring within cells. As eukaryotic cells, the types and roles of the organelles in fungal cells are similar to those of most plant and animal cells. A review of the major organelles and their function in fungal cells is needed as a background for understanding the metabolic events and mode of toxicant actions discussed in later chapters. A *cytoplasmic membrane*, consisting of lipoproteins lies within the cell wall and regulates the entrance and exit of materials from the cells. In some fungi, the cell membranes contain vesicular-like structures called paramural bodies (lomasomes) that they may aid in absorption, secretion, or cell wall modifications. These structures been heavily studied in the yeasts, but have received less attention in the wood inhabiting fungi (Douglas and Konopka, 2014; Riquelme, 2013; Pantazopoulous, 2016). An *endoplasmic reticulum (ER)* is present throughout the cytoplasm, and generally concentrated in regions of metabolic activity and cell wall extension. The ER has net-like connections with the other cell membranes (nuclear membrane, tonoplast, and cytoplasmic

membrane) and plays a key transport role among the organelles. *Ribosomes* are the centers of protein synthesis and appear in the cytoplasm particularly in regions of hyphal growth. Associations between ribosomes and the ER termed *rough ER*, which are common in many eukaryotes, are infrequent in fungi. *Microtubules* are present in the cytoplasm where they may be associated with protoplasmic streaming, and play a role in nuclear division. The Golgi apparatus plays a role in movement of materials and in apical extension of the hyphae. Mitochondria are visible under the light microscope using an oil-immersion objective and are associated with respiration and energy release. Time-lapse microscopy indicates that mitochondria can rapidly change shape and position within the cell. *Vacuoles*, bounded by a membrane called the *tonoplast*, are abundant in older hyphae and generally absent in the tips. They often originate in cells adjacent to the hyphal tips and increase in size in the older hyphal cells.

Vacuolization has been proposed as one method by which fungi isolate toxic compounds liberated during the digestion of wood. Nuclei are often very small (2–3 μm) and appear to change both size and shape rapidly. At times, and particularly during spore formation, the nuclear membrane appears to be continuous with the ER.

A dark region can be seen in the hyphal apex of some septate fungi under the light microscope called the Spitzenkörper. In electron micrographs, this dark region is an electron-dense zone closely associated with the hyphal tip and is present only during periods of active growth. This region is believed to contain clusters of transport vesicles involved in cell apex synthesis. *Crystals* and *globules* containing lipids, glycogen, or other reserve foods are also observed commonly in older hyphae. The ultrastructural features of the hyphal tip region of a wood-decay fungus are shown in Fig. 3.5.

Specialized hyphae

Specialized hyphae have developed for a variety of purposes in some fungal groups. *Appressoria* are flattened, enlarged hyphal tips that adhere to surfaces and facilitate the penetration of fine hyphal pegs through cell walls. They are formed by many pathogens and some wood-staining fungi. *Haustoria* are enlarged, convoluted hyphal cells adapted for absorption that contact the plasmalemma of host cells after wall penetration. They are present in the rusts, powdery mildews, and some mycorrhizae fungi. Some biotrophic fungi that parasitize other fungi invade their hosts

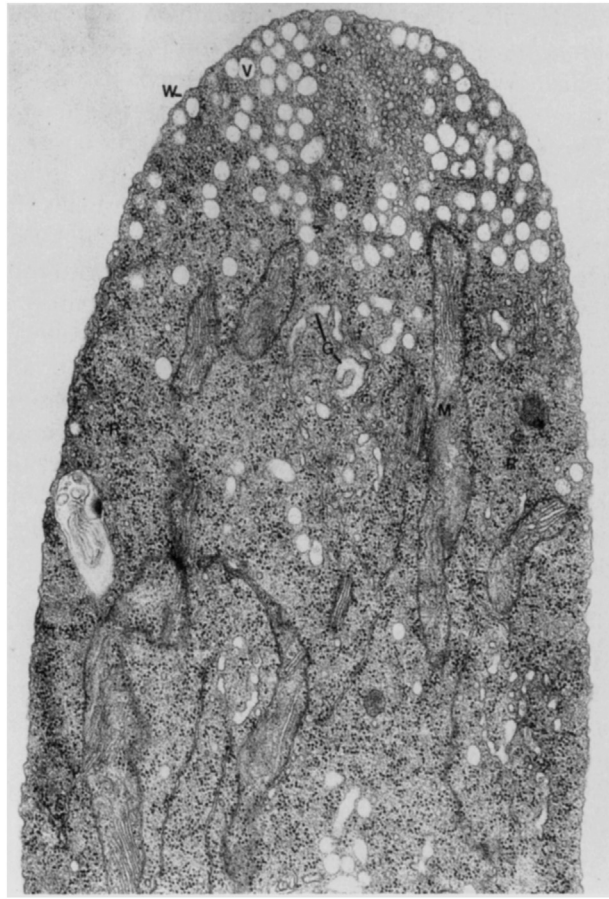


Figure 3.5 Electron micrograph of a hyphal tip of the fungus *Aspergillus niger*, showing the vesicles (V) clustered in the apical zone, ribosomes (R), mitochondria (M), smooth-surfaced Golgi cisternae (G), and the wall zone (W). X25000. *Courtesy of Dr. S. N. Grove and C.E. Bracker (1970) and permission of the Journal of Bacteriology.*

with haustoria-like hyphae. Unique cases of specialization include the hyphae that trap nematodes by the formation of sticky adhesion zones or capture them in rapidly constricting rings. *Chlamydospores* are specialized cells that develop thick walls and are able to withstand long periods of adverse environmental conditions. Some hyphae contain *melanin*, a dark pigment that protects hyphae growing on surfaces (paint mildew) from the ultraviolet light. Melanized hyphae are also present on the surface of some rhizomorphs and fungal propagules such as microsclerotia and are believed to play a protective function. *Spores* are the reproductive units of

fungi. They are specialized hyphae and because of their importance, are discussed later in a special section.

The large reproductive structures of some Ascomycota and Basidiomycota also contain specialized structural hyphae. Aggregations of tightly packed thin-walled hyphae resembling the parenchyma cells of higher plants, *called pseudoparenchyma hyphae*, occur in the soft tissues of some ascomata or fleshy sporophores. These hyphae are termed *sphaerocysts* when present in the gill or trama tissue of a sporophore and are circular in shape, and useful for identification. Three types of hyphae may form in the larger sporophores of the decay fungi. The thin sporophore eventually forms the hymenial layers where spores are produced. Leathery sporophores also contain thick-walled, many branched cells termed *binding hyphae* that intermesh with the *generative hyphae*. Large woody sporophores such as *Gandoderma applanatum* also contain skeletal hyphae that are elongate, thick-walled and rarely branched or septate. *Skeletal hyphae* resemble wood fibers and provide the hard, woody characteristics of some of the large, shelving sporophores. The three types of structural hyphae in sporophores were used as key criteria used in taxonomic revisions of the wood-decaying Basidiomycota based upon fruiting structures. Generative hyphae are present in all sporophores, while the fruiting bodies of some species may contain binding or skeletal hyphae or both. This topic will be discussed further in the section on classification. There have been dramatic changes in fungal identification, which has evolved from sporophore identification to cultural identification to more sophisticated sequencing of DNA. We will review the various methods to provide historical context and because they still appear in the older literature.

Cultural characteristics

Mycelial mats are produced in pure cultures on a culture medium but also are observed in nature on surfaces that are exposed to high moisture/humidity conditions. Many fungi, other than the obligate parasites, can be grown in the laboratory under axenic culture conditions on various natural or synthetic media. Media often contain a nutrient source mixed with agar, a substance obtained from algae. The agar provides a semi-solid substrate on which the fungus can grow. Fungi can also be grown in liquid cultures. The mycelium of fungi developing on a medium is often a mixture of the vegetative and reproductive hyphae. Some fungi, such as *Schizophyllum commune*, can develop typical sporophores after a few weeks

in culture. The growth characteristics (macroscopic and microscopic) of the mycelial mats are often distinctive and traditionally were useful in the identification of the fungus. We are coming to understand that many other fungi cannot be cultured on synthetic media, although their DNA can be detected in the wood.

DNA sequencing methods

Our understanding of fungal colonization of various wood substrates has changed dramatically as a result of the ability to sequence and identify the DNA present in a sample, although it has raised almost as many questions as it has answered as researchers attempt to determine the roles of these non-culturable fungi. The development of the polymerase chain reaction (PCR) and the identification of highly conserved sections of fungal r-DNA, known as Internal transcribed spacer (ITS) 1 or 4, that could be used as a template to amplify fungal DNA represented a major breakthrough in the ability to determine the organisms present in a system. In this process, DNA is extracted from an organism and purified. The DNA is denatured to separate the individual strands then this mixture is exposed to reactants that result in the production of numerous copies of the parent DNA. This material is then sequenced and the resulting sequences can be compared with previously identified sequences of known fungi. This allows identification of known fungi without the need for classic mycological methods. This approach still has its drawbacks because it requires the isolation of a single fungus, but the more recent development of high throughput sequencing techniques where all the DNA of a given group is sequenced and then identified has opened new horizons concerning the organisms present in a given substrate.

Classic fungal identification by culturing

The fruiting bodies of decay fungi are frequently absent, ephemeral, or difficult to detect, making it difficult to identify the causal agent. In these cases, the cultural characteristics of fungi isolated from the decayed material may be used for identification. Some useful characteristics employed in cultural identification manuals include mat color, texture, rate of growth, and microscopic features such as the presence of clamp connections, the types and features of specialized hyphae, and the types of asexual spores. Examples of some unique microscopic features of several important wood-decaying fungi are shown in Fig. 3.6. Oxidase reactions (the

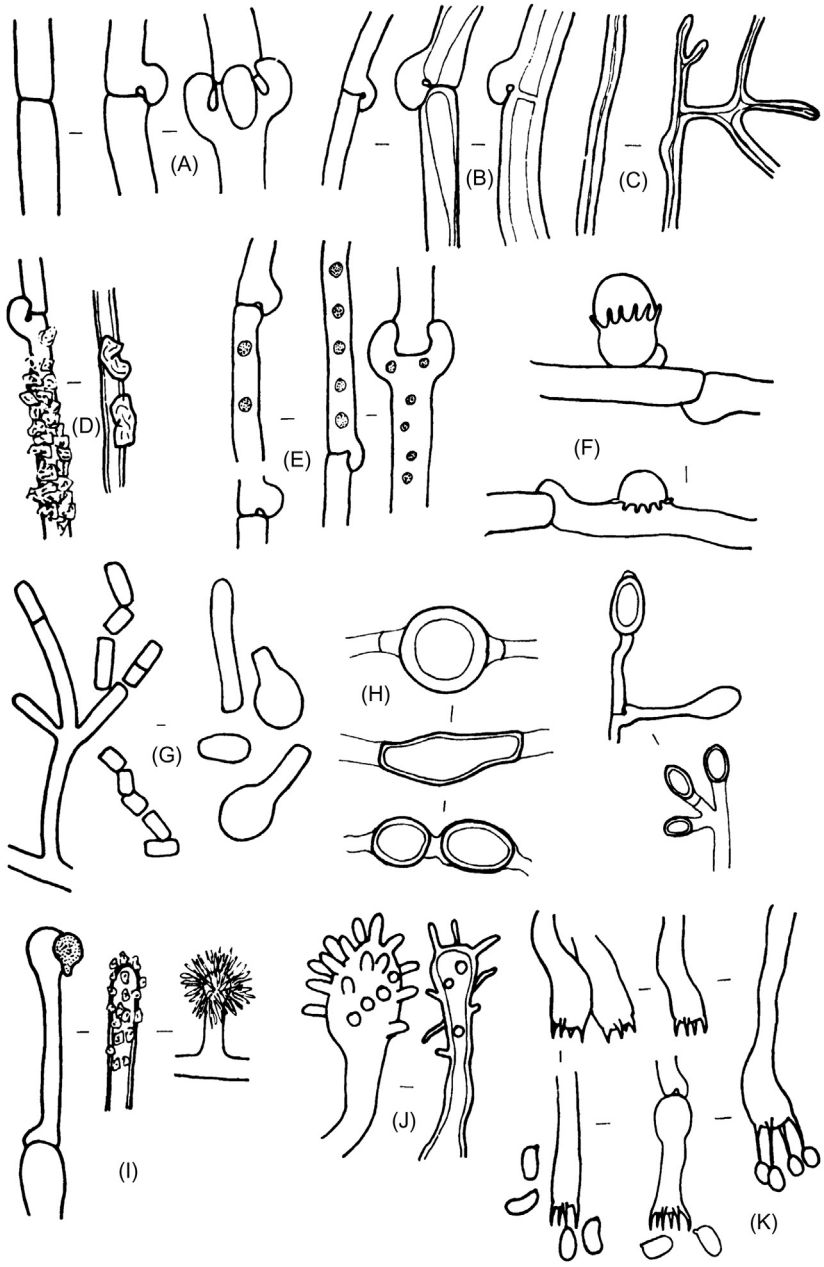


Figure 3.6 Examples of macroscopic features of Basidiomycota hyphae useful in cultural identification. (A) Variation in hyphal septation, (B) Variation in cell wall thickness, (C) Fiber hyphae, (D) Encrusted hyphae, (E) Variation in nuclear condition, (F) Stephanocysts (*Hyphoderma* spp.), (G) Arthroconidia (*Bjerkandra adusta* (left), *Gloeophylym sepiarium* (right)), (H) Chlamydospores (intercalary (left), terminal (right)), (I) Cystidia, (J) Acanthophyses (acanthohyphidia), (K). Variations in basidia: *Hyphodontia setulosa, Phlebia brevispora, Antrodia vaillantii* (top) and *Phlebia rufa, Sistotrema brinkmanii* complex (bottom). All X100. *Reproduced by permission of American Type Culture Collection and courtesy of Francis Lombard and Dr. George Chamuris.*

oxidation and discoloration of gallic and tannic acid media) and growth patterns of fungi on these two media are simple, important tests for separating most brown rot fungi (no oxidase reaction) from the white rot fungi (oxidase positive-discoloration of the media). Although isolation and sequencing of fungal DNA is now far more common for identification, a number of useful identification manuals based on fungal morphology are also available for various hosts, wood products, or fungal groups; they provide keys and cultural descriptions for many of the major wood-decay fungi (Davidson et al., 1942; Nobles, 1965; Stalpers, 1978; Wang, 1965; Wang and Zabel, 1990).

Fungal reproduction

Fungi reproduce by unicellular or multicellular spores that are formed from hyphae. They can be of asexual or sexual origin. Most spores provide a means of dissemination, but others are designed for surviving unfavorable conditions, and a few may function sexually as gametes. Many spores are very small and readily transported long distances by air currents, while others have shapes that allow them to attach to insects or float in water. Spores of some fungal groups are forcibly ejected into the air, while others are sticky and appear to be adapted for dissemination by insect vectors. Asexual spores are formed directly from a hypha without a meiotic division. Sexual spores are formed from hyphal cells where nuclear fusion (karyogamy) and meiosis have preceded the separation into spores.

Asexual spores: There are many types of asexual spores formed by fungi. This review will consider primarily the common types found in wood-inhabiting fungi. *Conidium* is the general term used for most asexual spores formed by members of the Ascomycota and Basidiomycota. Conidia have several very different modes of origin which are important in many classic taxonomic keys.

Arthrospores or *oidia* are conidia formed by the fragmentation of an existing hypha by separation of the septal walls. This type of conidial origin is termed thallic. Some wood decay fungi (*Gloeophyllum sepiarium*, *Phlebiopsis gigantea*, and *Bjerkandera adusta*) form abundant arthrospores in culture. In some fungi, such as the yeasts and the wood staining fungus *Aureobasidium pullulans*, the cell wall softens in a localized zone, balloons out, and forms *blastospores* by a budding process. This type of conidial origin is termed *blastic* and the conidiogenous cell enlarges generally

prior to septal formation and conidium release. There are several types of blastic conidial development. When the outer and inner walls of the conidiogenous cell are involved, conidia formation is called *holoblastic*. When only the inner wall is involved or a new wall is formed, the process is termed *enteroblastic*. Conidia formed by some wood-staining fungi (*Ophiostoma coerulescens*) represent enteroblastic development where spores formed inside an open-ended hyphae are ejected into the air when mature. Evolving concepts about the roles of sexual and asexual spores have markedly altered how these stages are addressed. With the advent of rapid molecular techniques for taxonomic classification, many fungi that had no readily identifiable sexual stage (called the Fungi Imperfectii or Deuteromycetes) are now classified as Ascomycota or Basidiomycota. Continued molecular investigations will ultimately shed further light on the relationships between these asexual stages and other fungal groups.

Sexual spores: Sexual spores result from the fusion of two haploid nuclei in a hyphal cell and subsequent meiosis. Sexual reproduction provides fungal progeny the great biological advantage of genotypic diversity as a result of crossing over or mixing of chromosome pair portions during meiosis. Genotypic diversity permits high variability for adaptive selections that can aid survival during changing conditions. In many groups of fungi, sexual spores are borne in highly specialized hyphal aggregates called fruiting bodies. Some typical examples of fruiting bodies are puffballs, mushrooms, perithecia, and the conks of wood-destroying fungi. Sexual spores formed inside a specialized terminal hyphal cell, termed the ascus, are called ascospores. This type of sexual spore formation is endogenous and characterizes the subdivision of fungi known as *Ascomycota* (Fig. 3.7A). This class of fungi contains many of the important wood stainers as well as a number of white rot fungi. A fertile layer of asci on or in an ascoma (ascocarp) is termed the *hymenium*. There are several types of ascomata (Fig. 3.7B). A closed ascoma is called a *cleistothecium*. A similar ascomata with an opening (ostiole) is termed a *perithecium*. The *perithecium* may be a free structure or embedded in a stroma. Perithecia embedded in stroma are often termed *ascostroma*. An open or cup-like ascome structure bearing the hymenium is called an *apothecium*. There are two types of asci; bitunicate asci with a rigid outer and a flexible inner wall and unitunicate asci with a single wall. This feature and the nature of the ascomata are the major distinguishing characteristics of the major wood-inhabiting ascomycetes.

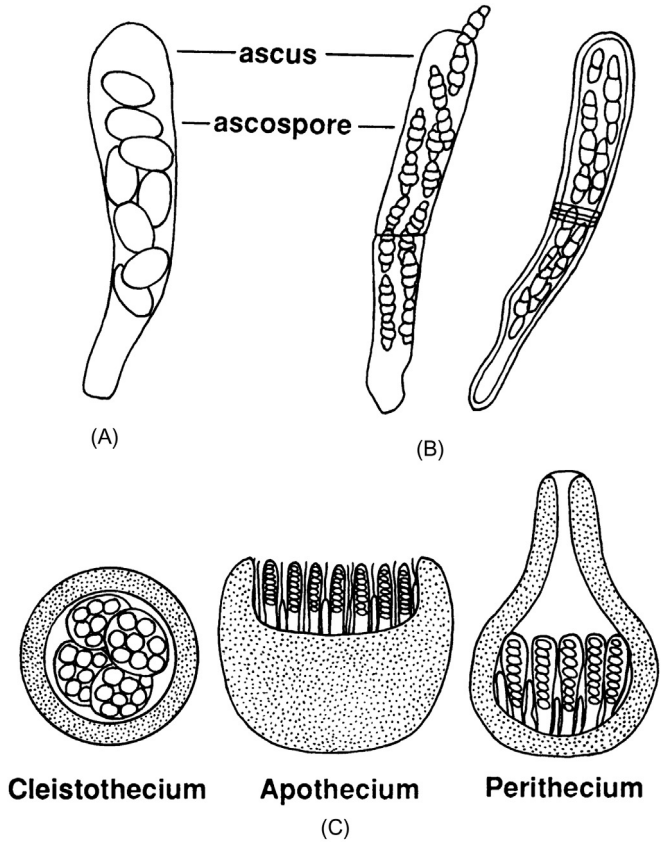


Figure 3.7 The sexual spore-bearing cells that characterize the Ascomycota. (A) A unitunicate ascus, (B) Several bitunicate asci- the left one is beginning to discharge ascospores, (C) Several types of ascomata. Sources: (A) Engler, A. and K. Prantl (1897) Die natürichen Pflanzenfamilien, Engelmann, Leipzig, (B) Luttrell, E.S. (1960), Mycologia 52, 64–69 with permission of the New York Botanical Garden, and (C). *Adapted from Figure 4.8, E. Moore-Landecker (1980), Fundamentals of Fungi with permission of Prentice-Hall, Inc. Drawings courtesy George Chamuris.*

Sexual spores formed externally on the tip of a swollen club-shaped hypha, termed a *basidium*, are called basidiospores and characterize the class of fungi known as the Basidiomycota (Fig. 3.8). This class of fungi contains most of the important wood-destroying fungi, and also contains the rusts and smuts, which are major plant pathogens worldwide. There are three sub-phylla: the Pucciniomycotina, the Ustilagomycotina and the Agaricomycotina. The former two sub-phylla represent the rusts and smuts, while the latter group contains most of the wood decay fungi.

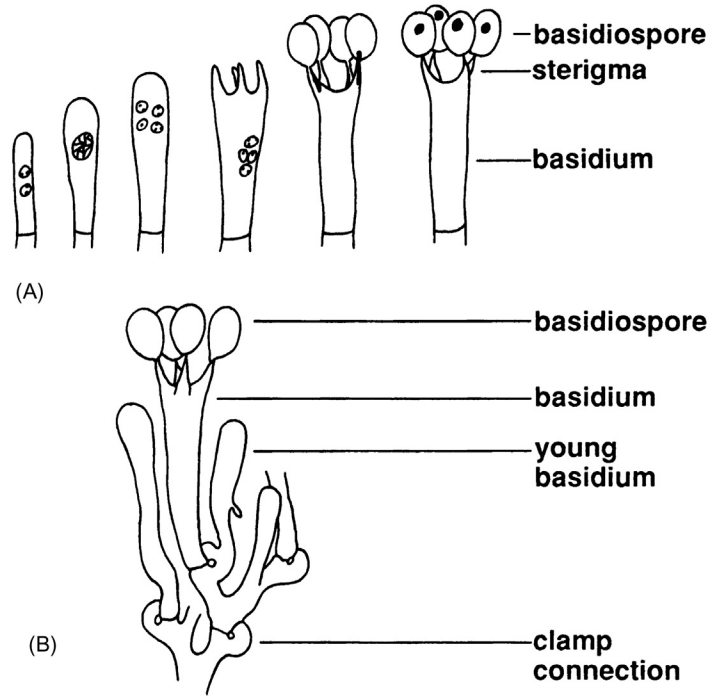


Figure 3.8 The developmental stages of the basidium in the Agaricomycetes of the Basidiomycota. (A) The developmental stages of a typical basidium, (B) The development of basidia in a representative zone of the hymenium. *(A) Adapted from Smith, A.H., 1934. Mycologia 26, 305–331 with permission of the New York Botanical Garden. (B) from Corner, E.J.H., 1950. Ann. Botan. Mem. 1, 1–74, with permission of Oxford University Press. Drawings courtesy of Dr. George Chamuris.*

Life cycles

Information on the life cycles of the major Phyla of fungi that attack wood is necessary to understand the nature and great benefits of genetic diversity to fungi. Genetic diversity permits rapid adjustments through selection to new conditions. There are enormous differences in the patterns of sexuality, sexual mechanisms, and the life cycles of the fungi (Raper, 1966). More detailed information is available in the various textbooks on mycology. (e.g. Webster and Weber, 2007; Talbot, 1971; Alexopolous et al., 1996; Snyder, 2019). Only a few highlights of the topic are presented here. Sexual reproduction is an effective means of developing genetic diversity that occurs in all fungal classes except the fungi that currently have only an asexual state. Sexuality basically involves the union of gamete protoplasts (plasmogamy) and fusion (karyogamy) of

two nuclei (n) to form a diploid nucleus (2 n). The diploid nuclei divides by meiosis and crossing over or exchange of genetic material among the paired or homologous chromosomes can occur. Subsequent mitotic divisions then maintain and multiply the new genotypic arrangement. In contrast, only mitotic divisions occur in asexual reproduction and there is less chance for genetic diversity. In some cases, high reproductive potential coupled with a high frequency for errors or mutations during mitosis can overcome the limitations presented by the absence of a sexual stage. The life cycle of a typical wood-decaying fungus (Agaricomycetes) can be described in steps as follows (Fig. 3.9):

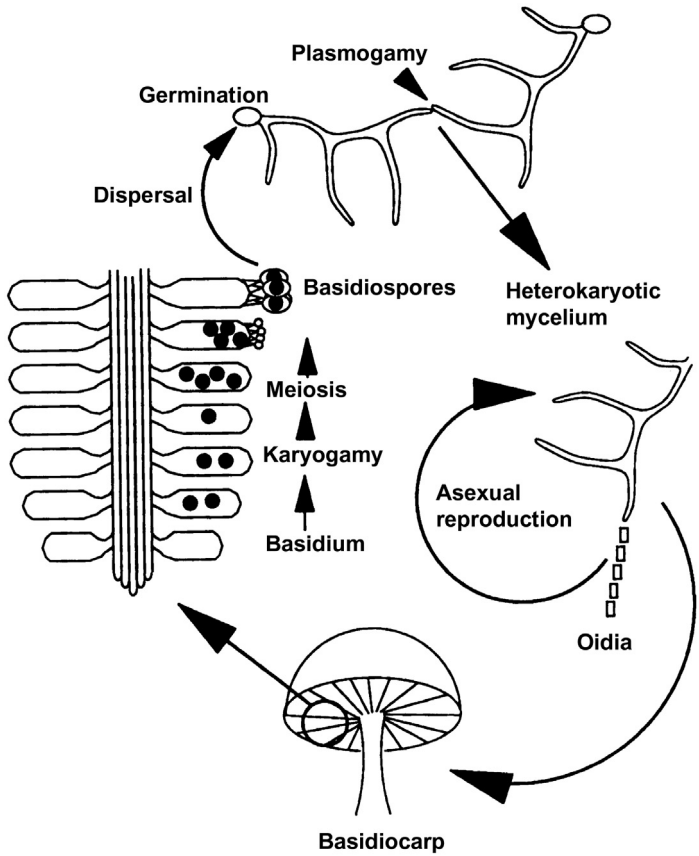


Figure 3.9 Life cycle of a typical fleshy Basidiomycota. The life cycle is monokaryon-dikaryon and then briefly diploid. The vegetative mycelium is binucleate and may be perennial. Sporophore (basidiocarps) develop sporadically. Hyphal fusions occur only among compatible mating types. *Courtesy Dr. J.J. Worrall.*

1. A *basidiospore* (haploid nuclear condition) germinates and forms hyphae. Since all the nuclei are the same genetically, it is a *homokaryon* and also a *monokaryon* (n) since each cell contains an identical nucleus.
2. The monokaryotic hypha may form branches that later separate at the septa forming spores (oidia or arthrospores) that germinate, forming new monokaryons. This is the asexual life cycle.
3. Two compatible monokaryotic hyphae fuse (anastomose) and their protoplasts, with two genetically different nuclei, become associated in a hyphal cell (plasmogamy). This is the beginning of the dikaryotic condition (n + n), which is unique to the Basidiomycota.
4. The dikaryonic condition in the hyphae maintains itself by simultaneous nuclear division with septa formation in new cells. This is facilitated by *clamp connections* in many Basidiomycota decay fungi. The major vegetative existence of the fungus is in the dikaryotic stage although the monokaryotic stage can also cause substantial decay.
5. When the energy reserves and environmental conditions are favorable, the mycelium aggregates and forms a sporophore.
6. Generative hypha mass on the surface of sporophore tissues and the terminal cells in the many hyphal chains become basidia. This zone of parallel-aligned basidia is called the hymenium.
7. Nuclear fusion of the two nuclei occurs and the basidium briefly enters the diploid stage (2 n).
8. The diploid (2 n) nucleus undergoes meiosis and four haploid nuclei are formed at the end of the two-stage process. The four nuclei move to the top of the basidium and migrate through sterigmata and into four exogenously produced basidiospores.
9. The basidiospores, when mature, are forcibly ejected. The life cycle begins again if one spore lands on a digestible substrate under favorable conditions.

There are many variations of this cycle. For example, hyphae in some Basidiomycota are heterokaryotic and become dikaryotic only in the cells basal to the basidium. Basidiospores are sometimes binucleate and germinate directly to form the dikaryotic stage (Raper, 1978). Occasionally, the homokaryon is self-fertile and clamp connections develop. Bipolar and tetrapolar compatibility patterns occur among the basidiospore isolates. Such patterns, when known, can be used to help determine the origins of decay columns in trees or products.

The heterokaryon is a common nuclear state of the hyphae in the Ascomycota and many asexual fungi. Openings in the septal pores appear

to facilitate the migration and accumulation of many nuclei in the cells of some species. In many Ascomycota, two compatible nuclei pair in an *ascogonium* and a series of complex nuclear divisions and wall formations leads successive ascogenous hyphae to form the asci, which usually contains eight haploid ascospores. Some asexual fungal species have a parasexual cycle that results in some genetic diversity. The process is complex and beyond our textbook purposes. In essence, it starts with heterokaryon formation, the fusion of some nuclei to form diploids, some crossing over of genetic material during their mitotic divisions, subsequent reversion to haploids, and their eventual isolation in conidia or hyphal tips.

It is readily apparent that fungi have developed a range of reproductive strategies to create and maintain genetic diversity allowing them to adapt to changing environmental conditions.

Reproductive capacity

Fungi can produce prodigious numbers of various sexual and asexual spores (Webster and Weber, 2007). The vast majority of these spores fail to land in locations with conditions suitable for growth and die. Some fungi adjust for this risk by limiting spore release to periods favorable for germination and subsequent host or substrate invasions. Others produce substantial numbers of resting spores that delay germination until substrate and environmental conditions are suitable for growth. A single sporophore of the wood-decay fungus *Gandoderma applanatum* has been estimated to produce billions of spores for extensive periods during the growing season. The large numbers of spores produced, coupled with their small size and ease of dissemination, means that any suitable substrates will be showered with spores within a short period. Prodigious reproductive capacity accounts for the high competitive effectiveness of both fungi and bacteria as carbon-compound decomposers. Microbial invasion of a substrate is certain to occur wherever and whenever suitable substrates and growth conditions occur.

Fungal variability

Fungi often exhibit wide ranges of variability in physiologic characteristics, appearances, and the ability to act as saprobes, pathogens, or mutualistic symbionts. This variability reflects, in part, the huge numbers of spores produced and the brief reproductive cycles of most fungi. A wide range of tolerances permits fungi and bacteria to respond quickly to

changing conditions. Examples include the rapid appearance of fungal strains resistant to new fungicides or bacterial strains tolerant of new antibiotics.

Variation represents, in part, a sorting out of the many strains or different genotypes comprising a species. Selection pressure then favors the steady increase of the strains whose genotype can cope with the new condition. Genotypic variability in fungi arises initially from mutations in the genome and crossing over or rearrangement of genes on the chromosome during meiosis or parasexuality. It is estimated that some fungi have 20 or more alleles on a single-gene pair, providing ample opportunity for developing new allele combinations. Aneuploidy, the addition or deletion of parts of a chromosome to the normal chromosome number, is reported to be common in fungi and may represent an important source of variation. The heterokaryotic condition is another source of variation since hyphae within a single mycelial mat may have many different combinations of nuclei with differing physiologic capabilities that affect appearance, growth, and survival. The sectoring of cultures and variations in the appearance of the fungal mats of a species often reflect this condition. For example, the monokaryon and dikaryon mycelial mats of some wood decay fungi differ in appearance, growth rates, and decay capabilities (Sexton, 1988).

The presence of plasmids or virus particles in a bacterium or fungus may provide an additional source of variation. Some of the most productive strains of penicillin-producing fungi are now known to be virus infected. Some viral infected bacterial strains are also more virulent. Conversely, plasmids can also render fungi less pathogenic. For example, plasmid-infected strains of the chestnut blight fungus, *Cryphonectria* (*Endothia*) *parasitica*, are less virulent. We are also learning that endosymbiotic bacteria can affect fungal fruiting and may affect many other fungal activities.

Other factors may be responsible for the variations observed among the isolates of a species. Some fungi are pleomorphic and produce different types of asexual spores depending on environmental conditions or age. Some fungi are dimorphic and may grow in yeast or mycelial stages depending on nutritional sources or environmental conditions.

The ability of fungi to rapidly adapt to new conditions substantially complicates disease and decay control programs. At the same time, however, this variation presents tremendous opportunities for industrial uses of fungi and future biotechnology developments.

Growth requirements

Decay fungi are able to grow on wood and damage or modify it under a wide range of environmental conditions. Four critical requirements for fungal growth in wood are:

Supply of free or unbound water
Favorable temperatures (0–42 °C)
Atmospheric oxygen
Digestible carbon compounds

These requisites and their relationship to decay prevention and control approaches are discussed in detail in Chapter 4.

A classification of fungi

For over a century, fungal classification was based primarily on differences in the types of the reproductive structures. As we will discuss, our ability to parse out the genetic make-up of fungi (and a host of other organisms) is radically altering our perceptions of classification. While physical features are still useful for identification, DNA sequencing has become a critical component in fungal identification.

The taxonomic study of fungi has two general purposes. One is identification and involves determining distinguishing features and characteristics that will lead to naming the fungal species. A critical first step in the control of most disease and biodeterioration problems is to determine the identities of the associated microorganisms. Fungal identities can also clarify ecologic roles, help determine structural and physiological relationships, and assist in the development of rational prevention or control programs. The need to identify fungal associates is even more understandable when we realize the wide array of fungal species and the small size as well as the similarity in general appearance.

A second important purpose of classification is the arrangement of organisms in the order of their phylogenetic origin. Such systems of classification are spoken of as natural systems. Related organisms in a natural system generally have many similarities and, in this way, detailed knowledge of a few often yields general information about many.

Early classifications of wood-decay fungi were based on convenient macroscopic characters such as the size and shape of the fruiting structures

and the nature of the surfaces bearing the hymenium (tubes, gills, spines, smooth, etc.). The limited fossil record of fungi made it difficult to suggest phylogenetic relationships. Another great difficulty was the evolutionary phenomena of convergence, where macroscopically similar fungi sometimes have very different origins. The result was a classification system based upon fruiting body features such as smooth, poroid, toothed or other arrangements of the hymenium that placed many similar looking, but phylogenetically distant organisms into the same groups. Techniques for assessing differences in DNA and RNA have markedly enhanced the ability to delineate between direct genetic connections and convergence.

Major revisions in the classifications of many groups will continue as new information on the microscopic, ultrastructural, physiologic, genetic and biochemical features of fungi accumulates. Similar changes are occurring in the classifications of the bacteria.

The general classification scheme we will use for fungi is in the current edition of the "Ainsworth & Bisby's Dictionary of Fungi" (Kerk et al., 2009). Emphasis in this section will be placed on the groups of fungi that inhabit, modify, or destroy wood. Drastic taxonomic revisions have occurred in all groups of fungi and will continue to occur as researchers examine the genetic basis for many groups.

Fungi are placed in a single group, the Eumycota, and are considered to have a common ancestor. One current classification scheme lists 7 Phyla within the Fungi: Microsporidia, Chytridiomycota, Blastocladiomycota, Neocallimastigomycota, Glomeromycota, Ascomycota, and Basidiomycota. Mycosporidia are internal parasites of other organisms and will not be considered here.

Chytridiomycota are often called chytrids are distinctive because they produce motile zoospores with a single flagellum. They are not important in wood degradation, but they have recently been implicated in a number of high-profile animal diseases notably with bats and many amphibians. Like the chytrids, Blastocladiomycota and Neocallimastigomycota produce motile zoospores, but primarily feed on decaying organic matter. The latter group lives in anaerobic environments including the digestive systems of many herbivores or other extreme environments where oxygen is limited. These groups, while important in other applications are not considered to be wood degraders.

Glomeromycota are arbuscular mycorrhizal fungi that have close symbiotic relationships with their plant hosts. These fungi form unique structures (arbuscles) and help plants capture nutrients from the soil such as

nitrogen, sulfur and phosphorous. These fungi are associated with almost all land-based plants and there has been speculation that this plant/fungus association was essential for the movement of plants from water to land. These fungi, while not wood decayers, play important roles in forest health.

Zygomycota formerly included the Glomeromycota and is currently a group in flux. For simplicity, we will continue to use the name but recognize that it is no longer valid. The group includes the genera *Mucor* and *Rhizopus* that are commonly found on freshly cut lumber. These fungi are characterized by rapid growth and abundant production of spores. Their role in wood is not clear, but they cause little or no damage beyond cosmetic effects on the wood surface.

The Ascomycotina and basidiomycotina are by far the most prevalent fungi in wood and play important roles in carbon recycling. There has been a revolution in fungal classification which has shifted from a morphological system based primarily on fruiting bodies to one that includes more detailed information on genetic composition.

For example, one of the most important morphological attributes for separating the Ascomycota was whether the ascus was unitunicate or bitunicate. However, this morphological separation has been replaced by a taxonomy based upon DNA sequences. The group is currently divided into three Sub-phylla: Pezizomycotina, Saccharomycotina, and Taphrinamycota. The Pezziomycotina includes all of the species that produce an ascocarp and includes some wood decay fungi. The Saccharomycotina includes single celled species such as yeasts that reproduce by vegetative budding (*Saccharomyces* spp., an industrial yeast and *Endomycopsis fasiculata*- a yeast growing in ambrosia beetle tunnels), while the Taphrinomycotina are a mixture species including some plant pathogens. For the purposes of this discussion, we will limit our discussion to the Pezizomycotina that are further divided into 4 groups.

Examples of groupings within the Pezizomycotina within the Ascomycota.

Pezizomycetes: formerly inoperculate cup fungi	Lecanoromycetes: Lichenized fungi	Leotiomycetes: Asci cylindrical- includes many plant pathogens	Sordariomycetes: Perithecial fungi includes many wood inhabitants

(*Continued*)

(Continued)
Examples of groupings within the Pezizomycotina within the Ascomycota.

Example:	**Example:**	**Example:**	**Examples:**
Morchella spp.- edible fungi and probable mycorrhizae	*Cladonia* spp, common symbiont of lichens	*Erisyphe graminis*- causes powdery mildew of wheat	*Xylaria* spp- white rot fungi that produce black zone lines *Daldinia concentrica*- white rot in slash piles *Ustilina deusta*- white rot causing root and saprots of hardwoods *Ophiostoma* spp- common sapstain fungi *Chaetomium globosum*- soft rot fungus

Basidiomycota

As with the Ascomycota, the Basidiomycota have seen a radical reclassification that is still underway. The group is still classified on the basis of the presence of a basidium and the production of basidiospores, but biochemical examinations of genetic relationships have led to considerable reclassification. Formerly, species were classified on the basis of the characteristics of their fruiting structures including the orientation of the hymenium. Potential wood inhabiting fungi were classified as either having an exposed hymenium (Hymenomycetes) or an enclosed hymenium (Gasteromycetes). The Hymenomycetes were classified as having a hymenium borne on the surface of the basidioma and were divided into two main groups based upon having a septate or non-septate basidium. The non-septate basidium species were further delineated into the Aphyllophorales, the Agaricales, Dacrymycetales, and the Septobasidiales. All but the last group contained some decay fungi with the majority being placed in the Aphllyophorales. This separation was based primarily upon

morphological features and genetic analysis has radically reclassified these fungi.

The Basidiomycota have been re-ordered into the Sub-Phyla Agaricomycotina, Pucciniomycotina and Ustilaginomycotina. The Pucciniomycotina include the rust fungi that attack many plants. White pine blister rust (*Cronartium ribicola*) is an excellent example of a member of this group that causes extensive losses in tree growth and mortality, while *Helicobasidium corticioides* causes a brown pocket rot on conifers.

Ustilaginomycotina includes the smut fungi that are pathogenic on many important crops, but does not include any wood degrading fungi. Corn smut (*Ustilago maydis*) is an excellent example. The Agaricomycotina includes the former Hymenomycetes as well as the Gasteromycetes, but also includes the jelly fungi that were formerly classified with the rusts and smuts. The group has been divided into Dacrymycetes, Tremellomycetes and the Agaricomycetes.

Examples of wood attacking fungi in the Dacrymycetes and Tremellomycetes

Dacrymycetes: jelly fungi with branched basidia	Tremellomycetes: gelatinous fruiting bodies grow on dead wood
Example: *Dacrymyces stillatus*. common wood decayer under wet conditions	**Examples:** *Tremella* spp- usually on dead portions of shrubs *Auricularia auricula-judae* causes a white rot on hardwoods

The Class Agaricomycetes contains the most genera within the Agaricomycotina including a majority of the common wood degrading fungi. There are 21 orders within this Class including nearly all of the species formerly classified as Hymenomycetes. Important members of this group include:

Agaricales

The Agaricales order includes the gilled mushrooms that we are most familiar with and contains over 400 genera. A number of important wood decayers including:

Schizophyllum commune: an important early colonizer of freshly fallen trees. It is a weak white rotter and tends to infect and produce fruiting bodies with distinctive split gills very quickly after a tree dies or on freshly sawn lumber.

Pleurotus spp. (Oyster mushroom): White rot fungus that produces edible fruiting bodies

Clavaria spp.: White rot fungus that produces distinctive coral-like fruiting bodies

Hymenochaetales

Includes 27 genera, many of which are important heart rot fungi including

Inonotus obliquus: a heartwood of standing hardwoods, notable birch. Its fruiting body is believed to have medicinal value

Phellinus weirii: causes laminated root rot of Douglas-fir and other trees

Phellinus pini: (red ring rot) causes a white rot of standing trees of many species

Phellinus igniarius (tinder fungus): Causes a white rot of standing hardwoods

Gloeophyllales

This order is defined by its ability to produce a brown rot of wood and contains several important species

Gloeophyllum trabeum and *G. sepiarium* are two important brown rotters of wood in service, particularly above ground. *G. trabeum* is an important test fungus and is tolerant of several organic wood preservatives.

Neolentinus lepideus: is an important brown rotter of many wood products and is well-known for its tolerance of creosote.

Boletales

Originally restricted to species that produced fleshy poroid fruiting bodies whose hymenium was easily separated from the cap, this group has been expanded to include many other fungi including a number of aggressive wood decayers.

Meruliporia incrassata and *Serpula lacrymans* are both called True Dry rot fungi. Both are aggressive brown rot fungi that can produce root-like rhizomorphs that translocate water from the soil to the wood.

Coniophora puteana is a common brown rot fungus that is common in buildings in Europe. It is also commonly used as a test fungus in decay tests.

Polyporales

Includes many of the species formerly called polypores because of the presence of pores on the hymenium, but now includes a number of other species including corticioid fungi that produce a crust-like hymenium on the wood surface.

Rhodonia placenta is one of the more common brown rots on wood products. It is also tolerant of copper-based preservatives and is commonly used as a test fungus when evaluating new wood preservatives.

Trametes versicolor is a white rot fungus and an important degrader of hardwoods. It is also commonly used as a test fungus when evaluating preservatives especially in hardwoods.

Ganoderma applanatum is a widely distributed white rot fungus found on living and dead trees. It produces a perennial conk or fruiting structure. The freshly formed hymenium is easily discolored and artists often create elaborate drawings on the surface leading to is also being called the Artist's Conk. The conk also has medicinal properties and is commonly used in traditional Asian medicines.

Phanerochaete chrysosporium is a white rot fungus found on hardwoods. It is relatively unimportant in decay of timber, but has been heavily studied because it can selectively remove lignin and decompose many complex organic pollutants. This makes it potentially useful for bio-pulping or for remediation of chemically contaminated sites.

Fibroporia vaillantii and *Fibroporia radiculosa* are both brown rot fungi that are common in decaying softwoods in soil contact. Both are exceptionally tolerant of copper and are being investigated to better understand the mechanisms of copper tolerance.

Wolfiporia (cocos) extensa is another brown rot fungus that produces a large, unique underground sclerotium that leads to it being called the "Tuckahoe fungus." Like *F. vallaintii*, this fungus has exceptional tolerance to copper compounds.

Bjerkandera adusta is a white rot that attacks living hardwoods. This species has also been studied for its ability to degrade organic pollutants.

Russulales

Includes a number of important wood degraders and tree pathogens including:

Heterobasidion annosum is a widely distributed root pathogen of conifers in the Northern Hemisphere. It causes a white rot and is especially problematic in pine plantations where it spreads between trees via root grafts.

Xylobolus frustulosa is a white rot found on many hardwoods and produces a white, crustose fruiting body. It is among the fungi used to test wood preservatives

Cantharellales

This group includes a number of edible mushrooms especially the Chantarelles (*Cantharellus* spp.), but also includes fungi that produce their hymenium on tooth-like projects. The group includes several fungi that grow on wood and have lignin-degrading enzymes.

Sistotrema brinkmannii is variously described as having no clearly categorized rot type. It is typically found on softwoods and appears to be important in non-soil contact exposures. Curiously, it is also known as an ectomycorrhizal species in some conifers.

Deuteromycetes or fungi imperfectii

This group contains fungi that are only known by their asexual state. This is no longer considered to be a valid group, but it persists until such time as the species within it can be properly categorized. Many of these species are likely to be Ascomycetes or Basidiomycetes, but the connections between the sexual and asexual stages have not been established. This group contains about 25,000 species and includes many important plant pathogens, sap-stain fungi, and soft rot wood decay fungi.

It is important to note that changes in taxonomy do not necessarily negate the value of older descriptions, but information must be carefully assessed for inaccuracies. Names assigned to genetic sequences using data bases such as FUNGuild are based upon prior morphological identifications. A number of descriptions and keys for the families in what was formerly the Aphyllophorales are available in Chapters by Talbot, Peterson, Harrison, and Pegler in Ainsworth et al. (1973). The two-volume textbook on the poroid fungi of North America by Gilbertson and Ryvarden (l986, 1987) also provides taxonomic keys, based on macroscopic and microscopic descriptions for family, genera, and species designations of many of the important members of this group in North America. These keys remain valuable, particularly for those without access to laboratories with molecular capabilities for routine fungal identification. Those who use these keys will then need to check the names against current taxonomic groupings, but the fungi themselves have not changed.

A classification of bacteria

Bacteria are everywhere and their roles in a variety of biological systems are still being elucidated. Bacteria have long been known to degrade the pit membranes in logs stored under water for long periods, leading to sinker logs (Elwood and Eklund, 1959) and they are important, if slow, degraders of submerged woods. Extensive studies by Nilsson highlighted the importance of tunneling bacteria in the early stages of wood decomposition in direct soil contact (Daniel, 2014). Bacteria that inhabit the gills of shipworms also may play a role in cellulose decomposition in the gut of these organisms, as the shipworm gut is largely sterile. Bacteria inhabit the gut of termites, and play important roles in the wood digestion by these animals. Bacteria are widespread and abundant in the environment, and although they decay wood more slowly than fungi in aerobic environments, they digest large amounts of woody biomass in terrestrial and aquatic ecosystems. However, it is not the intent of this chapter to fully describe bacterial taxonomy or their broad role in carbon cycling in different environments, but rather to provide sufficient background so that their possible roles in the decomposition process can be better understood.

This section will be limited primarily to descriptions of the several groups of bacteria where representatives are known to modify wood or attack plant cell wall constituents or aid other organisms in the decay process. Detailed information on the kinds of bacteria, their structures and functions, and roles in disease, industry, and the biosphere are available in textbooks on bacteriology or microbiology (e.g. Bender et al., 2017; Nester et al., 1983; etc.).

Bacteria are prokaryotes. There are three major groups of bacteria, based on their energy sources. Photosynthetic bacteria (phototrophs) obtain energy from sunlight, while autotrophic (cemolithotrophic) bacteria obtain energy from the oxidation of inorganic compounds including sulfur, ammonia, and hydrogen. Heterotrophic (chemoorganotrophic) bacteria obtain their energy from the oxidation of organic compounds and are most important in wood deterioration. This group plays a complementary role with the fungi as the major decomposers of carbon compounds in the biosphere.

Bacteria are grouped into genera and species based on morphological, physiological, biochemical and genetic characteristics. Bacteria traditionally

were segregated into groups based on their reaction to a Gram stain (positive and negative), their shape, color, presence of flagella, and their ability to grow on specific media. The ability to isolate and sequence DNA has radically changed the identification process and created a wealth of data that have been used to classify these organisms. Bergey's Manual of Determinative Bacteriology (George and Garrity, 2005) is the classic publication used for classifying and identifying bacteria. A more recent treatise on the Procaryotes by Rosenberg et al. (2014) is also useful. As with the fungi, the development of molecular techniques has dramatically changed and expanded our knowledge of the bacteria present in a variety of environments.

Many free-living bacteria can decompose cellulose in diverse environmental settings such as sea water, soils, and alimentary tracts (Schmidt et al., 1987; Greaves, 1968, 1970; Daniel and Nilsson, 1986; Drysdale et al., 1986; Holt et al., 1979; Rossell et al., 1973). The number of bacterial species that invade and decompose various wood constituents in living trees and wood products remains unknown simply because of the large number of bacteria that are still unknown and therefore untested. Some groups of bacteria of special interest to wood microbiologists are listed and described.

Cytophagales

Small, rod-shaped, chemo-organotrophs, without flagella, yet possessing the unusual property of gliding motion. They are morphologically similar to many gram-negative bacteria. *Cytophaga* is an important genus in the group and contains cellulosic decomposers. An interesting feature of various *Cytophaga* spp. is the regular alignment of the bacterial cells on the fiber surface oriented parallel to microfibrillar alignments of the cell wall. *Sporocytophaga myxoccoides* is reported to cause severe soft rot damage to wood exposed to ocean waters.

Gram-negative facultatively anaerobic rods

Enterobacter cloacae (*Erwinia nimipressuralis*) is associated with wet wood formation in elms, maples, willows, and poplars. Bacteria are commonly found in the heartwoods of many species. Methane is formed during metabolism, generating considerable pressure in stems and is partially responsible for copious exudate flows as well as a foul odor.

Gram-negative, aerobic rods, and cocci

The best-known genus in this group is *Pseudomonas*. Members of this genus are reported as the initial invaders in stem wounds in forest trees and have been proposed to play a role in the successional aspects of decay development in stems. Some species are antagonistic to fungi and have been explored for their potential as biological control agents.

Endospore-forming rods and cocci

Bacillaceae: Are gram positive bacteria. *Bacillus polymyxa* is reported to decompose the margo fibrils and torus in the bordered pits of submerged pine logs. The damage to the pits is substantial and increases liquid permeability to the degree that logs stored in water will sometimes sink. This species is also reported to fix nitrogen and has been explored for its biocontrol potential.

Clostridium quericcolum is reported as the cause of discoloration in living oaks. Many of the cellulolytic anaerobic bacteria in wet conditions are in the genus *Clostridium* which also includes *C. botulinum*, the cause of botulism poisoning in poorly prepared canned foods.

Actinobacteria

Gram negative bacteria are important in a variety of soil environments. Some form strand-like growth that resembles fungal hyphae. Many produce antibiotic compounds that inhibit fungi and have been explored for their biocontrol potential.

Coryneform humiferan is associated with wet-wood formation in poplars and bacteria from cedar have been found to stimulate *Phlebia brevispora* (Harry-Ascobarra and Kamei, 2019).

Micromonospora spp. have been isolated from the alimentary tract of termites and are presumed to play a role in cellulose decomposition. Similarly, *Enterobacter* have been associated with termite guts (Potrikas and Breznak, 1977). These are just a few examples of the bacteria associated with the decay process and it is likely that we will continue to find more important roles for these organisms.

Nocardia spp. and *Streptomyces* spp. are associated with soft rot development in pines submerged in marine environments.

The roles of fungi and bacteria in ecosystems and human affairs

A textbook focusing on wood decay and property changes caused by fungi and bacteria will naturally equate these organisms with their well-known negative roles of disease, death, and deterioration. It seems appropriate at this point, to briefly mention some of the broad, positive roles that microorganisms play in ecosystem stability and human affairs for a more balanced perspective. The industrial aspects of these roles are discussed in detail in Chapter 20 on Wood and Biotechnology.

Carbon cycle: Fungi and bacteria are the principal decomposers of organic materials and release carbon dioxide to the atmosphere that is indispensable for the continued photosynthetic activity of green plants. It is sobering to recognize that this fragile, balanced carbon cycle controls the major energy input to most living systems. The removal of accumulating organic debris is another part of this process. Soil fertility is improved and soil organic matter, an important soil constituent, formed. Increasing carbon dioxide levels as a result of anthropogenic inputs will place increasing importance in extending the useful life of wood to sequester carbon.

Nitrogen cycle: Bacteria play a major role in providing available nitrogen for green plants through nitrogen fixation and cycling. While the atmosphere is mostly nitrogen, it is in a state that is unavailable for plant growth. The ability of bacteria to fix nitrogen from the atmosphere is a major driver for the plant growth that sustains nearly all other organisms on the planet.

Mutualistic symbionts: Fungi and bacteria are beneficial symbionts with a wide range of other life forms. Symbiotic relationships include the mycorrhiza fungi associated with tree roots and agricultural plants, fungi with algae in lichens, and nitrogen fixing bacteria in the root nodules of legumes. The symbiotic activity of bacteria and fungi in the digestive tract of ruminant animals facilitates cellulose decomposition. Similarly, symbiotic relationships between shipworms and bacteria, and insects such as termites and bacteria, allow these organisms to digest large amounts of lignocellulose biomass in both terrestrial and aquatic environments.

Food sources: Fungi and bacteria are the principal food sources for a wide range of microfauna including protozoa, ambrosia beetles, and composting ants. Humans consume an estimated 60–70 million pounds of mushrooms per year.

Industrial chemicals and medicinals: Fungi and bacteria in controlled fermentation or transformation processes produce a wide range of useful chemicals and foods, including yeasts, vitamins, alcohols, glycerols, citric acid, enzymes, cortisone, antibiotics, and dairy products. The unique metabolic capabilities of some bacteria and fungi form the basis for a multibillion-dollar chemical industry. This industry will grow at a tremendous rate as pressures are put on the extraction of fossil resources, and we learn to more precisely exploit the genetic potential of heterotrophic microorganisms and lignocellulose substrates. We are also learning that many fungi produce potent anti-cancer compounds. We are just beginning to explore the potential for these organisms.

Biological control of diseases and pests (potential): Fungi and bacteria are pathogens of some insect pests and are antagonistic to other pathogens or harmful saprobes. Increasing use is being made of these organisms as biological control agents thereby reducing the need to use pesticides.

Biodegradation agents: Fungi and bacteria play major roles in detoxification of industrial poisons and in the breakdown of sewage sludges, garbage, and other xenobiotic wastes into industrially useful or harmless products. They also have great promise for cheaply and effectively modifying materials for subsequent industrial use, e.g. increasing wood permeability for preservative treatment or biological delignification of pulpwood.

Summary

1. Fungi that are involved in wood deterioration are characterized by filamentous eukaryotic cells. They are heterotrophic, have an external mode of digestion, and they reproduce via spore production. Virtually all fungi have chitin or chitosan as a component of the skeletal framework for their cell walls. These features place fungi in a separate kingdom, the Fungi.
2. The tubular cells of fungi, termed hyphae, are the basic cellular unit of all filamentous fungal structures. The "hyphal system" of many fungi appears to be uniquely adapted to penetrate, externally digest, absorb, and metabolize a wide range of plant materials, including wood.

3. Fungi reproduce primarily by spores that are one to several celled structures formed from hyphae. Many of the lignicolous fungi produce huge numbers of spores that, coupled with effective dissemination agents, such as wind, water, and insect vectors, assure that most wood substrates are continuously exposed to wood-inhabiting fungi.
4. The high reproductive potential of fungi, their short life cycles, and great variability give many fungal species the ability to adapt readily to changing conditions, such as changing environments, the introduction of new fungicides or resistant varieties of plants. This large reproductive potential and high variability also offers great potential for improving the capabilities of fungi in biotechnology developments and biological control programs.
5. The negative impacts of fungi as disease (pathogen) and biodeterioration agents (saprobes) are well known. Some of the advantageous roles of fungi are carbon and nitrogen cycling in the ecosystem, symbionts, as agents in the production of industrial chemicals and pharmaceuticals, as biological control agents, and as potentially important biodegradation or biotransformation agents.
6. An important first step in the understanding and prevention or control of many biodeterioration problems is the identification of the type of fungi involved.
7. Traditional classification of lignicolous fungi placed major emphasis on the macroscopic features of spore bearing structures, which made identification of fungi that did not readily produced sexual spores challenging. Advances in molecular technologies now permit rapid and accurate detection of many fungi. The major classes of wood-inhabiting fungi are the Ascomycota and the Basidiomycota. The Deuteromycotina are an invalid, but necessary category that is still in use practically until asexual fungi can be properly reclassified.
8. In the Basidiomycota, the Agaricomycetes contains many of the important wood-decaying fungi. Important orders are the Agaricales, Boletales, Cantharellales, Gloeophyllales, Hymenochaetales, Polyporales, and Russullales. Fungal taxonomy is still in flux as taxonomists probe the essence of fungal evolution and new lineages.
9. Key literature sources and references are provided in the text to facilitate the identification and correct naming of the important lignicolous fungi.
10. Information on the types and importance of bacteria that invade and damage wood and its products remains limited. This reflects the large

number of bacteria that are still unknown and that are not possible to isolate in pure culture. The roles of free-living and symbiotic bacteria relative to biodeterioration of wood, continue to be discovered.

References

Alexopolous, C.J., Mims, C.W., Blackwell, M.W., 1996. Introductory Mycology. John Wiley and Sons, NY.

Bartnicki-Garcia, S., 1968. Cell wall chemistry, morphogenesis and taxonomy of fungi, Ann. Rev. Microbiol., 22. pp. 87–105.

Becket, A., Heath, I.B., McLaughlin, D.J., 1974. An Atlas of Fungal Ultrastructure. Longman, London, 221 pp.

Bjurman, J., 1994. Ergosterol as an indicator of mould growth on wood in relation to culture age, humidity stress and nutrient level. Intl Biodeterior. Biodegrad. 33 (4), 355–368.

Davidson, R.W., Campbell, W.A., Vaughn, D.B., 1942. Fungi causing decay of living oaks in the eastern United States and their cultural identification. U.S. Dept. Agric. Bull. No. 785. 65 pp.

De Saad, D., DeSaad, D., Kinsey, G.C., Patterson, R., Gaylarde, G.C., 2003. Ergosterol analysis for the quantification of fungal growth on paint films. Proposal for a standard method. Surf. Coat. Int. Part. B Coat. Trans. 86 (2), 131–134.

Douglas, L.M., Konopka, J.B., 2014. Fungal membrane organization: the episome concept. Annu. Rev. Microbiol. 68, 377–393.

Gow, N.A.R., Latge, J.-P., Munro, C.A., 2016. The fungal cell wall: structure, biosynthesis and function. ASM Sci. Microbiol. Spectr. 5 (3), FUNK-0035-2016.

Harry-Ascobarra, J., Kamei, I., 2019. Bacterial strains isolated from cedar wood improve the mycelial growth and morphology of white rot fungus *Phlebia brevispora* on agar and liquid media. J. Wood Sci. .

Lindenmayer, A., 1965. Carbohydrate metabolism. 3. Terminal oxidation and electron transport. In: Ainsworth, G.C., Sussman, A.S. (Eds.), The Fungi, Vol. I. Academic Press, New York, pp. 301–348.

Margulis, L., 1981. Symbiosis in Cell Evolution. W.H. Freeman and Company, New York.

Montgomery, H.J., Montreal, C., Young, J.C., Seifert, K.A., 2000. Determination of soil fungal biomass from ergosterol analysis. Soil. Biol. Biochem. 32 (8), 1207–1217.

Nobles, M.K., 1965. Identification of cultures of wood-inhabiting Hymenomycetes. Can. Jour. Bot. 43, 1097–1139.

Pantazopoulous, A., 2016. The golgi apparatus: insights from filamentous fungi. Mycologia 108 (3), 603–622.

Parsi, Z., Gorecki, T., 2006. Determination of ergosterol as an indicator of fungal biomass in various samples using non-discriminated flash pyrolysis. J. Chromatogr. A 1130 (1), 145–150.

Potrikas, C.J., Breznak, A., 1977. Nitrogen fixing *Enterobacter agglomerans* isolated form the guts of wood eating termites. Appl. Environ. Microbiol. 33 (2), 392–393.

Raper, J.R., 1966. Life cycles, basic patterns of sexuality, and sexual mechanisms. In: Ainsworth, G.C., Sussman, A.S. (Eds.), The Fungi: An Advanced Treatise, vol. II. Academic Press, New York and London, pp. 473–511.

Raper, C.A., 1978. Control of development by the incompatibility system in Basidiomycetes. In: Schwalb, M.N., Miles, P.G. (Eds.), Genetics and Morphogenesis in the Basidiomycetes. Academic Press, New York, p. 168.

Riquelme, M., 2013. Tip growth in filamentous fungi: a road trp to the apex. Annu. Rev. Microbiol. 67, 587–609.
Snyder, D., 2019. Introductory Mycology. Laser and Keller Education, US.
Stahl, P.D., Parkin, T.B., 1996. Relationship of soil ergosterol concentration and fungal biomass. Soil. Biol. Biochem. 28 (7), 847–856.
Stalpers, J.A. 1978. Identification of wood-inhabiting fungi in pure culture. Centraalbureau voor Schimmelcultures, Baarn, Studies in Mycology No. 16, 248 pp.
Swift, M.J., 1973. The estimation of mycelial biomass by determination of the hexosamine content of wood tissue decayed by fungi. Soil. Biol. Biochem. 5, 321–332.
Whittaker, R.H., 1969. New concepts of kingdoms of organisms. Science 163, 150–160.
Wu, L.C., Stahmann, M.A., 1975. Chromatographic estimation of fungal mass in plant materials. Phytopathology 65, 1032–1034.

Classification of fungi

Ainsworth, G.C., Sparrow, F.K., Sussman, A.S., 1973. The Fungi, An Advanced Treatise. Volume IVB. A Taxonomic Review with Keys: Basidiomycetes and Lower Fungi. Academic Press, p. 504.
Rosenberg, E., DeLong, E.F., Lory, S., Stackebrandt, E., Thompson, F., 2014. The Prokaryotes, vol. 6. Springer-Verlag, Berlin.
Talbot, P.H.B., 1971. Principles of Fungal Taxonomy. Macmillan, New York, 274 pp.
Wang, C.J.K., 1965. Fungi of Pulp and Paper in New York. State University College of Forestry at Syracuse University Tech. Pub. 87.
Wang, C.J.K., Zabel, R.A. (Eds.), 1990. Identification Manual for Fungi from Utility Poles in the Eastern United States. American Type Culture Collection, Rockville, MD.
Webster, J., Weber, R.W.S., 2007. Introduction to Fungi, third ed. Cambridge University Press, Cambridge, 669 pp.

Characteristics and classification of bacteria

Bender, K.S., Buckley, D.H., Stahl, D.A., Madigan, M.T., Sattley, W.M., 2017. Brock Biology of Microorganisms. Pearson Education, London, U.K., 1136 pages.
Daniel, G., 2014. Fungal and bacterial biodegradation: white rots, brown rots, soft rots and bacteria. In: Schutz, T.P., Goodell, B., Nicholas, D.D. (Eds.), Deterioration and Protection of Sustainable Biomaterials. ACS, Washington, D.C., American Chemical Society Series 1158.
Daniel, G., Nilsson, T., 1986. Ultrastructural observations on wood degrading erosion bacteria. Intl. Res. Group on Wood Pres. IRG/WP/1283.
Drysdale, J.A., Rutland, P.J., Butcher, J.A., 1986. Isolation and Identification of Bacteria From Degraded Wood -a Progress Report. Intl. Res. Group on Wood Pres., Stockholm, Sweden, IRG/WP/192.
Garrity, GM (Ed.). 2005. Bergey's manual of systematic bacteriology. Auflage. In: Garrity, G.M. (Ed.), Volume 2: The Proteobacteria, Part B: The Gammaproteobacteria. Springer, NY.
Greaves, H., 1968. Occurrence of bacterial decay in copper-chrome-arsenic treated wood. Appl. Microbiol. 16, 1599–1601.
Greaves, H., 1970. The effect of selected bacteria and actinomycetes on the decay capacity of some wood rotting fungi. Mater. und Organismen 5, 265–279.
Holt, D.M., Gareth Jones, E.B., Furtado, S.E.J., 1979. Bacterial breakdown of wood in aquatic habitats. Record British Wood Preservers Association Annual convention. pp. 13–22.
Nester, E.W., Roberts, E.E., Lidstrom, M.E., Pearsall, N.N., Nester, M.T., 1983. Microbiology., third ed. Dryden Press, p. 875.

Rossell, S.E., Abbot, E.G.M., Levy, J.F., 1973. A review of the literature relating to the presence, action, and interaction of bacteria in wood. J. Inst. Wood Sci. 32, 28–35.
Schmidt, O., Nagashima, Y., Liese, W., Schmidt, U., 1987. Bacterial wood degradation studies under laboratory conditions and in lakes. Holzforschung 41 (3), 137–140.

Further reading

Alexopoulos, C.J., Mims, C.W., Blackwell, M., 1996. Introductory Mycology, third ed. John Wiley &Sons, Inc., New York, p. 869.
Aronson, J.M., 1965. The cell wall. In: Ainsworth, G.C., Sussman, A.S. (Eds.), The Fungi I. Academic Press, New York, pp. 49–76.
Ballou, C.E., 1976. Structure and biosynthesis of yeast mannan. Adv. Microbiol. Physiol. 14, 93–158.
Barnett, H.L., Hunter, B.B., 1972. Illustrated Genera of Imperfect Fungi. Burgess Publishing Co., Minneapolis, Minn.
Bracker, C.E., 1967. Ultrastructure of fungi. Ann. Rev. Phytopathol. 5, 343–374.
Burnett, J.H., 1968. Fundamentals of Mycology. St. Martin's Press, New York, 546 pp.
CABI, 2009. Ainsworth and Bisby's Dictionary of the Fungi (tenth ed.). Kirk, P.M., Cannon, P.F., Minter, D.W., Stalpers, J.A. (Eds.).
Corner, E.J.H., 1932. A *Fomes* with two systems of hyphae. Trans. Brit. Mycol. Soc. 14, 263–291.
Fries, E.M., 1874. Hymenomycetes Europaei. Upsala.
Gilbertson, R.L., Ryvarden, L., 1986. North American Polypores, vols. I and II. Fungiflora, Oslo, Norway, p. 433.
Griffin, D.H., 1981. Fungal Physiology. John Wiley and Sons, New York, 377 pp.
Harris, S.D., 2013. Golgi organization and the apical extension of fungal hyphae: an essential relationship. Mol. Microbiol. 89 (2), 212–215.
Hawker, L.E., 1965. Fine structure of fungi as revealed by electron microscopy. Biol. Rev. 40, 52–92.
Hawksworth, D.L., Sutton, B.C., Ainsworth, G.C., 1983. Ainsworth & Bisby's Dictionary of the Fungi, seventh ed. Commonwealth Mycological Institute, Kew, Surrey, 443 pp.
Holt, G., (editor-in-chief) 1989. Bergey's Manual of Systematic Bacteriology. Vol. 1 (Krieg, N.R. (Ed.) 964 pp); Vol. 2 (Sneath, P.H.A. (Ed.) 636 pp); Vol. 3 (Staley, J.T. (Ed.) 744 pp); and Vol. 4 (Williams, S.T. (Ed.) pp. 440). Williams and Wilkins, Baltimore, MD.
Holt, J.G. (Ed.), 1977. The Shorter Bergey's Manual of Determinative Bacteriology. eighth ed. Williams &Wilkins Co., Baltimore, MD.
Hughes, S.J., 1953. Conidiophores, conidia, and classification. Can. J. Bot. 31, 577–659.
Hunsley, D., Burnett, J.H., 1979. The ultrastructural architecture of the walls of some hyphal fungi. J. Gen. Microbiol. 62, 203–218.
Hunt, J., 1956. Taxonomy of the Genus *Ceratocystis*. Lloydia 19 (1), 1–58.
Kirk, P.M., Cannon, P.F., Minter, D.W., Stalpers, J.A. (Eds.), 2001. Ainsworth & Bisby's Dictionary of the Fungi. tenth ed. CABI Publishers, New York, NY.
Knuth, D.T., McCoy, E., 1962. Bacterial deterioration of pine logs in pond storage. For. Prod. J. 12 (9), 437–442.
Kobayashi, D.Y., Couch, J.A., 2009. Bacterial/fungal interactions: from pathogen to mutualistic endosymbionts. Annu. Rev. Phytopathol. 47, 63–82.
Lentz, P.L., 1955. *Stereum* and Allied Genera of Fungi in the Upper Mississippi Valley, Agri. Monograph No. 24. U.S. Department of Agriculture, Washington, D.C.
Lowe, J.L., 1942. The Polyporaceae of New York State (except *Poria*). New York State College of Forestry Tech. Pub. 60, Syracuse, N.Y.

Lowe, J.L., 1957. Polyporaceae of North America. The Genus *Fomes*. State University College of Forestry at Syracuse University Tech. Pub. 80, Syracuse, N.Y.

Lowe, J.L., 1966. Polyporaceae of North America. The Genus *Poria*. State University College of Forestry at Syracuse University Tech. Pub. 90, Syracuse, N.Y.

Matheny, P.B., Curtis, J.M., Hofstetter, V., Aime, M.C., Moncalvo, J.M., Ge, Z.W., et al., 2006. Major clades of Agaricales: a multilocus phylogenetic overview. Mycologia 98 (6), 982–95.

Moore, R.T., 1965. The ultrastructure of fungal cells. In: Ainsworth, G.C., Sussman, A.S. (Eds.), The Fungi, vol. I. Academic Press, New York, pp. 95–118.

Raven, P.H., Evert, R.F., Eichhorn, S.E., 1986. Biology of Plants, fourth ed. Worth Publishers, Inc, New York, p. 775.

Ruggiero, M.A., Gordon, D.P., Orrell, T.M., Bailly, N., Bourgoin, T., Brusca, R.C., et al., 2015. A higher level classification of all living organisms. PLoS One 10 (4), e0119248. Available from: https://doi.org/10.1371/journal.pone.0119248.

Schink, B., Ward, J.C., Zeikus, J.G., 1981. Microbiology of wet wood: Importance of pectin degradation and *Clostridium* species in living trees. App. Env. Microbiol. 42 (3), 526–532.

Slysh, A., 1960. The Genus *Peniophora* in New York State and Adjacent Regions. State University College of Forestry at Syracuse University Tech. Pub. 83.

Wessels, J.G.H., Sietsma, J.H., 1973. Wall structure and growth in Schizophyllum commune. In: Burnett, J.H., Trinci, A.P.J. (Eds.), Fungal Walls and Hyphal Growth. Cambridge University Press, London, p. 417.

CHAPTER FOUR

Factors affecting the growth and survival of fungi in wood (fungal ecology)

Contents

In a practical sense, factors affecting growth and development of fungi in wood substrates can generally be equated with the development of decay or discolorations. When the critical growth requisites of a wood damaging fungus are known, it is sometimes feasible to modify wood handling or use practices to adversely affect fungus growth, thereby achieving prevention or economic control of the problem. Information on the growth needs of fungi is also useful for optimizing cultural conditions for industrial fermentations or other emerging biotechnology processes.

The many chemical and physical factors affecting fungal and bacterial growth are covered in more detail in textbooks on fungal physiology (Lilly and Barnett, 1951; Cochran, 1958; and Griffin, 1981) and microbiology (Stanier et al., 1963; Brock et al., 1984). This chapter emphasizes the growth factors affecting decay fungi, placing emphasis on their limits and citing examples where changes in wood handling or use practices have or may achieve useful control of biodeterioration problems. The

Wood Microbiology.
DOI: https://doi.org/10.1016/B978-0-12-819465-2.00004-8

following chapter on metabolic activities of fungi demonstrates that most environmental factors affecting fungal growth are ultimately traceable to enzyme characteristics and reactions.

Major growth needs of wood-inhabiting fungi

Like all living organisms, fungi have certain requirements for growth and survival. The major growth needs of wood-inhabiting fungi are:

1. Water – free water on the surfaces of cell lumina.
2. Oxygen – atmospheric oxygen at relatively low levels for most fungi and very low levels or chemical oxygen only for some microaerobic and facultative anaerobic fungi.
3. Favorable temperature range – optima for most wood-inhabiting fungi range from 15 to 40 °C.
4. Digestible substrate (wood, etc.) – provides energy and metabolites for synthesis via metabolism.
5. Favorable pH range – pH optima for most wood-inhabiting fungi range from 3 to 6.
6. Chemical growth factors, such as nitrogen compounds, vitamins, and essential elements. The last two factors are often included with the substrate.

The absence of toxic extractives, while not a requirement, is necessary for growth by most fungi on wood. Visible light is needed by some fungi for the development of spore-bearing structures and may play a role in other physiologic functions. High levels of ultraviolet light are lethal to most fungi.

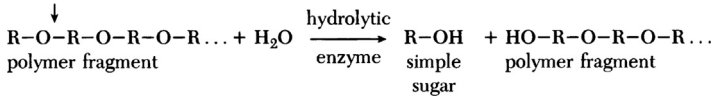
Water

Removing moisture or preventing wetting are simple decay prevention or control practices in many wood applications. Water serves four general purposes for fungal growth in wood:

a) *Reactant in hydrolysis*. Decay involves the exo-cellular digestion of plant cell wall carbohydrates by hydrolytic enzymes released by fungi. Water is one of the reactants in the chemical process. If R is used to designate simple sugar monomers connected by glycosidic bonds

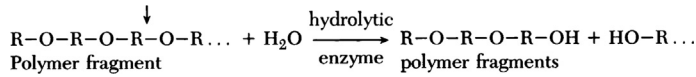
forming carbohydrate polymers, the reactions involved hydrolysis can be simplified as follows:

1) Endwise attack (one or two sugar units)

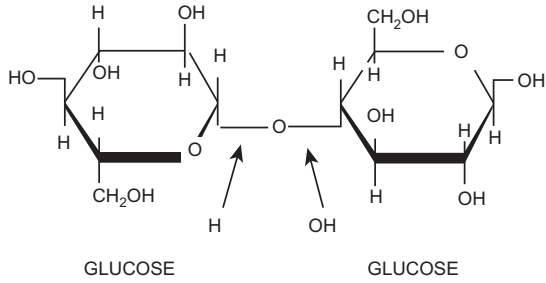


2) Hydrolytic enzyme

3) Random attack



These reactions release smaller fragments of the original polymer or glucose units. The digestion of a cellobiose (2 glucose units) in this hydrolytic reaction by a hydrolytic enzyme (β-glycosidase), as illustrated below, occurs by migration of proton ($H+$) and hydroxyl (OH^-) ions from water to the broken glycosidic bond, forming two glucose molecules.



Site of enzyme action on cellobiose.

b) *Diffusion medium for enzymes and solubilized substrate molecules*: Decay is an exo-cellular process that occurs outside the hyphal cell wall. Free (liquid) water is needed by most wood-inhabiting fungi both as a solvent and diffusion medium for movement of digestive enzymes (hydrolytic) out from the hyphae and the solubilized substrate decomposition products that are subsequently absorbed by the hyphae.

c) *Solvent or medium for life systems*. Water is the indispensable solvent for the many enzymes within the fungal hyphae that are involved in cell metabolic processes such as respiration, synthesis, and growth. It is also

the medium supporting colloidal suspensions that are critical to the structure and functioning of protoplasm necessary for all life forms.

d) *Wood-capillary swelling agent*. Water molecules in dry wood are attracted by hydrogen bonding to the hydroxyl groups on the carbohydrate macromolecules making up the cell walls. As the wood adsorbs water, it swells primarily in the radial and tangential planes. During swelling, small capillaries in the cell wall enlarge and attain a size range permitting the penetration of both free water and fungal enzymes deep into the so-called "transient capillary zone" of the cell wall. This process greatly facilitates the penetration of digestive enzymes (large protein macromolecules) into the cell wall at the ultrastructural level.

Extensive experience with wood in its many uses indicates that dry wood in protected environments or water-saturated wood seldom decays. Important questions for many users of wood have been to know the critical wood moisture limits when decay begins or stops, and how varying amounts of water in wood affect the rate of decay development. These are difficult questions since moisture gradients exist in wood from outer to inner zones. It is difficult to maintain precise moisture contents in wood test samples during the lengthy decay process, many fungi release metabolic water during the decay process, and there is considerable fungal variability in responses to moisture limits. Also, oxygen and carbon dioxide levels often become confounding factors at the upper moisture content ranges.

Wood moisture content is generally expressed as a percent of the oven-dry weight. This value is used in a physical property sense to indicate the total amount of water present in the wood. Fungal physiologists and wood microbiologists are also interested in the availability of the water in the wood to fungi. Water is tightly adsorbed to wood cell wall polymers and unavailable to most fungi at levels below the fiber saturation point. Units used to express the availability of water in wood to a fungus are osmotic pressure, the bar, or Pascal. A bar is defined as 0.987 atmospheres or 0.1 MPa (mega Pascal). Another useful unit is water activity, which is defined simply as the vapor pressure of water over the substrate divided by the vapor pressure of pure water (Griffin, 1981). By this measure, wood reaches essentially the fiber saturation point when stored in a confined space in a water-vapor saturated atmosphere. A more detailed discussion of various wood-water relationships is presented in Chapter 6.

Minimum moisture concentrations: There are many older reports on the minimum moisture levels in wood necessary to sustain fungus growth or decay development. These are briefly listed as to fungi, moisture levels, and sources.

Fungi	Moisture level	Source
Wood-decay fungi	25–32%	Snell et al., 1925
Xylobolus (*Stereum*) *Frustulosum*	16–17%	Bavendamm and Reichelt (1938)
Schizophyllum commune	16–17%	Bavendamm and Reichelt (1938)
Antrodia sinuosa	26%	Freyfeld (1939)
Common wood decay fungi:	22–24% (20% as safe control Level)	Cartwright and Findlay (1958)
Ophisotoma (*Ceratocystis*) *piliferum*	23%	Lindgren (1942)
"Wood-inhabiting fungi"	Slightly above the saturation point	Etheridge (1957)

Griffin (1977) analyzed the water needs of wood-inhabiting fungi for decay initiation in terms of water potential. Water potential measures available water in soil-water-plant root studies that may be analogous to fungal hyphae-wood water relationships. Water potential is defined as the sum of the matrix and osmotic potential of the wood-water system expressed in bar units. In a simplified sense, water potential measures the relative availability of the water in the wood to the passive osmotic capabilities of the fungal hypha in contact with it. Matrix potential depends on the dimensions in the transient capillary system and reflects the strong adhesion forces that retain free water in small capillaries. Osmotic potential depends on the amounts and types of solutes (sugars, extractives, etc.) in the free water in the wood. Griffin defined the fiber saturation point of wood as the condition when all voids of a radius greater than 1.5 μm are devoid of water. He stated that the limiting growth level for fungi is −40 bar, which is consistent with the 30% value often given as the approximate lower limit for decay. He also noted the extreme difficulty of measuring these values experimentally.

These data clearly show that fungi must have some free water present in the cell lumen to grow effectively in wood. This means that fungi are unable to grow effectively in wood below the fiber-saturation point (FSP). The FSP varies with wood species, but an average figure of

28–30% is generally assumed. Dry (seasoned) wood in structures protected from external water sources cannot exceed the FSP (see Chapter 7) providing a cardinal principle for decay control in wood uses in protected environments. As an added safety factor, the prudent decay control rule prescribed 30 years ago by Cartwright and Findlay (1958), which is still followed by many wood users is that to prevent the growth of microorganisms, wood moisture content may not exceed 20% based on oven-dry weight. It is important to note, however, the temporal nature of water in wood. For example, kiln dried lumber in shipping containers often develops mold in transit. This occurs because the wood is not absolutely dry (it is around 17% moisture content). Moisture evaporating from the wood as the container heats can later condense, leaving liquid moisture on the surface of the wood. Mold spores on the wood surface can germinate and sporulate within 24–48 hours. The wood later dries, leaving mold, dry wood and suggesting that the fungus was able to grow without free water.

Maximum moisture concentrations: Most wood-inhabiting fungi are unable to grow effectively in water-saturated wood. A principal reason is that these fungi are obligate aerobic organisms and require moderate amounts of oxygen for respiration. As moisture content increases above the f.s.p., water steadily replaces the air in the cell lumina (atmosphere-21% oxygen) and eventually oxygen becomes limiting. Since the void volume of wood varies inversely with specific gravity, the upper moisture levels limiting fungal growth will be much lower in denser woods (Fig. 4.1). Fungi vary in sensitivity to oxygen depletion. Reducing the void volume of the wood to about 20% of the original void volume causes a rapid reduction in decay rate. Water-saturated wood is resistant to fungal and insect attack and may be stored safely for many years without losses in most structural properties; however, more tolerant organisms, including bacteria, may begin to degrade the wood. Free water contains only a few parts per million of oxygen, and the rate of oxygen diffusion into large wood units such as logs is very slow and insufficient to support active fungal growth. The slow rate of oxygen movement into large wood members may help explain why ponding or continuous water spraying are effective for storing veneer bolts and high-quality saw logs. It should be stressed that most fungi will survive under these conditions, but not grow actively. Some soft-rot fungi decay wood submerged in the ocean or wood exposed continuously to near-saturated conditions such as the baffles in cooling towers or subterranean surfaces of utility poles on

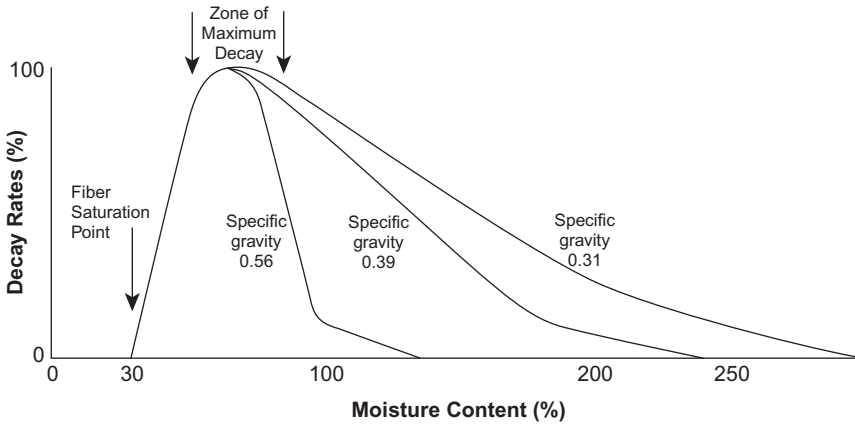


Figure 4.1 An approximation of the cardinal moisture contents for the rates of decay development of several common mesophilic basidiomycete decay fungi as judged from reports in the literature and experience. Maximum moisture contents are calculated based on data from Higgins (1957) and Skaar (1972).

wet sites. Some bacteria invade submerged wood and may cause extensive damage to parenchymatous tissue and cell wall pit membranes. This type of damage is often found in archeological wood and necessitates careful restoration to reinforce the weakened wood so that it does not collapse as it dries. Anaerobic bacteria are also associated with wet wood formation in living trees.

Optimum moisture concentrations: Optimal moisture levels for the growth of many wood-inhabiting fungi are not known, but experience with laboratory decay tests suggest that the optimum wood moisture levels for most decay fungi lie between 40–80% (Scheffer, 1973). Optimum moisture values probably vary considerably, and may help explain some of the substrate or condition specificities of fungal types and species. For example, laboratory decay tests with white-rot fungi require more moisture than brown-rot fungi to achieve optimum wood weight losses (Highley and Scheffer, 1970). The differences may partially explain the prevalence of white-rot fungi in wet coniferous chip piles in contrast to the high levels of brown-rot fungi found in drier coniferous slash in the forest or above-ground uses in structures.

Survival at various moisture levels: The effect of wood moisture content on the survival of the major types and species of wood-inhabiting fungi remains poorly defined. The survival of heartrot, early decay, or stain fungi during storage or seasoning in the wood product in service may be

important in wood uses such as house siding or utility poles where intermittent wetting occurs. Studies on survival of many wood decay fungi in ponded pulpwood bolts (*Pinus resinosa)* reported no visible growth, although fungi survived for up to 38 weeks (Schmitz and Kaufert, 1938).

Scheffer and Chidester (1948) evaluated the survival of both decay and sapstaining fungi in air-seasoned wood. Boards with viable heartrot fungi or invaded by decay or stain test fungi were stored at 27 °C and 65% relative humidity. These conditions would bring the boards to an equilibrium moisture content of approximately 12%. *Phellinus* (*Trametes*) *pini* and *Phaeolus schweinitzii*, two common heartrot fungi in conifers, survived six months or less, while some important wood products decayers such as *Rhodontia placenta*, *Gloeophyllum trabeum, Fomitopsis* (*Fomes*) *roseus*, and *G. saepiarium* were still alive after three years. *Meruliporia* (*Poria*) *incrassata*, an important building rot fungus, died within 25 days. Seven of the eight sapstain fungi died within seven months. The single exception for sapstain fungi was *Aureobasidium pullulans* which survived one year. This fungus is a common sapstainer in window sashes and a major cause of the "mildew" (pigmented mycelium) that often disfigures paint surfaces. The ability to survive at low wood moisture levels may explain its common presence in these two wood uses. A variety of fungi can invade timber during the period between cutting and final use of the product. Many decay fungi produce chlamydospores that allow them to survive in a dormant stage for many years. A survey of framing in buildings in California showed that many samples contained viable decay fungi that were attributed to survival of fungi through the harvesting and sawing process (Dietz and Wilcox, 1997). Moisture content plays a critical role in determining which species colonize wood and the extent to which these fungi utilize the substrate.

Oxygen

Most fungi are obligate aerobes that require free oxygen for several metabolic reactions involving energy release or synthesis. Aerobic respiration involves a major series of reactions that uses atmospheric oxygen as a reactant. The classic summation equation for respiration indicating direct oxygen use is as follows:

$$C_6H_{12}O_6 + H_2O \rightarrow 6\ CO_2 + 6\ H_2O + \text{energy}$$

Atmospheric oxygen serves as an electron and proton acceptor in the terminal respiration reactions. It is important to note that the sugar (glucose) is completely oxidized and a maximum amount of chemical energy released. Oxygen is also a direct reactant where oxidase enzymes are involved.

Some fungi, such as the yeasts, are able to obtain energy directly from chemical compounds by a series of internal redox reactions in a process termed fermentation, when oxygen levels are low or oxygen is absent. Organisms capable of fermentation are called facultative anaerobes. The classic simplified fermentation equation is as follows:

$$C_6H_{12}O_6 \rightarrow 2\ C_2H_sOH + CO_2 + 57\ \text{Kcal/mole}$$

In this case, the sugar (glucose) is only partially metabolized and the process is energy inefficient. Some bacteria and fungi are obligate anaerobes and require no oxygen. Anaerobic respiration uses chemical radicals such as NO_3, SO_4 or CO_3 to serve as the electron acceptors in the energy release process. According to evolutionists, primitive organisms developed anaerobic respiration and fermentation as energy release processes long before the accumulation of oxygen in the atmosphere as a by-product of photosynthesis by bacteria and plants. While most of the cellulose and related carbohydrates in the biosphere is oxidized by aerobic microorganisms (principally decay fungi) and are emphasized in this section, it should be remembered that an estimated 5–10% of the cellulose produced annually in the biosphere is converted to methane in the absence of oxygen in anaerobic environments such as animal rumens, sediments, and bogs (Vogels, 1979; Ljungdahl and Ericksson, 1986). These anaerobic reactions involve consortia of bacteria in fermentation sequences as summarized by the formula:

$$C_6H_{12}O_6 \rightarrow 3\ CH_4 + 3CO_2$$

Despite the current abundance of oxygen in the atmosphere (21%), these anaerobic processes are still of major importance in many specialized settings such as waterlogged soil, sediments, bogs, or specialized niches such as ruminant and termite digestion systems.

The effects of the atmosphere (with emphasis on O_2 and CO_2) on the growth of fungi in culture has been studied intensively as an important variable in the industrial production of fungal products such as antibiotics,

citric acid, and yeasts. A detailed review and extensive bibliography of this literature was assembled by Tabak and Cooke (1968).

The amount of oxygen consumed by aerobic fungi is directly related to the amount of carbon dioxide produced. The ratio of the volume of CO_2 used to the oxygen consumed is termed the respiration quotient and is a useful unit in metabolic studies of fungi. For example, a respiration quotient of 1 would imply that the nutrient source was a simple sugar. Respirometry has been applied in several other interesting ways in wood microbiology. The amount of carbon dioxide produced or oxygen consumed have been used to monitor decay development, to compare the decay rates of fungi on various substrates (Good and Darrah, 1967; Toole, 1972), and to evaluate wood preservatives (Halabisky and Ifju, 1968 and Behr, 1972). Smith (1975) compared an automated analytical procedure to measure respiration and the soil-block test procedure to compare the wood preservative possibilities of several agricultural fungicides with those of pentachlorophenol, chromated copper arsenate, and creosote. He concluded that the respiration method was quicker and provided reliable information in 4 weeks compared to the 12 weeks required by the standard soil-block preservative evaluation procedure.

The amount of oxygen in the wood or a cultural flask is often simply expressed as the atmospheric pressure in millimeters (mm) of mercury. Conversion of pressure in atmospheres to bars or pascals (Pa) can be made readily since one bar equals 0.987 atmospheres or 0.1 MPa (mega Pascal). For example, wood in equilibrium with the atmosphere at sea level would be assumed to contain oxygen at about 160 mm of pressure ($21/100 \times 760$ mm Hg).

Another useful measure is the ratio of the oxygen in the substrate to the amount or partial pressure of O_2 in the atmosphere, usually expressed as a percentage. In respirometry studies, the amounts of oxygen consumed or CO_2 formed are usually measured manometrically and expressed as volumes of gas per unit time (e.g., μL/hr). There are many reports on the effects of atmospheric gases (primarily O_2 and CO_2) on the growth of decay fungi or decay rates (Scheffer and Livingston, 1937; Thacker and Good, 1952; Jensen, 1967; Toole, 1972; Van der Kamp et al., 1979; Highley et al., 1983; Scheffer, 1986). These reports indicate that some gaseous oxygen is required for the growth of fungi in virtually all cases of wood biodeterioration; however, the amounts are very low (probably in the range of 1%). Many fungi have optimal growth rates at oxygen levels above 20% of ambient air (Fig. 4.2). Several papers of historical or special

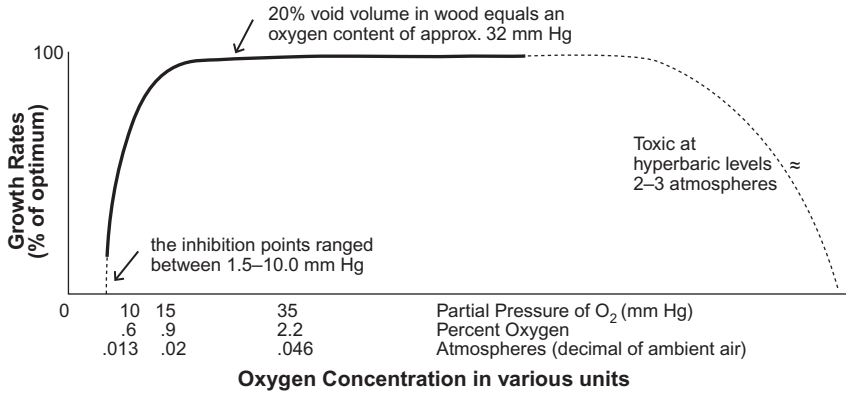


Figure 4.2 The growth responses of *Trametes versicolor* to various concentration of oxygen. *The lower ranges were taken from the data of Scheffer, T.C. 1986. O_2 requirements for growth and survival of wood-decaying and sapwood-staining fungi. Can. J. Bot. 64, 1957–1963.*

interest are briefly reviewed with emphasis placed on the amounts of oxygen needed or inhibitory carbon dioxide levels.

Minimum oxygen concentrations: In 1929, Snell reported on experiments on several commercial woods in a range of specific gravities to determine the optimum and inhibitional moisture levels for decay development. He concluded that decay development essentially ceased when the accumulating free water in the wood had reduced the air content or void volume to about 20% of the original air volume. Oxygen was assumed to be the limiting factor. This choice of 20% of the residual air volume as a decay limiting level became a frequently cited statistic (Boyce, 1961).

Scheffer and Livingston (1937) determined the effects of mixtures of nitrogen and oxygen on the growth of *Trametes versicolor* in malt agar cultures. Growth first began to decline at an oxygen partial pressure of 37 mm Hg, decreased rapidly below 15 mm Hg, and ceased between 10 and 1.5 mm Hg (1.3 to 0.2% oxygen).

In studies on the effects of various concentrations of oxygen, nitrogen, and carbon dioxide on the biomass of several oak heartrot fungi and *Trametes versicolor* grown in liquid cultures, Jensen (1967) showed that fungal biomass production decreased rapidly at an oxygen concentration of 15% and ceased in the absence of oxygen. Increasing levels of carbon dioxide acted as a growth inhibitor in oxygen and nitrogen mixtures (Fig. 4.3). Differences were also noted in the sensitivities of some of the fungi to varying concentrations of oxygen and carbon dioxide. Several

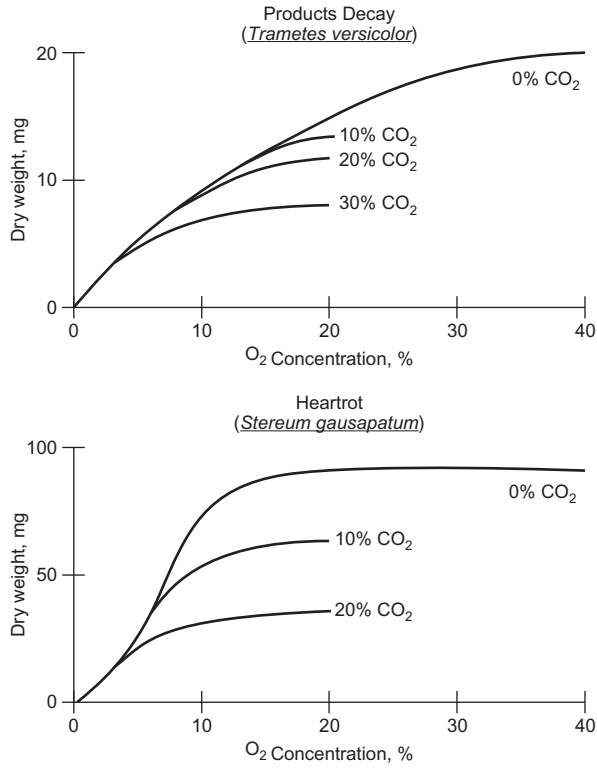


Figure 4.3 The effects of various concentrations of oxygen and carbon dioxide on the growth of a products decay (*Trametes versicolor*) and a heartrot fungus (*Stereum gausapatum*). *Data from Jensen, J.F. 1967. Oxygen and carbon dioxide affect the growth of wood-decaying fungi. For. Sci. 13 (4), 384–389.*

heartrot fungi were less sensitive to oxygen deficits and tolerant of the higher levels of carbon dioxide (Scheffer, 1986). Tolerance to low oxygen and high carbon dioxide levels in the heartwood zones of living trees may help explain the apparent selectivity of some fungi for this environment.

In subsequent studies, Highley et al. (1983) found that low O_2 levels (7.6 mm) and high CO_2 levels (76 mm) substantially reduced decay compared to ambient levels; however, there were no clear-cut differences between the saprot and heartrot fungi. Oxygen levels also strongly influenced decay rates. Only slight decay occurred below 0.01 atmospheres of oxygen and it increased progressively as oxygen pressures rose to 0.1 and 0.2 atmospheres. Scheffer (1986) studied the minimum oxygen requirements for growth by 48 decay fungi and 6 sapwood-stain fungi and found that no fungi grew at oxygen levels of 1.5 mm Hg (approximately 0.2%

volume). Fungal growth was moderately and then severely retarded at O_2 pressures of 11 mm and 2 mm Hg (1.3% to 0.3% volume), respectively.

Low oxygen levels in tightly stoppered decay chambers may be responsible for some of the puzzling variations observed in decay tests using similar procedures. Eades and Roff (1953) found substantial decay reductions when tight rubber stoppers were used to seal the decay chambers in laboratory studies. These rate variations were reduced significantly when uniform aeration or venting devices were used.

Optimum oxygen concentrations: Oxygen response curves for decay fungi indicate a sharp decrease in growth response between 1 to 2% oxygen (Fig. 4.2). Respiratory oxygen needs appear to be met at relatively low levels. A close inspection of the curves at higher oxygen levels indicates a slow increase in growth rates, suggesting that optimum growth may occur above current atmospheric oxygen levels.

Maximum oxygen concentrations: Since the growth of fungi or decay development at oxygen levels above the 21% of the atmosphere is unlikely to occur in nature, this aspect of fungal growth has been neglected. Highley et al. (1983) found that decay rates increased for several saprot fungi and decreased for two heartrot fungi at oxygen levels of 0.4 atmosphere. Reid and Seifert (1983) also showed that some fungi decayed wood more rapidly in pure oxygen at atmospheric pressure (100% O_2) than air (21% O_2), It is of special interest to note that decay rates increased in pure oxygen atmosphere, although there was no difference in fungal growth rate. These differences raise questions concerning the reliability of using hyphal growth responses of fungi to oxygen or other abiotic growth requisites as measures of the decay process. Hyperbaric levels of pure oxygen exceeding one atmosphere are toxic to several fungi including *Meruliporia lacrimans*; however, some *Penicillium* species grow slowly in three atmospheres of pure oxygen (Caldwell, 1963). Large accumulations of pyruvic acid were associated with the toxic action at high oxygen concentrations.

Survival of low oxygen concentrations: Effects of low oxygen levels are relevant to the major decay and stain fungi that are all aerobes. Decay tests of several heartrot fungi at near anaerobic levels found survival, but no wood weight loss after a 10-week incubation period, suggesting a method by which heartrot fungi survive the occasional anaerobic conditions that occur in living trees (Van der Kamp et al., 1979). In studies on the survival of fungi in sealed tube cultures, Scheffer (1986) showed that heartrot fungi survived longer than sap and products decay fungi. Two important

building decay fungi, *Meruliporia (Serpula) incrassata* and *Coniophora puteana*, died within a week, while several heartrot fungi survived two years or longer and the six sapstaining fungi in the test survived from one to six months. Oxygen depletion by respiration is a major factor in a sealed tube test, but other factors such as the accumulation of carbon dioxide or toxic metabolic products and nutrient exhaustion may also affect survival.

Oxygen and CO_2 levels in tree stems: The effects of heartrot on timber losses in the forest have focused considerable attention on stem conditions that might explain the apparent selectivity of this specialized group of fungi for this environment. Many studies (Chase, 1934; Thacker and Good, 1952; Hartley et al., 1961; Jensen, 1969a,b; Van der Kamp et al., 1979) have established that gases withdrawn from central stem tissues have lower levels of O_2 and higher CO_2 levels than those found in the atmosphere. The living cambium serves as an effective barrier to the diffusion of gases, effectively isolating the interior of the tree. Gas concentrations in stems varies with position and season of the year. Carbon dioxide accumulation and oxygen reduction presumably reflect respiration activities of bacteria, fungi, and wood parenchyma cells. Oxygen depletion does not normally reach levels limiting to fungal growth; however, decreases in wood permeability in wet wood formation further isolate tissues and complete anaerobic conditions sometimes develop. Wet wood in *Abies concolor* and other tree species is characterized by the presence of bacteria that may inhibit decay fungi by depleting oxygen and producing low molecular weight organic acids (Worrall and Parmeter, 1983). Wet wood in black cottonwood stems contained less than 0.1% oxygen for several weeks in the summer season (Van der Kamp et al., 1979). In addition, large volumes of methane may be formed in the wet wood zone by methanogenic bacteria in the genus *Methanobacterium* (Zeikus, 1974).

Soft-rot fungi: Soft rot fungi are often associated with water-soaked wood in baffles in cooling towers or the below-ground sections of posts and poles. In most cases, the soft-rot damage is shallow and limited to surfaces possibly reflecting anaerobic conditions deeper in the wood. The higher tolerance of soft rot fungi to low oxygen concentrations, compared to the white and brown rot groups, may explain their prevalence as decay agents in water-saturated woods (Duncan, 1961).

Carbon-dioxide relationships: Carbon dioxide is toxic to fungi at higher concentrations, but small amounts of CO_2 (0.05%) are essential for fatty acid synthesis in fungi (Griffin, 1981). This requirement suggests a critical growth need that may have been overlooked in previous studies, where

pure sources of oxygen and nitrogen were used to prepare gas mixtures; however, growth responses have been noted in the absence of carbon dioxide (Morrell, 1981). Carbon dioxide always increases in confined zones as O_2 is consumed in natural aerobic decay, and the two factors are confounded and interactions are possible. Increases in CO_2 can also elevate media acidity to undesirable ranges and this factor has often been overlooked in CO_2 toxicity studies. In addition to its combined toxicity and acidity effects, carbon dioxide also inhibits fruiting in basidiomycetes (Niederpruem, 1963; Taber, 1966). It will be interesting to observe how increasing levels of atmospheric carbon dioxide influence wood decay rates. Accelerated decomposition would confound efforts to use timber to sequester carbon dioxide as part of strategies to limit climate change.

Ponding and water sprays as storage practices: Long-term protection from insect and fungal attack can be achieved when wood is water saturated by immersion or spraying. These practices are often used to control decay in logs and pulpwood during storage (see Chapter 13). Bridge or building foundation piling also produce long service lives when the wood remains saturated. In such wood uses, the respiration of living cells in recently felled logs or bolts and/or the resident microbiota may rapidly deplete the oxygen in the stem gases and accumulate toxic levels of CO_2, The low solubility of O_2 in water and slow diffusion rates into the wood minimize replacement, near-anaerobic conditions develop, and aerobic fungal growth ceases. However, facultative anaerobic or microaerophyllic bacteria can slowly grow in some wood under these conditions. For example, the bacterium *Bacillus polymyxa* destroys pit membranes in ponded sugar pine logs and drastically increases wood permeability (Ellwood and Eckland, 1959). Subsequent work has shown that other species can damage ponded wood (Fogarty and Ward, 1973). Some soft rot fungi are able to attack limited portions of the cell wall on the surfaces of saturated wood when the water is constantly aerated. Soft rot damage may become important when the wood surface: volume ratio is high such as slats in water cooling towers (Levy, 1965).

Temperature

Temperature directly affects the many integrated metabolic activities of fungi such as digestion, assimilation, respiration, translocation, and synthesis that are mediated by enzymes. Within limits, metabolic reaction rates increase with increasing temperature until some reaction in the sequence

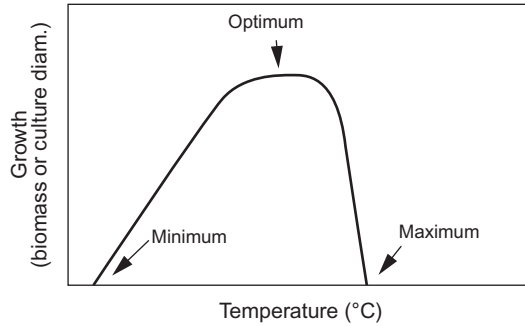


Figure 4.4 A hypothetical growth curve indicating the cardinal growth limits of a typical mesophilic wood-inhabiting fungus.

becomes rate-limiting or the heat denatures the enzymes. In the early logarithmic growth phases of many fungi, the approximate doubling of specific growth rate per 10 °C essentially follows the doubling of chemical reaction rates in a similar temperature range.

The effects of temperature on fungal growth and other metabolic activities have been studied extensively because of their many applications in the control of disease and biodeterioration and the industrial production of fungal metabolites. Temperature effects on fungi have generally been determined by growth as measured by hyphal extension rates on media surfaces or biomass accumulations. These measures, however, may not accurately reflect the role of temperature in wood degradation.

Cardinal temperature levels: Each fungus possesses three cardinal growth temperatures: a minimum level (growth begins), an optimum level (best growth), and a maximum level (growth ceases). Often the optimum temperature is skewed toward the maximum temperature, particularly for those fungi with the higher optimum growth levels (Fig. 4.4).

Growth curves for fungi must be viewed, in many cases, as approximations since many factors, including media, aeration, and method of growth measurement can alter fungal response. They have also generally ben developed through studies on synthetic media, not in wood.

In general, the temperature limits for the growth of most fungi lie between 0 and 45 °C (Fig. 4.5). Within these limits, fungi have adapted to utilize various substrates under difference temperature conditions. Psychrophiles are considered to have minima below 0 C and maxima of 20 °C with an optimum range between 0° and 17 C. A few psychrophilic (cold-loving) fungi in the genera *Cladosporium*, *Sporotrichum*, or

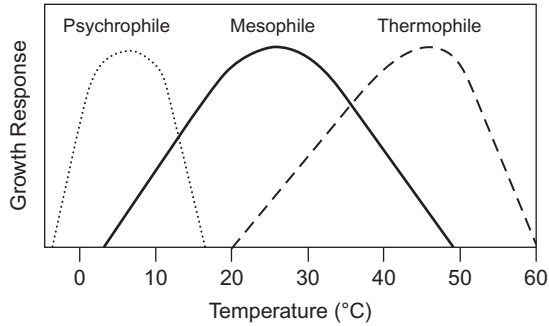


Figure 4.5 Approximate growth ranges of psychrophilic, mesophilic, and thermophilic wood-inhabiting fungi.

Thamnidium can cause serious problems on some refrigerated foods even a few degrees below 0 °C (Cochran, 1958). Snow mold fungi, *Typhula* and *Fusarium* species, can grow on cereals and turf grasses under snow cover and cause serious plant diseases (Agrios, 1988). At the other extreme, thermophilic fungi tolerate or grow at temperatures above 50 °C.

Thermophiles have been defined as having minima at or above 20 °C and maxima above 50 °C (Cooney and Emerson, 1964). *Chaetomium thermophile* and *Penicillium duponti* are found in decomposing compost or chip piles, where heat builds up as the biomass deteriorates. Several Actinomycetes (bacteria) are true thermophiles that tolerate high temperatures and cause wood damage in wood chip piles. Most fungi, including many wood-inhabiting fungi, are *meso*philic and grow best within a 15–40 °C temperature range. Their minima and maxima are considered to be above 0 °C and below 50 °C. Comparative growth rates of decay fungi at 28 °C on malt-extract agar are a widely used character in cultural identification. Wood microbiologists are particularly interested in the temperature requirements of wood-decay fungi that may explain their distributions and wood product selectivity, or predict the development and location of decay hazards. Optimum temperature selection is also important for comparative studies of decay fungi such as toxicity bioassays. Information on temperatures lethal to fungi is very important to the wood processor, since survival of these fungi in the product can lead to subsequent decay problems.

Temperature grouping of wood-inhabiting fungi: Information on the growth rates of 56 species (and several strains) of wood decay fungi on malt agar at temperatures ranging from 0 to 40 °C were assembled by Humphrey

Table 4.1 Optimal temperatures for common wood decay fungi.

Low temperature (<24 °C)		Intermediate temperature (24–32 °C)		High temperature (>32 °C)	
Serpula lacrimans	20	*Meruliporia incrassata*	28	*Gloeophyllum trabeum*	34
Phellinus pini	20	*Neolentinus lepideus*	28	*Gloeophyllum saepiarium*	36
Heterobasidion annosum	24	*Schizophyllum commue*	28	*Lentinus strigosus*	36
Hirschioporus pargamenus	24	*Trametes versicolor*	28	*Trametes hirsute*	34
Phaeolus schweinitzii	24	*Phellinus igniarius*	30	*Gloeophyllum striatum*	36

After Humphrey, C.J., Siggers, P.V., 1933. Temperature relations of wood destroying fungi. J. Agric. Res. 47, 997–1008.

and Siggers (1933). Growth measurements at 1-and 2-week intervals were used to determine the minimum, optimum, and maximum temperatures. Of the fungi tested, 12 had optima below 24 °C, 42 optima between 24–32 °C, and 10 optima above 32 °C. Growth was entirely inhibited or sharply retarded for all the species tested at 12 °C. No fungus grew above 46 °C, and most ceased growth at or above 40 °C. A few fungi, such as *Gloeophyllum trabeum* and *G. saepiarium*, had high optimums ranging from 34 to 36 °C. Both of these fungi attack timber exposed above ground and are also common in exposed wood in windows where temperatures can reach elevated levels. Several fungi exhibited bimodal optima. Some examples of common wood decay fungi are presented with their optimum temperature in three broad temperature groups as follows (Table 4.1)

Eslyn (1986) reported on the temperature relations and decay capabilities of 11 decay fungi (5–10 isolates per species) commonly associated with decay development in utility poles and found that growth rates varied widely within a species as well as among species. These differences suggest that caution be used when evaluating temperature optima for testing purposes when only a few isolates of a species are tested. An important conclusion from this study was that optimum decay capabilities of the fungi tested were related generally to their optimum growth rates.

Eleven isolates of blue stain fungi including seven geographic strains of *Ceratocystis coerulea* were found to have temperature and maxima ranging from 29 to 39 °C (Lindgren, 1942). Growth curves were similar in pattern to those for wood decay fungi.

Temperature and fungal wood products selectivity: Some fungi appear to be consistently associated with certain wood uses, possibly related in part to temperature optima. The predominance of *G. trabeum* and *G. saepiarium* in the exterior woodwork of buildings reflects one possible association (Eslyn, 1986). Many soft rot fungi have higher optima and temperature tolerances than basidiomycetes which may help explain their selectivity for wooden slats in cooling towers where high temperatures are common (Duncan, 1961). Soft rot fungi in chip piles demonstrate considerable tolerance to heat (Hulme and Stranks, 1976); however, some soft rot fungi commonly associated with below-ground zones of utility poles were more damaging at low than high temperatures (Morrell, 1981). It appears that generalizations concerning the temperatures of fungal groups are not as useful since even closely related taxa may vary greatly in decay optima.

Time and temperatures lethal to fungi in wood: In general, wood decay fungi are resistant to prolonged exposure to low temperatures and, conversely, readily killed by short exposure to high temperatures.

Staining fungi are particularly susceptible to high temperature, and some species are killed by prolonged storage at 35 °C. *Schizophyllum commune*, an important product rot fungus, exhibits unusual resistance to low temperatures. Fruiting bodies of this fungus, frozen at temperatures of −80 °C, will produce basidiospores within a few hours after warming.

Decay fungi often become established during storage of logs and later damage the wood products in service. Information on the time and elevated temperatures necessary to kill fungi in wood during processing such as kiln drying or preservative treatments is particularly important in large-sized logs, poles, piling, or beams where storage periods are often prolonged and heat diffusion slow. Chidester performed comprehensive tests to determine the time and temperature combinations necessary to kill fungi in wood samples previously decayed by several major wood products fungi (Chidester, 1937, 1939). Green southern pine colonized by *Meruliporia* (*Poria*) *incrassata*, *Neolentinus* (*Lentinus*) *lepideus, G. sepiarium*, *G. trabeum*, *Fomitopsis* (*Fornes*) *rosea*, or *Phlebia subserialis* was exposed to temperatures and times ranging from 40 to 100 °C and 6 minutes to 24 hours, respectively. Since wood rapidly loses strength at elevated temperatures, a principal purpose of this research was to determine the minimum temperature and time combination lethal to decay fungi (Table 4.2).

Temperatures below 65.6 °C were judged to be impractical because of the long exposure periods necessary to kill some heat-resistant fungi. *Gloeophyllum trabeum*, *G. sepiarium*, *Neolentinus lepideus* were the most

Table 4.2 Minimum times and internal wood temperatures that safely killed all fungi in greenwood during processing.

Temperature (°C)	Time (minutes)
65.6	75
76.7	30
82.2	20
93.3	10
100	5

After Chidester (1939); Chidester, M.S., 1937. Temperatures necessary to kill fungi in wood. Proc. Amer. Wood Pres. Assoc. 33, 316–324.

heat-resistant fungi, surviving 12 hours at 60 °C, 20 hours at 50.5, or 24 hours at 50 °C. As a result of these tests, 66 °C for 75 minutes was selected as the minimum time-temperature combination for wood sterilization.

Since the recommended temperature is based on the internal temperature of the wood, the wood treatment (kilning or preservative treatment) must provide an adequate surface temperature and heating time to permit heat transfer to the center of the wood products. This can be done by determining the rates of heat flow or transfer in woods of various species, sizes, and moisture conditions (Maclean: 1930, 1932, 1934, 1935).

In more recent tests, three of 23 Ascomycetes and microfungi isolated from chip piles including 2 soft rot fungi survived exposure to temperatures of 65 °C or greater for times ranging from 8 to 72 hours (Hulmes and Stranks, 1976). In agreement with the previous studies, nine Basidiomycetes (single exception *Phanerochaete chrysosporium*) were killed by exposure to temperatures equal to or greater than 50 °C. These results clearly illustrate the limits of the blanket sterilization rules. Some wood users also limit heating times because of concerns about possible heat effects on wood properties, especially strength. However, the temperatures recommended for sterilization are generally below the levels where strength would be significantly impacted.

Substrate (food sources)

As heterotrophs, fungi and most bacteria require a food source or substrate that provides three major needs.

(a) Energy from the oxidation of carbon compounds.
(b) A pool of metabolites for the synthesis of the wide range of compounds needed for growth and development (chitin, glucans, nucleotides, enzymes, proteins, lipids, etc.).
(c) Required vitamins, minor elements, CO_2, and nitrogen.

An indirect substrate requirement is the absence of various growth inhibitors and the physical access of microbial enzymes to the required substrate constituents.

Microbial carbon nutrition has become a large and complicated subject. Essentially all carbon-based compounds are subject to microbial degradation under some conditions. This subject will be discussed in greater detail in Chapter 5 on metabolism.

As a generalization, fungi are eucaryotes that appear to have evolved as scavengers of plant remains (selective for carbohydrates and low pH conditions). Bacteria, as procaryotes, are the major consumers of animal bodies (selective for proteins and neutral pH conditions). The same generalization holds for the diseases caused by bacteria and fungi. There are, however, many exceptions or crossovers where bacteria attack living plants or their remains or where fungi attack animals.

Many fungi can degrade and utilize carbohydrates including cellulose, but only the wood-inhabiting decay fungi—a few thousand species at best—are able to degrade and utilize carbohydrates in the cellulose-hemicellulose-lignin complex comprising the wood cell wall.

The monosaccharide D-glucose is utilized by essentially all fungi and is a common carbon source in many cultural media. Galactose, mannose, and fructose are used by many fungi, but appear to be initially converted to glucose-6-phosphate and then follow the same metabolic pathways as glucose in the respiration or fermentation processes.

The oligosaccharides maltose, cellobiose, and sucrose are also good carbon sources for many fungi. Malt extract is a preferred medium for many wood decay fungi, providing both glucose and vitamins.

Many fungi are able to utilize polysaccharides, such as cellulose, starches, and various hemicelluloses. The presence of small amounts of lignin as a barrier or shield around clusters of the carbohydrate components apparently drastically limits enzyme access and microbial attack to the small group of wood inhabiting micro-organisms. Some bacteria also degrade wood, but at a very slow rate.

Optimum nutrient sources vary widely for both fungi and bacteria. This variation is exploited in bacterial identification keys and was also explored for cultural identification of fungi. Determining optimum

nutrient sources and growth conditions for wood-inhabiting micro-organisms will help develop a better understanding of probable organismal successions (discussed in Chapter 11) in various stem invasions, heartrot developments, and preferential attack of various wood products.

Hydrogen ion concentration (pH)

Fungi usually have a pH for optimum growth and a minimum and maximum at which no growth occurs. In general, the optimum is skewed toward the maximum value in a manner similar to cardinal temperature requirements. In contrast to vegetative growth, sporulation and spore germination have more restrictive pH tolerances. As a substrate factor, external pH primarily affects substrate availability, rates of exo-enzymatic reactions, exo-enzyme stability, cell permeability, extracellular components and solubility of minerals and vitamins. It has little influence on the pH of cytoplasm. Hydrogen ion concentration does not always affect a single characteristic and low levels may alter exo-enzyme activity, while high levels might inhibit minor metal solubilities. These effects sometimes produce bimodal pH growth curves. In general, fungi grow best within a pH range of 3–6, while many bacteria and actinomycetes grow best at a pH of 7, but both groups often alter the pH of their substrate. Some optimum pH values for wood decay fungi are: *Heterobasidion annosum* 4.6–4.9; *Cerocorticium* (*Merulius*) *confluens* 4.0; and *G. sepiarium*, *Fomotopsis rosea*, *Serpula lacrimans* , and *Coniophora* (*Cerebella*) *puteana* at 3.0. Many plant pathogens have optimal growth within a pH range of 5–6.5. Wood-decaying Basidiomycetes have pH optima ranging from 3 to 6. Brown rotters have the lowest optima (around pH 3). Wood stain fungi are highly pH sensitive and their growth often diminishes or (ceases) as pH exceeds 5. Wood decay fungi lower the pH of wood during the decay process, and this characteristic forms the basis of several chemical indicator tests proposed for detecting incipient decays in pulpwood and utility poles (Eslyn, 1979). Woods of many species are already lightly acidic.

Chemical growth factors

Nitrogen: Fungi, like other organisms, require substantial amounts of nitrogen for the synthesis of proteins and other cell constituents or

products such as nucleoproteins, lipoproteins, enzymes, and the chitin in hyphal cell walls. Many fungi are able to use ammonia, nitrates, nitrites, and urea as sole sources of nitrogen. Ammonia is often the best nitrogen source, but the form in which it is supplied may affect media pH and, hence, growth responses. Certain L-amino acids are utilized by fungi as both carbon and nitrogen sources while others are poor nutrient sources. L-glutamine, L-asparagine, L-arginine, and L-proline are all good sole sources of nitrogen while D-amino acids are poor nitrogen sources.

Wood decay fungi can utilize many types of nitrogen but appear to most effectively utilize the amino forms found in wood (Huntgate, 1940). Wood-inhabiting fungi are unique in their ability to obtain their nitrogen needs from the very small amounts generally available in wood. The nitrogen content of wood ranges from 0.03 to 0.1%, while it ranges from 1 to 5% in herbaceous and other plant forms (Cowling and Merrill, 1966).

The capacity of decay fungi to meet nitrogen needs wholly from the low amounts available in wood is even more surprising given the prodigious number of spores released (nitrogen contents of about 3%) and the massive basidiocarps formed by some species that require substantial amounts of nitrogen-containing chitin in the hyphal walls.

Early researchers suggested that some wood-inhabiting fungi were capable of fixing atmospheric nitrogen. Critical measurements of nitrogen availability in wood and fungi at various stages of decay in pure culture studies have ruled out direct nitrogen fixation (Klingstrom and Oksbjerg, 1963), which appears to be a metabolic capability limited to procaryotes.

A series of studies on the availability and roles of nitrogen in wood deterioration (Cowling and Merrill, 1966; Merrill and Cowling, 1966a,b, c) summarized by Cowling (1970) showed that decay fungi probably conserve nitrogen by hyphal autolysis and recycling nitrogen toward hyphal tips. Nitrogen contents of the hyphal wall under low nitrogen levels are reduced prior to any reduction in exoenzyme formation. The close regulation of the cellulose enzyme system in some wood decay fungi may also serve as a nitrogen conservation method. Studies have shown that the degree of decay increases with increasing nitrogen content of the wood. Distribution of nitrogen in wood indicates reductions in amounts of nitrogen from outer to inner sapwood with the lowest amounts in the heartwood. The pith section often contains large amounts of nitrogen. The changes in nitrogen content of a growing tree stem at various developmental stages (Fig. 4.6) may account for some of the puzzling variability

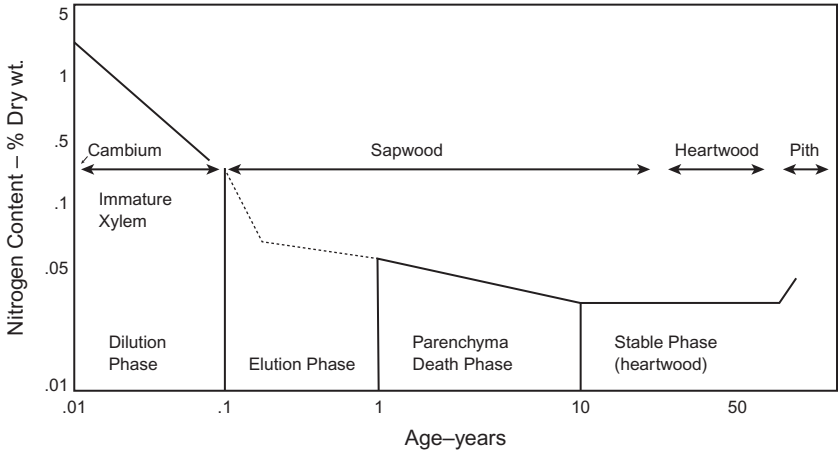


Figure 4.6 Changes in nitrogen content with radial position and during various development stages of a tree stem. *From Cowling, E.B., 1970. Nitrogen in forest trees and its role in wood deterioration. Abstracts of Uppsala Dissertations in Science, 164.*

experienced in standardized wood decay tests. Nitrogen content variations may also help to explain the variability and growth patterns of decay fungi in living stems.

While axenic tests of wood decaying fungi have ruled out nitrogen fixation, bacteria are often associated with fungi in the decay process and may play an important interactive role in some natural wood decay processes in both nitrogen cycling and fixation. Nitrogen fixation by bacteria associated with fungi in the decayed wood is apparently an important indirect nitrogen source in some ecosystems (Aho et al., 1974; Larsen et al., 1978).

Vitamins and minor metals

Vitamins and minor metals are often necessary components of enzyme systems. The vitamin and minor metal requirements for many wood decay fungi when grown in liquid media have been determined by Jennison et al. (1955). Some fungi can synthesize their needed vitamins, but other fungi such as *G. trabeum*, *H. annosum*, and *N. lepideus* are apparently thiamine deficient and require external sources. Many fungi appear to require thiamine (B,) and biotin, while pyridoxine (B_6) is less

frequently required. The destruction of thiamine in wood by heat and alkaline treatments has been reported as a potential decay control measures (Baechler, 1959), but subsequent work showed that the effects were due to factors other than thiamine reduction (Highley, 1970).

Major mineral elements required by fungi are phosphorus, potassium, magnesium, and sulfur. Trace amounts or minor elements of iron, zinc, copper, manganese, and molybdenum are also required. Many minor metals play essential roles in enzymatic reactions; for example, iron (ferric ion) and manganese have been hypothesized to play key roles on the decay by brown and white rot fungi. There is emerging evidence that many decay fungi produce siderophores that function to capture metals for use in fungal metabolism. The provision of minor metals and vitamins is often necessary in critical microbial studies using synthetic media. The reduction or elimination of these minor elements by certain sequestering chemicals or wood treatments has been proposed as a potential method for developing new wood preservatives or protective treatments.

Light

Generally, light is assumed to be harmful to vegetative growth of wood decay fungi and causes some growth reduction, probably due to the lethal effects of the ultraviolet portion of light at high intensities. However, some studies suggest that periodic exposure to light may increase decay rates (Duncan, 1967). In these studies, wood blocks close to a periodic light source developed nearly twice the weight loss of blocks furthest from the light source. No explanation for this carefully verified phenomena is known, but a slight temperature increase due to a subtle greenhouse effect cannot be overlooked.

Miscellaneous factors

Many other environmental factors may affect the growth and reproduction of wood decay fungi and their capacity to degrade wood such as osmotic concentrations, atmospheric pressure, sound vibrations,

gravitational forces, and radioactivity. Little is known about the effects of these agents on fungal growth and reproduction or the decay process, and they appear to offer interesting research avenues. It is, however, extremely difficult to perform experiments that completely isolate any of these factors.

Summary

- A review of the factors affecting fungal growth indicates that moisture level, oxygen content, and temperature are the growth requisites that can be most easily altered to adversely affect fungal growth and delay stain or decay development.
- Keeping wood dry or below the fiber saturation point (~28–30%) eliminates the possibility of effective microbial growth.
- Immersion of wood or the constant spraying of greenwood reduces the oxygen supply necessary for microbial growth and is another effective method for controlling development of stain and decay in stored logs.
- Initiation of storage piles of logs and pulpwood in the cold seasons is also an economical way to reduce biodeterioration damage.
- The use of lethal temperatures during wood processing and treatments is desirable to eliminate possible pretreatment invasions of wood-damaging fungi in standing trees, stored, or seasoning material. The addition of poisons or the use of woods containing natural toxicants is the principal method widely used to minimize growth of fungi in wood.
- Further studies on the growth requisites of fungi and interactions with other wood inhabiting organisms are certain to provide better answers to fungal product selectivity and help to identify more effective means for delaying the inevitable biodeterioration processes.

References

Agrios, G.N., 1988. Plant Pathology, third ed. Academic Press Inc, New York, p. 803.
Aho, P.E., Seidler, R.J., Evans, H.J., Raju, P.N., 1974. Distribution, enumeration, and identification of nitrogen-fixing bacteria associated with decay in living white fir trees. Phytopathology 64, 1413–1420.
Baechler, R.H., 1959. Improving wood's durability through chemical modification. For. Prod. J. 9, 166–171.

Bavendamm, W., Reichelt, H., 1938. Die Abhangigkeit des Wachstums holzersetzenden Pilz vom Wassergehalt des Nahrsubstrates. Arch. Mikrobiol. 9, 486–544.
Behr, E.A., 1972. Development of respirometry as a method for evaluating wood preservatives. For. Prod. J. 22 (4), 26–31.
Boyce, J.S., 1961. Forest Pathology, third ed. McGraw-Hill, New York, p. 572.
Brock, T.D., Smith, D.W., Madigan, M.T., 1984. Biology of Microorganisms, fourth ed. Prentice-Hall Inc, Englewood Cliffs, N.J, p. 847.
Caldwell, J., 1963. Effects of high partial pressures of oxygen on fungi. Nature 197 (4869), 772–774.
Cartwright, K.St.G., Findlay, W.P.K., 1958. Decay of Timber and its Prevention, second ed., Her Majesty's Stationery Office, London.
Chase, W.W., 1934. The composition, quantity, and physiological significance of gases in tree stems. Minn. Agric. Exp. Stat. Tech. Bull. 99, 1–51.
Chidester, M.S., 1937. Temperatures necessary to kill fungi in wood. Proc. Am. Wood Pres. Assoc. 33, 316–324.
Chidester, M.S., 1939. Further studies on temperatures necessary to kill fungi in wood. Proc. Am. Wood Pres. Assoc. 35, 319–324.
Cochran, V.W., 1958. Physiology of Fungi. John Wiley and Sons, Inc, New York, NY, p. 524, San Francisco, pp. 188.
Cooney, D.G., Emerson, R., 1964. Thermophilic Fungi. WH Freeman, San Francisco, CA.
Cowling, E.B., 1970. Nitrogen in forest trees and its role in wood deterioration. Abstracts of Uppsala Dissertations in Science, 164.
Cowling, E.B., Merrill, W., 1966. Nitrogen in wood and its role in wood deterioration. Can. J. Bot. 44, 1533–1544.
Dietz, M., Wilcox, W.W., 1997. The role of pre-infection of green Douglas-fir lumber in aboveground decay in structures in California. For. Prod. J. 47 (5), 56–60.
Duncan, C.G., 1961. Relative Aeration Requirements by Soft Rot and Basidiomycete Wood-destroying Fungi. U.S. Forest Service Forest Prod. Lab. Rept. No. 2218.
Duncan, C.G., 1967. Effect of light on the rate or decay of three wood destroying fungi. Phytopathology 57 (10), 1121–1125.
Eades, H.W., Roff, J.W., 1953. The regulation of aeration in wood: soil contact culture technique. For. Prod. J. 3 (3), 94–95.
Ellwood, E.L., Eckland, B.A., 1959. Bacterial attack on pine logs in pond storage. For. Prod. J. 9 (9), 283–292.
Eslyn, W.L., 1979. Utility pole decay. Part 3. Detection in pine by color indicators. Wood Sci. Tech. 3, 117–126.
Eslyn, W.E., 1986. Utility pole decay Part 4: Growth-temperature relations and decay capability of eleven major utility pole decay fungi. Ho1zforschung 40, 69–77.
Etheridge, D.E., 1957. A method for the study of decay resistance in wood under controlled moisture conditions. Can. J. Bot. 35, 615–618.
Fogarty, W.M., Ward, O.P., 1973. Growth and enzyme production by *Bacillus subtilis* and *Flavobacterium pectinovorum* in *Picea sitchensis*. Wood Sci. Technol. 7, 261–270.
Freyfeld, E.E., 1939. The effect of humidity on the growth of wood-destroying fungi in timber (In Russian). Abstract in. Rev. Appl. Mycol. 18, 644.
Good, H.M., Darrah, J.A., 1967. Rates of decay in wood measured by carbon dioxide production. Ann. Appl. Biol. 59, 463–472.
Griffin, D.M., 1977. Water potential and wood decay fungi. Ann. Rev. Phytopathol. 15, 319–329.
Griffin, D.H., 1981. Fungal Physiology. John Wiley &Sons, 383 pp.
Halabisky, D.O., Ifju, G., 1968. Use of respirometry for fast and accurate evaluation of wood preservatives. Am. Wood Preserv. Assoc. 64, 215–223.

Hartley, C., Davidson, R.W., Crandall, B.S., 1961. Wetwood, Bacteria, and Increased pH in Trees. U.S. For. Serv., For. Prod. Lab., Rep. No. 2215.
Higgins, C.N., 1957. The equilibrium moisture content-relative humidity relationship of selected native and foreign woods. Forest Prod. J. 7, 371–377.
Highley, T., 1970. Decay resistance of four wood species treated to destroy thiamine. Phytopathology 60 (11), 1660–1661.
Highley, T.L., Scheffer, T.C., 1970. A need for modifying the soil-block test for testing natural resistance to white rot. Mater. Org. Beih. 5, 281–292.
Highley, T.L., Bar-Lev, S.S., Kirk, T.K., Larsen, M.J., 1983. Influence of O_2 and CO_2 on wood decay by heartrot and saprot fungi. Phytopathology 73, 630–633.
Hulme, M.A., Stranks, D.W., 1976. Heat tolerances of fungi inhabiting chip piles. Wood Sci. 8, 237–241.
Humphrey, C.J., Siggers, P.V., 1933. Temperature relations of wood destroying fungi. J. Agric. Res. 47, 997–1008.
Huntgate, R.E., 1940. Nitrogen content of sound and decayed coniferous woods and its relation to loss in weight during decay. Bot. Gaz. 102, 382–392.
Jennison, M.W., Newcomb, M.D., Henderson, R., 1955. Physiology of wood rotting Basidiomycetes. 1. Growth and nutrition in submerged culture in synthetic media. Mycologia 47 (3), 275–304.
Jensen, J.F., 1967. Oxygen and carbon dioxide affect the growth of wood-decaying fungi. For. Sci. 13 (4), 384–389.
Jensen, F.F., 1969a. Effect of constant and fluctuating temperature on growth of four wood-decaying fungi. Phytopathology 59, 645–647.
Jensen, F.K., 1969b. Oxygen and carbon dioxide concentrations in sound and decaying red oak trees. For. Sci. 15, 246–251.
Klingstrom, A., Oksbjerg, E., 1963. Nitrogen source in fungal decomposition of wood. Nature 197, 97.
Larsen, M.J., Jurgensen, M.F., Harvey, A.E., 1978. N_2 fixation associated with wood decayed by some common fungi in western Montana. Can. J. For. Res. 8, 341–345.
Levy, J.F., 1965. The soft rot fungi -their mode of action and significance in the degradation of wood. Adv. Bot. Res. 2, 323–357.
Lilly, V.G., Barnett, H.L., 1951. Physiology of the Fungi. McGraw-Hill Book Co., Inc., New York, NY, p. 464.
Lindgren, R.M., 1942. Temperature, Moisture, and Penetration Studies of Wood Staining *Ceratostomellae* in Relation to their Control. U.S.D.A. Tech. Bull. 807.
Ljungdahl, L.G., Ericksson, K.-E., 1986. Ecology of microbial cellulose degradation. In: Marshall, K.C. (Ed.), Advances In Microbial Ecology, vol. 8. Plenum Press, New York, pp. 277–299.
Maclean, J.D., 1930. Studies of heat conduction in wood. Results of steaming green round southern-pine timbers. Proc. Am. Wood Pres. Assoc. 26, 197–219.
Maclean, J.D., 1932. Studies of heat conduction in wood. Part II. Results of steaming green round southern pine timbers. Proc. Am. Wood Pres. Assoc. 28, 303–330.
Maclean, J.D., 1934. Temperatures in green southern pine timbers after various steaming periods. Proc. Am. Wood Pres. Assoc. 30, 355–373.
Maclean, J.D., 1935. Temperature and moisture changes in Coast Douglas-fir. Proc. Am. Wood Pres. Assoc. 31, 77–103.
Merrill, W., Cowling, E.B., 1966a. Role of nitrogen in wood deterioration. Amounts and distribution of nitrogen in tree stems. Can. J. Bot. 44, 15551580.
Merrill, W., Cowling, E.B., 1966b. Role of nitrogen in wood deterioration. IV. Relationship of natural variation in nitrogen content of wood to its susceptibility to decay. Phytopathology 56, 1324–1325.

Merrill, W., Cowling, E.B., 1966c. Role of nitrogen in wood deterioration. Amount of distribution of nitrogen in fungi. Phytopathology 56, 1083–1090.
Morrell, J.J., 1981. Soft Rot Fungi: Their Growth Requisites and Effects on Wood (Ph.D. thesis). State University of New York, College of Environmental Science and Forestry, Syracuse, N.Y., p. 161.
Niederpruem, D.J., 1963. Role of carbon dioxide in the control of fruiting of *Schizophyllum commune*. J. Bact. 85, 1300–1308.
Reid, I.D., Seifert, K.A., 1983. Effect of one atmosphere of O_2 on growth, respiration, and lignin degradation by white-rot fungi. Can. J. Bot. 60, 252–260.
Scheffer, T.C., 1973. Microbiological degradation and the causal organisms. In: Nicholas, D.O. (Ed.), Wood Deterioration and its Prevention by Preservative Treatment, Volume 1: Degradation and Protection of Wood. Syracuse University Press, p. 380.
Scheffer, T.C., 1986. O_2 requirements for growth and survival of wood-decaying and sapwood-staining fungi. Can. J. Bot. 64, 1957–1963.
Scheffer, T.C., Livingston, B.E., 1937. Relation of oxygen pressure and temperature to growth and carbon dioxide production in the fungus *Polystictus versicolor*. Am. J. Bot. 24, 109–119.
Scheffer, T.C., Chidester, M.S., 1948. Survival of decay and blue stain fungi in air-dry wood. South. Lumberm. 177, 110–112.
Schmitz, H., Kaufert, F., 1938. Studies in wood decay. VII. How long can wood-destroying fungi endure immersion in water? Proc. Am. Wood Pres. Assoc. 34, 83–87.
Smith, R.S., 1975. Automatic respiration analysis of the fungitoxic potential of wood preservatives including an oxathiin. For. Prod. J. 25 (1), 48–53.
Skaar, C., 1972. Water in Wood. Syracuse University Press, Syracuse, New York.
Snell, W.H., Howard, N.O., Lamb, M.V., 1925. The relation of moisture contents of wood to its decay. Science 62, 377–379.
Stanier, R.Y., Dondoraff, M., Adelberg, L.A., 1963. The Microbial World. fifth ed., pp. 753, Prentice-Hall, New York.
Tabak, H.H., Cooke, W.B., 1968. The effects of gaseous environments on the growth and metabolism of fungi. Bot. Rev. 34, 126–252.
Taber, W.A., 1966. Morphogenesis in basidiomycetes. In: Ainsworth, G.C., Sussman, A.S. (Eds.), Fungi, vol. 2. Academic Press, New York, pp. 387–412.
Thacker, D.G., Good, H.M., 1952. The composition of air in trunks of sugar maple in relation to decay. Can. J. Bot. 30, 475–485.
Toole, L.R., 1972. Oxygen utilization and weight loss associated with decay by wood-decaying fungi. Wood Sci. 6 (1), 55–60.
Van der Kamp, B.J., Gokhale, A.A., Smith, R.S., 1979. Decay resistance owing to near-anaerobic conditions in black cottonwood wetwood. Can. J. For. Res. 9 (1), 39–44.
Vogels, G.D., 1979. The global cycle of methane. Antonie Leeuwenhoek. J. Microbiol. Seral. 45, 347–352.
Worrall, J.J., Parmeter Jr., J.R., 1983. Inhibition of wood decay fungi by wetwood of white fir. Phytopathology 73, 1140–1145.
Zeikus, U.G., 1974. Methane formation in living trees: a microbial origin. Science 184, 1181–1183.

Further reading

Boutelji, J.B., Kiessling, H., 1964. On water-stored timber and its decay by fungi and bacteria. Arkiv. Mikrobiol. 49, 305–314.
Cartwright, K.St.G., Findlay, W.P.K., 1934. Studies on the physiology of wood -destroying fungi. II. Temperature and rate of growth. Ann. Bot. (London) 48, 481–495.

Knutson, D.M., 1973. The bacteria in sapwood, wetwood, and heartwood of trembling aspen (*Populus tremuloides*). Can. J. Bot. 51, 498–500.
Shigo, A.L., Hillis, W.E., 1973. Heartwood, discolored wood, and microorganisms in living trees. Ann. Rev. Phytopath. 11, 197–221.
Snell, W.H., 1929. The relation of the moisture contents of wood to its decay. III. Am. J. Bot. 16, 543–546.
Wells, J.M., Uota, M., 1970. Germination and growth of five fungi in low oxygen and carbon-dioxide atmospheres. Phytopathology 60, 50–53.
Zabel, R.A., Lombard, F.F., Kenderes, A.M., 1980. Fungi associated with decay in treated Douglas-fir transmission poles in northeastern United States. For. Prod. J. 30 (4), 51–56.

CHAPTER FIVE

Fungal metabolism in relation to wood decay

Contents

A brief review of selected aspects of fungal metabolism is necessary to set the stage for an understanding the enzymatic nature of wood decay by microorganisms and why preservatives are toxic to fungi. Emphasis is placed on fungal metabolism since it is similar to that of the principal wood-damaging bacteria.

Metabolism is a complex topic and broadly includes all the chemical reactions occurring in living systems. General reviews of the energy release and synthesis aspects of cell and organism functioning are available in many modern textbooks on biology and microbiology (Raven et al., 1986; Madigan et al., 2018; Wiley et al., 2008; Cano and Colome, 1986; Brock et al., 1988). More comprehensive coverage is available in textbooks on biochemistry (Stryer, 1981) or fungus physiology (Griffin, 1981; Garraway and Evans, 1984). These sources provide references to the many specialized topics comprising the vast literature that has accumulated on microbial metabolism. In this chapter, emphasis will be placed on digestion and energy release from organic carbon compounds since these two processes play key roles in understanding the decay process.

Wood Microbiology.
DOI: https://doi.org/10.1016/B978-0-12-819465-2.00005-X

Biochemistry is the principal discipline dealing with the study of the chemical processes occurring in living systems (metabolism).

In the *1800s*, many scientists studying the chemical functioning of cells believed in a mysterious and unique property of living materials that they termed "vitalism." As the knowledge of cell systems improved, the concept of vitalism declined and scientists accepted that all metabolic reactions were explainable as chemical reactions that follow chemical laws.

Metabolic reactions can be classified as catabolic reactions that release energy or anabolic reactions that require an energy source. Respiration represents a series of catabolic reactions. Photosynthesis is a classic anabolic reaction whereby light energy (photons) is transformed and stored in carbon compounds as chemical energy and becomes the major energy source for most living organisms.

Some examples of major metabolic activities occurring, often simultaneously, in microorganisms are digestion (an external process in fungi leading to substrate degradation), absorption, respiration, synthesis of cell components, growth, storage of food reserves, and reproduction. In terms of the specific growth requisites, discussed in Chapter 4, metabolism includes the synthesis of digestive enzymes; their transport and release through cell membranes and walls; the external digestion of wood; the absorption of H_2O, O_2 and the solubilized wood components; the release of CO_2; the synthesis of needed vitamins, and the absorption of minor metals. All of these reactions occur within a relatively narrow range of temperature and pH conditions.

Energy sources, transfer, and storage

Fungi and most bacteria are heterotrophs that require external organic energy sources. They obtain this needed energy from the respiration of organic compounds as chemical bonds are broken and reformed in chemical reactions. In this process, electrons drop to a lower energy level and generally are transferred to another element or compound. In many metabolic reactions, the chemical energy associated with these electron changes is temporarily transferred to special compounds termed electron carriers. Electron carriers play a key role in the timely transfer, storage, and use of chemical energy in many metabolic reactions. The free energy change (ΔG) involved in a chemical reaction between reactants and their products determines whether energy will be released (exergonic) or

required (endergonic). The reaction products in respiration are at a lower energy state and energy is released ($-\Delta G$).

Oxidation -reduction reactions: In chemical reactions, the loss of electrons is termed oxidation and is associated with energy release. Oxidation of organic materials (carbon compounds) often also involves loss of protons (hydrogen ions). The gain of electrons is termed reduction and requires energy. In organic compounds, reduction also often involves the simultaneous gain of protons. In reduction, the electrons involved in the chemical bonds of the products are raised to higher energy levels. The transfer of electrons between atoms, elements, or compounds is always coupled. One reactant in the reaction gains electrons while the other loses them in oxidation-reduction reactions (the term is usually abbreviated to redox reactions). Redox reactions are emphasized in this section because of their importance in respiration. Carbohydrates are good electron donors that are easily oxidized in cell respiration and serve as a prime energy source for many fungi. Atmospheric oxygen (O_2) is an excellent electron acceptor, relative to most other elements or compounds, explaining its universal role in aerobic respiration and the term oxidation.

Electron carriers and high-energy compounds: Several complex molecules function as electron carriers between donor and acceptor compounds in metabolic reactions. Some carriers appear to facilitate enzyme function and are also termed coenzymes. Coenzymes may serve as carriers of small molecules from one enzyme to another. Most are synthesized from vitamins. Electron carriers and high-energy compounds temporarily store and transport chemical energy released during respiration so it can be available when needed for critical cell functions requiring energy such as enzyme synthesis, active solute transport, or growth.

Adenosine triphosphate (ATP) serves as the major energy provider for most living organisms. The structure of ATP consists of ribose, adenine and ester linkages connecting three phosphates (Fig. 5.1). Energy is stored in the last two phospho-diester bonds. Successive loss of these groups in phosphorylation reactions with electron transfer to other compounds forms first the related compounds ADP (adenosine diphosphate) and then AMP (adenosine monophosphate). Phosphorylation reactions, whereby a phosphate group is added enzymatically to another compound, are one of the key energy transfer mechanisms in respiration.

An important electron carrier in respiration is nicotinamide adenine dinucleotide (NAD) and a phosphorylated form NADP (Fig. 5.1). These

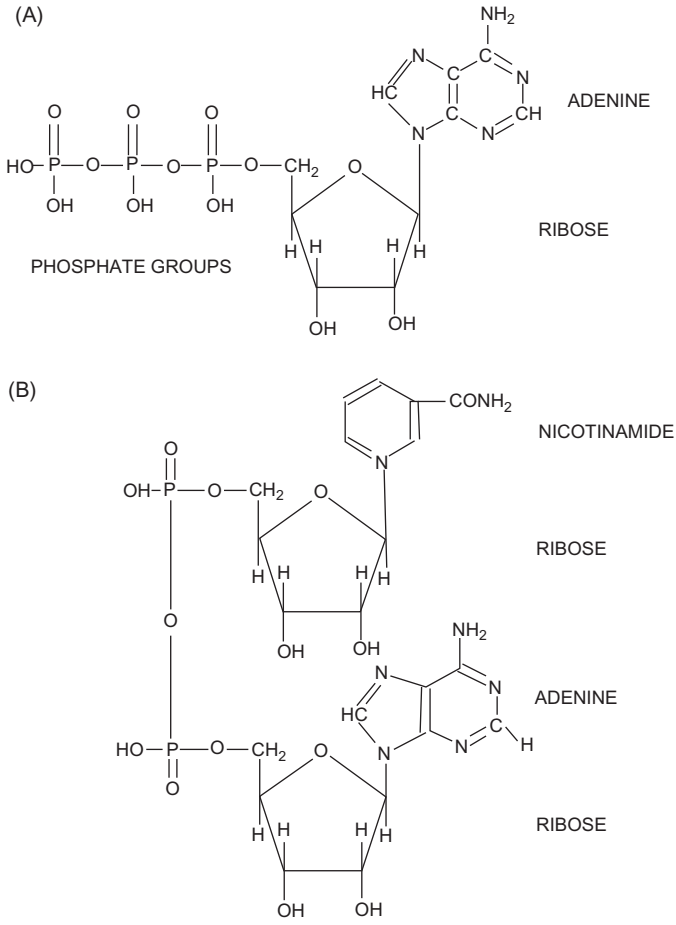


Figure 5.1 The structural formulas of two major compounds involved in energy transfers in respiration showing their constituents and close relationship. (A) Adenosine triphosphate (ATP) has two high energy phosphate bonds, which are major energy sources for many metabolic reactions, (B) Nicotinamide adenine dinucleotide (NAD) is an important electron carrier.

two compounds also transport protons. NAD is a major transporter of protons during respiration, while NADP plays a similar role in many synthesis reactions.

Enzymes

Catalysts in chemical reactions are substances that accelerate reaction rates without being permanently altered themselves. Biocatalysts or enzymes are involved in most chemical reactions in living systems.

Structure and mode of action: Enzymes are proteins consisting of one or more polypeptide chains folded in a complex tertiary structure connected by disulfide linkages. Binding sites control enzyme specificity and a unique topography corresponding to the reactive zones of the substrate determines the active site. Recent studies have substantially improved our understanding of the structure and function of enzymes, but the "lock-and-key" model proposed by Emil Fisher half a century ago still provides a useful way to illustrate the close structural relationship between enzyme and substrate (Fig. 5.2A). Critical regulators or cofactors that alter the enzyme structure so the active site meshes more precisely with a portion

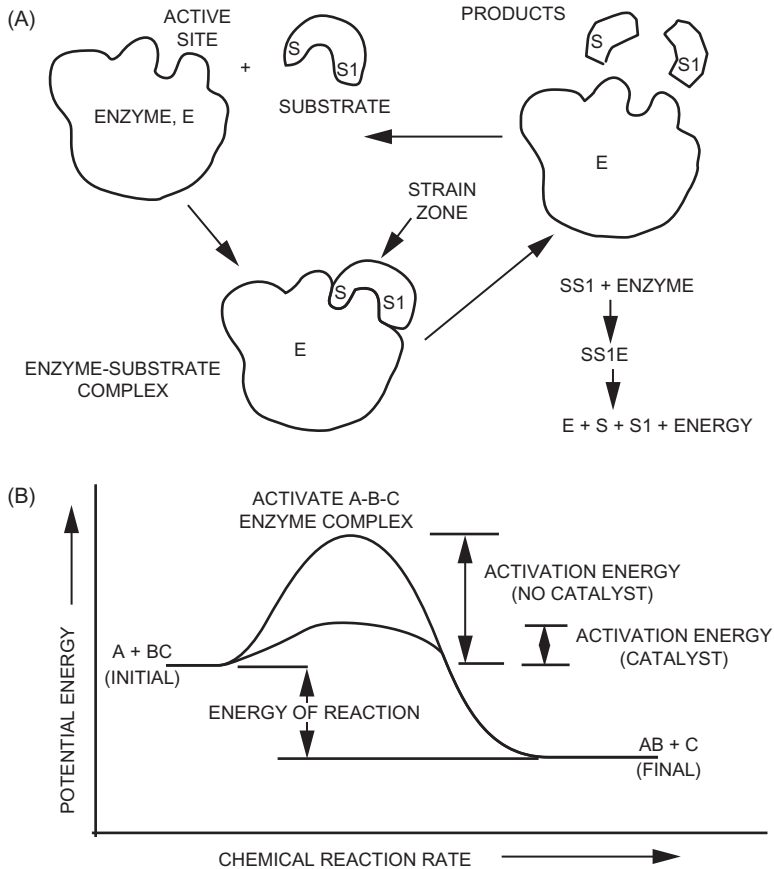


Figure 5.2 (A) The traditional lock-key model suggesting the mechanism of enzyme action. (B) Representation of the energy required to start the reaction with and without a catalyst.

of the substrate molecule are often associated with enzyme. Copper or iron are two common enzyme cofactors.

The energy required to break a chemical bond so a reaction can proceed is the energy of activation. A loose analogy may be the push necessary to start a large boulder rolling down a steep hill causing a large amount of potential energy to be released as kinetic energy at the bottom. Enzymes reduce the activation energy dramatically, thus increasing the rate of the reaction (Fig. 5.2B). The number of substrate molecules catalyzed per second per enzyme molecule has been reported to be as high as 10^5 (also known as turnover) for some enzymes. Enzymes also mediate many reactions at room temperatures and near pH neutrality that otherwise could occur only under extreme temperature or pH conditions that would be detrimental to the organism. Enzymes are highly specific to the substrate used and the reactions that they catalyze. They are easily activated, modified, or inactivated by a variety of cell controls. The range of reactions carried out by different types of enzymes is broad, and collectively they are very versatile.

Enzymes also enable organisms to transform energy from one form to another. Autotrophic plants can convert light energy (photons) into chemical bond energy, providing a basic energy pool that sustains heterotrophic life forms.

While enzymes are highly versatile, they cannot alter the direction of a reaction, which is determined by other factors or the equilibrium, but can only greatly accelerate the reaction rate. Many enzymes are fragile and inactivated by temperatures above 50 °C, changes in pH, or the presence of heavy metal cations such as Cu, Hg, and Pb.

The mechanisms by which enzymes function are still not fully understood, although major advances in protein chemistry, especially those related to three-dimensional modeling have greatly expanded our understanding of these critical cell constituents. The enzyme binds to the substrate forming an intimate union with it called the enzyme-substrate complex. Presumably, certain chemical groups on the substrate are stressed by the electrostatic forces involved and bonds are strained and broken. The active sites on the enzyme are presumably where this molecular distortion occurs (Fig. 5.2A). Prosthetic groups attached to the enzyme are sometimes necessary for activation. Vitamins such as thiamine may play this role. Other compounds associated, but not attached, that are necessary for the reaction to proceed are termed co-factors. Magnesium cations, for example, are a necessary co-factor for phosphorylation reactions catalyzed

by kinases. Important factors affecting enzyme reaction rates are enzyme and substrate concentrations, pH, and temperature.

Types and classifications of enzymes: The first few enzymes discovered were unique and assigned trivial names such as diastasis, pepsin, or ferment. In 1878, Kuhn proposed that the suffix "ase" be added to the substrate modified. This practice was quickly adopted and is still in use for the general or trivial name of many enzymes or enzyme complexes, e.g., cellulases and hemicellulases. By the 1950, many enzymes had been discovered and considerable confusion developed with similar names used for different enzymes and the use of different nomenclature schemes. In 1955, the International Union of Biochemistry established a Commission on Enzymes to develop a uniform classification system and standardized nomenclature that is still in use today. The principal grouping was based on the type of chemical reaction involved. The six main groups are as follows:

Oxido-reductases – oxidation-reduction reactions involving basically the loss and gain of electrons.

Transferases – transfer of radicals such as amino, methyl, or acetyl from one compound to another.

Hydrolases – compounds are separated at the oxide bridge and often into monomers or dimers by the addition of water.

Lyases – removal of groups leaving a double bond or the reverse: to add groups to a double bond

Isomerases – catalyzing an internal rearrangement in a molecule to form an isomer.

Ligases (synthetases) – catalyze the joining of molecules to form a new compound and usually requiring chemical energy from ATP.

Hydrolases and oxido-reductases are the major enzyme types involved in decay and cell respiration. Some other common descriptive groupings of enzymes in general use are the following:

Site of action: Endocellular and exocellular enzymes act inside or outside the cell, respectively (hydrolases are exocellular while most oxido-reductases are endocellular)

Constitutive vs Induced: Whether the enzymes are always present in the cell (constitutive) or appear only in the presence of a specific substrate (induced).

Terminology using name of substrate and the reaction: Terms combining the name of the substrate altered and the chemical reaction involved such as glucose oxidase, pyruvate decarboxylase, and glyceraldehyde

3-phosphate dehydrogenase. Knowledge about enzymes and their function has increased enormously in the past few decades. Where a single volume summarized enzymology in the early 1960s (Dixon and Webb, 1964), nearly 600 volumes in a series currently present the methods of enzymology and over 40 volumes now comprise a definitive treatment titled "The Enzymes".

Digestion and hydrolases

The major cell-wall constituents of wood are all large macropolymers that are insoluble in water. The principal source of nutrition for most wood inhabiting microorganisms are the carbohydrates present in cell walls and storage tissues. Cellulose, hemicelluloses, and pectins are the major polymeric carbohydrates, comprising about 65–75% of the cell wall. Lignin, an aromatic heteropolymer, makes up most of the remaining cell-wall substance. Small amounts of lipids and proteins are also used by some bacteria and fungi. Starches and lipids are the principal nutrition sources in the storage tissues. A wide range of compounds are present in the wood extractives in relatively small amounts. Lignin forms a shield or barrier around the cellulose and hemicellulose microfibrils in the cell wall that must be altered or removed before the microfibrils can be enzymatically digested. For many years, lignin was seen as a barrier that the fungus had to remove or modify to gain access to the cellulose; however, the fact that some fungi selectively remove lignin has altered this assumption. We have now begun to recognize that many fungi alter, but do not utilize lignin as a carbon source.

Cellulose, hemicelluloses and lignin are complex polymers that must be reduced to small diffusible units before they can be absorbed and utilized. This process of external digestion is carried out largely by hydrolases and oxidases. This is an exocellular process that requires that the fungal hypha or bacterial cell be in a film of water for the diffusion and transport of the breakdown products. The enzymes attacking the various substrates also require either a water film to diffuse to the active sites on the substrate molecules or direct contact between the hyphal surface or bacterial walls and the wood cell wall. Hydrolases are enzymes that break the polymer into smaller units by the addition of H_2O (proton and hydroxyl). Simplified hydrolytic (digestive) reactions are shown for a sugar, polypeptide, and lipid in Fig. 5.3.

In general, large polymers are attacked by several specific hydrolytic enzymes. First, the polymer complex in the cell wall is exposed from the

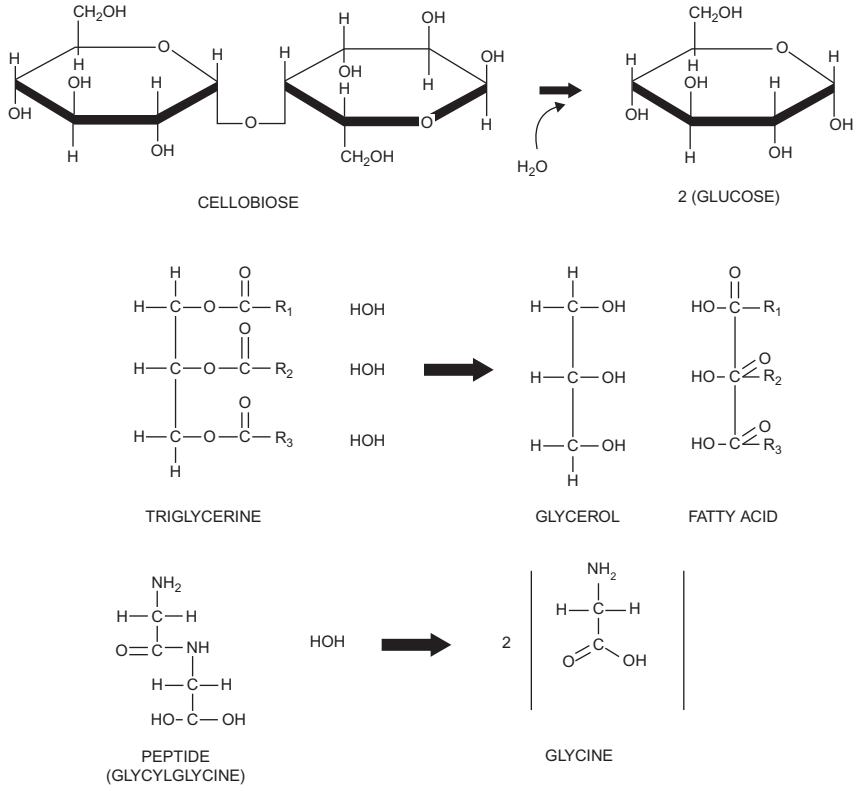


Figure 5.3 Hydrolytic reactions involving an oligosaccharide, a lipid, and a peptide.

others and simplified by chain separations and removal of side chains. The exact nature of these initial steps remains poorly defined, but non-enzymatic steps that create non-specific free-radicals are likely involved. Once the polymer is exposed, enzymes known as exohydrolases attack the ends of the chains usually releasing monomers or dimers. Other enzymes known as endohydrolases randomly attack the chains and in effect form many more ends that are accessible to exohydrolases, greatly accelerating the decomposition process. This brief review of digestion is limited to cellulases, hemicellulases, and ligninases because of their central role in wood decay. This topic is considered in more detail in a later chapter on wood decay.

Cellulose: This polymer is decomposed by a multi-enzyme complex known generally as cellulase that consists of at least 3 enzymes that may vary with fungi (decay type) and type of substrate. Endo-1,4 β-D glucanases act randomly on exposed molecule surfaces, randomly cleaving the

cellulose polymer. The exposed ends are attacked by exo-l,4 β glucanases (cellobiohydrolase) forming glucose or cellobiose. A 1,4-β-glucosidase then converts the cellobiose into glucose that can be absorbed by the microorganism. In some fungi, oxidative enzymes may also be involved.

Hemicelluloses: These polymers are also decomposed by a multi-enzyme complex. Hemicelluloses are complex heteropolymers containing xylose, galactose, glucose, and mannose with acetyl, methyl, and short oligosaccharide units as side chains. Many of the hemicellulase enzymes are poorly understood although efforts to use hemicelluloses in woody tissues for the production of ethanol have encouraged more research. The hemicellulases that attack the polymer backbones are presumed to be β-D galactanases, β-D-mannanases, and β-D-xylanases. In a pattern similar to cellulose, exo-enzymes probably remove oligosaccharide units from the ends while endo-enzymes attack the polymers randomly. Separate enzymes may be necessary to remove the side chains, but the coordination between polymer decomposition and side chain attack is uncertain. Exo-glycosidases then attack the oligosaccharide residues to produce xylose, mannose, glucose, and galactose units that can be absorbed and utilized.

Lignin: Lignin biodegradation is the unique capability that distinguishes wood-decay fungi from most other microorganisms. Enzymatic breakdown of lignin is an oxidative process in sharp contrast to the hydrolytic enzymes that predominate in the degradation of cellulose and hemicelluloses. Lignin is included in this section primarily because its enzymatic removal or alteration is necessary for carbohydrate digestion (degradation) to proceed. A key part of the breakdown is the oxidative separation of carbon to carbon bonds and ether linkages among certain phenyl propane units by a lignin peroxidase. An interesting feature of this enzymatic reaction is the requirement for an extracellular source of H_2O_2. The enzymatic breakdown of lignin will be covered in some detail in Chapter 9 on the chemical aspects of wood decay.

Absorption of digestion products

Digestion begins and ends at the plasmalemma (cytoplasmic membrane) just within the cell wall. It begins with the release of exoenzymes by exocytosis and diffusion into the surrounding liquid medium to the substrate. Digestion ends when the soluble products of digestion diffuse

back to the hyphal wall. These polymer residues of the decay process are primarily simple sugars (glucose, xylose, galactose, etc.) which are small enough to readily pass through the cell wall, but not the plasmalemma which is semi-permeable and highly selective. There is some uncertainty as to the membrane transport mechanism used to move sugars into the hyphae. In yeasts and some molds, both facilitated diffusion and active transport are reported, depending on the type of simple sugar and fungal species. Solute concentrations during wood decay are very low and active transport is the probable method. In this process, the sugar is phosphorylated and transported by a protein carrier (permease) across the plasmalemma into the cytoplasm, where respiration can begin. This process requires energy that is obtained generally from the high energy compound, ATP. This process provides a rich array of metabolites for further synthesis.

Aerobic respiration

Most fungi undergo aerobic respiration in the presence of atmospheric oxygen (O_2) by synthesizing ATP (storage of chemical energy) from the oxidation of glucose. ATP is the principal energy source for a wide range of energy-dependent metabolic activities. Aerobic respiration has three major phases, glycolysis, the citric acid cycle, and the electron-transport chain (Fig. 5.4). The process begins with glucose and ends with CO_2, H_2O, and chemical energy stored in ATP. The process also provides a rich array of metabolites for synthesis.

Glycolysis: Glycolysis takes place in the cytoplasm. This phase of respiration begins with the enzymatic phosphorylation of glucose at the 1 and 6 carbons, successively, using ATP. These reactions are carried out by kinase enzymes with $Mg++$ as a critical co-factor. Thus, magnesium is a critical growth requisite for some fungi. The phosphorylated glucose is then changed into fructose that is broken into two triose sugars. A series of enzymatic oxidations and molecular rearrangements converts the triose sugar glyceraldehyde-3-phosphate to pyruvic acid. In this process, 2 moles of ATP are gained per mole of glucose and one mole of NAD is reduced (protons acquired). Pyruvic acid is a key intermediate in several biochemical pathways and has been termed a "biochemical turntable" in cell metabolism. It is interesting to note that the oxidation/reduction reactions

releasing chemical energy for transfer to ATP in glycolysis occur within the glucose molecule. Some portions become highly oxidized, forming CO_2, while other portions become further reduced to methyl groups in the acetyl radical. In aerobic respiration, the pyruvate is further oxidized and then decarboxylated, forming CO_2 and an acetyl radical (CH_3CO-). This process produces two molecules of reduced NAD (NADH +) per mole of glucose.

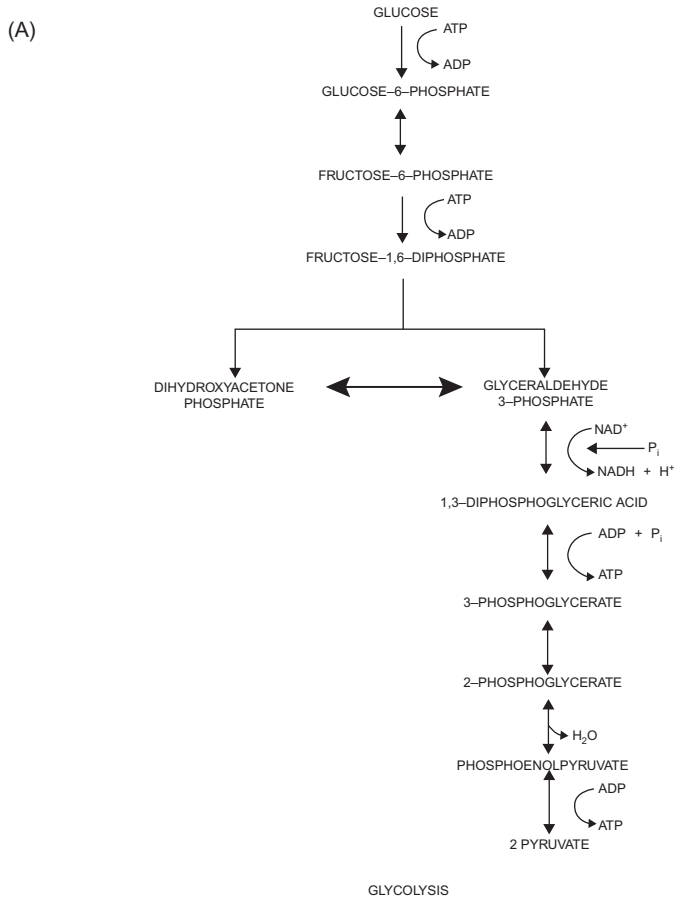


Figure 5.4 The three major biochemical pathways in aerobic respiration. (A) In glycolysis, glucose is oxidized to pyruvate. (B) In the citric acid cycle, the electron carriers NAD and FAD are reduced and donate electrons to the electron transport system. (C) In the electron transport system the electrons passed through the various cytochromes provide the energy to synthesize ATP. These cycles produce 38 molecules of ATP per molecule of glucose oxidized to CO_2 and H_2O. *Figures courtesy R. Cano and Colome (1986) and permission West Publishing Co.*

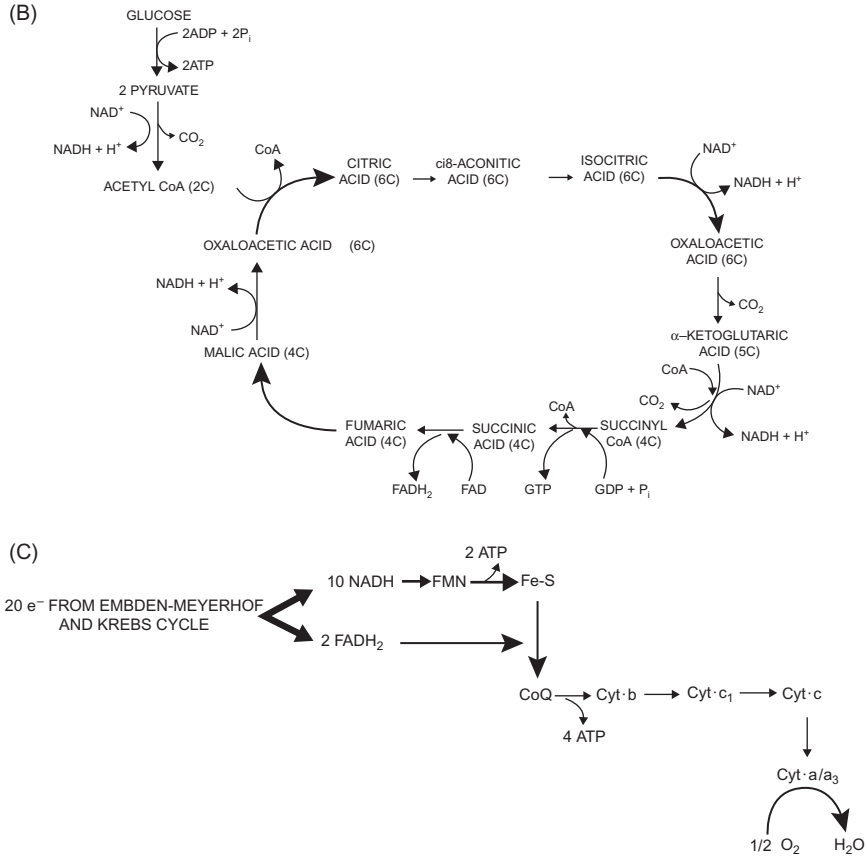


Figure 5.4 (Continued)

Citric acid cycle: The citric acid or Krebs cycle takes place in the inner matrix of the cristae in the mitochondria. In this phase of aerobic respiration, a coenzyme (coenzyme A) combines with an acetyl group temporarily forming an acetyl coenzyme A complex. The acetyl group in this complex then combines with oxaloacetate (a 4-carbon acid) to form citrate (a 6-carbon acid). In a further series of oxidations and molecular rearrangements, 2 moles of ATP are formed and 6 moles of NAD reduced per mole of glucose. Two moles of another electron carrier, FAD, are reduced per mole of glucose. The oxalocetate is regenerated and available for further reactions with the acetyl radical.

Electron transport chain: At this stage of respiration, the electrons and protons released during the oxidation of glucose are transported by the

electron carriers NAD and FAD to a series of electron carriers embedded in walls of the inner mitochondrial membranes. These electron carriers differ in the energy levels at which the electrons are held. The electrons move down the sequence of carriers and at the end are accepted by oxygen. Oxygen then combines with the available protons to form water. The major electron carriers are the cytochromes, iron-sulfur proteins, and quinones. The quinones are able to transport protons across the mitochondrial membranes. The proton ($H+$) gradient or proton-motive force established across these membranes is another energy source for some metabolic reactions and is believed to play a role in energy transfer during oxidative phosphorylation.

The energy released at various levels in this sequence of electron energy levels in the presence of the enzyme ATP synthetase regenerates ADP to ATP in a process termed oxidative phosphorylation. Most of the energy obtained from glucose respiration is derived from the electron and proton transfers that occur in this final stage of aerobic respiration. This phase of aerobic respiration occurs in an array of enzymes and enzyme carriers on the surface of the cristae in the mitochondria.

Pentose shunt: Another important aerobic respiratory pathway for the utilization of glucose is the hexose-monophosphate pathway or pentose shunt. The pentose shunt provides the 5-carbon sugars, ribose and deoxyribose needed for the synthesis of DNA, RNA, ATP, CoA, and $NAD+$. It also is an energy yielding process and reduces NADP to NADPH, which serves as the major proton and electron donor in many synthetic reactions. In this pathway, glucose 6-phosphate is oxidatively decarboxylated in several steps to form ribose-5-phosphate and CO_2. In these reactions, NADP is reduced to $NADPH+$ and utilized as a readily available source of reducing power for many biosynthetic reactions. NADP can also be oxidized in the mitochondria and its chemical energy transferred to ATP. Later stages of this pathway form intermediates that are precursors for the synthesis of the aromatic amino acids or glyceraldehyde 3-phosphate that connects the pathway with glycolysis. The pentose shunt is present in most organisms and is an interesting example of a pathway playing an important role both in catabolism and anabolism. This is also an excellent example of how many of the major metabolic pathways are connected by common intermediate compounds creating alternate routes and regulation possibilities.

Fermentation

Simple sugars such as glucose and some other carbon compounds can be utilized by some bacteria and fungi such as the yeasts in the absence of oxygen in the process termed fermentation. There is no external electron acceptor in this type of metabolism and the oxidation-reduction reaction that releases energy takes place between parts of the same substrate molecule. As such, fermentation is an inefficient process that yields only a portion of the potential energy available in a compound. Fermentation is believed to be a primitive biochemical pathway that originated in microorganisms prior to the accumulation of oxygen in the atmosphere from photosynthesis. The glycolysis phase of fermentation, however, is still retained as a critical first step in aerobic respiration. The biochemical pathway for glucose fermentation is the same as in aerobic respiration until the formation of pyruvic acid, but there are differences in subsequent steps depending on the fermenting organisms. Some yeasts enzymatically decarboxylate pyruvate, releasing CO_2, and acetaldehyde is subsequently reduced by NADH to form ethanol. The fermentation process generates only a net of 2 moles of ATP per mole of glucose. Other compounds that may be produced fermentatively are alcohols such as butanol, organic acids such as acetic acid, and ketones such as acetone. For some bacteria and animals, the final product of glucose fermentation under anaerobic conditions is lactic acid.

Anaerobic respiration

Anaerobic respiration is important in the decomposition of cellulose and other carbon compounds that accumulate in oxygen deficient environments, e.g., animal rumens, wetwood in trees, and intestinal tracts (herbivores and termites). Anaerobic respiration can be defined as a nutritional biochemical pathway where compounds other than oxygen are used as electron acceptors.

The denitrifying bacteria that reduce NO_3 to NO_2 or nitrogen in their decomposition of organic materials are an interesting example of anaerobic respiration and an important source of fertility loss in some soils. The methanogenic bacteria are of special importance in decomposing cellulose

in anaerobic environments such as animal rumens, bogs, and water-logged sediments. In these fermentations, methane (CH_4) gas is most commonly formed from the reduction of either CO_2 or acetic acid (CH_3COOH). Cellulose breakdown is actually carried out by a consortium of anaerobic bacteria and understanding their relative roles could be useful in bioconversion schemes.

Sulfate (SO_4) reduction is also carried out by a variety of anaerobic bacteria which utilize organic acids and alcohols as electron donors. Some bacteria also can use hydrogen as the donor. Sulfate reducing bacteria often accumulate in environments which become anaerobic as a result of active microbial decomposition. There are two major groups that reduce sulfate (SO_4) to a gas (hydrogen sulfide) or sulfides.

Anaerobic respiration also occurs in the inner stem zones of trees when a condition known as "wetwood" occurs. In this condition, considerable amounts of methane are formed and an exudate known as "slime flux" develops on the trunk surface from cracks in the stem.

Enzyme inhibitors

The extreme complexity of metabolism and the many processes involved provide numerous potential points that can be chemically altered or blocked to limit decay.

Many chemical compounds are known to inactivate enzyme activity. Selective enzyme inhibitors or poisons represent potential fungicides or preservatives where other properties such as cost, ease of handling and treatment, and safety to other life forms, are acceptable. Many heavy metals such as $Hg+^{2}$, $Pb+^{2}$, $Cr+^{3++}$, and $Ag+$ are broad-spectrum toxicants that disrupt many enzymes and damage the physical structure of proteins. More specific toxic actions include blocking of electron and ions exchange across mitochondrial membranes in the electron transport process by tributyltin oxide; pentachlorophenol uncoupling and disrupting the formation of ATP from ADP; and arsenic blocking the pyruvate dehydrogenase enzyme system in glycolysis and also serving as a competitive inhibitor of phosphorous in both substrate and oxidative phosphorylation reactions. Cytochalasins inhibit the transport of cellulase across cell membranes while antibiotics,

such as penicillin, disrupt cell wall synthesis in some types of bacteria. This topic will be discussed in more detail in the chapter on wood preservatives.

Nutrition in relation to fungal growth requisites and decay control

A general diagram of the nature and sites of these many degradative and respiratory activities is summarized in Fig. 5.5. In this model, the related roles of the ecological factors affecting the growth of fungi and decay rates are re-emphasized. It is interesting to note that the effect of each growth factor can be explained at the molecular level by an enzymatic reaction or its requirements. Important points to stress are:

1. Water is the diffusion medium for enzymes and oxygen, a reactant in hydrolysis, and the solution medium for all cell chemistry.
2. Oxygen (free) is the ultimate electron and hydrogen acceptor in the energy yielding aerobic oxidation-reduction reactions, forming H_2O.
3. Temperature controls reaction rate and, at higher levels, disrupts the stability of enzyme structures.
4. The substrate provides the basic energy, the pool of metabolites for synthesis, and in many cases, the vitamin and nitrogen sources for fungi.
5. Minor metals and vitamins play critical roles as cofactors or coenzymes in the many enzymatic reactions.
6. Hydrogen ion concentration (pH) defines the optimal level for many enzyme reactions and protein stability.

Summary

1. Metabolism broadly includes all the chemical reactions occurring in living systems. Nutrition involving the digestion, absorption, and respiration of energy rich organic compounds is particularly important to the process of wood decay and its control.
2. Fungi and most bacteria are heterotrophs that require energy from organic sources. This energy is released in the process of respiration. The basic source is electron donors (organic compounds) and

involves the release and transfer of energy when chemical bonds are broken and reformed during the exchange of electrons in redox reactions.

3. Adenosine triphosphate (ATP) is a major energy-rich compound that stores and provides the energy released by respiration for many cellular activities. Nicotinamide adenine dinucleotide (NAD) is one of the important electron carriers in respiration reactions and a phosphorylated form (NADP) plays a similar major role in many synthetic reactions.
4. Many chemical reactions in metabolism are facilitated by enzymes. Enzymes are biocatalysts that accelerate reaction rates without being permanently altered themselves. Enzymes are complex proteins that can be readily inactivated or destroyed by adverse temperatures, pH, and many toxic chemicals.
5. Hydrolases and oxido-reductases are the major types of enzymes involved in decay and cell respiration.
6. Wood decay can be considered to be the external digestion of the large macropolymers (cellulose, hemicelluloses, and lignin) of the cell wall that are insoluble in water. Digestion is carried out primarily by hydrolases and oxidases that reduce the complex polymers to diffusible units that can be absorbed and respired for energy and metabolites for synthesis.
7. Most fungi utilize the aerobic respiration pathway, that requires atmospheric oxygen (O_2) as the final electron acceptor in the oxidation of glucose and the synthesis of ATP (storage of chemical energy). The three major phases of respiration are glycolysis, the citric acid cycle, and the electron-transport chain.
8. Some fungi and bacteria utilize nutrients by fermentation. There is no external electron acceptor in this biochemical pathway and the electron transfers that release energy take place between portions of the substrate molecules.
9. Some bacteria utilize an anaerobic respiration pathway where compounds other than oxygen are the electron acceptors. Some bacteria in this group are able to decompose cellulose in anaerobic environments and animal rumens.
10. The requisites for fungal growth can often be explained at the molecular level by the related enzymatic reactions involved or its requirements. Practical prevention or control of decay can often be achieved by adversely affecting a critical growth factor (Fig. 5.5).

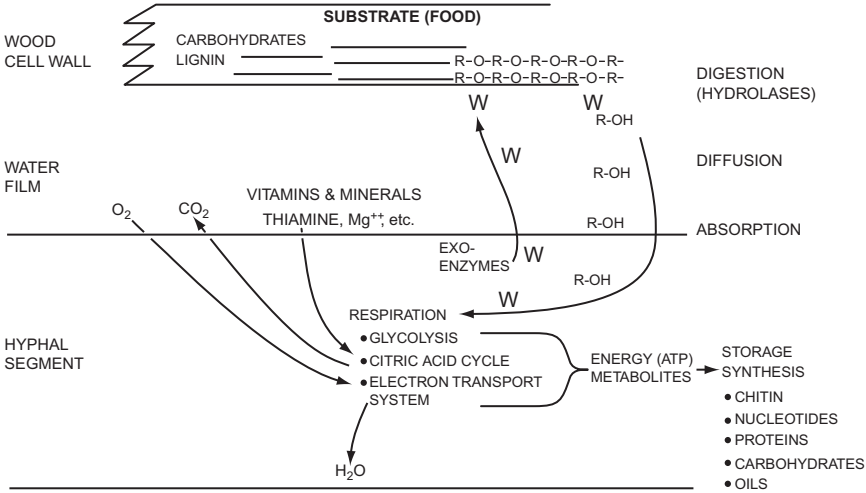


Figure 5.5 A simplified model of the major nutritional activities of a decay fungus, including digestion, absorption, and respiration and the relationships between growth requisites and decay control practices.

References

Brock, T.D., Smith, D.W., Madigan, M.T., 1988. Biology of Microorganisms, fifth ed. Prentice-Hall, Inc, Englewood Cliffs, NJ, p. 835.

Cano, R.J., Colome, J.S., 1986. Microbiology. West Publishing Co., New York, p. 888.

Dixon, M., Webb, L.C., 1964. Enzymes, second ed. Academic Press, New York, p. 950.

Garraway, M.O., Evans, R.C., 1984. Fungal Nutrition and Physiology. John Wiley & Sons, New York, p. 401.

Griffin, D.H., 1981. Fungal Physiology. John Wiley and Sons, Inc, New York, p. 383.

Madigan, M.T., Martinki, J.M., Bender, K.S., Buckley, D.H., Stahl, D.A., Brock, T., 2018. Brock Biology of Microbiology. Pearson/Prentice Hall, NY.

Raven, P.H., Evert, R.F., Eichhorn, S.L., 1986. Biology of Plants, fourth ed. Worth Publishers, Inc, New York, p. 775.

Stryer, L., 1981. Biochemistry, second ed. W.H. Freeman and Co., San Francisco, p. 949.

Wiley, J., Sherwood, L., Woolverton, C.J., 2008. Prescott's Microbiology. McGraw-Hill, NY.

Further reading

Abelson, J.N., Simon, M.L., 1990. Methods in Enzymology. Volume 175. Cumulative Subject Index. Academic Press, San Diego, p. 272.

Ainsworth, G.C., Sussman, A.S., 1965. The Fungi., vol. 1. Academic Press, New York.

Blumenthal, H.J., 1965. Carbohydrate metabolism. In: Ainsworth, G.C., Sussman, A.S. (Eds.), The Fungi: An Advanced Treatise. Academic Press, New York, pp. 229–348.

Colowick, S.P., Kaplan, N.O., 1988. In: Wood, W.A., Kellogg, S.T. (Eds.), Methods in Enzymology Volume 160 Biomass Part A Cellulose and Hemicellulose. Academic Press, Inc, New York, p. 774.

Corbett, J.J., 1974. The Biochemical Mode of Action of Pesticides. Academic Press, New York, p. 330.

Fersht, A., 1977. Enzyme Structure and Mechanism. W.H. Freeman and Company, San Francisco, p. 371.
International Union of Biochemistry, 1961. Report of the Commission on Enzymes. Pergamon Press, New York, p. 159.
Siegel, M., 1981. Sterol-inhibiting fungicides: effects on sterol biosynthesis and sites of action. Plant Dis. 65 (12), 986–989.
Sigman, D.S., Boyer, P.O. (Eds.), 1990. The Enzymes. Volume XIX. Mechanisms of Catalysis. third ed. Academic Press, San Diego.
Smith, J.E., Berry, D.R., 1976. The Filamentous Fungi. Volume II. Biosynthesis and Metabolism. John Wiley and Sons, New York, p. 520.

CHAPTER SIX

The decay setting: Some structural, chemical, and moisture features of wood features of wood in relation to decay development

Contents

In order to understand the anatomical and chemical aspects of the decay process, a brief review of wood structure is needed, emphasizing the types and locations of the major chemical constituents of cells walls, and the dimensions of the various openings and capillaries of the wood system that control enzyme access and water accumulations.

These topics are presented in detail in the various books and review articles cited on wood anatomy, wood ultrastructure, wood technology, wood-liquid relations, reaction woods, and wood chemistry (Côté, 1965, 1981; Côté et al., 1968; Core et al., 1976; Fengel and Wegener, 1984; Higuchi, 1985; Hoadley, 2000; Panshin and deZeeuw, 1980; Skaar, 1972;

Wood Microbiology.
DOI: https://doi.org/10.1016/B978-0-12-819465-2.00006-1

Siau, 1984; and Timell, 1982, 1986). For our purposes wood will be defined broadly as the accumulated xylem of the aerial stems of perennial plants. Commercial woods come primarily from the Gymnosperms (softwoods) and dicotyledons (hardwoods) of the Angiosperms. A few monocotyledons (palms and bamboo) in tropical zones are also used for structural purposes. As a plant material originating from the stems of many different species, wood is a complex and highly variable material both structurally and chemically.

Wood functions

The functions of wood in the living plant provide useful insights about its structure and help to explain how saprobes discolor and decay wood products. The wood tissue system consists of clusters of specialized xylem cells carrying out four major functions (Fig. 6.1).

(a) Conduction of water and various solutes through tracheids or vessels from the roots to the leaves.

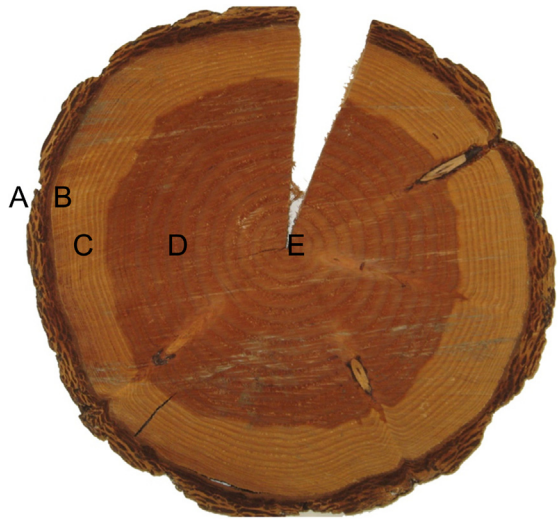


Figure 6.1 (A) Cross section (transverse) of a Douglas-fir stem (*Pseudotsuga menziesii*) stem showing the (A) bark, (B) cambial zone, (C) sapwood, (D) heartwood, and (E) pith zone.

(b) Support to hold erect a large assemblage of branches and leaves in a competitive position with those of other trees for light in drastically varying weather conditions. This function requires a supporting material with high tensile, compressive and bending strength. The stem of a tree closely resembles a loaded vertical cantilever beam in carrying out this function.

(c) Storage of water and various translocates (e.g., reserve foods, hormones) in both radial and longitudinal parenchyma tissues.

(d) Protection (durability) of the energy rich accumulating stem tissues from pathogen invasion and disease or destruction by decay fungi or insects.

Tracheids carry out both fluid conduction and strength functions in conifers. Conduction is carried out primarily by vessels, while the fibers provide support in hardwoods. The parenchyma cells in both groups serve a storage function and provide protection against biological attack by deposition of protective toxicants when the stem is injured or during heartwood formation.

The structural features of wood growth patterns and microscopic features

Wood represents the annual accumulations of cone-shaped increments of xylem cells originating from a lateral meristem or cambium. The cambium is a continuous ring of meristematic cells that form around the outer circumference of the developing stem. Cambium originates from the inter-fascicular cambium of the procambial strands and the ground meristem. The procambial strands and ground meristem originate from the apical meristem in the growing tip of the stem and are primary tissues (Esau, 1965). The cambial cells divide periclinally and form xylem cells (wood) to the inside and phloem cells (bark) to the outside. One cell of the dividing pair remains meristematic. Occasional anticlinal divisions of the cambial cells allow the stem to increase in girth. There are two general types of cambial cells based on shape and the tissues formed. Vertically elongated cambial cells, termed fusiform initials, form the longitudinal parenchyma, the tracheids in conifers, and the vessels and fibers in hardwoods. Horizontally elongated or cuboid cells, termed ray initials, form the radially aligned wood rays consisting of ray parenchyma and, in conifers, also ray tracheids.

The vascular cambium divides only during the growing season. In temperate zones, the annual accumulations of xylem often result in abrupt

annual rings, usually consisting of a zone of rapid growth (earlywood) in the spring and early summer and slower growth (latewood) later in the growing season. In tropical zones, annual rings are not as apparent in many species and the differences in growth accumulations may closely reflect regular seasonal patterns in rainfall. Annual ring elements in conifers consist primarily of tracheids in uniform radial rows. In angiosperms annual rings consist of vessels, tracheids and fibers that are arranged in ring-porous, semi porous or diffuse porous patterns depending on the size and distribution of the vessels. Parenchyma cells are intermixed with these conductive and strength providing tissues in longitudinal and ribbon-like radial distributional patterns. Adjacent wood cells are interconnected by thin zones in the wall and cell ends called pits that are discussed in a later section on the cell wall.

The outer zone of the stem contains many living parenchyma cells and is termed the sapwood. Sapwood is white in color in most species and functions for conduction, food storage and stem protection. The pith is a small zone at the center of the stem, consisting of parenchyma cells and originating as a primary tissue from the ground meristem. As the girth of the tree expands and the inner sapwood tissues age and recede from the phloem, increasing numbers of parenchyma cells slowly die, and the tissue develops into non-living heartwood. The transition zone between the sapwood and heartwood is sometimes termed intermediate wood. Heartwood is the internal core of dead tissue in the stem that may or may not be colored. It slowly expands outward as the tree ages and growth diminishes. The transition zone may be abrupt or gradual and does not necessarily form uniformly in the same annual ring. The heartwood is deeply colored in some species of *Quercus*, *Juglans*, and *Fraxinus* and similar to sapwood in color in species of *Abies, Populus, Picea*, and *Tsuga*. The sapwood is wide in some species such as ponderosa pine (*Pinus ponderosa*) and loblolly pine (*Pinus taeda*) and narrow in Douglas-fir (*Pseudotsuga menziesii*) and longleaf pine (*Pinus palustris*). Current theory on heartwood origins suggests successive formation of small air bubbles (embolisms) in the older vessels and tracheids that isolate the adjacent parenchyma cells from food sources, ultimately resulting in their death. While most cells are dead in the older heartwood, a few isolated parenchyma cells are reported to remain alive for years in some species. Significant chemical and structural changes during the transformation of sapwood into heartwood include the loss of starch, the deposition of extractives, and the aspiration of pits in conifers or the formation of tyloses in hardwoods. These changes may render the wood more resistant to biological attack or decrease the

permeability, making preservative treatment more difficult. The common macroscopic features of wood are illustrated in Fig. 6.1.

Coniferous wood consists of tracheids and parenchyma cells. Tracheids function in both conduction and structural support. Hardwoods are more complex and are considered to be taxonomically more advanced. Short barrel-shaped cells called vessels form the specialized conductive tissue for hardwoods and long, thick-walled cells called fibers provide strength. Parenchyma cells carry out food storage, translocation, and protective functions in both wood types. The major cell types in wood are illustrated in Fig. 6.2. Parenchyma cells are arranged radially in wood rays, but are also axially oriented in woods that form longitudinal parenchyma that often surround the vessels. Longitudinal parenchyma in some conifers, such as the pines, larches, spruces, and Douglas-fir, form large resin canals that may be visible to the unaided eye. Characteristic features of conifers are the arrangement of tracheids in radial rows of uniform width and inconspicuous wood rays. In contrast, the wood rays are a prominent feature of many hardwoods, approaching 25% of the wood volume in some species such as the oaks and beeches.

It is convenient to describe wood anatomically as the tissues appear in the 3 planes of a precisely cut wood cube. The cross or transverse section represents the plane oriented at a right angle to the longitudinal axis of the stem. The transverse face exposes the open ends of the longitudinal aligned or axial cells. The radial section is the surface exposed by a plane running from the outer stem and bisecting the stem center or pith. This plane exposes the radial surface of the wood rays. The tangential plane is oriented tangent to the outer stem surface, at a right angle to the radial section. These wood planes are illustrated by photomicrographs of a typical conifer and hardwood (Fig. 6.3).

Cell Wall Ultrastructure: Wood cell walls consist primarily of the large biopolymers cellulose, hemicellulose, and lignin. The chemical structures and interrelationships of these components are discussed later in this chapter. Cellulose molecules in the cell wall form microfibrils that are surrounded by hemicellulose. Microfibrils are laid down successively and form layers or laminae in the cell wall as growth occurs. Lignin deposition occurs after cell maturation. Wardrop (1964) proposed the useful terms of framework for the cellulose microfibrils, matrix for the hemicellulose, and encrustant for the lignin to explain both their function and interrelationships in cell wall structure. Tracheids in conifers and fibers in hardwoods are the predominant cell types affecting strength and the many use properties of wood. This Chapter will be limited to their cell wall structure.

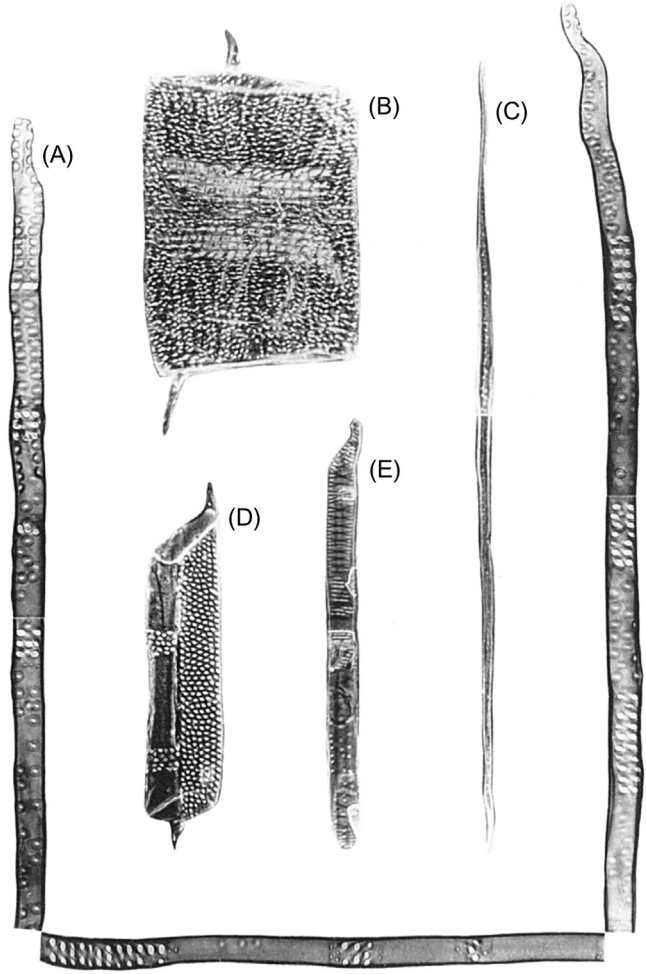


Figure 6.2 A radial view of the principal cell types of a conifer and a hardwood indicating their relative sizes and the types and frequency of pitting: (A) a hardwood vessel, (B) a hardwood fiber, (C) a conifer tracheid, (D) a hardwood ray parenchyma cell, and (E) a conifer ray parenchyma cell. Note the greater length of the conifer tracheid and the abundance of pitting in the vessels. The zones of abundant pitting in the tracheid occur where ray parenchyma were in contact. *Courtesy:* Dr. W.A. Côté, from Kollman and Côté, 1968 with permission.

A cross section of a typical fiber or tracheid cell wall reveals a common organizational pattern. The middle lamella (ML) is a narrow zone between contiguous cells that consists primarily of pectins and lignin. The middle lamella is derived from the new cell plate that forms during the

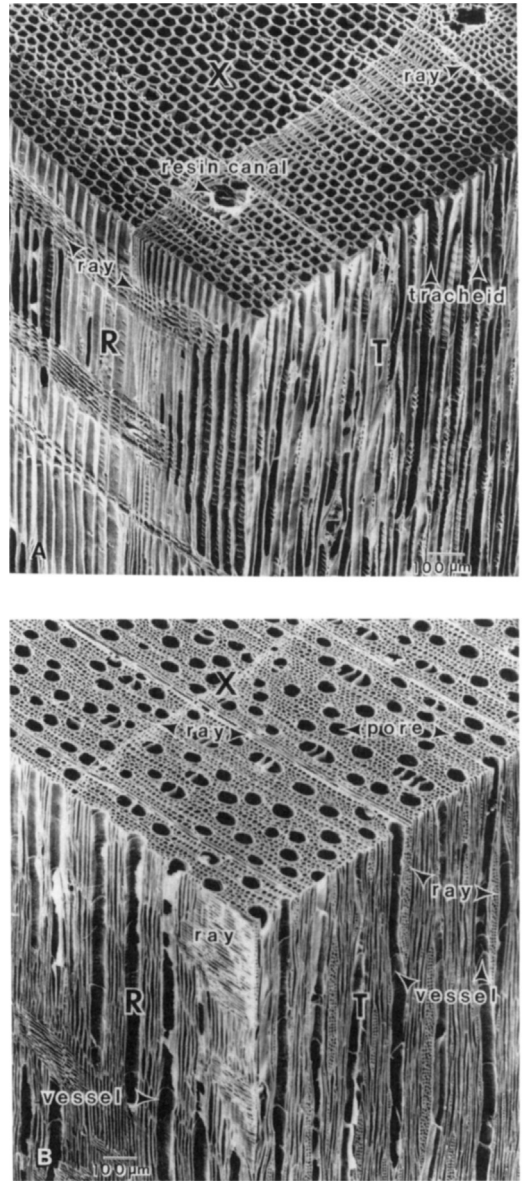


Figure 6.3 Cubes of eastern white pine (*Pinus strobus*) and red maple (*Acer rubrum*) illustrating the cross, radial and tangential planes and important microscopic features. *From Côté, W.A. (1981). Ultrastructure - Critical Domain for Wood Behavior. Wood Sci. Technol. 15, 1—29.*

mitotic division of a cambial cell into another cambial cell and a cambial initial (that develops into either a xylem cell on the inside or phloem cell on the outside). The middle lamella in stained sections may also be seen as a fine dark line between contiguous cells. A thin primary wall (PW) is initially laid down during enlargement and maturation of the cambial initial into a tracheid or fiber. The wall consists of a loose network of mostly axially oriented cellulose microfibrils. The secondary wall (SW) develops next and consists of three successively formed layers termed the S1, S2, and S3. The S1 and S3 are narrow zones where the cellulose microfibrils are arranged in a flat helix. The S2, which comprises the bulk of the cell wall, consists of microfibrils that are arranged in a steep helix oriented nearly parallel to the longitudinal axis of the cell. The S2 layer is the most important zone of the cell wall and is responsible for a majority of wood strength properties, particularly its remarkable tensile strength. The S2 cell wall comprises most of the cell wall seen in cross sections under the light microscope. A warty layer also develops on the S-3 surface of some woods. This layer represents either additional depositions of the S3 wall material or accumulations of protoplasmic debris upon cell death. Discerning individual layers (PW and the S1, S2, and S3 of the SW) requires the higher magnification of the electron microscope. The various parts and layers of the cell wall are illustrated in Fig. 6.4. The inner cell cavity is termed the cell lumen. The lumen is inert space occupied by air or water in the living tree or the wood product. The lumen volume, collectively, in most woods is large, and will be seen later as the critical cell wall zone where most fungi initiate the decay process.

Cell wall pitting

Conduction of water and various solutes between adjacent cells occurs across contiguous thin zones in the wall termed pits. Pits provide an interconnected network between cells. Pits are also the principal cell wall zone initially penetrated by hyphae during wood colonization and degradation by stain and decay fungi. Pits are gaps in the secondary wall containing a modified portion of the adjacent primary walls called the pit membrane. The types of pits vary with cell types and plant species.

The tracheids of many commercial conifers (Pinaceae) and the fibers in hardwoods possess bordered pits. The membrane in a bordered pit consists of cellulosic strands (margo) and a thickened central portion termed a torus (Fig. 6.5). The spaces between the strands of the membrane are large

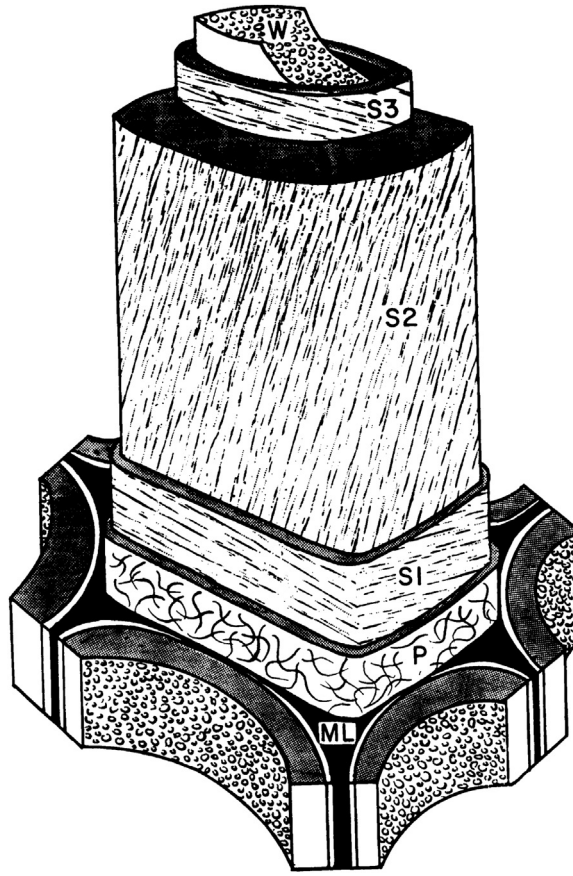


Figure 6.4 A model of several contiguous wood cells showing the organization and microfibrillar orientation of the major cell wall layers. The layers are identified from the middle lamella (ML) and outward as primary wall (P), the S1, S2, and S3 comprising the secondary wall, and the warty (W) lining the lumen surface. *From Wilfred A. Wood Ultrastructure: An Atlas of Electron Micrographs. pp. 33. © 1967. Reprinted with permission of the University of Washington Press.*

enough to permit liquid flow and small particle passage (up to 1 μm) between adjacent cells. These openings may become occluded with extractives during heartwood formation. The secondary wall of each adjacent cell in the pit pair forms a partial arch around the connecting opening or aperture of each cell member. The torus may block the aperture upon cell death, due to changes in moisture content or alterations in air pressure. Wood with many blocked (aspirated) pits is difficult to season or treat with preservatives.

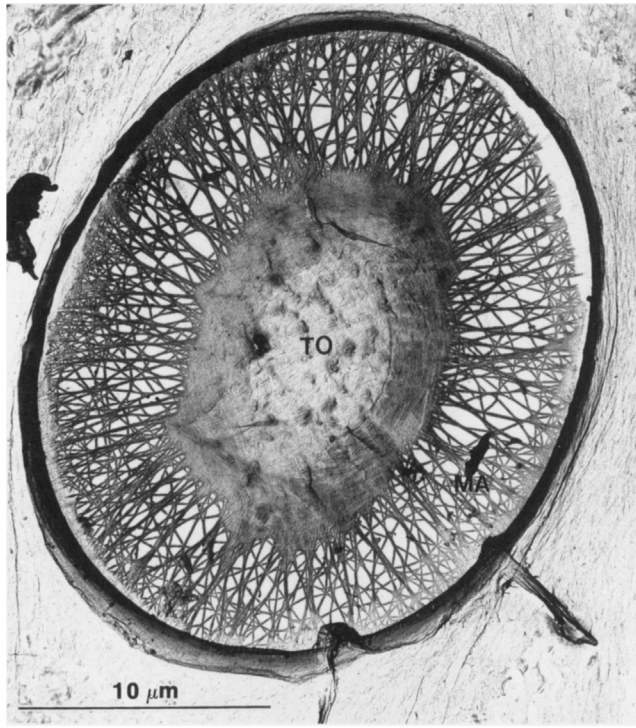


Figure 6.5 Transmission electron micrograph of radial surface replica of a bordered pit membrane from an earlywood tracheid of *Tsuga canadensis*. Note the nature and dimensions of the openings among the margo (MA) and the thickened center or torus (TO). *Reproduced with permission from Côté, W.A., 1977. Wood Ultrastructure in Relation to Chemical Composition. In: Loewus F.A., Runeckles V.C. (eds) The Structure, Biosynthesis, and Degradation of Wood. Recent Advances in Phytochemistry, vol 11. Springer, Boston, MA.*

The pits between ray parenchyma cells and adjacent tracheids are termed semi-bordered pits. The membranes in these pits lack a torus and are continuous. The pits between parenchyma cells are termed simple pits and contain numerous small plasmodesmatal pores (Harada and Côté, 1985). The pits between vessels and adjacent parenchyma cells in some hardwoods form tyloses. These tyloses are formed when the pit membrane bulges or balloons into the vessel lumen occluding fluid flow. In some species such as the white oaks, tyloses may completely block the large earlywood vessels (Fig. 6.6). Tyloses formation followed by wilting is a common symptom of many vascular diseases of forest trees such as oak wilt or Dutch-elm disease.

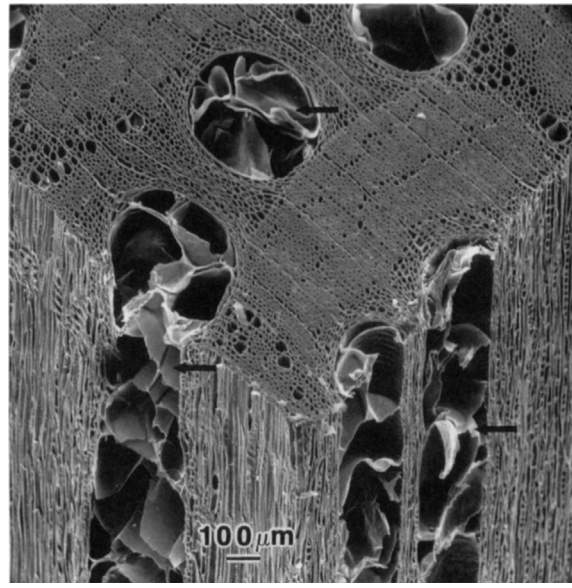


Figure 6.6 A scanning electron micrograph of a white oak (*Quercus alba*) cube showing numerous tyloses occluding the large vessels in the earlywood zone. *Courtesy W. A. Côté.*

The major chemical constituents of wood

Cellulose, the hemicelluloses, and lignin are the three major types of chemical constituents of wood cell walls. They are large macromolecules or biopolymers that are closely associated physically or covalently bonded in the case of lignin and the hemicelluloses. Collectively, these cell wall polymers represent the major organic compounds in the biosphere and are a principal carbon sink in terrestrial ecosystems. Cellulose and the hemicelluloses are carbohydrates that are readily digested by many organisms. Lignin is an aromatic heteropolymer consisting of condensed phenylpropane units and is a recalcitrant compound that can be degraded by only a few groups of specialized fungi (the wood decayers) or bacteria and often over long time periods.

Cellulose

Cellulose is a long, linear homopolymer consisting of β-D-glucose residues connected by (1−4) glycosidic linkages (Fig. 6.7A). The surfaces of the cellulose molecules contain three exposed hydroxyl groups per anhydroglucose unit that control the structural properties in the cell wall as well as

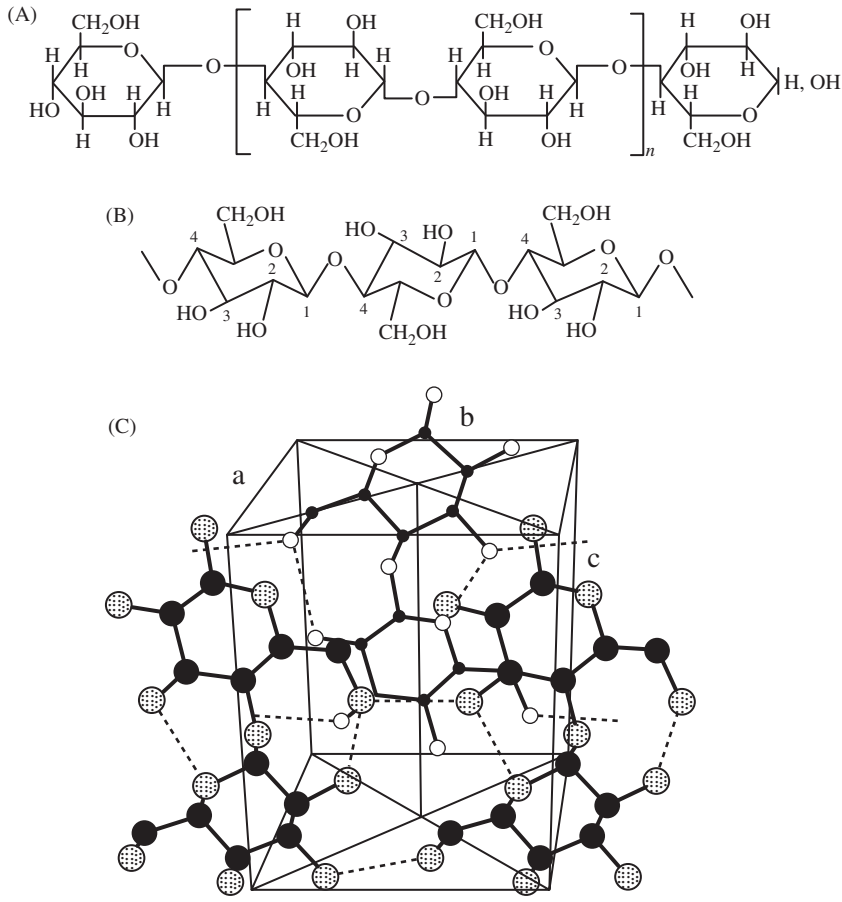


Figure 6.7 (A) A portion of a cellulose molecule showing the (β1-4) glycosidic linkage between two glucose units. (B) The same portion drawn in a chair configuration showing the equatorial position of the hydroxyls on the number 2, 3, and 6 carbons. (C) An elementary unit of crystalline cellulose showing the dimensions and relative positions of the glucose units in the crystalline structure.

many physical and chemical properties of wood (Fig. 6.7B). An average degree of polymerization (DP) of around 10,000 has been determined for bark and wood cellulose (Goring and Timell, 1962), but the degree of polymerization of plant celluloses can vary between 3000 and 26,000 units. The cellulose chains are aligned parallel to each other and combine to form the microfibrils discussed above. Microfibrils can be observed using the electron microscope (TEM), and these structures seem to be the common form of all natural or native celluloses. Microfibrils consist of

highly organized or crystalline zones interspersed with non-crystalline or unorganized zones. The degree of crystallinity for cellulose in wood ranges from 60 to 70%.

A single cellulose molecule in a microfibril may continue through several crystalline and non-crystalline zones. X-ray diffraction studies of the crystalline zones have defined the precise dimension of a unit cell in the crystal (Fig. 6.7C). The surfaces of the microfibrils are surrounded by hemicelluloses (Fig. 6.8A). The polysaccharides are laid down successively

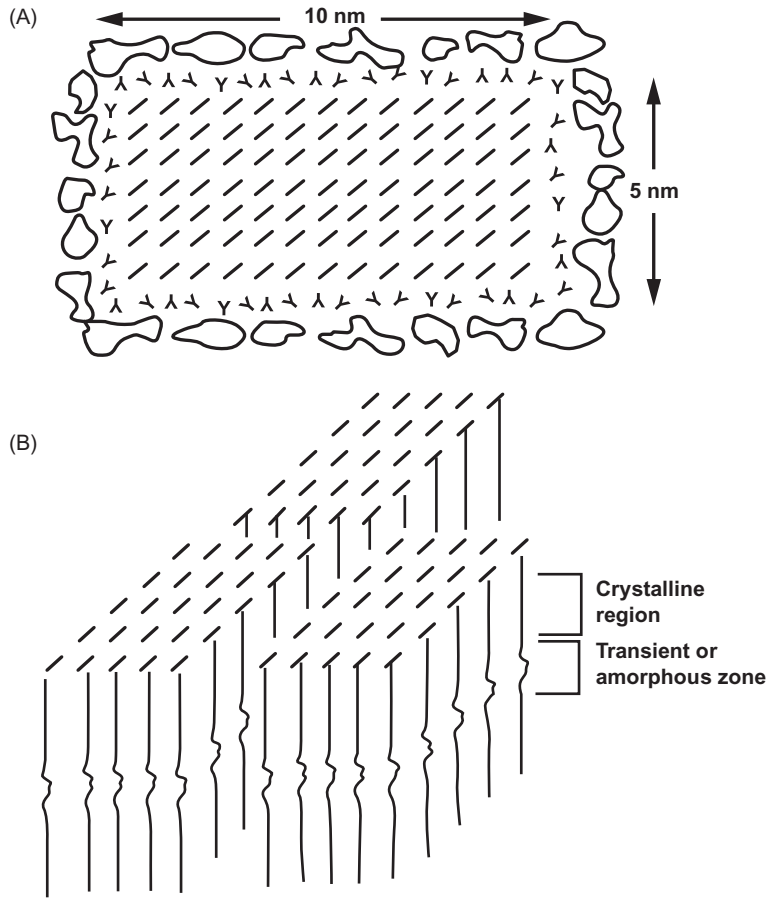


Figure 6.8 A schematic illustration of the postulated arrangement of the long linear cellulose molecules in a microfibril. (A) A cross section through the crystalline zone of a microfibril. (B) A longisection of several microfibrils showing the associations and relative sizes of the crystalline and amorphous zones. The hemicellulose and lignin are omitted in this sketch.

and form layers or lamina in the cell wall as growth and cell enlargement occurs. As the cell matures, lignin deposition occurs. The hemicellulose and lignin are associated primarily with the non-crystalline zones that occur within and between the microfibrils (Fig. 6.8B).

Hemicellulose and other carbohydrates

Hemicelluloses are polymers of various pentose and hexose sugar units. The major sugar residues in the polymer backbones are glucose, xylose, mannose, galactose, arabinose, rhamnose, and uronic acids. These polymers differ from cellulose in having shorter chain lengths, side chains that are sometimes branched, and sugar monomers other than glucose. The types and amounts of the hemicelluloses present in the cell walls differ between hardwoods and conifers. Softwoods contain less hemicelluloses than hardwoods, with mannose being the most common constituent of conifer hemicellulose. Xylose is a major constituent of hardwood hemicelluloses which also contain more acetyl groups than conifers (Table 6.1).

Xylan, the predominant hemicellulose in hardwoods, is a homopolymer of β-D-xylose monomers connected by (β1-4) glycosidic bonds. Side branches of 4-0-methyl-α-D-glucuronic acid are attached by (1−2) linkages to some xylose units and O-acetyl groups substitute some hydroxyIs (Timell, 1964, 1965) (Fig. 6.9A). Glucomannan, the major hemicellulose in conifers (up to 20% of cell wall) is a heteropolymer with a backbone containing β-D-glucose and β-D-mannose units connected by (1−4) glycosidic bonds (Fig. 6.9B). Acetyl groups and galactose residues are attached to some monomers in the backbone (Timell, 1965). The xylan in conifers has arabinose side chains instead of acetyl groups. Larch trees often contain large amounts of an arabinogalactan in their heartwood.

Table 6.1 The major differences in the sugar residues from hemicelluloses of three conifers and three hardwoods[a].

Species	Content in extract-free stemwood (%)		
	Xylose	Mannose	Acetyl group
Abies balsamea	5.2	10.0	1.4
Pinus sylvestris	7.6	12.4	1.6
Picea glauca	7.0	12.0	1.2
Fagus grandifolia	21.7	1.8	4.3
Populus tremuloides	12.2	3.5	3.9
Betula papyrifera	23.9	2.0	3.9

[a]Summarized from the data of Côté et al., 1966 and Timell, 1969.

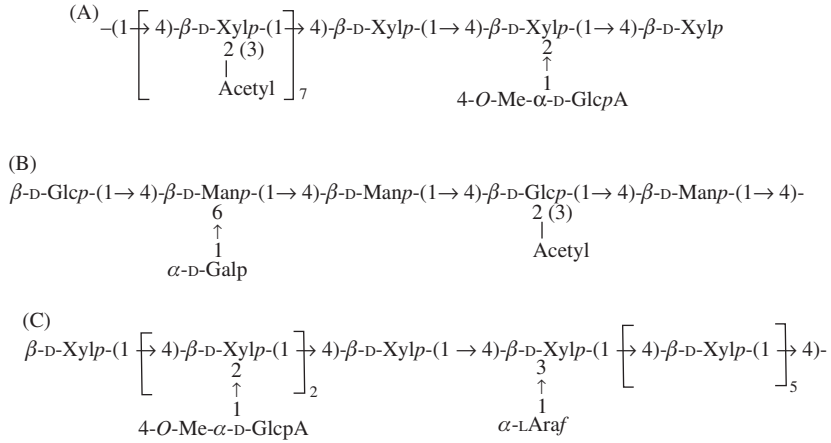


Figure 6.9 Examples of hemicelluloses commonly found in wood. (A) A xylan (O-acetyl-4-0-methylglucuronoxylan) commonly found in hardwoods and the abbreviated formula, (B) a galactoglucomannan (O-acetyl-galactoglucomannan) commonly found in softwoods and the abbreviated formula, and (C) a xylan (arabino-4-0-methylglucuronoxylan) also found in softwoods. *Source: Fengel, D., Wegener, G., 1984. Wood Chemistry, Ultrastructure, Reactions. Walter de Gruyter, New York, pp. 613.*

Many other hemicelluloses occur in smaller amounts in wood materials. Pectin is found in the middle lamella zone and in the torus of bordered pits. A glucan, starch, is an important food reserve in plants and is often found in abundance in wood parenchyma cells. One constituent of starch, amylose, consists of α-D-glucose units linked by (1−4) glycosidic bonds and is linear, while amylopectin, the major starch component is branched at position C-6.

Hemicelluloses in the wood cells probably serve a structural function by coating and binding the cellulose microfibrils into a common matrix, and may also serve to prevent the cellulose from becoming too crystalline. There is considerable evidence that fungal attack begins with decomposition of hemicelluloses and these changes have profound effects on the material properties of wood. The short chain lengths (DP 200 in most cases) that increase hemicellulose solubility and the outer exposed position on the surface of the microfibrils may explain why these polymers are among the first cell wall components attacked by decay fungi.

Lignin

Lignin, the most complex of the cell wall constituents, is a polyphenolic polymer formed from three types of phenyl propane units (Fig. 6.10A).

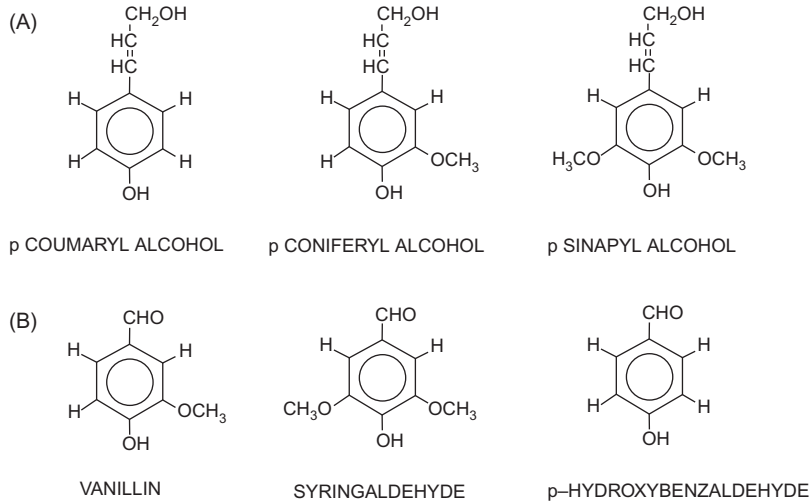


Figure 6.10 The principal precursors of lignin: (A) Coniferyl, sinapyl, and *p*-Coumaryl alcohols and (B) Several typical lignin degradation products.

These monomeric units condense by free radical polymerization to form a huge, heterogenous aromatic biopolymer. Lignin comprises about 20–30% (in a few species the lignin content approaches 40%) of the wood cell wall and is a constant feature of all vascular plants (ferns, fern allies, and seed plants). Lignin provides both mechanical strength and protects (durability) stem tissues against degradation. Lignification may be viewed as a remarkable evolutionary event that permitted the development of aerial plants from which the major timber species originated with their large vertical, perennial stems.

The structure of lignin varies between conifers and hardwoods. Guaiacylpropane units are the principal repeating monomer in conifers, while both guaiacyl and syringylpropane units are present in hardwoods. Lignins also contain small amounts of p-hydroxyphenylpropane units, a lignin monomer that is common in monocotyledonous plants. The principal precursors of lignin formation in the cell wall are three p-hydroxycinnamyl alcohols; coniferyl, sinapyl, and p-coumaryl alcohols (Fig. 6.10B). Dehydrogenation of these alcohols forms phenoxy-radicals that, by subsequent dehydrogenation, polymerize to form lignin. The polymerization process of these phenoxy radicals is random and the lignin macromolecule formed has none of the predictable fixed or repeating structures that are found in the other cell wall constituents, cellulose and hemicelluloses.

The principal linkages among the various phenylpropane units are carbon to carbon (C-C) and ether (C-O-C) bonds. Of the many inter-unit linkage types present in lignin, the β-aryl ether linkage *(β-0—4)* occurs more than 50% of the time (Fig. 6.11). Lignin deposition begins after the new xylem cell has enlarged and the polysaccharides have been laid down in the outer part of the secondary wall. The process proceeds slowly

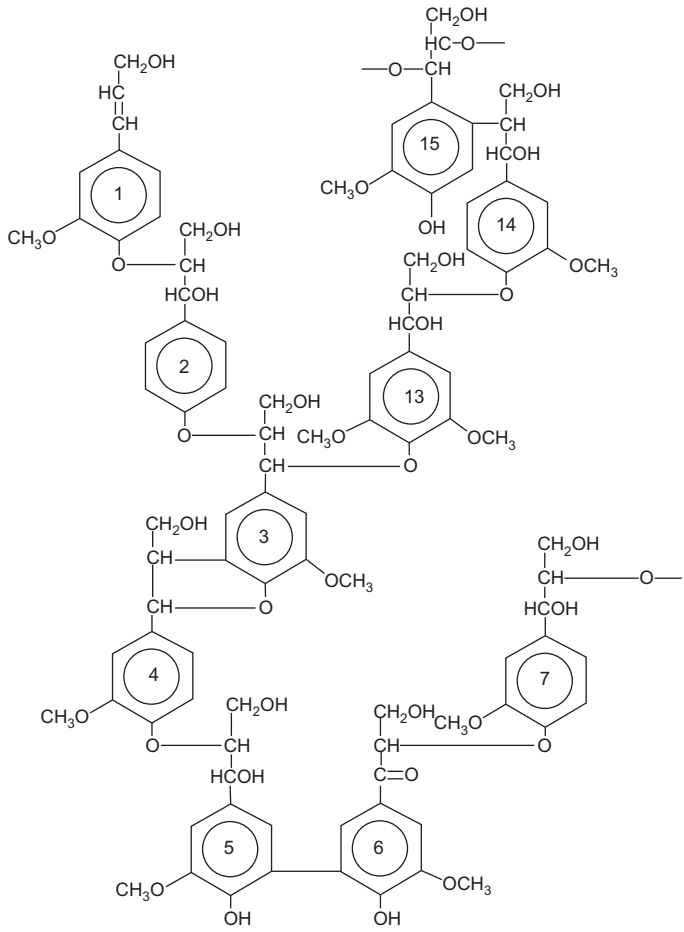


Figure 6.11 A structural model of spruce lignin. Examples of the common guaicyl glycerol-β-aryl ether linkage (β-0—4) can be seen between units 1 and 2. A syringyl phenyl propane unit with two methoxyl groups (OCH_3) in the benzene ring can be seen in unit 13, and biphenyl linkages (C-C) can be seen between units 5 and 6. *From Adler, E., 1977. Lignin chemistry --past, present, and future. Wood Sci. Technol. 11 (3),169—218.*

following successive formation of the layers of the secondary wall (S1, S2 & S3) and then progresses more rapidly after these layers are complete. Lignin is probably interspersed in the spaces within and between the hemicellulose-coated cellulose microfibrils, forming an inter-penetrating polymer system with the hemicelluloses. Lignin links covalently with some hemicellulose units. Lignification provides cohesion and strength to the cell wall and serves as an effective barrier against microbial access and digestion of the carbohydrates. Lignin is an amorphous polymer whose chemical structure can only be modeled on the basis of the kinds of units, frequency of linkages, and the nature of some of its degradation products. Models of lignin as it may occur in a conifer and a hardwood are shown in Fig. 6.11. Expanded models of lignin, based on a computerized analysis program involving 94 phenylpropane units were developed (Glasser et al., 1981).

Miscellaneous cell wall chemicals

Extractives -A number of other compounds are present in the cell wall, including extractives. Extractives are mostly low-molecular weight compounds that are readily removed or extracted from wood by solvents such as water, alcohol, benzene, or ether. These compounds are primarily found in parenchyma cells and in the lumens of other cells in amounts ranging from 1 to 5% of the total wood weight. In exceptional cases, extractives may represent 10–40% of the wood weight. Extractives represent many classes of compounds including a large number that are species-specific. A few examples are carbohydrates such as starch, glucose, fructose, and sucrose; phenolic compounds such as stilbenes, tannins, phlobaphenes, flavonoids, and lignans; oils and waxes; esters of organic acids; alkaloids; and tropolones.

Extractives affect many wood properties and two of special concern are susceptibility to sapstains (both biotic and abiotic stains) and natural durability of the heartwood. These effects will be considered in more detail in the later chapters on "Wood Sapstains" (Chapter 14) and "Wood Durability" (Chapter 18).

Ash Content -The inorganic or mineral content of wood is low and rarely exceeds one percent in temperate zone species. Some tropical hardwoods contain high levels of silica that improves resistance to marine borers. Mineral content is determined by incinerating the wood under controlled temperature conditions to reduce the losses of volatile ash

components. The principal elements present are calcium, potassium, and magnesium. Other common elements include manganese, sodium, phosphorus, and chlorine, as well as low concentrations of trace elements (Young and Quinn 1966). Several minerals play significant direct or indirect roles in the development of decay and wood defects. Mineral stain, a serious discoloration in some hardwoods, is characterized by an abnormally high calcium carbonate content and excessive warping. It is believed to originate as an injury response to outer sapwood in the living tree. Calcium is reported to increase the resistance of tissues in the living stem to pathogen attack in herbaceous plants (Agrios, 1978). Iron and manganese have been proposed to play roles as oxidants in some decays and are discussed further in Chapter 9. Release of "cell wall cations" during decay is purported to be one of the factors responsible for increases in electrical conductivity (Shortle, 1982), and these changes have been employed in at least one decay detection device (Shigo and Shigo, 1974). Historically, it is interesting to note that the alkaline nature of leached wood ash (Ca, K) was used in colonial times to saponify fats for the preparation of soap and large acreages of the original eastern forests were burned for commercial potash production.

Proteins: Small amounts of protein are present in wood cells, with the largest amounts in the cambium, early xylem derivatives, and parenchyma cells. Only trace amounts of protein are present in the dead xylem cells. Protein is the principal source of the nitrogen in wood, which is very low compared with other plant forms, ranging from only 0.03% in heartwood to as high as 0.1% in the young sapwood. Nitrogen is discussed in more detail in Chapter 4 as a necessary growth factor for fungi in wood.

Amounts and Distributions of Cell Wall Components: Wood is not only highly heterogeneous in the various arrangements of the cell types, but also in the amounts and distributions of the major cell wall components. Information on the distribution of cell wall components helps explain the differing ways various microorganisms attack and differentially utilize cell wall parts. For example, some bacteria selectively attack the cellulosic strands in the margo in the bordered pits of some pines; some soft rot fungi selectively attack the S2 of the secondary walls in conifers, and individual wood cells are detached in the intermediate stages of white rot caused by *Phellinus* (*Fomes*) *pini*.

Although the distribution of chemicals in cell walls differs by species, stem position, rate of growth, or the presence of heartwood/sapwood, some generalizations concerning wood chemistry can still be made (Table 6.2). Cellulose content is relatively constant across all species and

Table 6.2 Chemical composition of wood from five hardwoods and five conifers[a].

Component	Hardwoods				
	Acer rubrum L.	*Betula papyrifera* Marsh.	*Fagus grandifolia* Ehrh.	*Populus tremuloides* Michx.	*Ulmus americana* L.
Cellulose	45	42	45	48	51
Lignin	24	19	22	21	24
O-Acetyl-4-*O*-methyl-glucurono-xylan	25	35	26	24	19
Glucomannan	4	3	3	3	4
Pectin, starch, ash, etc.	2	1	4	4	2

Component	Conifers				
	Abies balsamea (L.) Mill	*Picea glauca* (Moench) Voss	*Pinus strobus* (L.) Carr.	*Tsuga canadensis* L.	*Thuja occidentalis* L.
Cellulose	42	41	41	41	41
Lignin	29	27	29	33	31
Arabino-4-*O*-methyl-glucurono-xylan	9	13	9	7	14
O-Acetyl-galacto-glucomannan	18	18	18	16	12
Pectin, starch, ash, etc	2	1	3	3	2

Data from Kollman and Côté (1968). Results provided by T. E. Timell.

[a]All values given in percentage of extractive-free wood.

comprises 40–50% of cell wall substance. In temperate zone woods, lignin is present in higher levels in conifers than hardwoods and the difference is made up by the higher hemicellulose content in hardwoods. The chemical components of five hardwoods and five conifers from the temperate zone illustrate the relative uniformity of cellulose levels in all the species studied as well as the higher lignin and lower hemicellulose contents in the conifers in comparison with lower lignin and higher hemicellulose contents in hardwoods (Table 6.3). Tropical hardwoods have higher lignin and ash contents than most temperate zone woods.

Distribution of the major chemicals in the wood cell wall

Distribution of individual chemical components vary widely across cell walls, but some generalizations can be made regarding the distribution of the principal chemical constituents among cell types and within cell wall layers. Cellulose is present at the highest levels in the secondary wall and is least abundant in the compound middle lamella (middle lamella and the adjoining primary walls). Hemicellulose levels are highest in the **S1** and lowest in the S2 of the secondary wall of tracheids and fibers. The hemicellulose content is higher in parenchyma cells, but hemicellulose types and distributions vary greatly among species. Ray parenchyma cells contain more xylans than do tracheids and fibers.

The removal of cellulose by acid hydrolysis and careful sectioning for electron microscopic study of the residual lignin clearly shows that high levels of lignin are present in the middle lamella and primary wall with lower and relatively uniform lignin distribution in the secondary wall (Fig. 6.12). The selective removal of carbohydrates by brown-rot fungi has also been used to prepare residual lignin for study (Côté et al., 1966). Detection techniques for studying lignin use either ultraviolet microscopy or bromination followed by dispersive x-ray analysis (TEM-EDXA) (Saka and Goring, 1985) permitted detailed mapping of lignin composition in the cell wall layers of conifers and hardwoods. Substantial differences occur in various cell types. Ray parenchyma cells and the secondary walls of hardwood fibers primarily contain a syringyl type lignin, while vessel walls contain mostly a guaicyl type lignin. In addition to the major components, pectins are present in the middle lamella zone. Starch and

Table 6.3 Major chemical components of a representative group of temperate zone hardwoods and conifers contrasted with several tropical hardwoods[a].

Name	Amount of chemical component (%)							
	Holocellulose	Cellulose	Hemicellulose	Pentosans	Lignin	Ethanol-benzene extracts	Hot-water extracts	Ash content
Temperate zone—conifers								
Abies balsamea	70.0	49.4	15.4	7.0	27.7	4.3	3.6	0.4
Picea abies	80.9	46.0	15.3	8.3	27.3	2.0	2.0	—
Pinus sylvestris	14.3	52.2	13.5	8.2	26.3	—	—	—
Pseudotsuga menziesii	67.0	50.4	—	6.8	27.2	4.4	5.6	0.2
Sequoia sempervirens	71.8	49.9	16.7	—	37.0	13.5	8.7	0.2
Temperate zone—hardwoods								
Populus tremuloides	80.3	49.4	21.2	17.2	18.1	3.8	2.8	0.4
Fagus sylvatica	85.8	44.5	30.2	20.6	22.2	—	—	—
Quercus sp.	73.2	40.5	23.3	17.5	22.2	—	—	—
Acer rubrum	71.0	44.5	—	17.1	22.8	2.5	4.4	0.7
Robinia pseudoacacia	81.7	50.1	—	23.7	20.6	2.8	4.6	0.3
Tropical hardwoods[b]								
Obeche	77.2	47.8	20.1	16.8	21.3	12.6	4.2	1.8
Kefe, awari	78.1	44.9	25.1	15.8	22.7	2.6	2.6	1.3
Teak	—	39.1	—	13.0	29.3	13.0	1.8	0.7
Mahogany	—	43.9	—	16.0	28.2	3.5	3.3	1.1
Balsa	—	52.0	—	19.0	24.5	2.6	2.8	1.6

[a]Data selected from Fengel and Wegener (1984).
[b]Scientific names for the tropical hardwoods selected are obeche (*Triplochiton sceroxylan* K. Schum.); Kefe, awari (*Pterogota macrocarpa* K. Shum.); teak (*Tectona grandis* L.); African mahogany (*Khaya anthotheca* C.D.C.); and balsa (*Ochroma lagopus* SW).

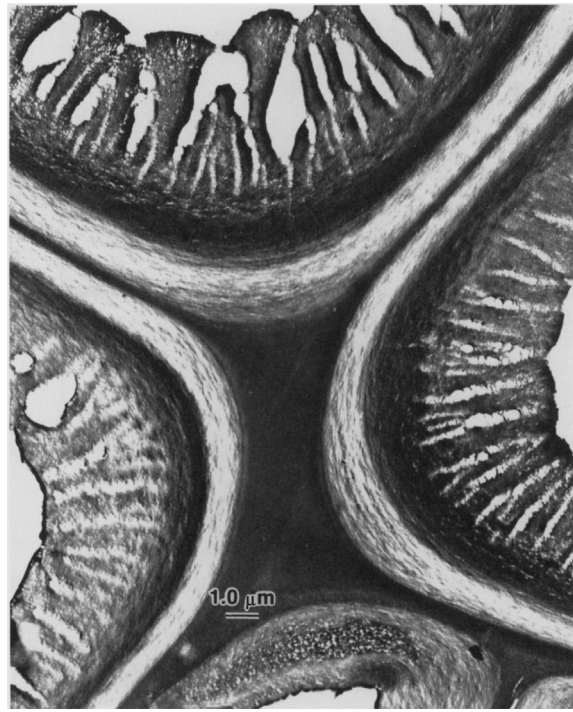


Figure 6.12 Cross-section of a Douglas-fir tracheid, revealing the lignin skeleton which remains after removal of the carbohydrates by successive exposures to hydrogen fluoride. Lignin concentrations in the middle lamella and network distribution in the secondary wall are indicated. *With permission from Wilfred A. Wood Ultrastructure: An Atlas of Electron Micrographs. pp. 33. © 1967. Reprinted with permission of the University of Washington Press.*

extractives are found predominantly in the parenchyma cells of the living sapwood, while the extractives generally occur in the greatest amounts in the outer zones of the heartwood.

In addition to the variations in polymer distribution in cell walls, trees can react to changing in growing stress to produce reaction woods that differ in the amounts of cellulose or lignin. Reaction wood in conifers, commonly known as compression wood, develops on the lower or compression side of leaning stems or branches. In hardwoods, reaction wood is termed tension wood and develops on the upper or tension side of leaning stems or branches. Compression wood is characterized macroscopically by a dark color, wide growth rings, and microscopically by intercellular spaces between the rounded tracheids. Cells in these zones contain excessive amounts of lignin in the secondary wall and the S3 layer is

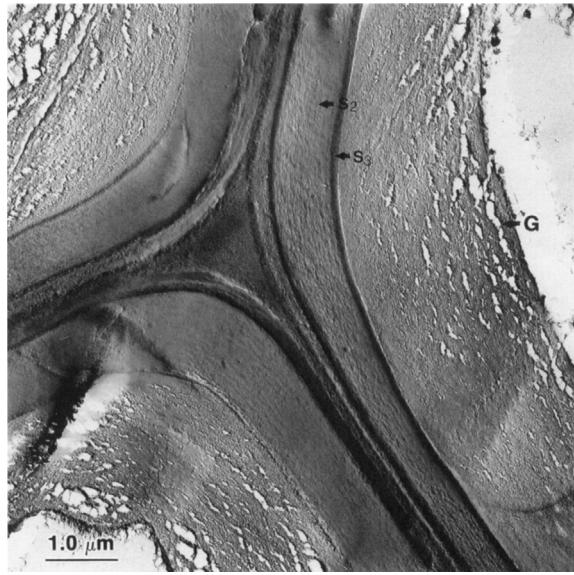


Figure 6.13 A cross-section of several gelatinous fibers of *Celtis occidentalis* showing the deposition of a G layer after formation of the S2 and S3. TEM, 14,400X. *Reproduced by permission of the Forest Products Journal and courtesy of W.A. Côté and Day (1962).*

lacking. The cellulose content is low. Finished lumber containing tension wood has a silvery sheen and a fuzzy surface due to pulled fibers. At the anatomical level, tension wood contains so-called gelatinous fibers. These cells have a cellulose rich cell wall layer (G-layer) that is lignified. The G-layer may develop after deposition of any one of the three wall layers, S-1, S-2 and S-3 (usually S2) (Fig. 6.13). Tension wood is more susceptible to some decay fungi, while compression wood is more resistant than normal wood, probably because of its high lignin content (35–40%).

The literature concerning the origins, anatomical features, and chemical constituents of compression wood and the effects of these characteristics on its utilization were summarized by Timell (1982, 1986).

Organizational levels in the cell wall

There are three general levels of organization in the cell wall and each profoundly affects access of microbial enzymes and moisture to the

chemical constituents. The gross capillary zone consists of the cell lumen, pit chambers, pit apertures and pit pores and has general size dimensions ranging from about 10 μm to 2000 Angstroms for the smallest pit pores. It has been estimated that a gram of wood possesses a square meter of gross capillary surface. The amorphous or transient capillary zone between the crystalline portions of the microfibrils has very small capillaries (up to 200 A in size). This zone is accessible to water in vapor form and is the chemically reactive portion of cell wall material. The transient zone is accessible to some enzymes and is the area where initial decay often develops. A gram of wood at this level is estimated to possess 300 square meters of surface area. The crystalline zone within the highly ordered cellulose crystals is initially inaccessible to water or enzymes. The dimensions of this zone are the distance between cellulose chains in unit cells and are in the order of 8XIO Å (Fig. 6.7). The mode of access of the enzyme molecules to the inner structure of the cell wall is an interesting question and can be introduced briefly at this point. Potential modes for accessing the cell wall include progressive digestion inward through the wall from the lumen surface, or rapid enzyme diffusion in a water film into the openings and cavities in the cell wall. The relative sizes of the available openings and the digestion enzymes are discussed further in Chapters 8 and 9 on wood decay.

Wood-water relationships

As previously discussed (Chapter 4), certain levels of moisture in wood are required for decay development. Moisture levels also determine the degree of wood swelling and shrinking. These changes can lead to the development of deep checks, particularly in roundwood products or poorly fitting joints in structures that we will see in the later chapters on decay are zones with increased risk of decay development. Understanding wood-moisture relationships is the key for decay control in many wood uses.

The moisture properties of wood result from both its chemical constituents and the capillary nature of the amorphous zones of the microfibrils that make up the cell walls. Water may occur in wood in liquid or vapor phases in the lumina of the cells and the pit cavities (gross capillary system). Water also may occur in a bound state (hydrogen bonding) on

wood surfaces, within and between the microfibrils in the amorphous zones. The zone between the microfibrils is termed the transient capillary system since it contracts when wood is dried and expands when it is moistened. This capillary system has a large surface area estimated to be at $6 \times 10\ cm^2/cm^3$ (Côté et al., 1968). Changing moisture levels in this zone affect many wood properties such as strength, swelling/shrinking, and electrical conductivity. This zone is also the point where fungal enzymes gain access inside the cell wall to initiate decay.

The principal water sources in wood are residual water that was in the living tree, liquid water from the atmosphere, contact with wet porous materials such as soil, condensation, and water vapor from the atmosphere.

It is useful to start with a wet board with a moisture content of 100% in order understand the forms of water in wood in relation to various uses and treatments. Water can enter wood in either liquid or vapor form, but primarily leaves as a vapor. Liquid water is removed from the surface under low humidity and/or high temperature conditions via capillary flow mechanisms and by evaporation from the capillary menisci as water vapor. The point where water is depleted from the cell lumina (gross capillary system), but the cell walls remain saturated and swollen, is termed the fiber saturation point (f.s.p.). The f.s.p. generally ranges from 25% to 35%, depending upon wood species and extractive content. As we will see later, the f.s.p. is a transition zone and might better be defined simply as the level where many wood properties change. Wood begins to shrink below the f.s.p. and most strength properties increase. Bound water in the transient capillary system evaporates from the wood below the f.s.p. via the now open gross capillary system as water vapor. This process requires that humidity near the surface be lower than that of the wood and is termed desorption. Initially, the water molecules removed are attached to other water molecules by hydrogen bonding, but the residual water molecules are held tenaciously to the surface of the microfibrillar capillary zone and their removal requires lower humidities or higher temperatures. About one-third of the moisture below the fiber saturation point is held tenaciously as a monomolecular layer by hydrogen bonding to the exposed hydroxyls on the cellulose and other biopolymers in the cell wall. The final bound water can be removed only by prolonged heating or after long time periods over desiccants and under vacuum conditions. Elevated temperatures increase the kinetic energy of the bound water molecules enhancing their release from the wood. Wood in which all the bound water is moved is called oven dry and the term oven-dry-weight (o.d.w.) designates this condition.

The reverse process of exposing oven-dry wood to water vapor is termed adsorption. A typical adsorption-desorption curve for wood at a range of relative humidities from 0% to 100% is shown in Fig. 6.15A. Wood below the f.s.p. reaches a series of equilibrium moisture contents (e.m.c.) at various temperatures and relative humidities termed sorption isotherms (Fig. 6.14).

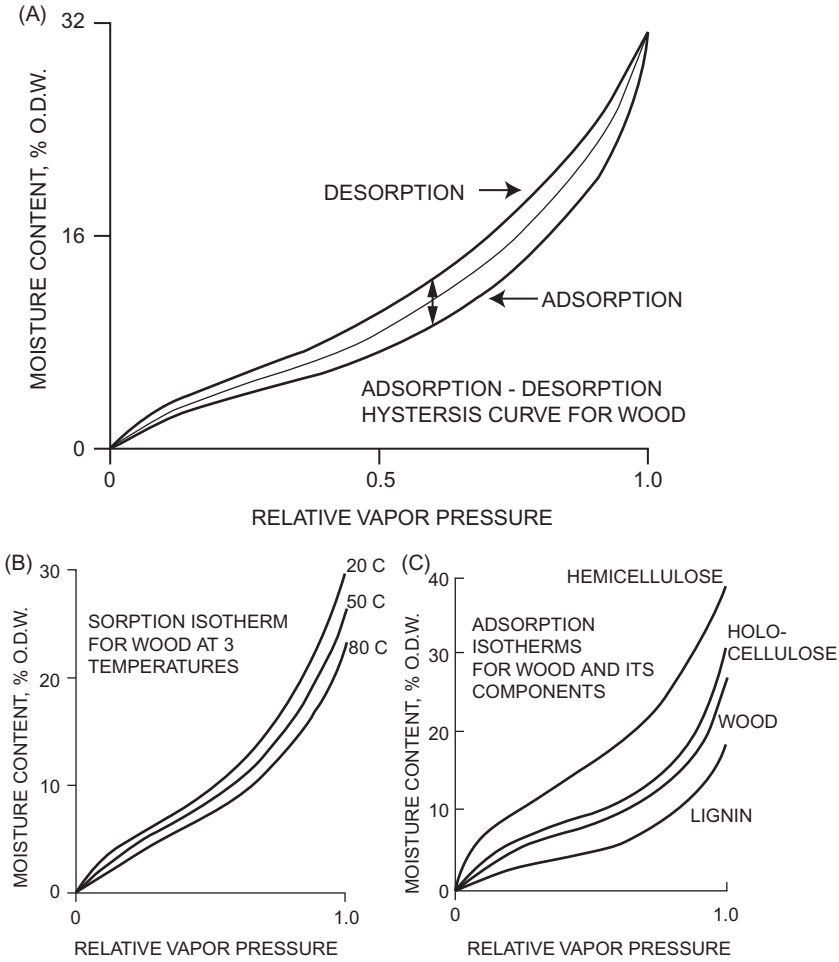


Figure 6.14 Wood-water sorption curves showing: (A) equilibrium moisture content curves for wood illustrating desorption and adsorption curves at various water vapor pressures at a given temperature, (B) effects of temperature on the sorption characteristics of wood, and (C) curves indicating the different adsorption isotherms for the major wood cell wall components. *From Skaar, C., 1972. Water in Wood. Syracuse, University Press, Syracuse, N.Y., pp. 218, with permission from Syracuse University Press.*

The e.m.c.'s attained are 1−2% higher when wood is equilibrated by desorption (from f.s.p. to e.m.c.) than absorption (o.d.w. to e.m.c.). This is termed hysteresis and is a characteristic of other colloidal materials such as agar (Fig. 6.14A). The hysteresis effect is a common source of error in wood decay experiments when differences in e.m.c. weights before and after decay are used to determine decay capabilities without repeating the original drying direction.

The various chemical constituents in the wood cell differ substantially in e.m.c. Hemicelluloses equilibrate at the highest and lignin the lowest e.m.c. values (Fig. 6.14C). Some other interesting features of wood sorption isotherms include:

(a) Wood at low moisture contents is a very hygroscopic material and gains weight rapidly in the presence of water vapor.

(b) Wood which that has been heated or kiln-dried subsequently attains lower e.m.c. values than non-heated wood.

(c) Wood attains higher e.m.c. values at low temperatures than at high temperatures at the same temperature-relative humidity conditions.

(d) Wood with high extractive contents generally attains lower emc values.

(e) Hardwoods attain higher emc values than conifers, probably reflecting the higher hemicellulose and lower lignin contents of hardwoods.

(f) The slope of the sorption isotherm curves are sigmoidal, which may reflect the difficulty of breaking hydrogen bonding of water molecules to cell wall constituent hydroxyls at the low end of the curve and the ease of water removal from the gross capillary system at the high end of the curve.

The sorption characteristics of wood reflect the strong attraction of water vapor molecules for the exposed hydroxyl groups (OH) that cover the surfaces of the cellulose and hemicellulose molecules in the non-crystalline (amorphous) zones of the microfibrils. Examples of hydrogen bonding between two hydroxyl groups and between an OH and H_20 molecules are diagrammed in Fig. 6.15. Intermolecular hydrogen bonding between hydroxyls on adjacent cellulose molecules plays a role in hysteresis and in the aggregations of cellulose molecules into the supramolecular structures termed "microfibrils". The amorphous zone retains about one water molecule per available hydroxyl, accounting for approximately 25% of the f.s.p.

Wood swells or shrinks as it gains or loses moisture. The swelling begins just above the o.d.w. condition and ends when the f.s.p. condition

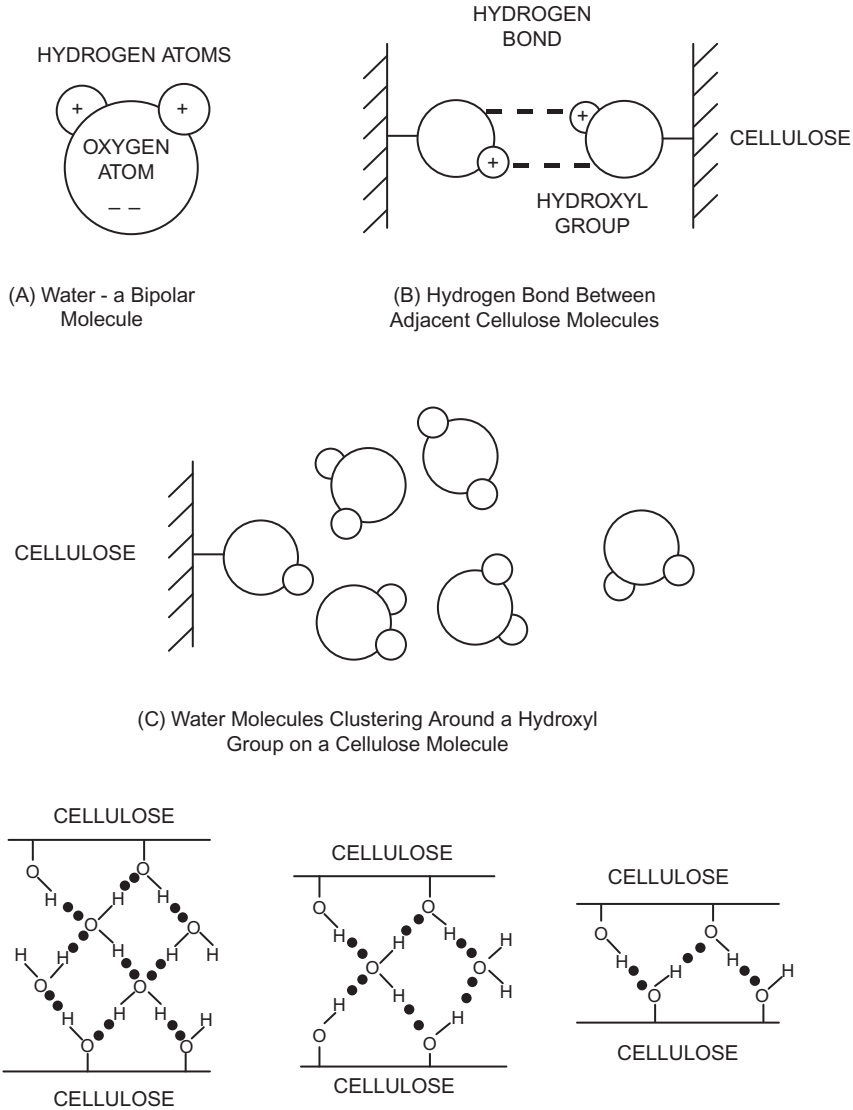


Figure 6.15 Diagrams illustrating the causes and nature of hydrogen bonding. (A) The bipolar nature of the water molecule, (B) Hydrogen bonding between hydroxyls on adjacent cellulose molecules, (C) Water molecules clustering by hydrogen bonding around a hydroxyl on a cellulose molecule, and (D) Reduction of hydrogen bonding as water molecules evaporate during wood drying. *Sources Skaar, C., 1972. Water in Wood. Syracuse. University Press, Syracuse, N.Y., pp. 218, with permission Syracuse University Press; Fengel, D., Wegener, G., 1984. Wood Chemistry, Ultrastructure, Reactions. Walter de Gruyter, New York, pp. 613, with permission Walter de Gruyter & Co.*

Table 6.4 Examples of equilibrium moisture contents of wood exposed inside a building in various locations in the U.S.

City	Moisture content of interior woodwork (% oven dry weight basis)	
	July	January
Atlanta, Georgia	11.5	8.5
Albuquerque, New Mexico	6.0	7.0
Boston, Massachusetts	13.0	7.0
Madison, Wisconsin	10.0	6.0
New Orleans, Louisiana	13.5	12.5
San Francisco, California	10.5	10.5
Washington, D.C.	11.0	8.0

Source: Peck, E.C., 1950. Moisture Content of Wood in Use. U.S.D.A., Forest Service Forest Service, Forest Products Laboratory Report 768. Madison, Wisconsin.

is attained. Shrinkage begins at the f.s.p. and ends at the o.d.w. condition. The dimensional changes associated with variations in moisture content are essentially limited to the tangential and radial planes of the wood and are caused by swelling by the transient capillary system as it adsorbs or desorbs water molecules. No significant dimensional changes occur in the longitudinal wood plane of normal wood since the cellulose, a long linear polymer, are primarily oriented longitudinally and their chain length does not change appreciably with wetting.

Repeated differential dimensional changes in wood tangentially and radially create differential stresses that lead to the formation of radial splits or checks during the seasoning or use of some wood products. Checks may become severe in roundwood materials such as poles or piling, particularly when they develop after preservative treatment. Checks in relation to decay development are discussed in Chapters 13 and 15.

A major decay control principle is evident from the water sorption characteristics. Wood protected from free water sources never exceeds the f.s.p. and wood kept below the f.s.p. cannot decay (Table 6.4).

Wood variability

Wood is an extremely variable material that often requires the application of substantial safety factors in critical structural uses. As reviewed in

this chapter, wood variability is both structural and chemical. Structural variability reflects the many cell types and organizational patterns, while chemical variation represents differences in the types and amounts of cell wall constituents. The selection of certain species from the hundreds of tree species for special uses such as lumber, pulpwood, veneer, piling, and poles reflect properties such as strength, grain pattern, density, hardness, treatability, and form that are specific for that species.

Many wood properties are based on genetic traits, but they may still vary considerably within a species. These traits form the basis for tree breeding or tree improvement programs and the selection of seed stock with rapid growth rates, high density, disease resistance, durability, or improved stem form.

Variations in wood properties within a single stem are of special interest because they may affect patterns of decay development or decay susceptibility in various wood products. These variations also make it important to carefully select representative wood samples for various testing purposes. Some of the many variations in wood properties include:

(a) Stemwood is very different structurally and chemically from rootwood and upper branch wood.

(b) Nitrogen distribution varies with radial position and is highest in the outer sapwood and pith zones.

(c) Extractive contents show extreme variability within the heartwood both radially and longitudinally.

(d) Many species contain a juvenile wood in the stem center that has lower fiber lengths and densities.

(e) Reaction wood, depending on stem position and wood origin, can severely alter wood properties.

(f) Wood densities vary with growth rates and, within certain ranges, rapid growth is associated with high density woods in some hardwoods (typically ring porous woods) and low density in conifers.

(g) The handling and processing of wood may alter properties, e.g. high temperature decreases hygroscopicity and pond storage may increase porosity.

(h) Prior invasion of wood by non-pigmented mold fungi or the early stages of invasion by decay fungi may significantly affect properties such as porosity and strength.

These examples highlight wood variability and stress the need for having a thorough knowledge of wood properties whenever designs incorporating this material are considered.

Summary

1. Wood consists primarily of a series of thick walled, elongated cells, with the dual functions of fluid conduction and structural support for the tree stem. Both of these functions are carried out by tracheids in conifers and by vessels (conduction) and fibers (strength) in hardwoods. About 90% or more of the biomass of most commercial woods are composed of tracheids or fibers.
2. Parenchyma cells perform storage (starch and oils) or secretory functions (resins and gums) and form the wood rays that are radially aligned tissues interspersed between the axial elements.
3. Wood cells are interconnected by a series pits in the secondary walls that permit passage of water and solutes among contiguous cells. The pits are the primary invasion routes for many wood-inhabiting fungi. The parenchyma cells are the major colonization site for the fungi that discolor the sapwood.
4. Wood cell walls consist primarily of the large macropolymers, cellulose, hemicelluloses, and lignin. Cellulose is a long linear polymer consisting of β-D-glucose units connected by (1-4) glycosidic linkages that forms the cell wall framework. Cellulose occurs in plant cell walls as bundles of parallel-aligned molecules termed microfibrils. Microfibrils contain alternating crystalline and non-crystalline or amorphous zones. Hemicelluloses are shorter, linear molecules containing hexose or pentose sugars as the monomer units. The monomer units in the main chain are, as in cellulose, connected by (1-4) glycosidic linkages. Some hemicelluloses are branched and most contain side chains. The hemicelluloses are deposited around the microfibrils and form the cell wall matrix. Lignin is an aromatic polymer formed by free radical polymerization of three types of cinnamyl alcohols. This constituent is a huge amorphous polymer without a regular structure and forms an interpenetrating polymer system around and between the hemicellulose coated microfibrils of cellulose. As a cell wall encrustant, lignin provides stiffness and strength. Lignin also is a very durable material and acts as a barrier against microbial attack of the more vulnerable carbohydrates in the cell wall.
5. The cell walls of tracheids or fibers consist of a primary wall (PW) laid down initially during cell maturation followed successively by the three layers of the secondary wall *(Sl, S2, S3)*. The S2 is the

major cell wall zone and the micro fibrils in this zone are oriented nearly parallel with the stem axis. The S2 layer of the wall has the most significant impact on wood strength, density, and moisture properties.

6. The types and distribution of the major chemical constituents in the wood cell wall vary among and within species, as well as within a single cell. Cellulose content is relatively constant across all wood species and makes up about 40–50% of wood biomass. Hemicelluloses are more abundant in hardwoods than conifers, while lignin is present at higher levels in conifers than hardwoods. Exceptions to these trends occur in some tropical woods. In addition to varying levels of constituents, the types of hemicelluloses and lignin differ between conifers and hardwoods. Cellulose levels are highest in the secondary cell wall, hemicelluloses levels are highest in the S1, and lignin is present at highest levels in the middle lamella and primary wall (compound middle lamella).
7. The three organizational levels in the cell wall are the gross capillary, transient capillary, and crystalline zones. Liquid water or water vapor can occur in the gross capillary zone that consists of the lumina of cells and pit cavities. Water occurs in the transient capillary zone as bound water (hydrogen bonding). This zone includes the amorphous or non-crystalline zones of the microfibrils. The transient capillary zone contracts as wood dries and expands as it is moistened between the oven dry and fiber saturation levels.
8. Wood attains a series of equilibrium-moisture contents at various temperature-vapor pressure conditions within the oven-dry and fiber saturation point range.
9. Many special properties of wood can be explained in part by the structure of cellulose and its primary orientation in the cell wall. The anisotrophy or swelling and shrinking of wood in only two planes (radial and tangential) is due to the unique exposed position of the hydroxyls on the cellulose molecules and their hydrogen bonding with hydroxyls on adjacent cellulose molecules or available water molecules. The high tensile strength of wood is due to the longitudinal orientation of the cellulose molecules, their high degree of polymerization, and the covalent bonds between the glucose units.
10. The sorptive properties of wood clearly indicate that wood protected from free water sources cannot attain sufficient moisture to sustain microbial colonization.

References

Adler, E., 1977. Lignin chemistry --past, present, and future. Wood Sci. Technol. 11 (3), 169–218.

Agrios, G.N., 1988. Plant Pathology 3rd Edition, Academic Press.

Core, H.A., Côté, W.A., Day, A.C., 1976. Wood Structure and Identification. Syracuse University Press, Syracuse, N. Y.

Côté Jr., W.A. (Ed.), 1965. Cellular Ultrastructure of Woody Plants. Syracuse University Press, Syracuse, N.Y.

Côté Jr., W.A., 1977. Wood ultrastructure. In: Loewus, F.A., Runeckles, V.C. (Eds.), Recent Advances in Phytochemistry vol. II. The Structure, Biosynthesis, and Degradation of Wood. Plenum Publishing Corp., New York.

Côté, W.A., 1981. Ultrastructure - critical domain for wood behavior. Wood Sci. Technol. 15, 1–29.

Côté, W.A., Day, A.C., 1962. The G layer in gelatinous fibers - electron microscope studies. For. Products J. 13 (7), 333–338.

Côté, W.A., Timell, T.E., Zabel, R.A., 1966. Studies on compression wood. Part 1. Distribution of lignin in compression wood of red spruce, *Picea rubens* Sarg. Holz als Roh-Werkstoff 24, 432–438.

Côté, W.A., Day, A.C., Timell, T.E., 1968. Distribution of lignin in normal and compression wood of tamarack. Wood Sci. Technol. 2, 13–37.

Esau, K., 1965. On the anatomy of the woody plant. In: Côté Jr., W.A. (Ed.), Cellular Ultrastructure of Woody Plants. Syracuse University Press, Syracuse, New York, pp. 35–50.

Fengel, D., Wegener, G., 1984. Wood Chemistry, Ultrastructure, Reactions. Walter de Gruyter, New York, p. 613.

Glasser, W.G., Glasser, H.R., Morohoshi, N., 1981. Simulation of reactions with lignin by computer (SIMREL). Macomolecules 14, 253–262.

Goring, D.A.I., Timell, T.E., 1962. Molecular weight of native celluloses. Tappi 45, 454–460.

Harada, H., Côté Jr., W.A., 1985. Structure of wood. In: Higuchi, T. (Ed.), The Biosynthesis and Biodegradation of Wood Components. Academic Press, New York, pp. 1–42.

Higuchi, T. (Ed.), 1985. Biosynthesis and Biodegradation of Wood Components. Academic Press, New York.

Hoadley, R.B., 2000. Understanding Wood. The Taunton Press, Newton, CT.

Kollman, F.F.P., Côté Jr., W.A., 1968. Principles of Wood Science and Technology, Vol. I. Solid Wood. Springer-Verlag, Berlin, p. 592.

Nimz, H., 1974. Beech lignin-proposal of a constitutional scheme. Angew. Chern. (Int. Ed. Engl.) 13, 313–321.

Panshin, A.J., deZeeuw, C., 1980. Textbook of Wood Technology, fourth ed. McGrawHill Book Co., New York, p. 772.

Peck, E.C., 1950. Moisture Content of Wood in Use. U.S.D.A., For. Serv Forest Service, Forest Products Laboratory Report 768, Madison, Wisconsin.

Saka, S., Goring, D.A.I., 1985. Localization of lignins in wood cell walls. Chapter 3. In: Higuchi, T. (Ed.), Biosynthesis and Biodegradation of Wood Components. Academic Press Inc, New York, pp. 51–62.

Shigo, A.L., Shigo, A., 1974. Detection of Discoloration and Decay in Living Trees and Utility Poles. USDA, Forest Service Research Paper, NE-294. Northeastern Forest Experiment Station, Forest Service, U.S.D.A, Broomall, PA.

ShortIe, W.C., 1982. Decaying Douglas-fir wood: ionization associated with resistance to a pulsed current. Wood Sci. 15 (1), 29–32.

Siau, J.F., 1984. Transport Processes in Wood. Springer-Verlag, New York, p. 245.
Skaar, C., 1972. Water in Wood. Syracuse University Press, Syracuse, N. Y, p. 218.
Timell, T.E., 1964 Wood hemicelluloses. Part I. Advances in Carbohydrate Chemistry 19:247–302.
Timell, T.E., 1965. Wood hemicelluloses. Part II. Advances in Carbohydrate Chemistry 20: 409–483.
Timell, T.E., 1969. The chemical composition of of tension wood. Svenska Papperstidning 72:173–181.
Timell, T.E., 1982. Recent progress in the chemistry and topochemistry of compression wood. Wood Sci. Technol. 16, 83–122.
Timell, T.E., 1986. Compression Wood in Gymnosperms (in 3 Volumes). Springer-Verlag, New York, p. 2150.
Wardrop, A.B., 1964. In: Zimmerman, M.H. (Ed.), The Formation of Wood in Forest Trees. Academic Press, New York, p. 8788., pp. 562.
Young, H.E., Quinn, V.P., 1966. Chemical elements in complete mature trees of seven species in Maine. Tappi 49, 190–197.

Further reading

Browning, B.L. (Ed.), 1975. The Chemistry of Wood. Krieger Pub. Co., Huntington, N.Y.
Côté, W.A., 1976. Wood Ultrastructure -An Atlas of Electron Micrographs. University of Washington Press, Seattle, WA, p. 64.
Cowling, E.B., 1965. Microorganisms and microbial enzyme systems as selective tools in wood anatomy. In: Côté Jr., W.A. (Ed.), Cellular Ultrastructure of Woody Plants. Syracuse University Press, Syracuse, New York, pp. 341–368.
Crawford, R.L., 1981. Lignin Biodegradation and Transformation. John Wiley and Sons, New York, p. 154.
Kirk, T.K., Farrell, R.L., 1987. Enzymatic "combustion": the microbial degradation of lignin. Annu. Rev. Microbiol. 41, 465–505.
Timell, T.E., 1965. Wood and bark polysaccharides. In: Côté, W.A. (Ed.), Cellular Ultrastructure of Woody Plants. Syracuse University Press, Syracuse, New York, pp. 127–156.
Wardrop, A.B., 1965. Cellular differentiation in xylem. In: Côté Jr., W.A. (Ed.), Cellular Ultrastructure of Woody Plants. Syracuse University Press, Syracuse, N.Y, pp. 61–124.
Zimmerman, M.H. (Ed.), 1964. The Formation of Wood in Forest Trees. Academic Press, New York, N.Y.
Zimmerman, M.H., Brown, C.L., 1971. Trees Structure and Function. Springer-Verlag, Berlin, p. 336.

CHAPTER SEVEN

General features, recognition, and anatomical aspects of wood decay

Contents

The characteristic softening, discoloration, and eventual disintegration of wood in some uses is well known as decay or rot to most wood processors and users. Colloquial terms in some regions used to describe decay include doze, dote, punk and stack burn. A broad dictionary definition of decay implies slow changes in a material that reduce its useful properties resulting from organism actions, e.g. insects, borers, and fungi or abiotic factors such as weathering.

Decay has a more restrictive and specific meaning in wood microbiology. In this textbook, we will define decay as significant changes in the physical and chemical properties of wood caused by the chemical

Wood Microbiology.
DOI: https://doi.org/10.1016/B978-0-12-819465-2.00007-3

(primarily enzymatic) activities of microorganisms (primarily higher fungi). Since rot is commonly used by laymen to describe decay we will continue to use these terms interchangeably.

This chapter will review the general features of wood decay with an emphasis on decay types, decay detection from various macroscopic and microscopic evidence, and the anatomical features of decay development.

The dual nature of decay

It is important to note that the term decay is used to describe both a process and an outcome. For example, an inspector judges the physical condition of a board to be sound or decayed. In this sense, wood decay is viewed as a property of a material related to its usefulness. The second, broader meaning is the process of wood digestion by microorganisms including the enzymes, the oxidants and the changes in the wood constituents involved. In this sense, decay is a verb and can be viewed as the external digestion of a complex organic material by microorganisms. Decay is generally a slow process requiring weeks and sometimes years to disintegrate wood, but, small wood blocks, under ideal decay conditions, may be consumed totally within a month. At the other extreme, decays developing in the heartwood (heartrots) of durable trees may sometimes develop over hundreds of years. Decay, by definition, must cause significant changes in wood properties. We generally consider these changes to include measurable losses in cell wall biomass.

General features of wood decay stages

The decay process, under ideal conditions, is a continuum that begins when a few innocuous spores land on a substrate and ends when the wood is destroyed or mineralized. Points along the continuum have been selected arbitrarily to designate various stages in decay development that, as we will see later, can be related to various use properties (Fig. 7.1). The decay process begins when the hyphae of a decay fungus penetrates wood, initiates colonization, and releases enzymes. Damage is

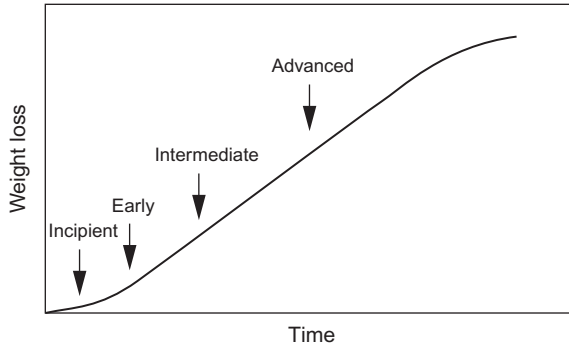


Figure 7.1 Diagram illustrating the continuum of the decay process under ideal decay conditions and its subdivision into several decay stages.

limited in the early colonization phase, and since there is no visible evidence of damage, it is termed the incipient or hidden stage of decay. As decay develops, slight changes in color, wood texture, and fiber brashness may appear and these changes constitute the early stage when decay is detectable, but not obvious. Obvious changes in wood color and texture become evident as decay continues to the intermediate stage, but the gross wood structure still remains intact. The late stage is characterized by total disruption of the wood structure leaving the residual wood a brownish amorphous, whitish punky or fibrous material. Some fungi (white rots) may completely degrade the wood, producing weight losses approaching 96–97%. Other decay fungi degrade only the carbohydrate portions of the wood cell walls and cause maximum weight losses of 60–70%. Given the myriad of factors involved in the decay process including wood type, fungal species, and environmental conditions, it is virtually impossible to accurately estimate when decay in a given piece of wood in service began.

Macroscopic features: The appearance of wood at the intermediate and late stages of decay is extremely variable reflecting the differences in how different fungi attack wood and the characteristics of the wood being degraded. The residual wood may be a white, gray, or brown and the color consistent or mottled. The wood texture may be soft, spongy, feathery, or fibrous for the white rot decayers or consist of loosely adhering soft-brown cubes for the brown rot decayers. Narrow black to brownish zone lines or the black lines of decay may be associated with the white rot fungi. These lines can represent either boundaries of non-decayed wood between genetically different fungi or mycelial responses to surface

drying (Rhoads, 1917; Campbell, 1933; Hopp, 1938). The wood may be uniformly decayed or decay can be concentrated in localized pockets. In some cases, the decay may be concentrated along the earlywood bands of the annual ring, forming a condition call red ring rot. Decay can lead to separations along the annual rings or laminated rot. In other decays, wood rays or fibers may be selectively removed, forming shot-holes or producing a stringy texture in the residual wood. Many macroscopic features of decay appear to be characteristic of the causal fungi. For example, *Phellinus* (*Fomes*) *pini* causes a white-pocket decay and often a ring shake in conifers; *Postia amara* (*Polyporus amarus*) causes a brown-cubical pocket decay in incense cedar; *Echinodontium tinctorium* forms a brown-stringy decay in western hemlock; and *Ganoderma* (*Fomes*) *applanatum* causes a white-mottled decay and produces abundant zone lines in hardwoods. Some of these macroscopic features of decay and their causal fungi are shown in Fig. 7.2.

Decay patterns: The location and stages of decay in various wood types and uses may vary greatly with degree of decay hazard and the initial invasion point. Stem decays in living trees are often concentrated in the more susceptible heartwood. A cross section through a decaying stem often shows all decay stages ranging from none in the outer sapwood to complete wood removal in the heartwood (Fig. 7.3). In treated woods such as railroad ties or poles, deep-drying checks often penetrate through the outer-treated shell and a characteristic decay pattern forms between the treated and untreated zones punctuated by advanced decay where the check enters the untreated wood. Decay patterns will be discussed in more detail in the subsequent chapters on decays associated with various wood uses. Understanding decay patterns provides useful clues to the wood user on when and where a decay fungus entered or why a protective treatment failed.

Major disadvantages of decay: Strength and volume (biomass) reductions are the principal losses associated with decay. Drastic strength losses occur in the incipient (hidden) decay stage with some decays. Failure to detect early decay can lead to serious strength losses in some high stress uses of wood such as ladder rails or poles. Wooden ladders provide an excellent example of the consequences of failure to detect early decay. Incipient decay in a ladder reduces strength to the point where a ladder rung fails during use. Personal injury lawsuits resulting from these failures resulted in the nearly complete shift to fiberglass or aluminum ladders.

Drastic alterations in the dimensions or chemical and physical properties of wood at the intermediate and late stages of decay adversely affect

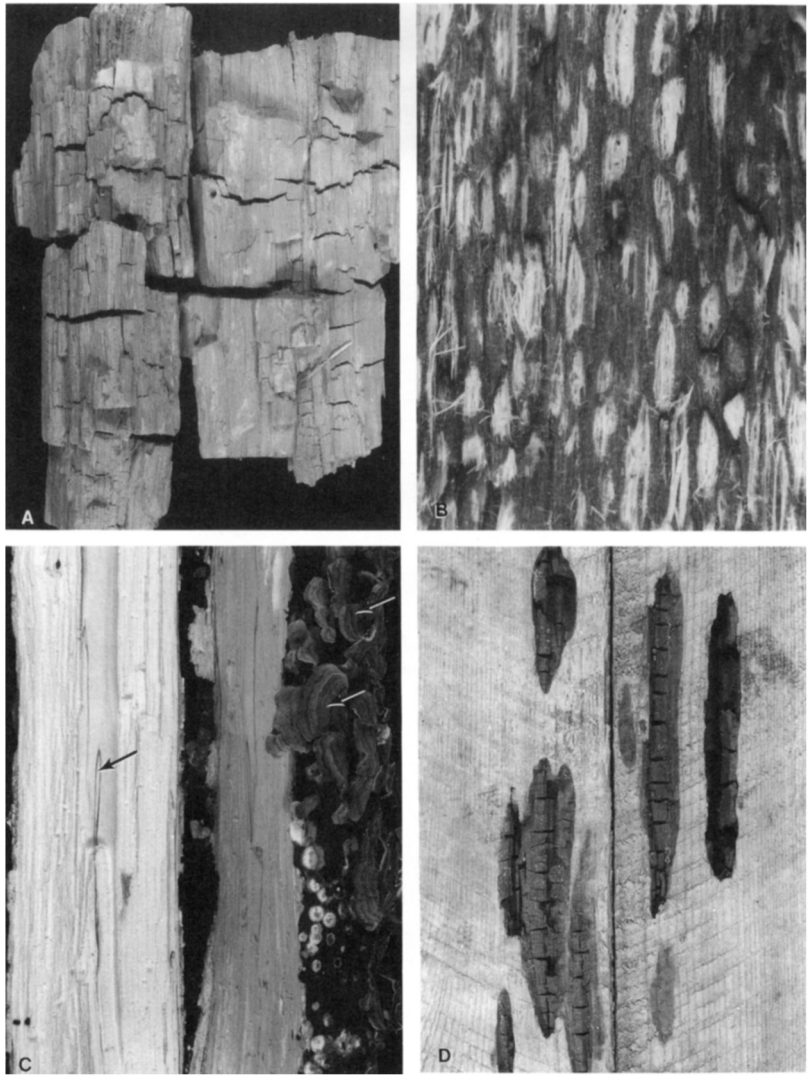


Figure 7.2 Some typical features of several major types of decay based upon color and texture (A) brown cubical rot, (B) white pocket rot, probably an advanced stage of *Phellinus pini* decay, (C) a white spongy rot caused by *Trametes versicolor* (arrows indicate basidioma and zone lines), and (D) a brown pocket rot in incense cedar caused by *Postia amara*.

many use properties. These changes include decreased volume and mass resulting in dimensional collapse of supporting timbers or loss of raw materials for chemical processes, reductions in many strength properties (tension, bending, and compression), increased hemicellulose solubility

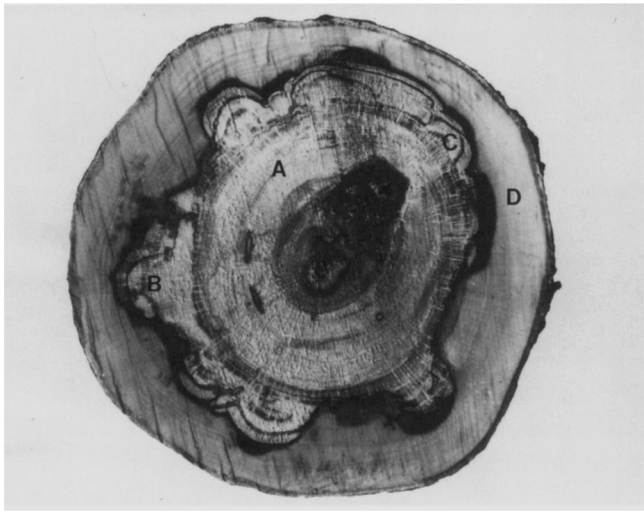


Figure 7.3 Cross section of American beech (*Fagus grandifolia*) stem showing various decay stages and macroscopic features of *Phellinus igniarius*, a major heart-rot fungus of hardwoods. Letters indicate (A) advanced decay, (B) early decay, (C) zone lines, and (D) sound sapwood (Magn. X33).

leading to substantial losses during pulping and related water pollution problems, increased permeability to liquids leading to excessive absorption of preservatives or coatings, changes in electrical properties (increased conductivity), and aesthetic losses due to abnormal colors, rough textures, or pulled fibers. These property changes are discussed in more detail in Chapters 8–10.

Decay losses: Collectively, decay losses are very large and include those incurred during timber growth, wood storage, processing, and in many product uses. Losses can be difficult to quantify because even damaged wood may be sold for some other use and there is no system for collecting loss data for timber in service.

Thus, loss estimates are available only for some parts of the total. Losses in standing timber due to stem decays are estimated to be 15–25% of total volume. These losses have likely decreased as over-mature stands were utilized and the average timber stand age decreased in more intensively managed forests. Losses during the storage and conversion of logs, pulpwood, and chipwood piles are estimated to be 15% of total timber harvested. The major decay losses, however, occur in wood products and have been estimated to represent a replacement cost of 10% of the annual cut. This cost does not include the labor of replacement, service

interruptions, inconvenience, or the serious injuries and losses that can occur when a structure fails. Decay in structures often occurs in locations where access and repair are difficult (e.g. foundation plates, floor joists, support beams, etc.) and replacement costs can sometimes approach twenty-five times the original wood cost. The specific decay losses associated with various applications are discussed in subsequent chapters. Accurate decay loss estimates are necessary to justify decay reduction programs in the major wood-use industries and encourage research on more effective decay controls. Substantial reductions in decay losses could be effected if known information and recommendations on decay control were applied by wood users and the designers of wooden structures.

While this textbook stresses the economic losses associated with some wood uses, in the aggregate, it should be remembered, that decay is beneficial in forest ecosystems. Slash removal, soil enrichment, and returning carbon dioxide to the atmosphere for green plant use in the carbon cycle are all dependent upon wood decay to recycle these nutrients. This recycling is both beneficial and potentially detrimental since wood sequesters carbon from the atmosphere, while decay returns this carbon to the atmosphere. Forests have been projected to play a role in reducing carbon dioxide levels through sequestration and retaining this carbon in wood products offers the added advantage of prolonging that sequestration.

Recognition of decay (visual evidences)

The rapid detection of decay in the field is important during the purchase of unprocessed wood (e.g. logs, poles, pulpwood, posts, etc.), and the selection of wood for some products (e. g. veneer, structural lumber, etc.). Decay detection is particularly important during the grading and selection of wood destined for high stress uses such as ladder rails, laminated beams and poles. Early decay detection is also important during the periodic inspections of wood used under high decay hazard conditions such as utility poles, pilings, and cooling towers. Failure to recognize visible evidence of decay in the wood product often becomes a key issue in litigation, where it is used to determine responsibility for wood failure and related damage.

Decay detection in wood is intrinsically difficult since it often occurs in the interior of an opaque material, frequently in inaccessible zones

(below ground). Decay evidence in the incipient and early stages may be subtle and difficult to distinguish from natural variations present in sound wood.

Visually detecting the later stages of decay when the interior wood is exposed on cut surfaces such as log ends, boards, or increment cores can be relatively easy. However, visual detection of decay in the early stages is difficult and requires both a familiarity with the macroscopic features of early decay as well as an awareness of the natural variations in wood.

Macroscopic decay evidences

Useful evidence of early decay on wood surfaces include:

a) *Changes from normal wood color patterns*. Wood in the early stages of decay can be discolored red, pink, purple, yellow, gray, brown, or white. Color changes generally do not follow annual rings, often distinguishing them from sapwood-heartwood color differences. A bleached or mottled appearance is a common feature of the early stages of some white rots (e.g. *Trametes versicolor* or *Gandoderma applanatum*).

b) *Black lines of decay* (zone lines) between the sound and decayed wood or between different decay fungi (Fig. 7.4A). Zone lines are a positive evidence of early decay that occur most commonly with white rots on hardwoods.

c) *Abnormal shrinkage patterns*, as evidenced by wood collapse and development of numerous small surface checks. The checks generally form at right angles to the longitudinal plane of the wood and are most evident after the wood surface has dried. Cross-checks are a common feature of brown rots, reflecting the destruction of the long chains of cellulose (Fig. 7.4B), but are also common in soft rotted wood.

d) *Brashness or brittleness* of the wood as evidenced by roughened-saw cuts on exposed log ends, pulled fibers during lumber surfacing, or extreme brittleness of long slivers lifted from the wood surface with a probe or pick are all indicators of decay (Fig. 7.4C,D).

e) *Physical presence of the fungus* on the wood can include mycelial fans on board surfaces, punk knots in boards, or basidiocarps on logs. The mycelial fans that form on wood surfaces under moist conditions or between boards often assume intricate fan-like patterns and are known among wood tradesmen as "fungal flowers". A characteristic fungus odor (similar to commercial mushrooms) is often associated with

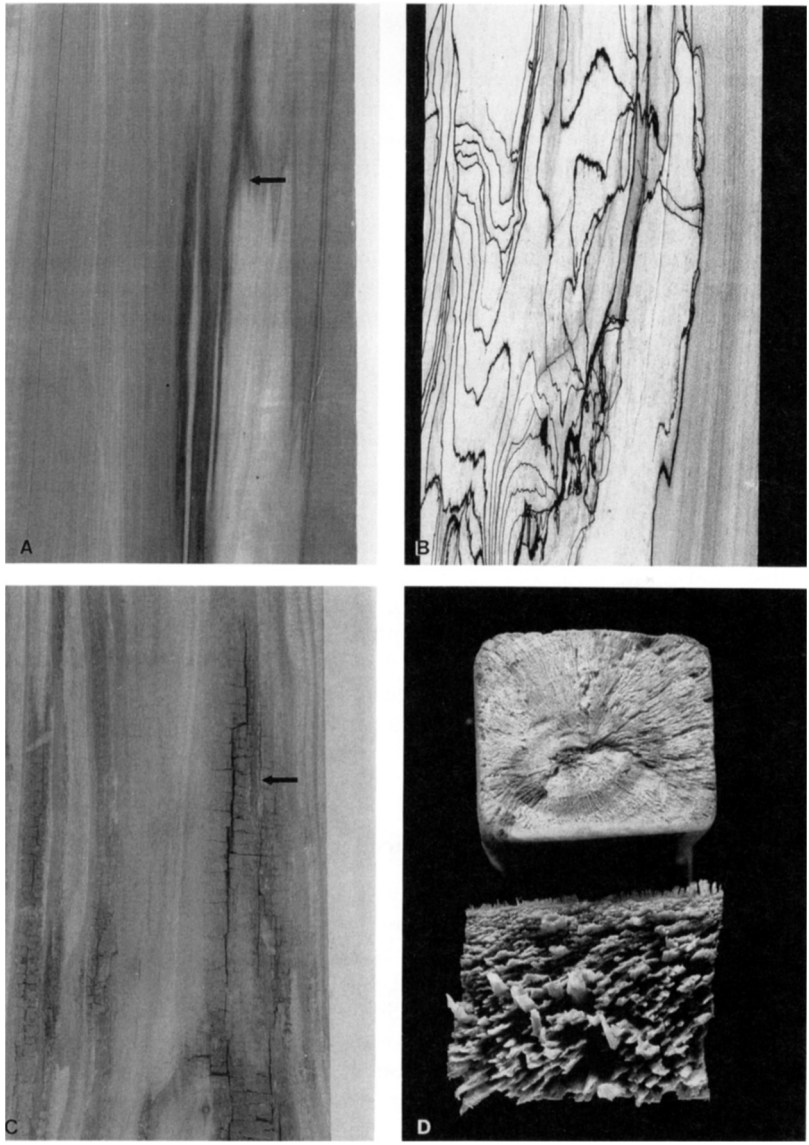


Figure 7.4 Macroscopic features of wood decay including (A) mottled white discoloration and zone lines associated with stem decay in sweet gum, (B) black zone lines associated with the white rot *Xylaria polymorpha*, (C) abnormal shrinkage cracks on the surface of a Douglas-fir board solid piled for several months with inadequate seasoning, and (D) a brash break in decayed beech (top) contrasted with a fibrous or splintered break in sound wood (bottom).

mycelial fans. Decayed knots in boards associated with abnormal discolorations often signal extensive zones of incipient decay. Color changes and brashness are only presumptive evidences of early decay.

Zone lines and the physical presence of the fungus can generally be safely assumed to be definitive indicators of early decay (Fig. 7.5). Fruiting bodies, while an indicator of fungal colonization are not necessary a sign of advanced decay nor are they an indicator that the fungus producing the fruiting bodies necessarily caused the decay. Combinations of decay indicators often occur, increasing the reliability of a positive decay diagnosis.

Microscopic decay evidences

Microscopic features of early decay, such as removal of pit membranes, cell wall erosion, bore holes, or hyphae with clamp connections, are definitive evidences of early decay (Fig. 7.5). Localized destruction of ray-parenchyma cells or the formation of axially-aligned diamond shaped or long linear cavities in the secondary wall are also useful decay detection criteria. Microscopic features of decay are discussed in more detail later in this chapter under the anatomical aspects of decay. Microscopic features can reliably confirm decay and are useful to confirm presumptive macroscopic features. It should be remembered that decay does not occur uniformly throughout the wood. Reliable decay detection depends on adequate sampling to minimize the chances of failing to detect early decay when it is present.

Other decay detection procedures

Detection of early decay can also be accomplished by several non-visual methods. Isolation of the associated fungi and subsequent identification from cultural features has long been a useful method for detecting viable decay fungi in wood (Wang and Zabel, 1990; Eslyn, 1979; Graham and Corden, 1980). More recent developments in DNA sequencing that allow identification of fungal taxa using biochemical procedures have greatly reduced the need for specialists with extensive knowledge of specific taxonomic characteristics. Isolation of fungi on specialized media has long been the preferred method for assessing fungal colonization of wood; however, it has long been known that not all fungi are culturable.

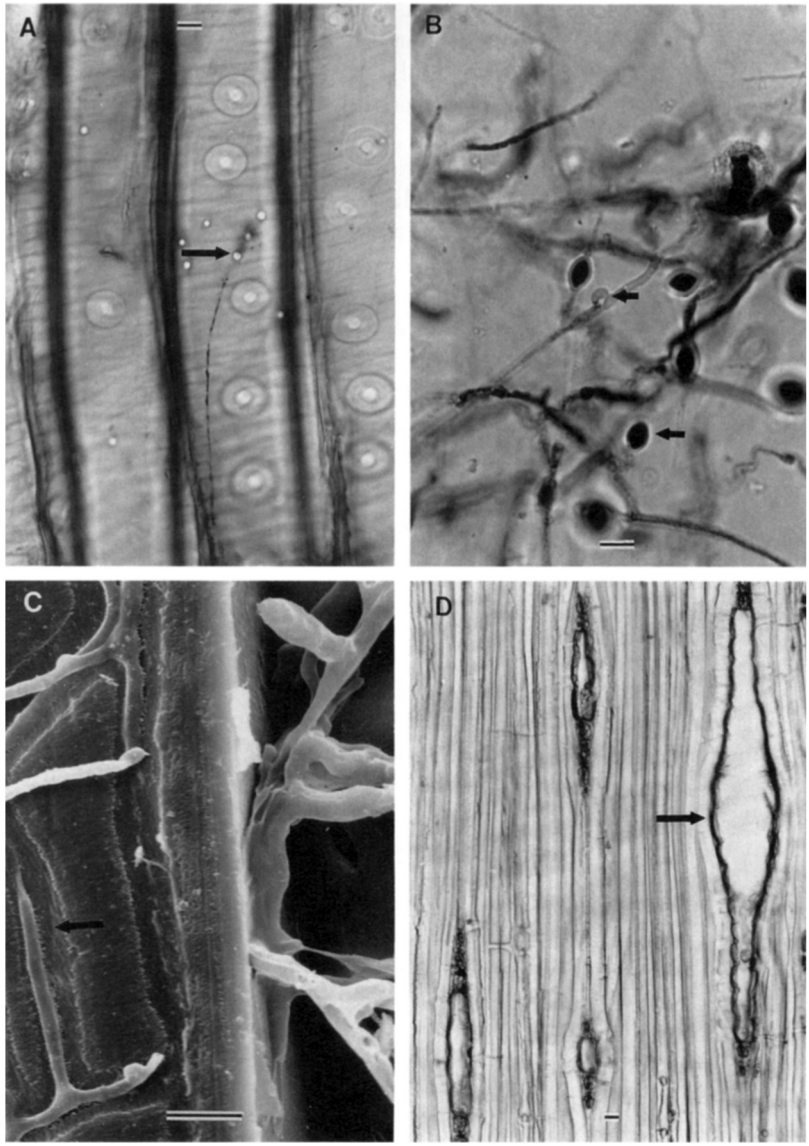


Figure 7.5 Microscopic features of decay including (A) bore holes associated with early decay in Douglas-fir caused by *Rhodonia placenta*, (B) hyphae of *Antrodia carbonica* showing a typical clamp connection and numerous ovoid-shaped chlamydospores, (C) transmission electron micrograph of cell wall erosion associated with early decay in southern pine caused by *Trametes versicolor*, and (D) selective destruction of parenchyma cells by *R. placenta* in early decay of southern pine. Bars = 10 μm. *(C) Reprinted by permission Electric Power Research Institute, Palo Alto, CA.*

Emerging techniques for sequencing all fungal DNA present in a sample create enormous opportunities to detect these other organisms, although they cannot yet determine if these fungi are active on the substrate.

Chemical-spot tests with various dyes have been proposed for detecting brown rots in some wood products (Cowling and Sachs, 1960; Eslyn, 1979). Various physical tests based on changes in the acoustic, X-ray, electrical, and strength properties of decayed wood have also been proposed (Miller, et al., 1965; Shigo and Shigo, 1974; Stoker, 1948). The theory and methodologies of decay detection are presented in detail in Chapter 16 (Detection of decay).

Types and classifications of wood decay

Decays have been grouped in a variety of often confusing and overlapping ways in the literature. The major subdivision into white and brown rots was made in 1874 by Hartig, based on the colors of the residual woods and the assumptions that the whitish material was cellulose (white rot) and the brownish material lignin (brown rot). Subsequent research established that white-rot fungi actually utilized all cell-wall constituents (cellulose, hemicelluloses, and lignin) (Campbell, 1932; Scheffer, 1936), but the early dogma persisted for many years. The grouping of decays into white-and brown-rot categories remains a major subdivision reflecting very different chemical processes in the decay of wood. As with any natural property, delineations between white and brown rot fungi are more of a continuum. As will be discussed later, our understanding of the decay processes by these two groups of fungi has dramatically changed over the past 20 years, especially in the last decade as we begin to understand how the basidiomycetes evolved.

Another important early contribution of Hartig that greatly impacted decay classification was the demonstration that a specific fungus caused a specific type of decay and that the decay was similar in other woods. This observation opened the door for decay detection in standing timber since identification of basidiocarps on the stem permitted estimates of the associated internal-cull columns. The early interest of forest pathologists and foresters was primarily the heartrots in standing timber. Decay classifications proposed by Hubert (1924) were based on decay color (white or

brown), textural features (e.g. stringy, spongy, pocket, cubical, mottled), and stem location. The early textbooks in forest pathology essentially followed these decay groupings and emphasized the macroscopic features of advanced decay, basidiocarp appearances, and associated cull (Hubert, 1931; Boyce, 1948, 1961; Baxter, 1952). The classic publication by Davidson et al. (1938) on the differentiation of wood-decay fungi into two groups by their staining reactions when grown on gallic and tannic acid media (originally determined by Bavendamm in 1928) defined the white and brown rot groups and further substantiated the basic chemical differences between them.

In 1958, Cartwright and Findlay published the first comprehensive textbook on timber decay. The decays were grouped into various timber type and wood use categories as follows:

1. *Decays of standing timber (conifers and hardwoods):*
2. *Decays of felled timber and timber in service in the open*
3. *Decays of timber in buildings and structures*
4. *Decays in timber during storage, conversion, and shipment*
5. *Decays of timber in various uses*

Their treatment again emphasized identification of the causal organisms and decay descriptions; however, this was the first comprehensive grouping of decays by major wood-processing and use categories.

Soft rots - a new decay type

In 1954, Savory described an unusual type of decay in wood caused by *Chaetomium globosum*, an ascomycete. The decay was characterized macroscopically by surface softness and microscopically by selective attack of the interior zone of the secondary wall. Savory coined the term "soft rot" to describe this new decay type, although the damage had been first described in the mid-1800s but not associated with fungal attack. Prior to this discovery, it had been assumed that only Basidiomycetes were capable of decaying wood. Soft rots were initially judged to be an interesting oddity primarily because of the unique shapes and patterns of the longitudinal bore holes.

The discovery of soft rots was soon followed by a series of studies, indicating that many Ascomycotina were capable of decaying wood (Liese, 1955; Duncan, 1960; Lundstrom, 1973; Nilsson, 1974). Soft rot

fungi were initially defined by the formation of unique longitudinal bore holes in the secondary cell wall; however, Corbett (1965) showed that some non-Basidiomycotina eroded the secondary wall in a manner similar to some white rotters and proposed that cavity formers be called Type 1 soft rotters and the cell-wall eroders the Type 2 soft rotters. Ultrastructural studies by Liese and Schmid (1962) complicated this classification by showing that small cavities were formed in the secondary walls at later decay stages by several Basidiomycete decayers. These studies illustrate the trend for organisms from different taxonomic groups to evolve similar mechanisms for accessing the energy-rich polymers in wood.

Some groupings have limited the term decay to wood destruction by Basidiomycetes and proposed soft rot as a category for wood damaged by other fungi and bacteria. This is unsatisfactory since it creates different terms for cases where wood is severely and similarly damaged by microorganisms in different taxonomic groups.

Liese (1970) reviewed woody-tissue disintegration by microorganisms and presented a classification of decay types that we will follow with some modifications in this textbook. In this separation, major reliance is placed on the order or sequence of cell-wall constituents utilized or altered. Secondary emphasis is placed on the mode of hyphal penetration of prosenchyma cells (fibers and/or tracheids) and the types of tissue damaged. The major types of woody tissue destruction by fungi and bacteria are listed in decreasing severity of cell-wall damage and examples given of typical causal organisms. The major non-decay categories of wood-inhabiting-microorganisms are also included to emphasize their distinctive features. The wood-decay group are characterized by significant weight and/or strength losses resulting from destruction of prosenchyma tissue and hyphal bore holes as large as, or larger than hyphal width. The non-decay group (wood modifiers) are characterized by tissue destruction limited primarily to parenchyma cells and hyphal bore holes absent or, when present, narrower than hyphal width.

A classification of wood-modification by microorganisms wood decayers

1. *Simultaneous-white rotters*: Attack all cell-wall constituents, essentially uniformly during all decay stages (e.g. *Trametes versicolor* or *Irpex lacteus*).
2. *Sequential-white rotters*: Attack all cell wall constituents; however, initial attack is selective for hemicelluloses and lignin (e.g. *Phellinus pini* or

Heterobasidion annosum). Other sequences of attack of cell-wall constituents may occur.

3. *Brown rotters*: Primarily attack cell-wall carbohydrates, leaving a modified lignin at the end of the decay process (e.g. *Gloeophyllum sepiarium* or *Meruliporia lacrimans*).
4. *Soft rotters*: Preferentially attack cell-wall carbohydrates in the S2 layer of the secondary cell wall, forming longitudinal cavities (Type 1) or eroding the wood cell wall from the lumen surface in hardwoods (Type 2) or the S2 in conifers (e.g. *Chaetomium globosum* or *Alternaria alternata*). Some bacteria are known to cause typical soft-rot cavities, and related tunnel-or cavitation-type cavities in cell walls.

Non-decaying wood-inhabitors

1. *Sap stainers*: Primarily attack the parenchyma cells and discolor the sapwood by the presence of pigmented hyphae. Hyphal penetration of the wood occurs primarily through pits. Weight losses caused by these fungi are minimal (e.g. *Ophiostoma piliferum* or *Aureobasidium pullulans*).
2. *Molds:* Describes the surface discolorations of wood caused by colored spores or mycelial masses (e.g. *Trichoderma* spp.). The hyaline hyphae of these fungi also grow through the parenchyma cells, but do not discolor the wood. Their primary damage is the production of pigmented spores on the wood surface that visually mar the wood appearance.
3. Scavengers: Primarily utilize the simple carbon compounds stored in the wood rays, longitudinal parenchyma, and lumen surfaces or are released during decay by other organisms. Hyphal penetration of the wood also occurs primarily through pits. No appreciable weight losses occur; however, pit penetration may increase wood permeability. A wide range of wood-inhabiting fungi and bacteria fall into this group and their roles and interactions are discussed in Chapter 5 (e.g. *Penicillium* spp., *Gliocladium virens*, or *Rhinocladiella atrovirens*). Some bacteria cause erosion channels on the lumen surface, erode pit membranes, and may cause considerable wall damage over long time periods, but these organisms are included with the scavengers since this appears to be their major, but somewhat limited role in wood decomposition.

There are some inconsistencies and neglected areas in the grouping of the four major decay types. For example, some basidiomycetes form small cavities in the cell wall that resemble Type 1 soft rot attack and the

cell-wall erosion of the Type 2 soft rotters in hardwoods resembles an early stage of brown rot. Studies of decays caused by several Xylariaceae (Ascomycetes) have shown that the characteristics of wood damage by these fungi are similar to that produced by the simultaneous-white rots, but also contains typical soft-rot cavities (Kistler and Merrill, 1968; Nilsson et al., 1989). Other groups of fungi associated primarily with slash decomposition in the forest floor such as the Tremellales, Dacrymycetales, Auriculariales, and the Gasteromycetes have been neglected as probable agents of decay and studying fungi in these groups may provide new insights on the chemical aspects of decay. In a sense, the similarities in decay patterns produced by divergent microorganisms probably reflects a convergence in attack strategies that is determined by wood chemistry and structure. As more becomes known about the chemical processes involved in decay, the decay groupings will certainly change to more closely follow the phylogenetic groupings of fungi and provide insights on the origin and development of the decay process itself.

Other common wood decay groups

Several other common decay groupings based wholly on the status of the wood in the forest or during its processing and use are largely self-explanatory, but warrant a listing and brief description as follows:

a. *stem decays in standing timber* are grouped into heartrots and sapwood decays of the living trees;
b. *slash rots* are those that develop in down timber, branches, and logging slash;
c. *storage decays* occur during the storage and processing of pulpwood, pulp chips, logs, ties, lumber, poles or piling;
d. *special commodity and use categories* are the building-rot decays, decays of utility poles, mine timbers, and piling;
e. *conifer decays are often contrasted with hardwood decays*;

Manion (1991) separated decays into heartrots in living stems and saprobic decays in wood products. As can be seen, there are numerous ways to categorize decay on the basis of wood appearance, commodity attack or the organisms involved. It is important to recognize that these are all arbitrary systems created for the convenience of those involved with assessing the damage.

Some anatomical features of wood decay

Anatomical study of wood decay is one of the oldest aspects of wood microbiology and curious observers probably were looking at decayed wood shortly after the discovery of the microscope (circa 1650). Early observers assumed the hyphal filaments were a stage of the decayed wood rather than the cause. As indicated in Chapter 1, Hartig first clearly established the causal role of fungi (hyphae) in decay in 1874 and in 1878 described and illustrated, in detail, hyphal distribution, bore holes, and cell wall erosion associated with a major building rot fungus, *Meruliporia* (*Serpula*) *lacrimans*. A large literature has accumulated on the anatomical features of decay. The older literature has been reviewed (Hubert, 1924; Wilcox, 1968, 1970). We will limit this section to a review of the significant contributions that led to a clearer understanding of microscopic features of decay, a summary of the anatomical features of the three major decay types (white, brown, and soft rots), and end with some additional research needs. The related ultrastructural aspects of decay are covered later in Chapter 10.

An early history and major contributions to the anatomy of decay

Hubert (1924) assembled a classic publication on the anatomical features and diagnosis of decay in wood, describing many decays (heartrots, slash rots, and products rots) microscopically, and suggested that the important decay fungi could be identified from anatomical features such as bore-hole types, cell-wall erosion patterns, the sequences of tissues attacked, and hyphal distribution patterns in the wood.

A popular approach to research in forest pathology between 1915 and 1940 was preparation of the biology of an important decay fungus. Some of papers that contain detailed observations on the microscopic features of several products rot fungi include: Buller (1905) on *Neolentinus* (*Lentinus*) *lepideus*; Hirt (1928–1932) on *Phellinus* (*Polyporus*) *gilvus* and *Trametes suaveolens*; Rhoads (1918) on *Trichaptum biforme* (*Polyporus pargamenus*); Spaulding (1911) on *Gloeophyllum* (*Lenzites*) *sepiarium*; and White (1920) on *Ganoderma* (*Fomes*) *applanatum*.

In 1936, Scheffer reported on the progressive effects of *Trametes* (*Polyporus*) *versicolor* decay in red gum, studying, for the first time, the anatomical, physical and chemical features of decay. He observed a uniform thinning of cell walls from the lumen toward the middle lamella and found that the majority of cell wall penetrations took place through pits. Chemical analysis of the wood at various stages of decay clearly showed uniform consumption of all cell-wall constituents by this white-rot fungus and finally dispelled the stubborn myth that white rotters decayed only lignin.

In 1937, Bailey and Vestal carefully described longitudinal bore holes in the secondary walls of several decayed woods and reported that the cavities often contained or were connected by small hyphae. The unique diamond shapes of the cavities and their periodicity were a curiosity that stimulated much research interest. Several years later, Barghoorn and Linder (1944) culturally confirmed several fungi as the cause of similar longitudinal cavities in wood submerged in the ocean.

In 1954, Savory demonstrated that several Ascomycetes caused surface decay of wood in cooling towers. The damage was characterized by the presence of unique longitudinal cavities. He coined the term soft rot for the new decay type because it was associated with a surface softening of the wood. These studies clearly showed that non-Basidiomycetes could also decay wood; decay was no longer the unique capability of some Hymenomycetes.

In 1961, Cowling completed a comparative study of the anatomical, physical, and chemical properties of sweet gum sapwood colonized by a brown rot *Rhodonia* (*Poria*) *placenta* and a white rot *Trametes* (*Polyporus*) *versicolor*. Based on anatomical and chemical changes in cell walls, hypotheses were developed concerning the relative sizes of the cellulose and lignin destroying enzymes involved in the two decay types and the sequences in which they degraded the wood. This paper stimulated extensive chemical and ultrastructural studies of decay.

Several researchers between 1959 & 1968 (Ellwood and Ecklund, 1959; Knuth and McCoy, 1962; Greaves and Levy, 1965; Boutelje and Bravery, 1968) clearly demonstrated that bacteria were able to damage pit membranes and cause some surface erosion of wood under some environmental conditions.

In 1968, Nicholas and Thomas demonstrated that filtrates and enzymes from decay fungi could induce typical decay damage in wood. This evidence opened the door to fundamental studies of the anatomy of decay and future biotechnological applications.

Wilcox (1964, 1970) improved procedures for the microscopic study of decayed wood and compared the major anatomical features of wood damaged by white, brown, and soft rot fungi and bacteria.

The principal anatomical features of decay

There are many conflicts and inconsistencies in the literature on some anatomical features of decay. Some of these differences reflect the extreme difficulty studying the anatomy of decay using light microscopy. Most fungal hyphae are hyaline and require special stains for detection against the wood background. Some hyphal features are at the resolving limit of the light microscope. It can be difficult to maintain the hyphal filaments in their precise original position when preparing sections for study and to distinguish decay from damage caused by sectioning or prior wood damage. Some microscopic features may also vary with stage of decay since the decay is a continuum. Finally, there has been a tendency to study a limited number of fungi representative of a decay type, and to expect all species in the type to have similar features.

In this section, we will limit the review to the white, brown, and soft rots. Anatomical features of the sap stains and molds are covered in Chapter 14.

Entrance and early colonization -Wood inhabiting microorganisms enter wood primarily through the torn cell walls of the wood rays and axial cells (e.g. tracheids, vessels, etc.) exposed on the surfaces of various wood products or damaged wood in the living tree. Microfauna and insects are also effective vectors for some decayers (e.g. horntail wasps vector *Amylostereum chailetti* into dying and dead balsam fir) and many sap-stain fungi. The inoculum sources are airborne spores or spores and mycelial fragments that are carried to the wood with soil, water, or wood processing machinery. Spore germination and/or mycelial growth occurs when suitable conditions for decay are present (Chapter 4). Occasional reports of fungi in the monokaryon condition suggest that single basidiospores are sometimes able to initiate decay (Zabel and Kenderes, 1980; Morrell et al., 1987; Przybylowicz et al., 1987). The entrance and early colonization phases are sustained by food reserves in the inoculum and the utilization of available simple carbon compounds in the lumina of the parenchyma cells. The hyphae of wood-inhabiting fungi in the

colonization phase appear to grow longitudinally along the lumina surface and pass from cell to cell through the pits. Growth rates longitudinally through the wood, under ideal conditions such as wood blocks in decay chambers, approach growth rates on malt agar medium. Rapid growing fungi, such as *Irpex lacteus*, can penetrate a 1 cm wood block longitudinally in several days. Whether decay begins directly after hyphal entrance or is delayed until the available simple carbon compounds are utilized is unknown and probably varies with the fungal species and its wood-inhabiting role. *Chaetomium globosum* was reported to cause extensive soft-rot in beech sapwood blocks within seven days in malt-agar decay chambers (Jutte and Zabel, 1974), while some decay fungi appear to have a long lag period before causing extensive wood degradation (Smith et al., 1992). There is also some selection of the cell types initially invaded. In hardwoods, white-rot fungi often develop initially in the vessels and wood rays, while the wood rays and longitudinal parenchyma are first colonized in conifers. These cells typically contain the majority of the readily accessible nutrients. Brown-rot fungi are less selective and hyphae are often present in most cell types. There are exceptions, such as *Rhodonia placenta* which initially attacks the wood rays. Sap-stain fungi primarily confine their invasion to the parenchyma cells but will also move though other cells as needed. Soft-rot fungi also initially appear to invade primarily wood rays and axial parenchyma. Hyphae are often more abundant in the cells damaged by white rots than brown rots or soft rots. No correlation is evident between the abundance of hyphae in cells and decay severity.

Cell wall penetration

There has long been an interest in how the fragile appearing hyphae penetrate woody cell walls. Nutman (1929) described in detail the initial cell wall penetrations of *Coriolopsis gallica* (*Polyporus hispidus*) in ash. A fine hair-like penetration peg emerged at the point of contact between the hyphal tip and the wood cell wall. This peg enlarged after wall passage. After penetration, cell-wall erosion began on each corresponding wall. An hour-glass shaped bore hole eventually formed and the initial fine penetration peg swelled to regular hyphal size. The penetration process was believed to be enzymatic because of the nature of the hour-glass like bore hole. Cartwright (1930) in studies of decay in spruce caused by *Rhodonia placenta* (*Poria monticola*), reported that hyphal tips developed minute projections that formed nicks on lumen wall contact and then rapidly

penetrated. The bore holes later enlarged uniformly to diameters several times greater than the original hyphal size, suggesting that the penetration process was primarily enzymatic due to the bore hole enlargement.

In a major study of cell-wall penetration and hyphal tip contact zones, Procter (1941) used six decay fungi, four wood species, and both ultraviolet microscopy and polarized light techniques. He concluded that enzymes were secreted primarily at the hyphal tip zone and that dissolution of the cell wall by enzymes preceded any physical hyphal contact. A transpressorium (a small stalk with a pointed tip) has been observed on the hyphal tips of some stain fungi, suggesting primarily a mechanical mode of cell wall penetration (Liese, 1970). This approach is similar to that found with many plant pathogens as they penetrate into leaves.

The question of the cell wall penetration method, however, still lingers. It is well known that some stain fungi are able to penetrate thin silver or aluminum foils. This seems to indicate that mechanical pressure also may play a role in those stain and decay fungi that form appresoria at hyphal tips or form penetration pegs. There may be a close analogy between the penetration made through the thick cuticle and epidermal cell walls of plants and that made by wood-inhabiting fungi through woody cell walls. In the case of plant pathogens, fine hypha or penetration pegs develop from the contact surface of an appresorium and penetrate the cell wall by mechanical force and enzymatic softening of the cell wall. The penetration peg resumes the normal hyphal size after the wall passage. The cell wall passages of many soft rot and sapstain fungi appear to be similar. The penetration modes of wood-inhabiting fungi are probably primarily enzymatic for the decay fungi since they often form bore holes similar to hyphal diameters and primarily mechanical for the sap stainers, soft rotters (Type 2), and others that form narrow penetration pegs (Fig. 7.6A). The type of bore hole formed is a useful way to separate colonization by stains, molds and decay fungi. Enlargement of bore holes during or after penetration to normal hyphal diameter appears to be a special trait of the decay fungi. This type of bore hole appears to develop primarily enzymatically. Bore holes formed by fine penetration pegs characterize the other wood-inhabiting fungi. These holes do not enlarge after penetration and their development appears to be primarily mechanical. Bore holes through the pits are also enlarged enzymatically and it is often difficult to distinguish degraded pits from the larger bore holes. Enlarged bore holes through pits are commonly associated with some of the white rots.

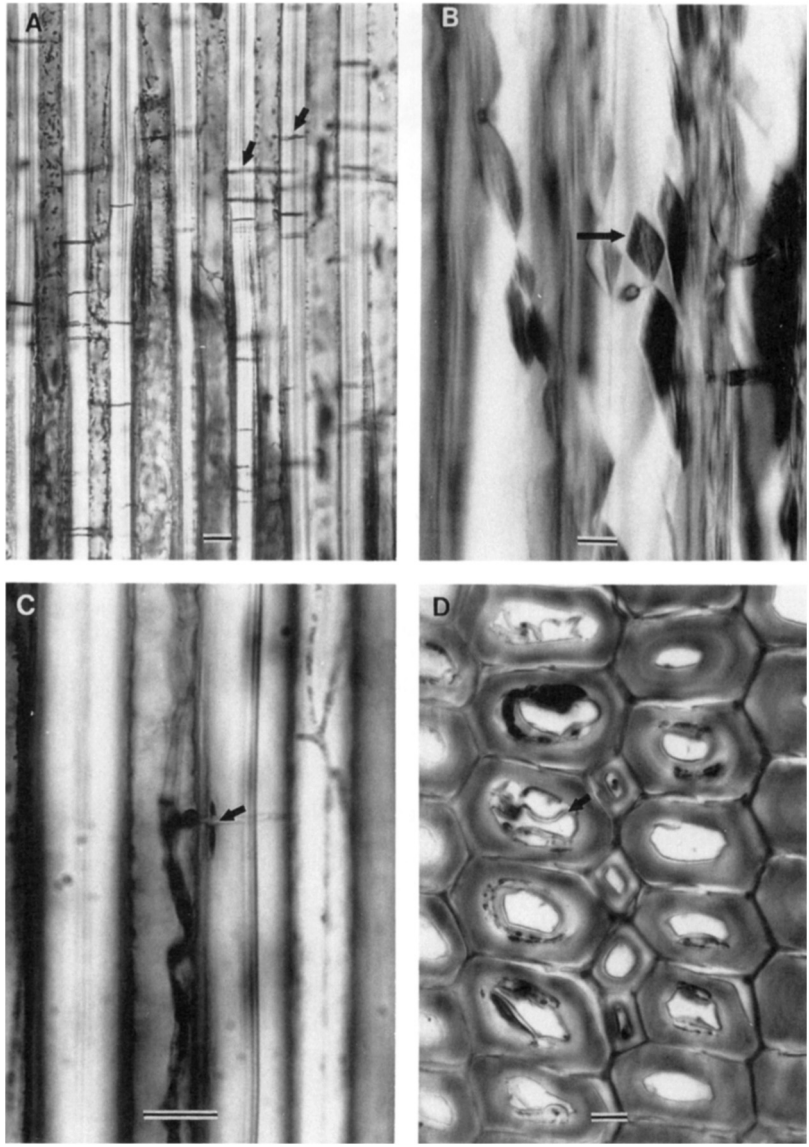


Figure 7.6 Several modes of hyphal cell wall penetration showing (A) formation of penetration pegs by *Alternaria alternata* in successive wood cell walls, (B) diamond shaped cavities formed by *Phialophora* sp., (C) formation of a T-cell and initiation of longitudinal bore hole, and (D) Type 2 soft rot caused by *A. alternata*, indicating separation of the S3 cell wall layer (Bar = 10 μm).

Soft-rot fungi (Type 1) initially form penetration pegs into the cell wall. When the tip of the peg reaches the S2 layer of the cell wall, it may branch or turn at a right angle and continue to penetrate the S2 longitudinally. Hyphal tips that divide in the S2 form structures termed "T-cells" and initiate two penetrating hyphae that then grow longitudinally through the S-2 cell wall layer (Fig. 7.6C). Periodically, enzymes are secreted from the tips of these hyphae as they grow through the S-2 cell wall layer and their action on the wood produces the typical longitudinal soft-rot cavities described elsewhere (Fig. 7.6B).

It should be stressed that the formation of longitudinal bore holes also occurs in some Basidiomycetes. Minute pockets and cavities with conical ends have been reported in the secondary wall for several white rots (Liese and Schmid, 1962), but these cavities are much smaller and more irregular than typical soft-rot cavities. Duncan (1960) reported that *Rigidoporus crocatus* (*Poria nigrescens*) formed longitudinal cavities in the secondary wall. White-rot fungi produce a gradual thinning of the cell walls from the lumina toward the middle lamella as decay advances. This attack pattern occurs most commonly in hardwoods and has been related to complete utilization of all cell-wall constituents as the decay develops. Thinning implies that the principal decay action occurs on a surface. Cell detachment and shape changes occur in pockets in the early decay stages in some sequential white rots (e.g. *Phellinus* (*Fomes*) *pini*) (Fig. 7.7). In contrast, brown rots show no change in wood cell wall thickness until the late decay stages when the residual lignin develops shrinkage cracks (initially in the vicinity of pits) and collapses. This implies that the carbohydrate portions of the wall are quickly removed, while the lignin initially maintains the cell shape and dimension. The decay damage in white rots also appears to be rather uniform in adjacent cells when viewed microscopically in cross section. In contrast, decay damage is often erratic and localized in both brown and soft rots. Soft rots in conifers appear to preferentially attack the S2 of the secondary wall. Type 1 soft-rot fungi form the longitudinal bore holes described above, while Type 2 soft rotters selectively attack the secondary cell wall in a manner very similar to brown rots. The S3 may become detached in Type 2 attack in conifers and appear as a small ring in the decayed cell wall material (Fig. 7.6). Soft rots in hardwoods are generally more severe and Type 1 longitudinal bore holes predominate in the S2, particularly in fibers. Both the S2 and S3 are eroded by Type 2 soft-rot fungi and the damage resembles white rot attack (Fig. 7.8).

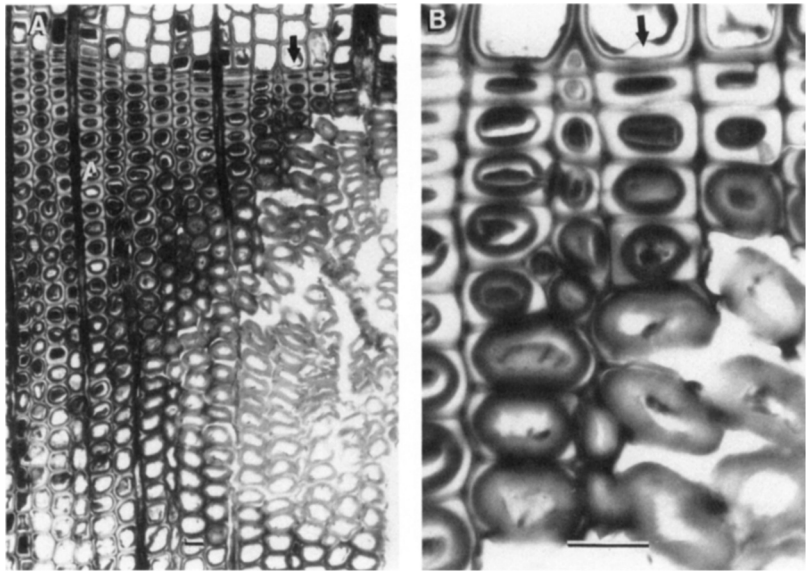


Figure 7.7 Sequential stages of decay development in a white pocket rot of red spruce caused by *Phellinus pini*. Vertical arrows identify the same radial row of tracheids. (A) Cross section through a decay pocket, (B) successive delignification of tracheid walls from the lumen towards the middle lamella and then detachment and rounding of cells (Bar = 20 μm). *Courtesy Dr. Susan Anagnost, SUNY College of Environmental Science & Forestry.*

The cell-wall layers exhibit striking differences in decay susceptibility. Cowling (1965) proposed that the selective action of some fungi against various cell-wall layers could be useful for their selection and structural study. The S2 is more susceptible to degradation; likely because this cell wall layer contains the majority of cellulose. In conifers, the compound-middle lamella and S3 are more resistant to both brown and soft rots. In contrast, the compound-middle lamella zone in conifers is quickly decayed by the sequential white rotters, often resulting in cell detachment early in the decay process. In hardwoods, the compound-middle lamella zone is also resistant to brown rotters.

Some research needs

While scientists have studied wood decay for over a century, much of the process remains poorly understood.

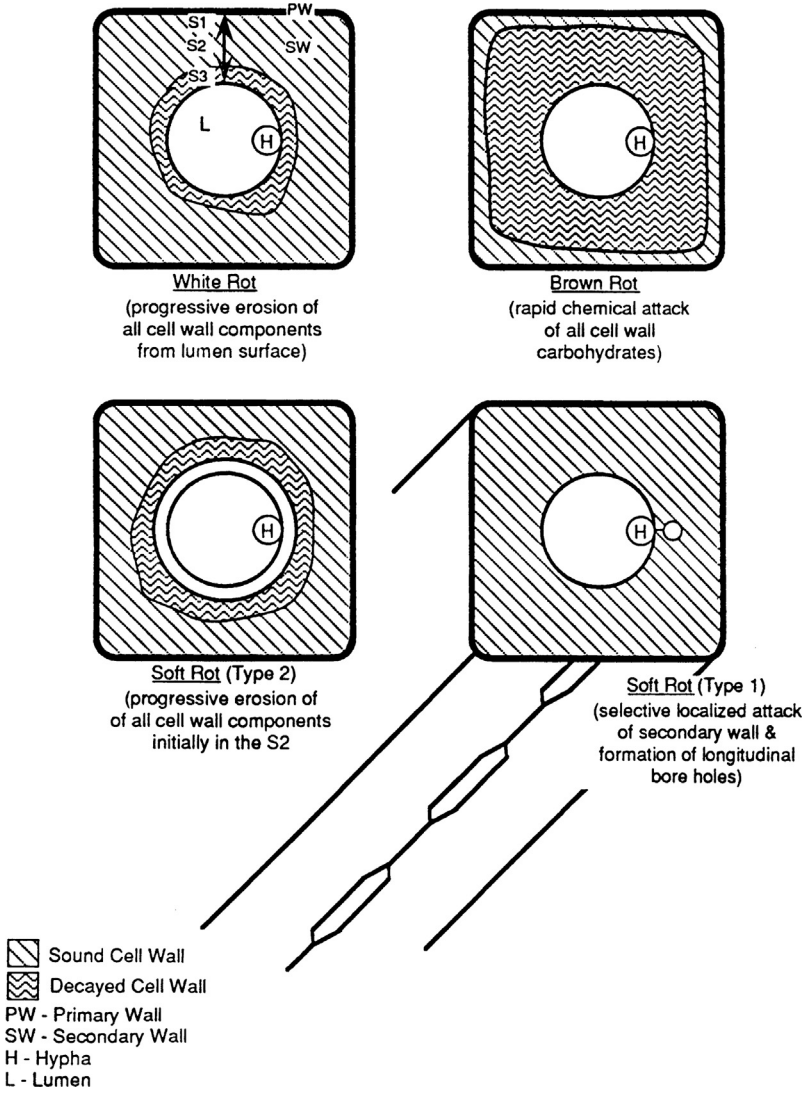


Figure 7.8 Diagram showing the various modes of cell-wall destruction for white-rots, brown rots, and the two types of soft rots.

A major research need is the development of a more rational system of decay classification that harmonizes chemical changes in cell wall constituents with taxonomy.

The current decay classification system is based on chemical studies of a rather limited number of fungi and neglects several important taxonomic

groups. This system fails to clearly separate some Type 2 soft rots from brown rots and some white rots (Xylariaceae) from Type 1 soft rots. There is a need to study the major representatives of all major decay groups and search for a more definitive classification system that better reflects current taxonomic classification.

Some additional research needs can be posed through a series of questions as follows:

1. What are the fundamental differences in the nature of the enzymes/non-enyzmatic factors produced by white rots and brown rots that limit the former to surface action while the latter rapidly and deeply penetrate the wood-cell wall?
2. What limits the effective distance an enzyme can diffuse from a hyphae and still be effective?
3. How do brown rot and the other wood-inhabiting fungi penetrate cell walls in the absence of lignin destroying enzymes?
4. How and where are enzymes released by the hyphae?
5. How can the conical ends and puzzling periodicity of successive longitudinally aligned cavities be explained for the soft rotters?
6. Is there an evolutionary sequence hidden in the many types and degrees of cell-wall disintegrations caused by microorganisms?

A final research need is to develop methodologies to simplify and increase the accuracy of early decay detection.

Summary

1. Decay is defined as those significant changes in the physical and chemical properties of wood that are caused by the chemical (enzymatic) activities of microorganisms.
2. The term decay is used both as a description of the condition of wood and the process of its external digestion by microorganisms.
3. The decay process has stages that are arbitrarily designated as incipient (hidden), early, intermediate, and late or advanced.
4. The major disadvantages of decay are losses in strength and biomass, but changes in color and texture can also occur.
5. Decay losses are large in the aggregate and estimated at 15–25% of the gross volume of standing timber, 15% of the storage volume annually, and 10% of the annual cut as replacements for decay of wood in service, plus the replacement costs.

6. Decay can be recognized macroscopically by color and textural changes and microscopically by characteristic hyphal features, bore holes, and cell wall erosion.
7. Common decay groups based on the wood location and use are stem decays, heartrots, slash rots, storage decays, and products rots, including special uses such as building rots. Wood-inhabiting fungi can be grouped into decay and non-decay categories.
8. Decay groups, based on the cell-wall constituents utilized and the type of causal agents, are the white rots (attack of all cell components by Basidiomycetes), brown rots (attack primarily of carbohydrates by Basidiomycetes), and soft rots (attack primarily of carbohydrates by Ascomycetes).
9. The major categories of the non-decay wood-inhabiting fungi are sapstains, molds, scavengers, and bacteria.
10. During early colonization, most wood-inhabiting fungi pass from cell to cell through the pits. Decay fungi are characterized by producing large bore holes directly through cell walls primarily enzymatically in the later decay stages. Bore holes are a useful microscopic feature of decay.
11. Non-decay fungi may form narrow threadlike hyphal filaments called "penetration" pegs that pass through cells mechanically and enzymatically.
12. White rots utilize all cell wall constituents and are characterized by a uniform thinning of the cell walls from the lumen toward the middle lamella as decay develops.
13. Brown rots display no change in wall thickness until the late decay stages when the residual lignin shrinks and collapses.
14. Soft rots are subdivided into Type 1 soft rots characterized by production of longitudinal bore holes in the S2 of the secondary wall and Type 2 soft rots that erode the cell wall in a manner similar to brown rots in hardwoods, but are more confined to the S2 in conifers.

References

Bailey, I.W., Vestal, M.R., 1937. The significance of certain wood-decaying fungi in the study of the enzymatic hydrolysis of cellulose. J. Arnold Arbor. 18, 196–205.

Barghoorn, L.S., Linder, D.H., 1944. Marine fungi: their taxonomy and biology. Farlowia 1, 395–467.

Bavendamm, W., 1928. Uber das Vorkommen und den Nachweis von Oxydasen bei holz zerstorenden Pilzen. Z. Pflanzenkrankeheiten u. Pflanzenschutz 38, 257–276.

Baxter, D.V., 1952. Pathology in Forest Practice, second ed. John Wiley &Sons, New York, p. 601.

Boutelje, J.B., Bravery, A.F., 1968. Observations on the bacterial attack of piles supporting a Stockholm building. J. Inst. Wood Sci. 20, 47–57.
Boyce, J.S., 1961. Forest Pathology. McGraw-Hill Book Co., Inc., New York, p. 572.
Buller, A.H.R., 1905. The destruction of wood paving blocks by the fungus, *Lentinus lepideus* Fr. Jour. Econ. Biol. 1, 2–13.
Campbell, W.G., 1932. The chemistry of the white rots of wood. III. the effect of wood substances of *Gandoderma applanatum* (Pers.) Pat., *Fomes fomentarius* (Linn.) Fr., *Polyporus adustus* (Willd.) Fr., *Pleurotus ostreatus* (Jacq.) Fr., *Armillaria mellea* (Vahl.) Fr., *Trametes pini* (Brot.) Fr., and *Polystictus abietinus* (Dicks.) Fr. Biochem. J. 26, 1829–1838.
Campbell, A.H., 1933. Zone lines in plant tissues: I. The black lines formed by *Xylaria polymorpha* (Pers.) Grev. in hardwoods. Ann. Appl. Biol. 20 (123), 145.
Cartwright, K.St.G., 1930. A decay of Sitka spruce timber, caused by *Trametes serialis*, Fr. A cultural study of the fungus. Dept. Sci. and. Ind. Res. For. Prod. Res. Bull. 4, 332 (London).
Cartwright, J.St.G., Findlay, W.P.K., 1958. Decay of Timber and Its Prevention. Her Majesty's Stationery Office, London, p. 332.
Corbett, N.H., 1965. Micro-morphological studies on the degradation of lignified cell walls by Ascomycetes and fungi imperfecti. Jour. Inst. Wood Sci. 14, 18–29.
Cowling, E.B., 1961. Comparative biochemistry of the decay of sweetgum sapwood by white-rot and brown-rot fungi. U.S. Dept. Agr. Tech. Bull. 1258, 79.
Cowling, E.B., 1965. Microorganisms and microbial enzyme systems as selective tools in wood anatomy. In: Cote, W. (Ed.), Cellular Ultrastructure of Woody Plants. Syracuse University Press.
Cowling, E.B., Sachs, I.B., 1960. Detection of brown rot. For. Prod. J. 10 (11), 594–596.
Davidson, R.W., Campbell, W.A., Blaisdell, D.J., 1938. Differentiation of wood decaying fungi by their reaction on gallic or tannic acid medium. J. Agric. Res. 57, 683–695.
Duncan, C.G., 1960. Wood-attacking capacities and physiology of soft-rot fungi. For. Prod. Lab. Rep. 2173, 28.
Ellwood, E.L., Ecklund, B.A., 1959. Bacterial attack of pine logs in pond. For. Prod. J. 9, 283–292.
Eslyn, W.E., 1979. Utility pole decay. Part 3: Detection in pine by color indicators. Wood Sci. Technol. 13, 117–126.
Graham, R.D., Corden, M.E., 1980. Controlling Biological Deterioration of Wood With Volatile Chemicals. Electric Power Research Institute Final Report EPRI El-1480, Palo Alto.
Greaves, H., Levy, J.F., 1965. Comparative degradation of the sapwood of Scot's pine, beech, and birch by *Lenzites trabea*, *Polystictus versicolor*, *Chaetomium globosum* and *Bacillus polymyxa*. J. Inst. Wood Sci. 15, 55–63.
Hartig, R., 1874. Wichtige Krankheiten der Waldbaume. Springer-Ver1ag, Berlin.
Hartig, R., 1878. Die Zerzetzungersche" inungen des Holzes der Nadelholz baume und der Eiche, Berlin.
Hirt, R.R., 1928. The biology of *Polyporus gilvus* (Schw.) Fries N.Y. State Coll. For. Syracuse. Tech. Pub. 22, 27.
Hirt, R.R., 1932. On the biology of *Trametes suaveolens* (L) Fries. N.Y. State Coll. For. Syracuse. Tech. Pub. 37, 36.
Hopp, H., 1938. The formation of colored zones by wood-destroying fungi in culture. Phytopathology 28, 601–620.
Hubert, E.E., 1924. The diagnosis of decay in wood. J. Agr. Res. 29, 523–567.
Hubert, E.E., 1931. An Outline of Forest Pathology. John Wiley and Sons, New York, p. 543.
Jutte, S.M., Zabel, R.A., 1974. Initial wood decay stages as revealed by scanning electron microscopy. Proc. Scanning Electron. Microscopy (Part II) 445–452.

Kistler, B.R., Merrill, W., 1968. Effects of *Strumella coryneoidea* on oak sapwood. Phytopathology 58, 1429–1430.
Knuth, D.T., McCoy, E., 1962. Bacterial deterioration of pine logs in pond storage. For. Prod. J. 12, 437–442.
Liese, W.K.F., 1955. On the Decomposition of the Cell Wall by Microorganisms. Rec. Ann. Conv., Brit. Wood Pres. Assoc.
Liese, W., 1970. Ultrastructural aspects of woody tissue disintegration. Ann. Rev. Phytopathol. 8 (231), 258.
Liese, W., Schmid, R., 1962. Elektronmikroskopische Untersuchungen uber den Abbau des Holzes durch Pilze. Angew. Bot. 36, 291–298.
Lundstrom, H., 1973. Studies on the wood-decaying capacity of the soft rot fungi *Allescheria terrestris*, *Phialophora* (*Margarinomyces*) *luteo-viridis*, and *Phialophora richardsiae*. Research Note R 87. Dept. of Forest Prod., Royal College of Forestry, Stockholm.
Manion, P., 1991. Tree Disease Concepts, second ed. Prentice-Hall, New York, NY.
Miller, B.D., Taylor, F.L., Popek, R.A., 1965. A sonic method for detecting decay in wood poles. Proc. Amer. Wood Pres. Assoc. 61, 109–113.
Morrell, J.J., Corden, M.E., Graham, R.D., Kropp, B.R., Przybylowicz, P., Sexton, C.M., et al., 1987. Basidiomycete colonization of Douglas-fir poles during our seasoning and its effect on wood strength. Proc. Am. Wood Preserv. Assoc. 83, 284–296.
Nicholas, D.O., Thomas, R.J., 1968. The influence of enzymes on the structure and permeability of loblolly pine. Proc. Am. Wood Preserv. Assoc. 64, 70–76.
Nilsson, T., 1974. Studies on Wood Degradation and Cellulolytic Activity of Microfungi. STUD. For. Suec. 104. Royal College of Forestry, Stockholm.
Nilsson, T., Daniel, G., Kirk, T.K., Obst, J.R., 1989. Chemistry and microscopy of wood decayed by some higher ascomycetes. Holzforschung 43, 11–18.
Nutman, F.J., 1929. Studies of wood-destroying fungi. 1. *Polyporus hispidus* (Fries). Ann. Appl. Biol. 16, 40–64.
Procter Jr., P., 1941. Penetration of the Walls of Wood Cells by the Hyphae of Wood Destroying Fungi. Yale University School of Forestry Bull. No. 47.
Przybylowicz, P.R., Kropp, B.R., Corden, M.E., Graham, R.D., 1987. Colonization of Douglas-fir poles by decay fungi during air-seasoning. For. Prod. J. 37 (4), 17–23.
Rhoads, A.S., 1917. The black zones formed by wood-destroying fungi. N.Y. State Coll. For. Tech. Pub. 8, 49.
Rhoads, A.S., 1918. The biology of *Polyporus pargamenus* Fries. N.Y. State Coll. For. Tech. Pub. 11, 197.
Savory, J.G., 1954. Breakdown of timber by ascomycetes and fungi imperfection. Ann. Appl. Biol. 41, 336–347.
Scheffer, T.C., 1936. Progressive Effects of *Polyporus versicolor* on the Physical and Chemical Properties of Red Gum Sapwood. USDA Tech. Bull. No. 527.
Shigo, A.L., Shigo, A., 1974. Detection of Discoloration and Decay in Living Trees and Utility Poles. USDA, For. Ser. Res. Paper, NE-294, p. 11.
Smith, S.M., Morrell, J.J., Sexton, C.M., 1992. Effect of fungal colony size on residual strength of Douglas-fir sapwood and heartwood. For. Prod. J. 42 (9), 19–24.
Spaulding, P., 1911. The timber rot caused by *Lenzites sepiaria*. U.S. Dept. Agr. Bur. Plant. Indus. Bul. 214, 46.
Stoker, R.S., 1948. X-ray pole inspection. Proc. Amer. Wood Pres. Assoc. 44, 298–313.
Wang, C.J.K., Zabel, R.A., 1990. Fungi Inhabiting Utility Poles in the Eastern United States. American Type Culture Collection, Rockville, MD.
Wilcox, W.W., 1964. Preparation of Decayed Wood for Microscopical Examination. U.S.D.A. For. Ser., For. Prod. Lab. Res. Note FPL-056, p. 22.
Wilcox, W.W., 1970. Anatomical changes in wood cell walls attacked by fungi and bacteria. Bot Rev. 36 (1), 1–28.

White, J.H., 1920. On the biology of *Fomes applanatus* (Pers.). Wallr. Trans. R. Canad. Inst. 12, 133–174.
Zabel, R.A., Lombard, F.F., Kenderes, A.M., 1980. Fungi associated with decay in treated Douglas-fir transmission poles in northeastern United States. Forest Prod. J. 30 (4), 51–56.

Further reading

Bayliss, J.S., 1908. The biology of *Polystictus versicolor* fries. J. Econ. Biol. 3, 124.
Greaves, H., 1969. Micromorphology of the bacterial attack of wood. Wood Sci. Technol. 3 (150), 166.
Hubert, L.E., 1922. A staining method for hyphae of wood-inhabiting fungi. Phytopathology 12, 440–441.
Humphrey, C.J., 1923. The destruction of the fungus *Poria incrassata* of coniferous timber in storage and when used in the construction of buildings. South. Lumber J. 49 (3), 36–37, 49–53, 55.
Levi, M.P., 1964. The fungal degradation of wood. J. Lnst. Wood Sci. 12, 56–66.
Levy, J.F., 1965. The soft rot fungi: their mode of action and significance in the degradation of wood. In: Preston, R.D. (Ed.), Advances in Botanical Research, vol. 2. Academic Press, New York, pp. 323–357.
Liese, W., 1967. History of wood pathology. Wood Sci. Technol. 1, 169–173.
Liese, W., Smid, R., 1964. Uber das Wachstum von Blauepilzen durch verholtze Zellwande. Phytopathol. Z. 51, 385–393.
Lutz, J.G., Duncan, C.G., Scheffer, T.C., 1966. Some effects of bacterial action on rotary-cut southern pine veneer. For. Prod. J. 16 (8), 23–28.
Merrill, W., 1970. Spore germination and host penetration by heart-rotting hymenomycetes. Ann. Rev. Phytopathol. 8, 281–300.
Merrill, W., French, D.W., 1965. Wood fiberboard studies. 3. Effect of common molds on the cell wall structure of the wood fibers. Tappi 48, 653–654.
Merrill, W., Lambert, D.H., Liese, W., 1975. Important Diseases of Forest Trees and Phytopathology for Botanists and Foresters by Robert Hartig (Translation from 1874 Textbook. Phytopathology Classics No. 12). American Phytopathological Society, St. Paul, Minnesota.

CHAPTER EIGHT

Chemical changes in wood caused by decay fungi

Contents

The chemistry of wood decay is a subject of growing interest and importance. It is of interest because of the natural challenge of any complex problem. It is of great economic importance since increased knowledge of the successive chemical steps involved in the decay process may permit wood to be protected more effectively for high decay hazard uses. It may also lead to more economical methods of biopulping, pulp bleaching and the separation of carbohydrates from the wood complex for chemical purposes or new uses of "enzymatically modified wood" as cheap animal food or energy sources.

Great progress has been made toward understanding the chemistry of wood decay. These results are summarized in textbook chapters, textbooks, and symposia (Kirk, 1971, 1973; Loewus and Runeckles, 1977; Kirk et al., 1980; Crawford, 1981; Sjostrom, 1984; Fengel and Wegener, 1984; Higuchi, 1985; Eriksson et al., 1990). Key early contributions that either set the stage for a better understanding of the decay process or added

Wood Microbiology.
DOI: https://doi.org/10.1016/B978-0-12-819465-2.00008-5

significant new insights include the following: Reese (1963, 1977) characterized the enzymes involved in cellulose degradation; Cowling and Brown (1969) examined the dimensional relationships between the molecular architecture of the cell wall and the puzzle of its accessibility to enzyme molecules; Timell (1967) elucidated the complex structures of the hemicelluloses; Côté (1977) clarified the ultrastructure of wood; Adler (1977) studied lignin structure; Tien and Kirk (1983) identified the first enzymes involved in the enzymatic attack of lignin; Eriksson (1978) identified the "cellulase" enzymes associated with some white-rot fungi; Higuchi (1985) studied lignin biodegradation; and Ruel and Barnoud (1985) and Eriksson et al. (1990) described the micromorphological sequences of many decays suggesting that some non-enzymatic events occurred in white rot.

The major wood cell wall constituents involved in the decay process are cellulose, the hemicelluloses and lignin. Decay occurs primarily from the enzymatic activities of a few groups of specialized fungi.

Previous chapters have reviewed the general chemical nature and structure of the three major cell components, considered their interrelationships at various levels of organization, and the spatial dimensions of wood at the microscopic and ultrastructural levels. They have also reviewed enzyme functions and related them to the major metabolic events taking place in and around hyphae. The sequential chemical and enzymatic events involved in the decay process are, of course, the basic cause of the grosser anatomical and physical property effects discussed in Chapters 7 and 10.

A useful approach to this complex topic will be to look first at the effects of decay on the major wood components for the three major decay types (white, brown, and soft rots), then to consider the sites and sequences of action of the enzymes involved, and to end with a summary of current understandings of the decay process and its effects on various important wood properties.

Changes in cell wall components by decay type

The cell wall components of wood are utilized in varying orders and rates by different decay fungi. These differences are the basis for the chemical classification of decay presented in Chapter 7. The general depletion patterns of the cell wall components at progressive stages of decay provides

useful insights on the types and sequences of the enzymes involved. The depletion pattern can be monitored by comparing the weight of the cell-wall constituents in original wood to their weights in the residual decayed wood. Comparative weight-loss curves are developed by examining weight losses at various times. For example, lignin content steadily decreases as decay develops in a white rot, while essentially no lignin loss occurs with brown rots (Table 8.1). Another useful method for monitoring cell wall depletion is to express the level of each component at various decay stages on the basis of the original amount present in sound wood. This method makes it easy to compare the percent change in the cell wall constituents with the total weight loss percent to detect differences rates of component utilization (Tables 8.2–8.4). Precise gravimetric determination of the major cell-wall constituents is both time consuming and difficult, but more rapid techniques involving chromatographic quantification of the major sugars have been developed. For example, the analysis of glucan, xylan, and mannan in decayed coniferous wood provides estimates of cellulose, galactoglucomannan, and arabino-4-0-methylglucuronoxylan content (Tables 8.2 and 8.3). Hydrolysis of these carbohydrates and their groupings into glucan, xylan, and mannan does not segregate the arabinose or galactose and may lead to over-estimates of certain fractions, but minor analytical errors

Table 8.1 Chemical composition of sweetgum sapwood in progressive stages of decay caused by white and brown rot fungi. The weight loss percentages are based on the moisture-free weight of the original sound wood[a].

Average weight loss %	Composition of residual material (%) Total carbohydrate	Glucan	Galactan	Mannan	Xylan	Araban	Lignin
Trametes (*Polyporus*) *versicolor* (white rot)							
0	77.1	52.3	1.1	2.7	20.1	0.9	22.9
25.3	58.0	34.5	0.6	2.6	14.9	0.4	16.7
55.2	33.9	22.8	0.3	1.7	8.9	0.3	10.9
79.0	14.9	9.7	0.2	0.9	3.9	0.2	6.1
96.7	2.5	2.0	0.03	0.1	0.3	0.03	0.8
Rhodonia placenta (*Poria monticola*) (brown rot)							
0	77.1	52.3	1.1	2.7	20.1	0.9	22.9
20.1	56.8	40.1	0.5	1.3	14.5	0.4	23.1
44.8	32.6	22.2	0.2	0.7	0.2	0.2	22.6
69.5	9.2	5.4	0.1	0.2	3.4	0.1	21.3

[a]Data selected from Cowling (1961).

Table 8.2 Weight losses of the major cell wall components in conifers and hardwoods caused by several white-rot fungi.

Substrate	Fungus	Weight loss (%)[a]				
		Total	Glucose	Mannan	Xylan	Lignin
Conifers[b]						
Pinus monticola (western white pine)	Trametes versicolor	13	4	13	13	27
		22	17	22	21	33
		43	43	47	47	52
		61	65	68	67	62
		81	85	89	89	86
	Ganoderma applanatum	16	12	16	19	26
		43	42	52	51	57
Picea sitchensis (*Sitka spruce*)	*Trametes versicolor*	6	10	13	3	5
		21	27	29	27	20
		40	39	50	45	52
		61	65	72	66	65
	Ganoderma applanatum	11	13	18	17	11
		35	33	47	44	48
Hardwoods[c]						
Betula alleghaniensis (*Yellow birch*)	*Trametes versicolor*	21	20	26	26	31
		36	39	54	39	39
	Ganoderma applanatum	17	28	27	14	18
		32	38	50	36	37
	Bondarzewia berkeleyi	8	16	8	4	31
		22	31	33	30	42
		39	44	51	40	63
	Oschnoderma resinosum	11	17	25	10	35
		22	30	27	21	44

[a]Weight loss percentages based upon the original amount of each component in sound wood.
[b]Data selected from Kirk and Highley (1973).
[c]Data selected from Kirk and Moore (1972). Values rounded to nearest percent.

introduced are judged to be insignificant in relation to the overall mass changes (Kirk and Highley, 1973). Lignin determinations also vary substantially with method of preparation and these variations are discussed further in the lignin section. The more recent development of indirect spectroscopic measures such as Fourier Transform Infrared Spectroscopy (FTIR) allow researchers to probe changes in linkages between the three primary wood polymers as wood is degraded by either abiotic or biotic agents.

It is prudent to remember that the decayed sample itself contains all the different cell and tissue types that have varying levels of each wood component, that microoganisms may selectively attack specific cell types,

Table 8.3 Weight losses of the major cell wall components in conifers and hardwoods caused by several brown-rot fungi[a].

Wood species	Fungus	Weight loss (%)				
		Total	Glucan	Mannan	Xylan	Lignin
Conifers[b]						
Tsuga heterophylla (western hemlock)	*Rhodonia placenta*	6	4	11	16	8
		26	26	38	39	4
		46	79	88	75	2
Picea engelmanii (Engelmann spruce)	*Rhodonia placenta*	12	13	26	22	6
		26	29	41	69	10
		49	81	93	79	− 13
	Gloeophyllum trabeum	10	12	14	18	9
		19	22	47	37	4
		43	55	80	65	11
P. sitchensis (Sitka spruce)	*Neolentinus lepideus*	6	12	6	19	3
		27	37	68	49	− 4
		45	57	78	64	6
Pinus taeda (loblolly pine)	*Rhodonia placenta*	9	13	25	1	− 4
		24	29	58	26	2
		45	65	81	69	4
Hardwoods[c]						
Liquidambar styraciflua (Sweetgum)	*Rhodonia placenta*	20	23	52	30	0
		45	58	74	55	0
		70	90	93	85	− 9

[a]Weight loss percentages based upon the original amount of each component in sound wood.
[b]Data selected from Kirk and Highley (1973).
[c]Data selected from Cowling (1961). Values rounded to nearest percent.

that substantial decay gradients can occur even in small samples of decayed wood, and that the fungus may be simultaneously assembling storage sugars or polyphenols at later stages of decay that may be inadvertently detected as cell wall components.

The general depletion patterns of cell wall components for white, brown, and soft-rot fungi in conifers and hardwoods are presented in Tables 8.1–8.4. General information on each decay type is also briefly summarized.

White rot fungi are able to attack and metabolize all major wood constituents. In this regard, the white-rot fungi are unique among most microorganisms in their capacity to completely depolymerize and metabolize lignin. Numerous studies now indicate, however, that the major cell wall components are used to varying orders and rates by different white

Table 8.4 Weight losses of the major cell wall components of conifers and hardwoods caused by several soft-rot fungi[a].

Wood species	Fungus	Weight loss (%)[b]				
		Total	Glucan	Mannan	Xylan	Lignin
Conifers						
Pinus monticola (western white pine)	*Papulospora* sp.	15	18	13	18	12
	Paecilomyces sp.	10	7	6	8	14
	Thielavia terristrius	7	3	− 3	9	14
Hardwoods						
Alnus rubra (red alder)	*Papulospora* sp	10	15	14	6	9
		17	23	18	8	12
	Paecilomyces sp.	15	21	28	25	11
		25	37	20	23	13
		41	60	22	44	19
	Thielavia terrestrius	7	10	19	1	9
		28	40	21	29	17
Populus balsamifera (balsam poplar)	Papulospora sp.	10	14	14	17	0
		21	27	25	29	4
	Paecilomyces sp.	14	15	28	35	10
		28	41	30	37	11
	Thielavia terrestrius	10	11	29	12	6
		23	25	43	27	13

[a]Data selected from Eslyn et al. (1975).
[b]Weight loss percentages based on original amount of each compound in sound wood.

rot fungi, suggesting that fungi represent a heterogenous group with widely varying enzymatic capabilities. These differences were used by Liese (1970) to group white-rot fungi into the simultaneous rotters that utilized the components uniformly and white rotters that initially utilized lignin more rapidly than cellulose. The differences illustrate the largely artificial separation of fungi into white and brown rotters.

Early, differential lignin attack by some white rotters has important potential commercial applications and many attempts have been made to find isolates or develop new fungal strains for possible use in biopulping, biobleaching of pulps, bioremediation of hazardous chemicals, or increasing the biodegradability of wood for use in industrial fermentation or ruminant feed. *Trametes* (*Polyporus*) *versicolor* is an example of a fungus that uses all wood components essentially uniformly. *Phellinus* (*Fornes*) *pini*, *Heterobasidion* (*Fornes*) *annosum*, and *Bondarzewia* (*Polyporus*) *berkeley*i are

examples of fungi that remove the lignin at faster rates than cellulose or the hemicelluloses in early decay stages. *Ganoderma applanatum* is an interesting example of the few white-rot fungi that initially remove cell wall carbohydrates somewhat more rapidly than lignin. Some general features of utilization of wood constituents by white rot fungi are summarized as follows:

(a) All cell-wall components are presumably ultimately consumed with the exception of the minor minerals. There is considerable variation in the sequence and the rate at which components are utilized both by species and fungal strains within a species. In most cases, the hemicelluloses are utilized preferentially in early decay stages, but weight losses may approach 95–97% of the original wood material with prolonged exposures under optimum decay conditions.

(b) Residual wood at all stages of decay has low solubility in 1% sodium hydroxide (alkali solubility) suggesting that the breakdown products of decay are utilized by the fungi as rapidly as they are released.

(c) The cellulose, hemicelluloses and lignin remaining in the undecayed portions of the wood appear to be essentially unaltered, suggesting that white rot fungi concentrate their attack on exposed cell wall surfaces. Thus, the enzymes slowly erode their way into the cell walls from lumina surfaces.

Brown rot fungi primarily decompose the cell-wall carbohydrates, leaving behind a modified, demethoxylated lignin residuum (Tables 8.1, 8.3). For decades, brown rot fungi were largely considered to be incapable of substantial lignin degradation; however, recent developments indicate that these fungi actually actively depolymerize lignin through non-enzymatic activities at the early stages of attack. These activities expose hemicellulose and cellulose to enzymatic attack, but the lignin repolymerizes, leaving a modified lignin residual.

Selective removal of carbohydrates in the later stages of brown rot attack has been used to study the distribution of lignin in the cell wall (Côté et al., 1966). The hemicelluloses are removed more rapidly than cellulose by brown rot fungi in the early decay stages and these changes are believed to be the primary cause for the rapid losses in strength associated with early brown rot decay. Highley (1977) showed that carbohydrate supplements such as mannan are necessary for depolymerization of pure cellulose by *Rhodonia* (*Poria) placenta*, a brown-rotter. Brown rots differ from white rots in the extensive depolymerization of the carbohydrates in the secondary wall early in the decay process (Kirk and Highley, 1973).

While lysis zones are closely associated with white rot hyphae, substantial cell wall damage has been noted at distances up to several cell widths for some brown rots (Blanchette, 1984).

The ways brown-rot fungi modify wood during progressive decay development can be summarized as follows:

a. All carbohydrates are ultimately consumed, leaving a residuum of modified lignin in the cell wall.

b. Large increases in water solubility and solubility in 1% NaOH occur at early decay stages due to the rapid carbohydrate depolymerization in the early decay stages and increased lignin solubility in the later decay stages. Brown-rot fungi appear to depolymerize wood in the early decay stages much more rapidly than the decay products can be metabolized. The presence of excess wood decomposition products may help explain the presence of other wood scavengers in brown-rotted wood.

c. The decay process rapidly involves the S1 and S2 layers of the cell walls, but develops irregularly and lacks the hyphal-associated lysis zones typical of white rot fungi.

d. There appears to be much less variation in the sequential attack of cell-wall constituents by brown-rot fungi as compared to the white rotters.

Soft-rot fungi display considerable variation in their effects on cell-wall constituents during decay development (Tables 8.4 and 8.5). For many species, the principal targets are the carbohydrates, while lignin attack is believed to be limited to minor demethoxylation. Some soft rotters, however, selectively remove more lignin than carbohydrate from coniferous wood, in a manner similar to some white rotters (Eslyn et al., 1975). Most of the soft-rot fungi tested remove glucans at a faster rate

Table 8.5 Chemical composition of European beech in progressive stages of decay by the soft rot fungus, *Chaetomium globosum*. The weight loss percentages are based on the dry weight of the original non-decayed wood corrected for mineral content (Savory and Pinion, 1958).

	Composition of residual material %			
Average weight loss (%)	**Cross & Bevan cellulose[a]**	**Alpha cellulose[b]**	**Pentosans**	**Lignin**
0	56.2	42.2	27.8	21.6
29.9	34.3	26.5	18.4	19.4
31.7	34.7	25.7	17.6	19.3
40.6	28.2	19.9	14.6	17.6
49.1	22.4	16.3	12.5	16.75

[a]Roughly equivalent to total carbohydrate as listed in Tables 8.1–8.3.
[b]Roughly equivalent to glucan as listed in Tables 8.1–8.3.

than the hemicelluloses, although several exceptions were noted. Type 1 soft-rot fungi can degrade crystalline cellulose as reflected by the formation of characteristic cavities in the S2 zone of the secondary wall. Wood decayed by soft-rot fungi resembles that of white-rot degraded wood in having low alkali solubility, indicating that the degradation products are used at the same rate as they are released. In conifers, the S-3 zone of the secondary wall is resistant to soft rot attack, but delignification substantially increases decay susceptibility and may shift the fungus from cavity formation (Type 1) to the wall erosion (Type 2) mode (Morrell and Zabel, 1987). The soft rot fungi appear to be variable in their effects on the cell-wall components and share some features of both white and brown rots. It is probable that this broad group, now defined primarily as non-basidiomycete decayers, includes fungi typified by several different patterns of cell wall decomposition.

The chemical mechanisms of wood decay

While it is important to understand the relative differences in wood degradation among fungi, it is equally important to consider how these drastic changes in the various cell wall components are accomplished by the three major types of decay fungi. Carbohydrate components are depolymerized primarily by a series of hydrolytic enzymes, while lignin is degraded primarily by oxidative enzymes. There is also evidence, that powerful, non-enzymatic oxidizing agents initiate the first steps in the brown rots by depolymerizing the crystalline zones of the cellulose microfibrils. As we examine the various fungal enzymes, it is important to note that the enzymes active in decay are secreted and act outside of the hyphae (exo-enzymes). These enzymes must physically bind or form a complex with the portion of the polymer or macromolecule they attack. Since the large macromolecules comprising the cell wall are non-soluble, the enzymes involved in decay must either diffuse into the cell wall to contact the susceptible bonds or be delivered there through direct contact with fungal hyphae.

Cellulose decomposition

While many fungi can decompose modified cellulose products, only a limited number are able to decompose cellulose in its native, highly

crystalline form (cotton, ramie, and wood pulp). These species are termed cellulolytic fungi.

Initial concepts on cellulolytic enzymes

The fungi capable of cellulolytic attack of fabrics have been studied intensively since the 1950's. Lists of cellulolytic fungi and reviews of the early research on their enzyme systems were assembled by Siu (1951), Gascoigne and Gascoigne (1960), Reese (1963), and Norkrans (1963, 1967).

Reese et al. (1950) worked with several *Trichoderma* species and postulated that cellulolytic fungi secreted an enzyme (C_1) that was able to separate the cellulose molecules in the crystalline regions and enzymes (C_x) that then both randomly and endwise depolymerized the separated chains into glucose, cellobiose, and oligosaccharides. This concept stimulated additional studies to identify and characterize the enzymes in the "cellulose" complex of several species of *Trichoderma* and other cellulolytic fungi (Iwasaki et al., 1964; Halliwell, 1965; Selby and Maitland, 1967; King and Vessal, 1969).

Until recently, "cellulase" was considered to be a multi-enzymatic system consisting of at least three enzymatic components that converted cellulose to glucose. C_1 was posed as the component that attacked crystalline cellulose and separated the chains by an unknown mechanism. To function, the system requires the presence of co-enzyme consisting of two types of β 1−4 glucanases.

Exo-β-1,4 glucanase successively removes single cellobiose or glucose units from the non-reducing end of the cellulose chain, while endo-β 1,4 glucanases cleave cellulose at random sites along the chain. β-glucosidases are attached to the hyphal wall and further decompose cellulose fragments by hydrolyzing the various oligomers including cellobiose that is converted to two molecules of glucose. Endo-glucosidases hydrolyze the smaller cellulose fragments and exo-glucanases the larger ones. The exo-glucanases depolymerize by inversion and release β-glucose units.

It was generally presumed that cellulase enzymes were also involved in the initial cellulose breakdown in wood decay, even though their identities and modes of action were based largely on studies of pure cotton cellulose. However, there is some question about the validity of applying this model to cellulose decomposition *in situ* in wood, since cellulose microfibrils in wood are surrounded by hemicelluloses and high concentrations of lignin. Furthermore, the inability of purified cellulase enzymes

to depolymerize non-modified cellulose in the absence of C_1-enzymes was puzzling and raised questions concerning the role of C_x in decay initiation. When examining the role of cellulose in wood decomposition, it is helpful to evaluate systems for white, brown, and soft rot fungi separately.

White-rot fungi: Studies of the white-rot fungus, *Phanerochaete chrysosporium*, suggest that the C_x enzyme is actually the exoenzyme that attacks both glucose and cellobiose from the ends of the cellulose molecules (Eriksson, 1969). The major step in enzymatic breakdown of cellulose involves hydrolytic cleavage of the β1,4 glycosidic bonds connecting the glucose monomers (Fig. 8.1A) followed by a series of hydrolytic and oxidative reactions (Eriksson, 1978). These steps can be outlined as follows:

Strong synergism has been reported between the endo- and exo-glucanases on some substrates (Streamer and Peterson, 1975). The molecular weights of the exoglucanase are around 50,000 kd, while endoglucanases range from 30,000 to 35,000 kd. The two *β-1,4* glucosidases are larger enzymes with molecular weights of 165,000 and 182,000 kd. It is interesting to consider the ability of smaller enzymes to penetrate the wood cell wall. Several proteases formed by *P. chrysosporium* are reported to enhance the activity of the endoglucanase (Eriksson and Peterson, 1982), suggesting that modification of the native enzyme enhances activity.

A series of reactions appear to regulate the enzymatic decomposition of cellulose and control the rate of glucose release so that it matches the capacity of the fungus to utilize it. The regulation reactions controlling glucose release from cellulose degraded by white-rot fungi are briefly outlined below:

1. An excess of glucose inhibits the release of the β1,4 endoglucanases (and possibly the exo-glucanases) by catabolite repression.

$$\text{glucose excess} \xrightarrow{\text{glucose oxidase}} \underset{(a\ \beta\text{-glycosidase inhibitor})}{\text{gluconolactone}} \rightarrow \underset{(\text{may be absorbed by hyphae})}{\text{gluconic acid}}$$

2. An excess of glucose induces the formation of glucono-lactone.

$$\underset{(\text{excess})}{\text{Cellobiose}} \xrightarrow[\text{Transglycosylation}]{\text{Endogluconases}} \text{Oligosaccharides} + H_2O$$

3. When β-glycosidase is inhibited by gluconolactone, cellobiose accumulates

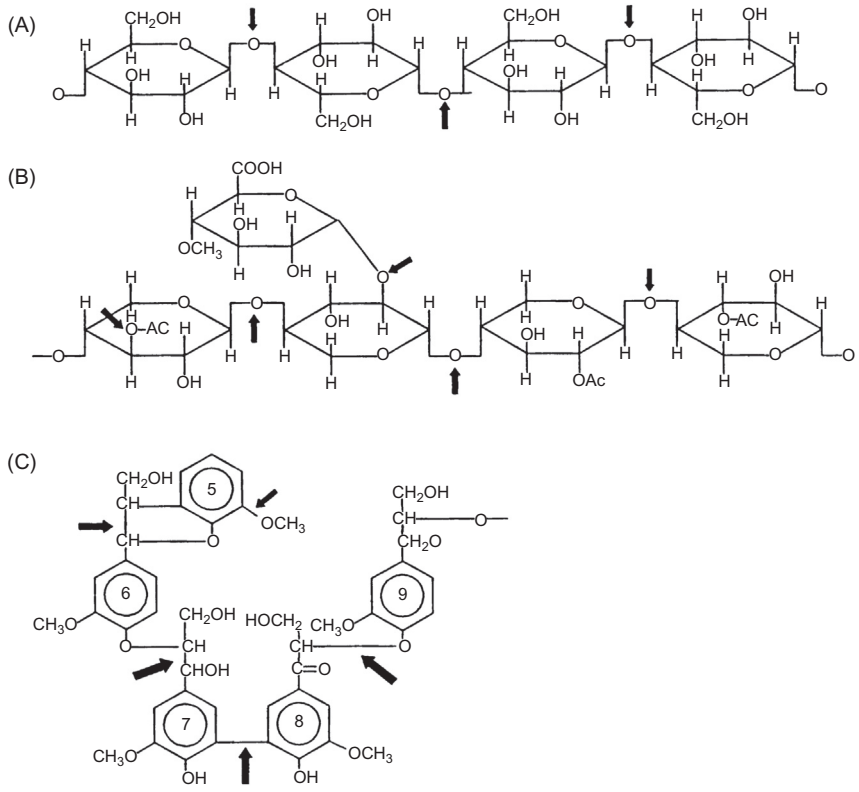


Figure 8.1 General sites for enzymatic cleavage of major wood cell-wall components during decay by a white rot fungus (A) hydrolytic cleavage of glucose, cellobiose, and glucose oligomer from cellulose by exo and endo-glucanases, (B) hydrolytic cleavage of xylose, xylobiose, and xylose oligomers from a xylan (O-acetl-4-O-methyl-glucuronoxylan) by endoxylanase and xylosidase. The methylglucose side branch and acetyl substituents are removed by other hydrolytic enzymes, (C) oxidative random cleavage of spruce lignin at C_{α}-C_{β} bonds by a ligninase that releases phenolic fragments. *(C) After Adler, E., 1977. Lignin chemistry --past, present, and future. Wood Sci. Technol. 11, 169–218.*

4. Transglycosylation and the formation of storage products (dextrins, etc.) may occur when an excess of simple sugars are present.
5. An excess of simple sugars may also inhibit the release of hemicellulases.

The regulation processes reduce excesses of simple sugars that may limit competition for the substrate from other wood-inhabiting saprobes. The absence of excess sugars also explains the absence of solubility charges in wood decayed by white-rot fungi.

Brown-rot fungi

As would be expected from the drastic strength losses and rapid drops in the degree of polymerization (DP) exhibited at early decay stages, the brown-rot fungi appear to extensively degrade cellulose in a manner that differs from the white-rot fungi. Brown-rot fungi utilize endo-β-1,4 glucanases, but lack exo-β-1,4 glucanases (Highley, 1975). As a result, the mechanism of attack on crystalline cellulose by brown-rot fungi is still under study. Since the dimensional constraints of the crystalline zones preclude enzyme access of the intact wood cell wall by diffusion, the initial attack on the cellulose is now believed to be through formation of free radicals that act as non-enzymatic depolymerizing agents (Cowling and Brown, 1969). Koenigs (1974) suggested that oxidation by an H_2O_2/Fe^{+2} system provided a non-enzymatic cellulose depolymerization agent. This process may also be related to early hemicellulose attack in brown rots. Lyr (1960) has shown that glucomannan, an important hemicellulose in conifers, is initially removed more rapidly than cellulose and its decomposition is associated with the release of H_20_2. Some brown-rot fungi, such as *Rhodonia* (*Poria) placenta* can utilize pure cellulose in culture in the presence of hemicelluloses (Highley, 1978). After the cellulose chains in the crystalline zones are separated, endo-β-1,4 glucanases similar to those in white-rot fungi randomly cleave the cellulose molecule and β-1,4 glucosidases convert the cellobiose to glucose. In contrast to the regulated process of cellulose decomposition associated with white-rot fungi, excesses of glucose and cellodextrins are present in brown rotted wood, but appear to play no role in cellulase regulation.

Soft-rot fungi

These fungi resemble the brown-rot fungi in that their attack is limited primarily to cell-wall carbohydrates, with demethoxylation of lignin. However, soft-rot fungi also resemble the white rotters in having similar hydrolytic enzyme systems.

Much of the early research on cellulolytic enzymes involved fungi now known to have soft-rot capabilities (e.g.*Trichoderma* spp.). Considerable efforts were devoted to developing mutant strains of *T. reesei* that produced high levels of "cellulase" for industrial use (Montenecourt and Eveleigh, 1977). Some soft-rot fungi utilize exo-1,4-β-glucanases, endo-l,4-β-glucanases, and 1,4-β glucanases to degrade cellulose. Other species utilize primarily the endo-1,4-β-glucanases and 1,4-β-glucosidases and appear to limit their attack

to the amorphous cellulose zones in the microfibrils. As in the white-rot fungi, synergism has been reported among the enzymes in the cellulase complex of soft rot fungi (Mandels and Reese, 1964). These differences suggest that the soft-rot fungi, now defined broadly as "non-Basidiomycete" decayers, are a cosmopolitan group of fungi with rather diverse mechanisms for degrading cellulose. This would suggest that a number of very different organisms have developed similar strategies to access cellulose.

Hemicelluloses decomposition

Many bacteria, yeasts, and fungi can degrade and utilize the hemicelluloses in plant tissues. Hemicelluloses are often the first cell wall components degraded by decay fungi, probably due to their shorter chain lengths, solubility, and exposed locations around the cellulose microfibrils.

Hemicellulose degrading enzymes are primarily hydrolytic in nature and the attack patterns are analogous to cellulase degradation of cellulose. However, exoenzymes are absent, probably reflecting the low DP (~ 200) of hemicelluloses. Since the major wood hemicelluloses are heteropolymers consisting of several sugars, side branches and substituent groups, the enzymatic processes involved in degradation are much more complicated and are just beginning to be elucidated. As a result, no comparisons among decay types can be made.

We will limit our attention to the enzymes involved in the digestion of xylan (O-acetyl-4-0-methyl-glucurono-β-D-xylan) and mannan (galactoglucomannans). Enzymatic degradation of the other hemicelluloses has been reviewed elsewhere (Dekker, 1985; Eriksson et al.,1990).

Xylans. The breakdown of xylan requires the action of several xylanolytic enzymes (Fig. 8.1). These enzymes and their functions are as follows:

a. Endo-l,4-β-xylanases separate the polymer backbone into xylose and its oligomers. Endo-xylanases have been isolated from *Trichoderma* spp., *Coniophora cerebella*, *Rhodonia* (*Poria*) *placenta*, and *Irpex lacteus*. These enzymes are analogous to the endo-glucanases in the "cellulase" enzyme complex.

b. 1,4-β-xylosidases hydrolyze the xylo-oligosaccharides or xylan fragments to xylose. These enzymes have been obtained from fungi in a

wide range of genera (Reese et al., 1973) and appear to be inducible enzymes. The enzymes are cell-wall bound and analogous to the B-glucosidases in the "cellulose" enzyme complex.

c. α-glucuronidase separates 4-0-methylglucuron sidechains from the xylan backbones and releases glucuronic acid units.

d. α-arabinosidase removes L-arabinose side chains, greatly increasing the effectiveness of the endoxylanases.

e. Acetyl esterase removes the acetyl substituent groups from the xylose. Xylan esterases have been obtained from *Trichoderma reesi* and *Schizophyllum commune*.

Mannans: The mannans in wood are heteropolymers and their degradation requires the combined action of several enzymes to remove the various sugars comprising the polymer backbone and the galactose and acetyl side chains. The enzymes required have modes of action similar to the xylanases and include endo1,4-β-mannase, β-mannosidase, β-glucosidase, α-galactosidase, and acetyl esterase.

Endomannases have been obtained from a wide range of organisms, including white, brown, and soft-rot fungi. The sequences of enzymatic attack of mannans are unknown, but various patterns of degradation have been proposed by Dekker (1985).

The utilization rates of the various wood hemicelluloses by white-rot fungi suggest that there are substantial differences among fungal species in the amounts of hemicellulase produced and the sequences by which these enzymes degrade the substrate.

Hemicellulases have come under more intensive study because of their potential commercial applications for removing residual xylans from pulp and for converting pentose sugars in agricultural wastes to proteins or release of sugars for subsequent fermentation to ethanol.

Lignin decomposition

Lignin is resistant to degradation by most microorganisms and, indeed, its primary role in the wood cell wall is to protect the carbohydrates from microbial attack. Lignin is efficiently degraded in nature primarily by white-rot fungi, including many litter-decomposing basidiomycetes.

The complex chemical structure of lignin has made it difficult to understand fungal mechanisms of decomposition.

As expected, there are major differences between fungal degradation of lignin and carbohydrates. Lignin breakdown is accomplished by a limited group of specialized fungi (white rotters), while many microorganisms successfully degrade and utilize wood carbohydrates. Lignin decomposition proceeds by oxidative reactions that separate carbon to carbon bonds or ether linkages and separate various functional groups, side chains, and aromatic rings randomly from the huge, amorphous lignin macromolecule (Fig. 8.1C) rather than the uniform hydrolytic cleavages of β-1,4 glycosidic bonds common to the carbohydrates. White-rot fungi can completely degrade lignin in wood in laboratory decay tests. Yet, the end-product of lignin decomposition in nature contains partially decomposed, fragmented lignin (humus) that enters the soil cycle, and remains in this layer for many years. Release of H_2O and CO_2 as decomposition products in this layer is extremely limited. Soil bacteria, microfauna, and even physical processes may play important roles in the final breakdown of humus. This is a slow process and some lignin degradation products have soil residence times of centuries (Ziekus, 1981).

Great progress has been made in the past decade toward understanding the biodegradation of lignin.

A number of review articles or textbook chapters presented detailed accounts of the many chemical changes in decayed lignin, the enzymes involved, and tentative biodegradation pathways [Kirk and Farrell; 1987; Chen and Chang, 1985; Higuchi, 1985; Eriksson et al., 1990]. For our purposes, coverage of this complex topic will be limited to the major chemical changes; the enzymes involved in lignin degradation and a comparison of lignin degradation by the three major groups of decay fungi.

Lignin determination

Caution should be exercised in judgments on lignin degradation and metabolism since lignin determination can vary considerably with the analytical method employed. Gravimetric determination of the sulfuric-acid insolubles in the wood as lignin (Klason lignin) is widely used because of its simplicity, but this method includes some extractives and degraded products in the soluble fraction, artificially inflating lignin losses. This method also dramatically alters the chemical structure of the residual lignin, making it less useful for studying the structure of decomposed lignin.

As an alternative, a lignin can be synthesized by oxidation of the lignin precursors with hydrogen peroxidase. This material, called dehydrogenation polymerizate (DHP) lignin, has particular advantages in that carbohydrates that are difficult to remove from native lignins are absent. Lignin structures have also been studied using milled wood lignin (MWL) (Bjorkman lignin) that is extracted from finely ground or milled wood by solvents. MWL is generally accepted as a preferred lignin preparation method for critical studies. Other lignin preparation methods and their advantages and disadvantages have been reviewed by Crawford (1981). Useful clues on the lignin degradation process have come from the analysis of residual products in partially decayed lignin, the identification of the many low-molecular weight degradation products, and the use of isotope labeled dimeric lignin model compounds.

Chemical modification in decayed lignin

The analysis of residual lignin at various stages of decay by white-rot fungi indicates a steady loss of methoxyl groups and increases in oxygen and hydroxyl content. The major structural changes suggested by Kirk and Chang (1975) include degradation products in the low molecular weight solubles include vanillin, syringaldehyde, coniferyl aldehyde, vanillic acid, syringic acid, and a wide range of aliphatic or aromatic acids and phenols (Figs. 8.2 and 8.3).

Lignin degrading enzymes from white-rot fungi

Remarkable progress has been made in determining the enzymatic nature and probable pathways of lignin digestion since the initial identification of a ligninase (Tien and Kirk, 1983, 1984; Gold et al., 1984). The enzymes now believed to play a role in this complicated process are the lignases (lignin peroxidase), manganese-peroxidases, phenol-oxidizing enzymes such as laccase, and peroxidase producing enzymes (Kirk and Shimada, 1985).

Ligninases are peroxidase enzymes that require H_2O_2 as a reactant in their catalytic reactions with lignin. These enzymes have been detected in cultures of *Phanerochaete chrysosporium*, *Trametes versicolor* and several other white-rot fungi, and function by cleaving the C_α-C_β bond in the phenyl propane units (Fig. 8.3). Liginase oxidation may also cleave the phenolic rings. The non-specific oxidation of the lignin units by this enzyme produces a wide array of products. The non-specificity of these reactions and their variety led Kirk and Farrell (1987) to describe the process with the

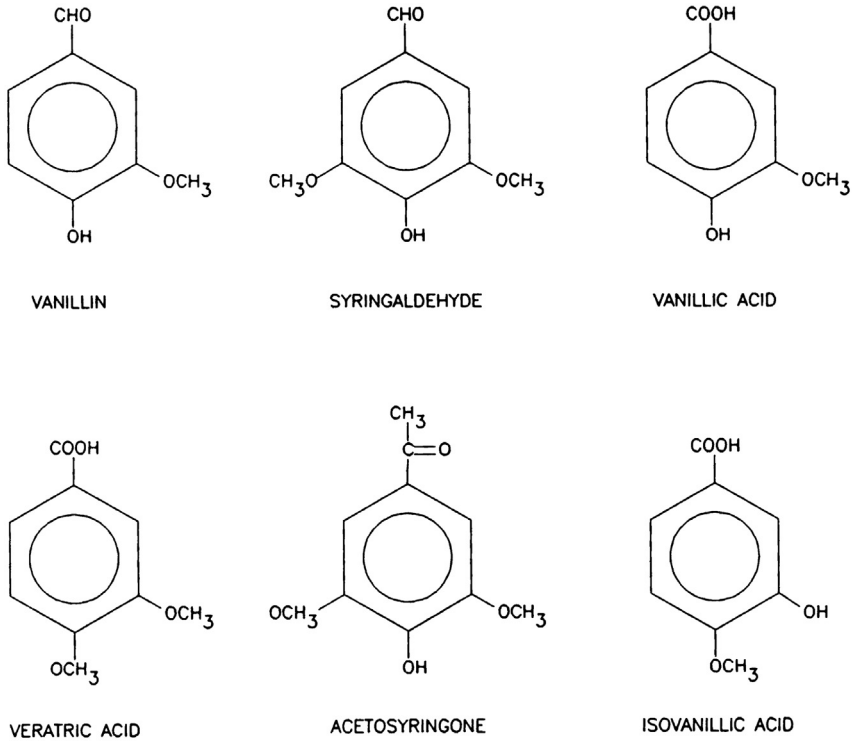


Figure 8.2 Examples of some common low-molecular weight products obtained from partially degraded lignin.

novel term of "enzymatic combustion". Cultural studies indicate that ligninase is a secondary metabolite released in liquid cultures when nitrogen becomes limiting. Most woods contain very little nitrogen and enzyme induction at low nitrogen levels probably reflects an adaptation for this environment.

Manganese peroxidases oxidize the syringyl compounds better than guaiacyl compounds and require the presence of Mn + 2, but the role of this enzyme in lignin degradation is still unclear (Glenn and Gold, 1985). Manganese peroxidases may play a role similar to the phenol-oxidizing enzymes and function in peroxide formation. Manganese accumulation has been found in white-rotted wood (Blanchette, 1984), suggesting that this metal may play significant, but as yet unknown, roles in lignin decomposition.

Laccase: Other phenol oxidizing enzymes are produced by most white-rot fungi and their presence has long suggested a role in lignin

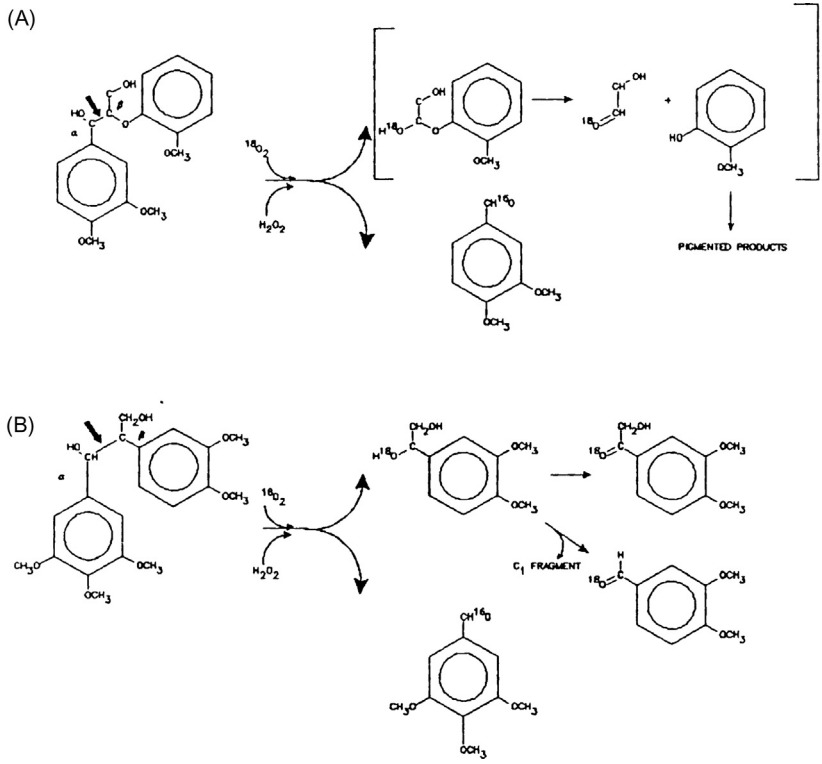


Figure 8.3 Cleavage of the C_α-C_β chemical bond between the carbons in the propane chains of two dimeric models representing two major linkage patterns between phenylpropane units in lignin and their probable degradation products. (A) Cleavage in a β-O-4 model representing the syringyl glycerol-aryl-ether bond and (B) cleavage in a β-1 model representing the diarylpropane bond (Tien and Kirk, 1984).

decomposition (Kirk and Kelman, 1965). The color changes induced by these enzymes in gallic and tannic acid media are the basis of the well-known Bavendamn reaction for separating the white rot fungi from the brown rotters (Davidson et al., 1938).

Laccase is reported to cause C_α-oxidation, demethoxylation, cleavages in phenyl groups and C_α-C_β cleavage in syringyl structures (Kirk and Farrell, 1987). Laccase also provides the phenoxyradicals and quinones from lignin decomposition that play key roles in the decomposition of cellobiose by cellobiose dehydrogenase. One puzzle to the function of laccase is the mechanism that prevents the rapid repolymerization of the phenoxyradicals back into lignin since phenoxyradicals are starting materials for lignin biosynthesis. The removal of toxic quinones may prevent

repolymerization in the degradative reaction. A number of white-rot fungi lack laccase, but other polyphenol oxidases can apparently carry out the same reactions.

H_2O_2 -forming enzymes are required for the ligninase reactions described above. Glucose oxidase and other oxidases have been suggested as the enzymes that produce H_2O_2 from various sugars possibly in peroxisome-like organelles in the periplasmic zone adjacent to the hyphal cell wall.

Lignin degradation by non-white-rot fungi

While brown-rot fungi are not considered to be lignin degraders, chemical analyzes of residual lignin from brown-rotted wood indicates some degradation and substantial increases in solubility. There is emerging evidence that brown rot fungi produce considerable depolymerization of lignin but these products them subsequently repolymerize, leaving decreased methoxyl content, oxidations of some alcohol and aldehyde groups to carboxyls, and the introduction of some phenolic hydroxyls (Kirk,1975). There appears to be, however, no significant permanent separation of the guaiacyl and syringyl units comprising the amorphous macromolecule. Lignin losses during decay have been reported for some brown-rot fungi (Enoki et al., 1988). Whether these losses represent natural variation in lignin degradation within brown-rot fungi or differences in the method of lignin detection is uncertain. Degradation studies of labeled dimeric lignin models also indicate a substantial variation among brown-rot fungi (Tanaka et al., 1986). *Gloeophyllum trabeum*, *Neolentinus lepideus*, and *Pholiota adiposa* were the most active of the 11 brown rotters tested, suggesting that more detailed studies may reveal some intermediates between the white-rot and brown-rot fungi. The classification of white and brown-rot fungi is an artificial system, and the occurrence of species with characteristics intermediate between both groups is not surprising.

Soft-rot fungi have been reported to cause considerable lignin degradation (Savory and Pinion, 1958; Levi and Preston, 1965; Eslyn et al., 1975). Some species degrade more lignin than the brown-rot fungi (Seifert, 1966) and preferential attack of the syringyl units has been observed (Nilsson et al., 1989). The enzymes involved in soft rot attack of lignin are not known, but the yeast phases of many soft rot fungi may be extremely useful in lignin degradation schemes since filamentous fungi

often pose problems in fermentation systems. As a group, the soft-rot fungi exhibit considerable variation in their attack on cell wall components and appear to be intermediate in their lignin degrading capabilities between the white-and brown-rot groups.

Bacterial roles in wood decomposition

Bacteria are commonly present in wood during all the stages of its decomposition and their many roles can be grouped conveniently into four general categories, (a) primary log invaders that damage pits in parenchymatous tissues increasing wood permeability, (b) heartwood invaders in living stems that cause wetwood and often decrease wood permeability, (c) species associated with other wood-inhabiting flora or fauna that may antagonistically or synergistically interact with them; and (d) species capable of degradation of wood or its isolated major cell-wall components. This latter role is discussed in this section, while the others are reviewed in Chapter 11.

While some bacteria can degrade wood, the damage is often slight and develops very slowly compared to fungal attack. Bacteria are unicellular and are unable to invade wood substrates as rapidly as filamentous fungi. Soil inhabiting actinomycetes; however, can assume a mycelial form and some species cause soft-rot damage to wood in marine usages or under extreme nutrient/moisture regimes.

Many bacteria decompose the pectins and hemicelluloses in plant materials. Cellulose is also readily decomposed by some specialized aerobic bacteria and also by consortia of bacteria with other microorganisms, anaerobically during ruminant and termite digestion. Bacteria growing on hemicelluloses and cellulose are a major source of the troublesome slime deposits that develop during the pulp and paper making processes. Bacteria are also able to utilize many of the low-molecular weight lignin degradation products in partially decayed lignin and pulping waste liquors. Bacteria can cleave benzene rings and some species are being proposed as agents for detoxifying chlorinated phenols. Crawford (1981) reported that some strains of bacteria degraded lignin, lignin model compounds, and DHP lignin in a manner similar to the white rot fungi by a process involving substituent oxidations and cleavage of both 2 carbon fragments from propyl side chains and aryl ether linkages.

In wood, however, bacterial damage is generally minor compared to fungal damage, and is largely limited to cell wall surface etching, erosion in sapwood zones, and degradation of pit membranes. Decay tests on large numbers of wood-inhabiting bacteria have generally been negative (Schmidt and Dietrichs, 1976). Lignification appears to be the major factor limiting significant bacterial attack in most woods and delignification of wood substantially increases the severity of bacterial degradation.

Several studies have shown that *Streptomyces* species (Actinomycetes) cause soft rot (Type 1) in wood in some marine exposures (Cavalcante and Eaton, 1980; Baecker and King, 1982; Holt, 1983). A unique tunneling type of bacterial damage has been noted in wood cell walls and has been attributed to unidentified bacteria (Daniel et al., 1987). The damage to the cell wall is severe and cultural studies demonstrate degradation of radioisotope labeled lignin.

These reports suggest that bacteria in the Actinomycetes and Eubacteria may play important roles in wood decomposition in certain environments. The significance of these organisms in large-scale deterioration of wood products deserves further studies to more accurately quantify their roles.

A decay model and related research needs

A tentative model of the major steps involved in the decay process for a typical brown and white rot can both emphasize current views and demonstrate how much is still unknown and awaiting additional research.

1. The decay process begins when the hyphal tip of a fungus with decay capabilities penetrates a wood tracheid and grows along the lumen surface in a moisture film (above the f.s.p.) that bathes the hypha and serves as a transport medium.
2. Under favorable growth conditions, the presence of certain soluble cell wall compounds elicit synthesis and secretion of enzymes through the hyphal walls by exocytosis. The enzymes or chemical agents either diffuse with moisture to the wood cell wall or are carried there in a hyphal sheath.
3. Non- enzyme agents, characteristic of all decay fungi, alter the chemical bonds in the lignin shield or those between lignin and the

hemicelluloses to facilitate enzyme access to carbohydrates; the lignin barrier around the hemicelluloses and cellulose microfibrils is not complete and some enzymes are able to access the unprotected zones; or the concerted action of several enzymes operating simultaneously exposes carbohydrates.

4. Hemicelluloses in the amorphous. zones of the microfibrils are attacked initially. Whether the removal of substituent groups, and polymer debranching occurs before, during, or after depolymerization of the polymer backbone is unknown. The sequences of the removal of various sugars in the several heteropolymers comprising the different hemicelluloses in various wood species remain poorly understood.
5. Subsequent events vary with decay type and are discussed in parallel to emphasize both differences and similarities of the two systems.

Brown rot fungi	White rot fungi
a. H_2O_2 is formed from hemicellulose sugars and may react with Fe^{+2} to form an unidentified oxidizing agent that diffuses into microfibrils to separate cellulose molecules in the crystalline region and randomly break some glycosidic bonds.	aa. H_2O_2 is formed from oxidases (GLOX) associated with hemicellulose degradation and ligninase (LIP) and manganese peroxidase (MnP) are synthesized and released. Free radicals are formed and lignin degradation begins. This involves the separation of propyl chains between C_α-C_β carbons, aryl ether bond separation and some benzene ring splitting
b. Glucose induces cellulase formation	bb. Cellobiose induces cellulase formation. Cellulase and additional hemicellulases are then released as the lignin barrier is disrupted. Synergism occurs between 1.1,44-β exo and endo-gluconases. Several control mechanisms appear to limit accumulation of excess sugars.

c. Cellulose and hemicellulase enzymes are released and are primarily endo-enzymes. Synergisms probably occurs between enzymes types for the 1,4-β glycosidic bonds. Some fungi produce these enzymes in a larger complex.	cc. Cellobiose is oxidized to cellobionic acid and other compounds by a oxido-reductase enzyme via the reduction of lignin fragments such as quinones by laccases. This may be more of a detoxification mechanisms to remove toxic lignin breakdown products
d. Glycosidases on the hyphal wall depolymerize cellobiose and the dimers of hemicellulose fragments into glucose or equivalent simple sugars and excesses accumulate.	dd. Laccase and other oxidase enzymes also play a role in lignin degradation
e. Lignin degradation, via free radical activity occurs by demethoxylation, some propyl side chain splitting, oxidation involving formation carbonyl groups and hydroxylation of benzene rings. While lignin monomer groups remain linked, some fragmentation occurs and the protective role of lignin for carbohydrates is compromised	ee. Lignin is degraded and metabolized. Syringyl monomer units appear to be degraded more readily than guaiacyl units. It remains unclear whether lignin degradation is energetically positive or negative for the fungus
f. A degraded lignin is all that remains at the end of the decay process which consumes up to 60–70% of the original cell mass	ff. Only a few residual minerals remain at the end of the decay process

6. The simple sugars and hydrocarbon fragments from the decay process are absorbed by active hyphal transport processes completing the external digestive process (decay). The simple carbon compounds enter

various metabolic pathways and are converted to ATP for synthesis and energy purposes or stored as food reserves.

7. Differing sequences and amounts of the various enzymes and chemical agents involved in the decay process probably explain the many types of decay. Since the enzymes are under gene control, the developing methods of biotechnology offer exciting possibilities for developing decay fungi for specific commercial purposes.

Most decay mechanism studies have been based on single enzyme tests on substrates in fermentation flasks. The natural decay processes probably involves a cascade of simultaneous reactions that are highly interactive. The situation is further complicated by microbial interactions that are either competitive or synergistic. Thus, while great progress has been made toward understanding decay, it is clear that much remains to be elucidated.

Summary

1. White-rot fungi degrade and metabolize all major cell-wall constituents proceeding from the lumen surface inward while leaving the residual cell wall material intact. Wood degraded by white rot fungi is characterized by low alkali solubility and only minor reductions in the degree of polymerization (DP) of the cellulose. White-rot fungi vary in the degree of early attack on lignin and cellulose. These differences may be exploited by identifying species or developing strains that preferentially remove lignin for biopulping and biobleaching possibilities. Strength losses in white rotted wood are approximately proportional to specific gravity reductions.
2. Brown-rot fungi primarily depolymerize and metabolize the structural carbohydrates in the wood cell wall. Lignin is partially degraded, but remains as an un-utilized residuum. Brown-rot fungi develop rapidly in the S1 and S2 zones of the cell wall causing dramatic reductions in DP and increasing alkali solubility. These changes are also associated with early, drastic reductions in wood strength properties.
3. The soft-rot fungi have capabilities that appear to lie between the white-and brown-rot fungi. These fungi concentrate their attack on

the cell-wall carbohydrates, but cause more lignin degradation than brown-rot fungi. Soft-rot degradation is primarily localized in the S2 zone of the cell wall. As with the white rots, soft rotted wood is characterized by only minor reductions in alkali solubility.

4. Structural carbohydrates in all decays are depolymerized by hydrolytic enzymes. The major cellulose degrading enzymes are endo-l,4-β-glucanase, exo-1-4-β glucanase, and 1,4-β-glucosidase. Hemicelluloses are more structurally complex and require a more complex set of enzymes to debranch and remove substituent groups as well as depolymerize the polymer backbone. Some of the enzymes involved are endo-I1,4 -4-β-xylanase, 1 1,4-4-β xylosidase, their mannan equivalents and several acetyl esterases.
5. Small, "diffusible" oxidants that separate and depolymerize cellulose molecules in the crystalline zones and are responsible for the early sharp reductions in wood strength and DP are believed to be produced by brown rot fungi.
6. Lignin is degraded by all three decay types, but the degradation is minor in the brown-and soft-rots and involves primarily demethoxylation. Lignin is completely degraded and metabolized by white rot fungi. Oxidation by several peroxidase enzymes is the primary breakdown mode, although methoxyl groups are hydrolyzed by methyl esterases. The enzymatic degradation of lignin is just beginning to be understood and has been studied for only a few white-rot fungi. The major enzymes involved directly or indirectly in the random oxidation process are ligninase peroxidase, manganese peroxidase, other polyphenol oxidizing enzymes, and H_2O_2 forming enzymes.
7. While great progress has been made to determine the degradative sequences and the enzymes involved in the decomposition of cellulose, the hemicelluloses, and lignin, much remains to be clarified. Major research is still needed to better understand the concerted action and coordination of all the constituent enzymes on the wood. There are also questions concerning the nature and mechanism by which rapid cellulose depolymerization occurs in early stages of brown rot development. Finally, the protective role of lignin and the occurrence of uneven deposition zones around the microfibrils which may provide sites for enzyme contact merit further research. It is prudent to remember that our current ideas on the chemical mechanisms of wood decay are based on detailed studies of only a few fungi in each major decay type. As more fungi are studied, including the neglected

higher Ascomycetes, the litter-decomposing Basidiomycetes, and soil fungi, we may find that the fungi have solved the lignin barrier to energy rich carbohydrates in a variety of other interesting and novel ways.

References

Adler, E., 1977. Lignin chemistry --past, present, and future. Wood Sci. Technol. 11, 169–218.

Baecker, A.A.W., King, B., 1982. Soft rot in wood caused by *Streptomyces*. J. Inst. Wood Sci. 10, 65–71.

Blanchette, R.A., 1984. Manganese accumulation in wood decayed by white-rot fungi. Phytopathology 74, 725–730.

Cavalcante, M.S., Eaton, R.A., 1980. The Isolation of Actinomycetes From Wood in Ground Contact and the Sea. Int. Res. Group Wood Preserv. Document IRG/W P/1110, Stockholm, Sweden.

Chen, C.-L., Chang, H.-M., 1985. Chemistry of lignin biodegradation. In: Higuchi, T. (Ed.), Biosynthesis and Biodegradation of Wood. Academic Press, New York, pp. 535–556. , 679 pp.

Côté, W.A., 1977. Wood ultrastructure in relation to chemical composition. In: Loewus, F.A., Runeckles, V.C. (Eds.), Recent Advances in Phytochemistry: The Structure, Biosynthesis and Degradation of Wood. Plenum Press, NY.

Côté, W.A., Timell, T.E., Zabel, R.A., 1966. Distribution of lignin in compression wood of red spruce (*Picea rubens* Sarg.). Holz Roh-Werkst. 24, 432–438.

Cowling, E.B., 1961. Comparative Biochemistry of the Decay of Sweetgum Sapwood by White-rot and Brown-rot Fungi. U.S. Dept. Agric. Tech. Bull. No. 1258.

Cowling, E.B., Brown, W., 1969. Structural features of cellulosic materials in relation to enzymatic hydrolysis. In: Hajny, G.J., Reese, E.T. (Eds.), Cellulases and Their Applications. Chap. 10, Advanced Chemistry Series, 95. American Chemical Society Press, Washington, DC, pp. 152–187.

Crawford, R.L., 1981. Lignin Biodegradation and Transformation. John Wiley and Sons, New York, 137 pp.

Daniel, G.F., Nilsson, T., Singh, A.P., 1987. Degradation of lignocellulosics by unique tunnel-forming bacteria. Can. J. Microbiol. 33, 943–948.

Davidson, R.W., Campbell, W.A., Blaisdell, D.J., 1938. Differentiation of wood decaying fungi by their reactions on gallic or tannic acid medium. J. Agric. Res. 57, 683–695.

Dekker, R.F.H., 1985. Biodegradation of hemicellulose. In: Higuchi, T. (Ed.), Biosynthesis and Biodegradation of Wood Components. Academic Press, New York, pp. 505–533.

Enoki, A.H., Tanaka, Fuse, G., 1988. Degradation of lignin-related compounds, pure cellulose and wood components by white rot and brown rot fungi. Holzforschung 42, 85–93.

Eriksson, K.L., 1969. In discussion of the paper by J.M. Leatherwood. Cellulose complex of *Rumincoccus* and a new mechanism for cellulose degradation. In: Hajny, G.T., Reese, E.T. (Eds.), Cellulases and Their Applications, Advanced Chemical Series, 95. American Chemical Society Press, Washington, DC, pp. 58–59.

Eriksson, K.E., 1978. Enzyme mechanism involved in cellulose hydrolysis by the white rot fungus, *Sporotrichum pulverulentum*. Biotechnol. Bioeng. 20, 317–332.

Eriksson, K.E., Peterson, B., 1982. Purification and partial purification of two acidic proteases from the white-rot fungus *Sporotrichum pulverlentum*. Eur. J. Biochem. 124, 635–642.

Eriksson, K.-L.L., Blanchette, R.A., Ander, P., 1990. Microbial and Enzymatic Degradation of Wood and Wood Components. Springer-Verlag, New York, p. 407.
Eslyn, W.E., Kirk, T.K., Effland, M.J., 1975. Changes in the chemical composition of wood caused by six soft-rot fungi. Phytopathology 65, 473–476.
Fengel, D., Wegener, G., 1984. Wood Chemistry, Ultrastructure, Reactions. Walter de Gruyter, Berlin, 613 pp.
Gascoigne, J.A., Gascoigne, M.M., 1960. Biological Degradation of Cellulose. Buttersworth, London, 254 pp.
Glenn, J.K., Gold, M.H., 1985. Purification and characterization of an extracellular Mn (II)-dependent peroxidase from the lignin-degrading basidiomycete, *Phanerochaete chrysosporium*. Arch. Biochem. Biophys. 242, 329–341.
Gold, M.H., Kuwahara, M., Chin, A.A., Glenn, J.K., 1984. Purification and characterization of an extracellular H_2O_2-requiring diarylpropane oxygenase from the white-rot basidiomycete, *Phanerochaete chrysosporium*. Arch. Biochem. Biophys. 234, 353–362.
Halliwell, G., 1965. Total hydrolysis of celluloses by cell-free preparations from soil micro-organisms. Biochem J. 94, 270–289.
Highley, T.L., 1975. Properties of cellulases of two brown-rot fungi and two white-rot fungi. Wood Fiber 6 (4), 275–281.
Highley, T.L., 1977. Requirements for cellulose degradation by a brown-rot fungus. Mater. Org. 12, 25–36.
Highley, T.L., 1978. Degradation of cellulose by culture filtrates of *Poria placenta*. Mater. Org. 12 (3), 161–174.
Higuchi, T. (Ed.), 1985. Biosynthesis and Biodegradation of Wood Components. Academic Press, New York, 679 pp.
Holt, D.M., 1983. Bacterial degradation of wood cell walls in aerobic aquatic habitats: decay patterns and mechanisms proposed to account for their formation. J. Inst. Wood Sci. 9, 212–223.
Iwasaki, T., Hayashi, K., Funatsu, M., 1964. Purification and characterization of two types of cellulase from *Trichoderma konigii*. J. Biochem. 55, 209–212.
King, K.W., Vessal, Ml., 1969. Enzymes of the cellulase complex. In: Hajny, G.J., Reese, E.T. (Eds.), Cellulases and Their Applications, Advanced Chemistry Series, Chapt. 2, 95. American Chemical Society, Washington, DC, pp. 7–25.
Kirk, T.K., 1971. Effects of microorganisms on lignin. Ann. Rev. Phytopathol. 9, 185–210.
Kirk, T.K., 1973. Chemistry and biochemistry of decay. Chap. 4. In: Nicholas, D.O. (Ed.), Wood Deterioration and Its Prevention by Preservative Treatments. Syracuse University Press, Syracuse, NY, pp. 149–181.
Kirk, T.K., 1975. Effects of a brown-rot fungus, *Lenzites trabea*, on lignin in spruce wood. Holzforschung 29 (3), 99–107.
Kirk, T.K., Kelman, A., 1965. Lignin degradation as related to the phenol oxidases of selected wood-decaying basidiomycetes. Phytopathology 55, 739–745.
Kirk, T.K., Moore, W.E., 1972. Removing lignin from wood with white-rot fungi and digestibility of resulting wood. Wood Fiber 4, 72–79.
Kirk, T.K., Highley, T.L., 1973. Quantitative changes in structural components of conifer woods during decay by white-and brown-rot fungi. Phytopathology 63 (11), 13381342.
Kirk, T.K., Chang, H.-M., 1975. Decomposition of lignin by white-rot fungi. II. Characterization of heavily degraded lignins from decayed spruce. Holzforschung 29, 56–64.
Kirk, T.K., Shimada, M., 1985. Lignin biodegradation: the microorganisms involved and the physiology and biochemistry of degradation by white-rot fungi. In: Higuchi, T. (Ed.), Biosynthesis and Biodegradation of Wood Components. Academic Press, Orlando, FL.

Kirk, T.K., Farrell, R.L., 1987. Enzymatic "combustion": The microbial degradation of lignin. Ann. Rev. Microbiol. 41, 465–505.
Kirk, T.K., Higuchi, T., Chang, H.M., 1980. Lignin Biodegradation: Microbiology Chemistry and Potential Applications, vols. 1, 2. CRC, Boca Raton, FL, 241 pp., 255 pp.
Koenigs, J.W., 1974. Hydrogen peroxide and iron: a proposed system for decomposition of wood by brown-rot basidiomycetes. Wood Fiber 6, 66–80.
Levi, M.P., Preston, R.D., 1965. A chemical and microscopic examination of the action of the soft-rot fungus *Chaetomium globosum* on beechwood (*Fagus sylvatica*). Holzforschung 19, 183–190.
Liese, W., 1970. Ultrastructural aspects of woody tissue disintegration. Ann. Rev. Phytopath. 8, 231–258.
Loewus, F.A., Runeckles, J.C. (Eds.), 1977. The Structure, Biosynthesis, and Degradation of Wood. Plenum Press, NY.
Lyr, H., 1960. Formation of ecto-enzymes by wood destroying and wood inhabiting fungi on various culture media. V. Complex medium as carbon source. Arch. Microbiol. 35, 258–278.
Mandels, M., Reese, E.T., 1964. Fungal cellulases and the microbial decomposition of fabrics. Dev. Ind. Microbiol. 5, 5–20.
Montenecourt, B.S., Eveleigh, D.E., 1977. Preparation of mutants of *Trichoderma reesii* with enhanced cellulase production. Appl. Environ. Microbiol. 34, 777.
Morrell, J.J., Zabel, R.A., 1987. Partial delignification of wood: its effect on the action of soft-rot fungi isolated from preservative-treated southern pines. Mater. Org. 22, 215–224.
Nilsson, T., Daniel, G., Kirk, T.K., Obst, J.R., 1989. Chemistry and microscopy of wood decay by some higher Ascomycetes. Holzforschung 43, 11–18.
Norkrans, Birgitta, 1963. Degradation of cellulose. Ann. Rev. Phytopathol. 1, 325–350.
Norkrans, Birgitta, 1967. Cellulose and cellulolysis. Adv. Appl. Microbiol. 9, 91–130.
Reese, E.T., 1963. Advances in Enzymatic Hydrolysis of Cellulose and Related Materials. Pergamon, New York.
Reese, E.T., 1977. Degradation of polymeric carbohydrates by microbial enzymes. In: Loewus, F.A., Runeckles, J.C. (Eds.), Recent Advances in Phytochemistry: The Structure, Biosynthesis and Degradation of Wood. Plenum Press, NY.
Reese, E.T., Siu, R.G.H., Levinson, H.S., 1950. The biological degradation of soluble cellulose derivatives and its relationship to the mechanism of cellulose hydrolysis. J. Bacteriol. 59, 485–497.
Reese, E.T., Maguire, A., Parrish, F.W., 1973. Production of β-D-xylopyranosidases by fungi. Can. J. Microbiol. 19, 1065–1074.
Ruel, K., Barnoud, 1985. Degradation of wood by microorganisms. In: Higuchi, T. (Ed.), Biosynthesis and Biodegradation of Wood Components. Academic Press, New York, 679 pp.
Savory, J.G., Pinion, L.C., 1958. Chemical aspects of decay of beech wood by *Chaetomium globosum*. Holzforschung 12 (4), 99–103.
Schmidt, 0., Dietrichs, H.H., 1976. Zur Aktivitat von Bakterien gegenuber Holzkamponenten, Mater. Org. Suppl, 3. pp. 91–102.
Seifert, K., 1966. Die chemische Veranderung der Bucherholz-Zellwand durch Moderfaule (*Chaetomium globosum* Kunze). Holz als Roh-und Werkst. 24 (185), 189.
Selby, K., Maitland, C.C., 1967. The cellulase of *Trichoderma viride* cellulolytic-complex. Biochem. J. 104 (716), 724.
Siu, R.G.H., 1951. Microbial Decomposition of Cellulose. Reinhold Pub. Corp., NY.
Sjostrom, E., 1984. Wood Chemistry: Theory and Applications. Academic Press, New York.

Streamer, M.K.-E., Peterson, B., 1975. Extracellular enzyme system utilized by the fungus *Sporotrichum pulverlentum* (*Chrysosporium lignorum*) for the breakdown of cellulose. IV. Functional characterization of five-endo-l,4-B-glucanase. Eur. J. Biochem. 59, 607–613.

Tanaka, H., Enoke, A., Fuse, G., 1986. Correlation between ethylene production from α-*oxo*—gamma-methylthiobutyric acid and degradation of lignin dimeric model compounds by wood-inhabiting fungi. Mokuzai Gakkaishi 32, 125–135.

Tien, M., Kirk, T.K., 1983. Lignin-degrading enzyme from the Hymenomycete *Phanerochaete chrysosporium* Burds. Science 221, 661–663.

Tien, M., Kirk, T.K., 1984. Lignin-degrading enzyme from *Phanerochaete chrysosporium*: purification, characterization, and catalytic properties of a unique H202-requiring oxygenase. Proc. Natl. Acad. Sci. USA 81, 2280–2284.

Timell, T., 1967. Recent advances in the chemistry of wood hemicelluloses. Wood Sci. Tech. 1, 45–70.

Ziekus, J.G., 1981. Lignin metabolism and the carbon cycle: polymer biosynthesis, biodegradation, and environmental recalcitrance. Adv. Microb. Ecol. 5, 211–243.

CHAPTER NINE

Ultrastructural features of wood decay

Contents

The general anatomical features of decay as seen with the light microscope were presented in Chapter 7. This chapter reviews the ultrastructural features of wood decay and stresses the new insights they offer on the chemical nature of the decay processes.

The nature of fungal attack through an opaque, solid material such as wood makes it extremely difficult to follow the sequences of cell wall degradation. Most evidence of early fungal attack is invisible to the naked eye, hence the term incipient decay. Examination of thin sections cut from decaying wood, using the light microscope, increases our ability to study the characteristics of decayed wood and to examine the hyphae associated with this damage; however, hyphae are generally very small and their thickened cell walls have limited the detailed studies of internal function that have been performed with animals and higher plants. Wood cell wall changes associated with the early stages of fungal colonization are generally subtle and not easily detected with the light microscope. Furthermore, determining the spatial relationships between associated

Wood Microbiology.
DOI: https://doi.org/10.1016/B978-0-12-819465-2.00009-7

fungi in the wood tissues is difficult using thin sections. While modifications such as the UV microscopy, confocal microscopy and phase contrast optics have enhanced the usefulness of the light microscope, these techniques are still limited to useful maximum magnifications of about 1500X.

Electron microscopy

Many of the difficulties of studying fungal attack of wood can be overcome by the use of Transmission Electron Microscopy (TEM) or Scanning Electron microscopy (SEM). In electron microscopy, a beam of electrons is substituted for the wavelengths of light. This provides for better resolution and a larger magnification factor. For TEM, electrons passing through a thin specimen reflect the relative electron density of a given specimen, while electron scattering by thicker specimens is used to construct surface images for SEM. The principles of operation and practical protocols for routine laboratory usage of electron microscopy are available in several textbooks and laboratory manuals (Goldstein et al., 2017; DeGraef, 2003; Dawes, 1979; Postek et al., 1980; Hyat, 1981; and Robards, 1985).

Transmission electron microscopy (TEM)

TEM was first described in 1932 by Knoll and Ruska, but the resolution of early models was no better than existing light microscopes. TEM resolution exceeded that of light microscopy within three years and, by 1946, resolution to ten Angstroms (1×10^{-10} m) had been achieved. In TEM operation, a tungsten filament, heated to incandescence, emits electrons that are accelerated by high voltage. A series of condenser lenses are then used to focus the electron beam to a small zone on the specimen (3–5 μm). The lenses used in electron microscopy are electromagnetic, not the glass or quartz lenses used in light or UV microscopy. After penetrating the specimen, the beam passes through intermediate and projective lens, and finally the image is viewed on a fluorescent screen or photographic plate. TEM now permits examination of samples at magnifications exceeding 1,000,000 ×. Unlike light microscopy, where absorption of light decreases image brightness, deflection of electrons from the beam as it passes through the specimen plays a major role in image contrast.

Heavier elements are more electron dense and are often used as stains (e.g. potassium permanganate ($KMn0_4$), uranyl acetate, and lead citrate) to improve contrast in biological samples.

The successful development of TEM opened a vast new dimension (from microns to Angstroms) to microscopists studying wood and its degradation, permitting observations of microbial sequences and interactions that could only previously be imagined. Initially, techniques for embedding were imprecise and it was sometimes difficult to distinguish between real differences in structures and artifacts that occurred from the embedding process or damage from the electron beam. As researchers began to understand the system, however, artifacts were more easily detected and TEM became an extremely powerful tool for studying wood and fungal ultrastructure and interactions between wood and fungi during the decay process.

There were some disadvantages and difficulties with TEM. All materials must be dehydrated and viewed under high vacuum. The process required extensive, time-consuming preparatory steps. In some instances, researchers were primarily interested in biological events on surfaces and not internal structure. The replica technique, whereby samples were coated with thin layers of heavy metal and then treated to dissolve the original sample, permitted such studies but the technique was still time-consuming and limited to rigid materials that could withstand the required coating and stripping steps. Furthermore, these techniques were limited by the shallow depth of field of the TEM.

Scanning electron microscopy (SEM)

In SEM, an electron beam is focused to a small diameter (10–20 nm) onto the specimen surface by two condenser lens. Biological specimens are often coated with either carbon or a heavy metal (gold-palladium) to reduce the buildup of electrical charges that obscure the surface image. Two pairs of scanning coils in the second condenser lens deflect the focused beam so it can scan a square region of the sample surface. As the beam penetrates the sample, secondary electrons, emitted from the surface, are collected and counted by an electron detector located above and to one side of the specimen. The detector uses a grid with accelerating potential in front of a scintillation counter to convert electrons to light. The light is conducted through a photo-multiplier tube and visualized with a cathode ray tube. The SEM produces useful magnifications ranging

from 50 to 20,000× (more than 10 times the light microscope). The main advantages of SEM are direct observation of surfaces at great magnifications with excellent depth of field and easier sample preparation. Originally, samples had to be dry prior to examination which still required substantial preparation, but advances in SEM now permit examination of non-fixed biological specimens, further reducing the risk of artifacts.

In addition to its topographic capabilities, the electron beam generates x-rays characteristic of the various elements present in the material being observed. Energy Dispersive X-ray Analysis (EDXA) is a technique that collects these X-rays and identifies the various elements present in the specimen. EDXA is extremely useful for determining relative levels of various elements in a sample and has been used, for example, to study distribution of some preservatives in the wood.

In addition to STEM and SEM, various other techniques are increasingly used to better understand the changes occurring in wood during microbial degradation. Fourier transform infrared spectroscopy has been used to examine changes in wood chemistry during the decay process and atomic force microscopy has been used to better understand changes in surface characteristics of wood materials. Confocal microscopy allows more direct investigations but has greater depth of field than conventional light microscopy and has proven useful for studying fungal growth through wood. These non-destructive techniques allow researchers to better understand the subtle changes associated with the early stages of both biotic and abiotic degradation.

The combination of light microscopy, TEM and SEM now permits detailed examination of fungal-wood interactions and a wealth of literature related to the ultrastructure of the decay process has developed.

Some wood and fungal ultrastructural features

Several ultrastructural discoveries in wood and fungi that helped develop a deeper understanding of the decay process are reviewed briefly here (See also Chapters 3 and 6). Electron microscopes were largely unavailable commercially until the early 1950s. The electron microscope permitted unimagined explorations of wood ultrastructure, including studies of differences between the cell types of each wood species, the

presence of crystals in the wood, a warty layer on the lumen surface, pit membrane structure, and, most importantly, clear delineation of the cell wall layers (Côté, 1967; Core et al., 1976; Parham and Gray, 1984). Information on the cell wall architecture set the stage for studies of enzymatic degradation of wood and illustrated the important effects of variations of the major chemical constituents in the cell wall layers. Studies examining the cellulose microfibril (Harada and Côté, 1985) permitted precise delineation of microfibrillar orientation in each cell wall layer. This orientation explained, for example, the characteristic spiraling of soft-rot cavities as they closely followed the microfibrillar angle of the S2 cell wall layer. Subsequent research examined the relative distribution and localization of chemical components in the wood cell wall (Kerr and Goring, 1975; Parameswaran and Liese, 1982). For example, staining with potassium permanganate provided important clues concerning the distribution of lignin in the wood cell wall (Bland et al., 1971). In later studies, bromination of the lignin was used to study its distribution in various cell wall layers (Saka and Goring, 1985; Otjen and Blanchette, 1988). The distribution of hemicellulose, which is an important component in the initial phases of some fungal attack, has been studied by first treating the wood with xylanase then staining with thiocarbohydrazide and silver proteinate (Ruel and Joseleau, 1984; Joseleau and Ruel, 1985). The weak reactivity of cellulose to the PAT Ag staining technique for the hemicelluloses also provided a useful contrast for inferring cellulose locations. These techniques allowed the three major cell wall constituents to be delineated so that changes could be detected microscopically as decay developed.

Fungal ultrastructure: At the same time that wood anatomists were unlocking the secrets of the wood cell wall, mycologists were also using electron microscopy to study the ultrastructure of fungal hyphae (See Chapter 3). Most early studies were taxonomic in purpose and descriptive in nature. Some were of limited value since it was often difficult to separate fungal activity from preparative artifacts; however, these early studies provided much useful information on the structure and physiology of fungal hyphae. The ultrastructural aspects of fungi have been the subject of extensive research, for example, even by 1967, Bracker cited nearly 180 papers dealing with this subject. Mycologists exploring fungal ultrastructure were able to determine the structure of the rigid cell wall, the plasmalemma, the formation of clamp connections, nuclear behavior, and septa (Beckett et al., 1974; McLaughlin, 1982; Thielke, 1982). An atlas of fungal ultrastructure was assembled by Beckett et al. in 1974. While there

have been numerous ultrastructural studies, it is prudent to remember that thousands of fungi remain unexplored. The wide array of structural variations noted in those species already examined suggest that many new variations in fungal structure remain to be discovered. The widespread availability of electron microscopes and the general familiarity of many researchers with electron microscopic techniques resulted in a virtual explosion in the use of electron microscopy to explore changes in fungal structure and function over the course of wood degradation. The large literature on this subject makes it difficult to provide a comprehensive review. Our approach will be to briefly review those TEM and SEM studies that initially provided insights on the decay process for the three major decay types.

Wood ultrastructural changes during decay

White-rot fungi

Of the major decay types, the white-rot fungi, owing to their potential value in biopulping, bleaching and other lignin removal schemes, have been most extensively studied (Fig. 9.1). The reaction of phenolic groups in lignin with osmium tetroxide was used in early TEM studies to monitor lignin removal from the wood cell wall (Liese, 1970). These studies illustrated the gradual loss of lignin from the lumen outward associated with *Trametes* (*Polyporus*) versicolor and the rapid, widespread lignin removal noted with *Phanerochaete chyrososporium* (Highley and Murmanis, 1989; Blanchette, 1984; Blanchette et al., 1984, 1987; Ruel and Barnoud, 1985). These studies suggested a selective removal of lignin prior to cellulose attack. Further studies with cellulase-less mutants suggested that some cellulose attack was necessary for uniform lignin removal (Ruel et al., 1986; 1984; Eriksson et al., 1980; Eriksson, 1981). Seemingly widespread lignin degradation in the cell wall, often associated with the presence of electron dense particles, led some researchers to suggest that the lignin degrading enzymes were more mobile than previously believed (Ruel et al., 1981). Examination of wood decayed by white rotters also revealed that hemicellulose was removed prior to or concurrently with lignin (Hoffmann and Parameswaran, 1976; Blanchette and Abad, 1988; Blanchette et al., 1989a) (Fig. 9.2). The utilization of hemicellulose has

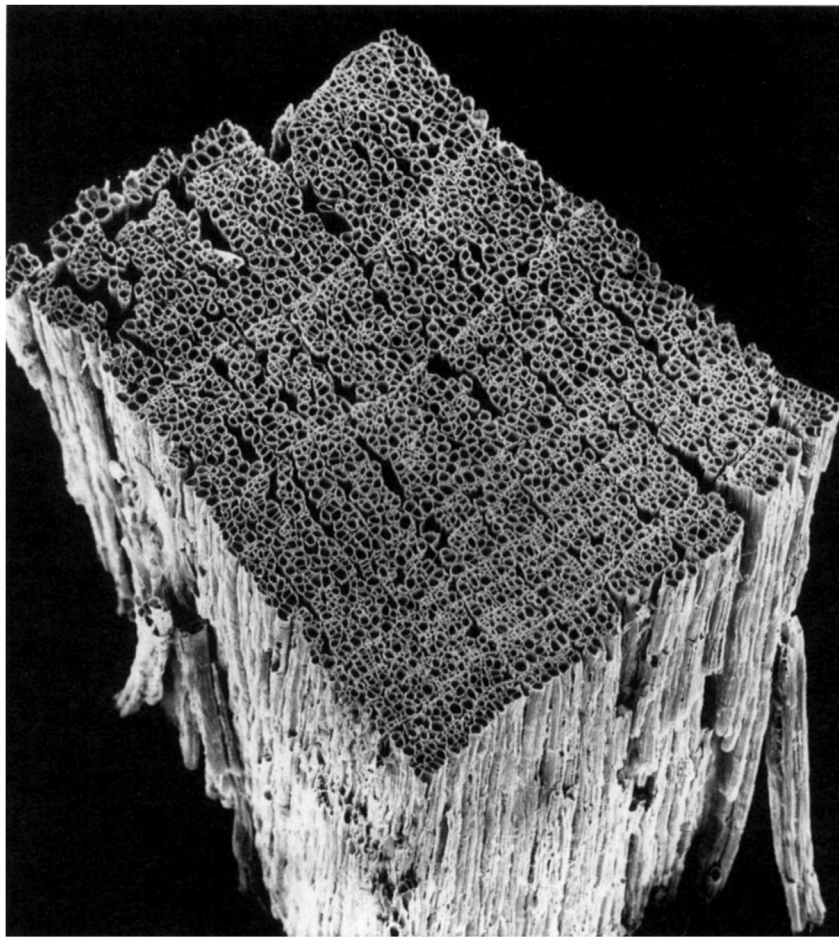


Figure 9.1 Scanning electron micrograph of *Acer* sp. colonized by a white rot fungus causing a white stringy rot with nearly complete removal of the fibers and parenchyma (30 ×). *From Blanchette, R.A., Obst, J.R., Hedges, J.I., Weliky, K., 1988. Resistance of hardwood vessels to degradation by white rot basidiomycetes. Can. J. Bot. 66, 1841–1847.*

been proposed as an important first step in the degradation of lignin (Ruel et al., 1981) and its removal has profound effects on timber properties (Winandy and Morrell, 1993). This suggests that a decay sequence *for* many white-rot fungi may be hemicellulose, lignin and then cellulose degradation.

Along with the chemical studies, a large literature has developed to describe the varying patterns by which fungi cause white rot (Adaskaveg

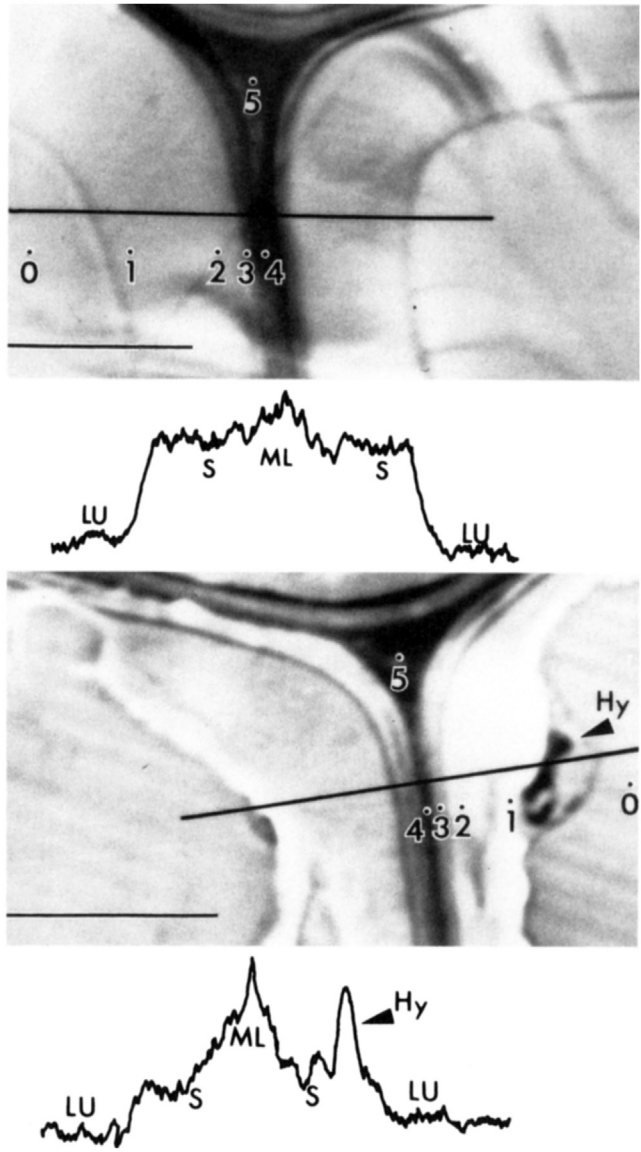


Figure 9.2 Lignin distribution in *Betula papyrifera* before and after colonization by *Trametes versicolor* as shown by EDXA-STEM analysis following bromination of the lignin residues where Lu = lumen, S = secondary cell wall, ML = middle lamella, and Hy = hyphae. *From Blanchette, R.A., Otjen, L., Carlson, M.C., 1987. Lignin distribution in cell walls of birch wood decayed by white rot basidiomycetes. Phytopathology 77, 684–690.*

and Gilbertson, 1986; Blanchette, 1980; Blanchette and Reid, 1986; Blanchette et al., 1988; Dill and Kraepelin, 1986; Murmanis et al., 1984; Otjen and Blanchette, 1986, 1987; Reid, 1985). Some of these studies indicated that extensive cell wall degradation occurred away from the vicinity of fungal growth. Earlier light microscopic and chemical studies suggested that white-rot fungi caused damage nearer to the zone of fungal growth, while brown-rot fungi produced enzymes that diffused for greater distances through the wood (Cowling, 1961). Ultrastructural studies now seem to negate these observations for white-rot fungi and suggest that cell-wall constituents are affected to a significant degree at the early stages of colonization by both brown-and white-rot fungi.

Studies of fungal hyphae associated with white-rot fungi have shown the presence of an extensive hyphal sheath around the hyphal tip (Highley and Murmanis, 1984; Foisner et al., 1985a,b) and the presence of osmophilic particles (Messner and Stachelberger, 1984). The sheath structure may retain fungal enzymes near the hyphae, prevent desiccation, and provide a diffusion channel for movement of wood decomposition products into the fungal cell. The sheath also could provide a contact point between the wood and the fungi, thereby serving as a concentrating point for enzymatic attack to minimize dilution or an external point of assembly for fungal enzymes that are too large to be excreted directly through the hyphal wall.

The effects of fungal enzymes on the wood cell wall have also come under increasing scrutiny as biochemical tools were developed for identifying and quantifying fungal enzymes (Forney et al., 1982). A number of studies suggest that fungal enzymes that attack lignin, cellulose, or hemicellulose are too large to diffuse directly into sound wood (Srebotnik et al., 1988a,b). Thus, small, non-enzymatic entities were proposed that initiate wood cell wall decomposition and expand the capillary system. The production of free radicals has been proposed as one mechanism for this process. Once the wood structure becomes modified and accessible, fungal enzymes can then begin to directly interact with the various cell wall components.

Perhaps the most dramatic influence of ultrastructural studies on wood degradation has been the development of immunocytological techniques. Formerly, TEM or SEM observations were limited to describing changes in the wood structure or fungal hyphae sometimes aided by staining with specific chemical reagents, but it was generally not possible to determine the biochemical nature of the changes at specific sites in the wood. The

development of antibodies with high specificity for fungal enzymes, coupled with the development of specific chemical staining or colloidal gold coupled antibody techniques permitted detection of enzyme activity within specific areas of the fungus or the wood cell (Daniel et al., 1989a, b; Blanchette et al., 1989a,b; Garcia et al., 1987; Srebotnik and Messner, 1988; Srebotnik et al., 1988b; Blanchette et al., 1989a,b). These techniques were used to identify lignin peroxidase in the fungal cell and have shown that this enzyme is capable of moving into decayed wood, but not sound wood, supporting the existence of non-enzymatic decay factors discussed above (Fig. 9.3). Despite the high sensitivity of immunocytological techniques, caution must still be exercised when preparing materials for these techniques. For example, the fixation process can alter immunoreactivity, dramatically influencing the results (Srebotnik et al., 1988b).

Ultimately, immunocytological studies helped delineate the pathways for enzyme synthesis and secretion, providing important clues for the selection of control chemicals.

Brown rots

The ultrastructure of brown rotted wood has received less attention, but similar studies to delineate the nature of brown rot decay have important applications in the control of many important wood products fungi. Such information may also prove useful for altering wood or modifying cellulose to make it more digestible for ruminant animals. Studies show some cellulose dissolution adjacent to the hyphal tip (Fig. 9.4) as well as the depletion of cellulose in the secondary cell wall at great distances from the sites of hyphal growth. This latter loss is reflected in large decreases in wood strength at very early stages of fungal colonization. Early TEM studies suggested that enzymes diffused into the cell wall from the margins of bore holes and not from hyphae in the lumen (Chou and Levi, 1971). They also noted the presence of a gelatinous sheath around the hyphae. The hyphal sheath has been detected in other studies (Palmer et al., 1983a,b, 1984), but its function also remains undefined (Fig. 9.5). TEM studies showed that the hyphal sheath contained low molecular weight β-1,3 glucans (Green et al., 1989). Other studies suggested that the sheath contained contents from autolyzed hyphae or provided a site for translocation of enzymes (Highley et al., 1983a,b; Palmer et al., 1983a,b). The sheath may also function to retain and concentrate enzymes released from the hyphae in a manner similar to that described for the white rots.

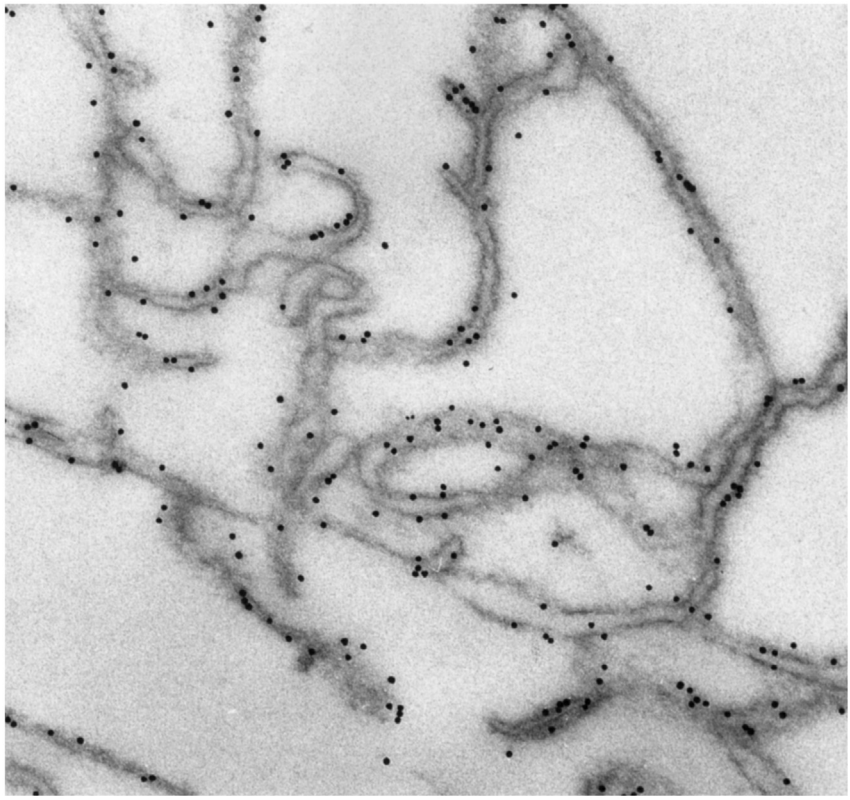


Figure 9.3 TEM of a thin section through pine sapwood colonized by *Phanerochaete chrysoporium* showing localization of colloidal gold-coupled lignin peroxidase within the fungal associated extracellular slime and membranous structure within the lumina of degraded wood fibers. *From Daniel, G.F., Nilsson, T., Pettersson, B., 1989a. Intra-and extracellular localization of lignin peroxidase during the degradation of solid wood and wood fragments by* Phanerochaete chrysosporium *by using transmission electron microscopy and immuno-gold labeling. Appl. Environ. Microbiol. 55 (4), 871–881.*

Early in the study of brown rotted wood, researchers noted that large quantities of modified lignin with a reduced methoxyl content and increased solubility remained in the wood cell walls, which were largely devoid of cellulose and hemicelluloses. These results suggested that brown-rot fungi lacked significant lignin degrading capability, while rapidly utilizing cellulose. However, this premise was confounded by the inability to grow brown-rot fungi on pure cellulose, either in cotton fibers (Highley et al., 1983b) or in wood delignified by white rot fungi

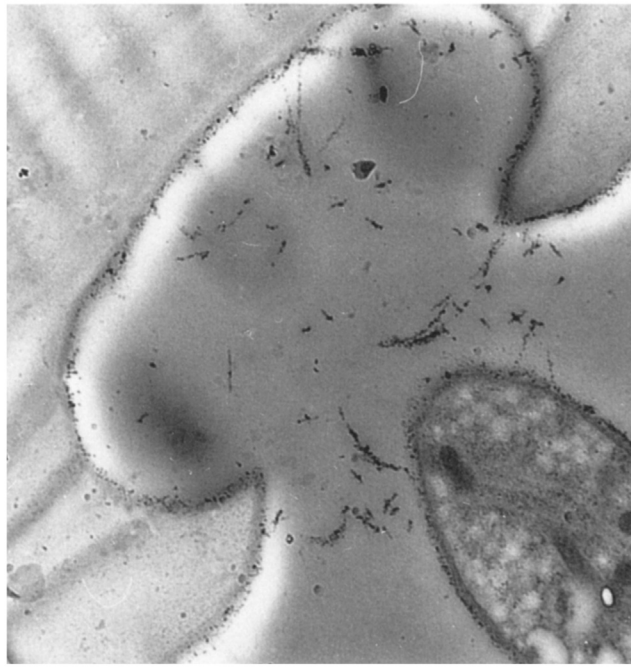


Figure 9.4 Dissolution of a tracheid wall of eastern white pine associated with the hyphal tip of *Rhodonia placenta* showing a lysis zone around the hyphal tip (TEM micrograph 9225 ×) from unpublished data from Jutte and Zabel (1974).

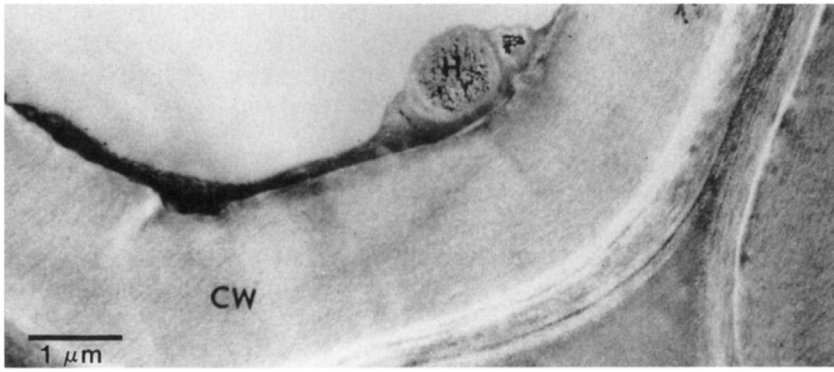


Figure 9.5 TEM of *Ganoderma applanatum* hyphae showing hyphal sheath attached to wood cell wall of *Tsuga heterophylla* (× 19,800) (Murmanis et al.,1985).

(Blanchette, 1983). These results suggested that some lignin or other growth factor must be present for these fungi to initiate the cellulose attack. TEM studies have suggested that the cellulase molecules are too

large to diffuse into sound wood and support the theory that a non-enzymatic factor also initiates brown-rot decay (Srebotnik and Messner, 1988). Furthermore, evidence suggests that brown rot fungi profoundly alter, but do not utilize lignin as they modify the wood to create access to the carbohydrate fraction of the wood.

The nature of brown rot enzymes has also become the subject of extensive research. Several studies have detected electron dense particles in wood attacked by a variety of brown-rot fungi or by cellulase preparations obtained from *Trichoderma reesi* (Murmanis et al.,1987, Messner and Stachelberger,1982,1985). The electron dense particles have been suggested to be lignin decomposition products (Srebotnik and Messner, 1988) and these older results align with recent observations that brown rot fungi extensively degrade lignin, but that it later repolymerizes, leaving a demethoxylated residue.

The effects of brown-rot fungi on lignin are becoming better understood as researchers more fully explore the changes induced by brown rot attack as well as the genetic make-up of these fungi. The presence of a peroxide based lignin degradation system has been proposed for the white-rot fungi (see Chapter 8), but no peroxide was detected in cultures of *Rhodonia placenta* (Highley and Murmanis, 1985, 1986; Murmanis et al., 1988a,b). However, weak reactivity to *Phanerochaete chrysosporium* lignin peroxidase has been reported at the hyphal tips of *Fomitopsis pinicola* (Blanchette et al., 1989a) and peroxide detected in *Antrodia* (*Poria) carbonica*. This suggests that brown-rot fungi have a more profound effect on lignin that is masked by the fact that this dissolution appears to be followed by some reassembly leaving a modified, but largely intact lignin.

Brown rot fungal attack of hemicellulose remains the most poorly understood process in the degradation of wood by these fungi, despite the premise that some hemicellulose degradation is essential for and may precede the attack of other wood components. Extensive hemicellulose degradation has been noted with *F. pinicola* using a silver staining technique, with higher levels of attack in the middle lamella (Blanchette and Abad,1988). However, xylanase, and endoglucanase activity were not detected in wood colonized by this fungus (Blanchette et al.,1989b). These studies highlight the difficulty of using electron microscopy for chemical and immunocytological analyzes. It is readily apparent that our knowledge of the enzyme systems responsible for brown rot degradation has many gaps. Furthermore, even this knowledge is limited to studies on a few species that are easily manipulated under laboratory conditions (e.g.

Rhodonia placenta). Yet brown-rot fungi, because of their prevalence in many wood products and their drastic effects on wood strength reduction, are an important group that merits more intensive study. Elucidating the initial pathways of fungal degradation may provide improved strategies for preventing or controlling colonization. Conversely, an improved knowledge of the enzyme systems could be exploited to produce bioenergy, chemical feedstocks or to improve the digestibility of wood for animal feeds.

Soft rot

The diamond shaped cavities formed within the S-2 cell wall layer by Type 1 soft rot fungi have long intrigued wood anatomists who have used light and electron microscopy to explore the unique morphology of these cavities (Figs. 9.6, 9.7). Most studies have been descriptive in nature,

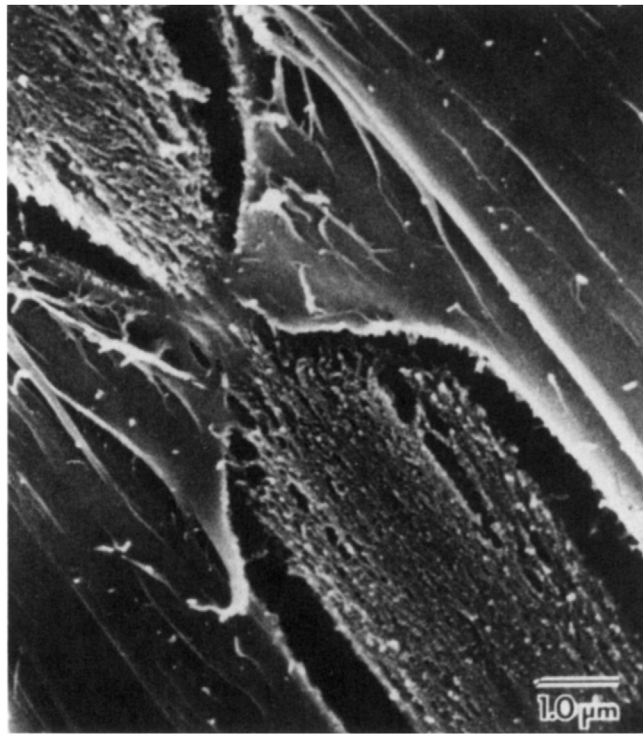


Figure 9.6 SEM micrograph of Type 1 soft rot damage in a creosote-treated southern pine pole showing an enlarged section of a hypha in two contiguous cavities and the typical conical zones. *Zabel et al. (1985) and permission of the Society of Wood Science and Technology.*

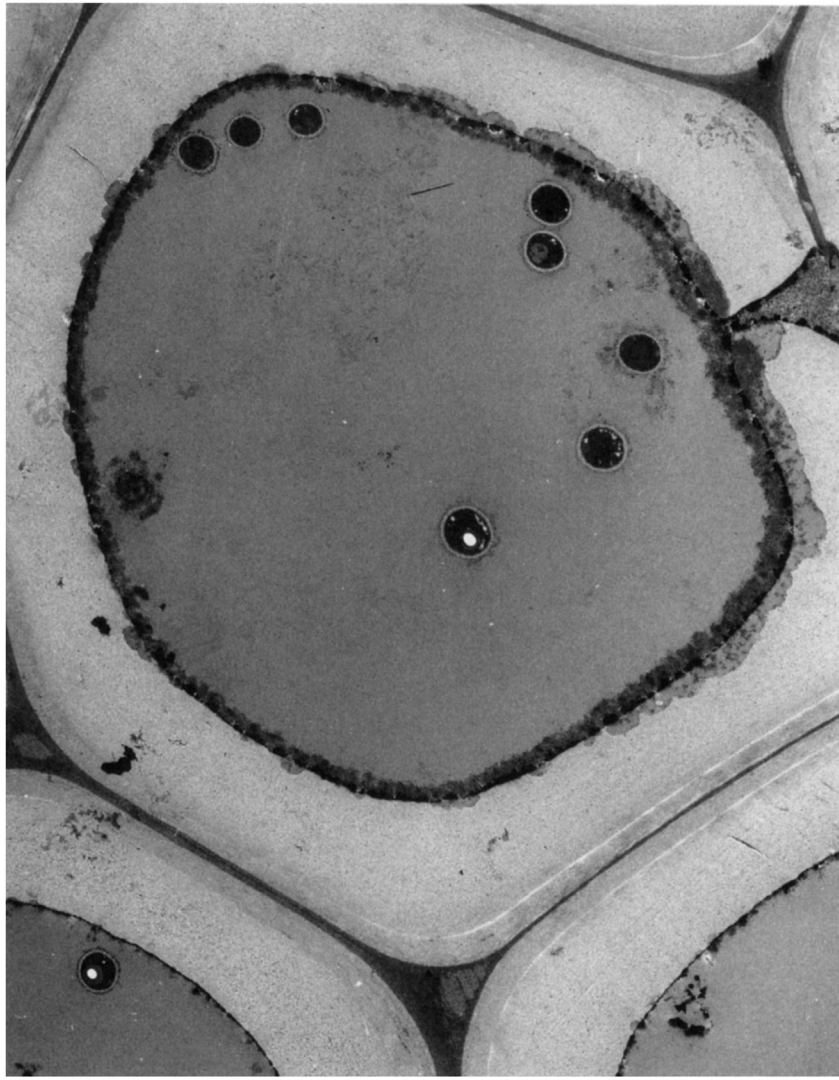


Figure 9.7 TEM of CCA treated *Betula verrucosa* after a one-month exposure to *Phialophora mutabilis* showing erosion of the S-2 cell wall layer away from fungal hyphae as well as CCA residue along the S-3 cell wall layer. *From Daniel, G.F., Nilsson, T., 1989. Interactions between soft rot fungi and CCA preservatives in* Betula verrucosa. *J. Inst. Wood Sci. 11 (5), 162–171.*

owing to the relative scarcity of research on the enzyme systems of the soft rot fungi (Mouzouras, 1989; Unligil and Chafe, 1974; Daniel and Nilsson, 1988, 1989; Nilsson et al., 1989; Jutte and Zabel, 1974).

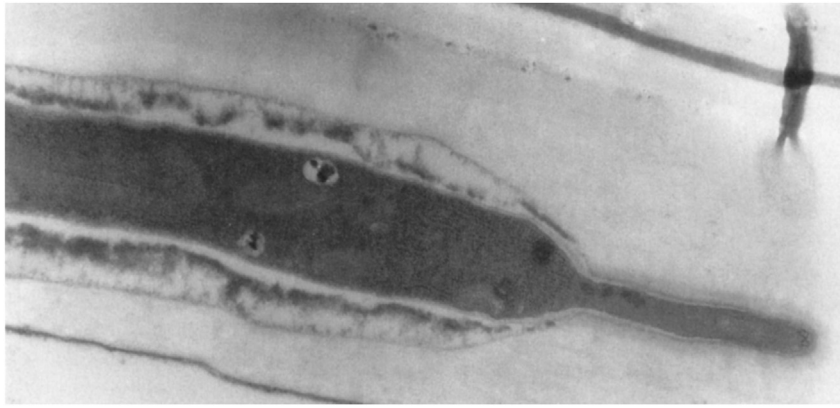


Figure 9.8 TEM of a Type 1 soft rot cavity produced by *Phialophora hoffmannii* in *Betula pendula* showing a diamond-shaped cavity with fine proboscis hyphae and halo surrounding the hyphal tip and the tip of the hyphal cavity prior to. *From Hale, M.D., Eaton, R.A., 1981. Soft Rot Ultrastructure. The International Research Group on Wood Preservation Document No IRG/WP/1138. Stockholm, Sweden.*

For example, Crossley and Levy (1977) noted a mucilaginous sheath that was extruded by the cavity hyphae. Earlier, Unligil and Chafe (1974) had noted that T-branch formation by soft-rot fungi involved enzymatic activity. Cavity initiation was reported to occur by dissolution, not physical pressure and the wood and fungus in these cavities were connected by fine strands of amorphous material (Hale and Eaton, 1981, 1985a,b). Numerous ribosomes were found in the hyphal tip, suggesting active enzyme synthesis. An electron opaque halo was detected around the hyphal tips that was proposed to contain an exocellulase (Fig. 9.8). These cavities also appeared to contain lignin residues and extracellular polysaccharides.

Electron microscopy has also been used to understand the selectivity of soft rot attack on certain treated wood species. Ryan and Drysdale (1988) found that the relationship between CCA loading in the S-2 cell wall layer and soft rot attack varied with wood species. Daniel and Nilsson (1988, 1989) examined wood attacked by *Phialophora mutabilis* and reported that electron dense particles in the soft rot cavities contained high levels of CCA and phenolics.

Bacterial erosion

While basidiomycetes cause more dramatic wood changes, the nature of bacterial attack has also been studied at the ultrastructural level. Bacteria

were not considered to be capable of wood damage until the late 1950, when studies on sinker logs (from log storage ponds) showed that bacterial attack of the pit membranes was the cause of excessive water absorption (Ellwood and Ecklund, 1959). SEM examination of wood infected with bacteria clearly showed severe pit membrane damage (Levy,1975; Liese and Greaves,1975; Greaves,1968,1969; Schmidt et al., 1987). Further examination of bacterially infected wood from marine environments has shown that bacteria cause damage similar to soft-rot fungi, although the rates of attack were extremely slow. Initially, bacteria were shown to form erosion channels on the lumen surface of the cell wall in a manner similar to that found with Type 2 soft rot fungi on hardwoods (Holt, et al., 1979; Greaves, 1969; Holt, 1983). Bacteria have also been found to produce tunnels similar to Type 1 soft rot cavities in the wood, particularly under conditions where excessive moisture and nutrients are present (Nilsson and Singh, 1984). Cavitation and tunneling produce numerous cavities in the S-2 cell wall layer as well as degradation of the primary cell wall and middle lamella. These studies indicate that some bacteria have the capability to degrade wood components under special conditions.

While the economic importance of bacteria in wood degradation is somewhat limited, these organisms have rapid life cycles and are easily manipulated in the laboratory. The ability of some species to degrade all wood components may make them useful for further elucidating mechanisms of decay.

Microbial interactions in decay

TEM and SEM can provide important clues about the decay process and the nature of microbial interactions (see Chapters 8 and 11). Ultrastructure may be particularly useful for exploring the interactions that occur when organisms compete for the same substrate and may provide important clues concerning the outcomes of various microbial combinations. Observations of hyphal coiling, cell lysis, and the production of toxic metabolites may all be visualized using electron microscopy. Electron microscopy may be especially useful for studying microbial interactions to detect potential biological control, particularly when immunocytological techniques are used to localize various enzymatic components of the interacting organisms in natural systems.

While SEM has been used to study spatial interactions among fungi in wood, there are relatively few TEM studies of decay in mixed cultures.

Murmanis et al. (1989) examined *Trichoderma harzianum*, *Trichoderma polysporum*, and several basidiomycetes in mixed cultures and found evidence of cell disruption in the basidiomycete. More detailed studies, in combination with biochemical tests, may provide important clues concerning the mechanisms by which biological control agents function. In the laboratory, mixed cultures of bacteria, yeasts and basidiomycetes associated with conifers have been shown to both stimulate mycelial growth and increase decay weight losses (Blanchette and Shaw, 1978).

Detection and quantification of wood preservatives

In addition to its usefulness for studying the ultrastructure of decay, SEM has also been used to determine distribution of various preservatives in the wood cell wall and the subsequent effects of these preservatives on fungal colonization and wood degradation. Resch and Arganbright (1971) evaluated EDXA for determining the distribution of pentachlorophenol in the wood cell wall and reported some penetration into the secondary cell wall layers when liquefied petroleum gas was used as the carrier, as did Wilcox and Parameswaran (1974) and Wilcox et al. (1974). Similar studies have been performed on fungi in wood treated with CCA (Chou et al., 1973; Daniel and Nilsson, 1989; Ryan and Drysdale, 1988; Ryan, 1986). More recently, these techniques have been used to examine copper particle distribution in treated wood in order to better understand performance (Evans et al., 2012; Feng et al., 2019).

Summary

1. The resolution of the TEM permitted researchers to view the internal structure of the fungal-wood interactions at the molecular level. However, the process of dehydrating, embedding, and ultrathin sectioning requires extensive and time-consuming procedures and results in a static two-dimensional image. In those instances where researchers were primarily interested in biological events on surfaces and not internal structures, the replica technique was utilized. This technique produces a *3-D* view of the surface. As a result of its great resolving power, the TEM is an extremely powerful tool for studying wood

and fungal ultrastructure and the interactions between wood and fungi during the decay process.

2. Electron microscopic studies of wood have provided a wealth of new and confirming information about the layered nature of the cell wall and microfibrillar orientations that provide the locations of cellulose, hemicelluloses, and lignin in the cell wall when used in conjunction with various staining techniques. This information has set the stage for an improved level of understanding of the decay process.
3. Ultrastructural studies of white rots suggest that hemicellulose attack precedes lignin decomposition and that cellulose decomposition follows early lignin attack. Cell wall decomposition occurs both in the vicinity of some hyphae and at considerable distances away, inferring the presence of diffusible enzymes or short-lived non-enzymatic factors. Staining tests specific for lignin suggest that the ligninase enzymes do not penetrate sound wood, supporting the hypothesis of a non-enzymatic, presumably oxidative factor that modifies sound wood prior to enzymatic entry and attack.
4. Ultrastructural studies of brown rotted wood indicate substantial decomposition of cellulose at considerable distances from the zone of hyphal growth. This early cellulose loss is very important in the brown rots since it is associated with drastic strength reductions at very early decay stages. TEM studies suggest that cellulases are too large to enter sound wood until it has been modified by a non-enzymatic decay agent. They also suggest that some brown rots, while they do not metabolize lignin, may substantially modify it and may retain low levels of lignin peroxidase activity. This implies that the decay mechanisms in white and brown rotters may not be as different as generally believed.
5. Ultrastructural studies indicate the presence of a "gelatinous" sheath around the hyphae of brown rotters and the hyphal tips in white rotters whose functions are undefined. They may play some role in enzyme or non-enzymatic factor secretion and delivery to the cell wall surfaces.
6. Ultrastructural studies of soft rotted wood (Type 1) have been largely descriptive in nature. TEM studies indicate that T-branches and cavities form enzymatically and that the debris within some cavities is residual lignin.
7. Ultrastructural studies have confirmed that, under some conditions, bacteria erode cell wall surfaces in a manner similar to Type 2 soft

rotters in hardwoods. Some bacteria can also tunnel in the cell wall and form cavities.

8. Electron microscopy is a useful way to locate antagonists in wood microorganism interactions with biological control potential and also to detect and quantify some of the metallic preservatives in wood.

References

Adaskaveg, J.E., Gilbertson, R.L., 1986. In vitro decay studies of selective delignification and simultaneous decay by the white rot fungi *Ganoderma lucidum* and *G. tsugae*. Can. J. Bot. 64, 1611–1619.

Beckett, A., Heath, I.B., McLaughlin, D.J., 1974. An Atlas of Fungal Ultrastructure. Longman Group Limited, London, p. 221.

Blanchette, R.A., 1980. Wood decomposition by *Phellinus* (*Fomes*) *pini:* a scanning electron microscopy study. Can. J. Bot. 58, 1496–1503.

Blanchette, R.A., 1983. An unusual decay pattern in brown-rotted wood. Mycologia 75 (3), 552–556.

Blanchette, R.A., 1984. Screening wood decayed by white rot fungi for preferential lignin degradation. Appl. Environ. Microbiol. 48 (3), 647–653.

Blanchette, R.A., Shaw, C.G., 1978. Associations among bacteria, yeasts, and basidiomycetes during wood decay. Phytopathology 68, 631–637.

Blanchette, R.A., Reid, I.D., 1986. Ultrastructural aspects of wood delignification by *Phlebia* (*Merulius) tremellosus*. Appl. Environ. Microbiol. 52 (2), 239–245.

Blanchette, R.A., Abad, A.R., 1988. Ultrastructural localization of hemicellulose in birch wood (*Betula papyrifera*) decayed by brown and white rot fungi. Holzforschung 42 (6), 393–398.

Blanchette, R.A., Otjen, L., Carlson, M.C., 1987. Lignin distribution in cell walls of birch wood decayed by white rot basidiomycetes. Phytopathology 77, 684–690.

Blanchette, R.A., Obst, J.R., Hedges, J.I., Weliky, K., 1988. Resistance of hardwood vessels to degradation by white rot basidiomycetes. Can. J. Bot. 66, 1841–1847.

Blanchette, R.A., Abad, A.R., Farrell, R.L., Leathers, T.D., 1989a. Detection of lignin peroxidase and xylanase by immunocytochemical labeling in wood decayed by basidiomycetes. Appl. Environ. Microbiol. 55 (6), 1457–1465.

Blanchette, R.A., Abad, A.R., Cease, K.R., Lovrien, R.E., Leathers, T.D., 1989b. Colloidal gold cytochemistry of Endo-l,4-β-Xylanase: ultrastructure of sound and decayed birch wood. Appl. Environ. Microbiol. 55 (9), 2293–2301.

Bland, D.E., Foster, R.C., Logan, A.F., 1971. The mechanism of permanganate and osmium tetroxide fixation and the distribution of lignin in the cell wall of *Pinus radiata*. Holzforschung 25 (5), 137–143.

Bracker, C.E., 1967. Ultrastructure of fungi. Annu. Rev. Phytopathol. 5, 343–374.

Chou, C.K., Levi, M.P., 1971. An electron microscopical study of the penetration and decomposition of tracheid cell walls of *Pinus sylvestris* by *Poria vaillantii*. Holzforschung 25 (4), 107–112.

Chou, C.K., Preston, R.D., Levi, M.P., 1973. Fungitoxic action of copper-chromium arsenate wood preservatives. Phytopathology 64, 335–341.

Core, H.A., Côté Jr., W.A., Day, A.C., 1976. Wood Structure and Identification. Syracuse University Press, Syracuse, N.Y, 166 p.

Côté Jr., W.A., 1967. Wood Ultrastructure-an Atlas of Electron Micrographs. University of Washington Press, Seattle, WA, p. 64.

Cowling, E.B., 1961. Comparative Biochemistry of the Decay of Sweetgum Sapwood by White-rot and Brown-rot Fungi. U.S. Dept. Agr. Tech. Bull. 1258, p. 79.

Crossley, A., Levy, J.F., 1977. Proboscis hyphae in soft rot cavity formation. J. Inst. Wood Sci. 7, 30–33.
Daniel, G.F., Nilsson, T., 1988. Interactions Between Soft Rot Fungi and CCA Preservatives in *Betula verrucosa*. The International Research Group on Wood Preservation, Document No. IRG/WP/1367, Stockholm, Sweden.
Daniel, G.F., Nilsson, T., 1989. Interactions between soft rot fungi and CCA preservatives in *Betula verrucosa*. J. Inst. Wood Sci. 11 (5), 162–171.
Daniel, G.F., Nilsson, T., Pettersson, B., 1989a. Intra-and extracellular localization of lignin peroxidase during the degradation of solid wood and wood fragments by *Phanerochaete chrysosporium* by using transmission electron microscopy and immunogold labeling. Appl. Environ. Microbiol. 55 (4), 871–881.
Daniel, G., Pettersson, F., Nilsson, T., Volc, J., 1989b. Cytoplasmic and Extracellular Localization of Manganese II Dependant Peroxidase(s) in White Rot Fungi During Degradation of Wood Materials. The International Research Group on Wood Preservation, Document No. IRG/WP/1416, Stockholm, Sweden.
Dawes, C.J., 1979. Biological Techniques for Transmission and Scanning Electron Microscopy. Ladd Research Industries, Inc., Burlington, Vt., p. 303.
De Graef, M., 2003. Introduction to Transmission Electron Microscopy. Cambridge University Press, Cambridge, UK.
Dill, I., Kraepelin, G., 1986. Palo Podrido: model for extensive delignification of wood by *Ganoderma applanatum*. Appl. Environ. Microbiol. 52 (6), 1305–1312.
Ellwood, L.L., Ecklund, L.A., 1959. Bacterial attack of pine logs in pond storage. For. Prod. J. 9, 283–292.
Eriksson, K.-L., 1981. Fungal degradation of wood components. Pure Appl. Chem. 53, 33–43.
Eriksson, K.-E., Grunewald, A., Nilsson, T., Vallander, L., 1980. A scanning electron microscopy study of the growth and attack on wood by three white rot fungi and their cellulase-less mutants. Holzforschung 34 (6), 207–213.
Evans, P.D., Limaye, A., Averdunk, H., Turner, M., Senden, T.J., Knackstedt, M.A., 2012. Use of Computer X-ray Tomography to Visualize Copper in Preservative Treated Wood. International Research Group on Wood Protection Document No. IRG/WP/12-20488, Stockholm, Sweden.
Feng, D., Turner, M.L., Limaye, A., Knackstedt, M.A., Evans, P.D., 2019. Accumulation of Copper in Parenchyma Cells in Southern Pine Treated With Micronized and Amine Copper. International Research Group on Wood Protection Document No. IRG/WP/12-20488, Stockholm, Sweden.
Foisner, R., Messner, K., Stachelberger, H., Rohr, M., 1985a. Wood decay by basidiomycetes: extracellular tripartite membranous structures. Trans. Br. Mycol. Soc. 85 (2), 267–278.
Foisner, R., Messner, K., Stachelberger, H., Rohr, M., 1985b. Isolation and characterization of extracellular three-lamellar structures of *Sporotrichum pulverulentum*. J. Ultrastruct. Res. 92, 32–46.
Forney, L.J., Reddy, C.A., Pankratz, H.S., 1982. Ultrastructural localization of hydrogen peroxide production in ligninolytic *Phanerochaete chrysosporium* cells. Appl. Environ. Microbiol. 44 (3), 732–736.
Garcia, S., Latge, J.P., Prevost, M.C., Leisola, M., 1987. Wood degradation by white rot fungi: cytochemical studies using lignin peroxidase immunoglobulin-gold complexes. Appl. Environ. Microbiol. 53 (10), 2384–2387.
Goldstein, J.I., Newbury, D.E., Michael, J.R., Ritchie, N.W.M., Scott, J.H.J., Joy, D.C., 2017. Scanning Electron Microscopy and X-ray Microanalysis. Springer, New York.
Greaves, H., 1968. Occurrence of bacterial decay in copper-chrome-arsenic treated wood. J. Appl. Microbiol. 16, 1599–1601.

Greaves, H., 1969. Micromorphology of bacterial attack of wood. Wood Sci. Technol. 3, 150–166.
Green, F., Larsen, M.J., Murmanis, L.L., Highley, T.L., 1989. Proposed Model for the Penetration and Decay of Wood by the Hyphal Sheath of the Brown Rot Fungus *Postia placenta*. The International Research Group on Wood Preservation, Document No. IRG/WP/1391, Stockholm, Sweden.
Hale, M.D., Eaton, R.A., 1981. Soft Rot Ultrastructure. The International Research Group on Wood Preservation Document No IRG/WP/1138, Stockholm, Sweden.
Hale, M.D., Eaton, R.A., 1985a. The ultrastructure of soft rot fungi. I. Fine hyphae in wood cell walls. Mycologia 77 (3), 447–463.
Hale, M.D., Eaton, R.A., 1985b. The ultrastructure of soft rot fungi. II. Cavity forming hyphae in the wood cell wall. Mycologia 77 (4), 594–605.
Harada, H., Cote Jr., W.A., 1985. Structure of wood. In: Higuchi, T. (Ed.), Biosynthesis and Biodegradation of Wood Components. Academic Press, New York, pp. 1–42.
Highley, T.L., Murmanis, L.L., 1984. Ultrastructural aspects of cellulose decomposition by white-rot fungi. Holzforschung 38 (2), 73–78.
Highley, T.L., Murmanis, L.L., 1985. Involvement of Hydrogen Peroxide in Wood Decay by Brown-rot and White-rot Fungi. The International Research Group on Wood Preservation, Document No. IRG/WP/1256, Stockholm, Sweden.
Highley, T.L., Murmanis, L.L., 1986. Determination of hydrogen pyroxide production in *Coriolus versicolor* and *Poria placenta* during wood degradation. Mater. Org. 20 (4), 241–252.
Highley, T.L., Murmanis, L.L., 1987. Micromorphology of degradation in western hemlock and sweetgum by the white-rot fungus *Coriolus versicolor*. Holzforschung 41 (2), 67–71.
Highley, T.L., Murmanis, L.L., Palmer, J.G., 1983a. Electron microscopy of cellulose decomposition by brown rot fungi. Holzforschung 37 (6), 271–277.
Highley, T.L., Palmer, J.G., Murmanis, L.L., 1983b. Decomposition of cellulose by *Poria placenta:* light and electron study. Holzforschung 37 (4), 179–184.
Hoffmann, P., Parameswaran, N., 1976. On the ultrastructural localization of hemicelluloses within delignified tracheids of spruce. Holzforschung 30 (2), 62–70.
Holt, D.M., 1983. Bacterial degradation of lignified wood cell walls in aerobic habitats, decay patterns and mechanisms proposed to account for their formation. J. Inst. Wood Sci. 9, 212–223.
Holt, D.M., Gareth-Jones, E.B., Furtado, S.E.J., 1979. Bacterial breakdown of wood in aquatic habitats. Record, British Wood Preserver's Association Annual Convention. pp. 13–22.
Hyat, M.A., 1981. second ed. Principles and Techniques of Electron Microscopy Biological Applications, vol. 1. University Park Press, Baltimore, p. 522.
Joseleau, J.P., Ruel, K., 1985. Anew cytochemical method for ultrastructural localization of polysaccharides. Biol. Cell 53, 61–66.
Jutte, S.M., Zabel, R.A., 1974. Initial wood decay stages as revealed by scanning electron microscopy. Scanning Electron. Microsc. (Part. II) 445–452.
Kerr, A.J., Goring, D.A.I., 1975. The ultrastructural arrangement of the wood cell wall. Cellulose Chem. Technol. 9, 563–573.
Levy, J.F., 1975. Bacteria associated with wood in ground contact. In: Liese, W. (Ed.), Biological Transformation of Wood by Microorganisms. Springer-Verlag, Inc, New York, pp. 64–73.
Liese, W., 1970. Ultrastructural aspects of woody tissue disintegration. Annu. Rev. Phytopathol. 8, 231–257.
Liese, W., Greaves, H., 1975. Micromorphological of bacterial attack. In: Liese, W. (Ed.), Biological Transformation of Wood by Microorganisms. Springer-Verlag, New York, pp. 74–88.

McLaughlin, D.J., 1982. Ultrastructure and cytochemistry of basidia and basidiospore development. In: Wells, K., Wells, L.K. (Eds.), Basidium and Basidiocarp Evolution, Cytology, Function and Development. Springer-Verlag, New York, pp. 37–74.
Nilsson, T., Singh, A., 1984. Cavitation Bacteria. International Research Group on Wood Preservation Document No. IRG/WP/1235, Stockholm, Sweden.
Messner, K., Stachelberger, H., 1982. Extracellular Osmiophilic Particles in Connection With Brown-Rot. The International Research Group on Wood Preservation, Document No. IRG/WP/1157, Stockholm, Sweden.
Messner, K., Stachelberger, H., 1984. Transmission electron microscope observations of white rot caused by *Trametes hirsuta* with respect to osmiophilic particles. Trans. Br. Mycol. Soc. 83 (2), 217–221.
Messner, K., Stachelberger, H., 1985. Transmission electron microscope observations of brown rot caused by *Fomitopsis pinicola* with respect to osmiophilic particles. Trans. Sr. Mycol. Soc. 83 (1), 131–137.
Mouzouras, R., 1989. Soft rot decay of wood by marine microfungi. J. Inst. Wood Sci. 11 (5), 193–201.
Murmanis, L., Highley, T.L., Palmer, J.G., 1984. An electron microscopy study of western hemlock degradation by the white-rot fungus *Ganoderma applanatum*. Holzforschung 38 (1), 11–18.
Murmanis, L., Palmer, J.G., Highley, T.L., 1985. Electron-dense particles in wood decayed by *Ganoderma applanatum*. Wood Sci. Technol. 19, 313–321.
Murmanis, L., Highley, T.L., Palmer, J.G., 1987. Cytochemical localization of cellulases in decayed and non-decayed wood. Wood Sci. Technol. 21, 101–109.
Murmanis, L., Highley, T.L., Palmer, J.G., 1988a. The action of isolated brown-rot cell-free culture filtrate, H_2O_2-Fe^{++}, and the combination of both on wood. Wood Sci. Technol. 22, 59–66.
Murmanis, L., Highley, T.L., Ricard, J., 1988b. Hyphal interaction of *Trichoderma harzianum* and *Trichoderma polysporum* with wood decay fungi. Mater. Org. 23, 271–279.
Nilsson, T., Daniel, G., Kirk, T.K., Obst, J.R., 1989. Chemistry and microscopy of wood decay by some higher ascomycetes. Holzforschung 43 (1), 11–18.
Otjen, L., Blanchette, R.A., 1986. A discussion of microstructural changes in wood during decomposition by white rot basidiomycetes. Can. J. Bot. 64, 905–911.
Otjen, L., Blanchette, R., 1987. Assessment of 30 white rot basidiomycetes for selective lignin degradation. Holzforschung 41 (6), 343–349.
Otjen, L., Blanchette, R.A., 1988. Lignin distribution in wood delignified by white-rot fungi: x-ray microanalysis of decayed wood treated with bromine. Holzforschung 42 (5), 281–288.
Palmer, J.G., Murmanis, L., Highley, T.L., 1983a. Visualization of hyphal sheaths in wood-decay hymenomycetes. I. Brown-rotters. Mycologia 75 (6), 995–1004.
Palmer, J.G., Murmanis, L., Highley, T.L., 1983b. Visualization of hyphal sheaths in wood-decay hymenomycetes. I. White-rotters. Mycologia 75 (6), 1005–1010.
Palmer, J.G., Murmanis, L., Highley, T.L., 1984. Observations of wall-less protoplasm in white-and brown-rot fungi. Mater. Org. 19 (1), 39–48.
Parameswaran, N., Liese, W., 1982. Ultrastructural localization of wall components in wood cells. Roh-und Werkst. 40, 145–155.
Parham, R.A., Gray, R.L., 1984. Formation and structure of wood. In: Rowell, R.M. (Ed.), Chemistry of Solid Wood. American Chemical Society, Washington, D.C, pp. 1–56.
Postek, M.T., Howard, K.S., Johnson, A.H., McMichael, K.L., 1980. Scanning Electron Microscopy: A Student's Handbook. Ladd Research Industries, Inc., Burlington, Vt, p. 305.

Reid, I.D., 1985. Biological delignification of aspen wood by solid-state fermentation with the white-rot fungus *Merulius tremellosus*. Appl. Environ. Microbiol. 50 (1), 133–139.
Resch, H.R., Arganbright, D.G., 1971. Location of pentachlorophenol by electron microprobe and other techniques in Cellon-treated Douglas-fir. For. Prod. J. 21 (1), 38–43.
Robards, A.W. (Ed.), 1985. Botanical Microscopy. Oxford University Press, Oxford.
Ruel, K., Joseleau, J.P., 1984. Use of enzyme-gold complexes for ultrastructural localization of hemicelluloses in the plant cell wall. Histochemistry 81, 573–580.
Ruel, K., Barnoud, D., 1985. Degradation of wood by microorganisms. Chapter 16. In: Higuchi, T. (Ed.), Biosynthesis and Biodegradation of Wood Components. Academic Press, Inc, New York, pp. 441–467.
Ruel, K., Barnoud, F., Eriksson, K.-E., 1981. Micromorphological and ultrastructural aspects of spruce wood degradation by wild-type *Sporotrichum pulverulentum* and its cellulase-less mutant Cel 44. Holzforschung 35 (4), 157–171.
Ruel, K., Barnoud, F., Eriksson, K.-E., 1984. Ultrastructural aspects of wood degradation by *Sporotrichum pulverulentum*. Holzforschung 38 (2), 61–68.
Ruel, K., Barnoud, F., Joseleau, J.P., Johnsrud, S.C., Eriksson, K.E., 1986. Ultrastructural aspects of birch wood degradation by *Phanerochaete chrysosporim* and two of its cellulase deficient mutants. Holzforschung 40 (Suppl), 5–9.
Ryan, K.G., 1986. Preparation techniques for x-ray analysis in the transmission electron microscope of wood treated with copper-chrome-arsenate. Mater. Org. 21, 223–234.
Ryan, K.G., Drysdale, J.A., 1988. X-ray analysis of copper chromium and arsenic within the cell walls of treated hardwoods-new evidence against the microdistribution theory. J. Inst. Wood Sci. 11 (3), 108–113.
Saka, S., Goring, D.A.I., 1985. Localization of lignins in wood cell walls. Chapter 3. In: Higuchi, T. (Ed.), Biosynthesis and Biodegradation of Wood Components. Academic Press, Inc, New York, pp. 51–62.
Schmidt, O., Nagashima, Y., Liese, W., Schmidt, U., 1987. Bacterial wood degradation studies under laboratory conditions and in lakes. Holzforschung 41 (3), 137–140.
Srebotnik, E., Messner, K., 1988. Spatial Arrangement of Lignin Peroxidase in Poi Ne Decayed by *Phanerochaete chrysosporium* and *Fomitopsis pinicola*. The International Research Group on Wood Preservation, Document No. IRG/WP/1343, Stockholm, Sweden.
Srebotnik, E., Messner, K., Foisner, R., 1988a. Penetrability of white-rot degraded pi ne wood by the 1ignin peroxidase of *Phanerochaete chrysosporium*. Appl. Environ. Microbiol. 54 (11), 2608–2614.
Srebotnik, E., Messner, K., Foisner, R., Pettersson, B., 1988b. Ultrastructural localization of ligninase of *Phanerochaete chrysosporium* by immunogold labeling. Curr. Microbiol. 16, 221–227.
Thielke, C., 1982. Meiotic divisions in the basidium. In: Wells, K., Wells, E.K. (Eds.), Basidium and Basidiocarp Evolution, Cytology, Function, and Development. Springer-Verlag, Inc, New York, pp. 75–91.
Unligil, H.H., Chafe, S.C., 1974. Perforation hyphae of soft rot fungi in the wood of white spruce (*Picea glauca* (Moench) Voss.). Wood Sci. Technol. 8, 27–32.
Wilcox, W.W., Parameswaran, N., 1974. A method for electron microscopical demonstration of pentachlorophenol within treated wood. Holzforschung 28, 153–155.
Wilcox, W.W., Parameswaran, N., Liese, W., 1974. Ultrastructure of brown rot in wood treated with pentachlorophenol. Holzforschung 28 (6), 211–217.
Winandy, J.E., Morrell, J.J., 1993. Relationship between incipient decay, strength, and chemical composition of Douglas-fir heartwood. Wood Fiber Sci. 25 (3), 278–288.

Zabel, R.A., LKombard, F.F., Wang, C.J.K., Terracina, F., 1985. Fungi associated with decay in treated southern pine utility poles in the eastern United States. Wood Fiber Sci. 17 (1), 75–91.

Further reading

Blanchette, R.A., Otjen, L., Effland, M.J., Eslyn, W.E., 1985. Changes in structural and chemical components of wood delignified by fungi. Wood Sci. Technol. 19, 35–46.

Daniel, G., Nilsson, T., 1985. Ultrastructural and T.E.M. EDXA Studies on the Degradation of CCA Treated Radiata Pine by Tunneling Bacteria. International Research Group on Wood Preservation Document No. IRG/WP/1260, Stockholm, Sweden.

Daniel, G., Nilsson, T., 1986. Ultrastructural Observations on Wood-Degrading Erosion Bacteria. International Research Group on Wood Preservation Document No. IRG/WP/1283, Stockholm, Sweden.

CHAPTER TEN

Changes in the strength and physical properties of wood caused by decay fungi

Contents

Microbial invasion and colonization of wood causes numerous changes in a wide range of strength (mechanical) and physical properties. These changes range from drastic effects on wood strength to subtle modifications in properties such as density, hygroscopicity, electrical conductance, acoustics, caloric values, and dimensions. These changes and their rates of development vary with the wood species, the organisms involved and the environmental conditions. These changes are often hidden, poorly defined, and may partially explain the high variability of many wood properties. A review of the major microbial effects on wood properties will facilitate their detection and help define the roles and interactions of causal agents in the biodeterioration process.

It is important to remember that changes in one property are invariably associated with changes in other properties. For example, a loss in wood weight from decay decreases its caloric value and reduces strength properties. Subtle changes in properties such as permeability coupled with changes in nutritional factors, may increase decay susceptibility and set the stage for further colonization by other, more destructive agents of decay. It should be remembered that most changes in physical properties are

Wood Microbiology.
DOI: https://doi.org/10.1016/B978-0-12-819465-2.00010-3

simply reflections of the more basic anatomical and chemical changes in wood associated with decay development, reviewed in Chapters 7 and 8.

Strength is critical in most structural uses and has been the central concern of much wood decay research. Important changes in wood properties associated with microbial colonization include reduced strength, biomass loss (weight), hygroscopicity, increased permeability, and decreased dimensions.

This chapter will emphasize these topics and also review changes in electrical or acoustical properties that may be useful in non-destructive tests to detect early decay in wood products where strength is critical. This chapter places major emphasis on the effects of decay fungi on wood properties (The effects of sapstain and mold fungi are covered in Chapter 14). While insects can also cause substantial damage through the galleries they create, they have received much less attention because it is generally difficult to accurately estimate the extent of physical damage so that it can be related to changes in properties.

Wood weight loss (biomass loss)

Most wood-inhabiting microorganisms utilize the various components of the wood cell wall as they grow through the wood, reducing overall wood weight. Some fungi only utilize easily accessible nutrients in storage tissues or extractives, causing relatively minor weight losses (1−3%) and minimal damage. Others attack the more chemically complex components of the wood cell wall, eventually metabolizing these to carbon dioxide and water. Wood weight losses can approach 70% with brown-rot fungi, 96−97% for white rot fungi, and 3−60% for soft-rot fungi. The magnitude of weight loss depends on fungal type and wood species evaluated (Fig. 10.1). Weight loss is the most commonly used measure of decay capability and is generally expressed on an oven dry basis as: why does the formula appear twice

$$\text{Weight Loss (\%)} = [(\text{Original Weight} - \text{Decayed Weight})/\text{Original Weight}]100$$

$$\text{Weight Loss (\%)} = [(\text{Original Weight} - \text{Decayed Weight})/\text{Original Weight}]100$$

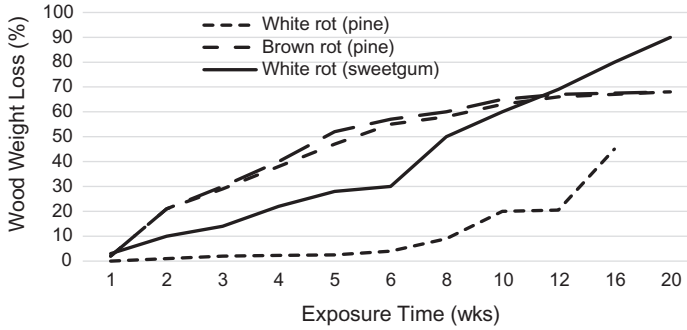


Figure 10.1 Relationships between incubation time and weight loss as affected by decay type and substrate. *From the data of Wilcox, W.W., 1968. Changes in Wood Microstructure Through Progressive Stages of Decay. U.S. Forest Service Research Paper FPL-70. Madison, WI.*

Weight loss is generally expressed at a given wood moisture content, most frequently on an oven-dry basis (ODW). Unfortunately, drying wood at high temperatures (100 °C) can degrade the wood-cell wall, potentially altering susceptibility to microbial attack. This problem can be overcome by conditioning the wood to an equilibrium moisture content (EMC) (usually 12%) over saturated salt solutions or in a controlled humidity-temperature room. In this method, the weight loss due to decay is the difference between the EMC weights before and after decay fungus exposure. The original ODW necessary for the weight loss percent determination is calculated from the moisture content of reference blocks kept under the same EMC conditions. However, wood EMC can vary with the degree and type of decay (Fig. 10.2) and become an error source unless accounted for in critical studies. Wood is also an extremely hygroscopic material and appreciable errors can be introduced during weighing when small test specimens with high surface-volume ratios are used unless the EMC conditions are carefully controlled (See Chapter 6). In such cases, the use of critically defined temperature, relative humidity conditions and weighing bottles are necessary for accuracy.

The use of control blocks not exposed to a decay fungus to account for any changes in equilibrium moisture content conditions between the original and the post-decayed blocks and to monitor any decay chamber effects such as autoclaving or nutrient migration are also necessary to achieve accuracy in laboratory decay determination tests, particularly in those cases where the weight losses due to decay are low.

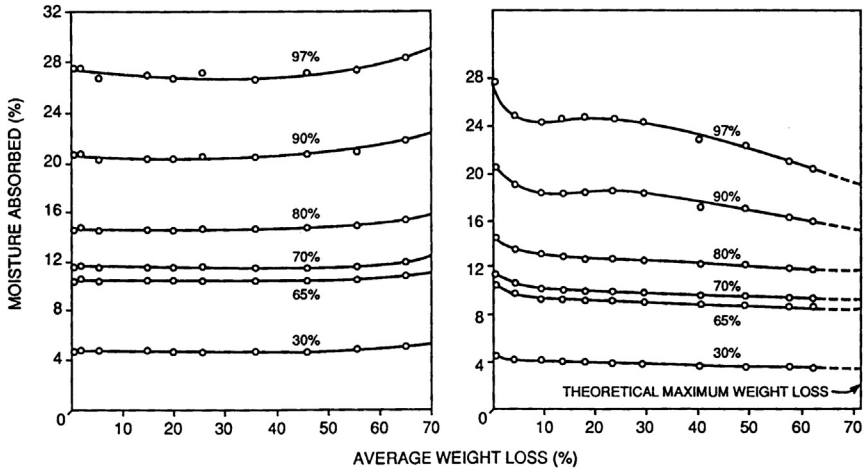


Figure 10.2 Moisture absorbed at various relative humidities at 26.5 °C for wood in progressive stages of colonization by (left) the white rot fungus *Trametes versicolor* and (right) the brown rot fungus *Rhodonia placenta*. *From data of Cowling, E.B., 1961. Comparative Biochemistry of the Decay of Sweet Gum Sapwood by White-rot and Brown-rot Fungi. USDA Forest Service Technical Bulletin No. 1258, Madison, WI.*

Wood weight loss is extremely useful under laboratory conditions, where the original wood weight can be obtained, but is of limited use for naturally decayed wood since the original weight of the wood can only be roughly approximated. Wood weight loss remains a useful comparative measure of decay capability (Hartley, 1958), but it does not accurately measure the magnitude of decay effects on many other wood properties such as strength (Wilcox, 1978).

Density/specific gravity loss

Density (wood mass/unit volume at a specific moisture content) and specific gravity (oven-dry wood weight/weight of displaced unit volume of water) are also used to measure the effects of microbial attack. Both examine changes in mass relative to either the direct volume (density) or the volume of water the test piece displaces water (specific gravity). The disadvantages of these methods are the difficulty of accurately measuring wood volume due to its sensitive hygroscopic properties and the higher variability since two wood property measurements are involved. White-rot

fungi cause substantial weight losses with little change in wood volume, while brown rot fungi cause substantial volume reductions as the wood tends to collapse as it is degraded. Thus, changes in density are not comparable between decay types. Low density, measured as abnormal lightness is often used as a rough decay indicator by lumber graders since density is closely correlated with some strength properties, such as bending. Density can also be measured, indirectly, using x-rays that pass at different rates through sound and decayed wood (Mothershead and Stacey, 1965); but this technique is both expensive and difficult to interpret. Density scans are, however, used in some mills to estimate timber properties as part of the grading process. Density can also be inferred acoustically, using the time it takes for sound to pass through a wood sample (time of flight), but this method requires prior testing to establish the relationship between time of flight and density for a given wood species. Surface density can also be indirectly estimated using mechanical devices such as the Pilodyn, a spring-loaded pin penetration device (Cown and Hutchinson, 1983; Hoffmeyer, 1978); however, readings often vary with wood species and must be corrected for wood moisture content (Smith and Morrell, 1986).

Strength (mechanical) properties

The effects of decay fungi on wood strength have been intensively studied. As fungi grow through the wood, they modify the chemical structure and remove mass, thereby altering the mechanical properties of wood. Wood derives its strength from a combination of highly oriented cellulose microfibrils and encrusting hemicellulose. Any changes in arrangements of these carbohydrates often causes sharp reductions in wood strength.

As a result of their many obvious deleterious effects on wood use, a large literature has accumulated on the effects of microbial colonization on wood strength. Unfortunately, most of this research examined the effects of a limited microbial flora on a few commercially important wood species using a variety of different measures of wood strength.

Early studies on the effects of decay emphasized heartrots of standing trees and developed estimated of losses of usable volumes of lumber

(Colley, 1921). The effects of these fungi were further quantified by testing beams cut from clear and colonized wood (Scheffer et al., 1941; Hartley, 1958). As the effects of these fungi were quantified, the emphasis shifted to the roles of various fungi associated with the decay of buildings and other wood products (Cartwright et al., 1931). These early studies were dependent on naturally decayed material, but as techniques became more refined, individual fungi were isolated and tested for their ability to cause weight and strength losses under controlled laboratory conditions. One of the first carefully controlled studies of the effects of a single decay fungus on wood strength was performed by Scheffer, who evaluated the effects of *Trametes versicolor* on red gum (1936).

The mechanical properties of wood can be measured by a variety of methods. Wood strength is covered in detail in several textbooks (Panshin and deZeeuw, 1980; Hoyle, 1972; Bodig and Jayne, 1982; U.S. Department of Agriculture, 2010). Virtually every country on the planet has procedures for standard strength tests on wood. In the U.S., these standards are promulgated by ASTM International, a consensus standards writing body (American Society for Testing and Materials ASTM, 2017). There are numerous strength properties that can be measured, but those most often used to measure the effects of decay include modulus of rupture (MOR), modulus of elasticity (MOE), work to maximum load in bending, maximum crushing strength, compression perpendicular to the grain, impact bending, tensile strength parallel to the grain, toughness, hardness, and shear strength (Brown, 1963; Henningsson, 1967; Kennedy and Ifju, 1962; Mulholland, 1954; Toole, 1969,1971; Armstrong and Savory, 1959). Of these many strength properties, work to maximum load, toughness, and impact bending are reported to be the most sensitive to detect early decay (Wilcox, 1978).

Most studies of microbial effects on wood strength have used bending tests of small clear specimens that had been exposed to the desired test organism for varying time periods. The beams can range from matchsticks to full scale lumber.The load applied to the wood and the deflection of the specimen are measured during the test and then plotted to determine MOR and MOR (Fig. 10.3). Bending tests of simple beams (supported at both ends and. center loaded) measure the effects of microbial colonization at the center of the span, and any effects of colonization away from this zone on the measurement are sharply diminished. Thus, uneven colonization of even small test beams can produce variable results. Other studies have used tensile strength, wherein wood samples are pulled apart with

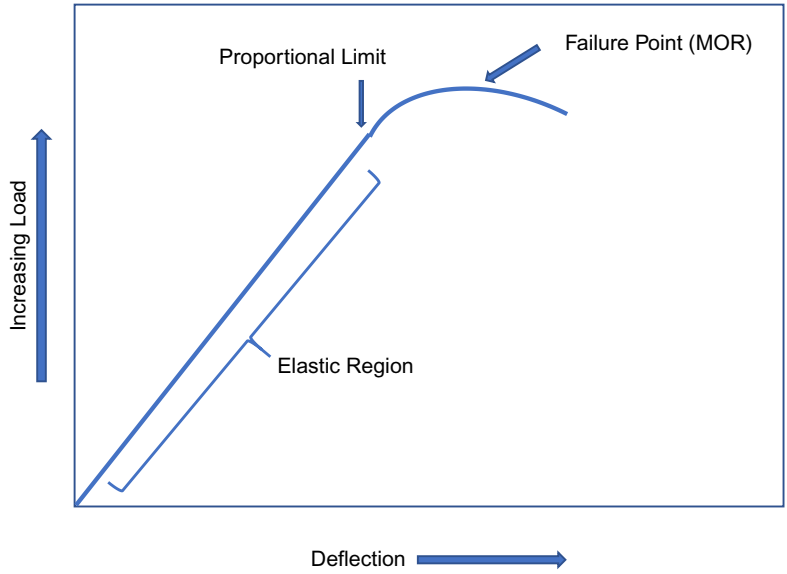


Figure 10.3 Example of a typical load/deflection plot for a wood beam in bending showing the elastic region, the proportional limit, and the failure point.

the load applied parallel to the fiber orientation. Wood has tremendous strength in this orientation, but tensile strength is also very sensitive to the early stages of decay.

While bending and tensile tests are useful, many researchers have employed other strength tests using smaller, more easily handled specimens to accelerate the decay process and increase replication. These tests include tension parallel to the grain, radial or longitudinal compression testing, and small-scale tests such as the breaking radius and pendulum tests (Graham and Safo-Sampah, 1976; Scheffer, 1978). In the latter tests, small wafers are exposed to the test fungus and tested either by dropping a heavy pendulum into the center of the wafer and measuring the force absorbed or by bending the wafer successively around a series of mandrils with smaller diameters until failure is achieved. In both instances, the variation is high and many specimens must be tested to achieve reasonable accuracy (Sexton, 1988).

Another useful method for rapid strength tests on small beams is use of the toughness testing machine developed by the U.S. Forest Products Laboratory (Forest Products Laboratory, 1941). This test measures the kinetic energy absorbed by a swinging pendulum during the sudden

breaking of a small beam. Toughness values are measured in inch-pounds and comparative data have been developed for a number of commercial wood species. Reductions in toughness are a particularly useful and sensitive indicator of the early decay stage. An advantage of the method is that large numbers of test beams can be measured rapidly. A disadvantage is that the measuring unit (inch-pounds) cannot be related directly to other strength values. Nevertheless, pendulum or mandril tests are useful methods for rapidly screening large numbers of microorganisms to roughly approximate decay capability.

Radial and longitudinal compression strength tests (RCS or LCS, respectively) can be performed on small blocks or samples removed from larger wood members. RCS is especially sensitive to the early stages of decay, particularly for the brown rot fungi (Kubiak and Kerner, 1963; Smith and Graham, 1983), while LCS is affected by density changes and more closely relate to bending properties (Smith and Morrell, 1987).

Wilcox (1978) surveyed the literature on the effects of fungi on strength properties and reported that toughness or resistance to impact loading, was the property most sensitive to the early stage of decay, followed by static bending properties (as work) (Table 10.1, Fig. 10.4). In general, brown rots are perceived to be more damaging because they are associated with greater strength losses at lower weight losses than white rots. These rapid decreases in strength at early stages of decay can be attributed to the fact that brown rot fungi produce enzymes that diffuse well away from the fungal hyphae. As a result, the damaging effects of colonization by a brown rot fungus extend far beyond the limited occurrence of hyphae present in the incipient stages of decay. White-rot fungi generally produce enzymes that are more closely associated with hyphal tips and, therefore, the strength effects associated with these fungi are less significant in the early stages of attack (<5% weight loss). White rot fungi also tend to utilize decomposition products at approximately the same rates they are produced, leading to a more gradual loss in strength. However, there is little difference in the degree of strength loss between the two fungal groups at the later stages of attack where the fungal mycelia are well distributed through the wood structure. A bias in the current literature is the preponderance of strength testing performed using only a few brown rot fungi (e.g. *Rhodonia* (*Poria*) *placenta*, *Gloeophyllum* (*Lenzites*) *trabeum*) since they are more commonly associated with decays in structural applications using coniferous woods and are easily grown under laboratory conditions. While the strength effects of decay by brown and

Table 10.1 Estimated values for strength losses in softwoods and hardwoods at early stages of decay (as indicated by weight loss) by brown and white rot fungi[a].

Approximate weight loss (%)	Toughness	Impact bending	Static bending				Compression perpendicular (radial)	Compression parallel	Tension parallel	Shear parallel	Hardness
			Bending	Work to max load	Modulus of rupture	Modulus of elasticity					
Brown rot-softwoods											
1	57	20–38	–	–	–	–	–	–	–	2	–
2	–	20–50	5	27	13–50	4–55	18–24	10	23–40	–	–
4	75	25–55	–	–	–	–	25–35	–	–	6	7
6	–	62–72	16	–	61	65	48	25	60	–	–
8	–	78	–	–	–	–	48–60	–	50	15	21
10	–	85	36	–	70	–	66	45	–	20	–
Brown rot-hardwoods											
1	–	6–27	–	–	–	–	–	–	–	–	–
2	36	31–50	–	54	32	–	6–10	–	56	–	–
4	–	60–70	–	69	49	–	–	–	–	–	–
6	–	80	–	75	61	–	16–25	–	–	–	–
8	–	9–89	13–34	–	–	–	19	–	82	–	–
10	60	70–92	–	–	–	–	–	–	–	–	–
White rot-softwoods											
1	55	–	–	–	–	–	–	–	–	–	–
2	–	–	–	–	–	–	10–20	–	4–38	–	–
4	–	–	–	–	–	–	–	–	8–43	–	–
6	75	–	–	–	–	–	32–61	–	10–49	–	–
8	–	–	–	–	–	–	–	–	14,058	–	–
10	85	–	–	–	–	–	–	–	20–63	–	–

(*Continued*)

Table 10.1 (Continued)

Approximate weight loss (%)	Toughness	Impact bending	Static bending				Compression perpendicular (radial)	Compression parallel	Tension parallel	Shear parallel	Hardness
			Bending	Work to max load	Modulus of rupture	Modulus of elasticity					
White rot-hardwoods											
1	–	21	–	–	–	–	4	–	–	–	–
2	–	26	–	28–35	13–14	4	5	–	22–42	–	–
4	70	44	–	38	20	–	–	–	17–44	–	–
6	75	50	–	45–53	20–27	10	12–27	14	12–58	–	–
8	–	–	–	–	–	–	–	–	14–49	–	–
10	85	60	–	58	24	14	55	20	20–50	–	–

[a]As a percentage of the values for non-decayed samples. From Wilcox (1978). Values obtained from published experimental results and adjusted to equivalent weight loss levels.

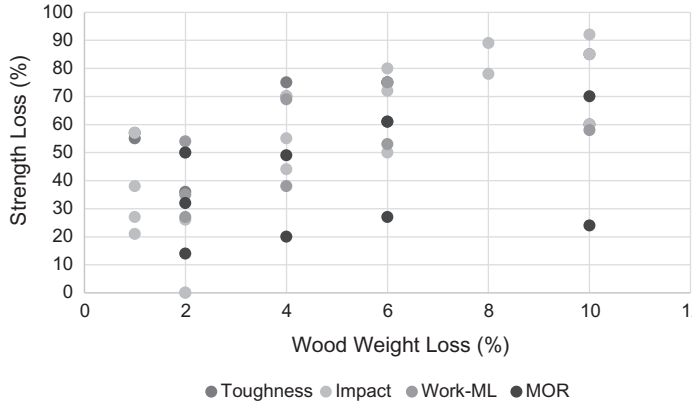


Figure 10.4 Relationship between wood weight loss and strength for selected white, brown and soft rot fungi. *Data from Table 10.1*.

white rot fungi have been studied, the effects of other decay types on wood strength are less clearly defined.

Soft-rot fungi are similar to brown rot fungi in that they cause large strength losses at low wood weight losses. Some soft rot fungi (Type 1) also remove sizeable sections of the wood cell wall during cavity formation, forming many failure zones and magnifying the strength losses in very small areas of the wood cell wall. Furthermore, hyphal penetration of multiple cell walls has been noted with some soft-rot fungi (Type 2) and may also create planes of weakness (Morrell and Zabel, 1985). At present, soft rot damaged wood, when detectable visually, is presumed to have no residual strength (Hoffmeyer, 1978); however, further research on the enzyme systems associated with soft rot fungi and their progressive effects on wood strength are needed to better quantify the relationship between types of microbial colonization and strength effects.

One weakness of the many laboratory studies on the effects of various decays on strength is the use of copious quantities of inoculum in an axenic system, generally in the presence of an excess of sugar compounds, to accelerate fungus growth. In natural systems, fungal colonization originates from spores or hyphal fragments that must germinate and grow through the wood, generally in the absence of exogenous nutrients. These fungi must compete with other microbes that are also attempting to colonize and utilize the substrate. Tests using *Stereum sanguinolentum* and *Rhodonia placenta*, indicated that microbial colonization of the wood was relatively rapid, while effects on wood properties occurred more gradually

(Smith et al., 1992). These results suggest that there is a delay between colonization and strength effects.

One area of decay-associated strength effects that has received less intensive study is the effect of microbial attack on fiber properties during pulping for paper production. As the fungus attacks the wood, cellulose microfibrils become more susceptible to chemical and physical damage during the pulping process. While the effects of microbial attack on pulp yield and paper properties have been studied (McGovern et al., 1951; Lindgren and Eslyn, 1961; Rothrock et al., 1961; Christie, 1979), most studies have depended upon natural colonization in pulpwood or chip piles. Thus, the microflora and its potential effects in a particular study might vary widely with season, wood moisture content, pulpwood species, or other environmental conditions. Microbial colonization, as measured by wood density losses with storage time, was associated with a 15% decrease in burst strength and a 24% decline in folding over a 6-month period (McGovern et al., 1951). Similarly, tear strength declined by 35% with a 7% decline in density over a 5-month period (Rothrock et al., 1961). The effects of microbial colonization on other pulpwood and pulp properties are discussed in Chapter 13 and 14. Losses in pulping properties may be difficult to detect because degraded fibers may be lost in the pulping process resulting in yield loss, but there is relatively little change in the fibers that are retained.

The effects of microbial agents on wood strength properties vary widely with the microbial agents involved, wood species, and environmental conditions, but the previous studies suggest that most changes are readily measurable before the wood has exceeded a 5% weight loss. These effects highlight the importance of timely and accurate detection of early decay since substantial reductions in the strength properties in brown rots have already occurred. Improving our knowledge about the types and rates of these strength losses and their detection will enhance greatly the reliability of wood systems and increase the safety of these structures. It may also reveal other property changes highly sensitive to early decay changes that may improve the effectiveness of early decay detection. This becomes especially important in wood structures where inspectors must first detect decay and then, more importantly, estimate its effects. Many inspectors take a conservative approach and reject any wood showing evidence of wetting or decay.

Hygroscopicity

As microbial enzymes degrade the ligno-carbohydrate matrix, they induce changes in the moisture holding capacity of the wood cell wall. These changes, in turn, alter the equilibrium moisture content (EMC) and can influence the results of decay studies where wood weight loss measurements are made by equilibrating wood at constant temperature-relative humidity conditions (Fig. 10.2). Generally, the EMC of brown-rotted wood is lower than sound wood, while the EMC of white rotted wood is higher when induced weight losses exceed 60% (Cowling, 1961). Increased EMC begins at about 40% weight loss for white-rotted wood, while brown rotters are associated with sharp drops in EMC at the early stages of decay. The sharp drop in EMC associated with brown-rot fungi probably reflects preferential attack of the amorphous cellulose which retains higher levels of adsorbed water than the crystalline cellulose regions. Early loss of amorphous cellulose decreases the overall moisture holding capacity of the wood. The absence of EMC changes in the early stages of white rot attack probably reflects uniform removal and near simultaneous utilization of all wood components, while increases in EMC during the later stages of decay by these fungi may reflect selective attack of crystalline cellulose.

Caloric value

As microbial agents colonize and utilize the wood substrate, they remove and convert wood substance to microbial biomass, carbon dioxide, water, and metabolic waste products. Although microbial biomass makes a slight contribution to the caloric value, the net energy content of the decayed wood declines as substantial amounts of cell wall polymers are converted to carbon dioxide (Fig. 10.5). Caloric value becomes particularly important wherever wood is used for heating, since additional biomass must be used to generate the same quantity of heat. This can be problematic if a wood-heating facility is limited in the volume that can be processed over time. In general, losses in caloric value of decayed wood should be directly proportional to the density reduction (Scheffer, 1936). On a weight basis, conifers due to their higher lignin content have a

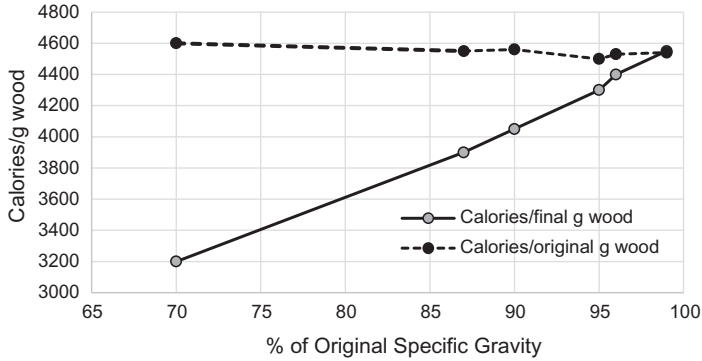


Figure 10.5 Net caloric value of wood decayed to various weight losses on original or final wood weight basis. *After Scheffer, T.C., 1936. Progressive Effects of* Polyporus versicolor *on the Physical and Chemical Properties of Sweet-gum Sapwood. USDA Tech. Bull. No. 527.*

higher caloric value (e.g. pines: 8836 b.t.u.'s per lb. or 20,535 kJ/kg) than hardwoods (e.g. Oak: 8556 b.t.u.'s per lb. or 19,883 kJ/kg). This higher caloric value is usually masked by the lower density of conifers. On this basis, brown rotted wood should have a higher caloric value (at equal weight loss) than white rotted wood, but no confirming data are available. Differential scanning calorimetry provides a rapid laboratory method for detecting the relative amounts of cellulose, hemicellulose, and lignin remaining in decayed wood (Reh et al., 1986).

Permeability

Although some wood-inhabiting fungi directly penetrate the wood cell wall to move from cell to cell, the majority of the decay organisms initially move through the wood primarily by direct pit penetration. Since pits represent the major limiting factor in fluid flow through fibers and tracheids, removal of pit membrane makes the wood markedly more receptive to fluid movement. As a result of these changes, decayed wood absorbs and desorbs liquids more readily than sound wood.

Excessive absorption can result in sinking of logs during storage, reduces the rate of kiln drying time for some species, and can result in uneven absorptions of preservative treatments. The process of pit removal due to bacterial action in ponded logs is a relatively slow process

(Ellwood and Eckland, 1959); however, over many years, sinker logs can represent a substantial loss in volume. In fact, companies regularly salvage or mine ponds at old lumber mills to recover these sinker logs. While the outer sapwood is often degraded by bacteria, the interior is still sound. The presence of differing areas of permeability can also influence the rate of lumber drying due to the variable distribution of decayed and sound wood (Ward, 1986). The bacterially attacked wood for example, is markedly wetter than uninfected wood. These materials have differing drying rates than the uninfected wood that may be present in the same board (Ward, 1986).

Increased permeability will also increase the sorption of various preservatives, glues and coatings applied to the wood. Excessive uptakes increase preservative costs and serious preservative bleeding or coating retention problems can develop. In gluing, more permeable wood can lead to dry joints and premature failures. In coatings, color variations and retention problems may develop as the more permeable wood absorbs solutions differently than the sound zones. The increased permeability of fungal colonized wood has been explored to improve the treatability of difficult to treat species (Lindgren, 1952; Graham, 1954; Schultz, 1956; Lindgren and Wright, 1954). Fungi, such as *Trichoderma viride*, a non-decaying scavenger of carbon compounds, were inoculated on the wood surface and allowed to colonize the freshly peeled wood. While treatability was improved near the surface, the results were inconsistent and treatment of the heartwood was not markedly improved (Johnson and Gjovik, 1970).

In addition to the obvious effects on end-uses, increased permeability may also alter the ecology of the decaying wood. More permeable wood will wet more readily and, as a result, conditions in the wood will be conducive to microbial colonization for longer time periods.

Electrical properties

Wood has a much lower electrical conductivity than other construction materials such as steel, and this is one reason for its common use to support electric distribution systems. As the wood is degraded, however, its electrical conductivity increases (Richards, 1954). As a result, electric resistance type moisture meters can sometimes over-estimate the moisture content in decayed wood that is below the fiber saturation point.

Changes in electrical properties due to microbial colonization using pulsing electric currents have been used to detect early decay, particularly in live trees (Shigo and Shigo, 1974). In this process, voltages are pulsed through a twisted wire probe that is insulated along its length, but exposed at the tip. Electrical resistance is measured as the probe is inserted into a predrilled hole. Sound wood has a high electrical resistance, while decayed or discolored wood registers resistance readings that are 50–75% lower than the sound material. The Shigometer has been extensively used to detect decay and discoloration in living trees (Shigo and Shigo, 1974; Shigo et al., 1977; Shigo and Berry, 1975; Shortle et al., 1978; Skutt et al, 1972), but has been less consistent in wood products probably because variations in wood moisture content affect readings to a greater extent than any microbially-mediated changes in resistance (Shigo et al, 1977; Shortle et al., 1982; Piirto and Wilcox, 1978; Zabel et al, 1982; Graham and Inwards, 1980). A disadvantage of the Shigometer is the limitation of accurate usage to wood that is above the fiber saturation point. Electrical resistance is presumed to increase over the course of decay due to the release of ions and organic acids during decay. These changes are easily detected in the standing tree since wood moisture contents generally exceed the fiber-saturation point; however, moisture contents in wood products vary widely with position in the sample. For example, moisture contents around water trapping joints will generally be higher than in zones away from these sites. Furthermore, moisture content in wood products varies with season. Thus, moisture content variations will minimize the usefulness of a tool that is heavily dependent on moisture. Finally, wood in soil contact or near metal fasteners will often experience inward migration of various salts that can also influence electrical resistance.

The results to date suggest that, while changes do occur in electrical resistance over the course of microbial colonization of wood, these changes may be masked by the larger effects of migration of chemicals from outside the wood or by variations in wood moisture content.

Acoustic properties

Wood is an excellent transmitter of sound waves and also produces characteristic acoustic emissions as it is stressed mechanically. Musicians

have long depended on the tonal quality of various wood species to create various wood instruments. As wood is colonized by microbial agents, its ability to transmit or emit sounds is generally altered (Pellerin et al., 1986; Noguchi et al., 1986). Alterations in acoustic properties associated with fungal attack can be exploited to detect various stages of decay. As sound waves move through wood, they will pass around decay pockets or voids. The time of flight (velocity) of a given pulse of sound across the cross section of a timber can be used to determine the internal condition of the wood. There are a variety of possible uses for sound measurements in wood, but the natural variations in the wood structure make velocity measurements the easiest to use for detecting advanced decay (>25% wood weight loss) where voids in wood structure are present.

A second approach to measuring changes in acoustic properties is analyzing the characteristics of a sound wave after it has passed through the wood. As a sound wave moves through the wood, the characteristics of the wood (growth rings, knots, checks, etc.) modify the wave pattern. The modified wave represents an acoustic "fingerprint" of the wood. Small changes in the structural integrity of the wood due to microbial activity should be discernable in the resulting fingerprint. At present, the wave form analysis of wood is still relatively crude; however, progress in other areas of material science suggest that this technique has promise to non-destructively monitor changes in the wood over the course of decay (Pellerin et al., 1986; Ross and Pellerin, 1990).

Summary

1. The major effects of decay development on the physical properties of wood are weight and density losses, strength reductions, and changes in permeability, hygroscopicity, electrical conductance, caloric content, acoustics, and dimensions.
2. Important changes in wood properties associated with decay development affecting many wood uses are weight loss, strength reductions and increased permeability.
3. Weight loss is nearly linear over time when the decay fungi are established and decay conditions optimal.

4. Total wood weight losses may approach 96–97% for white rot fungi, up to 70% for brown rot fungi, and range from 3% to 60% for soft-rot fungi.
5. Of the many strength properties used to measure the effects of decay, work to maximum load, toughness, and impact bending have proven to be most sensitive to the detection of early decay.
6. Of the fungi tested, brown rots cause substantially greater strength losses at lower weight losses than white rots. In brown rots, strength reductions of up to 80% can occur with weight losses of less than 5%.
7. The equilibrium moisture content (EMC) of brown rotted wood is lower than sound wood at all weight losses, while it is higher for white rots at weight losses exceeding 60%.
8. In general, the caloric values of decayed wood are highest with the brown rots and are directly related to weight loss or density reductions.
9. Decayed wood has a higher electrical conductivity than sound wood but moisture content is a significant confounding factor at levels below the fiber saturation point.
10. Sound moves through decayed wood differently than non-decayed wood. These patterns can be used as non destructive tests for detecting early decay.
11. Most studies of the physical changes in wood properties associated with decay have utilized a limited number of brown or white rot fungi that are easily maintained in cultures, particularly those isolated from conifers. There is a danger in generalizing from these few fungi and a need to determine the effects of other fungi and decay types on the major use properties of wood.

References

American Society for Testing and Materials (ASTM), 2017. Volume 4.10 Wood. ASTM Annual Book of Standards. ASTM International, West Conshohocken, PA.

Armstrong, F.H., Savory, J.G., 1959. The influence of fungal decay on the properties of timber. Holzforschung 13, 84–89.

Bodig, J., Jayne, B.A., 1982. Mechanics of Wood and Wood Composites. Van Nostrand Reinhold, N.Y, 712p.

Brown, F.L., 1963. A tensile test for comparative evaluation of wood preservatives. For. Prod. J. 13 (9), 405–412.

Cartwright, K.St.G., F'indlay, W.P.K., Chaplin, C.J., Campbell, W.G., 1931. The Effect of Progressive Decay by *Trametes serialis* Fr. on the Mechanical Strength of Wood of Sitka Spruce. Great Britain Dept. Sci. Ind. Res. For. Prod. Res. Bull. No. 11.

Christie, R.D., 1979. The use of decayed wood in pulping processes. Chapter 6. In: Hatton, F.V. (Ed.), Chip Quality Monograph. Pulp and Paper Technology Series. TAPPI, Atlanta, Georgia.
Colley, R.H., 1921. The effect of incipient decay on the mechanical properties of airplane timber. Phytopathology 11, 45.
Cowling, E.B., 1961. Comparative Biochemistry of the Decay of Sweet Gum Sapwood by White-Rot and Brown-Rot Fungi. USDA Forest Service Technical Bulletin No. 1258, Madison, WI.
Cown, D.J., Hutchinson, J.D., 1983. Wood density as an indicator of the bending properties of *Pinus radiata* poles. N. Zealand J. For. Sci. 13 (1), 87–99.
Ellwood, L.A., Eckland, B.A., 1959. Bacterial attack of pine logs in pond storage. For. Prod. J. 9 (9), 283–292.
Graham, R.D., 1954. The preservative treatment of Douglas-fir post sections infected with *Trichoderma* mold. For. Prod. J. 4, 164–166.
Graham, R.D., Safo-Sampah, S., 1976. Rapid agar stick breaking-radius test to determine the ability of fungi to degrade wood. Wood Sci. 9 (2), 65–69.
Graham, R.D., Inwards, R.D., 1980. Comparing methods for inspecting Douglas-fir poles in service. Proc. Am. Wood Preserv. Assoc. 76, 283–289.
Hartley, C., 1958. Evaluation of Wood Decay in Experimental Work. USDA Forest Products Laboratory General Technical Report 2119, Madison, WI.
Henningsson, B., 1967. Changes in impact bending strength weight, and alkali solubility following fungal attack of wood. Studia. Forestalia Suecica (Stockholm) No. 41. special reference to wooden poles. Soft Rot in Utility Poles Salt-treated in the Years 1940–1954. Swedish Wood Preservation Institute, Nr 11.
Hoffmeyer, P., 1978. The Pilodyn as a non-destructive tester of the shock resistance of wood. Proceedings 4th Symposium on Non-Destructive Testing of Wood. Washington State University, Pullman, WA.
Hoyle Jr., R.J., 1972. Wood Technology in the Design of Structures. College of Engineering, Washington State University, Pullman, Washington.
Johnson, B.R., Gjovik, L.R., 1970. Effect of *Trichoderma viride* and a contaminating bacterium on microstructure and permeability of Loblolly pine and Douglas-fir. Proc. Am. Wood Preserv. Assoc. 66, 234–240.
Kennedy, R.W., Ifju, G., 1962. Applications of microtensile testing to thin wood sections. Tappi 45 (9), 725–733.
Kubiak, M., Kerner, G., 1963. Die Veranderungen einiger physikalischer Eigerschaffen der Druckfestigkeit und der chemischen Zummensetzing des Buchenholzes im anfangsstadium des Holzabbaues durch *Coniophora cerebella* Pers und *Stereum hirsutum* Willd. Drev. Vysk. 1963 (4), 181–193.
Lindgren, R.M., 1952. Permeability of Southern pine as affected by mold and other fungus infection. Proc. Am. Wood Assoc. 48, 158–174.
Lindgren, R.M., Wright, E., 1954. Increased absorptiveness of moldy Douglas-fir poles. For. Prod. J. 4, 162–164.
Lindgren, R.M., Eslyn, W.E., 1961. Biological deterioration of pulpwood and pulp chips during storage. Tappi 44 (6), 419–429.
McGovern, J.N., Martin, J.S., Hytinner, A., 1951. Effect of storage of slash pine pulpwood on sulfate and ground wood pulp quality. For. Prod. Res. Soc. Proc. 5, 162–169.
Morrell, J.J., Zabel, R.A., 1985. Wood strength and weight losses caused by soft rot fungi isolated from treated southern pine utility poles. Wood Fiber Sci. 17 (1), 132–143.
Mothershead, J.S., Stacey, S.S., 1965. Applicability of radiography to inspection of wood products. Proceedings 2nd Annual Symposium on Non-Destructive Testing of Wood.

Spokane, WA. Published by Washington State University, Pullman, WA, pp. 307–336.
Mulholland, J.R., 1954. Changes in weight and strength of Sitka spruce associated with decay by a brown rot fungus. For. Prod. J. 4 (6), 410–416.
Noguchi, M., Nishimoto, K., Imanura, Y., Fujii, Y., Okamura, S., Miyauchi, T., 1986. Detection of very early stages of decay in western hemlock using acoustic emissions. For. Prod. J. 36 (4), 35–36.
Panshin, A.J., deZeeuw, C., 1980. Textbook of Wood Technology, fourth ed. McGraw Hill, New York.
Pellerin, R.F., DeGroot, R.C., Esenther, G.R., 1986. Nondestructive stress wave measurements of decay and termite attack units. Proceedings 5th Nondestructive Testing of Wood Symposium. Washington State University, Pullman, WA.
Piirto, D., Wilcox, W.W., 1978. Critical evaluation of the pulsed-current resistance meter for detection of decay in wood. For. Prod. J. 28 (1), 52–57.
Reh, U., Kraepelin, G., Lamprecht, I., 1986. Use of differential scanning calorimetry for structural analyses of fungally degraded wood. Appl. Environ. Microbiol. 52 (5), 1101–1106.
Richards, O.B., 1954. Physical changes in decaying wood. J. For. 52, 260–265.
Ross, R.J., Pellerin R., 1990. Non destructive evaluation of wood: past, present, and future. Proceedings Symposium on non-destructive evaluation of materials. Annapolis, MO. (Conference draft, unpublished).
Rothrock Jr., C.W., Smith, W.R., Lindgren, R.M., 1961. The effects of outside storage on slash pine chips in the South. Tappi 44 (1), 65–73.
Scheffer, T.C., 1936. Progressive Effects of *Polyporus versicolor* on the Physical and Chemical Properties of Sweet-gum Sapwood. USDA Tech. Bull. No. 527.
Scheffer, T.C., 1978. Bending tolerance of veneer as related to its toughness. For. Prod. J. 29 (2), 53–54.
Scheffer, T.C., Wilson, T.R.C., Luxford, R., Hartley, C., 1941. The Effect of Certain Heartrot Fungi on the Specific Gravity and Strength of Sitka Spruce and Douglas-fir. U.S.D.A. Technical Bulletin 779, Washington, D.C, 24 p.
Schultz, G., 1956. Exploratory test to increase preservative penetration in spruce and aspen by mold infection. For. Prod. J. 6 (2), 77–80.
Sexton, C.M., 1988. The Development and Use of Breaking Radius and Impact Bending Test for Measuring Wood Strength was Caused by Basidiomycetes Isolated From Air-seasoning Douglas-fir (M.S. thesis). Oregon State University, Corvallis, OR, 65p.
Shigo, A.S., Shigo, A., 1974. Detection of Discoloration and Decay in Living Trees and Utility Poles. USDA For. Service Paper. NE-294, Washington, D.C.
Shigo, A.L., Berry, P., 1975. A new tool for detecting decay associated with *Fornes annosus* in *Pinus resinosa*. Plant. Dis. Rep. 59 (9), 739–742.
Shigo, A.L., Shortle, W.C., Ochrymowych, J., 1977. Detection of Active Decay in at Groundine and Utility Poles. USDA Forest service General Technical Report NE-35, Washington, D.C.
Shortle, W.C., 1982. Decay of Douglas-fir wood: ionization associated with resistance to a pulsed electric current. Wood Sci. 15 (1), 29–32.
Shortle, W.C., Shigo, A.L., Ochrymowych, J., 1978. Patterns of resistance to a pulsed electric current in sound and decayed utility poles. For. Prod. J. 28 (1), 48–51.
Skutt, H.R., Shigo, A.L., Lessard, R.M., 1972. Detection of discolored and decayed wood in living trees using a pulsed electric current. Can. J. For. Res. 2, 54–56.
Smith, S.M., Graham, R.D., 1983. Relationship between early decay and radial compression strength of Douglas-fir. For. Prod. J. 33 (6), 49–52.
Smith, S.M., Morrell, J.J., 1986. Correcting Pilodyn pin measurement of Douglas-fir for different moisture levels. For. Prod. J. 36 (1), 45–46.

Smith, S.M., Morrell, J.J., 1987. Longitudinal compression as a measure of ultimate wood strength. For. Prod. J. 37 (5), 49–53.
Smith, S.M., Morrell, J.J., Sexton, C.M., 1992. Effect of colony size on residual wood strength of Douglas-fir sapwood and heartwood. For. Prod. J. 42 (4), 19–24.
Toole, L.R., 1969. Effect of decay on crushing strength. For. Prod. J. 19 (10), 36–37.
Toole, L.R., 1971. Reduction in crushing strength and weight associated with decay by rot fungi. Wood Sci. 3 (3), 172–178.
U.S.D.A., 1941. Forest Products Laboratory Toughness Testing Machine. Report No 1308, Madison, WI.
U.S. Department of Agriculture, 2010. Wood Handbook. U.S. Forest Products Laboratory, Madison, WI.
Ward, J.C., 1986. The effect of wetwood on lumber drying times and rates: an exploratory evaluation with longitudinal gas permeability. Wood Fiber Sci. 18 (2), 288–307.
Wilcox, W.W., 1968. Changes in Wood Microstructure Through Progressive Stages of Decay. U.S. Forest Service Research Paper FPL-70, Madison, WI.
Wilcox, W.W., 1978. Review of literature on the effects of early stages of decay on wood strength. Wood Fiber 9 (4), 252–257.
Zabel, R.A., Wang, C.J.K., Terracina, F.C., 1982. The Fungal Associates, Detection and Fumigant Control of Decay in Treated Southern Pine Poles. Final Report EL2768. Electric Power Research Institute, Palo Alto, CA.

CHAPTER ELEVEN

Colonization and microbial interactions in wood decay

Contents

In this chapter we consider how decay fungi colonize wood and effectively compete with other wood-inhabiting microorganisms. An emphasis is placed on the colonization strategies of decay fungi and their associations and interactions with other wood-inhabiting microorganisms. These topics are a part of the expanding discipline of microbial ecology. Topics of related interest to the wood microbiologist include soil microbiology, ruminant digestion, decomposition processes, and microbial waste conversions. The broad outlines of microbial ecology are covered in current textbooks on microbiology and microbial ecology (Pugh, 1980; Atlas and Bartha, 1998; Brock et al., 1984; Campbell, 1983). This chapter provides a background for the next five chapters that discuss the major decays and discolorations that can develop during the processing and some uses of wood.

First, a brief review of some of the major concepts and terms developed by ecologists is provided to describe organismal roles and interactions in the environment.

Wood Microbiology.
DOI: https://doi.org/10.1016/B978-0-12-819465-2.00011-5

Some ecological concepts and terminology

Most organismal interactions involve competition for space, nutrients, or energy. The organisms associated with wood decay are heterotrophs and the competition is primarily nutritional. Wood in the living tree, during conversion into products, and in its many product forms, represents a rich potential nutrient source or useful habitat for many microorganisms. The bark of the living tree and the cambium beneath provide trees with a reasonable array of defenses against fungal and insect attack. Extreme microbial competition for the substrate is the common rule when wood suddenly becomes available for use after tree wounding or felling or when untreated wood is used in conditions conducive to decay. Colonization is the term used to describe the initial invasion and occupancy as fungi and other microorganisms invade and utilize wood components (Levy, 1975, Rayner and Todd, 1979, 1982; Rayner and Boddy, 1988; Swift, 1977, 1984). These organisms create a continuum of changing conditions from the wood surface inward, both radially and longitudinally. The zones of changing conditions created in the wood due to microbial action and related abiotic factors are called *niches*. A *niche* might be defined as the physical and biological conditions of a zone in the wood favoring the development of a species or its population. Microorganisms, by the process of natural selection, have adapted to compete for the special nutritional and ecological conditions of a given niche.

The number of individuals of the same species occupying a niche is termed the *population*. Populations of different organisms living and interacting at the same time in the many niches comprising the habitat or substrate, make up a *community* (e.g., fallen log, stump, pole, etc.). The term substrate is preferred in the case of wood decay where the non-living wood is primarily a nutrient source and does not respond to colonization. The progressive change in species composition and frequency occurring over time in the various communities of the habitat is termed *succession*. The concept of succession usually implies a predictable direction in community development. Among some higher plant and animal communities, succession may lead to rather stable assemblages called climax communities. In contrast, colonization and succession events during wood decomposition lead to eventual destruction and in some cases, total mineralization. Colonization and succession patterns in wood products remain poorly understood primarily because of the difficulty of studying organisms in an opaque material. The process appears to be more erratic and varies considerably with the abundance of inoculum

available at the time of initial exposure, the wood used, and the presence of any chemical treatment. Emerging genetic sequencing techniques that allow a majority of the genetic material in decaying wood to be categorized will eventually help to unravel these mysteries, although interpreting the results of early studies remains problematic.

Severe competition for the available nutrient resource can develop when a new wood substrate becomes available for microbial colonization and abiotic conditions are favorable (i.e. moisture content and temperature), Interactions among competing species range from exclusion to cooperation and co-existence. The conflict with small organisms such as bacteria tends to be among populations, whereas competition may be between the thalli of individuals in larger organisms such as fungi. A complex terminology has developed to describe and categorize the many possible interactions. As early as 1879, Anton deBary (the so-called father of Plant Pathology) introduced the term symbiosis to describe, in a broad sense, any close interspecific associations between two organisms.

Initially, symbiotic relationships were separated into those that were beneficial (mutualistic symbiosis) or harmful (parasitic symbiosis). Classic examples of mutualistic symbiosis that received major attention were the lichens and mycorrhiza, where fungi form beneficial associations with algae and plant roots, respectively. Studies of the many interactions among the higher plants and animals, new insights into the complexities of ruminant and termite digestion, and the discovery of antibiotics greatly expanded the categories needed to describe the wide range of interactions. The term symbiosis is now narrowly defined by some microbiologists to describe primarily beneficial associations (Salle, 1961; Brock et al., 1984).

The possible first order interactions of two microorganisms competing for the same niche in wood are illustrated in Table 11.1.

It is important to realize that the effects of the participants on each other are judgmental, so controversy can occur about categorizing interactions.

Types of microbial interactions in wood

The principal categories of microbial interaction of direct relevance to wood microbiology include:

(a) *Mutualism*: both organisms benefit. For example, *Amylostereum chailetti*, a decay fungus, is vectored by a horntail (*Sirex cyaneus*) during oviposition into the dying sapwood of fire killed or defoliated balsam fir. The fungus benefits because the insect places it in an ideal position to invade

Table 11.1 An illustration of the types of first order interactions that may occur between two microorganisms competing for the same niche during the decay of wood.

		Effects on species 1 when it competes with species 2		
		No effect	Harmful	Beneficial
Effect on species 2 when it competes with species 1	No effect	0, 0[a] Neutralism	−, 0 Amensalism (Antibiosis)	+, 0 Commensalism
	Harmful	−, 0 Amensalism (Antibiosis)	−, − Competition	−, + Predation and Parasitism
	Beneficial	+, 0 Commensalism	+, − Predation or Parasitism	+. + Mutualism

[a]Where 0 = no effect, + = enhanced effect and − = negative effects. Species 1, right and species 2, left.

the wood before the arrival of competing fungi. The horntail larvae benefit by ingesting fungal mycelium and acquiring the cellulase enzymes needed for softening and digesting the wood (Slippers et al., 2012; Kukor and Martin, 1983).

(b) *Commensalism*: one is benefited and the other unaffected. In the later decay stages, many brown rot fungi release more simple carbon compounds than they use or store. Some secondary scavenger fungi such as *Trichoderma* spp. or *Mucor* spp. invade in the late decay stages and use the excess sugars.

(c) *Amensalism* (antibiosis): the toxic products of one organism inhibit or retard the growth of another. Prior colonization of logs by *Trichoderma* spp. has been shown to inhibit subsequent basidiomycete colonization (Hulme and Shields, 1975). A microfungus, *Scytalidium* sp., commonly present in seasoning Douglas-fir utility poles was reported to produce an antibiotic that delayed colonization by *Antrodia carbonica* (Ricard and Bollen, 1968). This antibiotic association was proposed as a potential biological control approach to reduce decay development in utility poles (Ricard, 1976, Morris et al., 1984, Bruce and King, 1986). Highly variable performance of the *Scytalidium* made this approach infeasible. Some wood-inhabiting bacteria have been shown to produce toxic metabolites that inhibit the growth of white, brown, and soft rot fungi in laboratory tests (Preston et al., 1982).

(d) *Parasitism*: one organism obtains its nutrition directly from another and the relationship is generally harmful to one. Some fungi are pathogens of other fungi and insects. Some white-rot fungi attack and utilize other wood inhabiting fungi, apparently as a nitrogen source (Rayner et al., 1987). Such fungi are called mycopathogens and are of special interest as possible biological control agents.

(e) *Predation*: one organism physically captures and consumes part or all of another. Usually the predator is larger than the prey, but some fungi trap nematodes using hyphal coils and digest them as a food source (Thorn and Barron, 1984; Stirling, 1988).

(f) *Neutralism*: the organisms occur together with no discernible effect on each other. Bacteria are commonly associated with the gelatinous coating on the outer hyphal wall of some soft-rot fungi. It has been suggested that these bacteria may interfere with the feed-back inhibition of cellulase and accelerate decay. It is generally difficult to prove that organisms do not interact or affect one another.

There are many other interesting types of interactions between organisms in wood during the decay process. *Synergism* occurs when the combined effects (growth, decay rate, etc.) of both organisms are greater than either alone. The combination of some yeasts and decay fungi has been shown to greatly accelerate decay rates (Blanchette and Shaw, 1978). Co-metabolism occurs when one organism modifies a complex substrate, such as a paint polymer, permitting its subsequent digestion by another. Neither organism can completely utilize the substrate alone.

Common wood inhabitants during decay

While fungi play a dominant role in decay development, many other microorganisms are often present and may affect the fungi. They include the bacteria, protozoa, algae and small animals such as nematodes, mites, and insects. Their roles in decay development range from significant to minor and, in many cases, are unknown. Insects are important vectors of many stain and decay fungi. Some bark beetles carry nitrogen-fixing bacteria that can enhance the nutritional quality of the wood (Bridges, 1981). Bacteria are also emerging as constant associates of some fungi and may play a role in certain physiological activities such as fruiting. Other insects harbor fungi that grow on tunnel walls and are

consumed by grazing larvae. By tunneling and comminution, insects and other microfauna increase both the aeration and accessibility of the wood to other microorganisms. These roles are common in the later decay stages of logs and stumps in the forest.

Bacteria are ubiquitous in decaying wood and an important food source for protozoa and nematodes. Predator/prey relationships occur among the microfauna in the later decay stages. Mycorrhizal roots of forest trees are often found growing inside decayed wood on the forest floor. Decayed wood is a favorable site for nitrogen fixation by bacteria and an important nitrogen source for the invading mycorrhizal feeder roots (Larsen et al., 1982).

Microecology of wood decay

It is clear that many organisms directly and indirectly interact during the decay process. However, the microecological aspects of wood decay remain poorly understood and their study has lagged far behind ecological studies of higher plant and animal systems. A major reason is the difficulty of measuring decay development and the microorganisms involved in a non-destructive fashion. The decay process occurs over time periods often measured in years in an essentially opaque material that usually must be destructively sampled for analysis. Early studies of the sequences of fungi associated with decay relied primarily on the presence of basidiomycete fruiting structures to determine identities and changes in species composition (Spaulding and Hansbrough, 1944). Such studies tended to favor the basidiomycetes that were better known taxonomically at the time and neglected less visible organisms, particularly the ascomycetes. The appearances of fruiting bodies can simply reflect differences in maturation times and have little or no relationship to when the fungi colonized the material or their impact on the substrate.

The use of cultural characteristics (Campbell, 1938; Davidson et al., 1938; Nobles, 1948; Stalpers, 1978) was an effective way to identify isolated fungi from their vegetative or cultural characteristics. This was an important step in setting the stage for progress in fungal ecology.

Further developments in systematic sampling (Butcher, 1971; Sharp, 1974; Sharp and Levy, 1973) and improved isolation procedures, coupled with advances in cultural taxonomic data have improved our ability to define the identities (or taxa descriptions) and relative abundance of the organisms associated with stages of the decay process. More recently

developed DNA sequencing methods have accelerated the identification process and reduced the need for highly specialized taxonomic specialists to identify fungi. Emerging methods for directly sequencing fungal DNA from wood without the need to isolate fungi on specialized media will further expand our abilities to better understand the ecology of the decay process. Each of these methods provides us with a different view of the complex interactions occurring within decaying wood.

Laboratory tests can be used to determine the decay capability and type, as well as the antibiotic, or mycoparasitic abilities of the isolates. Nevertheless, determining the relative role and importance of individual species in the decay process remains difficult because of the complexity of the system. There is always the underlying question of whether the laboratory results, often from axenic tests under idealized conditions, are relevant to decay events in the natural system where organisms are competing for a substrate.

Colonization strategies

A useful approach for understanding the roles and interactions of microorganisms during the decay process is to consider the characteristics or strategies used to occupy a niche, retain it, or in some cases, capture it from a competing microorganism.

The major microorganism types present at various stages of decay development and their roles can often be traced to nutritional capabilities. Some microorganisms can only utilize the relatively simple carbon compounds that occur in untreated, sound wood. Some can utilize hemicelluloses and cellulose, but not lignified cell wall material. Some, such as the white-rot fungi utilize all cell-wall components.

Rayner and Boddy (1988) have proposed three types of wood-inhabiting fungi to explain their appearances and sequences in colonization. These types are listed and discussed with examples as follows:

Opportunists: These fungi are scavengers and appear to have adapted to rapidly exploit a new wood substrate (e.g., broken branches, felled trees, freshly sawed boards, checks in seasoning poles, untreated sapwood, etc.). They are analogous to the pioneer species in the colonization of disturbed soil sites by plants. Opportunists are characterized by rapid growth rates and prolific production of air borne spores. A few examples are: (a) the molds and sapstain fungi that rapidly invade freshly sawn lumber, (b) the bacteria and microfungi that initially colonize

stem wounds, (c) some decay fungi in wounded stems, slash, stumps and wood products (e.g. *Schizophyllum commune*, *Sterum sanguinolentum*, *Irpex lacteus* , and *Phanerochaete* (*Peniophora*) *gigantean)*.

Stress resistors: These fungi have adapted to adverse growth conditions such as the presence of toxicants or allopathic chemicals, low aeration levels, minimal moisture levels, and high temperatures. A few examples are: (a) the molds and soft-rot fungi that tolerate high levels of some toxicants, (b) the heartrot fungi that tolerate the low aeration and allopathetic environment of some living stems, (c) the thermotolerant and thermophillic fungi and bacteria that survive in wood chip piles, (d) the heartrot fungi with cryptic invasion strategies that may remain dormant for years in the sapwood of small dead branches, (e) the bacteria that invade ponded logs where low oxygen levels limit fungi.

Combatants: These fungi are able to repel invaders or take the niches of other fungi. A few examples are: (a) fungi that produce antibiotics that may repel or assist in wood invasion (Etheridge, 1961) and (b) fungi that are mycopathogens and parasitize other wood-inhabitants (Barnett, 1964). For example, *Trametes* (*Lenzites) betulina* parasitizes and replaces some other decay fungi (Rayner et al., 1987).

Many wood inhabiting microorganisms overlap these categories or their strategies remain unknown. However, these categories are useful for understanding the appearances, sequences, and interactions of microorganisms involved in wood colonization and decay development. Wood colonization patterns vary with wood types, uses, and exposure conditions. Common colonization patterns are discussed in the next section.

Colonization patterns of wood by fungi

A variety of microbial sequences are reported for various wood products or wood under various exposures. These studies were related primarily to the salvage of trees killed by disease, insects, fire, or windthrow, the storage of pulpwood and logs, the decomposition rates of slash and branches under various silvicultural practices, branch stubs, and the service lives of treated posts and poles. Shigo assembled a comprehensive review of the early literature on the types and sequences of microorganisms occurring during the discoloration and decay of wood with emphasis on wounded living trees and diseased or dead standing trees (Shigo, 1967). Kaarik (1974, 1975) also summarized the research on fungal successions in various wood substrata with emphasis on slash, logs, stumps, or posts in soil contact.

These studies are now summarized for a range of wood uses and conditions. The topic is discussed further in the subsequent chapters on stem decays, wood storage problems, sapstains, and decays in the major wood products.

Standing tree: Wood in the living tree represents a hostile environment for most microorganisms because the tree can respond, but when branches and roots die or wounds occur, some organisms invade the roots, stem, or branches (Shigo, 1972; Manion, 1975; Good and Nelson, 1962). Heartrot fungi are believed to invade the central non-living portions of the stem through wounds or dead branch stubs. These fungi are examples of stress-tolerant organisms because they can survive in a low oxygen environment that may also contain toxic extractives. Sapwood decays originate from wounds that destroy the protective bark and reduce the moisture content of the exposed sapwood. Extensive studies of microbial sequences in stem wounds and decay development were made by Shigo and associates (Shigo, 1967, 1972) (see Chapter 12). Many decays in the outer sapwood become heartrots as they are later enveloped by continued growth in girth. A progression from bacteria to microfungi to decay fungi represents the proposed succession of microorganisms invading stem wounds of living trees.

Standing dying or dead trees: Large volumes of standing timber become susceptible to decay when defoliated, subjected to fire, or killed by insects such as the bark beetles, gypsy moths or spruce budworm. The sequences of fungi invading stems have been determined in studies to identify salvage schedules timed to remove trees before substantial degradation makes them less valuable. Basham (1959) proposed a sequence of microfungi, a weak decayer *Amylostereum chailletii*, and finally a severe decayer, *Hirchioporus abietineus* in spruce budworm-killed balsam fir in eastern Canada. Warren (1989) suggested that the appearances of these two decayers was independent, with *A. chailletii* vectored by a Sirex wood wasp and the *H. abietineus* invasion related to subsequent severe bark beetle damage to the stem. Understanding the sources of fungi in standing trees can be important since some of these organisms can continue to survive into the final product.

Seasoning lumber : Freshly sawn and stacked, untreated lumber represents a rich nutrient source that is suddenly unprotected and available to the first pioneer microorganisms that can invade and exploit the storage materials in the parenchyma cells. Sapstain, is a serious source of lumber degrade for many natural finish wood uses that can develop within a week of the sawing in the absence of proper piling and protective dip

treatments. A few stain fungi such as *Ophiostoma* (*Ceratocystis) picea* may eventually also produce soft rot upon prolonged exposure. If the lumber is excessively wet, molds in genera such as *Rhizopus*, *Penicillium*, *Aspergillus*, or *Trichoderma* are favored. These fungi can be precursors to invasion by wood decay fungi, but they can also create issues because wood users are concerned about the possible risk of mold spores on lumber. When lumber drying is restricted or in cases of bulk piling, the stain fungi may be followed by brown rot decayers such as *Rhodonia placenta*. This often-observed sequence of stainers followed by decayers may not reflect a colonization pattern, but rather differing growth and mycelial development rates. Sapstains are discussed later in Chapter 14.

Untreated wood in ground contact: Merrill and French (1966) reported the major initial colonizers of buried wood were the microfungi *Fusarium solani* and *Trichoderma viride*, followed by the appearance of soft rot within 6 weeks and pockets of brown rot after 10 weeks. *Sistotrema* (*Trechiospora*) *brinkmanni*, a brown-rot fungus, was isolated after 12 weeks. The generalized sequence in this and related studies suggests an initial invasion by some microfungi acting as aggressive pioneers, followed by soft-rotters and later basidiomycete decayers. Torres (2017) found few basidiomycetes and an abundance of Ascomycetes over a 24 month period in red alder, western redcedar and Douglas-fir lumber using classic isolation methods. However, she found much higher basidiomycete levels when high throughput sequencing was used to assess all of the recoverable DNA in the same samples. These results suggest that we need to carefully evaluate our notions about microbial sequences in the decay process in soil contact using all available methods to determine the organisms that are present and their possible roles.

Corbett and Levy (1963) reported a similar colonization pattern of the fast growing species with minimal effects on the wood (*Trichoderma* spp., *Penicillium* spp., etc.), followed by Sphaeropsidales (including some soft-rot fungi), and finally basidiomycetes in the development of decay in untreated woods (*Pinus* and *Betula*) in soil contact.

In 1968, Butcher studied fungal successions over a 13-month exposure period in *Pinus radiata* sapwood stakes in New Zealand. He reported a successional sequence of blue stain to molds above ground and a pattern similar to that shown by Merrill and French (1966) and Corbett and Levy (1963) below the groundline.

Kaarik (1968) found that fungal isolations varied substantially with soil type and with position below and above the ground in studies of fungal colonization of pine and spruce poles in Sweden over a four-year period.

Many different fungal species were isolated from many zones in the poles, suggesting random initial invasions and severe competition to determine survivors in the succession. In agreement with the other studies, the major decay fungi appeared in wood already occupied by the non-decay fungi and competed effectively with them.

In a 10-year study of the succession of microorganisms into posts of birch and Scotch pine in England, Banerjee and Levy (1971) reported the fungal sequences at the soil line to be sapstain fungi with soft rot characteristics followed by basidiomycetes.

Treated wood in ground contact: The presence of preservative substantially slows and alters microbial colonization and tends to select more chemically tolerant species (Greaves and Savory, 1965, Henningsson and Nilsson, 1976, Nilsson and Henningsson, 1978, Sorkhoh and Dickinson, 1975).

Studies on the sequences and prevalence of fungi isolated from a series of preservative treated poles representing a wide range of service ages indicated that the probable invasion sequence of fungi at the groundline began with microfungi and soft rotters followed by primarily white-rot fungi. The incidence of microfungi increased in the older pole age groups. The initial invading fungi were tolerant to wood preservatives and some tolerant fungi were associated with specific preservative treatments, e.g. *Cladosporium resinae* from creosote-treated poles and *Rhinocladiella atrovirens* from penta-treated poles (Zabel et al., 1985).

Other wood conditions: While there are numerous reports concerning the major decay fungi associated with many important wood products there are few studies of the sequences of fungi that invade treated wood in its many above-ground uses. Freitag et al. (1995) found basidiomycetes slowly invaded mortise and tenon joints in a tropical exposure even though untreated pine joints failed within 2–3 years. The more limited above ground microflora likely slows the process even under ideal conditions for decay development. Studies on microorganism sequences in chip piles involving bacteria and thermos-tolerant fungi are discussed in Chapter 13 on Wood Storage Losses.

Succession in wood decay

Microbial succession during the decay process, although often postulated, is difficult to unequivocally establish due to the uncertainty of reliably isolating all the principal microorganisms involved. Important

organisms may be missed because of the isolation procedures, types of selective media used, the absence of a critical associate, or even the conditions employed for incubation of the isolated samples. Even more recently developed DNA sequencing techniques can be misleading because of the potential for heavily sporulating fungi to dominate and the fact that the technique cannot discern between active vs inactive organisms.

The thallus character and indeterminate growth pattern of many decay fungi presents difficulties in comparative enumeration. Prevalence of a single species may simply reflect greater reproductive potential. Isolation frequency can be very misleading since a single fungus mycelium may dominate several cubic meters of decaying wood yet appear minor if compared numerically to the more frequent isolations of prolific spore producing scavenger fungi or bacteria. The difficulty of properly identifying each isolate and the tendency to study the roles of the known have also stymied ecological studies of the decay process. The wood-inhabiting bacteria and ascomycetes are often difficult to identify and hence have been subject to notorious neglect. Important organisms in the various decay processes of wood may still lurk in the unidentified isolates of many earlier studies. As a brief aside, these are some of the reasons why the microbial ecology of wood inhabiting microorganisms remains largely in a descriptive stage compared to the highly developed quantitative and statistical approaches to population dynamics and understandings of succession and climax communities in the higher plants and animals.

Successions of microorganisms involved in the decay of various wood materials are exceedingly complex and variable events. It is probable that variations in inoculum sources, substrate changes by prior inhabitants, and changes in abiotic conditions such as wood aeration or moisture all influence the decay process. There are several problems with using the term succession to describe the sequences of microorganisms observed in the initial decay of wood. First, as mentioned earlier, succession as used in higher plant and animal ecology, implies the steady state condition of communities at the end of a predictable series of changes. With few exceptions, most claims of succession in the wood decay process start with sound wood and end with the appearance of decay, or when the practical use of the wood product under study is over. Thus, the process being studied is really early colonization. The continuing later stages of decay are rarely studied either in wood products or wood under natural conditions such as decaying logs. Observations of advanced decay in the forest floor or structures indicate that the microfauna play a significant

comminuting role. Studies of soil also show that humus development is traceable to recalcitrant lignin and microbial residues that may remain in soil for decades and represent significant carbon reservoirs in the biosphere. Bacteria probably play a key role in the final decomposition of the lignin residues originating from wood. Complete mineralization of wood represents the end of the decay process from the ecological viewpoint. Second, there is uncertainty about the actual causes of the changes in fungal communities reported in the early stages of decay development. The successional concept implies that sequences of communities modify the substrate to more favorable conditions for subsequent communities. Uncertainty exists about whether the community changes observed in early decay sequences are the cause or simply a direct reflection of changing abiotic conditions under seasonal control such as moisture content, temperature, or even the phenology of the microorganisms involved.

Research needs on ecology of decay fungi

Our present limited understanding of organism types, roles, interactions and sequences (succession) in the decay process of major wood materials and products stresses the need for a new emphasis in research on the ecology of decay fungi. On a broader base, an increased role by mycologists in ecological research and theory development is important to more clearly define the decomposer role of fungi in the biosphere. Efforts to examine the potential for carbon sequestration as part of plans to use forests and forest products as part of an overall strategy to combat rising carbon dioxide levels will only increase the importance of decomposer fungi.

There are also immediate and practical applications for information on the sequences and interactions of the microorganisms involved in decay development. Such information is useful to the wood microbiologist and wood products engineers concerned with controlling decay. Important questions to the wood microbiologist about the development of decay in various wood forms or products include the identities and numbers of the microorganisms involved (who); their time of appearance (when); locations (where); and roles (why).

Answers to these questions can lead to improved decay prevention practices for some wood uses or suggest decay vulnerable locations or periods in other wood uses.

Summary

1. Colonization is the term used to describe the invasion and occupancy of wood by microorganisms.
2. Wood-inhabiting microorganisms have adapted to compete for the various nutritional and ecological zones in the wood termed niches.
3. Competition develops among wood-inhabiting microorganisms for the available nutrients when a new wood substrate becomes available and growth conditions are favorable.
4. Microbial interactions between competing wood-inhabiting microorganisms include mutualism (both benefit), commensalism (one benefits, one is unaffected), amensalism (one poisons or chemically repels the other), parasitism (a harmful relationship where one obtains nutrients from the other), predation (one captures and consumes another), and neutralism (no discernible effects).
5. Common wood inhabitants during decay development in addition to fungi are bacteria, protozoa, algae and small animals such as nematodes, mites, and insects.
6. Colonization strategies are a useful way to explain the appearances and sequences of microorganisms that invade wood. Opportunists are the pioneers (scavengers) that have adapted to rapidly exploit a new wood substrate; stress-resistors are able to withstand adverse growth conditions or tolerate poisons and alleopathic chemicals; combatants are able to repel invaders or capture the niches of other fungi with antibiotics or by acting as mycopathogens.
7. The sequences of microorganisms often reported in the colonization of untreated and treated wood in ground contact are microfungi, soft-rot fungi, and basidiomycete decayers.
8. Microbial succession during the decay process, though often claimed, is difficult to prove because of the uncertainty of reliably isolating, identifying, quantifying, and determining the interactive roles of the many microorganisms often involved. Emerging DNA and RNA techniques offer great promise for unraveling these sequences.
9. Additional research on the microbial ecology of the decay process may lead to future effective biological controls of decay and provide useful information to the wood microbiologist about when and where various wood products are most vulnerable to attack *by* destructive biotic agents.

References

Atlas, R.M., Bartha, R., 1998. Microbial Ecology Fundamentals and Applications. Benjamin/Cummings Publ. Co., Menlo Park, CA, p. 533.

Banerjee, A.K., Levy, J.F., 1971. Fungal succession in wooden fence posts. Mater. Org. 6, 1–25.

Barnett, H.L., 1964. Mycoparasitism. Mycologia 56, 1–19.

Basham, J.T., 1959. Studies in forest pathology II. Investigations of the pathological deterioration of killed Balsam fir. Can. J. Bot. 39, 291–326.

Blanchette, R.A., Shaw, C.G., 1978. Associations among bacteria, yeasts, and basidiomycetes during wood decay. Phytopathology 68, 631–637.

Bridges, J.R., 1981. Nitrogen-fixing bacteria associated with bark beetles. Microb. Ecol. 7, 131–137.

Brock, T.O., Smith, O.W., Madigan, M.T., 1984. Biology of Microorganisms, fourth ed. Prentice-Hall, Inc, Englewood Cliffs, NJ, p. 847.

Bruce, A., King, B., 1986. Biological control of decay in creosote treated distribution poles. II. Control of decay in poles *by* immunizing commensal fungi. Mat. Org. 21, 165–179.

Butcher, J.A., 1968. The ecology of fungi infected untreated sapwood *of Pinus radiata*. Can. J. Bot. 46, 1577–1589.

Butcher, J.A., 1971. Techniques for the analysis of fungal floras in wood. Mater. Org. 6, 209–232.

Campbell, W.A., 1938. The cultural characteristics of the species of *Fomes*. Bull. Torrey Bot. Club 65, 31–69.

Campbell, R., 1983. Microbial Ecology, second ed. Blackwell Scientific Pub, Oxford, London, p. 191.

Corbett, N.H., Levy, J.F., 1963. Ecological studies on fungi associated with wooden fence posts. Nature Lond. 198, 1322–1323.

Davidson, R.W., Campbell, W.A., Blaisdell, D.J., 1938. Differentiation of wood-decaying fungi by their reactions on gallic or tannic acid medium. J. Agr. Res. 57, 683–695.

Etheridge, D.E., 1961. Factors affecting branch infection in aspen. Can. J. Bot. 39, 799–816.

Freitag, C.M., Morrell, J.J., Archer, K.J., 1995. Colonization of Treated and Untreated Ponderosa Pine Exposed in Hilo, Hawaii. International Research Group on Wood Preservation, Stockholm, Sweden, IRG/WP/95-20068 11 pages.

Good, H.M., Nelson, C.D., 1962. Fungi associated with *Fomes igniarius* var. *populinus* in living poplar trees and their probable significance in decay. Can. J. Bot. 40, 615–624.

Greaves, H., Savory, J.G., 1965. Studies of the microfungi attacking preservative-treated timber, with particular reference to methods for their isolation. J. Inst. Wood Sci. 15 (1965), 45–50.

Henningsson, B., Nilsson, T., 1976. Some aspects on microflora and the decomposition of preservative-treated wood in ground contact. Org. Holz. Int. Symp. Berlin-Dahlem (1975) Mat. Org 3, 307–318.

Hulme, M.A., Shields, J.K., 1975. Antagonistic and synergistic effects for biological control of decay. In: Liese, W. (Ed.), Biological Transformation of Wood by Microorganisms. Springer-Verlag, New York, pp. 52–63.

Kaarik, A., 1968. Colonization of pine and spruce poles by soil fungi after twelve and eighteen months. Mat. Org. 3, 185–198.

Kaarik, A., 1974. Decomposition of wood. In: Dickinson, H., Pugh, G.J. (Eds.), Biology of Plant Litter Decomposition., vol. 1. Academic Press, London, pp. 129–174.

Kaarik, A., 1975. Successions of microorganisms during wood decay. In: Liese, W. (Ed.), Biological Transformation of Wood by Microorganisms. Springer-Verlag, New York, pp. 39–51.

Kukor, J.J., Martin, M.M., 1983. Acquisition of digestive enzymes by siricid wood wasps from their fungal symbiont. Science 220, 1161–1163.
Larsen, M.J., Jurgensen, M.F., Harvey, A.E., 1982. N_2 fixation in brown-rotted soil wood in an intermountain cedar-hemlock ecosystem. For. Sci. 28 (2), 292–296.
Levy, J.F., 1975. Colonization of wood by fungi. In: Liese, W. (Ed.), Biological Transformation of Wood by Microorganisms. Springer-Verlag, New York, pp. 16–23.
Manion, P.O., 1975. Two infection sites of *Hypoxylon mammatum* in trembling aspen (*Populus tremuloides*). Can. J. Bot. 53 (22), 2621–2624.
Merrill, W., French, D.W., 1966. Colonization of wood by soil fungi. Phytopathology 56, 301–303.
Morris, P.I., Dickinson, D.J., Levy, J.F., 1984. The nature and control of decay in creosoted electricity poles. Br. Wood Preserv. Assoc. Annu. Conv. 42–53.
Nilsson, T., Henningsson, B., 1978. *Phialophora* species occurring in preservative-treated wood in ground contact. Mat. Org. 13 (40), 297–313.
Nobles, M.K., 1948. Studies in forest pathology. IV. Identification of cultures of wood-rotting fungi. Can. J. Res. C, Bot. Sci. 26, 281–431.
Preston, A.F., Erbisch, F.H., Kramm, K.R., Lund, A.E., 1982. Developments in the use of biological control for wood preservation. Proc. Am. Wood Preserv. Assoc. 78, 53–61.
Pugh, G.J.F., 1980. Strategies in fungal ecology. Trans. Br. Mycol. Soc. 75, 1–14.
Rayner, A.D.M., Todd, N.K., 1979. Population and community structure and dynamics of fungi in decaying wood. Adv. Bot. Res. 7, 333–420.
Rayner, A.D.M., Todd, N.K., 1982. Population structure in wood-decomposing basidiomycetes. In: Frankland, J.E., Hedger, J.N., Swift, M.J. (Eds.), Decomposer Basidiomycetes: Their Biology and Ecology. Cambridge University Press, Cambridge, pp. 109–128.
Rayner, A.D.M., Boddy, L., 1988. Fungal communities in the decay of wood. In: Marshall, K.C. (Ed.), Advances in Microbiological Ecology, vol. 10. Plenum Press, New York, pp. 115–166.
Rayner, A.D.M., Boddy, L., Dowson, G.G., 1987. Temporary parasitism of *Coriolus* spp. by *Lenzites betulina*: a strategy for domain capture -in wood decay fungi. FEMS Microbiol. Ecol. 45, 53–58.
Ricard, J.L., 1976. Biological control of decay in standing creosote treated poles. J. Inst. Wood Sci. 7, 6–9.
Ricard, J.L., Bollen, W.B., 1968. Inhibition of *Poria carbonica* by *Scytalidium* sp., an imperfect fungus isolated from Douglas-fir poles. Can. J. Bot. 46, 643–647.
Salle, A.J., 1961. Fundamental Principles of Bacteriology. McGraw Hill Book Co., Inc., p. 812.
Sharp, R.F., 1974. Some nitrogen considerations of wood ecology and preservation. Can. J. Microb. 20, 321–328.
Sharp, R.F., Levy, J.F., 1973. The isolation and ecology of some wood colonizing microfungi using a perfusion culture technique. Mat. Org. 8 (3), 189–213.
Shigo, A.L., 1967. Successions of organisms in discoloration and decay of wood. Int. Rev. For. Res. 2, 237–299.
Shigo, A.L., 1972. Successions of microorganisms and patterns of discoloration and decay after wounding in red oak and white oak. Phytopathology 62 (2), 256–259.
Slippers, B., DeGroot, P., Wingfield, M.J., 2012. The *Sirex* Woodwasp and its Fungal Symbiont. Springer, London, p. 301.
Sorkhoh, N.A., Dickinson, D.J., 1975. The effects of wood preservative on the colonization and decay of wood by microorganisms. Org. Holz. Int. Symp. Berlin-Dahlen 1975 Mat. Org 3, 287–293.
Spaulding, P., Hansbrough, J.R., 1944. Decay of Logging Slash in the Northeast. Tech. Bull. U.S. Dept. Agric. 876, 22 pp.

Stalpers, J.A., 1978. Identification of wood-inhabiting fungi in pure culture. Centra'ibureau voor Schimmelcultures, Baarn. Stud. Mycol. 16, 1–248.
Stirling, G.R., 1988. Prospects for the use of fungi in nematode control. In: Burge, M.N. (Ed.), Fungi in Biological Control Systems. Manchester University Press, New York, NY, p. 269.
Swift, M.J., 1977. The ecology of wood decomposition. Sci. Prog. Oxf. 64, 175–199.
Swift, M.J., 1984. Microbial diversity and decomposer niches. In: Klug, M.J., Reddy, C.A. (Eds.), Current Perspectives in Microbial Ecology. American Society for Microbiology.
Thorn, R.G., Barron, G.L., 1984. Carnivorous mushrooms. Science 224, 76–78.
Torres, P.A., 2017. Characterization of Fungal Communities in Selected Wood Species by Field Exposure Tests (PhD. Dissertation). Oregon State University, Corvallis, OR, 253 p.
Warren, G.R., 1989. The Identities, Roles, and Interactions of Tree Fungi Associated With Stem Biodeterioration of Balsam Fir Stressed and Killed After Spruce Budworm Defoliation in Newfoundland (Ph.D. thesis). State University of New York, College of Environmental Science and Forestry, Syracuse, NY.
Zabel, R.A., Lombard, F.F., Wang, C.J.K., Terracina, F., 1985. Fungi associated with decay in treated southern pine utility poles in the eastern United States. Wood Fiber Sci. 17 (1), 75–91.

CHAPTER TWELVE

Decays originating in the stems of living trees

Contents

While this textbook is primarily about the decays of wood products, a general understanding of the features and origins of the many decays that develop in living trees is important to wood users, wood scientists, and wood engineers for several reasons.

We emphasize, initially, that some of the decays and discolorations that appear commonly in lumber and other wood products in use have their origins in living stems. Undetected incipient decay helps to explain some of the high variability experienced in some wood uses. It is particularly important to detect such decay in roundwood and lumber destined for critical strength uses such as poles, piling, and structural lumber as early as possible. The physiological and structural responses in the living stem following injury and subsequent decay development can reduce

Wood Microbiology.
DOI: https://doi.org/10.1016/B978-0-12-819465-2.00012-7

preservative treatability and increase decay susceptibility in products. Information on the mechanisms by which trees resist invading wood-decay fungi may yield clues toward more effective preservatives and decay prevention practices during the storage, conversion and use of wood.

Descriptions and general information on the major decays of standing timber are available in various timber decay and forest pathology textbooks (Hubert, 1931; Cartwright and Findlay, 1958; Boyce, 1961; Sinclair et al., 1987; Manion, 1991; Tainter and Baker, 1996) and information on their identities and prevalence in the major timber species in North America was compiled by Hepting (1971). Descriptions of most of the causal fungi are available in taxonomic studies by Gilbertson and Ryvarden (1986/1987), although the taxonomy has changed dramatically. Monographs on heart rots in living trees have been prepared by Wagener and Davidson (1954) and Shigo and Hillis (1973).

In this chapter, a special emphasis will be placed on types, origins, and developmental patterns of stem decays that are important for the detection and understanding of their various effects on many wood use properties.

Historical highlights

As indicated in Chapter 1, forest pathology had its origin in the study of stem decays. In 1884, R. Hartig of Germany first showed that a specific fungus caused a specific decay and a predictable amount of cull. Forest pathology research began in the United States around the 1900 in response to growing fears of a timber famine and deep concerns about extensive decay damage developing in railway ties and other wood construction (Williams, 1989). For these reasons, the study of stem decays was a major concern of the early forest pathologists (von Schrenk, 1900; Weir and Hubert, 1918; Boyce, 1920, 1932). Emphasis was placed on estimating the extent of internal decay from the locations of external indicators such as basidiomata and decayed knots (Hubert, 1924). Cull-detection manuals were developed for most of the major timber producing regions to provide foresters with information on the principal decay indicators and associated cull (Murphy and Rowatt, 1932; Hepting and Hedgcock, 1937; Silverborg, 1954; Lockhard et al., 1950; Roth, 1959; Shigo and Larson, 1969; Kimmey and Hornebrook, 1952; Partridge and Miller, 1974).

Other important studies addressed rates of decay development and pathologic rotations (when decay development begins to exceed annual growth) for important timber species. The effects of various important decays on wood uses and strength properties were determined for some principal timber species (Scheffer et al., 1941 Stillinger, 1951; Atwell, 1948). Extensive studies also demonstrated the close association between fire scars and decay development in southern hardwoods (Hepting, 1935).

More recent stem-decay studies have focused on the infection process (Haddow, 1938; Etheridge and Craig, 1976; Manion, 1967; Manion and French, 1967; Merrill, 1970). Contributions toward understanding the types and sequences of fungi involved in decay initiation were made by Davidson and Redmond (1957); Basham (1958); Good (1959), Shigo (1967, 1976, 1984) and, more recently, Rayner and Boddy (1988).

Prevention practices for stem decays have been indirect and largely associated with silvicultural operations because of cost limitations. Some of the measures recommended have been sanitation cuttings, early branch pruning (before heartwood forms), sound prevention practices, fire control, and silvicultural treatments favoring rapid growth and the natural pruning of small branches. Restricting the amount of times heavy logging equipment enters a stand can also reduce the risk of wounding.

Stem decay types

There are some important differences between the decays that develop in living stems (pathogenic decays) and wood products (saprobic decays). Decay development in the living stem is a disease and the tree often is able to respond to the initial infection and defend itself. Some tree (host) responses provide useful evidences (symptoms) of the internal decay. The outer sapwood in the living tree is resistant to decay, and the heartwood less so, while heartwood in the wood product is often resistant and the sapwood universally susceptible.

Stem decays can be grouped conveniently into heart rots and sap rots based on the principal stem zones invaded and destroyed. Heart rots are the predominant type of stem decay in mature forests and occupy primarily the physiologically inactive central portion of the stem (heartwood).

The sap rots generally develop in the outer sapwood after injury and death of localized bark and xylem tissues.

These positional distinctions are not clearcut and may intergrade in the later stages of decay development. We will see later that many heart rots in the central zone of stems originated as localized sap rots around wounds that were later enveloped by the radial growth of the tree. Some heart-rot fungi such as *Phellinus* (*Fomes) pini* and *Ionotus* (*Poria*) *obliquus* may invade and kill the outer living sapwood and form cankers.

Stem decays can be also subdivided into those that occupy the top, central, or basal (butt) portions of the tree trunk. Some stem decays are localized, while others may occupy entire heartwood zones from the basal roots to the larger branches. While both brown and white rots are important in standing timber, white rots are more common (Gilbertson, 1980).

Stem decay origins

The manner in which decay fungi enter xylem tissues in living stems still remains a perplexing and intriguing question which is difficult to study and answer because of the long infection periods involved, the complex growth patterns of trees, and the many microorganisms involved. The anatomical nature of bark and the high moisture and low aeration levels of outer sapwood tissues in uninjured trees are effective barriers to the entry and growth of many decay fungi. Most dying tissues during the natural shedding of branches (self-pruning) are effectively sealed from inner living tissues anatomically by callus formation and chemically by infiltration with resins and gums further limiting fungal ingress.

Heart rots

Early forest pathologists (beginning with Hartig in the late 1800s) assumed that all heart-rot infections occurred through the exposed heartwood in branch stubs. This belief, which dominated forest pathology for nearly fifty years, was based on the observed continuity of the central decay columns with heartwood zones in the branch stubs. Decay fungi were believed to be saprobes that could grow only in dead heartwood tissues. The assumptions made were that the basidiospores of decay fungi germinated on the surface of dead branch stubs, grew into the exposed

heartwood and eventually invaded the heartwood zone of the central stem. No experimental evidence to date has verified this infection route; however, evidence that has accumulated indicates that heart-rot fungi use multiple pathways to enter trees. In 1938, Haddow showed that *Phellinus* (*Fornes*) *pini* often infected small branches or twigs of eastern white pine at the base of pole-sized trees or weeviled tips in larger trees. Decay development could be traced from these sapwood zones to the central stem. His research clearly established that sapwood injury and small branches (sapwood) were important infection courts for this major heart-rot fungus of conifers, worldwide. Studies of *Echinodontium tinctorium* on western hemlock also showed that small sapwood twigs and branches on the lower stem were the initial infection courts (Etheridge and Craig, 1976). The fungus entered the base of twigs, remained dormant for years in ray tissues until the branch tissues were enveloped by the growing stem. Subsequent stem injury or tree stressing appeared to initiate decay development. Extensive studies of stem decays in northeastern hardwoods have also shown that many of the apparent heart rots and central discolored zones have their origins as wound initiated sapwood decays that were compartmentalized and entrapped in heartwood after subsequent stem growth (Shigo et al., 1977). Some root rot pathogens such as *Heterobasidion* (*Fomes*) *annosum*, *Armillaria mellea*, and *Phellinus* (*Poria*) *weirii* are also able to invade the heartwood in the basal section of older trees from roots.

While dead branch stubs are still often posed as probable infection courts for many heart-rot fungi, the evidence is primarily circumstantial and proof is still lacking. It is clear that small dead branches, sapwood wounds, broken tops, and roots are entry points for many major heart-rot fungi.

Sap rots

Sap rots in the living stem primarily originate from wounds that expose and kill xylem tissues. Many sap rots in young vigorous stems are localized. Others are transitory and when contained by compartmentalization, become heart rots when enveloped later by the expanding heartwood. Other sap rots are traceable to drastic injury to stems or injuries to low vigor trees, where the callus tissue cannot enclose the damaged zone and decay slowly expands in the dead sapwood tissues. Some sap rots develop in the stems of highly stressed and moribund trees when bark tissues die.

Localized sap rots are also commonly associated with, or caused by, some stem cankers (e.g. *Cryptosphaeria, Eutypella, Strumella*, and *Hypoxylon cankers*). Sap rots are a common cause of stem breakage in the forest since the outer decayed zone is located where bending stresses during storms are at a maximum.

Types of stem wounds

Injuries to the bark and outer sapwood are common and occur throughout the life of most trees. Wounds that expose only sapwood contain a boundary of living tissues that may react to the injury and small wounds are commonly contained by callus development. Larger wounds or those that expose heartwood close more slowly and the chances of containing infections is much reduced.

Injuries that expose heartwood include: large dead branches and branch stubs, broken tops from windstorms, pruning wounds, and large broken branches or roots from windstorms.

Injuries that expose sapwood arise from: fire scars; logging and related mechanical damage; frost cracks; animal injury (i.e., rodent gnawing, bird pecks, deer, and elk damage); insect tunnels; sunscald; lightning strikes; bark cankers; and bark tears, and abrasions caused by falling trees or broken branches during wind storms.

Stem tissue reactions to wounding

The general observation that trees can live for centuries, even though stem wounds are common indicates that most timber species have evolved effective defense mechanisms to limit decay. Small sapwood injuries often heal rapidly, particularly in young vigorous trees. Wounding stimulates the production of growth hormones that in turn, stimulate the formation of callus cells from the cambium at the outer margin of the injury. The callus or wound parenchyma form new phloem and xylem tissues that gradually cover the wound surface with healthy tissue. Live tissues internal to the damage also react to isolate the dead and damaged

tissues. The isolation of diseased tissue by histological and chemical defenses is a common method many plants have developed to defend themselves against pathogens (Agrios, 2005). Major changes include the formation of tyloses and deposition of resins, gums and other toxicants (phytoalexins) in the living cells surrounding the injury (Shain, 1979). The term compartmentalization has been used by Shigo (1984) to describe this process in injured tree stems and will be discussed in the next section.

Another effective defense developed by some tree species is the infiltration of dying tissues with toxicants. Examples are the infiltration of branch stubs in many conifers and hardwoods with resins or gums and the deposition of highly toxic extractives in the heartwood of some timber species to create a hostile ecological niche that inhibits most decay fungi. Characteristics of these extractives will be addressed in more detail in Chapter 18.

Compartmentalization and succession

Shigo and his associates (Shigo and Marx, 1977; Shortle, 1979) used studies based on the dissection and analysis of the decay patterns in thousands of stems to develop a new interpretation of decay development that involved the ideas of compartmentalization and succession. They explained the "decay containment process" in some stems and the eventual formation of serious heartrot in others. The acronym CODIT (Compartmentalization Of Decay In Trees) was used to describe the process. The related term of succession described the sequence of microorganisms that invaded the damaged tissues, modified them, and initiated decay. Four barriers or stem zones restrict the invasion of microorganisms from sapwood wounds (Fig. 12.1). Wall 1 incorporates gums, tyloses, and resins that plug the cells above and below the injury. Wall 2 is the inner wall consisting of a series of latewood bands of cells where pitting is infrequent and the cell walls are thickest. Wall 3 includes the radially aligned wood rays, where living cells may react metabolically to the injury and release toxic phenolic compounds. Walls 1, 2, and 3 resist the upward, inward, and lateral spread of microorganisms. Wall 4 represents the new xylem tissue that subsequently develops over the wound surface and protects the new sapwood from infection. Walls 1 and 3 rely on parenchyma

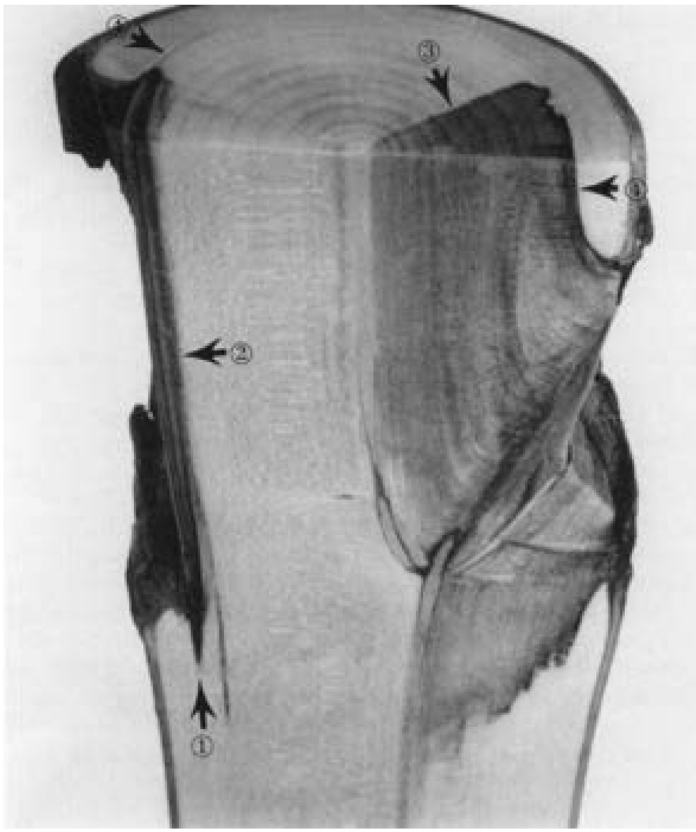


Figure 12.1 Example of walls (walls 1, 2, 3 and 4) that formed to compartmentalize two pruning wounds and limit invasion in a red maple (*Acer rubra*) stem. The flush cut wound on the right was not contained, whereas the pruning cut on the left retained the branch protection zone was successfully contained. *From Shigo, A. L. (1989). A New Tree Biology: Facts, photos and philosophies on trees and their problems and proper care, second ed. Shigo and Trees, Associates, Durham, New Hampshire. www.ShigoandTrees.com.*

cells with limited resources for response. Wall 4 is more intimately tied to the dynamic vitality (cambium) of the tree and is therefore the most effective. It is termed the barrier zone.

Succession, or the sequential involvement of other microorganisms associated with decay fungi, is the second premise of the expanded concept of stem decay. Based on isolations from many types and ages of stem injuries and inoculation experiments, Shigo (1967) concluded that bacteria were the first wound invaders, followed by a series of microfungi in genera such as *Phialophora, Gliocladium*, and *Ceratocystis*. These microorganisms generally preceded the appearance of decay fungi and were assumed to

play a role in conditioning the wound tissue for decay fungus invasion. Whether these organisms are required for infection by decay fungi, or just opportunistic remains uncertain and difficult to prove. Other studies showed that the entry of decay fungi into wounded stem tissues may not be immediate, but follows other microorganisms that presumably modify the micro-ecological conditions of the infection court (Etheridge, 1961; DeGroot, 1965). In contrast, decay fungi appear to be among the first fungi colonizing sweetgum and yellow-poplar wounds and are associated with tissue discoloration (Shortle and Cowling, 1978). Manion and French (1969) showed that *Phellinus tremulae* is one of the first fungi to colonize aspen wounds. Host wound defenses apparently inhibit spore germination of competing microorganisms and stimulate the germination of *P. tremulae* basidiospores.

Rayner and Boddy (1988) believed that the presence of microorganisms was not necessary to initiate host defense reactions and proposed that tissue drying and aeration of the injury surface were the initiating factors.

It is clear that much more needs to be known about when, where and how major decay fungi enter living stems and the roles of associated microorganisms (Manion and Zabel, 1979). Factors and conditions affecting spore germination of decay fungi on branches and injury courts are still largely unknown (Merrill, 1970). Clarification of the roles and sequences of other microorganisms involved in the invasion process may lead to effective "biological" methods for preventing invasion by decay fungi.

Rates of decay development

Very little is known about the rates of decay development in single stems since it is difficult to accurately determine the times of infection. Artificial inoculations of stems have indicated varying annual rates of longitudinal spread, ranging from a few centimeters to a meter in maples and aspen (Silverborg, 1959). Analysis of decay development in bottomland oaks indicated vertical spreads ranging from 25 to 225 mm annually (Toole and Furnival, 1957).

The rate of decay development in stands varies greatly with species and site. Comparisons of pathologic rotation periods provides some

insights on decay rate differences among species, but there still is the uncertainty of knowing when decay began or whether the rates of decay are uniform. Some examples of pathological rotation ages determined for mature timber are as follows: 40–50 years for aspen in Minnesota, (Schmitz and Jackson, 1927), 150 years for Douglas-fir in Washington (Boyce, 1932), 170 years for eastern white pine in Nova Scotia (Stillwell, 1955), and 250–300 years for Sitka spruce in the Queen Charlotte Islands (Bier et al., 1946). The long pathologic rotation for many timber species clearly illustrates that heart rots develop very slowly in most tree stems.

Recognition of stem decays

The detection of internal stem decay from external evidences is difficult and often presumptive at best. Many of the evidences used to detect decay during timber cruising is also useful for roundwood (logs, pulpwood, and poles). Wood users responsible for decay detection during purchasing or inspections should become familiar with regional cull manuals for the species involved. Useful macroscopic evidences of decay (reviewed in Chapter 7) that apply to timber and rough-wood are the presence of decayed knots, fungal fruiting bodies, and discolored or decayed wood in the exposed ends. A roughened sawcut often coincides with the exposed decayed zone in the log end. Instruments such as the Shigometer and increment borer are also often very useful for detecting internal decay in stems (see Chapters 10 and 16 for details). Many other useful clues to field detection of decays are often host or causal fungus-specific and are mentioned in regional cull manuals. For example, swollen knots in eastern white pine and beech containing a core of a brown spongy fungus material are evidences of extensive stem decay by *Phellinus pini* and *Inonotus glomeratus*, respectively. Swollen, resin-soaked knots are also presumptive evidence of *Phellinus pini*. A depression below knots in incense cedar, called a "shot-hole cup" is a reliable indicator of extensive decay caused by the brown pocket-rot fungus, *Oligoporus* (*Polyporus*) *amarus* (Boyce, 1961). Shigo and Larson (1969) prepared a photograph guide of many northeastern hardwoods showing that common decays, discolorations, and other defects are often associated with bark evidences of prior wounds.

Some common stem decays

Information on the identities of the many stem-decay fungi reported as damaging commercially important timber trees in North America was assembled by Hepting (1971) and the literature sources cited. Detailed descriptions of many of the major decays and their external evidences are also available (Boyce, 1961). The decay fungi reported as causing serious losses in the major timber species in the United States and Alaska are listed in Table 12.1. Several decays that develop in living stems and are later often found in roundwood, lumber, and commercial wood products are described briefly in this section.

Major heart rots

Phellinus (*Fomes*) *pini* is reported to be the major cause of stem decays in conifers in the northern temperate zone. It is particularly severe on Douglas-fir in the northwest and white pine in the east. The common names for this decay are white-pocket rot, white spec, or red-ring rot since the small decayed pockets in some hosts are concentrated in the earlywood bands. The wood is a pink-reddish to purplish color in the incipient and early decay stages.

In the early stages of the decay, the hemicelluloses and lignin are attacked selectively and in the later decay stages cellulose remains in many, small-lenticular shaped pockets. Wood in the early stages of the red-ring rot is reported to be suitable for general construction uses until the decay pockets are visible (firm pocket stage). Wood in the firm-pocket stage is also acceptable for low-grade construction lumber and plywood. The fungus invades the living sapwood in the advanced stages of decay and sometimes forms cankers. Infected wood in the outer-sapwood zones is often resin infiltrated. The decay fungus dies out rapidly once the tree is cut and seasoned.

External evidences of red-ring rot in standing trees are the presence of swollen knots containing brownish punky masses of fungal material, extensive resin flow from the base of branch stubs, and the presence of characteristic brownish-shelving perennial basidiomata (Fig. 12.2). Boyce (1961) reported that heart-rot columns extended a meter or more above

Table 12.1 A list of some common stem-decay fungi associated with major timber species in the United States.

Wood species	Important stem-decay fungi	Appearance	Type of decay	Location
Abies sp. (white, grand, California, red firs)	*Echindontium tinctorium*	Brown, laminated	White rot	Central stem
	Heterobasidion annosum	White, pocket	White rot	Lower stem
	Armillaria mellea	Light, brown	White rot	Basal stem
	Fomitopsis pinicola	Brown, cubical	Brown rot	Central stem
Abies balsamea (balsam fir)	*Stereum sanguinolentum*	Brown, stringy	White rot	Central stem
	Scytinostroma galactinum	Yellow, stringy	White rot	Basal stem
	Piloderma bicolor	White, stringy	White rot	Basal stem
Acer saccharum (sugar maple)	*Ionotus glomeratus*	Tan, spongy	White rot	Central stem
	Phellinus igniarius	White, mottled, springy	White rot	Central stem
	Oxyporus populinus	Yellow, stringy	White rot	Lower stem
	Cerrena uniicolor	White, soft	White rot	Central stem
	Ganoderma applanatum	White, mottled	White rot	Lower stem
	Climaccodon septentrionalis	White, spongy	White rot	Central stem
Betula allegeniensis (yellow birch)	*Phellinus igniarius*	White, mottled, spongy	White rot	Central stem
	Ionotus obliquus	White, mottled, spongy	White rot	Central stem
	Stereum murrui	Light brown	White rot	Central stem
	Ionotus hispidus	White, spongy	White rot	Upper stem
	Pholiota adiposa	Brown, mottled	Brown rot	Central stem
	Ganoderma applanatum	White, mottled	White rot	Lower stem
	Fomes fomentarius	White, mottled	White rot	Central stem
Chaemacyparis spp. (Alaska and Port Orford Cedar)	*Phellinus pini*	White, pocket	White rot	Central stem
	Fomitopsis pinicola	Brown, cubical	Brown rot	Central stem
Fagus grandifolia (American beech)	*Phellinus igniarius*	White, mottled, spongy	White rot	Central stem
	Ganoderma applanatum	White, mottled, spongy	White rot	Lower stem
	Ionotus glomeratus	Light brown, spongy	White rot	Central stem
	Armillaria mellea	Light brown	White rot	Lower stem
	Climacodon septentrionalis	White, spongy	White rot	Central stem
	Cerrena unicolor	White, soft	White rot	Central stem
	Ustilina deusta	White, soft	White rot	Basal stem

Juglans nigra (black walnut)	*Phellinus igniarius*	White, mottled, spongy	White rot	Central stem
	Phellinus everhartii	White, soft-flaky	White rot	Lower stem
	Ionotus hespidus	White, spongy	White rot	Upper stem
Larix occidentalis (western larch)	*Fomitopsis officinalis*	Brown, cubical	Brown rot	Central stem
	Phellinus pini	White, pocket	White rot	Central stem
	Laetiporus sulphureus	Brown, cubical	Brown rot	Central stem
Leriodendron tulipifera (yellow poplar)	*Pleurotus ostreatus*	White, flaky	White rot	Central stem
	Hydnum, erinaceus	White, spongy	Brown rot	Central stem
	Armillaria mellea	Light brown	White rot	Lower stem
Picea spp. (red, white, black spruce)	*Stereum sanguinolentum*	Brown, springy	White rot	Central stem
	Phellinus pini	White, pocket	White rot	Central stem
	Ionotus tomentosus	White, pocket	White rot	Lower stem
	Scytinostroma galactinum	Yellow, stringy	White rot	Basal stem
	Rhodoformes cajanderi	Brown, cubical	Brown rot	Upper stem
	Fomitopsis pinicola	Brown cubical	Brown rot	Central stem
	Perreniporia subacida	White, spongy	White rot	Lower stem
Picea sitchensis (Sitka spruce)	*Phellinus pini*	White, pocket	White rot	Central stem
	Fomitopsis pinicola	Brown, cubical	Brown rot	Central stem
	Phaeolus schweinitzii	Brown, cubical	Brown rot	Lower stem
	Oligoporus placenta	Brown, cubical	Brown rot	Central stem
	Neolentus kauffmanii	Brown, cubical, pocket	Brown rot	Lower stem
	Fomitopsis officinalis	Brown cubical	Brown rot	Central stem
	Laetiporus sulphureus	Brown cubical	Brown rot	Central stem
Pinus spp. (white, western red, and sugar pines)	*Phellius pini*	White, pocket	White rot	Central stem
	Phaeolus schweinitzii	Brown, cubical	Brown rot	Lower stem
	Heterobasidion annosum	White, pocket	White rot	Lower stem
	Fomitopsis officinalis	Brown, cubical	Brown rot	Central stem

(*Continued*)

Table 12.1 (Continued)

Pinus spp. (southern pines)	*Phellinus pini*	White, pocket	White rot	Central stem
	Phaeolus schweinitzii	Brown, cubical	Brown rot	Lower stem
	Heterobasidion annosum	White, pocket	White rot	Lower stem
Pinus ponderosa (ponderosa pine)	*Dichomitus squalens*	White, pocket	White rot	Central stem
	Laetiporus sulphureus	Brown, cubical	Brown rot	Central stem
	Phellinus pini	White, pocket	White rot	Central stem
	Crytopous volvatus	Gray, soft	White rot	Central stem
	Fomitopsis officinalis	Brown cubical	Brown rot	Central stem
Populus tremuloides (quaking aspen)	*Phellinus tremulae*	White, mottled, spongy	White rot	Central stem
	Ganoderma applanatum	White, mottled	White rot	Lower stem
	Armillaria mellea	Light, brown	Brown rot	Lower stem
	Phellinus prunicola	White, mottled, spongy	White rot	Central stem
	Phellinus laevigata	White, mottled, spongy	White rot	Central stem
Pseudotsuga menziesii (Douglas-fir)	*Phellinus pini*	White, pocket	White rot	Central stem
	Fomitopsis officinalis	Brown, cubical	Brown rot	Central stem
	Fomitopsis rosea	Brown, cubical	Brown rot	Upper stem
	Phaeolus schweinitzii	Brown cubical	Brown rot	Lower stem
	Oligoporus placenta	Brown, cubical	Brown rot	Central stem
Quercus spp. (white and red oaks)	*Hydnum erinacea*	White, spongy	White rot	Central stem
	Xylobolus frustulosum	White, pocket	White rot	Lower stem
	Pleurotus ostreatus	White, flaky	White rot	Upper stem
	Phellinus everhartii	White, soft, flaky	White rot	Lower stem
	Ionotus hispidus	White, spongy	White rot	Upper stem
	Stereum gausapatum	White, mottled	White rot	Central stem
Sequioa sempervirens (coastal redwood)	*Postia sequoiae*	Brown, cubical, pocket	Brown rot	Central stem
	Ceriporiopsis rivulosa	White, laminated	White rot	Central stem
Taxodium distichum (bald cypress)	*Stereum taxodii*	Brown, cubical, pocket	White rot	Central stem

Thuja plicata (western redcedar)	*Phellinus weirii*	Yellow, laminated	White rot	Lower stem
Tsuga heterophylla (western hemlock)	*Phellinus pini*	White, pocket	White rot	Central stem
	Heterobasidion annosum	White, pocket	White rot	Central stem
	Ganoderma applanatum	White, mottled	White rot	Lower stem
	Echinodontium tinctorium	Brown, stringy	Brown rot	Central stem
	Fomitopsis pinicola	Brown, cubical	Brown rot	Central stem
	Fomitopsis officinalis	Brown, cubical	Brown rot	Central stem

Principal information sources: Hepting, G.H., 1971. Diseases of Forest and Shade Trees of the United States. U.S. Dept. Agric., Forest Service, Agriculture Handbook, Number 386, Washington, DC; Gilbertson, R.L. Ryvarden, L., 1986/1987. North American Polypores, vol. I (1986), vol. 2 (1987), Fungiflora, Oslo, Norway. Names of fungi were obtained from Farr et al. (1989).

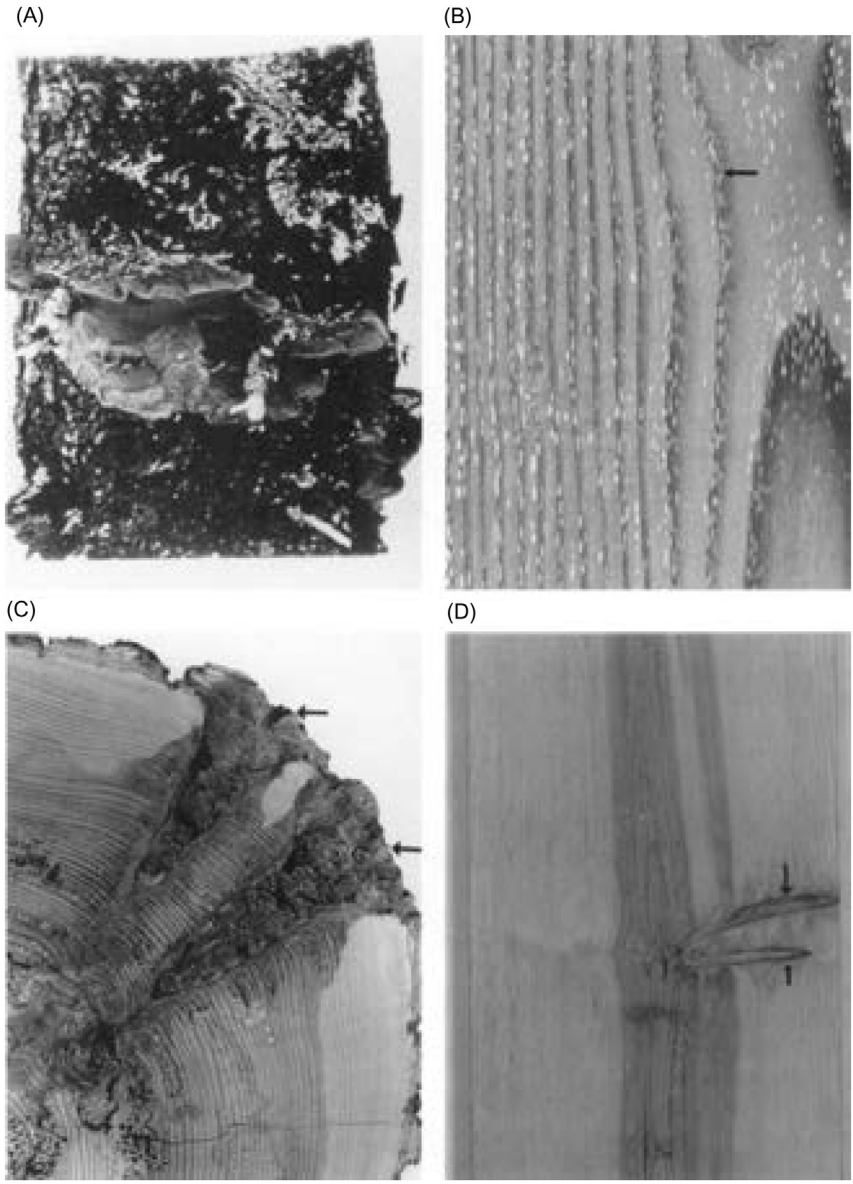


Figure 12.2 Evidence of *Phellinus pini* decay in logs or lumber can include (A) a typical basidioma on red spruce, (B) an early stage (firm pocket decay) of decay in Douglas-fir showing decay concentrated in the earlywood, (C) a punk knot indicating extensive decay, (D) buried weeviled tips (arrows) are common infection courts for *P. weirii* in eastern white pine and a useful indicator of decay. *Photo (C) courtesy From Shigo, A. L. (1989). A New Tree Biology: Facts, Photos and Philosophies on Trees and Their Problems and Proper Care, second ed. Shigo and Trees, Associates, Durham, New Hampshire. www.ShigoandTrees.com.*

the top basidioma or punk knot and about 1.3 m below the lowest. Haddow (1938) showed that small branches or buried weeviled tips were common infection courts on eastern white pine in the East. In studies of the infection process, DeGroot (1965) reported that the weeviled tip preference may reflect lower concentrations of pinosilvin in weeviled tips compared to recently dead branches. Pinosilvin levels decreased as branch stubs age and eventually basidiospores of *Phellinus pini* were able to germinate on these substrates; however, heartrot infections were not obtained.

Echinodontium tinctorium is the major cause of a serious heartrot in the true firs and western hemlock in the Pacific Northwest. The importance of this fungus varies with region and site, with losses as high as 30% of the gross merchantable volume on some poor sites. This fungus is a brown rot that causes a stringy or fibrous-textured decay. Wood in the incipient decay stage is a light-tan color and may extend 1−2 m longitudinally beyond the point of visible decay. In some cases, the decay may extend from the roots to the larger branches. Perennial, hoof-shaped basidiomata form beneath branch stubs (Fig. 14.4) and are readily recognized by the toothed pore surface and the bright-red context that gives the fungus its common name "Indian paint fungus". A single conk on a stem means essentially total stem cull. The earliest appearance of heart rot in stems has been estimated to range from 45 to 75 years. Infection is believed to originate some years earlier at the base of small branchlets on shade-killed lower branches. The fungus remains dormant for years as the branch tissues are slowly enveloped by stem growth. The fungus becomes activated later in response to surface wounds or loss of tree vigor (Etheridge and Craig, 1976). Managerial controls appear to be limited to determining regional pathologic rotations and silvicultural treatments to limit suppression of tree growth.

Phaeolus (*Polyporus*) *schweinitzii* causes a serious brown-cubical rot in the roots and the butt log of all conifers species in the north temperate zone with the exception of cedars, junipers, and cypresses. This fungus is a particularly dangerous heart-rot pathogen, since the early decay stages are barely detectable in the wood, yet associated with sharp reductions in tensile strength and toughness (Scheffer et al., 1941). There is a slight yellowing of the wood in vertical spires at the early stages of attack and incipient decay may project a meter or more above the zone of visible decay. The advanced decay stage is characterized by a brown cubical rot with near total tissue collapse. Decay develops in the roots and butt portions of logs and reaches a height of 2.4 m, although the decay column

may extend 4.5–6.0 m above ground in some cases. Occasionally, young trees are killed by extensive decay in the roots and wind throw is common in severely diseased stands. The only external evidence of this heart rot is the occasional presence of the characteristic brown, velvety textured, stipitate basidiomata arising from the forest floor near the base of the infected tree or, rarely, sessile conks attached to basal wounds (Fig. 14.4). Presumed entry points of the fungus are through broken roots or deep basal wounds. Conifer roots previously infected by *Armillaria mellea* may be predisposed to colonization by *P. schweinitzii* (Barrett, 1970). Decayed trees often occur in clusters in pine stands, but the decay may not be detectable until felling. Managerial controls are not known other than following pathologic rotations and avoiding planting on sites where *A. mellea* is established. Heavy stand stocking to minimize root breakage during wind storms has been suggested as a potential method for decreasing infection.

Fomitopsis officinalis occurs on conifers in North America and Europe. It is important primarily on Douglas-fir, sugar pine, ponderosa pine and western hemlock in western North America and produces a brown cubical rot in the late stages that is often characterized by the presence of thick mycelial felts in the shrinkage cracks. Like P. *schweinitzii*, this fungus is dangerous in wood products since the early stage (a faint yellow to brown) is almost imperceptible and incipient decay may extend a meter or more longitudinally beyond the zone of visible decay. Infection pathways are uncertain but are presumed to occur through large broken branches in the upper bole and broken tops, but some infections are also associated with fire scars and logging wounds. Large chalky white, hoof-shaped perennial basidiomata are found in wound faces or knots (Fig. 14.4) and the presence of a single basidioma indicates total cull.

Phellinus (*Fornes*) *igniarius* is an important major cause of heart rot in birches, maples, beech, and oaks in North America. *Phellinus tremula*, a closely related species, is the major cause of heart rot in aspens. This fungus causes a white-spongy rot in the heartwood, but can also invade and kill living sapwood. The rot, as seen on stem-cross sections, is characterized by a yellowish-green to brownish-black outer invasion zone that surrounds a core of irregularly mottled, white spongy wood. The advanced decay often contains fine, concentrically arranged zone lines. The decayed wood is still usable for pulping purposes. The fungus may continue to decay stored wood for some months, but eventually dies out in the finished product. Perennial hoof-shaped basidiomata commonly develop in

late decay stages at wound margins or the base of branch stubs. The presence of a single conk generally indicates a cull tree with decay columns ranging from 3 to 5 m in length. Wounds are believed to be the principal infection sites. Minimizing stem injuries and following regional pathological rotations are the recommended management practices for limiting damage by this fungus.

Inonotus (*Poria*) *obliquus* and *I.* (*Polyporus*) *glomeratus* cause a mottled white rot that is very similar to decays caused by *Phellinus igniarius*. *Inonotus obliquus* is a major cause of heart rot in birches in the northern hemisphere, while *I. glomeratus* is important primarily on maples and beech in eastern North America. Both decay fungi form large, black, coal or clinker-like abortive basidiomata that are sterile and perennial on living hosts. Both fungi also invade the living sapwood and may form stem cankers. The presence of a single sterile conk indicates extensive heart rot and a cull tree. Swollen knots or dead bark on canker faces in beech, may partially conceal the sterile conks of *I. glomeratus*. Wounds and cankers appear to be the principal infection courts for these fungi. *Inonotus obliquus* forms a brown resupinate basidioma in the decayed sapwood of dead hosts splitting off the outer sapwood and bark layers to expose the hymenial surface, while *I. glomeratus* forms large effused-reflexed basidiomas on the lower surface of logs (Zabel, 1976). The basidioma of *I. obliquus* is also called "chaga" and is reputed to have medicinal properties. Neither fungus continues to decay wood in the product form.

Common sap rots

Sap rots are more localized in the stem than most heart rots and are usually associated with dead tissue from large stem wounds such as logging injuries, fire scars, frost cracks, sunscald, and lightning injuries that fail to callus over or injuries on trees of low vigor. Most sap rots are white rots and occur more commonly on hardwoods. These fungi are of special importance because of their location in the lower, outer stem zones where the most valuable lumber grades are located and strength most critical. Sap-rot fungi are primarily facultative pathogens on severely wounded, low vigor, or moribund trees. Most sap-rot fungi in tree stems are also important decayers of forest slash and wood products. Some of the

common sap rots occurring on the trunks of living trees are described below. Descriptions of the associated basidiomata are available in the taxonomic literature (Gilbertson and Ryvarden, 1986/1987).

Fomes fomentarius causes a white-mottled rot primarily in beech and the birches. While found commonly on slash and dead stubs, it also causes sapwood decay in moribund and severely injured trees. *Gandoderma applanatum* causes a white-mottled rot in butt logs of oaks and other hardwoods. *Ustilina* (*vulgaris*) *deusta*, an ascomycete, causes a white rot in the basal roots and butt of maples and beech. The decayed wood contains numerous zone lines that often remain as black, brittle sheets of fungal tissue in badly decayed stumps. Interestingly, logs with these zones lines produce lumber that is highly prized by wood workers because of its attractive colors. Black perithecial stromata often cover the remnants of decayed stumps.

Daedaleopsis (*Daedalea*) *confragosa* is a common white rotter that is associated with severe upper stem wounds on maples, willow, and other hardwoods. *Cerrena* (*Daedalea*) *unicolor* causes a white rot and a canker on maples, although it is unknown whether this fungus causes the canker or invades the dead tissues. *Irpex lacteus* causes a soft-white rot and commonly fruits on dead branches and severe wounds in hardwoods. *Bjerkandera adusta* causes a soft-white rot in beech sapwood after the bark has been damaged by the beech scale (*Cryptococcus fagisuga*) and *Nectria* canker. *Amylostereum chailletii* causes a red stain and white rot in balsam fir that has been stressed and killed by the spruce budworm (*Choristoneura fumiferana)*. This fungus is vectored by a wood wasp (*Sirex* sp.) after the tree is damaged by insect defoliation or fire (Warren, 1989; Slippers et al., 2012). *Hirschioporus abietinus* forms a white-pocket rot in the large wounds and associated dead tissue in conifers.

Host specific stem decay fungi

Some heart rot fungi appear to be important in only a limited number of hosts. For example, *Oligoporus* (*Polyporus) amarus* causes a brown cubical rot in incense cedar throughout the range of the species. Decay pockets are localized and surrounded by firm wood. The presence of a basidioma (rare and rapidly consumed by insects) or the characteristic shot-hole cup indicates a rot column throughout the stem heartwood. *Laurilia*

Figure 12.3 Example of the typical brown cubical pocket rot on cypress, known in the lumber trade as "pecky cypress".

(*Stereum*) *taxodii* causes a brown-cubical pocket rot in cypress, known as pecky cypress in the lumber trade. The decayed pockets are small, and, as in the case of *Q. amarus*, the wood surrounding the pockets appears sound. Both woods species produce very durable heartwood that performs well in posts and greenhouse benches. Pecky cypress has textural qualities that are aesthetically pleasing and is a popular paneling product (Fig. 12.3).

Phellinus robiniae (*Fomes rimosus*) causes a soft-white rot in black locust. The perennial basidiomata are conspicuous and indicate the presence of extensive heartwood decay. Black locust has a very durable heartwood and is often used for fence posts.

Some colonization strategies of stem decay fungi

Stem decay fungi display a wide variation in host selectivity. Some are limited to the heartwood zones of a few durable species, while others have broad host ranges. Some heart-rot fungi only develop in living trees and cannot survive in the wood once it is cut. Some sap rotters are predominately saprobes in slash or wood products and mostly occupy the dead tissue of severe wounds. Colonization strategies may explain some of these differences (see Chapter 11). Some heart-rot fungi, such as *Stereum taxodii* and *Postia sequoiae* appear to have selected for "stress resistance" as a colonization strategy. Tolerance to the toxic heartwood extractives of the host tree would be an important selective trait (Wilcox, 1970). Other heartwood characteristics that restrict fungal access are low nitrogen levels and high CO_2/O_2 ratios (Highley et al., 1983; Rayner, 1986). Stress resistance to adverse heartwood environmental conditions or unique infection routes, as shown by *E. tinctorium*, may explain the host selectivity of some

heart rot fungi. Many of the fungi that cause severe basal rots are potent pathogens that penetrate root tissues from large inoculum bases in the soil. The stem decay fungi that primarily colonize damaged sapwood tissues appear to be "substrate opportunists". Sapwood injury exposes a nutrient rich substrate that rapidly dries and aerates to levels favorable for microbial growth (Rayner and Boddy, 1988). It is interesting to note that many of the sap rotters in living stems are also important slash and products rot fungi, while many of the heart-rot fungi are unable to survive or compete outside of the unique environmental niche provided in heartwood of the living tree. The exposed dead tissues in large stem wounds provide a substrate for many opportunistic fungi that is similar to sapwood in slash or wood products.

Undoubtedly, there are many other explanations for host selectivity by heart rot fungi. The many microbial sequences and interactions involved in the infection process in stem decays may be another important cause of host selectivity. Methods for circumventing the containment defenses of a tree may also be important. The host selection process can be viewed as a part of the long-term co-evolution of hosts and pathogens in plant diseases.

Summary

1. Some decays and discolorations that appear in wood products develop in the living tree. It is particularly important to detect early decay in wood destined for critical strength uses or applications where decay adversely affects important use properties. A general understanding of the origins and types of decay that originate in the living tree will facilitate its detection in products and encourage more effective wood uses.
2. Decay fungi in living stems are pathogens and their damages are grouped as heart rots or saprots based on the stem zones occupied. Heart rots develop primarily in the physiologically inactive central zone of the stem and may occupy the entire heartwood of a tree. Sap rots in the physiologically active outer sapwood usually develop after wounding and are often localized. In contrast to stem decayers, the fungi that decay wood products are saprogens.

3. Stem decays may be either white or brown rots. While the white rots predominate, brown rots are a special concern since the incipient stage is often difficult to detect and associated with large strength losses.
4. Dead branches, wounds, and root tissues appear to be the principal infection courts for many stem-decay fungi. Sapwood tissues in branches are the point of entry for these fungi. While exposed heartwood is also a logical entry point, entry through this wood has not been decisively demonstrated.
5. Trees have evolved effective defenses against the invasion of most wood inhabiting fungi. The defenses involve the impregnation of branch stubs with resins and gums, the development of callus to form new bark and xylem tissues over the wounds, and the isolation of injured tissues.
6. The term compartmentalization has been coined to describe the "infection containment process" and utilizes, in part, the natural structure and functioning of the stem. Compartmentalization involves the deposition of gums and resins, the formation of tyloses to restrict longitudinal microbial spread, production of successive layers of thick-walled latewood tissues to restrain radial spread, and the release of toxic chemicals (phytoalexins) from radial tissues to reduce tangential spread. Subsequent xylem tissues laid down by callus limit outward spread. The barrier zone formed by the callus is the most effective infection restraint. Compartmentalization effectively isolates infections through most small wounds, particularly in vigorous trees, and walls them off from the rest of the tree.
7. Tissue drying and aeration in the boundary zones of stem wounds have been proposed as possible causes of the defense reactions and not necessarily the actions of microorganisms.
8. Some heart rot decays are traceable to compartmentalized infections that became active after they were included in the heartwood or when the outer spread was contained, but the fungus continued to colonize tissues towards the pith.
9. Succession or the sequential involvement of bacteria and microfungi with the decay fungi is believed to be an important part of the infection process. These organisms are presumed to modify the tissue to favor decay fungi.
10. The presence of characteristic fruiting bodies, decayed (punky) knots, and discolored or decayed wood in the exposed ends of logs and pulpwood are useful macroscopic evidences of decay in logs and

roundwood. A roughened sawcut often coincides with the exposed decay in the log end.

11. The major stem decays in the United States and Alaska are briefly described with emphasis on their recognition, decay type, and associated cull.
12. Future practical controls of stem decays may rest on developing a better understanding of microbial colonization sequences and interactions in infection courts and the physiologic actions involved in tree responses to injury. Such information could lead to biological protection strategies and provide useful information for tree selection for genetic improvement.

References

Agrios, G.N., 2005. Plant Pathology, fifth ed. Elsevier, New York, p. 952.
Atwell, E.A., 1948. Red-Stain and Pocket-Rot in Jackpine: Their Effect on Strength and Serviceability of the Wood, 63. Department of Mines & Resources, Dominion Forest Service Forest Product Laboratory Circular, pp. 1–23.
Barrett, O.K., 1970. *Armillaria mellea* as a possible factor predisposing roots to infection by *Polyporus schweinitzii*. Trans. Br. Mycol. Soc. 55, 459–462.
Basham, J.T., 1958. Decay of trembling aspen. Can. J. Bot. 36, 491–505.
Bier, J.E., Foster, R.E., Salisbury, P.J., 1946. Studies in Forest Pathology IV. Decay of Sitka Spruce on Queen Charlotte Islands. Can. Dept. Agr. Publ. 783, Tech. Bull. 56, pp. 1–35.
Boyce, J.S., 1920. A Dry-Rot of Incense Cedar. U.S. Dept. Agr. Bul. 871-1-58.
Boyce, J.S., 1932. Decay and Other Losses in Douglas-fir in Western Oregon and Washington. U.S. Dept. Agr. Tech. Bull. 286, pp. 1–59.
Boyce, J.S., 1961. Forest Pathology, third ed. McGraw-Hill, New York, p. 572.
Cartwright, K.St.G., Findlay, W.P.K., 1958. Decay of Timber and Its Prevention, second ed. Her Majesty's Stationery Office, London, p. 332.
Davidson, A.G., Redmond, D.R., 1957. Decay of spruce in the maritime provinces. For. Chron. 33, 373–380.
DeGroot, R.C., 1965. Germination of the basidiospores of *Fomes pini* on pine wood extract media. For. Sci. 11, 176–180.
Etheridge, D.E., 1961. Factors affecting branch infection in aspen. Can. J. Bot. 39, 799–816.
Etheridge, D.E., Craig, H.M., 1976. Factors influencing infection and initiation of decay by the Indian paint fungus (*Echinodontium tinctorium*) in western hemlock. Can. J. For. Res. 6, 299–318.
Farr D.F., Bills G.F., Chamuris G.P. and Rossman, A.Y., Fungi on Plants and Plant Products in the United States, 1989, American Phytopathological Society Press, St. Paul, MN.
Gilbertson, R.L., 1980. Wood-rotting fungi of North America. Mycologia 72, 1–49.
Gilbertson, R.L., Ryvarden, L., 1986/1987. North American Polypores, volume I (1986), volume 2 (1987). Fungiflora, Oslo, Norway.
Good, H.M., 1959. Fungi associated with *Fomes igniarus* in decay of poplar. Proc. Ninth Int. Bot. Congr. 2, 136.
Haddow, W.R., 1938. The disease caused by *Trametes pini* (Thore) Fries in white pine (*Pinus strobus* L.). Trans. R. Can. Inst. 22, 21–80.

Hepting, G.H., 1935. Decay Following Fire in Young Mississippi Delta Hardwoods. U.S. Dept. Agr. Tech. Bull. 494, 32 pp.
Hepting, G.H., 1971. Diseases of Forest and Shade Trees of the United States. U.S. Dept. Agric., Forest Service, Agriculture Handbook, Number 386, Washington, D.C.
Hepting, G.H., Hedgcock, G.G., 1937. Decay in Merchantable Oak, Yellow Poplar, and Basswood in the Appalachian Region. U.S. Dept. Agricultural Technical Bulletin 570-1-20, Washington, D.C.
Highley, T.L., Bar-Lev, S.S., Kirk, T.K., Larsen, M.J., 1983. Influence of O_2 and CO_2 on wood decay by heartrot and saprot fungi. Phytopathology 73, 630–633.
Hubert, E.E., 1931. An Outline of Forest Pathology. John Wiley & Sons, New York, 534 pp.
Hubert, E.E., 1924. The diagnosis of decay in wood. J. Agr. Res. 29 (11), 523–567.
Kimmey, J.W., Hornebrook, E.M., 1952. Cull and Breakage Factors and Other Tree Measurement Tables for Redwood. U.S. Dept. Agr., Forest Service, California Forest and Range Exp. Sta., Forest Survey Release 13, pp. 1–28.
Lockhard, C.R., Putnam, J.A., Carpenter, R.D., 1950. Log Defects in Southern Hardwoods. U.S. Dept. Agr. Handbook 4, pp. 1–37.
Manion, P., 1991. Tree Disease Concepts, second ed. Prentice Hall, New York, NY.
Manion, P., 1967. Factors Affecting the Germination of Basidiospores of the Heartrot Fungus *Fomes igniarius* in the Infection of Aspen (Ph.D. thesis). Univ. Minnesota.
Manion, P.D., French, D.W., 1967. *Nectria galligena* and *Ceratocystis fimbriata* cankers of Aspen in Minnesota. For. Sci. 13, 23–28.
Manion, P.D., French, D.W., 1969. The role of glucose in stimulating germination of *Fomes igniarius* var. *populinum* basidiospores. Phytopathology 59, 293–296.
Manion, P.D., Zabel, R.A., 1979. Stem decay perspectives are introduction to the mechanisms of tree defense and decay patterns. Phytopathology 69 (10), 1136–1138.
Merrill, W., 1970. Spore germination and host penetration by heart rotting hymenomycetes. Ann. Rev. Phytopathol. 8, 281–300.
Murphy, T.G., Rowatt, H.H., 1932. British Columbia Softwoods: Their Decays and Natural Defects. Dept. of Interior, Canadian Forest Service, Bulletin No. 80, Ottawa, Canada.
Partridge, A.D., Miller, D.I., 1974. Major Wood Decays in the Inland Northwest. Idaho Research Foundation, University Station, Moscow, Idaho.
Rayner, A.D.M., 1986. Water and origins of decay in trees. In: Ayres, P.G., Boddy, L. (Eds.), Water, Fungi, and Plants. Cambridge University Press, Cambridge, pp. 321–341.
Rayner, A.D.M., Boddy, L., 1988. Fungal Decomposition of Wood. Its Biology and Ecology. John Wiley and Sons, Chichester, United Kingdom.
Roth, E.R., 1959. Heart Rots of Appalachian Hardwoods. U.S.D.A. Forest Service, Forest Pest Leaflet 38.
Scheffer, T.C., Wilson, T.R.C., Luxford, R.F., Hartley, C., 1941. The Effect of Certain Heart-Rot Fungi on Specific Gravity and Strength of Sitka Spruce and Douglas-fir. Tech. Bull. U.S. Dept. Agric., No. 779, 24 pp.
Schmitz, H., Jackson, L.W.R., 1927. Heartrot of Aspen With Special Reference to Forest Management in Minnesota. Minn. Agr. Expt. Stat. Tech. Bull., 50, pp. 1–43.
Schrenk, H. Von., 1900. Some Diseases of New England Conifers. Bull. U.S. Dept. Agric. No. 25, pp. 31–40.
Shain, L., 1979. Dynamic responses of differentiated sapwood to injury and infection. Phytopathology 69, 1143–1147.
Shigo, A.L., 1984. Compartmentalization: a conceptual framework for understanding how trees grow and defend themselves. Ann. Rev. Phytopathol. 22, 189–214.

Shigo, A.L., 1976. Microorganisms isolated from wounds inflicted on red maple, paper birch, American beech, and red oak in winter, summer, and autumn. Phytopathology 66 (5), 559–563.
Shigo, A.L., 1967. Successions of organisms in discoloration and decay of wood. Intl. Rev. For. Res. 2, 237–249.
Shigo A.L., A New Tree Biology: Facts, Photos and Philosophies on Trees and Their Problems and Proper Care, 1989, Shigo and Associates, Durham.
Shigo, A.L., Hillis, W.E., 1973. Heartwood, discolored wood, and microorganisms in living trees. Ann. Rev. Phytopathol. 11, 197–222.
Shigo, A.L., Larson, E., 1969. A Photo Guide to the Patterns of Discolorations and Decay in Living Northern Hardwood Trees. U.S.D.A. Forest Service Research Paper NE-127, Northeastern Forest Experiment Station, Upper Darby, PA.
Shigo, A.L., Marx, H., 1977. CODIT (Compartmentalization of Decay in Trees). U.S. Dept. Agric. Inf. Bull. 405, 73 p.
Shigo, A.L., Shortle, W.C., Garrett, P.W., 1977. Genetic control suggested in compartmentalization of discolored wood associated with tree wounds. For. Sci. 23, 179–182.
Shortle, W.C., 1979. Mechanisms of compartmentalization of decay in living trees. Phytopathology 69, 1147–1151.
Shortle, W.C., Cowling, E.B., 1978. Development of discoloration, decay, and microorganisms following wounding of sweetgum and yellow poplar trees. Phytopathology 68, 609–616.
Silverborg, S.B., 1954. Northern Hardwoods Cull Manual. State University of New York College of Forestry at Syracuse University, Bull. 31, p. 1045.
Silverborg, S.B., 1959. Rate of decay in northern hardwoods following artificial inoculation with some common heartrot fungi. For. Sci. 5, 223–228.
Sinclair, W.A., Lyon, H.H., Johnson, W.T., 1987. Diseases of Trees and Shrubs. Cornell University Press, Ithaca, N.Y, p. 574.
Slippers, B., DeGroot, P., Wingfield, M.J., 2012. The *Sirex* Woodwasp and Its Fungal Symbiont. Springer, New York, 301 pages.
Stillinger, J.R., 1951. Some Strength and Related Properties of Old-Growth Douglas fir Decayed by *Fomes pini*. ASTM Bulletin No. 173, pp. 52–57.
Stillwell, M.A., 1955. Decay of yellow birch in Nova Scotia. For. Chron. 31, 74–83.
Tainter, F.H., Baker, F.A., 1996. Principles of Forest Pathology. John Wiley and Sons, New York.
Toole, L.R., Furnival, G.M., 1957. Progress of heartrot following fire in bottom-land red oaks. J. For. 55, 20–24.
Wagener, W.W., Davidson, R.W., 1954. Heartrots in living trees. Bot. Rev. 20, 61–134.
Warren, G.R., 1989. The Identities, Roles, and Interactions of the Fungi Associated With Stem Biodeterioration of Balsam fir Stressed and Killed After Spruce Budworm Defoliation in Newfoundland (Ph.D. thesis). State University of New York College of Environmental Science and Forestry, Syracuse, N.Y., p. 227.
Weir, J.R., Hubert, E.E., 1918. A Study of Heart-Rot in Western Hemlock. U.S. Dept. Agr. Bull. 722.
Wilcox, W.W., 1970. Tolerance of *Polyporus amarus* to extractives from incense cedar heartwood. Phytopathology 60, 919–923.
Williams, M., 1989. Americans and Their Forests. A Historical Geography. Cambridge University Press, Cambridge, United Kingdom, p. 599.
Zabel, R.A., 1976. Basidiocarp development in *Ionotus obliquus* and its inhibition by stem treatment. For. Sci. 22 (4), 431–437.

Further reading

White, L.T., 1953. Studies in forest pathology. X. Decay of white pine in the Timagami Lake and Ottawa Valley areas. Can. J. Bot. 31, 175–200.

CHAPTER THIRTEEN

Biodeterioration of stored wood and its control

Contents

Exposed wood surfaces rapidly dry when a living tree is felled, debarked, and cut into logs or bolts. Drying reduces the risk of fungal attack, but the sapwood portions become susceptible to fungal colonization until the wood dries. In some cases, bark also loses its protective properties and is invaded soon after felling by both insects and fungi. In an ecological sense, the freshly exposed wood tissues provide a new energy-rich substrate for rapid colonization by opportunistic microorganisms. Essentially all timber species are subject to serious damage by invading microorganisms within a few months after cutting during warm seasons in the temperate zones and at nearly all times in the tropics. Chipping trees or wood waste for paper pulp or fuel has many advantages, but also provides a greatly expanded, unprotected wood surface for rapid microorganism colonization.

Round wood (logs, poles, and bolts) and chips are the common raw material forms for most primary wood-processing operations. Many operations need to maintain large inventories to achieve seasonal transportation or purchase price advantages to insure continuous operation of large mills. In other cases, there may be seasonal harvesting limitations or the need to air season materials prior to treatment. These storage periods may

Wood Microbiology.
DOI: https://doi.org/10.1016/B978-0-12-819465-2.00013-9

sometimes extend for several years and can result in serious deterioration losses unless remedial steps are taken.

In this chapter, we review the principal types of storage losses, their causal agents and the factors affecting their development in logs, pulpwood and chip piles stored in the United States. An emphasis will be placed on management to prevent or reduce losses. Particularly useful sources for additional information on log and chip wood storage practices are Hajny (1966), Scheffer (1969), Cowling et al. (1974), Hulme (1979), and Fuller (1985).

Types of storage loss

The principal losses from prolonged wood storage are decay, discoloration (both chemical and biological), insect damage, and deep checking during drying. Other subtle losses that are difficult to detect and quantify can also be important in some products. Fungal colonization during storage causes variability in wood properties such as permeability, strength, color and decay susceptibility in structural uses. The magnitude of decay losses varies greatly with length of storage, region, wood species, season, and end use. The ultimate application for a given piece of timber will also influence how damage is perceived.

Insect tunnels and associated stains may be very damaging in veneer bolts, but of little concern in pulpwood. Pulp yield losses from decay and stains are of special concern during pulpwood and chip storage for paper products. End checking may be a minor defect in logs destined for poles or piling, but causes serious losses in lumber or veneer recovery. Some conifers are very susceptible to sap stain damage while many hardwoods are relatively immune. White rots, which are more common in hardwoods, may cause modest losses in chips used for paper production, whereas the brown rots, which are more common in conifers may be very damaging. Bark beetles rapidly attack logs and veneer bolts stored in the Gulf region of the United States and cause serious damage, but are generally less important in cooler, northern zones. Particularly severe losses from both insects and fungi can occur rapidly in tropical regions. In northern locations, logs and wood chips may be stored beginning in the winter for a year or so with minimal deterioration losses.

Actual wood losses for the various industries are difficult to measure for a number of reasons. These include the fact that wood is continually added and removed from the same storage pile, unit measures often change between the raw material and product, it is difficult to measure large bulk volumes of mixed species of wood, the high percentage of waste products involved in many wood product conversions, subtle changes in quality, and the huge volumes of continually changing raw materials. Degraded wood may not be a complete loss because it can still be used for other purposes and thus is not counted as a loss.

Storage losses are often accepted due to the low cost and abundance of wood and the absence of precise information on the cost of the losses. Some control practices followed to reduce losses in high value products include fall cutting and winter storage, ponding logs, and limiting the storage periods to several months in the south and less than a year in northern regions.

Studies of wood storage losses in the United States began shortly after the depletion of the eastern forests and the rapid expansion of the pulp and paper industry in the northern states (Humphrey, 1917; Kress, 1925).

General control practices

The relative value of the end products generally determines the nature and cost of the control treatments that are feasible. For example, the high value of a black walnut peeler log for veneer justifies costly control treatments for each log, while the low value of pulpwood severely limits treatment possibilities.

The general methods used for protecting green wood during storage are: rapid utilization; arranging the major storage period to begin during a cold season; storage under water; water sprinkling or spraying; chemical treatments with fungicides and/or insecticides, applications of end-checking compounds, and conversion to chips.

The actual controls practiced vary greatly with species, periods of storage, end products, region, and the size and sophistication of the, industry. Short-term storage practices for low value items generally try to keep the wood wet to limit oxygen levels and for long-term storage, the wood is peeled so it can dry out as rapidly as possible. Chemical treatments or water immersion are necessary for high cost items. Basic control rules are rapid utilization and maintaining the minimum volume of wood in storage permitted by the economics of the operation. This is difficult because

mills must purchase on anticipated demand that might fail to materialize. This results in wood remaining in storage for long periods of time under conditions conducive to fungal and insect attack. These times pose the greatest risk to wood in storage.

Logs and bolts

Logs of all North American species can be damaged when stored under warm, moist conditions (Fig. 13.1). These losses are often severe when late fall and winter cut logs are stored more than a year in northern and western zones or three months or more in the south. While these losses are accepted as substantial by the industry and needing control, published data documenting the volume and grade losses are meager. Visible decay can develop 30–45 cm into each end of birch bolts stored during a single summer season in the Northeast (Isaacson, 1958), while the bark on beech bolts rapidly loses its protective qualities allowing decay to develop throughout the bolt from the sides of the logs (Scheffer and Zabel, 1951; Scheffer and Jones, 1953). Scheffer and Eslyn (1976) also noted relatively shallow decay zones in birch exposed for 3 months in Wisconsin and Michigan, but substantial attack after a full summer of exposure (Table 13.1). Similar decay rates can occur in the sapwood zones of most other species stored through a summer season unless preventative measures are taken.

Sapstain fungi and ambrosia beetles may become major problems within a month or so of summer storage in *Pinus* spp. or hardwoods and seriously degrade high quality lumber (Fig. 13.2). Neither of these agents causes substantial losses in physical properties, but they mar the appearance of the finished products. End checking of logs or stored lumber results in reduced recovery in some species during periods of drying stress and solar exposure. A more serious consequence of drying checks is their function as infection courts for decay and stain fungi which greatly prolongs the colonization period of seasoning logs.

General practices followed in most small operations to reduce seasoning losses include:

(a) Pile logs off ground on well-drained sites
(b) Facilitate air movement around the piles
(c) Avoid exposing log ends to the south to minimize solar heating and end checking or end-seal valuable logs.
(d) Rapid utilization

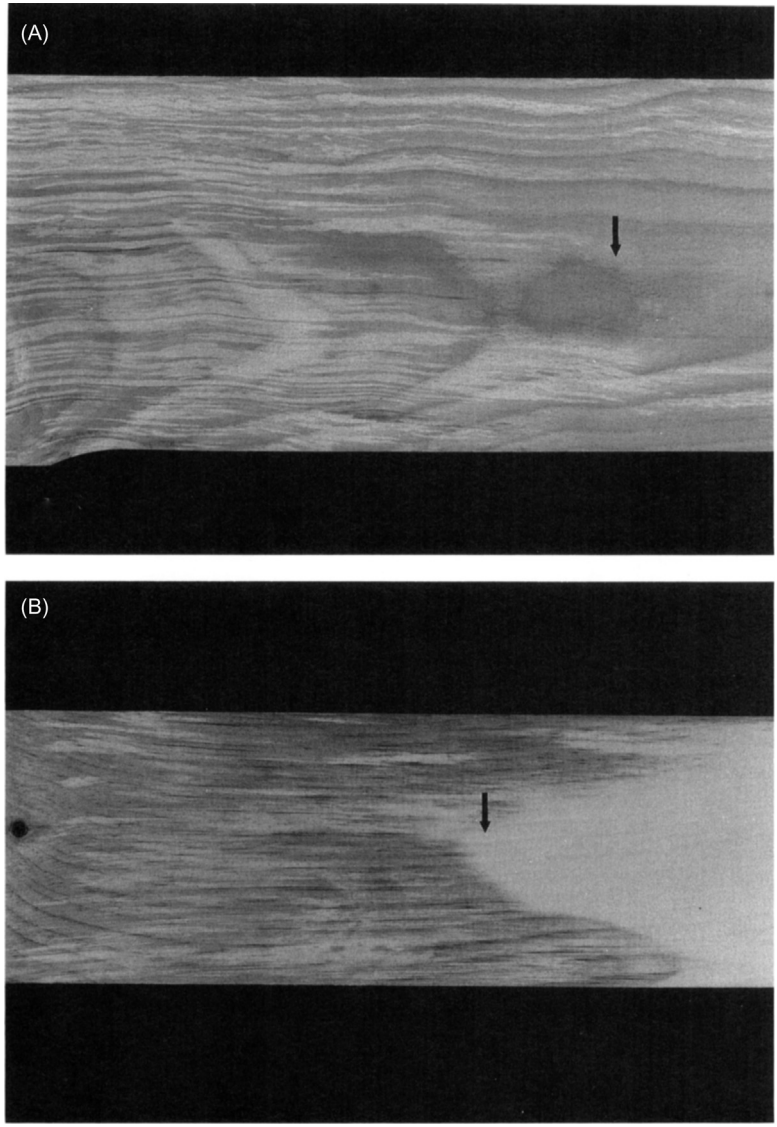


Figure 13.1 Boards sawn from the ends (arrows) of logs showing penetration of early decay of (A) beech (*F. grandifolia*) and sapstain on (B) eastern white pine (*P. strobus*) after 2 months of summer storage in northern New York (X 0.5).

(e) Initiate storage during colder seasons
(f) Follow the practice that first logs in are the first logs out for processing

Table 13.1 Degree of decay in birch logs stored for varying periods of time in the Upper Midwest[a].

Exposure periods	Decay penetration from ends (cm)	
March – July	5	–
March – October	27.5	35
April – October	25	32.5
November – October	32.5	35

[a]Data from Scheffer and Eslyn (1976).

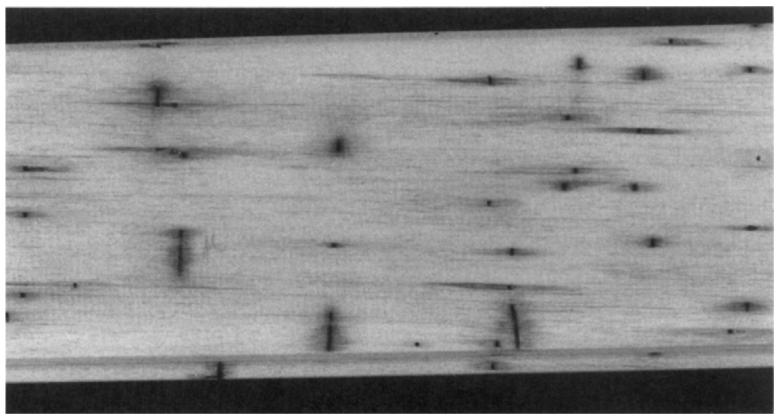


Figure 13.2 Sapstain and pinhole damage from ambrosia beetles that developed in eastern white pine (*P. strobus*) after summer storage in central New York.

Other practices followed in large operations for high value logs are water storage, water spraying or sprinkling, and chemical treatments.

Water storage: Submerging logs in water (ponding) provides superior protection and has long been used for tide-water mills and where the processing operation is adjacent to a suitable convenient water body (Fig. 13.3). Water storage provides several years of protection against decay, stains, insect attack, and checking. The presumed protective mechanism is oxygen deprivation of wood-inhabiting fungi because of the low concentrations and slow diffusion rate of oxygen in water. Despite the effectiveness of water immersion to control biologically derived storage defects there are disadvantages. It takes large volumes of water (about 3–4 acres of water surface per million bd. ft. of logs) to store adequate supplies of roundwood for large operations. It is difficult to keep the logs grouped by species, size, and grade for various uses. High density

Figure 13.3 A large raft of logs in the Pacific Northwest assembled for water protection against insects and fungi.

hardwood logs often sink and may be difficult to recover. Exposed portions of conifer logs are still subject to decay, stain, and insect damage unless held under water by baffles. A slight darkening of the wood occurs in some hardwoods and disagreeable odors may develop and permeate the wood in smaller ponds during warmer periods of the year. In some cases, serious permeability damage may occur in some conifers and especially the soft pines. Under prolonged or concentrated log storage conditions bacteria can invade and destroy parenchyma tissue (wood rays and resin canals) increasing the permeability of the wood (Ellwood and Ecklund, 1959; Knuth and McCoy, 1962). Logs storage ponds are also regulated by various agencies with regard to water quality and any releases of water are likely to have high levels of tannins and other water-soluble wood components that might affect water quality of the receiving body of water.

Water spraying: Water spraying or sprinkling protects decked logs of all species for an entire warm season (Hansbrough, 1953; Lane and Scheffer, 1960; Carpenter and Toole, 1963). There appears to be no difference between continuous mist sprays and intermittent sprinkling provided the volumes of water delivered are sufficient to keep the log surfaces saturated at all times. Spraying must begin while the logs or bolts are still green; otherwise, fungi and insects may already be inside and actively degrading the material. A thick film of bacteria and molds typically develop on the ends and surfaces of the logs, but any wood penetration is shallow and

removed as slabs or initial peelings during log processing. For example, longleaf pine logs protected by water spraying for a summer season in the south retained similar strength properties (toughness) to fresh-cut logs (Scheld and DeGroot, 1971). The protective mechanism is unknown, but water storage presumably deprives microorganisms of oxygen. Surface microflora (yeasts, bacteria, molds, and algae) may also form physical or chemical (antibiotic) barriers to invasion by airborne decay and stain spores or the parenchyma cells in the bark and sapwood may remain viable longer in water soaked wood to serve a protective function. Spraying also appears to limit beetle attack, possibly because it upsets normal egg laying behavior (Garcia and Morrell, 1999). Spraying costs are nominal, but can often require the acquisition of appropriate environmental permits.

The advantages of sprinkling or spraying are effectiveness and nominal cost. The disadvantages are the need to initiate spraying promptly while the logs are still green, the availability of large supplies of water, the need to provide drainage to avoid excessively muddy conditions to access the piles, and the need to assure that runoff does not adversely affect nearby surface water.

Chemical treatments: Chemical treatments can also limit fungal or insect attack for many species for several months even where storage hazards are high. They can also limit end-checking when combined with waxes.

Treatments are applied by spraying or brushing fungicides or insecticides on the exposed wood on log ends and debarked areas, spraying the bark with insecticides where ambrosia beetles and wood borers are a problem, or applying sealants to the log ends (after the fungicide treatment) to reduce end checking. It is recommended that log ends and sides be sprayed with fungicides and insecticides in areas below 40° N latitude when summer storage periods up to seven weeks are anticipated (Scheffer and Eslyn, 1976). Hardwood logs stored in the northern zones usually require only a fungicidal treatment. End coatings are necessary to reduce checking for longer storage periods. Chemical treatments are particularly useful for small operations where stock piles accumulate rather irregularly and the log values are high. The fungicides used are the anti-stain chemicals commonly used for lumber dipping (see Chapter 14 on wood sap stains). The principal insecticide used initially was the gamma isomer of benzene hexachloride but this chemical was banned for wood protection uses by the Environmental Protection Agency (EPA) and substitutes such as synthetic pyrethroids are now used. Appropriate State or Federal

regional agencies should be consulted to determine which insecticides and fungicides are approved currently by the EPA or state agencies for local wood protection uses. End coatings include waxes, hardened gloss oil, coal tar pitches, and asphalt mixtures. A variety of proprietary end dressing compounds designed to reduce checking are also available.

The principal disadvantages of depending on chemical methods for log protection are the critical need to treat the logs within 24 hours of tree felling to avoid cases where the rapid growth of fungi in the exposed xylem cells exceeds the subsequent depth of fungicide penetration and the potential environmental impacts of chemical loss from the treated wood. Chemical treatments may also be applied prior to shipping debarked logs in containers, although the value of these treatments is difficult to assess because many fungi may have already colonized the wood between the time of felling and treatment.

Biological controls: The growth of innocuous molds on exposed wood surfaces that inhibit colonization by damaging decay and stain fungi has long intrigued wood microbiologists. *Trichoderma viride* is one example of a pioneer fungus that rapidly invades exposed wood tissues and forms green masses of spores on the surface. The hyphae that penetrate the wood are colorless and do not adversely affect wood strength or color. Lindgren and Harvey (1952) reported that applying *T. viride* provided considerable protection against stain and decay development in pine bolts stored in the south. Spraying the wood with solutions of ammonium bifluoride appeared to favor the growth of *T. viride* on the log surfaces. Similar *Trichoderma* inoculations have also protected several hardwood species (Shields and Atwell, 1963).

The erratic nature of the protection has limited large scale applications. The potential for bioprotection against both stain and decay fungi has been the subject of periodic renewed interest, but the variability has largely limited commercial application (Seifert et al., 1988, Benko and Highley, 1990). An interesting alternative that has seen some success is the application of pigmentless *Ophiostoma piliferum* (Blanchette et al., 1992; Berendt et al., 1995). The idea was to inoculate logs with the pigmentless isolate so that it outcompeted and excluded the pigmented fungi. This strategy has been used with some success on radiata pine. It also has the added advantage of removing resins that can interfere with pulping for paper. Biological control of damaging fungi during log storage remains an intriguing future possibility warranting additional research.

Poles and piling

Air-seasoning of poles for periods of a few months in the south and years in the northwest prior to preservative treatment is practiced by many wood-treating companies. Particular advantages of air-seasoning to reduce pole moisture contents are the low cost and avoidance of potential damaging strength effects of the high temperatures associated with some kiln drying schedules. Practices to hasten air seasoning are prompt debarking and shaping, stickered piling on foundations, and, sometimes, overhead structures to protect against rainfall. Air seasoning poles in the south for periods of even a few weeks during periods of wet weather can lead to rapid colonization by stain and decay fungi. The decay fungi enter the pole ends and any deep checks that may form on the pole surfaces. Zones of early internal decay with prolonged storage may develop between the inner sapwood and heartwood. The use of elevated temperatures or deep preservative penetration usually kills the invading fungi in the smaller poles, but cannot restore the original properties of the wood subjected to fungal attack. So-called internal "pre-treatment decay" in poles is particularly insidious because it is difficult to detect, may seriously reduce pole bending strength, and results in erratic preservative treatment. Decay increases wood permeability, and poles exposed to rain just prior to treatment will be poorly penetrated while those treated after prolonged dry weather will be over-treated. Over-treated poles tend to bleed in service, creating potential environmental hazards. Taylor (1980) made a number of recommendations for limiting pretreatment decay in utility poles. Dip or flood treatments of debarked and shaped poles in concentrated solutions of ammonium bifluoride have been shown to provide protection against decay and stain fungi during air-seasoning periods of up to a year (Panek, 1963), but the treatment is no longer available and the process is logistically difficult. Prompt kiln drying of southern pine poles prior to preservative treatment minimizes any decay and related storage problems and kiln drying is now widely used in the southern U.S.

Douglas-fir poles in the Pacific Northwest may be air-seasoned for periods of up to two years (Fig. 13.4). Air seasoning is practiced for economic reasons because of the large size of the poles and the energy costs required for the long kiln schedules necessary to minimize checking.

A study of Douglas-fir poles in the Eastern U.S. found a high incidence of decay in poles that had been air-seasoned several years prior to treatment with chromated copper arsenate (CCA) at ambient temperatures

Figure 13.4 Douglas-fir poles piled for air-seasoning in a Pacific Northwest prior to preservative treatment..

(Zabel et al., 1980). One particularly abundant fungus, *Antrodia carbonica*, had previously been found primarily in the Pacific Northwest, suggesting that the fungus was in the pole at the time of installation. Subsequent studies indicated that peeled Douglas-fir poles air-seasoned in the open for more than a year in the Pacific Northwest were extensively colonized by a variety of decay fungi (Przybylowicz et al., 1987; Morrell et al., 1987). However, no significant strength losses were noted in poles seasoned up to two years in the region (Smith et al., 1987). Chemical treatment with ammonium bifluoride delayed and reduced colonization by decay fungi, while similar treatments with boron were less effective. Both treatments were recommended when poles were air-seasoned longer than one year, but were never commercially used (Morrell et al., 1989). While minimizing fungal colonization is important, it is equally critical that air-seasoned poles should receive preservative treatments at temperatures that kill all established decay fungi (Newbill, et al., 1988). This is generally considered to require heating of a wood to 65.5 °C for at least 75 minutes (Chidester, 1939).

Railroad ties

Wood ties are used to support over 95% of rails in North America. Wood is ideal for this purpose because it is relatively inexpensive, easily replaced and its properties closely match those required for rail applications.

Most ties are hardwoods, but the wood species vary widely in their resistance to fungal attack as well as preservative treatment. In fact, some of the earliest studies of fungi on wood products were conducted on ties. Ties are usually air-seasoned for 6–12 months prior to preservative treatment. This period creates ample opportunity for fungal attack or so called "stack burn." Recent studies suggest that a majority of ties in a seasoning yard will contain at least one decay fungus at the end of the seasoning period, although the immediate effects of fungal attack on timber properties may be minimal. The guidelines for proper seasoning include creating piles that are at least 300 mm off the ground, using preservative treated timbers near the ground, creating adequate air flow, avoiding areas with standing water, and removing vegetation and woody debris. All these actions, reduce, but do not eliminate the risk of fungal attack. This makes it critical that air-seasoned ties be subjected to a sterilization process to eliminate fungi that invaded the ties during seasoning.

Pulpwood

Pulp mills require large inventories of raw wood to insure continuous operation. Initially, most mills were located in northern states and pulpwood was cut in the fall and winter seasons and placed in large pulpwood ricks or jack straw piles (Fig. 13.5). Wood losses were recognized after prolonged storage, but were overlooked because of· the relatively low value of pulpwood. In the 1950, pulp and paper operations began in the south where conditions were more conducive for decay and a series of studies were begun to quantify pulpwood storage losses and develop controls for these losses (Pascoe and Scheffer, 1950; Lindgren, 1953; Lindgren and Eslyn, 1961; Mason et al., 1963; Hajny, 1966). Decay and stain fungi are the principal destructive agents in stored pulpwood and reduce both pulp yield (specific gravity) and quality (color changes and losses in paper strength properties). This damage may also increase bleaching costs and contribute subsequently to slime control and wastewater problems in the paper mill. The fungi invading stored pulpwood are similar to those that invade forest slash and stored logs. The principal decay fungi invading coniferous pulpwood are *Phanerochaete* (*Peniophora*) *gigantea* and *Gloeophyllum* (*Lenzites) sepiarium*, while hardwood pulpwoods are degraded by *Hirschioporus pargamenus*, *Trametes versicolor*, and *Bjerkandera*

Figure 13.5 A large pulp mill showing the large piles of chips and pulpwood bolts required to maintain a constant supply. *Courtesy W.S. Fuller, Weyerhauser Co. and permission TAPPI (1985).*

adusta. Many members of the genera *Ceratocystis, Ophiostoma, Graphium*, and *Alternaria* also colonize pulpwood and cause stain damage that can increase bleaching costs. All of these fungi are common colonizers of fallen timber in the forest.

Some examples of the weight losses associated with several pulpwood species, stored for various periods in several locations include:

a. Southern yellow pine in the southern locations in rick piles, unbarked: in the summer season lost 2–4% weight in two months, 6–8% in

four months, 8–10% in six months. Weight losses in the winter season were 0–1.5% in two months, 1.5–3% in four months, 3–5% in six months, and losses for an entire year ranged from 11% to 15% (Lindgren, 1953; Lindgren and Eslyn, 1961). Subsequent studies produced similar results (Volkmann, 1966).

b. Jack pine (*Pinus banksianna*) in the Lake States lost 4.5% weight in one year and 9% in two years (Pascoe and Scheffer, 1950).

c. Aspen (*Populus tremuloides*) in the Lake States lost 6% weight per year (Scheffer, 1969).

d. Weight losses during six months of summer storage in the south were 9.6% for pine, 7.2% for oak, and 13.0% for gum (Hajny, 1966).

Estimates of storage losses in pulpwood must also consider the effects of decay type on pulp yields. Brown-rot fungi, such as *Gloeophyllum sepiarium*, selectively attack carbohydrates and associated increases in alkali solubility substantially decrease pulp yield, magnifying the storage decay loss. White rot fungi utilize wood components more or less uniformly and have little effect on yield on a weight basis. Their attack, may, however, require that a larger volume of wood be processed to maintain a given production level. Some white rot fungi selectively utilize lignin and these fungi have been intensively studied for use in biological pulping. While fungi such as *P. chrysosporium* can selectively remove lignin and might be useful in some pulping processes such as mechanical pulping, the logistics associated with treatment of large volumes of chips have largely precluded commercial applications.

General approaches to reducing pulpwood storage losses include rapid utilization, rick piling, and procurement schedules that maintain maximum volumes during the colder seasons. Peeling has been reported to substantially extend safe storage periods for aspen in the north. Lindgren (1953) recommended retaining bark, tight piles, and favoring large diameters and long lengths for short storage periods in the south (<3 months in the summer and 5 months in the winter). Debarking, open piles, favoring small diameters, and splitting large diameter bolts were recommended for storage periods exceeding these limits. A high initial moisture content can slow microbial growth for shorter storage periods, while rapid drying is the goal for longer periods.

Other controls, similar to those used for logs have been employed in some larger mills. Chemical treatments, again similar to those used for logs have been attempted, but proved to be expensive and often ineffective on peeled pulpwood in the south. Underwater storage is effective,

but is cumbersome because of the huge volumes of wood involved. Water sprays were introduced at some mills and proved to be very effective (Chesley et al., 1956; Volkmann, 1966). Large-scale experiments demonstrated that spraying could allow 12 months of safe storage at greatly reduced costs compared to conventional dry wood storage (Djerf and Volkman, 1969). Water spraying has been accepted generally as the most effective and economical way to store both conifer and hardwood roundwood in the south for up to 12 months without appreciable deterioration damage. Minor problems with spraying include loss of parenchyma cells in the outer 2.5 cm of sapwood after 4 months of storage and the occasional development of a sour odor. Prolonged storage, however, did not reduce the amounts of extractive by-products such as tall oil, turpentine, or rosins.

As an alternative to spraying, attempts were made to exclude oxygen by sealing large piles of green pulp in polyethylene enclosures (McKee and Daniel, 1966). Many fungi can survive at very low oxygen levels, making it difficult to completely limit growth. Furthermore, the material and handling costs of this technique were prohibitive. Biological protection of pulpwood by inducing *Trichoderma viride* growth on the surface was reported to reduce storage losses in some cases (Lindgren and Harvey, 1952); however, the treatment results were erratic.

Despite the effectiveness of decay control measures on pulpwood bolts, interest in round wood storage declined with the introduction of chip piling around 1950. Handling advantages and relatively low losses have made chips the predominant method of wood storage in pulp and paper mills.

Pulpwood chips

Chip storage of pulpwood reduces deterioration losses, decreases handling costs, and requires smaller storage areas. It also provides an economic use for slabs, edgings, and other forms of wood waste in large integrated wood conversion operations. Disadvantages include increases in dirt contamination and the loss of by-product yields (tall-oil and turpentine) in some pulping operations. The development of mechanized pulpwood harvesting and field chipping has also accelerated the use of chips for both pulp and as hog fuel.

Generally, bolts or wastewood are drum debarked, chipped and blown into large bins or carried by conveyor belts into huge conical piles (Fig. 13.5). Safe storage periods for chips of various species in regions of the United States were initially reported as follows: Southern yellow pines in the south up to four months; conifers in the northwest up to three years; hardwoods in the northwest up to one year; and hardwoods in the northeast up to 8 months. However, it was soon found that serious deterioration developed in some chip piles after prolonged storage and factors such as high sapwood proportion, green (wet) wood, susceptibility of some species (e.g. aspen, red alder), and excessive pile height or compaction could reduce safe storage periods. In general, conifer chips can be safely stored for longer periods than hardwood chips. Large diameter pulpwood contains more heartwood resulting in more durable chips. Seasoned pulpwood forms drier and more durable chips. Safe storage periods are longer in the cooler northern regions, particularly when the piles are assembled in the colder seasons.

The principal defects that develop in stored chips are chemical decomposition and discolorations (brownish), sap stains, molds, and decays. Temperature in the chip pile is an important factor, determining both the type of chemical damage and location of the microorganisms (Bjorkman and Haeger, 1963; Eslyn, 1967; Hulme and Stranks, 1976; Greaves, 1971; Wang, 1965). Many factors are involved in elevated temperatures that develop rapidly in large chip piles. The initial temperature rise (up to 7 days) is due to continued respiratory activity of living parenchyma cells in the sapwood that may remain viable for up to 6 months after cutting (Feist et al., 1971). Heat from the respiration of the huge bacterial population that rapidly develops on the chip surfaces and utilizes exogenous simple carbon sources (counts as high as 755×10^{11} bacterial cells per gram of wood have been measured) further increases pile temperature. Temperatures as high as 48.9 °C may be reached after 7 days (Springer and Hajny, 1970).

Further temperature rises depend, in part, on pile features. Tall piles with excessive compaction, or accumulated layers of fines have reduced air circulation that allow continued heating. These conditions may result in temperatures reaching between 60 and 110 °C. Slow heat decomposition of wood begins at these temperatures as acetic acid is released and begins to degrade the carbohydrates (Kubler, 1982). The exothermic reaction further elevates temperature and the acetic acid increases wood acidity which may reach a pH of 3. Chips exposed to these acidity and

Figure 13.6 Examples of heat/acid damaged wood chips caused by improper storage on top of fresh chips. The small pile on the right was exposed to temperatures in excess of 175 °F for 3–4 weeks, the pile on the left was exposed to the same temperatures for 6–8 weeks and smelled of acetic acid. *Courtesy W.S. Fuller, Weyerhauser Co. and permission TAPPI (1985).*

temperature levels turn brown and become friable (Fig. 13.6). Catastrophic losses from chemically degraded chips and occasional fires from spontaneous combustion can occur in piles under these high temperature conditions. The principal heat sources in chip piles and the factors affecting various temperature levels are illustrated in Fig. 13.7.

After the initial rise, temperatures decline in properly constructed and managed piles, but remain higher than external ambient temperatures (Feist et al., 1973a,b). Management practices generally strive to maintain pile temperatures below 60 °C. In an ecological sense, chips in a pile present a drastically different environmental niche to invading microorganisms than logs or roundwood during storage. Chips collectively create a huge wood surface that is inoculated with the spores and mycelial fragments of fungi and bacteria during chipping, transport and piling. They may also be contaminated with soil that carries many more organisms. The chip surfaces are smeared with extractives and ruptured cell contents that provide a moist film of simple carbon compounds. Chip piles are colonized rapidly within days by many of the same opportunists that invade

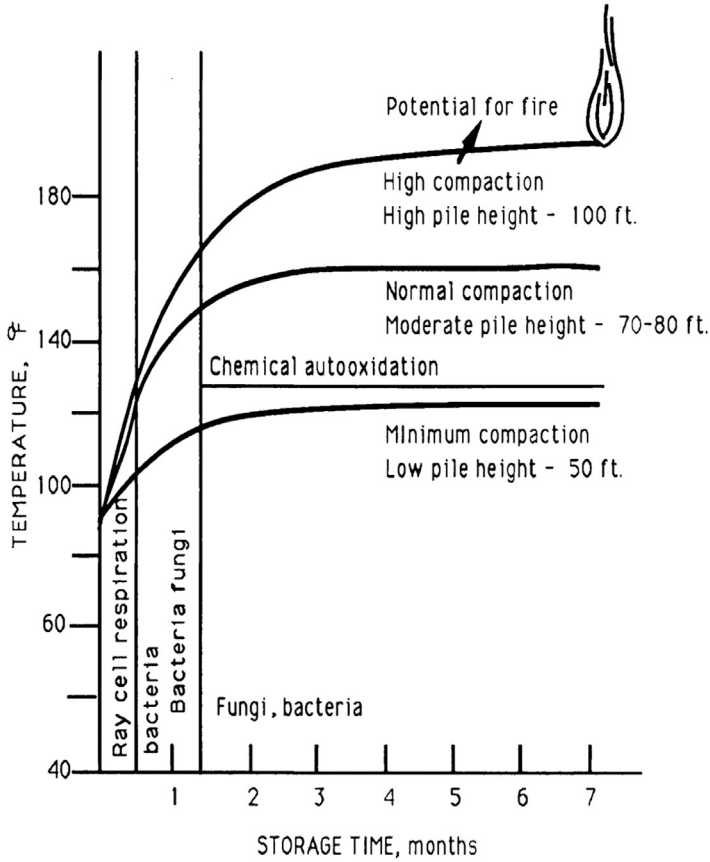


Figure 13.7 The principal heat sources in large piles of pulpwood chips and the various factors affecting the temperature levels and chip damage. *Courtesy W.S. Fuller, Weyerhauser Co. and permission TAPPI (1985).*

roundwood and slash. Respiration and fermentation release considerable heat. The small size and weight of chips leads to compaction in larger piles. The pile temperature rapidly rises and within a week or so the pile interior becomes a hostile site where colonization is limited to "stress-resistant" thermo tolerant or thermophilic fungi and bacteria. The outer shell of the pile is generally cooler and wetter than the inner zones. Chips in the outer layer of a pile are often stained, covered in mold spores or bleached. Common genera of the associated fungi are stainers such as *Ophiostoma, Graphium, Aureobasidium, Leptographium*, and *Alternaria*; as well as molds such as *Trichoderma, Aspergillus, Gliocladium*, and *Penicillium*

(Wang, 1965). The bleached chips, often at the early stages of attack by common white-rot fungi such as *Phlebiopsis gigantea*, *Hirschioporus* (*Polyporus*) *abietinus*, *Trametes versicolor*, and *Bjerkandera* (*Polyporus) adusta*. Other decayers such *as Gloeophyllum sepiarium* and *Fomes roseus,* are found in localized pockets. Common soft rot genera include *Chaetomium, Phialophora*, and *Humicola*. The wood-staining fungi (Eslyn and Davidson, 1976), microfungi (Shields, 1969), and thermophilic fungi (Tansey, 1971) in chipwood piles have been studied, although many of their interactions remain poorly documented.

Chips in the interior, warmer zones of the pile are characterized by light brown oxidative staining. Bacteria and thermophilic or thermotolerant fungi such as Actinomycetes and soft-rot fungi, predominate in the interior zone. Common thermophilic fungi include *Chaetomium thermophile* and *Talaromyces* (*Penicillium*) *duponti*. Major damage to the chips in the interior occurs through a combination of heat decomposition (acetolysis) and soft-rot decay.

As a secondary effect, the fungi colonizing stored chips can also pose health problems, both from the standpoint of fungal spores acting as allergens as well as the potential for human mycoses. Chemicals were initially evaluated to minimize microbial activity in storage. Eslyn (1973) evaluated a large number of chemicals and reported that dithiocarbamate and dinitrophenol showed particular promise. Propionic acid, which is used to control microorganisms in grain storage, has also been evaluated. Chemicals are sprayed on the chips as the pile is assembled; however, chemical control approaches to reduce chip storage losses are largely impractical due to high relative costs and environmental concerns associated with the potential runoff. However, the potential for chip treatment merits some attention. For example, a trial of radiata pine chips sprayed with a fungicide showed that the resulting pulp was brighter and required less bleaching.

Water spraying was evaluated as an alternative, but provided no benefits over dry storage (Djerf and Volkman, 1969). Current control approaches are to reduce storage times to absolute acceptable minimums and follow pile management practices to minimize conditions favoring chip deterioration. Current recommendations include limiting pile heights to 15 m (50 ft) and restricting tractor spreading to avoid compaction that reduces air circulation and sets the stage for precipitous temperature increases (Fuller, 1985). Chips from sources that deteriorate rapidly (hardwoods such as aspen and alder or whole tree chips) should be piled separately to avoid creating heat-generating pockets in the pile. Steps should

be taken to avoid layering of fines or sawdust in pile zones. Whole-tree chips, which are particularly subject to heating, should be stored separately in smaller piles (less than 8 m) and for periods not to exceed 4 weeks.

An important step in reducing chip deterioration is to regularly monitor pile condition and temperatures to catch problems before they become extreme. Rapid temperature rises and the strong odor of acetic acid are signs that a pile is out of control and heading for serious damage. Rapid utilization or ventilation of hot spots in the pile by trenching are then necessary. Prompt remedial measures are needed whenever pile temperatures reach 79 °C. Other useful procedures in large operations are reclaim systems that obtain chips from the base of pile using screw conveyors. These devices permit a first-in first-out practice, thereby reducing the storage time of individual chip batches. Blending of chips from storage piles of various ages just prior to use has been proposed as a useful way to avoid quality variations in chips due to differing storage periods, but this makes management more complex and costly (Schmidt, 1990).

A large literature on pulpwood chip storage exists on topics such as optimum size and packing of piles, types and sequences of microorganisms which occur in various pile locations and chip types, methods of monitoring weight losses in piles and the effects of microorganism types and storage periods on paper quality (Hatton, 1979). Interest in bioenergy and biofuels has encouraged continued studies, but these processes are very sensitive to the costs of other energy sources (White et al., 1983; Auchter, 1975). However, many mills have only a limited knowledge of their chip storage problems. Increasing wood prices often renews interest in chip quality, but current abundant natural gas prices have reduced interest in wood as a fuel source.

Summary

1. Logs, poles, bolts, and wood chips contain substantial amounts of sapwood that is susceptible to rapid colonization by microorganisms. Stored wood is subject to severe damage within months during warm periods unless properly managed.
2. The principal losses from prolonged or improper storage of raw wood are decay, chemical and biotic discolorations, insect damage,

deep checking, and in special cases, fires from internal combustion in chipwood piles.

3. General methods for reducing wood storage losses are rapid utilization, initiating storage piles during a cold season, storing under water, water spraying or sprinkling, and conversion to chips.
4. Visible decay can develop 30–45 cm in from each end in hardwood logs stored during the summer season. Sapstains and insect damage in *Pinus* spp. and susceptible hardwoods can seriously degrade high quality lumber within a month of storage during a warm season.
5. Submerging green logs in water (ponding) or continuous spraying protects wood from storage losses for an entire warm season.
6. Prompt kiln drying of southern yellow pine poles prior to preservative treatment is generally practiced in the south to eliminate pretreatment decay. In the Pacific Northwest, Douglas-fir poles are often air-seasoned, but the seasoning period should not exceed two years, and seasoning must be followed by preservative treatments that heat the wood to at least 65.5 °C for 75 minutes.
7. Decay and stain attack in stored pulpwood bolts, reducing both the yield and quality (brightness and strength) of the pulp. Density losses in southern pine pulpwood stored for a year in the south may range from 11% to 12%
8. Pulpwood storage losses can be reduced by rapid utilization, storing during colder seasons, and continuous water spraying or ponding.
9. The conversion of pulpwood to chips for storage has become common practice because of handling ease, the smaller storage spaces required, low deterioration losses, and the ability to use other wood wastes. Initial safe storage periods of pulpwood in chip piles were reported as months in the south and up to several years in the northwest. A disadvantage of chipping is the rapid loss of tall-oil, turpentine and other valuable by-products during storage.
10. Chemical decomposition and discoloration of chips can occur when the piles are too large and become compacted. Respiration of parenchyma cells and fermentation by bacteria rapidly elevate the interior temperature of piles within one week of initial assembly. Temperatures in large or compacted piles can reach levels that chemically decompose the wood and release acetic acid.
11. The elevated temperatures in chip piles create highly selective conditions for thermophilic and thermotolerant fungi and bacteria. Soft-rot fungi are the principal cause of biotic damage to stored chips.

12. Common practices for minimizing wood damage during storage in chip piles are rapid turnover, limiting pile heights (<15 m), avoiding compaction, and limiting accumulation of fines.
13. The development of procurement, storage, and handling systems to minimize deterioration losses will be increasingly important to wood using industries as wood raw material costs increase.

References

Auchter, R.L., 1975. Whole-tree utilization: fact or fancy. Tappi 58 (11), 4–5.
Benko, R., Highley, T.L., 1990. Evaluation of Bacteria for Biological Control of Wood Decay. International Research Group on Wood Preservation Document No. IRG/WP/1426, Stockholm, Sweden.
Berendt, C.J., Blanchette, R.A., Farrell, R.L., 1995. Biological control of blue-stain fungi in wood. Phytopathology 85, 92–97.
Bjorkman, E., Haeger, G.E., 1963. Outdoor storage of chips and damage by microorganisms. Tappi 46 (12), 757–766.
Blanchette, R.A., Farrell, R.L., Burnes, T.A., Wendler, P.A., Zimmerman, W., Brush, T. S., et al., 1992. Biological control of pitch in pulp and paper products by *Ophiostoma piliferum*. Tappi J. 75, 102–106.
Carpenter, B.E., Toole, E.R., 1963. Sprinkling with water protects hardwood logs in storage. South. Lumberm. 207 (Aug. 1), 25–26.
Chesley, K.G., Hair, L.C., Swartz, L.N., 1956. Underwater storage of southern pine pulpwood. Tappi 39 (9), 609–614.
Chidester, M.S., 1939. Further studies on temperatures necessary to kill fungi in wood. Proc. Am. Wood Preserv. Assoc. 35, 319–324.
Cowling, E.B., Hafley, W.L., Weiner, J., 1974. Changes in value and utility of pulpwood during harvesting, transport, and storage. Tappi 57 (12), 120–123.
Djerf, A.C., Volkman, D.A., 1969. Experiences with water spray wood storage. Tappi 52 (10), 1861–1864.
Ellwood, E.L., Ecklund, B.A., 1959. Bacterial attack of pine logs in pond storage. For. Prod. J. 9 (9), 283–292.
Eslyn, W.E., 1967. Outside storage of hardwood chips in the Northeast II. Microbiological effects. Tappi 50 (6), 297–303.
Eslyn, W.E., 1973. Evaluating chemicals for controlling biodeterioration of stored wood chips. For. Prod. J. 23 (11), 21–25.
Eslyn, W.E., Davidson, R.W., 1976. Some wood-staining fungi from pulpwood chips. Mem. N. Y. Bot. Gard. 28 (1), 50–57.
Feist, W.C., Springer, E.L., Hajny, G.J., 1971. Viability of parenchyma cells in stored green wood. Tappi 54 (8), 1295–1297.
Feist, W.C., Hajny, G.J., Springer, E.L., 1973a. Effect of storing green wood chips at elevated temperatures. Tappi 56 (8), 91–95.
Feist, W.C., Springer, E.L., Hajny, G.J., 1973b. Spontaneous heating in piled wood chips - contribution of bacteria. Tappi 56 (4), 148–151.
Fuller, W.S., 1985. Chip pile storage -a review of practices to avoid deterioration and economic losses. Tappi 68 (8), 48–52.
Garcia, S.M., Morrell, J.J., 1999. Fungal associates of *Buprestis aurulenta* in western Oregon. Can. J. For. Res. 29, 517–520.
Greaves, H., 1971. Biodeterioration of tropical hardwood chips in outdoor storage. Tappi 54 (7), 1128–1133.

Hajny, G.J., 1966. Outside storage of pulpwood chips - a review and bibliography. Tappi 49 (1O), 97a–105a.
Hansbrough, J.R., 1953. Storage of northern hardwood logs and bolts. J. For. Prod. Res. Soc 3 (2), 33–35. 92.
Hatton, J.V., 1979. Chip Quality Monograph. Tappi Pulp and Paper Technology Series No.5. Technical Association of the Pulp and Paper Industry, Atlanta, Georgia.
Hulme, M.A., 1979. Outside chip storage - deteriorating during OCS. Chapt. 11. In: Hatton, J.V. (Ed.), Chip Quality Monograph. Joint Textbook Committee of Paper Industry Publications, Atlanta, Georgia.
Hulme, M.A., Stranks, D.W., 1976. Heat tolerances of fungi inhabiting chip piles. Wood Sci. 8, 237–241.
Humphrey, C.J., 1917. Timber Storage Conditions in the Eastern and Southern States with Reference to Decay Problems. Bulletin 510. U.S. Department of Agriculture, Washington, D.C.
Isaacson, L.H., 1958. New water fog spray method reduces log losses in storage. Wood Wood Prod. 63, 1, 25-26, 60-62.
Knuth, D.T., McCoy, E., 1962. Bacterial deterioration of pine logs in pond storage. For. Prod. J. 12 (9), 437–442.
Kress, O., 1925. Control of Decay in Pulp and Pulpwood. Bulletin 1298. U.S. Department of Agriculture, Washington, D.C.
Kubler, H., 1982. Heat release in thermally disintegrating wood. Wood Fiber 14 (3), 166–177.
Lane, P.H., Scheffer, T.C., 1960. Water sprays protect hardwood logs from stain and decay. For. Prod. J. 10, 277–282.
Lindgren, R.M., 1953. Deterioration losses of stored southern pine pulpwood. Tappi 36 (6), 260–263.
Lindgren, R.M., Harvey, G.M., 1952. Decay control and increased permeability in Southern pine sprayed with fluoride solutions. J. For. Prod. Res. Soc. 2, 250–256.
Lindgren, R.M., Eslyn, W.E., 1961. Biological deterioration of pulpwood and pulp chips during storage. Tappi 44 (6), 419–429.
Mason, R.R., Muhonen, J.M., Swartz, J.N., 1963. Water sprayed storage of Southern pine pulpwood. Tappi 46 (4), 233–240.
McKee, J.C., Daniel, J.W., 1966. Long-term storage of pulpwood in sealed enclosures. Tappi 49, 47a–50a.
Morrell, J.J., Corden, M.E., Graham, R.D., Kropp, B.R., Przybylowicz, P., Smith, S.M., et al., 1987. Basidiomycetes colonization of air-seasoned Douglas-fir poles. Proc. Am. Wood Preserv. Assoc. 83, 284–296.
Morrell, J.J., Graham, R.D., Corden, M.E., Sexton, C.M., Kropp, B.R., 1989. Ammonium bifluoride treatment of air-seasoning Douglas-fir poles. For. Prod. J. 39 (1), 51–54.
Newbill, M.A., Morrell, J.J., Levien, K.L., 1988. Internal temperature in Douglas-fir poles during treatment with ammoniacal copper arsenate or pentachlorophenol. Proc. Am. Wood Preserv. Assoc. 84, 48–54.
Panek, E., 1963. Pretreatments for the protection of southern yellow pine poles during air-seasoning. Proc. Am. Wood Preserv. Assoc. 59, 189–202.
Pascoe, T.A., Scheffer, T.C., 1950. Jack pine pulpwood deterioration in yard storage. Paper Trade J. 131, 20–21.
Przybylowicz, P., Kropp, B.R., Corden, M.E., Graham, R.D., 1987. Colonization of Douglas-fir poles by decay fungi during air-seasoning. For. Prod. J. 37 (4), 17–23.
Scheffer, T.C., 1969. Protecting stored logs and pulpwood in North America. Mater. Org 4, Bd. Heft 3.

Scheffer, T.C., Zabel, R.A., 1951. Storage of Beech Logs and Bolts in the Northeast. Northeast Forest Experiment Station, Forest Service, USDA.
Scheffer, T.C., Jones, T.W., 1953. Control of Decay in Bolts and Logs of Northern Hardwoods During Storage. Paper 63. U.S.D.A. Northeast Forest Experiment Station, Broomall, PA, 15 pp.
Scheffer, T.C., Eslyn, W.E., 1976. Winter treatments protect birch roundwood during storage. For. Prod. J. 26 (1), 27–31.
ScheId, H.W., DeGroot, R.C., 1971. Toughness of sapwood in water-sprayed longleaf pine logs. For. Prod. J. 21 (4), 33–34.
Schmidt, R.L., 1990. The effect of wood chip inventory rotation policies on storage costs, chip quality, and chip variability. Tappi 73 (11), 211–216.
Seifert, K.A., Breuil, C., Rossignol, L., Best, M., Saddler, J.H., 1988. Screening microorganisms with the potential for biological control of sapstain on unseasoned lumber. Mater. Org. 23, 81–95.
Shields, J.K., 1969. Microflora of eastern Canadian chip piles. Mycologia 61, 1165–1168.
Shields, J.K., Atwell, B.A., 1963. Effect of a mold, *Trichoderma viride*, on decay of birch by four storage-rot fungi. For. Prod. J. 13, 262–265.
Smith, S.M., Graham, R.D., Morrell, J.J., 1987. Influence of air-seasoning on fungal colonization and strength properties of Douglas-fir pole sections. For. Prod. J. 37 (9), 45–48.
Springer, E.L., Hajny, G.J., 1970. Spontaneous heating in piled wood chips. 1. Initial mechanism. Tappi 53 (1), 85–86.
Tansey, M.R., 1971. Isolation of thermophylic fungi from self-heating industrial wood chip piles. Mycologia 63, 537–547.
Taylor, A., 1980. Pretreatment decay in poles. Proc. Am. Wood Preserv. Assoc. 76, 227–245.
Volkmann, D., 1966. Water spray storage of southern pine pulpwood. Tappi 49, 48A–53A.
Wang, C.J.K., 1965. Fungi of Pulp and Paper in New York. Technical Publication 87. State University of New York, College of Forestry, Syracuse, N.Y., 115p.
White, M.S., Curtis, M.L., Sarles, R.L., Green, R.W., 1983. Effects of outside storage on the energy potential of hardwood particular fuels: Part 1. Moisture content and temperature. For. Prod. J. 33 (6), 31–38.
Zabel, R.A., Lombard, F.F., Kenderes, A.M., 1980. Fungi associated with decay in treated Douglas-fir transmission poles in the northeastern United States. For. Prod. J. 30 (4), 51–56.

Further reading

Thornqvist, T., Lundstrom, J., 1982. Health hazards caused by fungi in stored wood chips. For. Prod. J. 32 (11/12), 29–32.

CHAPTER FOURTEEN

Wood molds, stains and discolorations

Contents

The natural beauty of wood is a major reason for its widespread use in construction and furniture and many users pay a premium for clear, defect-free wood. Unfortunately, a number of chemical and biological agents can discolor wood, reducing its aesthetic and, sometimes, structural value. The majority of objectionable discolorations appear in the sapwood during seasoning and storage and are primarily biological in origin.

Stains or discolorations can be broadly defined as abnormal color patterns that develop on or in wood and adversely affect its value. Most stains are readily delineated from normal color patterns. Recognition of stain depends on familiarity with normal wood color variations including heartwood/sapwood differences, color variations associated with buried sapwood, reaction wood, wound zones and annual ring variations.

Molds and stains cause significant losses in wood quality worldwide, but the exact cost of these losses is difficult to assess. Scheffer estimated that annual losses due to stain exceeded 50 million dollars in the United States (1973). Losses generally increase with increasing sapwood content and become more severe as climate becomes more conducive to fungal growth. Losses have become more severe in recent years due to an emphasis on natural finishes, a customer fear of moldy wood and the growth of an export market for clear, light wood timbers. In some regions, builders prefer green or unseasoned lumber because it is easier to nail. This material remains susceptible to biological attack until it dries.

Wood Microbiology.
DOI: https://doi.org/10.1016/B978-0-12-819465-2.00014-0

There can be considerable delays between the time a tree is cut and it dries inside a structure. Mold and stain issues in the U.S. have encouraged the installation of dry kilns at many facilities to remove moisture as quickly as possible after sawing, but this adds to costs and increases the use of energy, making the use of wood less environmentally attractive from a life-cycle analysis perspective.

This chapter will review the historical significance of stains, describe the major types of stains associated with different wood species, and emphasize methods for preventing stain development.

Historical

For many years, the presence of some stain was accepted by wood users, but the shift to second-growth timbers with higher percentages of stain susceptible sapwood along with the importation of tropical hardwoods that tended to stain in transit, encouraged efforts to determine stain causes, to identify the organisms associated with biological stains, and to develop effective prevention measures (Boyce, 1927). Four comprehensive publications summarized the status of sapstain research from 1935 to 1959 and presented effective control strategies, many of which are still practiced (Scheffer and Lindgren, 1940; Verrall, 1945; Verrall and Mook, 1951; Findlay, 1959). Most mold/stain prevention strategies depend upon either rapid processing to dry the wood below the fiber saturation point or surface application of prophylactic biocides to prevent microbial colonization of the wood. Sodiurn pentachlorophenate and ethyl mercury phosphate were the primary biocides used initially for this purpose, but increasing concerns about the safety of these chemicals stimulated research to identify safer materials. The protection period might be limited to the early months of air seasoning, but it can extend for as long as a year with solid-piled unseasoned lumber for export shipments.

Types of wood discoloration

Wood discoloration can be conveniently grouped into 4 general categories based upon the source of discoloration as follows:

- Color changes resulting from enzymatic and chemical activity that develops on the surface or deep in the wood.

- Color changes caused by contact with chemicals
- Color changes associated with the early stages of decay
- Color changes associated with the growth of fungi on the wood surface or deep within the sapwood

The first two types are commonly referred to as chemical, enzymatic or oxidative stains, while the latter two are biotic stains.

Enzymatic and chemical stains

Many lumber discolorations result from chemical changes in cell contents shortly after the wood is exposed to air. These oxidative stains have long been studied (Bailey, 1910), but they remain poorly understood. These reactions are analogous to the browning reactions that occur in freshly cut fruits and may be related to the defense reactions of wounded tissues in living stems.

Kiln brown or coffee stain - These dark discolorations develop on the surface or deep within ponderosa pine, white pines (eastern, western, and sugar), or western hemlock wood during kiln drying (Hubert, 1926; Barton and Gardner, 1966). The dark-brown color is frequently concentrated at the ends of the board, in the vicinity of knots and at the heartwood-sapwood interface. The stain develops in both the sapwood and heartwood and is characterized by longitudinally oriented lenticular-brown masses, with occasional white zones or bands between the discolored zones (Fig. 14.1A). Kiln-brown stain is believed to occur as the result of an enzymatic reaction involving a peroxidase and subsequent oxidation or polymerization of a leuco-product in a two-step chemical process (Stutz, 1959). High kiln temperatures then cause the polymerization and oxidation that produce the colored compounds (tannins and phlobotannins). Peroxidase activity on phenolic extractives is apparently accelerated at moisture and oxygen levels present in freshly sawn boards during such periods. The stain appears more frequently during warm humid periods and is exacerbated by bulk piling of freshly sawn lumber. The stain is deeply penetrating and cannot be removed by surface planing (Fig. 14.1B). The risk of stain can be minimized by using mild kiln schedules where the dry-bulb temperature does not exceed 150°F (65.5 °C). Formerly, dip applications of sodium azide and sodium fluoride were found to prevent kiln brown stain (Stutz, 1959; Stutz et al., 1961;

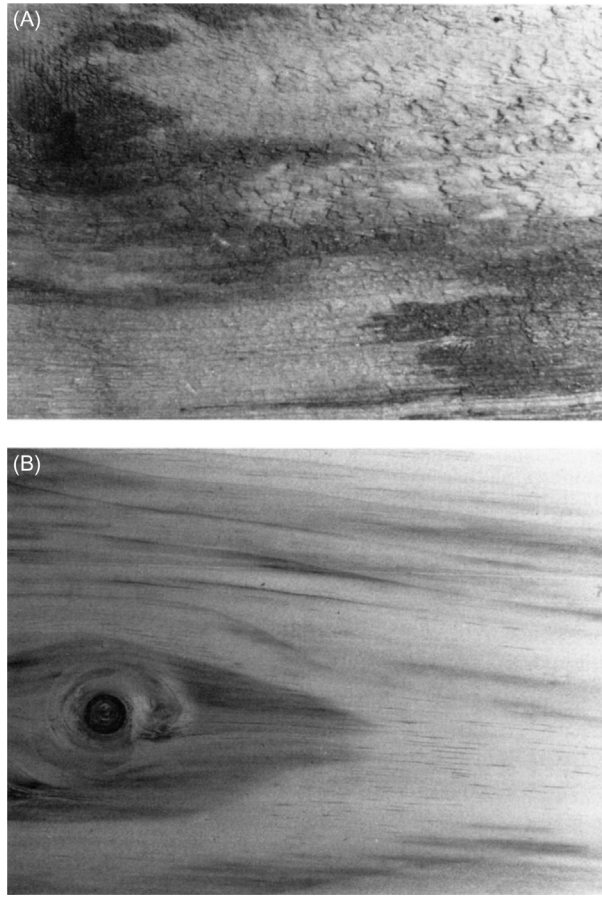


Figure 14.1 Chemical brown or coffee stain on kiln-dried white pine (A) the appearance on the unplanned board surface and (B) the same stain on a planed board, indicating that the stain develops deep in wood tissues. Note the more intense stain around the knot.

Cech, 1966); however, these compounds are toxic and safety concerns have largely limited their use. Ammoniacal zinc oxide (Shields et al., 1973) and several alkaline salts (Hulme, 1975) were shown to provide some control. Phosphoric acid and several β-hydroxyquinoline compounds are environmentally acceptable and control brown stain in sugar pine under laboratory conditions (Oldham and Wilcox, 1981). Hulme and Thomas (1983) found that dipping freshly sawn boards in 5% aqueous sodium sulfite or sodium thiosulfate was an effective and safe control of brown stain in eastern white pine.

A similar stain has also been noted on Douglas-fir and develops rapidly under moist, warm conditions (Miller et al., 1983). Water-soluble extractives migrate to the wood surface, where they undergo oxidation to produce a brown, polymerized pigment. The stain usually is close to the surface of the board, but has been observed deeper in bulk-piled boards. Brown stain appears sporadically, making it difficult to collect material for routine study.

Developing a more comprehensive understanding of the nature of oxidative stains might provide more useful insights into their prevention. One hypothesis for certain oxidative stains suggests that bacteria in the freshly harvested wood either alter wood pH or secrete enzymes that promote the formation of pigmented compounds (Yazaki et al., 1985). Attempts to confirm this effect in the brown-oxidative stain of Douglas-fir have, so far, proven elusive.

Brown stains are not always oxidative. Brown sapstain caused by a *Cytospora* sp. commonly occurs on several hard pines (Jackpine, red pine, and ponderosa pine) and occasionally on white pines (eastern, western and sugar) in northern regions. These biological stains can be confused with the chemical brown stain (Hubert, 1931; Fritz, 1952). Despite the similar brown colors, *Cytospora* stain is limited to the sapwood and produces numerous dark flecks in the wood. In addition, the whitish margins that characterize chemical brown stain are absent in the fungal colonized wood. An abundance of large, septate, brown hyphae in *Cytospora* stained wood cells readily separates the similarly colored stains microscopically. The *Cytospora* stain only develops in logs after prolonged storage and studies indicate that the stained wood is not weakened (Fritz, 1952).

Oxidative stains of hardwoods - Many hardwoods develop deep-yellow to reddish brown discolorations on the surface when exposed to air immediately after sawing or peeling. These discolorations are especially noticeable on cherry, birch, red alder, sycamore, oak, maple, and sweet gum. Stain develops in red alder, the oaks, birch and maple during air seasoning, and is intensified at the point where a sticker contacts the wood, hence the common name "sticker stain". This stain is absent in wood that is immediately kiln dried and appears less frequently under cool conditions. In 1910, Bailey had already established the probable enzymatic nature of this stain in alder and several birches and reported that it could be controlled by immersion in boiling water for several minutes to denature the enzymes that contributed to this process. This method has not proven practical and is rarely used. Kitchens (1997) showed that repeated

surface impacts sharply reduced the incidence of oxidative stains on the surfaces of hardwood lumber. They attributed this reduction to disruption of the ray parenchyma, thereby dispersing cell contents into the surrounding tissues. Limited commercial trials suggested that this process might be useful for higher value lumber. There are no other reported methods for preventing these stains, although treating stickers with a 4% solution of sodium hydrogen sulfite appears to minimize the intensity of sticker stain.

A related gray stain on several southern oaks also appears to be oxidative (Clark, 1956) and preliminary field studies suggested that it could be prevented by dipping boards in a 10% sodium bisulfite water solution and storing, solid-piled for 14 days under cover prior to seasoning (Forsyth, 1988).

Mineral stain or streak - This is a puzzling degrading stain that appears in several hardwoods, particularly maples in the Northeast and the Lake States (Scheffer, 1939, 1954). The stain is variable in occurrence and may appear in streaks or as a broad discoloration; moreover, it may be present in small clusters of stems in a stand and be absent elsewhere. Mineral stain appears in both the sapwood and heartwood of lumber, although it develops in the sapwood of the living tree. The stain may reflect a tree response to multiple or successive wounds in the sapwood zone; however, little is known about the cause and there is some confusion about the range of discolorations. Mineral stain on sawn maple generally appears as lenticular-shaped streaks ranging in color from a deep-olive to a green-black color. The streaks will often evolve small bubbles of CO_2 when treated with a mineral acid, denoting deposition of carbonates. The stained wood is denser and harder than normal wood and heavily stained wood tends to badly twist and warp when dried (Fig. 14.1C). The wood also tends to split when nailed and is essentially useless for construction purposes.

Examination of stained versus sound wood indicated that the mineral content of stained wood was about one-third greater and had a much higher pH than sound wood (Good et al., 1955). Levitin (1970) found that parenchyma cells in mineral stained wood were filled with numerous brown amber deposits, that soluble polyphenols were present at higher levels in stained wood, and found that condensed polyphenols deposited on the cell walls were not easily removed by solvents or bleaches. He also noted that tannate salts of Mg, K, and Ca formed compounds that ranged from brown to green in color. Based upon these findings, he proposed that wounding resulted in enzymatic hydrolysis of phenolic glycosides to

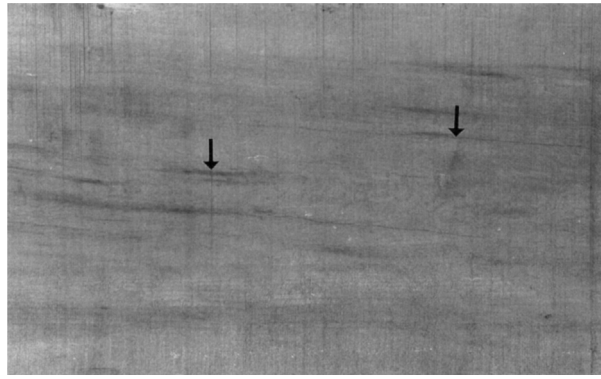


Figure 14.2 Mineral stain on the surface of an air-seasoned sugar maple board showing the characteristic lenticular-shaped streaks of a deep olive to green-black color (arrows). The board was also cupped, warped and unusually dense.

polyphenols that were, in turn, oxidized to form pigmented compounds. A similar yellow, purple, or brown-hued pattern of variegated colors may develop in the lower stem of living yellow poplar trees and is commonly termed "blue butt" or mineral stain by lumbermen. This stain is also believed to develop in the sapwood of living trees following wounding and is grouped with the oxidative stains (Roth, 1950).

Iron Stain - An intense black stain sometimes develops on the freshly sawn sapwood of some species when the woods come in contact with iron (Fig. 14.2). Iron tannate forms on the wood surface of lumber with high tannin contents such as the birches, cherry, sweetgum, and oaks, but is easily removed by planing (Fig. 14.1D). Iron stain is also common around nails in coniferous lumber. The blemishes can also develop when soils with high iron content fall on wet lumber. Dipping material in sodium carbonate solutions prevents the discoloration, while oxalic acid can be used to remove the stain. Alternatively, the use of non-ferrous metals or coating equipment wherever wood contacts machinery in the sawmill can eliminate iron stain; however, this is expensive and largely impractical.

Color changes associated with incipient decay

A number of subtle color changes may also occur as wood is degraded and the wood may develop shades of red, brown, purple, gray

or mottled white (see early decay in Chapter 8). In general, these discolorations may be distinguished from those caused by typical staining fungi by the presence of dark zone lines, textural changes on the sawn surface, or an irregular shape of the color patterns that may not coincide with annual rings.

Fungal stains or molds

Stains and molds are often used interchangeably. For our purposes, mold will refer to fungi whose clear (hyaline) hyphae grow through the wood cells and then produce pigmented spores on the wood surface. Stain fungi produce darker pigmented hyphae as they grow through the wood.

Molds - Molds were once considered to be of minor importance because their damage was mostly aesthetic. However, concerns over chemicals produced by some mold species as well as overall concerns about the possible role of fungal spores as allergens have heightened awareness. Molds are primarily a factor in very wet wood, such as wood that has been solid-piled or wood that has been covered to restrict aeration. Most molds are airborne-opportunistic fungi with hyphae that are normally colorless, but discolor the wood by forming masses of pigmented spores on the wood surface. Mold discoloration of coniferous woods can most often be removed by brushing or planing the wood surface; however, the discoloration on hardwoods is often deeper and may be more permanent. Common mold fungi and their discolorations include: *Aspergillus* spp. (black), *Fusarium* spp. (red or violet), *Gliocladium* spp. (green), *Monilia* spp. (orange), *Penicillium* spp (green), *Rhizopus* spp. (black), and *Trichoderma* spp. (green). Some molds, such as *Monilia* and *Aspergillus* species can cause both allergic reactions and worker health problems.

The molds of greatest concern are those species that produce aflatoxins. These chemicals have a variety of toxic effects and appear to be produced as metabolites by some fungi. *Stachybotrys atra* is, by far, the best known of these organisms. However, this fungus is much more common on cellulosic materials such as building paper. While *S. atra* aflatoxins are potent, there is little scientific evidence to suggest that this mold is any more of a risk. Despite the generally emerging view that mold effects are

largely limited to allergens, there have been a number of high-profile lawsuits over so-called toxic molds. The results has been public demand for mold free lumber.

Molds are particularly common on hardwoods when the wood is very wet, but some can develop on any wood held for long periods at high relative humidity. There is vigorous debate about the ability of mold fungi to grown on wood without free water. However, the difficulty in limiting condensation at the high relative humidity levels used in these tests makes it difficult to say with any certainty that mold fungi do not require free water to grow.

Molds tend to enter ruptured cells, vessels (hardwoods), and exposed rays, and spread from cell to cell via the pits. As they attack the pit membranes, these fungi make the wood more receptive to fluids. Treatments with molds have been proposed as a method for improving the permeability of Douglas-fir, the spruces and other difficult to treat species (Schulz, 1956, Lindgren and Wright, 1954). *Trichoderma* colonization, however, has also been shown to inhibit colonization of pine pulpwood by decay and stain fungi during storage (Lindgren, 1952; Hulme and Shields, 1972). The principles behind this deterrent effect have been explored for possible biological protection of wood from decay fungi with mixed success.

In addition to their protective and permeability effects, a number of molds have been shown to detoxify preservatives (Brown, 1953; Stranks and Hulme, 1975; Verrall, 1949). These include: *Penicillium (cyclopium) aurantiogriseum* on mercury compounds, *Scopulariopsis brevicaulis* on arsenic compounds, *Hormoconis* (*Cladosporium) resinae* on creosote, and *Trichoderma* sp. on sodium fluoride. The role of these fungi and the significance of their detoxification abilities on treated wood in the natural environment remains unknown, but under ideal conditions, they could detoxify preservative treatments, permitting decay fungi to colonize the treated wood. In general, there is a lack of knowledge on the roles of microfungi in wood and this remains a fertile area for further research.

Sapstains -Major sapstains are caused by fungi with pigmented hyphae that grow primarily in the parenchyma tissues of the sapwood (Fig. 14.3). Common sapwood discolorations are shades of blue, black, brown, or gray. Occasional stains of lesser importance are yellow, pink, purple, and green. These latter stains are the result of pigments secreted from the fungal hyphae. The predominance of a bluish discoloration and restriction to the sapwood have resulted in the major stains being termed blue stain or

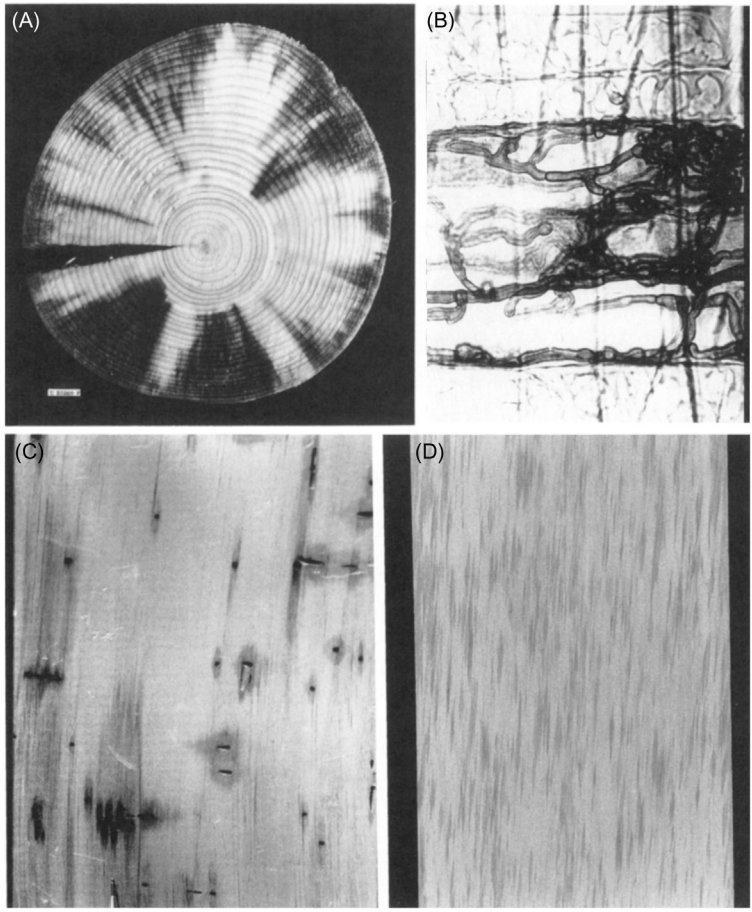


Figure 14.3 Sapstain in eastern white pine. (A) A cross section of pulpwood bolt showing the wedge-shaped pattern of stain development in the sapwood resulting from surface infections in shallow checks and rapid growth of the fungus through the ray tissues. (B) Photomicrograph of the radial section of wood stained by Aureobasidium pullulans showing preferential colonization of parenchyma cells in the wood rays by dark pigmented hyphae (~600 ×). (C) Ambrosia beetle attack of log showing general sapstain on the log as well as stain from the symbiont that is restricted to areas adjacent to the larval tunnels, (D) Multiple infection sites on the surface of a board that was solid piled for several weeks after sawing. *Photograph (A) courtesy: R.C. DeGroot, US Forest Service.*

sapstain. Fungal stains are worldwide in distribution, but become a more serious problem when species containing high percentages of sapwood are harvested and seasoned under warm, humid conditions that are suitable to rapid fungal growth.

Sapstain poses major challenges in the Southeastern United States due to the ideal climate for fungal growth, coupled with the large percentages of sapwood present in species such as the southern yellow pines, sweetgum, and yellow poplar. White pine in the Northeast and Lake states is similarly affected, but on a more seasonal basis. Similar conditions in the spring and fall in the Western United States make stain prevention difficult during air-seasoning of sugar and ponderosa pine. Sapstain is also a major hazard in western white pine and ponderosa pine in the Inland Empire (Idaho, Montana, Wyoming) during the warmer months. The export of solid-piled green lumber in the Pacific Northwest has led to increasing stain problems in Douglas-fir and western hemlock lumber (Cserjesi, 1977).

In all of these cases, fungi rapidly colonize the ray parenchyma to utilize the readily available storage carbohydrates (Fig. 14.3). A study of staining of lodgepole pine showed that proteins, fatty acids and other compounds stored in the parenchyma were important for growth of stain fungi (Alamouti et al., 2009; Kim et al., 2005). As they grew through the wood, the pigmented hyphae of the major fungi discolored the wood. Interestingly, hyphae often produce dark, melanin-based pigments (Zink and Fengel, 1988, 1989, 1990), but the effect of these darkened hyphae is to color the wood blue. Virtually all stain fungi are in the Ascomycetes. Several of the important staining fungi in the major timber species in the United States are listed in Table 14.1. Kaarik (1980) assembled a useful,

Table 14.1 Some important sapwood staining fungi in the United States[a,b].

Conifers	Hardwoods
Ceratocystis ips	*Ceratocystis pluriannulata*
Ophiostoma (*Ceratocystis*) *piliferum*	*Ceratocystis moniliformis*
Ophiostoma (*Ceratocystis*) *piceae*	*Lasiodiplodia theobromae* (*Diplodia natalensis*)
Aureobasidium pullulans	*Graphium rigidum*
Alternaria (*tenuis*) *alternata*	
Cephaloascus fragrans	
Cladosporium spp.	
Lasiodiplodia theobromae	
Phialophora spp.	

[a]Scientific names are based on current listings in Farr et al. (1989). In some cases, names that appear commonly in the older literature are placed in parenthesis.
[b]Sources: Scheffer and Lindgren (1940); Verrall (1939, 1942): Roff (1973); Davidson (1935, 1942, 1971): Davidson and Eslyn (1976); Zabel (1953); Kaarik (1980).

detailed list and described the principal features of the sapwood staining fungi in temperate zones.

Many stain fungi are specific to a region or wood species. For example, Kang and Morrell (2000) studied freshly sawn Douglas-fir lumber and found over 40 species colonized the wood with 6 weeks. Fungal flora also tended to shift with time. The staining fungi can be placed into two broad groups. Some of the fungi, particularly those in the genera *Ophiostoma* and *Ceratocystis*, are closely tied with the life cycles of bark beetles and other wood inhabiting insects (Verrall, 1941; Dowding, 1969, 1970). They produce sticky spores that are primarily transmitted by insects (vectors) as well as water splashing or aerosols (Fig. 14.3C). These fungi invade and damage wood primarily during log storage and the initial stages of lumber seasoning. The other group of stain fungi, such as *Aureobasidium pullulans*, *Alternaria alternata*, and *Cladosporium* sp. are general opportunists, whose dry spores are primarily air-disseminated. These fungi invade wood in a wide range of uses when conditions are conducive to fungal growth.

Like all biological agents, stain fungi require free water, adequate temperature, oxygen, and a food source. Mill operators have long exploited their need for oxygen by flooding or spraying logs to raise the moisture content and exclude oxygen from the wood surface (see Chapter 13 on Biodeterioration of Wood During Storage). Stain fungi grow at a broad range of temperatures (4–30 °C) making it difficult to avoid stain development without special measures except in the coolest and driest climates. Altering the wood through application of biocides essentially alters the nutritional value of the substrate.

Stain Development: Fungal stains most often originate from the germination of spores on the freshly-sawn wood surface. These spores are mostly airborne, or carried by insect vectors such as a bark or ambrosia beetles. Stain can also develop when hyphae from previously colonized stickers grow into the freshly-sawn wood. Sawmill machinery can also serve as an inoculum source infecting boards for some time after a badly stained log has been sawn. Under favorable staining conditions, a single board may develop scores of distinct stained zones in as little as 24–72 hours (Fig. 14.3). Under favorable conditions, spores germinate within hours of landing and penetrate the wood surface through ruptured tracheids and exposed wood rays. The hyphae then rapidly colonize the parenchyma cells in the wood rays or longitudinal parenchyma surrounding the resin canals via direct pit penetration. Stain fungi can grow up to

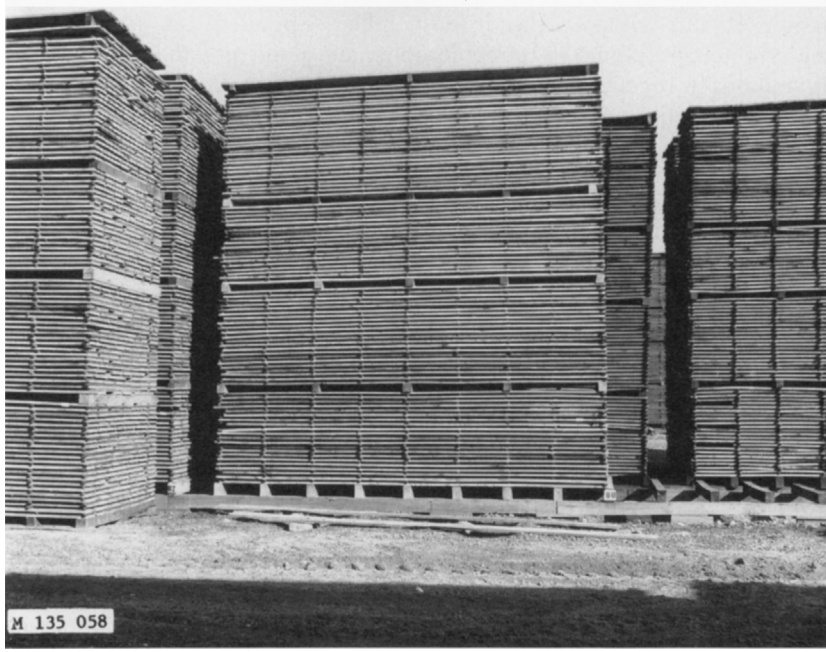


Figure 14.4 Example of a lumber pile showing proper piling procedures that include a roof with an overhang to shed water, separation between the soil and the wood, sticker spacing to allow for air flow and the use of narrow piles to facilitate air circulation. *Photo courtesy Forest Service, U.S. Department of Agriculture.*

0.5 mm in the tangential plane, 1 mm radially, and 5 mm longitudinally over a 24-hour period under ideal conditions (Lindgren, 1942). Movement inward along the rays from the sides is responsible for the wedge-shaped stain forms often seen on the cross-cut surfaces of logs or pulpwood bolts (Fig. 14.4). Mechanized logging practices that severely damage the bark have markedly increased the risk of stain development following felling. The rapid fungal growth rate permits extensive colonization of freshly sawn materials and highlights the importance of stain control at the earliest possible point after sawing. Many times, pigmentation develops 5–6 days after hyphal development, making it appear that the stain literally exploded throughout the wood when in fact it had been there all along. After several weeks, the parenchyma cell walls are often badly eroded and hyphae occasionally penetrate the wood cell walls directly to move between tracheid and fiber walls in the radial plane. A few stain fungi, upon prolonged incubation under ideal moisture

conditions, will act as typical soft-rot fungi and attack the S-2 layer of tracheid walls. These species include *Alternaria alternata*, *Phialocephala dimorphospora* and *Ophiostoma picea* (Levy, 1967; Wang and Zabel, 1990).

Effects of Fungal Stain on Wood Properties - Stain fungi produce both aesthetic and physical changes in wood value. The dark, melanistic pigments produced by stain fungi consume more bleach when stained wood is pulped, thereby increasing paper production costs. Conversely, stained wood has aesthetic value to some users who purchase "blue pine" panels that are actually stained wood. Some fungi such as *Scytalidium* species, produce pigments that color the wood green or red, and wood with these colors has been used in a wooden artform called intarsia (Vega-Gutierrez, and Robinson, 2017).

While the most obvious effect of stain is wood discoloration, some stain fungi also alter other wood properties. Reductions in toughness have been noted for some fungi, but the strength effects are not consistent among stain fungi (Chapman and Scheffer, 1940). For example, *Leptographium lundbergii* has no effect on toughness, while toughness losses associated with *Alternaria alternata* may approach 40% (Crossley, 1956). Stained wood is generally not recommended for structural purposes where strength is critical and is not used for structures such as utility poles, glue laminated timbers, ladders, or piling. Conditions favorable for stain development are also conducive to decay initiation, so decay and stain development in wood are often coupled. In addition to strength effects, stain fungi increase wood permeability by removing pit membranes. As a result, the wood will wet and dry more rapidly. These effects are particularly important in the finishing industry since stained wood will absorb excessive solution and will often finish unevenly. This material will also absorb water more quickly in service, increasing the development of checks that provide entry points for decay fungi (Bjorkman, 1947). Although stained wood appears to be no more susceptible to decay than sound wood, greater hygroscopicity in stained wood tends to create conditions conducive to fungal growth for longer periods of time, increasing the risk of fungal decay. Furthermore, stained wood may provide an ideal inoculum source for paint disfiguring fungi (Zabel and Terracina, 1980).

Sapstain Control: Stain control can be accomplished either through rapid drying to reduce wood moisture content or dipping or spraying with fungicidal solutions to protect the surface against fungal invasion. Dipping is relatively inexpensive: 1000 bd. ft. of one-inch rough lumber absorbs about 15 gallons of treating solution costing pennies per gallon.

Where stain conditions are severe, both fungicidal protection and good drying practices are required.

Drying procedures can include kiln drying or employing proper piling procedures during air-seasoning to promote extensive air-flow around the wood (Fig. 14.4). Kiln drying represents the best alternative in the southeastern U.S., although air-seasoning is often used for larger timbers such as railroad ties. Decay and stain will undoubtedly occur during air-seasoning and are often referred to as "stack burn". Air-seasoning is commonly used in the western United States, where drier conditions and wood species containing lower percentages of stain susceptible sapwood make these practices feasible. It is important to note that decay fungi can still invade wood in a properly configured air-seasoning yard and can survive for long periods afterwards. This makes it especially important to avoid wetting in structures once the wood is installed.

Chemical treatments for preventing stain have been employed since the early 1900's, when water solutions of sodium carbonate or borax were applied to the wood. These chemicals were relatively mobile and provided relatively short-term protection. In the 1930, significant losses to fungal stains in the southern and western pines led to an extensive chemical evaluation program by the U.S. Department of Agriculture, resulting in the testing and use of chlorinated phenols and organic mercury compounds (Scheffer and Lindgren, 1940; Verrall and Mook, 1951; Scheffer and Drow, 1960; Zabel, 1953; Zabel and Foster, 1949). These chemicals were traditionally applied to lumber by dipping in tanks or by passing through a spray of the chemical. Concerns about worker exposure and environmental contamination have led to the development of enclosed spray booths that minimize chemical exposure and reduce the amount of total liquid applied. These systems are generally located immediately after the planer. Spray systems allow the use of lower amounts of liquid, but they require more maintenance since a clogged nozzle can result in large volumes of wood being left unprotected. These errors are often only caught when the lumber has developed mold or stain further in the supply chain.

In the late 1960, concerns about the safety of mercury compounds resulted in the elimination of this chemical and most mills depended on sodium pentachlorophenate (Penta) or tetrachlorophenate, alone or with borax, for stain prevention. The presence of dioxins in some penta products led to use of alternative chemicals for dipping and spraying in North America.

The restriction of penta resulted in extensive evaluation programs to identify acceptable alternatives (Cserjesi, 1980; Cserjesi and Roff, 1975; Cserjesi and Johnson, 1982; Drysdale and Preston, 1982; Cassens and Eslyn, 1983; Eslyn and Cassens, 1983; Hulme and Thomas, 1979; Lewis et al., 1985; Presnell and Nicholas, 1990; Unligil, 1979; Miller and Morrell, 1989; Miller et al., 1989). Compounds currently used in anti-sapstain formulations include oxine copper, methyl thiobenzothiazole, methylene bisthiocyanate, propiconazole, tebuconazole, 3-iodo-2-propynyl butylcarbamate, and several quaternary ammonium compounds. These chemicals are usually employed in mixtures to overcome resistant species. In general, all of the acceptable alternatives are more expensive and none has been as completely effective as penta. Interestingly, although there has been an overwhelming effort to develop acceptable alternative anti-stain chemicals, studies related to the fundamental nature of fungal stain have been overlooked. The chemical aspects of anti-stain treatments are covered in more detail in Chapter 20.

Proper handling of wood during harvesting and seasoning is critical for limiting the development of stains. Some important practices include:

- Rapid utilization, ponding, or spray storage of logs prior to milling to insure stain-free logs at the mill
- Prompt treatment of wood with anti-stain chemicals within 24 hours after sawing to prevent germination and growth of fungi beyond the penetration depth of treatment, resulting in internal stain
- Maintaining treatment solutions at recommended levels and replenishing treatment solutions frequently to minimize dilution of the fungicide by selective absorption
- Protecting dipped lumber from rain to insure that a sufficient level of chemical remains on the wood and to prevent chemical contamination of the local environment
- Only using stickers made of heartwood or preservative-treated wood
- Using proper piling practices to maximize air-flow through and over the boards in the pile. Lumber should be far enough off the ground to prevent rain splashing.

In some cases, properly handled wood may still develop some stain. This stain is usually related to poor handling procedures prior to the log reaching the mill. For example, the log may have been stored in the woods for some time before transport, permitting fungi to colonize the end and debarked areas or insects to carry fungal spores into the wood. This stain developed further once the log was cut into lumber.

Treatment as soon as possible can limit this damage, but higher chemical levels are often required since the fungus is already established in the wood.

In addition to chemical control, some researchers have explored the use of other organisms that out-compete the stain fungus. Several organisms have been found that might be useful for this purpose and have performed well in vitro, but have generally performed poorly under field conditions (Bernier et al., 1986; Seifert et al., 1988; Benko, 1988). Alternatively, it may be possible to identify the mechanisms of pigmentation and develop methods for inhibiting this process. Researchers have also isolated pigmentless *Ophiostoma* species that colonize the wood and inhibit the growth of other stain species (Berendt et al., 1995). These fungi are primarily used to remove resin acids from paper chips to reduce chemical consumption during pulping. In this case, the fungus would still attack the wood, but would not cause discolouration. The use of solvents or other treatments to bleach or mask fungal stains have been explored, but are largely not feasible (Lee et al., 1995).

Summary

1. Stains can be chemical or biological in nature. Chemical stains are primarily oxidative, leading to formation of pigmented compounds in the wood.
2. Controlling chemical stains is difficult, although reducing compounds and anti-oxidants have sometimes proven useful. Biological stains include incipient decay, molds and sapstains.
3. Stains appear to be becoming an important issue because of the increasing proportions of sapwood in second growth woods, the increased use of exposed wood in decorative applications where appearance is important and customer preferences for clear wood.
4. In addition to the aesthetic damage, stained wood is more permeable, often has substantial losses in toughness and its presence may be an indicator of incipient decay.
5. Application of chemicals, either through dipping or spraying can prevent fungal attack, but care must be taken to ensure that the treatments are properly timed and performed.

References

Alamouti, S.M., Tsui, C.K.M., Breuil, C., 2009. Multigene phylogeny of filamentous ambrosia fungi associated with ambrosia and bark beetles. Mycological Res. 113 (8), 822–835.

Bailey, I.W., 1910. Oxidizing enzymes and their relation to "sap stain" in lumber. Bot. Gaz. 50, 142–147.

Barton, G.M., Gardner, J.A.F., 1966. Brown Stain Formation and the Phenolic Extractives of Western Hemlock (*Tsuga heterophylla* (Raf.) Sarg]. Publication No. 1147. Department of Forestry, Ottawa, Canada, 20 p.

Benko, R., 1988. Bacteria as Possible Organisms for Biological Control of Blue Stain. International Research Group on Wood Preservation Document No. IRG/WP/1339, Stockholm.

Berendt, C.J., Blanchette, R.A., Farrell, R.L., 1995. Biological control of blue-stain fungi in wood. Phytopathology 85, 92–97.

Bernier Jr., R., Desrochers, M., Jurasek, L., 1986. Antagonistic effect between *Bacillus subtilis* and wood staining fungi. J. Inst. Wood Sci. 10 (5), 214–216.

Bjorkman, E., 1947. On the development of decay in building-timber injured by blue stain. Satryck ur. Sven. Papperstidning 50, 11B.

Boyce, J.S., 1927. Sapstain and decay in Northwest lumber. Columbia Port. Dig. 5 (3), 5–6.

Brown, F.L., 1953. Mercury tolerant Penicillia causing discoloration in northern white pine lumber. J. For. Prod. Res. Soc. 5, 67–69.

Cassens, D.L., Eslyn, W.E., 1983. Field trials of chemicals to control sapstain and mold on yellow-poplar and southern pine lumber. For. Prod. J. 33 (10), 52–56.

Cech, M.Y., 1966. New treatment to prevent brown stain in white pine. For. Prod. J. 16 (11), 23–27.

Chapman, A.D., Scheffer, T.C., 1940. Effect of blue stain on specific gravity and strength of southern pine. J. Agric. Res. 61 (2), 125–134.

Clark, J.W., 1956. A gray non-fungus seasoning discoloration of certain red oaks. South. Lumberm. 193 (2417), 35–38.

Crossley, R.D., 1956. The Effects of Five Sapstain Fungi on the Toughness of Eastern White Pine (Master's thesis). State University of New York, College of Forestry, Syracuse, NY, 41 p.

Cserjesi, A.J., 1977. Prevention of stain and mould in lumber and board products. Proceedings ACS Symposium No. 43 on Wood Technology: Chemical Aspects, pp. 24–32.

Cserjesi, A.J., 1980. Field-Testing Fungicides on Unseasoned Lumber-Recommended Procedure. Technical Report 16. Forintek Canada Corp., Vancouver, BC, 10 p.

Cserjesi, A.J., Roff, J.W., 1975. Toxicity tests of some chemicals against certain wood-staining fungi. Int. Biodeterior. Bull. 11 (3), 90–96.

Cserjesi, A.J., Johnson, E.L., 1982. Mold and sapstain control: laboratory and field tests of 44 fungicidal formulations. For. Prod. J. 32 (10), 59–68.

Davidson, R.W., 1935. Fungi causing stain in logs and lumber in the southern states, including five new species. J. Agric. Res. 50, 789–798.

Davidson, R.W., 1942. Some additional species of *Ceratostomella* in the United States. Mycologia 31, 650–662.

Davidson, R.W., 1971. *Ceratocystis* Species in North America Recognized by Davidson. I International Congress. Exeter.

Davidson, R.W., Eslyn, W.E., 1976. Some wood staining fungi from pulpwood chips. Mem. N.Y. Bot. Gard. 28, 50–57.

Dowding, P., 1969. The dispersal and survival of spores of fungi causing bluestain in pine. Trans. Br. Mycol. Soc. 52 (1), 125–137.

Dowding, P., 1970. Colonization of freshly bared pine sapwood surfaces by staining fungi. Trans. Br. Mycol. Soc. 55 (3), 399–412.
Drysdale, J.A., Preston, A.F., 1982. Laboratory screening trials with chemicals for the protection of green timber against fungi. N. Zealand J. For. Sci. 12 (3), 457–466.
Eslyn, W.E., Cassens, D.L., 1983. Laboratory evaluation of selected fungicides for control of sapstain and mold on southern pine lumber. For. Prod. J. 33 (4), 65–68.
Farr, D.F., Bills, G.F., Chamuris, G.P., Rossman, A.Y., 1989. Fungi on Plants and Plant Products in the United States. American Phytopathological Society Press, St. Paul, MN.
Findlay, W.P.K., 1959. Sap-stain of timber. For. Abstr. 20 (1,2), 1–14.
Forsyth, P.G., 1988. Control of Non-Microbial Sapstains in Southern Red Oak, Hackberry, and Ash Lumber During Air-seasoning (Master of Science thesis). Mississippi State University, MS, 50 p.
Fritz, C.W., 1952. Brown stain in pine sapwood caused by *Cytospora* sp. Can. J. Bot. 30 (4), 349–359.
Good, H.M., Murray, P.M., Dale, H.M., 1955. Studies on heartwood formation and staining in sugar maple, *Acer saccharum* Marsh. Can. J. Bot. 33, 31–41.
Hubert, E.E., 1926. Brown stains of lumber. Timberman May.
Hubert, E.E., 1931. Outline of Forest Pathology. John Wiley and Sons, Inc, New York, p. 543.
Hulme, M.A., 1975. Control of brown stain in eastern white pine with alkaline salts. For. Prod. J. 25 (8), 36–41.
Hulme, M.A., Thomas, J.F., 1979. Control of fungal sap stain with alkaline solutions of quaternary ammonium compounds and with tertiary amine salts. For. Prod. J. 29 (11), 25–29.
Hulme, M.A., Thomas, J.F., 1983. Control of brown stain in eastern white pine with reducing agents. Forest Prod. Res. Soc. 33 (9), 17–20.
Hulme, M.A., Shields, J.K., 1972. Effect of primary fungal infection upon secondary colonization of birch bolts. Mater. Org. 7, 177–188.
Kaarik, A., 1980. Fungi Causing Sap Stain in Wood. The Swedish University of Agricultural Sciences. Report Nr R 114. International Research Group on Wood Preservation Document No. IRG/WP/199, Stockholm, Sweden.
Kang, S.M., Morrell, J.J., 2000. Fungal colonization of Douglas-fir sapwood lumber. Mycologia 92 (4), 609–615.
Kim, J.J., Allen, E.A., Humble, L.M., Breuil, C., 2005. Ophiostomatoid and basidiomycetous fungi associated with green, red, and grey lodgepole pines after mountain pine beetle (*Dendroctonis ponderosae*) infestation. Can. J. For. Res. 35, 274–284.
Kitchens, S.C., 1997. The Use of Mechanical Forces to Control Non-Microbial Enzymatic Stains in Hardwood Lumber (MSc thesis). Mississippi State University, MS.
Lee, B.G., Maristany, A., Brunner, C.C., Morrell, J.J., 1995. Removing fungal stain from ponderosa pine by caustic bleaching. For. Prod. J. 45 (3), 56–60.
Levitin, N., 1970. Lignins of sapwood and mineral stained maple. Can. For. Serv. Bi-Mon. Res. Notes 27 (4), 29–30.
Levy, J.F., 1967. Decay and Degrade of Wood by Soft Rot Fungi and Other Organisms. International Pest Control (November/December), pp. 28–34.
Lewis, D.A., Williams, G.R., Eaton, R.A., 1985. The development of prophylactic chemicals for the treatment of green lumber. Rec. Annu. Conv. Br. Wood Preserv. Assoc. 1985, 14–26.
Lindgren R.M., 1942. Temperature, Moisture and Penetration Studies of Wood Staining Ceratostomellae in Relation to their Control, USDA Bulletin No. 807, Washington, DC.

Lindgren, R.M., 1952. Permeability of southern pine as affected by mold growth and other fungus infection. Proc. Am. Wood Preserv. Assoc. 48, 158–174.
Lindgren, R.M., Wright, E., 1954. Increased absorptiveness of molded Douglas-fir posts. J. For. Prod. Res. Soc. 4 (4), 162–164.
Miller, D.J., Morrell, J.J., 1989. Controlling Sapstain: Trials of Product Group I on Selected Western Softwoods. Research Bulletin 65. Forest Research Laboratory, Oregon State University, Corvallis, Oregon.
Miller, D.J., Knutson, D.M., Tocher, R.D., 1983. Chemical brown staining of Douglas-fir sapwood. For. Prod. J. 33 (4), 44–48.
Miller, D.J., Morrell, J.J., Mitchoff, M.E., 1989. Controlling Sapstain: Trials of Product Group II on Selected Western Softwoods. Research Bulletin 66. Forest Research Laboratory, Oregon State University, Corvallis, Oregon.
Oldham, N.D., Wilcox, W.W., 1981. Control of brown stain in sugar pine with environmentally acceptable chemicals. Wood Fiber 13 (3), 182–191.
Presnell, T.L., Nicholas, D.O., 1990. Evaluation of combinations of low hazard biocides in controlling mold and stain fungi in southern pine. For. Prod. J. 40 (2), 57–61.
Roff, J.W., 1973. Brown mould (*Cephaloascus fragrans*) on wood, its significance and history. Can. J. For. Res. 34, 582–585.
Roth, E.R., 1950. Discolorations in living yellow poplar trees. J. For. 48, 184–185.
Scheffer, T.C., 1939. Mineral stains in hard maples and other hardwoods. J. For. 37 (7), 578–579.
Scheffer, T.C., 1954. Mineral Stain in Hard Maples and Other Hardwoods. Report 1981. U.S.D.A. Forest Products Laboratory, Madison, Wisconsin.
Scheffer, T.C., 1973. Microbiological degradation and the causal organisms. In: Nicholas, D.D. (Ed.), Wood Deterioration and Its Prevention by Preservative Treatments. Syracuse University Press, Syracuse, N.Y.
Scheffer, T.C., Lindgren, R.M., 1940. Stains of Sapwood Products and Their Control. Technical Bulletin 714. U.S. Department of Agriculture.
Scheffer, T.C., Drow, J.T., 1960. Protecting Bulk-Piled Green Lumber From Fungi by Dip Treatment. Report 2201. U.S.D.A. Forest Products Laboratory, Madison, Wisconsin.
Schulz, G., 1956. Exploratory tests to increase preservative penetration in spruce and aspen by mold infection. For. Prod. J. 6 (2), 77–80.
Seifert, K.A., Breuil, C., Rossignol, L., Best, M., Saddler, J.N., 1988. Screening for microorganisms with the potential for biological control of sapstain on unseasoned lumber. Mater. Org. 23 (2), 81–95.
Shields, J.K., Desai, R.L., Clarke, M.R., 1973. Control of brown stain in kiln-dried eastern white pine. For. Prod. J. 23 (10), 28–30.
Stranks, D.W., Hulme, M.A., 1975. The mechanisms of biodegradation of wood preservative. Mater. Org. Symp. Berlin-Dahlem 3, 346–353.
Stutz, R.E., 1959. Control of brown stain in sugar pine with sodium azide. For. Prod. J. 9 (11), 59–64.
Stutz, R.E., Koch, P., Oldham, M.L., 1961. Control of brown stain in eastern white pine. For. Prod. J. 11 (5), 258–260.
Unligil, H.H., 1979. Laboratory screening tests of fungicides of low toxic hazard for preventing fungal stain of lumber. For. Prod. J. 29 (4), 55–56.
Vega-Gutierrez, P.T., Robinson, S.C., 2017. Determining the presence of spalted wood in Spanish marquetry woodwork in the 1500's to 1800'. Coatings 7, 188. Available from: https://doi.org/10.3390/coatings7110188.
Verrall, A.F., 1939. Re1ative importance and seasonal prevalence of wood staining fungi in the southern states. Phytopathology 29, 1031–1051.

Verrall, A.F., 1941. Dissemination of fungi that stain logs and lumber. J. Agric. Res. 63 (9), 549–558.
Verrall, A.F., 1942. A comparison of *Diplodia natalensis* from stained wood and other sources. Phytopathology 32 (10), 879–884.
Verrall, A.F., 1945. The control of fungi in lumber during air-seasoning. Bot. Rev. 11 (7), 398–415.
Verrall, A.F., 1949. Some molds of wood favored by certain toxicants. J. Agric. Res. 78 (12), 695–703.
Verrall, A.F., Mook, P.V., 1951. Research on Chemical Control of Fungi in Green Lumber, 1940–1951. Technical Bulletin 1046. U.S. Department of Agriculture.
Wang, C.J.K., Zabel, R.A., 1990. Identification Manual for Fungi From Utility Poles in the Eastern United States. American Type Culture Collection, Rockville, MD, 356 p.
Yazaki, Y., Bauch, J., Endeward, R., 1985. Extractive components responsible for the discoloration of Ilomba wood (*Pycnanthus angolensis* Exell.). Holz Als Roh-und Werkst. 43, 359–363.
Zabel, R.A., 1953. Lumber stains and their control in northern white pine. J. For. Prod. Res. Soc. 3, 1–3.
Zabel, R.A., Foster, C.H., 1949. Effectiveness of Stain Control Compounds on White Pine Seasoned in New York. N.Y. State College of Forestry at Syracuse University. Tech. Pub. No. 71, 17p.
Zabel, R.A., Terracina, F.C., 1980. The role of *Aureobasidium pullulans* in the disfigurement of latex paint films. Dev. Ind. Microbiol. 21, 179–190.
Zink, P., Fengel, O., 1988. Studies on the colouring matter of blue-stain fungi. Part 1. General characterization and the associated compounds. Holzforschung 42 (4), 217–220.
Zink, P., Fengel, O., 1989. Studies on the colouring matter of blue-stain fungi. Part 2. Electron microscopic observations of the hyphae walls. Holzforschung 43 (6), 371–374.
Zink, P., Fengel, D., 1990. Studies on the colouring matter of blue-stain fungi. Part 3. Spectroscopic studies on fungal and synthetic melanins. Holzforschung 44 (3), 163–168.

CHAPTER FIFTEEN

Decay problems associated with some major uses of wood products

Contents

One of the major drawbacks associated with the use of wood products is their susceptibility to biological deterioration. This deterioration often occurs as we inadvertently duplicate natural conditions, such as elevated moisture levels, for decay in our structures. These conditions can sometimes be prevented by proper structural design, but wood is often used in structures where it is exposed to ground contact or periodic wetting. When this is unavoidable, economic realities dictate the use of preservative treated or naturally durable woods. Despite these efforts, a substantial percentage of the wood in service fails to the agents of decay.

Quantifying decay losses has long stymied researchers. Wood failures occur in many different applications such as buildings, utility poles, railroad ties, bridge timbers, piling, and a myriad of other unrelated uses. With the exception of utilities and railroads, most wood users lack a systematic method for quantifying their decay losses and even these groups have a relatively imprecise knowledge of their losses. Quantifying losses in residential structures is particularly difficult since there are no uniform procedures for reporting damage. As a result, many potentially important decay problems may be overlooked.

Wood Microbiology.
DOI: https://doi.org/10.1016/B978-0-12-819465-2.00015-2

The replacement of decayed wood alone was estimated to consume 10% of the timber cut annually in the United States (Boyce, 1961). To place this figure in perspective, the 10 largest softwood sawmills in the U.S. produced 17.771 billion board feet of timber in 2017. If we use an average price of $350 per thousand board feet (the standard unit of lumber production in North America), this represents an average loss of 6.22 billion dollars per year. While wood decay results in substantial losses, labor costs involved in replacing structures, productivity losses or liability stemming from poorly maintained wood far exceed the raw value of the wood. The total cost of insect and decay repairs in buildings in California was estimated to approach 400 million dollars per year (Brier et al., 1988). Even simple examples of decay can sometimes cause significant productivity losses. For example, decaying ties decrease the speed at which trains can safely travel without the risk of derailment, thereby decreasing track usage and increasing transit times for trains. These slow-downs were estimated to cost $18.60 per tie per year in main line track (Anonymous, 1985). There are nearly 3000 ties present in a single mile of mainline track, meaning that the cost of decay can rapidly add up. The cost to replace a single utility pole in California approaches $8000 including labor and may also trigger costly service interruptions. Individual utilities often have 100,000 or more poles within their system and incur rejection rates of 0.3–0.4% per year-amounting to 2.4–3.2 million dollars annually in replacement costs. Furthermore, the liability associated with the failure of a wood pole can easily exceed several million dollars if a serious injury or fire is involved. On a more personal note, decay or insect attack in residential homes can markedly reduce home value.

It is readily apparent that we accept a certain level of decay loss within specific commodities; however, the need to maximize wood performance and increasing raw material costs will necessitate a more careful evaluation of wood usage. In this Chapter, we review the causes of decay losses in the major commodities where wood is employed and stress the principles and practices to prevent or minimize these losses.

Decay hazard

If there is no decay hazard, wood will last indefinitely in most interior uses and many structural applications where the material is kept dry.

Decay hazards are generally related to external uses of wood subjected to atmospheric wetting or other moisture sources such as soil contact.

Before we address decay problems associated with specific wood uses, it is important to consider that the risk of decay varies widely with climate and geographic location. This premise is employed in the specifications of the American Wood Protection Association through the incorporation of different levels of chemical protection that the user can specify for their particular region (AWPA, 2017). It is readily apparent that the risk of decay is considerably greater in southern Florida than northern Wyoming and that the degree of exposure has a marked influence on performance. Decay problems are minimal in the dry southwestern U.S. or at higher elevations, but become quite significant in the southeastern U.S. It is, however, less apparent that decay risks can vary widely within closely situated sites. These hazards often necessitate the use of either species with naturally durable heartwood or wood that has been preservative treated.

The variations in exposure were used by Scheffer (1971) to develop a climate index for decay hazard for various exterior wood uses above the ground (Fig. 15.1). This index uses rainfall and temperature data to develop an index rating ranging from a 0 (no risk) to 100 (high hazard). These values are then adjusted on the basis of known service records of wood in the various regions. The index establishes three broad hazard zones: severe decay hazard (southeastern U.S. and the Olympic Peninsula), moderate decay hazard (northeastern U.S., north and central states, and western Oregon to California), and low decay hazard (southwest, Rocky Mountains, and the eastern Pacific Northwest). While the climate index is useful for predicting performance of wood in non-ground contact applications, it cannot predict ground contact performance since it cannot account for variations in soil type, water holding capacity, and vegetation. The climate index provides a simple method for assessing relative risk of decay out of soil contact and has been adapted in a number of countries. It has been updated based upon more recent climate data and served as the basis for prediction systems in other countries (Brischke and Rapp, 2007; Carll, 2009; Morris and Wang, 2008, 2011). The Scheffer index is limited to above ground applications. Predicting service life in direct soil contact is much more complicated because of the diversity of possible decay agents and the contributions of the soil in terms of moisture holding capacity and nutrients. Leicester et al (2003) developed models for predicting decay in soil contact in Australia based upon extensive field stake trials, but much remains to be done with regard to predicting performance in soil contact.

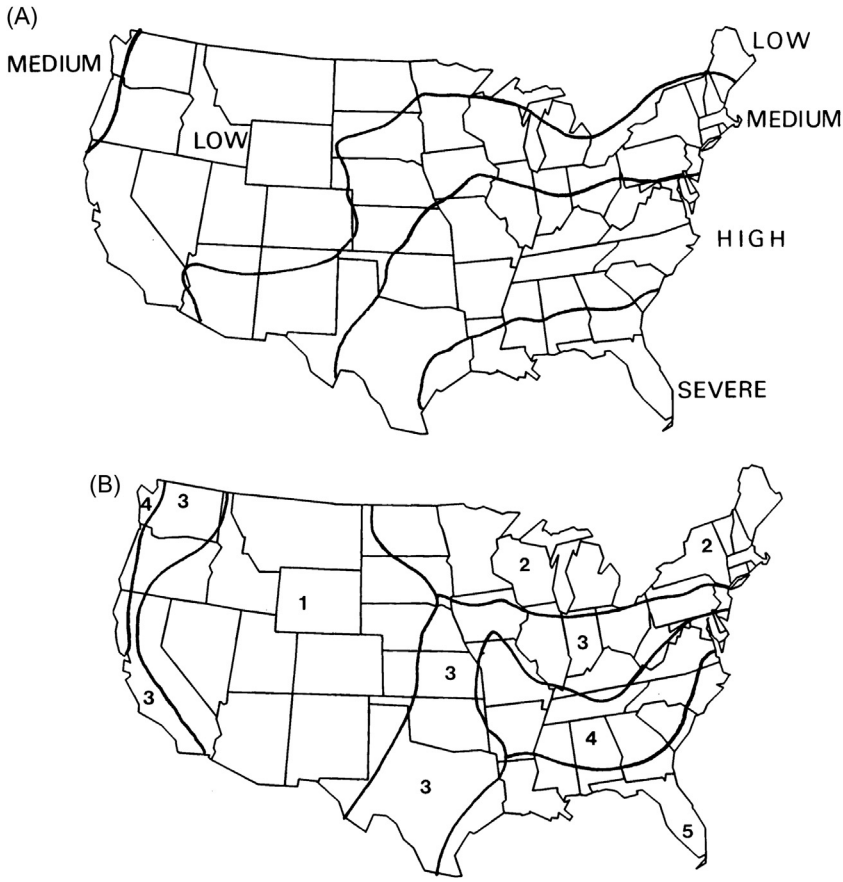


Figure 15.1 Decay hazards for (A) aboveground exposure and (B) utility poles in the United States where 1 represents low decay hazard and 5 represent severe exposure. *(A) From Scheffer, T.C., 1971. A climate index for estimating potential for decay in wood structures above ground. For. Prod. J. 21 (10), 25–31; (B) From Rural Electrification Administration (REA), 1973. Pole performance study. Staff Report, Rural Electrification Administration, Washington, DC.*

As an alternative to the climate index, the Rural Utility Services (RUS) developed a hazard index for utility poles based upon inspection data from across the United States (Fig. 15.1) (REA, 1973). Their system has five decay hazard categories and utilities who receive RUS loans must utilize these requirements. These zones, based upon actual pole failures, probably represent the most comprehensive public national data base of wood performance based on climate. This map is also included in the American Wood Protection Association Standard for specification of

wood poles. Even these guidelines; however, are imprecise because the relationships between climate, wood species/treatment, and soil characteristics remain poorly understood. As a result, these maps are, at best, general guidelines that need to be supported by local data.

It is interesting that there are relatively few guidelines available to assist in the specification of wood products; however, this void reflects the wide array of uses where wood is employed and the absence of an effective mechanism for monitoring wood losses in these many commodities.

Types of wood products decay fungi

Almost without exception, the principal wood products decayers are also common saprobic fungi that decay fallen timber and slash in the forest.

While a wide array of fungi colonize decaying wood in the forest, the number of species isolated from wood products is relatively limited. This decreased variety may reflect the more restrictive conditions within the wood product as well as the scarcity of extensive, systematic data on fungal colonization of wood. It may also represent an inability to recover all of the organisms involved in the decay process using selective media. Emerging high-throughput DNA sequencing methods will improve our ability to recover DNA from wood, but the results raise as many questions as they answer because we now need to determine if the organisms identified were actively colonizing the wood. In most instances, the fungi present in wood products probably reflect a close replication of an environmental niche present in decaying wood in the forest ecosystem.

A number of workers have reported on the incidence of fungi in various products (Richards, 1933; Silverborg, 1953; Davidson et al, 1947; Toole, 1973; Cowling, 1957; Eslyn, 1970; Eslyn and Lombard, 1983; Graham and Corden, 1980; Zabel et al., 1980; 1985). The most comprehensive listing was prepared by Duncan and Lombard (1965), that summarized fungi associated with wood products decays collected by the U.S. Forest Products Laboratory, Madison, WI, and the Forest Disease Laboratory, Beltsville, MD, over a 30-year period. These identifications tended to favor organisms that produced large, durable fruiting structures or that were readily cultured and identified from wood, but they do provide a relative guide to the major decayers of wood products in the United States. The Duncan and Lombard results indicate the following:

1. Brown rots constituted 76% of the nearly two thousand samples examined. These results probably reflect the extensive use of coniferous woods in many structures.
2. The 10 most prevalent fungi on conifers were: *Neolentinus lepideus*, *Gloeophyllum trabeum*, *G. sepiarium*, *Rhodonia* (*Poria monticola*) *placenta*, *Meruliporia* (*Poria*) *incrassata*, *Coniophora arida* var. *suffocata*, *Fibroporia* (*Poria*) *vaillantii, Antrodia* (Poria) xantha, *Coniophora puteana*, and *Fibroporia* (*Poria*) *radiculosa*.
3. The 6 most prevalent species on hardwoods were: *Gloeophyllum trabeum*, *Trametes* (*Coriolus, Polyporus*) *versicolor*, *Antrodia* (*Poria*) *oleracea*, *Meruliporia incrassata*, *Xylobolus frustulatus*, and *Schizophyllum commune*.
4. The principal fungi associated with wood decay in buildings were: *Meruliporia incrassata*, *Gloeophyllum trabeum*, *Tapinella* (*Paxillus*) *panuoides*, *Fibroporia vaillantii*, *Coniophora puteana*, *Serpula* (*Merulius*) *lacrimans, Rhodonia placenta*, and *Antrodia serialis*.

It is interesting to note that fungi with poroid basidiomata (previous family grouping - Polyporaceae) comprised 60% of the decay-associated fungi and those with resupinate basidiomata predominated (previous generic grouping - *Poria*).

One of the shortcomings of the available data on the identities of the wood-inhabiting fungi associated with wood decay is the over-emphasis on basidiomycetous decayers. Many members of the Ascomycetes have been shown to cause substantial soft rot or white rot in southern pine (Zabel et al., 1985), but most surveys of products decayers have excluded these fungi despite their importance in specific environments. Many members of the genera *Phialophora*, *Daldinia*, and *Xylaria* have been isolated from decaying wood products (Seifert, 1983; Nilsson et al., 1989; Wang and Zabel, 1990). These fungi produce less obvious fruiting structures and may be less obvious to inspectors seeking clues concerning the causes of decay. The role of these fungi in the decay of wood materials remains poorly understood, but it is readily apparent that they play important roles under specific environmental conditions.

Decay of wood products

Wood is used in a variety of applications, each with its own set of special conditions (niches) that may permit decay development as well as

support a specific fungal flora. While a majority of these problems reflect a failure to control one of the four requirements for decay fungi (moisture, air, favorable temperature, or food source), examining specific problems related to each commodity is helpful for determining controls and delineating future research needs.

Wood buildings (homes)

Wood will last for centuries when properly used in well-designed and maintained structures. Examples of this principle can be seen in the temples of Japan, the stave churches of Norway, and early houses in the U.S. The Hoxie house, built near Sandwich, Massachusetts in 1624, remains sound and contains many original timbers, framing, clapboards, and shingles. High foundations, a wide roof overhang, extensive overlapping of roof shingles and siding, and a well drained building site have all combined to provide this exceptional longevity. Maintenance also plays a key role in durability. Temples in Japan are often partially deconstructed and repaired on a regular basis. Thus, a 1000 year old temple contains much of the original material along with replaced wood in specific higher hazard zones.

More recently, decay losses in homes appear to be increasing at a substantial rate and this has stimulated extensive substitution of wood with aluminum, shingles, concrete/wood composites, and, more recently, plastics. Increasing decay problems reflect a number of changes in building design and services including:

1. The increased use of slabs or crawl space foundations in place of basements.
2. Designs with decreased roof pitch, narrowed roof overhangs, and extended decorative features such as roof beams, grills, or balustrades beyond the zone protected by the roof overhang.
3. Shifts away from naturally durable wood species and the use of woods with higher percentages of decay susceptible sapwood.
4. The use of green wood in framing that, once dried, results in open butt joints where moisture can penetrate and accumulate inside walls.
5. Increased water condensation resulting from indoor plumbing, dryers, air-conditioners and washers. This moisture causes particular problems in buildings that were tightly constructed to minimize heat loss or in those buildings where the vapor barriers were improperly installed.
6. Increased use of composites has introduced a range of materials that are inherently more sensitive to water uptake.

In most cases, these changes reflect economic decisions. For example, slabs or crawlspaces are less expensive to build than full basements. Changes in roof designs to reduce overhangs reflect a combination of saving costs by using less wood and a desire to reduce potential uplift forces on a roof during a strong wind event such as a hurricane.

Increasing decay issues also reflect decreasing numbers of builders and designers with extensive knowledge of wood properties. Most of these users have been educated on steel and concrete construction and fail to account for the inherent variability of wood. Examples of some of the common design errors in homes and wooden structures which led to serious decay problems were assembled by Rosenberg and Wilcox (1980).

The principles of decay and insect control in homes and wooden structures have been exhaustively covered in a number of excellent publications (Anon, 1969; Biesterfeldt et al., 1973; Scheffer and Clark, 1966; Scheffer and Verrall, 1973; Verrall, 1966, Verrall and Amburgey, 1977; Moore, 1979; Rambo, 1988). Amburgey (1971) prepared a useful annotated bibliography of the prevention and control of decay in homes and other wooden structures. The most important design principle emphasized in these publications is to exclude moisture from wood. Use of durable or preservative treated wood is essential when moisture exclusion is not possible but it is important to note that most houses in North America contain very little naturally durable or treated wood.

Moisture sources

Moisture in buildings can arise in the following ways:

1. Moisture can enter wood from rainfall during construction. Consider a building with 90 square meters of exposed surface under construction that received 150 mm of rainfall before the roof was finished. The water input represented over 14,000 kg of water to a structure containing just over 30,000 kg of wood. That water increased the average wood moisture content by 46% from its original value of 12–30%. This creates excellent conditions for fungal growth. Fungi present in green wood can also survive in a dormant state for long periods after installation leaving them ideally poised for regrowth should the wood become wet in service. Fungal-stained wood appears to be more frequently associated with decay damage to sheathing. The use of kiln-dried wood in construction can minimize these problems as can making sure that heat is applied to the interior to dry the wood before any sheathing is applied.

2. Direct wood contact with soil or damp masonry results in rapid moisture absorption. Examples include vertical beams in soil contact, grade levels above exterior woodwork, or wood forms left on concrete.
3. Wetting from rain or seepage due to roof leaks, splashing from the ground, or water flow through cracks or butt joints (Fig. 15.2). Water can also enter the roof in cold climates through ice dams that form near the roof edge. Although the immediate risk of decay is minimal due to the cool temperatures, this moisture often remains during warmer periods, creating ideal conditions for decay. Water enters the wood rapidly in liquid form, but must exit slowly as a vapor, leaving the wood wet for long periods after the initial wetting. Appreciable volumes of water may enter joints and cracks during periods of high winds and heavy rainfall. Water then spreads horizontally within walls by gravity flow, capillarity, and condensation of water vapors. Seepage

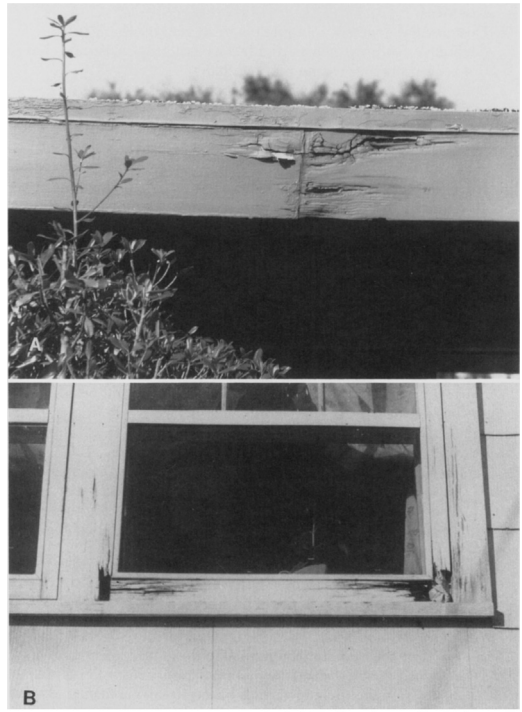


Figure 15.2 High decay hazard sites in homes where moisture accumulates include: (A) water in open-butt joints, (B) paint peeling and decay-in a facia board where water has seeped into a joint.

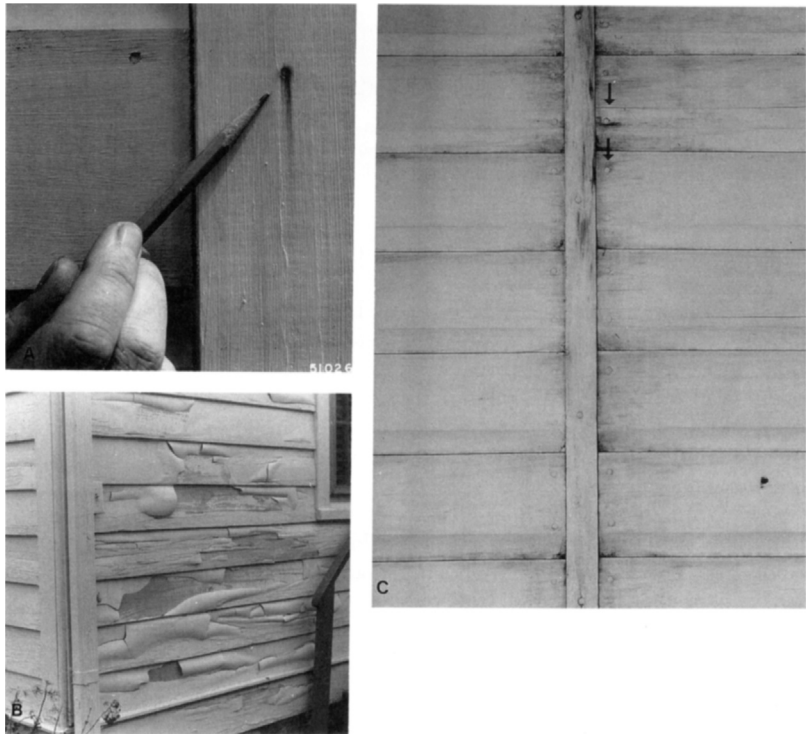


Figure 15.3 Some useful indicators of moisture accumulation in the walls of a home from leaks and condensation, setting the state for decay development. (A) A rust streak below a nail, (B) excessive paint peeling, and (C) paint discolorations, nail pulling and checks near joints. *From U.S. Forest Service and Southern Forest Experiment Station and courtesy Drs. Verrall and Amburgey (1977).*

and moisture accumulations can often be detected by the presence of rusting nail heads, paint discoloration or peeling, nails backing out, or siding buckling (Fig. 15.3). Damage can be minimized by using proper roof overhangs, installing gutters to minimize splashing, caulking wood joints, flashing horizontally exposed wood, installing drip lines on roof edges, and good paint maintenance.

4. Condensation on wood surfaces has long been recognized as a problem in cool climates where moisture vapor moving through the wood can condense on the cool outer surfaces (Fig. 15.2), but can also be a problem in air-conditioned homes in warmer climates (Anderson, 1972; Duff, 1971). Condensation varies with the volume of vapor diffusing and the magnitude of temperature drop. In cool climates,

the condensate may freeze during the winter and melt as the weather warms. This condensation may be relatively minor, but can lead to paint blistering, wood staining, compaction of insulation and corrosion of electrical conduits. More serious condensation can develop in subflooring over damp crawl spaces, especially in the corners of the building. Crawl space condensation can be minimized by installing adequate vents around the building and by covering the ground with an impermeable plastic barrier to limit moisture movement upward from the soil.

5. Water leaks from plumbing or inadequate sealing of joints around plumbing fixtures may cause extensive localized decay. Moisture can also come from outside sources, such as sprinklers that spray walls. Control rests mostly with correcting the leak or redirecting the water away from the wood.
6. Biotic sources may also contribute to moisture in wood. Some fungi, such as the notorious building rot fungi *Meruliporia incrassata* and *Serpula lacrimans*, form rhizomorphs that can transport water from damp soil across foundations and masonry into the wood. These fungi (also called True Dry Rot Fungi) can release a sufficient quantity of water through metabolism to sustain decay in poorly ventilated spaces. These fungi are best controlled by periodic inspection for the presence of the cable-like rhizomorphs and the removal of wood debris from the soil around the building (Verrall, 1968). Fortunately, these fungi are relatively rare in North America.

The two true Dry rot fungi are of special interest and importance. *Serpula* (*Merulius*) *lacrimans* is the major damaging building decayer in Europe and occurs also in Canada and the northern United States. Fortunately, this species appears to be limited to cool climates and occurs most commonly in crawl spaces or subflooring where the ventilation is poor. This fungus is widespread in Europe because of the prevalent practice in older buildings of embedding timbers in masonry. A similar major building decayer is *Meruliporia incrassata*. This fungus is more common in the southern coastal areas of the United States. Outbreaks of this house decay saprobe are often traceable to lumber that was invaded by the fungus during seasoning or storage (Verrall, 1968; Dietz and Wilcox, 1997). Both fungi occur most commonly in poorly ventilated locations and cause a similar-appearing brown cubical rot. They are able to transport water to the wood they colonize and invade the wood via mycelial fans, cords, and in some instances thick cable-like rhizomorphs (Fig. 15.4). The ability of

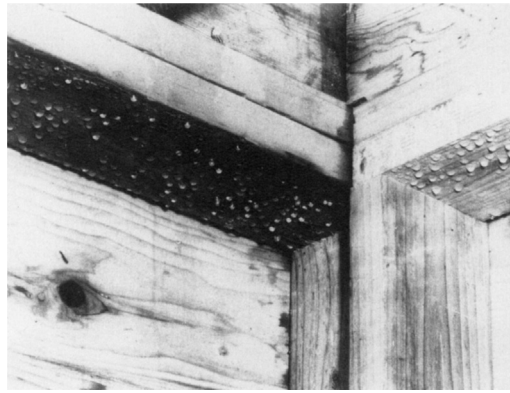


Figure 15.4 Condensation is an insidious source of moisture in some structures. Corners in crawl spaces or zones with restricted air flow are common locations for this problem in homes. *From U.S. Forest Service and Southern Forest Experiment Station and courtesy Drs. Verrall and Amburgey (1977).*

Table 15.1 Methods for preventing decay in buildings.

Use bright, kiln-dried lumber
Protect lumber from wetting during construction
Provide adequate roof overhang (>600 mm)
Provide gutters to move liquid water away from the building
Use well-drained building sites
Install preservative treated sill plates >200 mm above grade
Install ground covers on soil in crawl spaces
Ventilate crawl spaces (openings 1/160 of surface area)
Flash wood to shed water where it is exposed in a horizontal position
Use pressure treated wood for exposed design features such as posts or rails
Maintain coatings such as paints or stains and recaulk joints regularly
Periodically inspect building for signs of moisture
Use exterior finishes that shed water

these fungi to penetrate and survive in mortar makes them particularly difficult to control.

There are numerous simple methods for preventing wood damage in buildings (Table 15.1). Most require good initial design of the structure followed by a regular maintenance schedule to limit any damage that occurs. It is especially important to maintain paint films or other water repelling barriers that can minimize the creation of conditions suitable for microbial growth (Feist, 1984; Feist and Mraz, 1978; Rowell and Banks, 1985).

Utility poles

Utility poles (or electric poles) are used to support the wires that bring power to most of North America. There are an estimated 160 million wood poles in service in North America. Decay in utility poles has been extensively studied because of the high degree of reliability demanded of electrical service and the availability of utility research support.

A utility pole in direct soil contact is exposed to a high decay hazard, but must deliver 40 or more years of reliable service. Because of this hazard, most utilities use either naturally durable wood species such as western redcedar, or they pressure-treat less durable woods with preservative. Commercial wood pole species in North America include Douglas-fir, lodgepole pine, ponderosa pine, red pine, southern pine and western redcedar. In most regions, Douglas-fir is used for larger size poles, while southern pine comprises the majority of smaller poles.

Although wood poles have performed well for over 100 years, a certain percentage of poles develop decay problems. Examples of the major decay types and common locations in utility poles are illustrated in Figs. 15.5 and 15.6. Poles with low preservative retentions may slip through the inspection process and are responsible for some early failures. In a classic study, Lumsden (1960) showed that individual creosote retentions in a pole batch treated to a target of 128 kg/m^3 (gauge) ranged from 32 to 224 kg/m^3. These variations reflect the natural variability of wood.

Decay problems in poles are generally classified as internal or external decay. Internal decay can originate in the pole during storage or seasoning prior to treatment (Fig. 15.7). Such pretreatment decay is particularly insidious in the southern pines where the thick sapwood zone is responsible for much of the pole strength. This decay is usually inactivated by the preservative treatment, but the treatment can conceal existing damage nor restore the wood to its original capacity. Internal decay in larger poles may not be inactivated during the preservative treatment unless the wood is heated above 65 °C at the pith center. It is important to ensure that poles are heated to sterilize the wood during treatment. Internal decay also often develops in thin sapwood species that are difficult to completely protect with preservative. Poles are normally treated while the inner heartwood remains moist. As the pole seasons in-service, deep checks that penetrate beyond the depth of treatment expose the untreated heartwood to attack by decay fungi and insects. Untreated wood can also be exposed when holes or cuts are made after treatment. Internal decay normally occurs at or below the groundline, but can extend for considerable

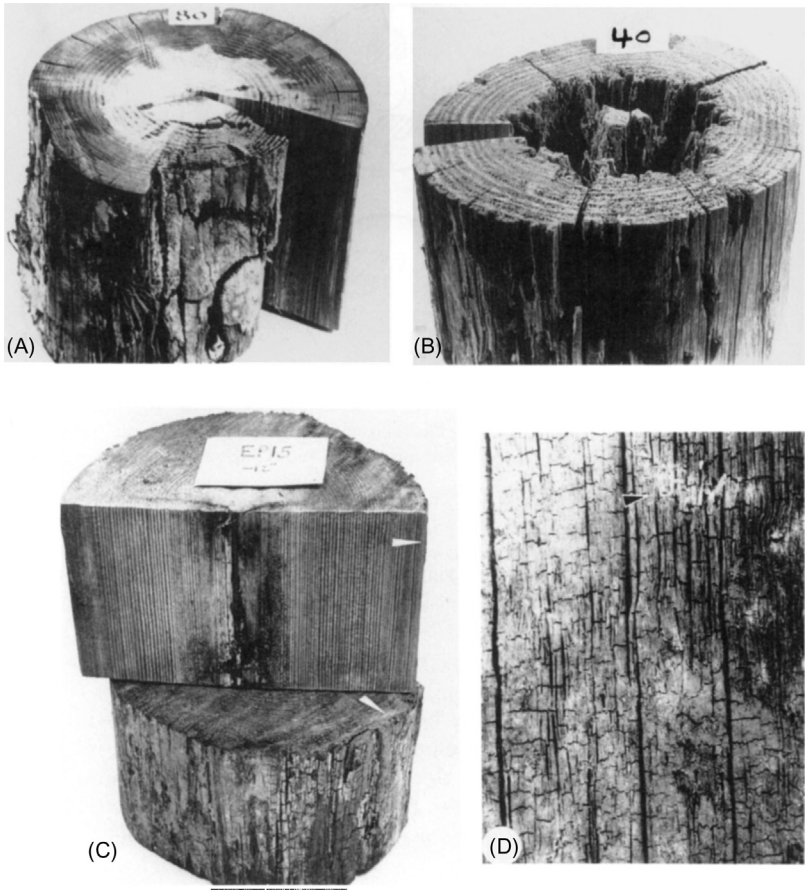


Figure 15.5 Examples of the major decay types in southern pine utility poles in service. (A) A brown cubical rot in the outer-treated lone, (B) advanced white rot in the untreated center, (C) soft rot as seen on surface of an older creosote-treated pole with the radial development indicated by white arrows, and (D) soft rot as seen on the below-ground surface of a cellon-treated southern pine pole displaying the typical cracking and exfoliation pattern. *From: Zabel, R.A., Wang, C.J.K., Terracina, F.C., 1982. The fungal associates, detection, and fumigant control of decay in treated southern pine poles. EPRI EL-2768, Final Report Project 1471-1. Electric Power Research Institute, Palo Alto, CA, (A and B) with permission: Electric Power Research Institute, Palo Alto, CA, and (C) and (D) Wang, C.J.K., Zabel, R.A. (Ed), 1990. Identification Manual for Fungi from Utility Poles in the Eastern United States. American Type Culture Collection, Rockville, MD, pp. 356, with permission of American Type Culture Collection, Rockville, MD.*

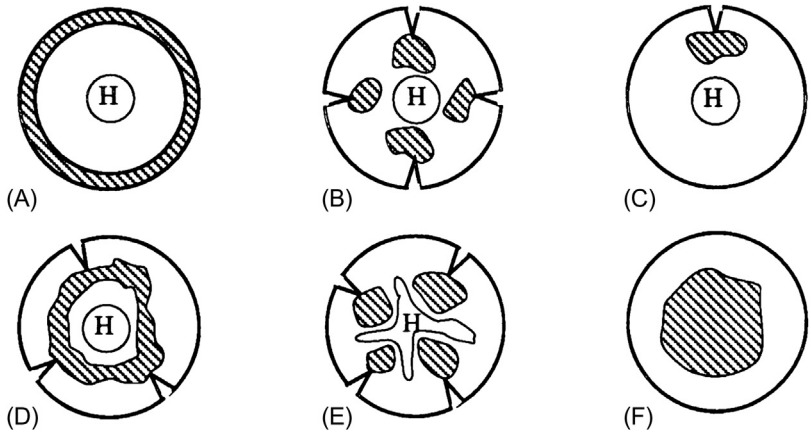


Figure 15.6 Illustrations of the common locations and patterns of decay development in the ground line zone of utility poles. (A) Surface soft rot, (B) and (C). Decay pockets associated with checks, (D) internal decay where the decay developed between the outer treated shell and the heartwood and usually originated from the base of deep checks, (E) similar to (D) but the cross section is at a stem Nodal zone, and (F) An internal decay hollow which may have resulted from an advanced case of (D) or pre-treatment decay that escaped the preservative treatment. *From: Zabel, R. A., Wang, C.J.K., Terracina, F.C., 1982. The fungal associates, detection, and fumigant control of decay in treated southern pine poles. EPRI EL-2768, Final Report Project 1471-1. Electric Power Research Institute, Palo Alto, CA; Wang, C.J.K., Zabel, R.A. (Ed), 1990. Identification Manual for Fungi from Utility Poles in the Eastern United States. American Type Culture Collection, Rockville, MD, pp. 356, with permission of Electric Power Research Institute, Palo Alto, CA and American Type Culture Collection. Rockville, MD.*

distances above the ground in regions where moisture levels are high or below the ground in arid regions. Internal decay can cause significant reductions in wood strength in relatively short periods of time (4–10 years). Internal decay can be minimized by limiting the degree of checking or by improving the depth of treatment in high decay hazard zones (the groundline). These topics are discussed in more detail in Chapter 20. Internal decay can also occur in species with thicker sapwood when the wood is treated while the moisture content remains high. This process results in pockets of inadequate treatment where fungal decay can develop.

External decay can develop in several ways (Fig. 15.8). When naturally durable woods are employed, the outer sapwood shell is generally left on the pole. This zone has no natural decay resistance and will begin to decay both above and below the groundline. This effectively decreases the circumference of the pole, thereby reducing bending strength.

Figure 15.7 A cross section in the groundline zone of a treated Douglas-fir transmission pole indicating the association between deep checking (arrows) and the development of internal decay. *Courtesy: R.W. Meyer.*

Figure 15.8 Typical soft rot damage in the groundline of a CCA treated Douglas-fir transmission pole that had been in service for approximately 10 years in a swampy are. Arrows (Left to right) identify the pole butt, zone of maximum soft rot development, and surface exfoliation and the groundline.

Above-ground decay of naturally durable woods can be controlled by full length preservative treatment prior to installation. It was formerly also controlled by flooding the surface of poles with preservative at 10–15 year intervals in the field. External flooding of pole surfaces with preservative led to considerable contamination of the surrounding environment and this process is no longer used. As an alternative, some utilities shave the sapwood off prior to installation to eliminate the need for chemical treatment.

External decay can also develop below the groundline in preservative treated poles. Southern pine, ponderosa pine and western redcedar are species considered to be susceptible to surface decay; however, Douglas-fir treated with pentachlorophenol in liquified petroleum gas or methylene chloride is also susceptible to this damage. Surface decay develops slowly as the outer shell of protective preservative is depleted by leaching or detoxified by pioneering fungi such as *Cladosporium resinae* or *Paecilomyces variottii* . Once these fungi have depleted the preservative below a toxic threshold level, soft-rot fungi capable of degrading the wood structure can invade. Soft rot damage can develop in pockets near the surface or may be associated with small surface checks. In some cases, soft rot fungi that are tolerant to a preservative may penetrate the entire treated shell (Zabel et al., 1991). The presence of this damage in the outer shell is critical since 90% of the bending strength of a pole lies in the outer 50 mm of the surface and any loss in pole circumference has drastic effects on strength.

The best methods for limiting decay in utility poles are performed before or during preservative treatment. They include adequate seasoning to remove moisture, predrilling of all holes for attachments, and strict adherence to specifications for preservative retention and penetration. In addition, the development of a regular program of inspection and remedial treatment can significantly extend pole service life (Morrell, 2012). As an example, Bonneville Power Administration once expected as little as 20 years service life from their Douglas-fir poles, but they now achieve an average service life far in excess of 50 years using a combination of improved specifications and maintenance (Lindgren, 1989). Early decay detection is important since the pole still retains adequate residual strength and remedial treatments are more effective at this point.

Foundation or marine piling

Wood piling have a long history of use to provide foundations for supporting buildings. Untreated piling under buildings in many major

European cities are hundreds of years old. Like wood poles, piling are subject to the same problems with checking and failure of the treated zone. In most instances, however, piling are exposed under high moisture conditions that exclude oxygen and therefore limit most fungal or insect attack. Problems do arise when fluctuating water levels permit oxygen to enter the wood and establish conditions for fungal growth. This can be a particular problem in older buildings supported by untreated foundation piling. Extensive fungal attack can occur as the water table fluctuates or declines. This damage is extremely difficult to correct or prevent because the building rests on top of the piling. Nearly all foundation piling are now preservative treated, further extending their useful life.

Piling are also used in fresh and salt water exposures to support docks and other marine structures. Wood submerged in fresh water will decay very slowly as a result of limited oxygen availability, but the portion above water can develop decay and insect issues. Piling in salt water may be susceptible to marine borer attack. Nearly all piling are preservative-treated to limit the risk of degradation.

Fungal attack can occur near the tops of marine piling above the water line or wherever joints with caps or stringers act as water traps. In many cases, builders remove the pile top, exposing the untreated wood beyond the treated shell and leading to extensive internal decay. Capping with fiberglass mesh or other suitable barrier will exclude moisture and protect the untreated wood from attack (Newbill and Morrell, 1990). There are also a number of corrective remedial treatments that can be applied to decaying pile tops to arrest and prevent decay.

In addition to decay problems, marine piling are exposed to varying degrees of marine borer attack. This attack is often concentrated near the mudline or around joints where the marine borers are more protected. Proper specification for the marine borer hazards present in the harbor and regular diving inspections can minimize the risk of marine borer damage. The American Wood Protection Association Standards include maps showing the levels required for various locations along the East and West Coasts of the U.S.

Railroad ties, mine timbers, and bridges

Railroads, mines, and highway departments employ extensive amounts of large dimension timbers that are susceptible to the development of deep checks that extend beyond the depth of preservative treatment and trap water.

Railroad ties are typically exposed on well-drained rock ballast that reduces the risk of moisture accumulation and limits soil contact. As a result, the decay rate should be slightly lower than would be found in direct soil contact. However, the development of deep checks beyond the depth of the original preservative treatment creates an excellent water trap and entry point for decay fungi. Ties are generally thought to fail most frequently due to mechanical failure as a result of repeated train loads or due to failure around the spikes (spike kill), but portions of this failure may reflect decay that occurred during air-seasoning (often called stack burn by wood treaters) or in service. Other activities such as drilling for new spikes expose untreated wood and may hasten failure. Decaying ties force lower train speed limits, reducing track efficiency.

Bridge timbers represent a slightly different exposure, but have many of the same problems experienced with ties. Bridge timbers can be either naturally durable species which are typically used in historic structures (mostly covered bridges) or preservative treated wood of less durable species. Because bridges are structural; however, decay poses significant safety risks. As a result, pretreatment handling is directed to limit the risk of decay and to developing checks prior to treatment. Despite these efforts, the use of large timbers with a portion of the pith virtually assures that some deep checks will develop and that these checks will penetrate beyond the depth of treatment. As a result of these problems and the expense of large dimension timbers, many timber bridges are now constructed using either glu-laminated timbers or using many smaller treated timbers in stress-laminated decks. Glue-laminated timbers are dried prior to treatment, minimizing later check development. In addition, individual laminations can be selected for high strength, improving the engineering properties of the resulting beam. The stress laminated deck bridges tend to perform well because there is relatively little risk of deep checking beyond the depth of the original treatment in the smaller wood members (Morrell et al., 2015).

In addition to the problems associated with checking, bridges experience a number of other decay problems, mostly relating to the collection of moisture around wood joints. Particular problems occur in the railings, where the bridge rests on the foundation, and in the deck. These problems often originate where debris collects in joints, creating ideal conditions for fungal growth. Careful design detailing to limit debris collection points and regular maintenance can minimize this type of damage.

Timbers are used to temporarily shore up tunnels in mines while the ore or coal is being removed. They are subjected to extremely wet and warm conditions, and can develop extensive decay issues. Initially, these timbers were used untreated, but declining supplies of durable species resulted in substitution of preservative-treated wood. More recently, mine timber usage has declined due to the development of alternative methods for tunnel support, but they are still a locally important product.

Cooling towers

Cooling towers are used to expose hot process water to air-flow to reduce the temperature. They usually employ extensive amounts of wood in lathing and for support. Basidiomycete attack can occur in portions of the tower above the splash line or in the whole tower when the tower is shutdown, but the high moisture conditions present during operation exclude most basidiomycetes. These conditions, however, are ideally suited for soft rot fungi that gradually erode the wood surface. In fact, the importance of soft rot fungi was first noted in cooling tower slats (Savory, 1954). In most cases, either naturally durable species such as redwood or preservative treated Douglas-fir are used in cooling towers. However, soft rot attack can even occur in timber of these species over long periods of time. Regular inspections and periodic retreatments with preservative sprays were used extend cooling tower service life (Baechler et al., 1966), but the ability to spray preservative solutions in these environments has been sharply curtailed because of the risk of water contamination. Almost all cooling tower wood is preservative treated with either chromated copper arsenate or acid copper chrome and this, coupled with regular inspections and replacement of parts as needed is now a more typical approach to maximizing cooling tower service life.

Wooden boats

Wooden boats have long experienced problems with decay. In fact, decay has played significant roles in maritime history-decimating the Spanish Armada prior to its planned attack on Britain and limiting the availability of ships for Britain during the American Revolution (Ramsbottom, 1937). Relatively few boats are constructed of timber in North America and most use durable timbers. However, there is a thriving market for restoration of older wooden sailing vessels or building replicas of these vessels often with many of the same issues present in the original vessel.

The large timbers in these ships are subjected to frequenting wetting and the slow drying rates create idea conditions for decay development.

Boat stems and transoms, frameheads, beam ends, and bilges are most susceptible to decay. The use of durable or preservative treated woods, ventilation of bilge areas, and careful maintenance practices can help minimize this damage. The fungi associated with this attack were the subject of much study during and after World War II (Hartley and May, 1943; Davidson et al., 1947; Savory and Packman, 1954; Roth and Hartley, 1957). The use of fungicides in the bilge water was recommended for limiting damage in that area (Scheffer, 1953), but this is not generally feasible. Similar decay problems have also been noted in commercial fishing vessels where the damage can reduce efficiency and endanger lives (Condon and Graham, 1975).

Pallets and boxes

Large quantities of wood are employed in pallets and boxes for transporting goods and a portion of this material is subjected to conditions conducive to fungal growth. In some instances, the material is treated by dipping in preservative at some point during or after assembly. The problems associated with these products for military uses was summarized by Greathouse and Wessell (1954). Chemical treatments for limiting damage have been explored by a number of workers (Blew, 1959; Verrall, 1959, 1965; Verrall and Scheffer, 1969; DeGroot and Stroukoff, 1986), and generally pressure treatment is required to protect wood in tropical environments, while dip treatments with water repellant preservatives are sufficient in the Southeastern U.S.

Pallets are a critical component in the global transportation network. The move to containerized shipping during the Second World War created a vast market for pallets. Pallets are mostly single-use items constructed using low value wood cut from the center of the tree. Pallet materials often contain decay pockets and active insect infestations. Insects present in pallets have accounted for a number of insect invasions into North America including the Asian longhorn borer and the emerald ash borer. Virtually all pallets are now subjected to a heat treatment of 56 °C for 30 minutes as per the International Standards for Phytosanitary Measures Standard ISPM 15 to minimize the risk that they contain live pests. While this treatment cannot eliminate everything in a pallet, it sharply reduces the presence of insects.

The other issue associated wood packing material (also termed solid wood packaging) is the risk that mold growing on the wood surface will contaminate the products being transported. Most pallet manufacturers expressly forbid the use of preservative treatments that would exclude mold because of fears that the chemicals will contaminate the products. As a result, mold can be a major issue on pallets. The use of heat treatments such as those required in ISPM 15 also increases the risk of mold. The treatment kills many fungi already in the wood, but does not reduce the moisture content, leaving a fertile surface for fast growing mold fungi to colonize. Mold remains a critical issue for wood packaging materials

Panel products

Composites represent an increasingly important segment of the forest products industry. These panel products are most often employed where the risk of decay is minimal; however, plywood, laminated veneer lumber, glued laminated timbers, and flakeboards are increasingly used under conditions conducive to fungal attack. The presence of resin in the panels can slow fungal attack, but composites decay, particularly when they are exposed under high moisture conditions. One important consideration with panel products is their susceptibility to wetting. Most panels swell as they sorb water and this swelling leads to permanent deformation and loss of properties well before fungal attack occurs. Many of the same fungi isolated from solid wood products appear to be common in panel products, but a number of other organisms including *Phanerochaete chrysosporium* and *Schizophyllum commune* appear to be more important colonizers of some products. As panel products see increased application, it is likely that a clearer picture will emerge concerning the fungal flora of these products. There is a general misconception that panel products are safe from fungal attack because they will never get wet; however, nearly all buildings experience some form of moisture intrusion through roof leaks, plumbing failures or condensation.

A decay control principle

Wood products are subject to attack under a variety of uses and environments; however, each of these situations accurately replicates one or more components of the natural environment for that particular decay organism. Thus, if we hope to limit losses due to decay, we must alter our

wood uses to eliminate these conditions. It is readily apparent that the prime factor in decay is moisture. Decay prevention depends on designs that keep moisture levels below the fiber saturation point. Alternatively, naturally durable or preservative treated woods must be used. In either instance, the structure must be properly maintained to continue to exclude moisture or to maintain an adequate level of preservative protection.

Wood can last for centuries if careful design practices are employed in properly maintained structures. As wood becomes increasingly valuable, wood microbiologists must educate the engineering and architectural communities to adhere to these decay prevention principles.

Summary

1. Decay of wood products causes losses amounting to billions of dollars each year.
2. Decay of wood products generally results from a failure to control one of the requirements for fungal growth (moisture, oxygen, temperature or an adequate food source). Moisture is the key factor and wood kept dry will not decay.
3. The risk of decay varies widely with geographic location, application and wood species. Attempts have been made to quantify these risks using maps describing decay hazard or decay index. These maps, while useful, provide only relative guidelines to decay risk.
4. A variety of fungi have been isolated from decayed wood products, but some species are particularly abundant across a variety of wood uses. Unfortunately, previous research has relied heavily upon the presumption that non-basidiomycetes are not important wood destroyers. Further research is necessary to better understand the role of all microorganisms in the deterioration process.
5. A variety of wood products are subject to decay. Most damage can be limited by using proper design principles, by using naturally durable or preservative treated wood, and by regular maintenance.

References

Amburgey, T.L., 1971. Annotated bibliography on prevention and control of decay in wooden structures (including boats). Southern Forest Expt. Stat., Forest Service, U.S. D.A., Gulfport, Mississippi. pp. 123.

American Wood Protection Association, 2017. Book of Standards. Birmingham, Alabama.
Anderson, L.O., 1972. Condensation problems: their prevention and solution. U.S. Dept. Agric. Forest Service Res. Paper FPL 132, Forest Prod. Laboratory, Madison, WI.
Anon, 1969. Wood decay in houses: how to prevent and control it. Home & Garden Bulletin No. 73, U.S. Dept. Agric., Washington, D.C.
Anonymous, 1985. Addressing bad tire clustering. Railway Track and Structures (August):21–25.
Baechler, R.H., Bendtsen, B.A., Roth, H.G., 1966. Preservative treatment of cooling tower slats. Heating, Piping, and Air Conditioning (April), 121–123.
Biesterfeldt, R.C., Amburgey, T.L., Williams, L.H., 1973. Finding and keeping a healthy home. Southern Forest Expt. Stat., U.S.D.A., Forest Service General Technical Report SO-1, New Orleans, Louisiana.
Blew, J.O., 1959. Preservatives for wood pallets. U.S.D.A., Forest Prod. Lab., Rept. No. 2166, Madison, WI.
Boyce, J.S., 1961. Forest Pathology, third ed. McGraw-Hill Book Company, New York, N.Y.
Brier, A.N., Dost, W.A., Wilcox, W.W., 1988. Characteristics of decay and insect attack in California homes. Calif. Agriculture 42 (5), 21–22.
Brischke, C., Rapp, A.O., 2007. Influence of wood moisture content and wood temperature on fungal decay in the field. Observations in different micro-climates. Wood Sci. Technol. 42, 663–677.
Carll, C.G., 2009. Decay hazard (Scheffer) index values calculated for 1971-2000 climate normal data. USDA Forest Products Laboratory General Technical Report FPL-GTR-179. Madison, WI.
Condon, E.J., Graham, R.D., 1975. Wood-boat maintenance: decay and its prevention. Extension Service Sea Grant Advisory Program Publication No. 23, Oregon State University, Corvallis, Oregon.
Cowling, E.B., 1957. A partial list of fungi associated with decay of wood products in the United States. Plant. Dis. Rptr. 41, 894–896.
Davidson, R.W., Lombard, F.F., Hirt, R.R., 1947. Fungi causing decay in wooden boats. Mycologia 39, 313–327.
DeGroot, R.C., Stroukoff, M., 1986. Efficacy of alternative preservatives used in dip treatments for wood boxes. U.S.D.A. Forest Products Laboratory Research Paper FPL-RP-481. Madison, WI.
Dietz, M., Wilcox, W.W., 1997. The role of pre-infection of green Douglas-fir lumber in above ground decay in structures in California. For. Products J. 47 (5), 56–60.
Duff, J.E., 1971. The effect of air conditioning on the moisture conditions in wood walls. U.S.D.A., Forest Service, Pres. Paper SE-78.
Duncan, C.G., Lombard, F.F., 1965. Fungi associated with principal decays in wood products in the United States. U.S. Forest Service Research Paper WO-4, Dept. Agric., Washington, DC.
Eslyn, W.E., 1970. Utility pole decay. Part 1. Basidiomycetes associated with decay in poles. Wood Science and Technology 4, 97–103.
Eslyn, W.E., Lombard, F.F., 1983. Decay in mine timbers. Part. II. Basidiomycetes associated with decay of coal mine timbers. For. Products J. 33 (7/8), 19–23.
Feist, W.C., 1984. The role of water repellants and chemicals in controlling mildew on wood exposed outdoors. U.S.D.A. Forest Products Laboratory Research Note FPL-0247. Madison, WI.
Feist, W.C., Mraz, E.A., 1978. Protecting millwork with water repellants. For. Products J. 28 (5), 31–35.
Graham, R.D., Corden, M.E., 1980. Controlling biological deterioration of wood with volatile chemicals. Electrical Power Research Institute El -1480, Project 212-1. Palo Alto, CA.

Greathouse, G.A., Wessel, C.J., 1954. Deterioration of Materials - Causes and Preventative Techniques. Reinhold Publ. Corp, NY.
Hartley, C., May, C., 1943. Decay of wood in boats. U.S. Dept. Agr. Forest Path. Spec. Release No.8. Washington, DC.
Leicester, R.H., Wang, C.-H., Nguyen, M.N., Thornton, J.D., Johnson, G., Gardner, D., Foliente, G.C., MacKenzie, C., 2003. An engineering model for decay of timber in ground contact. Discussion Document No. IRG/WP/03-20260. International Research Group on Wood Protection. Stockholm, Sweden.
Lumsden, G.Q., 1960. Fortified wood preservatives for southern pine poles. *Forest Products Journal* 10(9): 456–462.
Lindgren, P., 1989. Bonneville Power administration's wood pole management program. Proceedings International Conference on Wood Poles, Fort Collins, Colorado.
Moore, H.B., 1979. Wood inhabiting insects in houses: their identification, biology, prevention, and control. U.S. Department of Housing and Urban Development, Washington, DC.
Morrell, J.J., 2012. Wood Pole Maintenance Manual: 2012 Edition. Research Contribution 51, Forest Research Laboratory, Oregon State University, Corvallis, OR, 46 p.
Morrell, J.J., Norton, J., Powell, M., Gerber, C., Kleinschmidt, S., 2015. Assessment of an ACZA treated Douglas-fir stressed deck bridge in South Johnstone, Queensland, Australia: 22 year report. American Wood Protection Association 111, pp. 265–273.
Morris P.I., Wang, J., 2008. A new decay hazard map for North America using the Scheffer Index. Document No. IRG/WP/08-10672. International Research Group on Wood Protection. Stockholm, Sweden.
Morris, P.I., Wang, J.Y., 2011. Scheffer index as preferred method to define decay risk zones for above ground wood in building codes. Int. Wood Products J. 2 (2), 67–70.
Newbill, M.A., Morrell, J.J., 1990. Marine capping devices in combination with chemical treatments to prevent and arrest pile-top decay. Holzforschung 44 (l), 73–75.
Nilsson, T., Daniel, G., Kirk, T.K., Obst, J.R., 1989. Chemistry and microscopy of wood decay by some higher Ascomycetes. Holzforschung 43 (1), 11–18.
Rambo, G.W. (Ed.), 1988. Wood decay in structures and its control. National Pest Control Association, Dunn Loring, Virginia. 63 p.
Ramsbottom, J., 1937. Dry rot in ships. Essex Nat. 25, 231–267.
Richards, C.A., 1933. Decay in buildings. Proceedings American Wood Preserver's Association, 29, 389–398.
Rosenberg, A.F., Wilcox, W.W., 1980. How to keep your award-winning building from rotting. Wood Fiber 14 (1), 70–84.
Roth, L.R., Hartley, C., 1957. Decay prevention in wooden ships during storage. Division of Forest Disease Research, U.S.D.A., Forest Service Report NPO 1700S571. Beltsville, MD.
Rowell, R.M., Banks, W.B., 1985. Water repellency and dimensional stability of wood. U.S.D.A. Forest Products Laboratory General Technical Report FPL-50. Madison, WI.
Rural Electrification Administration (REA), 1973. Pole performance study. Staff Report, Rural Electrification Administration, Washington, DC.
Savory, J.G., 1954. Breakdown of timber by Ascomycetes and Fungi Imperfecti. Ann. Appl. Biol. 41, 336–347.
Savory, J.G., Packman, D.F., 1954. Prevention of wood decay in boats. Bull. For. Prod. Rep. No. 31, London.
Scheffer, T.C., 1953. Treatment of bilgewater to control decay in the bilge area of wooden boats. J. For. Products Res. Soc. 3, 72–78.

Scheffer, T.C., 1971. A climate index for estimating potential for decay in wood structures above ground. For. Products J. 21 (10), 25–31.
Scheffer, T.C., Clark, J.W., 1966. On-site preservative treatments for exterior wood of buildings. For. Products J. 17 (12), 21–29.
Scheffer, T.C., Verrall, A.F., 1973. Principles for protecting wood buildings from decay. USDA Forest Service Research Paper FPL 190.
Seifert, K.A., 1983. Decay of wood by the Dacrymycetales. Mycologia 75 (6), 10111018.
Silverborg, S.B., 1953. Fungi associated with the decay of wooden buildings in New York State. Phytopathology 43, 20–22.
Toole, E.R., 1973. Fungi associated with decay in utility poles. Information Series 15, Mississippi Forest Products Utilization Laboratory, Mississippi State College, State College, Mississippi.
Verrall, A.F., 1959. Preservative moisture-repellent treatments for wooden packing boxes. For. Products J. 9 (1), 1–22.
Verrall, A.F., 1965. Preserving wood by brush, dip, and short-soak methods. Forest Service, U.S.D.A. Tech. Bull. 1334, Madison, WI.
Verrall, A.F., 1966. Building decay associated with rain seepage. Tech. Bull. 1356. U.S. Dept. Agric. Forest Service, Madison, WI.
Verrall, A.F., 1968. *Poria incrassata* rot: prevention and control in buildings. U.S.D.A. For. Ser. Tech. Bull. No. 1385.
Verrall, A.F., Amburgey, T.L., 1977. Prevention and Control of Decay in Homes. Superintendent of Documents. U.S. Government Printing Office, Washington, D.C.
Verrall, A.F. and T.C. Scheffer. 1969. Preservative treatments for protecting wood boxes. U.S.D.A. Forest Service Research Paper FPL 106, Madison, Wisconsin.
Wang, C.J.K., Zabel, R.A. (Eds.), 1990. Identification Manual for Fungi From Utility Poles in the Eastern United States. American Type Culture Collection, Rockville, MD.
Zabel, R.A., Lombard, F.F., Kenderes, A.M., 1980. Fungi associated with decay in treated Douglas-fir transmission poles in the Northeastern United States. For. Products J. 30 (4), 51–56.
Zabel, R.A., Lombard, F.F., Wang, C.J.K., Terracina, F.C., 1985. Fungi associated with decay in treated southern pine utility poles in the Eastern United States. Wood Fiber Sci. 17 (1), 75–91.
Zabel, R.A., Wang, C.J.K., Anagnost, S.E., 1991. Soft-rot capabilities of some major microfungi isolated from Douglas-fir poles in the Northeast. Wood Fiber Sci. 23, 220–237.
Zabel, R.A., Wang, C.J.K., Terracina, F.C., 1982. The fungal associates, detection, and fumigant control of decay in treated southern pine poles. EPRI EL-2768, Final Report Project 1471-1. Electric Power Research Institute, Palo Alto, CA.

Further reading

Howard, J.L., Westby, R.M., 2013. U.S. Timber production, trade, consumption and price statistics 1965–2011. Research Paper FPL-RP-676. U.S. Department of Agriculture, Forest Service, Forest Products Laboratory, Madison, WI. 91 p.

CHAPTER SIXTEEN

The detection of decay

Contents

Wood Microbiology.
DOI: https://doi.org/10.1016/B978-0-12-819465-2.00016-4

The reliable detection of early decay is a major problem for many wood users and a principal research goal in wood microbiology. It is important to recognize decay early before serious damage occurs so corrective actions can be taken or remedial treatments applied.

Detecting wood decay before significant strength losses occur poses a critical challenge to managers of large wood systems such as utilities, railroads, and marine facilities. Emerging use of mass timber products in high-rise buildings will further exacerbate that challenge. While methods for rapidly arresting active decay are now highly effective and widely available (Morrell and Corden, 1986), most methods for detecting early decay remain unreliable and relatively unsophisticated (Morrell and Wilson, 1985). Where sophisticated devices have been developed, the need for trained operators to interpret results continues to limit their usefulness (Wilson, 1988).

Decay recognition from visual (macroscopic) and microscopic features was covered in Chapters 7, 9 and 10. This chapter focuses primarily on non-destructive or semi-destructive methods for detecting decay and estimating residual wood properties. The general principles of decay detection are presented in relation to practical approaches to detection in major wood products.

Location of decay

Decay can develop anywhere conditions are suitable for fungal growth, but we typically talk about external and internal decay. External decay generally often occurs below the ground and is caused by soft rot fungi although traditional decay fungi may also be involved, especially if the wood is not preservative treated. External decay can also occur above the ground, although moisture variations and ultra-violet light exposure often limit this damage to the soft rot fungi. Surface decay can be easily detected at the intermediate to advanced stages by probing to detect the depth of weakened wood.

Internal decay occurs in larger wood members and is typically caused by the more traditional basidiomycetes. The decay is often associated with a check or other opening in the wood that allows moisture and spores to enter the wood. The checks provide a more protected and stable environment for spore germination and fungal growth. The fungus continues to grow into the wood beneath the surface of the check gradually degrading the interior of the structure, but often leaving a relatively sound-appearing exterior.

Decay detection difficulties

Decay is difficult to detect since it often occurs internally or in hard to access locations. Sampling must be limited since it further damages and weakens the wood. Also, there is the inherent difficulty of detecting the beginning stages of decay when the property changes are small and subtle. Complicating the detection of decay are the differences in susceptibility of wood species to decay, the presence of normal wood growth characteristics such as knots or annual rings, and considerable variation in rate and pattern of attack by the many fungi that attack wood. Compounding these problems are the differences in the rates at which fungi degrade wood under the wide array of environmental conditions and the fact that subtle changes during the early stages of decay may resemble natural variations in wood quality.

Basic sampling for decay

In general, inspection for decay should concentrate on those locations that are most likely to become wet in service. Building inspectors typically call these conducive conditions such as evidence of wetting or direct soil contact. Moisture sources may be seasonal or continuous and the inspector must carefully observe the structure for signs of wetting. Points of direct contact with the ground, water-trapping joints such as those found in window frames and butt joints, crawl spaces where soil moisture can condense on wood in the foundation, condensation on ceilings or in attics, leaks from plumbing, and deep checks are all potential

sites for decay. Even when dry, these zones often have telltale white deposits where salts have diffused through the wood and precipitated on the surface.

An ideal decay detection device

It will be useful to briefly review the features of an ideal decay detection device. The ideal decay detection device would separate decay from natural wood defects, particularly at the early stages of decay when visible fungal attack is sparse, but effects on mechanical properties may be quite substantial (see Chapter 10). The device should be inexpensive, simple to use and require minimal training (i.e. be insensitive to operator variables). Ideally, the device would also assess residual wood strength. The cost of individual devices may vary, since an expensive device for some (~$10,000) may be quite acceptable to a large utility if it is rapid and highly reliable. Finally, but most importantly, the device must have a high probability of detecting decaying wood at the earliest possible timepoint, while minimizing the amount of sound wood that is labeled as decayed. The latter characteristic becomes particularly important for devices claiming to detect incipient decay since the inspector is incapable of confirming the device's accuracy in the field.

The ideal decay inspection device has not been developed yet and one capable of detecting incipient decay while providing a measure of residual strength is probably beyond the scope of any research program. However, many test methods and devices have been developed that give the inspector a reasonable chance of detecting the intermediate and advanced stages of decay, or estimating residual strength (Fig. 16.1). Unfortunately, wood has undergone substantive changes in properties long before it is visibly decayed (Wilcox, 1978). Losses in some strength properties approach 60% before the wood has lost 5% of its weight. A well-trained wood anatomist can detect the damage microscopically at this early stage, but the wood appears sound visually to the average field inspector. In addition to strength losses, previous studies also suggest that the efficacy of certain remedial treatments, particularly fumigants, decreases in decayed wood due to reduced binding (Morrell and Corden, 1986).

The absence of an ideal device for decay detection forces inspectors to employ several methods to improve the probability that they will detect

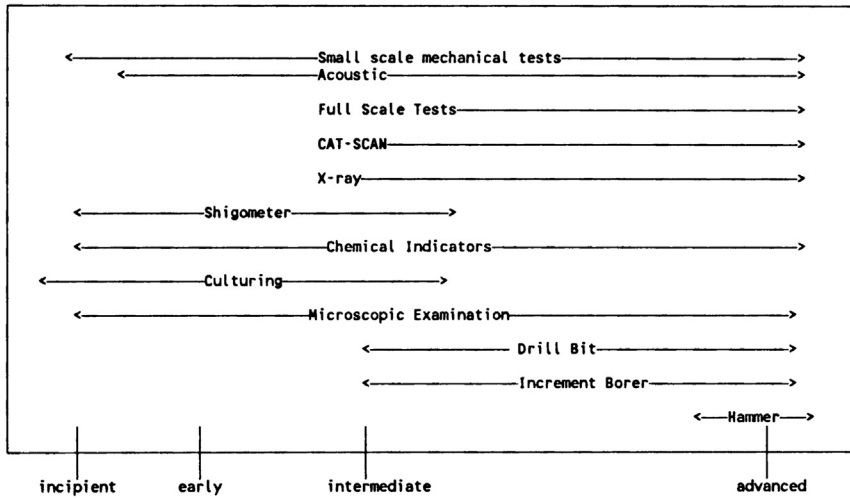


Figure 16.1 Applicability of various wood inspection techniques to the detection of incipient, intermediate, or advanced stages of internal decay.

damage. Current inspection methods can be classified as physical, mechanical, electrical, and acoustic/sonic.

Physical decay detection

Sounding

Sounding with a hammer is perhaps the simplest and most widely used physical wood inspection technique (Morrell, 2012; Inwards and Graham, 1980; Goodell and Graham, 1983). In this test, the experienced inspector pounds the structure along its length with the hammer and listens for changes in sound that signify the presence of advanced decay. This method can be useful for detecting advanced decay where large voids are present, but cannot detect earlier stages of decay when control measures would be most effective. Sounding provides the basis for more sophisticated acoustic inspection devices, but is not sufficiently sensitive to early decay.

Boring

Boring the wood with a drill bit readily detects soft zones or large internal voids. Torque release (changes in the drill speed) is usually evident in the

later decay stages. In addition, a trained inspector can examine the shavings for the evidence of decay such as changes in color or softening, thereby improving the ability to identify decaying poles. The inspection hole can also be used to determine the thickness of the remaining sound shell and can be used to apply remedial internal treatments. While boring the wood has these advantages, it only inspects a small portion of the wood pole, leaving the possibility that a decay pocket will be missed. In addition, each inspection hole removes a small area of the wood cross-section. While relatively minor in terms of overall strength, continued inspection (typically at 10-year intervals) reduces strength significantly if all the holes were drilled in the same plane of wood. Full scale tests found that two sets of inspection holes had no significant effect on flexural properties, but it would be prudent to limit further drilling (Morrell et al., 2014). Proper hole plugging is also necessary to avoid creating water entrapment zones.

Visual examination of increment cores

In this procedure, increment borer cores or plugs are removed and the intact internal wood tissues can be studied visually or with a hand lens for decay features such as pulled fibers, zone lines, variations in color patterns, core compaction, wet zones, and obvious intermediate and late decay stages. The location and extent of any decay on the core can be used to estimate the proportion of residual sound wood in the outer radius. Cores can also be collected aseptically and cultured to determine if viable decay fungi are present. The increment borer hole can be used to apply remedial internal treatments, although the volume of chemical that can be applied is small. Also, any torque release during the boring provides additional information on the wood condition. This type of inspection is not feasible on a regular basis, but it can be helpful for characterizing wood condition when just embarking on an inspection/maintenance program (Kenderes, 1990).

Mechanical decay detection

The primary concern of a wood inspection program is determining the amount of residual strength remaining in a given structure. Although some acoustic tests provide a relative measure of residual strength using

Modulus of Elasticity (MOE) to predict Modulus of Rupture (MOR) using statistical population estimates, only a few of the decay inspection devices currently available in the United States directly test the mechanical strength of the wood.

Compression tests

Small scale mechanical tests of cores, plugs, or veneers removed from wood in service represent one approach to measuring residual wood strength. Longitudinal compression has been shown to be well correlated with MOR of small clear beams and to predict the strength of western redcedar poles (Smith and Morrell, 1987). Radial compression of similar cores detects very early stages of decay (Smith and Graham, 1983); however, this technique measures the weakest growth ring and invariably measures earlywood properties, which are not well correlated to bending properties. While both of these techniques show some promise as tools for selecting high strength wood (Smith and Morrell, 1989), they require the removal of multiple samples from a given structure to develop a reliable strength estimate. Both techniques would also require the use of a mechanical test device in the field and an operator with some degree of training in wood properties. A more sensitive, but less useful property that could be tested on wood samples is toughness. While tests have been developed for evaluating toughness of veneers using breaking radius (Safo-Sampah and Graham, 1976), it is doubtful that these tests could be adapted for field use.

Penetration resistance

The Pilodyn is a spring-loaded, pin penetration device that has been extensively used in other countries to detect surface decay or to measure specific gravity in tree improvement programs (Taylor, 1981; Friis-Hansen, 1980; Hoffmeyer, 1978; Cown, 1978; Cown and Hutchinson, 1983). The depth to which a spring-loaded pin can be driven into the wood is measured and this value can be related to wood density. The Pilodyn has also been used to measure pole hardness in relation to various chemical treatments (Pierce, 1983). Attempts to use the Pilodyn to detect internal decay have been unsuccessful, but the device provides a means for measuring the severity of surface soft-rot attack. While the Pilodyn is relatively non-destructive, one problem that complicates its use is the

need to make moisture content corrections for each reading (Smith and Morrell, 1986).

Drilling torque release

Several internal inspection devices use a power drill equipped with a very fine drill bit and a sensor that detects changes in resistance to drilling. These devices merely reflect the fact that drilling through decayed wood is easier. An experienced inspector can also detect these changes using a traditional drill by listening for sound changes (torque release) as the drill bit penetrates the decayed zone. However, the meter output places a printed or electronic output in the hands of the inspector and provides a record that permits more detailed examination of the results as well as comparisons between multiple inspections over time. The inspection also produces only a small hole, sharply reducing potential strength effects, but the bit also tends to drift in denser woods and care must be taken when drilling at an angle. These devices are widely used for inspection of live trees, but are only used to a limited extent for timber. One drawback to this approach is that the inspector must still determine the nature of the defect (i.e. decay, ring-shake, etc.) using other methods.

Pick or splinter test

The pick test is a simple method for detecting surface decay in poles and timbers. In practice, a sharp screwdriver or awl is driven in the wood at an acute angle and bent back to snap a small piece of wood from the surface (Fig. 16.2). The break characteristics of the splinter removed are then examined. Brash breaks reflect low strength and the possible presence of decay, while splintery breaks reflect sound wood. The pick test measures toughness and is fairly sensitive to early decay (Scheffer and Verrall, 1979; Wilcox, 1983; Morrell et al., 1986a,b). Drawbacks of the pick test include the relatively large size sample removed and the inability to accurately assess internal condition.

Extensiometer

The pole extensiometer was developed in Europe and is a mechanical method for estimating bending strength. It consists of a lever frame that attaches to a tripod that braces the lever against the ground, and an extensiometer fixed between two screws affixed to the pole surface (Morris and Friis-Hansen, 1984). A screw in the tripod turns to mechanically force the

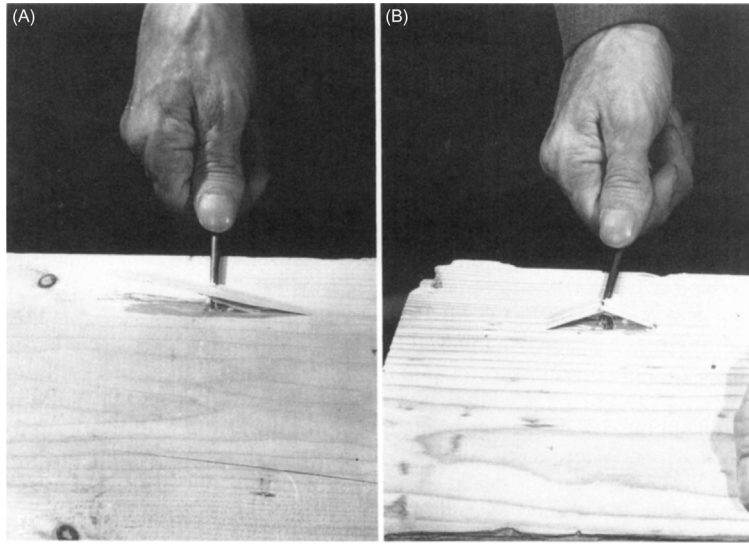


Figure 16.2 A pick or splinter test conducted on a board by Dr. T.C. Scheffer. (A) the splinter break indicates sound wood, (B) the brash break suggests the possible presence of decay. *Photograph M1240721-Courtesy of the U.S. Forest Service, Forest Products Laboratory, Madison, WI.*

lever up and the pole away from the tripod, giving the pole a bending moment between two points. A dial in the lever records the force required to produce a given extension that yields a predicted failure load in kNm. While the pole extensiometer test is non-destructive, the technique has not been widely used, possibly due to the set-up time per test, the inability to test specific sites for decay, or the difficulty of testing poles in line. The Mechanical Pole Tester was developed in Australia and used with some success in North America. These devices provide a measure of stiffness which can be used to estimate Modulus of Rupture. However, they cannot determine the cause of any changes in properties. For example, the device cannot delineate between a timber that was always weak and a strong timber that has become weaker as a result of active decay.

Vibration

Vibrational testing does not strictly measure mechanical properties; it does measure the ability of the wood to transmit energy. The wood is struck by a rubber mallet and the vibrational characteristics of the wood are measured (Murphy et al., 1987; Murphy and Taylor-Wilson, 1988). Sound wood vibrates more rapidly than decayed samples and these differences

can be used to determine residual wood strength. The vibrational technique does not determine the location or nature of damage and is not applicable to wood that contains hardware such as wires or connectors. It also requires considerable knowledge about the characteristics of the wood species being tested. For example, wood may differ in vibrational characteristics on the basis of density or ring characteristics and these must be characterized in order to understand how decay or other defects affect the results.

Full-Scale Testing

The best method for assessing wood strength is to test the structure to failure. Obviously testing structures to failure to determine their wood strength is not practical, but limited full-scale testing of structures does have a place in any large-scale decay detection program. Full-scale testing of some units provides a measure of reliability for the decay detection methods employed and provides a valuable training exercise as well as reference materials for inspectors. Wood users should consider routine testing of occasional selected structures to train their inspectors. In practice, the inspectors would inspect the wood using their approved methods, then make an assessment on wood condition and treatment recommendations. The structure would then be tested to failure and dissected to determine the nature and location of any defects present. In this way, the inspectors learn the accuracy of each device for detecting specific wood defects.

Electrical decay detection

A number of inspection methods have been developed that use the electrical properties of wood and the moisture often associated with wood in service.

Moisture meters

The simplest electrical inspection device is the moisture meter, which measures wood resistance in ohms between two metal pins driven in the wood (James, 1988). Since dry wood (<20% moisture content) will not decay, determining wood moisture content can provide a relative measure of decay risk. Moisture meters are often used by building inspectors to

identify locations where elevated moisture levels suggest possible fungal or insect attack. Wood pole inspectors assess moisture levels to ensure that post-treatment moisture requirements are met.

Wood moisture contents often vary seasonally. In addition, the region sampled by the moisture meter is limited by the length of the pins, which range from 2.5 to 7.5 cm long. Thus, the moisture meter is of little use for detecting internal decay or the conditions with which this damage is often associated. Furthermore, the moisture meter is sensitive to temperature (James, 1988) and the presence of inorganic salt-type wood preservatives (James, 1980). Currently available moisture meters represent a useful tool for the inspector, but cannot detect decay or determine the extent of any infestation.

Shigometer

The Shigometer measures changes in electrical resistance of wood using a twisted wire probe inserted into a hole drilled in the wood. In principle, the Shigometer measures changes in electrical resistance caused by the release of ions as fungi degrade the wood (Shigo and Shigo, 1974). There is considerable debate about the usefulness of this instrument, which some equate with the moisture meter (Piirto and Wilcox, 1978). The Shigometer appears to be very useful for detecting decay in living trees, where wood moisture contents are uniformly above the fiber saturation point (Shigo and Shigo, 1974), but has produced inconsistent results in timber products where moisture contents vary more widely (Shigo et al., 1977; Shortle et al., 1978; Shortle, 1982; Thornton, 1979). A limitation of the device is that at the time of the test the wood must be above the FSP for useful readings. In addition, several field trials indicate that the Shigometer requires a higher degree of experience to interpret the patterns of resistance changes characteristic of early delay than other currently used wood inspection devices (Zabel et al., 1982). The Shigometer was used commercially for a time to inspect utility poles, but its application has declined as users have seen little improvement over existing decay detection methods.

X-ray

X-ray inspection was once commercially used to detect internal voids in wood and has the ability to detect small changes in wood density related to decay or other defects (Mothershead and Stacey, 1965). In a comparison of methods of decay detection in poles, x-ray radiographs appeared to be

effective primarily when the wood was in the intermediate to advanced decay stages (Zabel et al., 1982). X-ray units have a high initial expense, require safety training for operators and generally require fairly sophisticated analyzes. In addition, many users are concerned about potential health risks associated with ionizing radiation. As a result, x-rays are primarily used to detect marine borer infestations in marine test panels, insect attack in museum test pieces, or for measuring wood density in relation to wood quality (Hoag and McKimmy, 1988; Yamamoto and Fujii, 1987). However, railroad track inspectors are evaluating a commercial device that x-rays ties as it passes overhead at speeds approaching 40 km/hour and identifies ties with voids or other defects. This system confines the x-rays to the area between the rails, but that is where most of the decay and damage occurs. Systems such as these will become increasingly common as improved processing technologies allow data to be processed in real time.

Tomography

Computer aided axial tomography scanning (CATSCAN) was developed in Europe to produce three dimensional maps of internal wood conditions (Hattori and Kanagawa, 1985; Morris and Friis-Hansen, 1984; Lindgren, 1987). One such device was attached around a structure and moved upward while performing a scan. The data were collected and analyzed to develop the internal map. CAT-SCANs represent a potentially useful tool, but the time required to collect and process the data, as well as the initial equipment costs, currently make this technique impractical for field purposes. Improvements in data processing and micro-circuitry will ultimately make this method practical, but high initial costs will probably prevent extensive field use of this technique.

Acoustic decay detection

Sound waves moving through a material are modulated by characteristics such as density, knots, growth rings and decay. It is possible to look at relatively simple characteristics such as the time it takes for a wave to move through the wood, but it is also possible to examine how the wave is modulated to assess changes in properties. Acoustic detection of decay and other wood defects has received much recent attention, as techniques have been refined for rapid data processing under field conditions.

Acoustic emissions

Acoustic emission refers to elastic waves produced by deformation and failure processes in stressed materials as a result of the presence of small defects (Knuffel, 1988). Acoustic emission has been used to evaluate incipient decay in western hemlock specimens that were stressed in bending (Noguchi et al., 1986). Acoustic emission appears to be useful for detecting defects in small samples that can be easily manipulated, but most structures such as poles present formidable challenges to the use of acoustic emission since they are already in tension and compression. In addition, any equipment attached to the structure or weather factors such as changes in wind velocity and direction during the test can affect the results. Thus, large wood structures do not appear to permit the controlled test conditions required for acoustic emission analysis. However, acoustic emissions have been used to detect insects in timbers, especially termites as the workers excavate new tunnels.

Stress wave timers

The use of stress wave timers that measure the time required for a sound wave to travel through a material, represented a major advance in the ability of wood users to detect internal defects. In principle, a sound wave is induced at one point on the wood and a transducer picks up that signal at a second point. The time between sending and receiving can be correlated with wood MOE and density (Agi, 1983; Bucur, 1983; Ross and Pellerin, 1988; Ross et al., 1994; Yang et al., 2017). These data can also be related to the presence of internal voids or other defects since the wave will travel faster through sound wood (Pellerin et al., 1985). While stress wave timers can be used to detect the presence of defects (Wilcox, 1988), they cannot distinguish between active decay, voids, ring shakes or other wood defects. One device, the PolTek, was developed for inspecting the groundline of wood poles. Several field trials indicated that this device was less sensitive than conventional inspection methods (Inwards and Graham, 1980). In addition, the device was unable to distinguish between defects that did not adversely affect strength and more serious defects such as decay or insect attack.

More recently developed devices appear to be more accurate and are often used for assessing the condition of poles that have been rejected by other inspection methods. The most common system uses time of flight to estimate a modulus of elasticity and then relates this value to MOR values developed using full scale testing on poles. Other devices develop

time of flight measurements at multiple points around a wood member and use these values to detect the presence of voids or other defects. The resulting map of internal voids can be used to estimate the thickness of the remaining sound wood and this can be used to calculate a residual strength based upon an assumed strength value for that wood species. While some of these tests claim to detect early, or incipient decay, there is no evidence that the can do so with any reliability.

Wave form analysis

Continued improvements in data processing have led a number of investigators to explore advanced sonic inspection techniques. In addition to velocity measurements, these devices attempt to analyze portions of the resulting sound wave (Anonymous, 1987). Sound waves passing through a material are modified or modulated by the characteristics of that material. Non-decayed wood transmits sound waves more efficiently and with less modification than decayed wood. These differences can be analyzed and incorporated with velocity measurements to develop estimates of residual wood strength (Wilson, 1988). These types of devices provide residual strength values which are based upon previous data collected from a large population of the same material (poles, piling, timbers) that were destructively tested and used as reference data (Bodig, 1986). While such devices represent a step forward in wood inspection, there are lingering questions concerning the performance of the currently available system. Sonic devices cannot detect the presence of decay, nor can they determine the exact location of any damage. Thus, they can provide an estimate of residual strength, but no estimate of future risk of increased damage, or the extent of existing decay damage in individual wood members.

As sonic technology improves, wave form analysis should eventually replace velocity measurement. At present, the complexity of the wave after it has been modified by the wood is too great to provide useful information; however, defining the components of the wave can permit more detailed analysis. Despite this anticipated improvement, it is doubtful that sonic testing will be capable of detecting incipient decay or determining whether this decay is active.

Ground Penetrating Radar

Developed for detecting differences in density and now more commonly used for detecting soil disturbances suggestive of historic sites, ground

penetrating radar has also been explored for detecting decay on both ground and helicopter mounted platforms. The system was accurate, but the processing times required necessitated return trips to verify the results. As a result, the efforts to use this technology were abandoned.

Spectroscopy tools

Infrared spectroscopy

Infrared (IR) light has a longer wavelength than visible light and is invisible to the naked eye. Light in this region can be used to indirectly measure temperatures. Infrared cameras are widely used for detecting temperature anomalies. For example, IR cameras at many international airports are designed to identify people who are entering with high fevers suggestive of carrying diseases. IR cameras have also been used to identify wet areas in structures. The premise is that wet and dry wood will heat differently and these subtle differences can be detected using IR. As with many other non-destructive tools, IR sensors cannot detect decay, only conditions that might be suitable for its development.

Near Infrared Spectroscopy (NIR)

NIR uses the wavelengths between 780 and 2500 nm to assess differences in materials. NIR spectroscopy has been used to estimate wood density, flexural properties and a host of other characteristics, including relative decay resistance with mixed results (Schimleck et al., 1996; Schimleck and Evans, 2002; Taylor et al., 2008). The NIR spectra is quite complex and difficult to understand and tools such as Principle Components analysis are used to identify trends for various properties.

Laboratory decay detection

While most laboratory techniques are not currently directly applicable for field use, they can provide valuable information for inspection programs and be used to develop more accurate decay detection devices.

Culturing

Culturing involves the isolation, growing, and identification of the fungi inhabiting a wood sample. The process normally takes 4–6 weeks and requires the services of a trained microbiologist as well as laboratory facilities. Culturing can determine the reliability of the decay criteria used in inspection programs and provide valuable clues concerning when and where some fungi entered the poles (Zabel et al., 1980; Eslyn, 1970; Graham and Corden, 1980). The technique can also authenticate uncertain decay diagnoses and indicate when preservative treatments are approaching threshold levels and remedial treatments are necessary (Zabel and Wang, 1988). Culturing can determine if viable decay fungi are present (Ricard and Mothershead, 1966; Graham and Corden, 1980; Morrell, 2012; Morrell et al., 1987). Culturing provides a useful measure of decay risk and the presence of viable fungi indicates that favorable decay conditions are present. Identification of the fungi isolated is an important feature of the culturing process and can provide useful information on the decay hazard and residual preservative effectiveness (Wang and Zabel, 1990; Stalpers, 1978; and Nobles, 1965). Culturing cannot distinguish between active decay and the presence of viable, but inactive fungi. Culturing may also artificially inflate the importance of fungi that sporulate heavily or those that grow well under the cultural conditions used. Because of the delay between inspection and cultural results, culturing probably is not feasible for routine wood inspection, but can provide a valuable supporting role.

DNA Based Identification: This is a broad and ever-expanding topic. At the least, culturing can be coupled with isolation of DNA from the resulting fungal cultures and this DNA can be amplified and sequenced for identification by comparison with various databases. The ability to identify a fungus without requiring extensive knowledge of the taxonomy of that fungus is a major advantage; however, care must be taken to ensure that the deposited sequence actually belongs to the fungus.

A more powerful tool that is just emerging is high-throughput sequencing. In this approach, DNA is extracted from a substrate such as soil, wood or plant tissue and sequenced using primers specific to various taxa. These sequences are then compared with databases to determine the organisms whose DNA is present. This approach results in a much greater number of organisms being identified as present; however, care must be taken since the technique cannot distinguish between organisms that might be present as only a few spores on or in the wood vs those that are

extensively degrading substrate. This technique; however, offers the opportunity to gain a better understanding of the microbial diversity present in various substrates especially those organisms that cannot be cultured on synthetic media.

Microscopy

Light microscopy has been effectively used to detect incipient decay and study the progressive stages of degradation (Wilcox, 1970, 1978; Bravery, 1971). While light microscopy detects decay at very early stages, it does not appear to be practical for field use and extensive training is generally required for this time-consuming procedure. Light microscopy can be useful along with culturing to identify the features of decay that can be difficult to detect by simpler, more practical methods in the field.

Chemical indicators

The use of chemical indicators for detecting decay has been proposed by a number of researchers (Lindgren, 1955; Cowling and Sachs, 1960; Eslyn, 1979; Line, 1982; Gilbertson et al., 1975). In general, these indicators are based upon changes in pH within the wood as the fungus alters wood structure or releases metabolic acids. In principle, indicators can be used to detect various stages of fungal attack. In practice, however, indicators have shown a wide degree of variability. Eslyn (1979) reviewed the use of nine indicators that showed some promise for detecting specific fungal agents in southern pine cores. More recently, a fluorescent dye, acridine-orange, has been used to detect attack by brown rot fungi (Krahmer et al., 1982). Subsequent studies have shown that the results of this technique were characterized by extensive operator variation. While currently available indicators have proved to be of limited use, successful development of such indicators would allow decay detection in the field.

Lectins

Another class of fungal indicators are the plant-derived lectins. These compounds are highly specific for sugars and have been used with great success as probes in cell biology (Leiner, 1976). Several lectins have been identified that are specific for components of the fungal cell wall. One lectin, wheat germ agglutinin (WGA), is highly specific for fungal chitin, which is a cell wall component of most higher fungi (Morrell et al., 1985). Lectins can be visualized in the wood by coupling to a fluorescent

or metal compound and observing it under a properly equipped microscope. Preliminary studies indicate that lectins improve the ability to detect fungi at very early stages of attack (Morrell et al., 1985; Krahmer et al., 1986). A wide array of non-dematiaceous (dark-pigmented) fungi can be detected using WGA (Morrell et al., 1986a,b). This technique, however, cannot distinguish between living and dead fungi.

Serological tests

Related to the development of indicators, serological techniques based upon the development of antibodies specific for the presence of a compound or class of compounds also show promise for decay detection (Goodell et al., 1988a,b; Daniel et al., 1988; Srebotnik and Messner, 1990; Garcia et al., 1987). The use of serological techniques to study wood decay was proposed over 50 years ago (Ricard and Mothershead,1966); however, the methodology has recently seen dramatic improvements. The presence of fungal hyphae or compounds specific to the decay process can be detected by coupling an antibody to a fluorescent compound or by using enzyme-linked immunosorbent assays (ELISA). The latter technique is extensively used as a diagnostic tool for the detection of viruses, such as that which causes acquired immune deficiency syndrome. ELISA could provide a high degree of reliability if a single factor common to wood decay by fungi could be identified. Unfortunately, decay types differ widely, making identification of a single factor difficult. If an enzyme unique to decay fungi could be identified and it was always present, serological detection of fungal decay could prove very useful. This technique would only be practical when the fungus was actively degrading the wood, since the enzymes produced are normally present at low levels and are relatively ephemeral. Thus, inspections during warm summer months when decay fungi are more active would be more likely to detect decay using such techniques. The ideal serological system will probably use an antibody specific for chitin (most higher fungi contain chitin) and a second antibody specific for an enzyme typically produced by wood decay fungi.

Analytical techniques

While chemical and cultural decay detection methods have been available for some time, recent advances in analytical techniques have permitted the use of more sophisticated chemical methods for decay detection.

As mentioned earlier, IR spectroscopy has been evaluated for detecting incipient decay in a number of conifers and hardwoods (Gibson et al., 1985; Nicholas and Schultz, 1987). IR-spectroscopy can detect relatively small shifts in the ratios of sugars and phenols in the wood that occur when wood is either actively attacked by fungi or subjected to photodegradation. A preliminary study indicated that this technique could detect incipient decay by brown rot fungi through the presence of a specific transmittance peak produced at the early stages of fungal attack; however, the technique was not as useful for detecting attack by white-rot fungi (Gibson et al., 1985). The peak correlated with brown rot was tentatively identified as a key lignin breakdown product but the method was not pursued (Holt, 1986).

IR spectroscopy has been improved by the use of Fourier transformations (FT-IR) that can increase the precision of analysis, permitting the detection of the more subtle shifts in IR spectra (Nicholas and Schultz, 1987). At present, the equipment required for IR detection of decay limits this procedure to the laboratory, but continued development of portable microprocessors may permit extension of this technique to the field. In addition, further studies are required to identify characteristics of decay by white rot and soft rot fungi that can be used for IR spectroscopy. This search is compounded by fact that these groups of fungi generally utilize wood breakdown products at rates that approximately correspond to the rate of degradation, leaving few breakdown products available for detection. In addition, the technique must be refined to distinguish between active and inactive decay, since incipient decay prior to treatment may be inactivated, but still produce a positive response with the IR.

In addition to these laboratory techniques, a number of other analytical methods may have application to decay detection. Infrared photo acoustic spectroscopy (FTIR-PAS) is a relatively recent development for examining solid wood samples (Kuo et al., 1988). In conventional FTIR, the samples are ground, extracted and compressed in a potassium bromide pellet (KBr). The process is fairly slow and not amenable to field sampling. FTIR-PAS permits solid state examination of relatively thick sections (400 μm). Although it has not yet been studied for detecting decay, the use of solid samples would permit more rapid analysis.

Scanning wood using microwaves has been proposed for detecting defects in lumber (Martin et al., 1987); however, the sensitivity of such a device in a thicker wood sections and its ability to distinguish between the various defects present are unknown.

Differential scanning calorimetry has also been proposed for detecting degraded wood (Reh et al., 1986; Baldwin and Streisel, 1985). This technique requires oven temperatures of approximately 600 °C and appears to be feasible only for laboratory studies of decayed materials.

One technique that may have some applicability to decay detection when used in conjunction with indicators, are the optical characteristics of the wood. Changes in the luster and color of wood as it decays have been noted by a number of researchers (Zabel et al., 1982). Optical measurements could be used to detect decay if the characteristics of color as wood decayed could be quantified. Indicators could also be used to enhance the chances of detecting the damage. Successful identification of color characteristics would require a high degree of background work since different decay fungi may cause different color changes, particularly between wood species. In addition, the method would have to be capable of detecting decay in the presence of preservative treatments that alter the wood color. While field practical optical devices for this purpose are not currently available, it is likely that devices used for other applications could be modified with relatively little effort.

Olefactory decay detection

As wood is degraded by fungi and insects, certain volatile chemicals are emitted. While these chemicals cannot be detected by humans, they can be detected analytically. At present, the current cost of instrumentation and the degree of training required for these purposes are beyond the means of most inspection agencies or utilities. However, some animals also have the ability to detect these emissions. The use of sniffer dogs to detect decay has been proposed in Europe and dogs have been used in the United States to detect termite infestations in buildings (Morris and Friis-Hansen, 1984). At present the use of dogs for decay detection remains largely an interesting idea with many uncertainties as to its feasibility under the wide range of conditions and treatments for wood in use.

As the fundamental aspects of decay are unraveled, it is likely that more sophisticated and accurate methods for detecting this damage will be developed.

Summary

1. The reliable detection of incipient and early decay is a major problem in many wood uses and remains an important research goal in wood microbiology.

2. The detection of internal decay in its early stages is difficult because of its occurrence in locations that are difficult to access. Detection of decay poses major challenges due to the sporadic patterns of deterioration, the range of effects, and the wide variety of natural defects present in wood.
3. Points where moisture can collect, such as deep checks, crawl spaces, soil contact, around window frames, or leaking pipes represent the most likely zones for detecting decay and should be investigated first.
4. There are a wide variety of inspection techniques but these methods can be classified as chemical, mechanical, electric, or acoustic. No universally effective inspection technique has been found and decay detection often requires the use of more than one device.
5. Mechanical methods such as drilling or boring are most frequently used for detecting internal decay, but efforts are underway to develop improved electric and acoustic detection devices. In addition, efforts to use serological techniques and other chemical indicators for detecting the presence of fungi or their degradation products have been studied.

References

Agi, J.J., 1983. Structural evaluation of marine structures. Proceedings, Specialty Conference on Port Modernization, Upgrading and Repairs. American Society of Civil Engineers, New Orleans, Louisiana.

Anonymous, 1987. Innovative wood-pole-management program uses NDE technique. Electr. World 39–43.

Baldwin, R.C., Streisel, R.C., 1985. Detection of fungal degradation at low weight loss by differential scanning calorimetry. Wood Fiber Sci. 17 (3), 315–326.

Bodig, J., 1986. Practical definition of wood pole strength: relation to construction standards. Proceedings, The Wood Pole Conference. Portland, OR.

Bravery, A.F., 1971. The application of scanning electron microscopy in the study of timber decay. Wood Sci. Technol. 19, 35–46.

Bucur, V., 1983. An ultrasonic method for measuring the elastic constants of wood increment cores bored from living trees. Ultrasonics 83, 116–126.

Cowling, E.G., Sachs, I.B., 1960. Detection of brown rot with osmium tetroxide stain. For. Prod. J. 10 (11), 594–596.

Cown, D.J., 1978. Comparison of the Pilodyn and torsiometer methods for the rapid assessment of wood density in living trees. New Zealand J. For. Sci. 8 (3), 384–391.

Cown, D.J., Hutchinson, J.D., 1983. Wood density as an indicator of the bending properties of *Pinus radiata* poles. New Zealand J. For. Sci. 13 (1), 87–99.

Daniel, G.F., Nilsson, T., Petterson, B., 1988. Immunolabelling Studies on the Detection of Enzymes During the Degradation of Wood *by Phanerochaete chrysosporium*. International Research Group on Wood Preservation Document IRG/WP/1364, Stockholm, Sweden.

Eslyn, W.E., 1970. Utility pole decay. Part 2. Basidiomycetes associated with decay in poles. Wood Sci. Technol. 4, 97–103.

Eslyn, W.E., 1979. Utility pole decay. Part 3. Detection in pine by color indicators. Wood Sci. Technol. 13, 117–126.

Friis-Hansen, H., 1980. A Summary of Tests and Practical Experiences With the Pilodyn Wood Testing Instrument. International Research Group on Wood Preservation Document No. IRG/WP/282, Stockholm.
Garcia, S., Latge, J.P., Prevost, M.C., Leisola, M., 1987. Wood degradation by white rot fungi: cytochemical studies using lignin peroxidase immunoglobulin-gold complexes. Appl. Environ. Microbiol. 53 (10), 2384–2387.
Gibson, D.G., Krahmer, R.L., DeGroot, R.C., 1985. Early detection of brown rot decay in Douglas-fir and southern yellow pine by infrared spectrophotometry. Wood Fiber Sci. 17 (4), 522–528.
Gilbertson, R.L., Lombard, F.F., Canfield, E.R., 1975. Gum Guaiac in Field Tests for Extracellular Phenol Oxidases of Wood-rotting Fungi and Other Basidiomycetes. USDA For. Sere Res. Paper FPL 269, Madison, WI.
Goodell, B.S., Graham, R.D., 1983. A survey of methods used to detect and control fungal decay of wood poles in service. Int. J. Wood Preserv. 3 (2), 61–63.
Goodell, B.S., Daniel, G., Jellison, J., Nilsson, T., 1988a. Immunolocalization of Extracellular Metabolites from *Poria placenta*. International Research Group on Wood Preservation Document No. IRG/WP/1361, Stockholm, Sweden.
Goodell, B.S., Jellison, J., Hosli, J.P., 1988b. Serological detection of wood decay fungi. For. Prod. J. 38 (3), 59–62.
Graham, R.D., Corden, M.E., 1980. Controlling Biological Deterioration of Wood With Volatile Chemicals. Electric Power Research Institute Report EL 1480, Palo Alto, CA.
Hattori, Y., Kanagawa, Y., 1985. Non-destructive measurement of moisture distribution in wood with a medical CT-scanner. J. Jpn. Wood Res. Soc. 31 (12), 974–982.
Hoag, M., McKimmy, M.D., 1988. Direct scanning x-ray densitometry of thin wood sections. For. Prod. J. 38 (1), 23–26.
Hoffmeyer, P., 1978. The Pilodyn instrument as a non-destructive tester of the shock resistance of wood. Proceedings, Non-Destructive Testing Symposium. Vancouver, WA.
Holt, K., 1986. Chemical Characterization of Breakdown Products Involved in Detection of Incipient Decay in Wood (M.S. thesis). Oregon State University, Corvallis, Oregon.
Inwards, R.D., Graham, R.D., 1980. Comparing methods for inspecting Douglas-fir poles in service. Proc. Am. Wood Preserv. Assoc. 76, 283–286.
James, W.L., 1980. Effects of Wood Preservatives on Electric Moisture-meter Readings. U.S. Forest Service, Forest Products Laboratory Research Note FPL0106, Madison, Wisconsin.
James, W.L., 1988. Electric Moisture Meters for Wood. U.S. Forest Service, Forest Products Laboratory General Technical Report FPL-GTR-6, Madison, Wisconsin.
Kenderes, A.M., 1990. Personal Communication. Wood Engineer. New York State Electric and Gas Corp., Binghamton, NY.
Knuffel, W.E., 1988. Acoustic emission as strength predictor in structural timber. Holzforschung 42 (3), 195–198.
Krahmer, R.L., DeGroot, R.C., Lowell, E.C., 1982. Detecting incipient brown rot with fluorescence microscopy. Wood Sci. 15 (2), 78–80.
Krahmer, R.L., Morrell, J.J., Choi, A., 1986. Double-staining to improve visualization of wood decay hyphae in wood sections. Int. Assoc. Wood Anatomists Bull. 7 (2), 165–167.
Kuo, M., McClelland, J.F., Luo, S., Chien, P.L., Walker, R.D., Hse, C.Y., 1988. Applications of photoacoustic spectroscopy for wood samples. Wood Fiber Sci. 20 (1), 132–145.
Leiner, I.E., 1976. Phytohemagglutinins (Phytolectins). Annu. Rev. Plant. Physiol. 27, 291–319.

Lindgren, R.M., 1955. Color Test for Early Storage Decay in Southern Pine. U.S.D.A. Forest Service, Forest Products Laboratory Report Number 2037, Madison, Wisconsin, 5 p.

Lindgren, O., 1987. Computerized Axial Tomography: a Nondestructive Method for Three-dimensional Wood Density/Moisture Content Measurements. International Research Group on Wood Preservation Document No. IRG/WP/2285, Stockholm, Sweden.

Line, M.A., 1982. Toward a Colour Assay of Wood Degradation. International Research Group on Wood Preservation Document No. IRG/WP/2180, Stockholm, Sweden.

Martin, P., Collet, R., Barthelemy, P., Roussy, G., 1987. Evaluation of wood characteristics: internal scanning of the material by microwaves. Wood Sci. Technol. 21, 361–371.

Morrell, J.J., 2012. Wood Pole Maintenance Manual: 2012 Edition. Research Contribution 51. Forest Research Laboratory, Oregon State University, Corvallis, OR, 46 pages.

Morrell, J.J., Wilson, J.B., 1985. Detecting and controlling decay of wood utility poles in service. In: Proceedings, Northwest Public Power Association Engineering and Operations Conference. Portland, Oregon.

Morrell, J.J., Corden, M.E., 1986. Controlling wood deterioration with fumigants: a review. For. Prod. J. 36 (10), 26–34.

Morrell, J.J., Gibson, D.G., Krahmer, R.L., 1985. Effect of fluorescent-labeled lectins on visualization of decay fungi in wood sections. Phytopathology 75, 329–332.

Morrell, J.J., Krahmer, R.L., Lin, L.C., 1986a. Use of Fluorescent-coupled Lectins as Probes for Studying Fungal Degradation of Wood. International Research Group on Wood Preservation Document No. IRG/WP/1288, Stockholm, Sweden.

Morrell, J.J., Smith, S.M., Newbill, M.A., Graham, R.D., 1986b. Reducing internal and external decay of untreated Douglas-fir poles: a field test. For. Prod. J. 36 (4), 47–52.

Morrell, J.J., Corden, M.E., Graham, R.D., Kropp, B.R., Przybylowicz, P., Smith, S.M., et al., 1987. Basidiomycetes colonization of Douglas-fir poles during air-seasoning and its effect on wood strength. Proc. Am. Wood Preserv. Assoc. 83, 284–296.

Morrell, J.J., Sinha, A., Clauson, M., Love, C.S., 2014. Effect of inspection holes on flexural properties of Douglas-fir utility poles. For. Prod. J. 64 (7/8), 300–313.

Morris, P.I., Friis-Hansen, H., 1984. Report on a Field Demonstration of Methods for Detecting Defects in Wood Poles. International Research Group on Wood Preservation Document No. IRG/WP/2232, Stockholm, Sweden.

Mothershead, J.S., Stacey, S.S., 1965. Applicability of radiography to inspection of wood products. In: Proceedings, Second Symposium on Non-Destructive Testing. pp. 307–336.

Murphy, M.W., Taylor-Wilson, J., 1988. Vibration analysis: an alternative to sonic testing. In: Proceedings, The Wood Pole Conference. Portland, OR.

Murphy, M.W., Franklin, D.E., Palylyk, R.A., 1987. A Non-destructive Testing Technique for Wood Poles. International Research Group on Wood Preservation Document No. IRG/WP/2293, Stockholm, Sweden.

Nicholas, D.D., Schultz, T.P., 1987. Detection of Incipient Brown Rot Decay in Wood by Fourier Transform Infrared Spectroscopy. International Research Group on Wood Preservation Document Number IRG/WP/2275, Stockholm, Sweden.

Nobles, M.F., 1965. Identification of cultures of wood inhabiting hymenomycetes. Can. J. Bot. 43, 1097–1139.

Noguchi, M., Nishimoto, K., Imamura, Y., Fujii, Y., Okumura, S., Miyauchi, T., 1986. Detection of very early stages of decay in western hemlock wood using acoustic emissions. For. Prod. J. 36 (4), 35–36.
Pellerin, R.F., DeGroot, R.C., Esenther, G.R., 1985, Non-destructive stress wave measurements of decay and termite attack in experimental wood units. Proceedings 5th Nondestructive Testing of Wood Symposium. Pullman, WA. pp. 319–352.
Pierce, B., 1983. Hardness-Penetration Tests on Three Species and Five Treatments (Unpublished Report). Arizona Public Service.
Piirto, D.O., Wilcox, W.W., 1978. Critical evaluation of the pulsed current resistance meter for detection of decay in wood. For. Prod. J. 28, 52–57.
Reh, U., Kraepelin, G., Lamprecht, I., 1986. Use of differential scanning calorimetry for structural analysis of fungally degraded wood. Appl. Environ. Microbiol. 52 (5), 1101–1106.
Ricard, J.L., Mothershead, J.S., 1966. Field procedure for detecting early decay. For. Prod. J. 16 (7), 58–59.
Ross, R.J., Pellerin, R.F., 1988. NDE of wood-based composites with longitudinal stress waves. For. Prod. J. 38 (5), 39–45.
Ross, R.J., Ward, J.C., TenWolde, A., 1994. Stress wave non-destructive evaluation of wetwood. For. Prod. J. 44, 79–83.
Safo-Sampah, S., Graham, R.D., 1976. Rapid agar-stick breaking-radius test to determine the ability of fungi to degrade wood. Wood Sci. 9 (2), 65–69.
Scheffer, T.C., Verrall, A.F., 1979. Principles for Protecting Wood Buildings From Decay. U.S.D.A. Forest Service Research Paper FPL-190.
Schimleck, L.R., Evans, R., 2002. Estimation of wood stiffness of increment cores by near infrared spectroscopy: the development and application of calibration based on increment cores. IAWA J. 23 (3), 217–224.
Schimleck, L.R., Mitchell, A.J., Vinden, P., 1996. Eucalyptus wood classification by near infrared spectroscopy and principal components analysis. APPITA J. 49 (5), 319–324.
Shigo, A.L., Shigo, A., 1974. Detection of Discoloration and Decay in Living Trees and Utility Poles. U.S.D.A. Forest Service Research Paper NE 294.
Shigo, A.L., Shortle, W.C., Ochrymowych, J., 1977. Detection of Active Decay at Groundline in Utility Poles. U.S. Forest Service General Technical Report NE-35, Upper Darby, PA.
Shortle, W.C., 1982. Decaying Douglas-fir wood: ionization associated with resistance to a pulsed electric current. Wood Sci. 15 (1), 29–32.
Shortle, W.C., Shigo, A.L., Ochrymowych, J., 1978. Patterns of resistance to a pulsed electric current in sound and decayed utility poles. For. Prod. J. 28 (1), 48–51.
Smith, S.M., Graham, R.D., 1983. Relationship between early decay and radial compression strength of Douglas-fir. For. Prod. J. 33 (6), 49–52.
Smith, S.M., Morrell, J.J., 1986. Correcting Pilodyn measurement of Douglas-fir for different moisture levels. For. Prod. J. 36 (1), 45–46.
Smith, S.M., Morrell, J.J., 1987. Longitudinal compression as a measure of ultimate wood strength. For. Prod. J. 37 (5), 49–53.
Smith, S.M., Morrell, J.J., 1989. Comparing full-length bending strength and small-scale test strength of western redcedar poles. For. Prod. J. 39 (3), 29–33.
Srebotnik, E., Messner, K., 1990. Immunogold Labelling of Size Marker Proteins in Brown Rot-Degraded Pine Wood. International Research Group on Wood Preservation. Document No. IRG/WP/1428, Stockholm, Sweden.
Stalpers, J.A., 1978. Identification of wood inhabiting Aphyllophorales in pure culture. Central Bureau Voor Schimmenlcultures, Baarn. Stud. Mycol. 16, 1–48.
Taylor, F.W., 1981. Rapid determination of southern pine specific gravity with a Pilodyn tester. For. Sci. 27 (1), 59–61.

Taylor, A.M., Freitag, C., Cadot, E., Morrell, J.J., 2008. Potential of near infrared spectroscopy to assess hot-water-soluble extractive content and decay resistance of a tropical hardwood. Holz Roh Werkst. 66, 107–111.
Thornton, J.D., 1979. Detection of decay in wood using a pulsed-current resistance meter (Shigometer). I. Laboratory tests of the progression of decayof *Pinus radiata* D. Don sapwood by *Poria monticola* Murr. and *Fomes lividus* (Kalch.) Sacco. Mater. Org. 14 (1), 15–26.
Wang, C.J.K., Zabel, R.A., 1990. Identification Manual for Fungi From Utility Poles in the Eastern United States. American Type Culture Collection, Rockville, MD, 356 pp.
Wilcox, W.W., 1970. Anatomical changes in wood cell walls attacked by fungi and bacteria. Bot. Rev. 36 (1), 1–28.
Wilcox, W.W., 1978. Review of the literature on the effects of early stages of decay on wood strength. Wood Fiber 9 (4), 252–257.
Wilcox, W.W., 1983. Sensitivity of the "pick test" for field detection of early wood decay. For. Prod. J. 33 (2), 29–30.
Wilcox, W.W., 1988. Detection of early decay stages of wood decay with ultrasonic pulse velocity. For. Prod. J. 38 (5), 68–73.
Wilson, J.B., 1988. An overview of non-destructive testing. Proceedings, The Wood Pole Conference. Portland, OR.
Yamamoto, K., Fujii, T., 1987. Application of soft-x-ray microdensitometry to 2-dimensional quantitative evaluations of density decreases by wood decay. J. Jpn. Wood Res. Soc. 33 (12), 974–982.
Yang, Z., Jiang, Z., Hsu, C.Y., Liu, R., 2017. Assessing the impact of wood decay fungi on the modulus of elasticity of slash pine (*Pinus elliottii*) by stress wave non-destructive testing. Intl. Biodeterior. Biodegrad. 117, 123–127.
Zabel, R.A., Wang, C.J.K., 1988. Utility pole decay problems in the Eastern United States: changing viewpoints. In: Proceedings, The Wood Pole Conference, Portland. OR, pp. 69–80.
Zabel, R.A., Lombard, F.F., Kenderes, A.M., 1980. Fungi associated with decay in treated Douglas-fir transmission poles in the Northeastern United States. For. Prod. J. 30 (4), 51–56.
Zabel, R.A., Wang, C.J.K., Terracina, F.C., 1982. The Fungal Associates, Detection, and Fumigant Control of Decay in Treated Southern Pine Poles (Final Report). Electric Power Research Institute. EPRI EL-2768, Palo Alto, CA.

Further reading

Anagnost, S.E., Zhou, S., Yeo, H., Wang, C.J.K., Smith, W.B., Roberts, D.M., 2006. Fungi inhabiting southern pine utility poles during manufacture. For. Prod. J. 56 (1), 53–59.
Graham, R.D., 1983. Improving the performance of wood poles. Proc. Am. Wood Preserv. Assoc. 79, 221–228.
Morris, C.O., 1960. Ultrasonics for Wood Industries. Report No. E-2. Oregon Forest Research Center, Corvallis, OR.
Muenow, R.A. The Ultrasonic Non-destructive Evaluation of Wood Utility Poles, Wood and Concrete Railroad Ties and Trestles, and Other Similar Structures. James Electronics, Inc., Chicago, IL.
Ricard, J.L., 1966. Detection, Identification, and Control of *Poria carbonica* and Other Fungi in Douglas-fir Poles (Ph.D. dissertation). Oregon State University, Corvallis, OR, 140 p.

Roberts, D.M., 1984. An Investigation of Decay Patterns and Strength Losses in Douglas-fir Transmission Poles (master's thesis). SUNY College of Environmental Science and Forestry, Syracuse, NY.

Smith, S.M., Graham, R.D., Morrell, J.J., 1987. Influence of air-seasoning on fungal colonization and strength properties of Douglas-fir pole sections. For. Prod. J. 37 (9), 45–48.

Zabel, R.A., Wang, C.J.K., Anagnost, S.E., 1991. Soft-rot capabilities of major microfungi isolated from Douglas-fir poles in the Northeast. Wood Fiber Sci. 23 (2), 220–237.

CHAPTER SEVENTEEN

Paint mildew and related degradative problems

Contents

Paints and other coatings applied to the surfaces of many building materials provide important protective and decorative benefits. A large multibillion-dollar industry has developed, worldwide, to provide the specialized coatings and application techniques needed for buildings and other materials (bridges, ships, automobiles, machinery, etc.). Coats of paint are particularly useful in most exterior uses of wood and in addition to a decorative function, protect the wood surface from weathering damage (see Chapter Two) and facilitate the shedding of rain water. Coatings used on buildings are often called architectural paints since they are primarily applied to building materials, including wood. They represent nearly one-half of the coverings volumes sold, annually.

In this chapter, we will cover the microflora of paint, the factors affecting mildew development, and general control practices. Emphasis will be on paint coverings over wood in exterior home uses. Brief mention will be made of mold and related biodeterioration problems in home and industry.

Comprehensive coverage of the principles and practices of coatings technology are available in textbooks, coatings journals and governmental

Wood Microbiology.
DOI: https://doi.org/10.1016/B978-0-12-819465-2.00017-6

publications (Weismantel, 1980; Martens, 1981; Lambourne, 1987; Williams, 2010). General reviews of biodeterioration problems in coatings on wood have been assembled (Ross and Hollis, 1976; Brand and Kemp, 1973; Bravery, 1988; Williams, 2010).

Hess's classic manual on Paint Film Defects is still a useful source of information on the nature, cause, and control of paint defects (Hamburg and Morgans, 1979).

Types of paint biodeterioration

Microorganisms (fungi and bacteria) are the principal biotic destructive agents of paints under some use conditions or locations although algae can be important in some environments. Paint disfigurement results primarily from the copious mycelial growth of pigmented fungi and the development of spore clusters on the paint surface. The general term used by industry for these growths is "mildew". Disfigurement or defacement are more preferable to avoid confusion in the minds of many with the mildew diseases of agricultural crop plants. The aesthetic properties of the paint film are often further reduced by accumulation of dirt and airborne detritus on the sticky exposed mycelia or spore clusters. Physical and chemical degradation (cracking, chipping, and exfoliation) of the paint film may also occur from the digestive and growth activities of microorganisms. Degradation of the primer coat by bacteria lodged at the wood-paint adhesion zone when the film remains wet is also believed to be an important contributor to loss of adhesion and peeling. Water-borne paints are degraded primarily by bacteria during the liquid storage phase leading to gas formation, foul odors, and losses in viscosity (Opperman, 1986). Millions of dollars are spent annually by the paint industry for the various chemicals needed to protect liquid paint (stored in cans) and paint films from microbial damage.

Paint types and compositions

A general idea of the chemical ingredients and structure of paint is necessary to understand its degradation by microorganisms. Paint is a

suspension of solid particles in a liquid that dries, hardens, and forms a durable, continuous surface film after application. The principal components of paint can be grouped by function into four general categories.

1. The vehicle or binder provides the matrix or major physical structure to the paint film. Common current vehicles are alkyd, acrylic, polyvinyl acetate, polyurethane, epoxy and other resins or polymers.
2. Pigments (TiO_2) and extenders (calcium carbonate, talc, mica) provide color and bulk the voids in the paint matrix. Semi-colloidal suspensions (emulsions) of the resins in water form the latex type of paints.
3. Various additives such as thickeners (for example: hydroxy-ethyl cellulose), emulsifiers, driers, plasticizers, and biocides confer special properties or application features to the paints.
4. A solvent (organic or water) is the carrier for the paint solids.

The coatings applied to homes for exterior and interior uses can be conveniently grouped into two general types: water-thinned paints (emulsions of resins, polymers, and pigments in water) and solvent-thinned paints (various drying oils, polymers and pigments in organic solvents such as turpentine). Concerns about the risks associated with the release of volatile organic compounds (VOC) during paint drying have led to a marked shift to water-thinned paints.

Both types of paint contain ingredients subject to microbial attack. Vehicles such as linseed oil, soya oil, safflower oil, and dehydrated castor oil in older, solvent-thinned paints can be degraded by fungi or bacteria with esterase enzymes. Resins in water thinned paints such as acrylic and polyvinyl polymers are resistant to microbial attack but thickeners such as ethylated cellulose are degraded by some fungi and bacteria.

The use of water-based latex paints has increased steadily in the past few decades due to their ease of application, rapid drying, and the presence of lower VOC's. In general, latex paints are softer films and more prone to microbiological attack than oil-based paints. The popularity of light colors in the latex paints also enhances the visibility of mildew.

The structure of paint films also affects their susceptibility to microbial damage. Internal voids in the film provide zones for water accumulation and penetration routes for fungi. It is important that all the voids around the binder matrix be occupied by pigment particles. This value is termed the "critical pigment volume concentration". Low porosity reduces the susceptibility of latex paints to fungal invasion. Chalking paints have been formulated so unbound pigment particles on the surface can slowly slough off and cleanse the surface. While biocides are a common component of

coatings, design of paint films specifically to minimize microbial damage remains a neglected research area of potential promise.

Paint microflora

It is surprising to note that until the late 1940, most paint disfigurements were believed to be unavoidable, detritus accumulations.

One of the earliest reports of fungi as the major culprits of paint mildew was made by Goll et al. (1952). They developed techniques to microscopically study the surfaces of many paint samples and found that most disfigurements were actually small patches of pigmented hyphae and clusters of spores. Comprehensive studies of the microflora on paint films soon followed (Klens et al., 1954; Rothwell, 1958; Drescher, 1958; Eveleigh, 1961; Smith, 1977) and were summarized by Brand and Kemp (1973) and Ross and Hollis (1976). While many genera of opportunistic microorganisms were reported, only a few were consistently obtained in large numbers. The major fungi were *Aureobasidium* (*Pullularia) pullulans*, *Cladosporium* spp., *Alternaria* spp., and *Phoma glomerata*. *Aureobasidium pullulans* was the predominant mildew agent and was found in all regions of the United States and subsequently worldwide. Of the other genera, *Phoma glomerata* was found primarily in the Pacific coast region and *Alternaria* sp. in the eastern region of the United States. The principal bacteria were species of *Flavobacterium* and *Pseudomonas* (Ross, 1964). *Pseudomonas aeruginosa* was isolated commonly from spoiled latex paints in liquid phase (storage) and this may account for its prevalence in latex films.

Studies of microorganisms isolated from the water-borne paint formulations and films indicated that changes in paint formulations and manufacturing practices created favorable niches for other fungi and bacteria (Jakubowski et al., 1983). In addition to *Pseudomonas* spp., various yeasts (*Torula* and *Saccharomyces*) were found to be important spoilage agents in stored latex paints. Also, while *A. pullulans* continued to be the predominant fungus associated with mildew on exterior latex paints, *Alternaria* spp. were also judged to be important. These shifts in microflora highlight the fact that a wide array of organisms that can exploit a niche as environmental conditions change.

The characteristics and growth features of *A. pullulans*

Some detail on the nature and capabilities of *A. pullulans* is necessary to explain its apparent global ubiquity on paint surfaces. As the accepted

major causal agent of paint mildew, *A. pullulans* has been studied intensely and a large literature has accumulated on its taxonomy, physiology, and role in paint films (Brand and Kemp, 1973).

Classification: *A. pullulans* is grouped in the Ascomycetes and is characterized by pigmented hyphae (usually black) and asexual reproduction by blastospores (Fig. 17.1). It is dimorphic and may assume yeast or mycelial forms depending on the substrate. As a heterokaryon, *A. pullulans* is extremely variable and isolates can assume a wide range of phenotypic forms. The fungus occurs commonly in the soil, wood, and on plant surfaces (phylloplane). This fungus forms an extracellular carbohydrate (pullulan) that facilitates tenacious adherence to surfaces.

Physiology: *A. pullulans* thrives over broad temperature and pH ranges and melanization of the hyphae is believed to confer resistance to ultraviolet light. It can utilize many simple carbon compounds and displays resistance to some toxicants (Stirling and Morris, 2010a,b). The fungus resists dessication and actinic exposure. It adheres tenaciously to substrates, appears to be well-adapted to surfaces and can utilize detritus and paint extractives whenever favorable moisture levels and temperatures occur.

Growth patterns in latex paint films: *A. pullulans* grows primarily in the hyphal form on paint surfaces. Small hyaline and pigmented hyphae grow on the surface and into any air bubbles or voids in the paint film (Fig. 17.2B and C). Large discreet clusters of blastospores and thick-walled chlamydospores commonly develop (Fig. 17.1). Copious amounts of a brownish hyphal exudate forms on the film surface and embeds the hyphae. Upon drying, the hyphae adhere tenaciously to the paint surface and removal attempts may cause the exfoliation of small paint fragments (Fig. 17.2).

Roles in Natur:. It is perhaps significant to note other known and suspect roles in nature of this interesting organism. *Aureobasidium pullulans* is a well-known causal agent of sapstain in lumber (Kaarik, 1974). Schmidt and French (1976) have shown this fungus to be responsible for discolorations that develop in shingles and shakes. Plant pathologists recognize the organism as a common phylloplane resident of leaf and bud surfaces (Baker and Cook, 1974) and this fungus has also been named as a probable symbiont in lichens (Ahmadjian, 1967).

Factors affecting mildew development

Both waterborne and organic-solvent paints may be severely damaged by microorganisms depending on factors such as geographic location, the

Figure 17.1 Some macroscopic and microscopic features of *Aureobasidium pullulans* showing (A) a colony on malt extract agar, (B) the fungus discoloring a wood sample, and (C) a composite of microscopic images showing hyaline hyphae, conidiogenous cells, and pigmented condia. *All images courtesy of FP Innovations*.

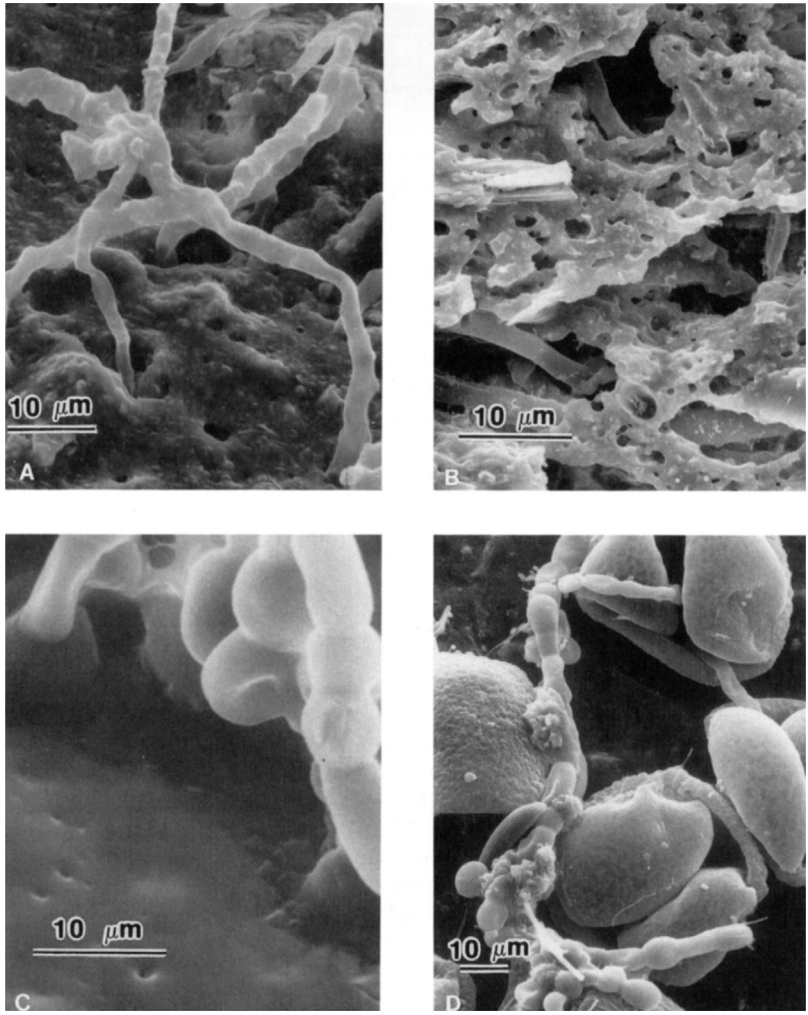


Figure 17.2 Microscopic features of several mildew fungi developing on a latex paint. (A) *A. pullulans* growing on the surface of a paint and penetrating into air holes, (B) *Cladosporium sphaerospermum* hyphae growing in the cavities and voids of a paint film prepared by freeze-drying and fracturing, (C) *A. pullulans* hyphae tenaciously adhering to a paint surface after sample drying, and (D) *A. pullulans* hyphae associated with a pine pollen grain on a paint surface. *Figures 17.2(C) and (D) from Zabel, R.A., Terracina, F., 1980. The role of Aureobasidium pullulans in the disfigurement of latex paint films. Dev. Ind. Microbiol. 21, 179–190, by permission of Society for Industrial Microbiology.*

material covered, the type of exposure, detritus sources, and the paint components.

Geographic location: Paint mildew problems are more severe in coastal and tropical regions where high humidities and temperatures prevail. They can also be a problem in areas receiving high amounts of rainfall.

Type of surface: In general, mildew develops more commonly on wood than on metals or masonry. Water soluble extractives from wood substrates may provide nutrients that diffuse into the paint film and can be a significant moisture and inoculum source. Film integrity and adhesion may also be affected by dimensional changes occurring with fluctuations in wood moisture content. Paint coats on more stable wood species such as coastal redwood and the cedars are less susceptible to mildew development than on less stable woods such as the southern yellow pines, Douglas-fir, and hemlock.

Type of exposure: Mildew is often heaviest on exterior surfaces of homes in locations where shade trees or shrubbery restrict rapid surface drying and on north-facing walls. Paint surfaces protected from direct rainfall accumulate more detritus and mildew. Ceilings over open garages and porches may be subject to severe mildew due to condensation caused by rapid temperature drops in the evening in humid regions. Unsightly mildew may develop on painted surfaces in bathrooms and window units due to condensation, particularly during the colder seasons. Poorly ventilated closets and damp basement rooms also allow for fungal growth during periods of high humidity. Spores and fungal fragments can be an overlooked significant source of allergens in some buildings and the presence of any mold growth in a building often raises concerns about occupant health and safety.

Detritus sources: Walls and ceilings of food-processing plants, paper mills, breweries, and dairies often experience severe mildew problems due to the combination of excessive moisture and detritus accumulations on the surfaces. In severe cases, mold (*Penicillium* spp., *Aspergillus* spp. and *Trichoderma* spp.) may also develop and require specially treated paints and frequent washings to control. In many instances, structures are built so they can be routinely power-washed to remove accumulated detritus.

Detritus accumulating on paint surfaces includes a variety of nutritive sources for mildew fungi such as plant exudates (aphid secretions) and pollen. Small amounts of pollen and simple sugars simulating natural conditions have been shown to stimulate mildew development on latex paints (Zabel and Terracina, 1980).

Nutrient sources: The nutritive status of paint ingredients varies greatly between and within paint types. Spoilage problems during paint storage occur primarily in waterborne paints. Vehicles consisting of natural oils are readily degraded by some paint-inhabiting microorganisms, while most of the synthetic resins are resistant. Some pigments, such as zinc oxide and barium metaborate, reduce mildew growth, while titanium dioxide appears to have no effect. Some paint additives such as ethylated cellulose can be degraded by bacteria during paint storage and may also be a minor nutrient source for some fungi on paint films. Biocides such as 3-iodo-2-propynylbutyl carbamate and substituted isothiazolones are often added to retard microbial attack.

Free moisture on the paint film appears to be the critical environmental requisite for mildew development. However, little basic information exists on the amounts and state of water necessary to initiate mildew growth on paints. Hill and April (1971) have postulated that there was critical moisture level to sustain mildew development in and on paint. It has been suggested that prolonged relative humidity above 75% is necessary for mildew formation. However, even minor decreases in temperature at high humidity would lead to a dew point and free water accumulation on the film. Controlled studies of temperature, relative humidity, and moisture suggest that mildew first begins to appear on latex films over wet wood blocks between 30% and 60% moisture content (Zabel and Terracina, 1980).

Postulated roles of *A. pullulans* in latex paints: Despite the importance of *A. pullulans* as a mildew agent, its nutritional sources remain uncertain. However, laboratory studies indicate that *A. pullulans* cannot sustain growth on a latex film under axenic conditions suggesting the need for exogenous nutrient sources such as pollen, plant exudates or other materials.

Studies on the succession of microorganisms on test paint panels have reported that bacteria and other fungi must precede and modify the nutritive status of the film before the appearance of *A. pullulans*, which then remains as the final dominant or climax species (Winters et al., 1975; Schmitt, 1974). Surface isolation difficulties and whether the microorganism sequences are fortuitous or obligatory raises uncertainties about these claims. Co-metabolism of some paint ingredients with other microorganisms has been proposed (Horvath and Esposito, 1979). Evolving DNA techniques should help to better elucidate the relationships between *A. pullulans* and other paint film inhabitants.

Similarities between *A. pullulans* growth patterns on paints, plastic greenhouse roofs (Durrell and Goldsberry, 1970), and cedar shingles, and its role in nature as a major phylloplane resident suggest it is uniquely adapted to utilize nutrients on these hostile sites (Stirling and Morris, 2010a,b; 2013a,b). In all cases, the fungus must adhere to a surface, withstand desiccation, resist high UV radiation and rely on detritus as the major nutrient source. This fungus is conspicuous and outlasts competitors making it appear as the dominant paint inhabitant. However, many other roles or sequences may occur among the many types of paint and paint-inhabiting microorganisms.

Exterior coatings for horizontal surfaces

A related, but slightly different exposure is horizontally exposed decking. The expansion of outdoor living in the 1970s was accompanied by a rapid increase in the construction of naturally durable or treated wood decks. These decks were subjected to direct, severe UV exposure coupled with regular rainfall that resulted in rapid weathering. A variety of deck coating materials were developed to help protect the wood against this damage. Most were originally oil-based, but changing VOC requirements resulted in a nearly complete shift to water-based systems. While pigmented finishes can be used for this application, most deck owners want to see the natural wood finishes. As a result, there are a variety of surface penetrating finishes containing waxes to serve as water repellents and some form of fungicide coupled with various resins that help retain the coating and limit UV degradation. Despite claims, few deck finishes provide more than 1–2 years of effective protection because of severe UV and weather conditions present (Morrell et al., 2001).

General control practices

The basic principles of mildew control are to minimize moisture and detritus accumulation on paint films and to use paints containing with

mildewcides where mildew hazards are severe. An analysis of the various factors affecting mildew development indicates a range of actions or decisions the homeowner can take to reduce the problem. Periodic washing with disinfectants (sodium hypochlorite) is sometimes helpful. Surface cleaning and disinfection is also necessary when repainting over a previously mildewed coat. Buildings designed to facilitate rapid water shedding and promote good aeration can be useful. Exhaust fans can reduce excessive condensation in bathrooms, windows, and other high humidity zones. Proper installation of insulating materials in ceilings and walls can reduce the moisture levels in walls and placing building membranes on the proper side of a wall to avoid condensation can reduce the risk of moisture accumulation. The selection of paints containing more "non-degradable ingredients" and that have an acceptable film hardness are recommended in all uses where mildew problems can develop.

However, these steps are still often insufficient for mold prevention. The most reliable practice for limiting mildew growth has been the addition of effective biocides (mildewcides) to the paint during formulation. These biocides provide protection to the paint during both storage and use. Two widely used pigments, barium metaborate and zinc oxide are known to be effective inhibitors when used at proper concentrations. Preservatives that were remarkably effective as mildewcides were the phenyl mercury compounds and chlorinated phenols; however, these compounds are no longer used because of concerns about their toxicity in this application.

Environmental restrictions on uses of mercury compounds (phenyl mercury acetate) and environmental concerns about the chlorinated phenols have stimulated a search for effective replacement compounds. A wide range of proprietary compounds are now marketed as mildewcides for various paint types and uses. A few examples of their toxic constituents are 3-iodo-2-prpynyl butyl carbamate, quaternary ammonium compounds, zinc dimethyl dithiocarbamate, isothiazoline compounds, substituted pyridines such as trichlorodicyanopyridine, triazines, copper-8-hydroxyquinolate (oxine copper), tributyltin oxide, tetraethyl-thiuram disulfide, and tetrachloropyridine-4-methyl sulfone.

Ideal mildewcides must be highly effective against mildew fungi, compatible with other paint components, possess limited solubility, produce no color changes, be non-volatile, be safe in handling and use, and be environmentally acceptable. All of these compounds must also be registered as fungicides with the U.S. Environmental Protection Agency.

Mildewcide evaluations

Agar plate toxicity assays are generally used for preliminary screening in the development of potential mildewcides. Promising compounds are then evaluated by chamber test procedures developed by the American Society for Testing Materials ASTM (2016). Painted wood panels are exposed in a closed chamber at 30 °C and 90–100% relative humidity. A bed of loam soil is seeded with selected test fungi and placed below the test panels. Air circulation is provided to insure a constant source of airborne propagules of the appropriate mildew fungi. The effectiveness of a compound is determined by the percent of mildew coverage compared with controls. Weatherometers expose samples to schedules of simulated rainfall and UV exposure prior to or intermittent with the chamber exposures. An accelerated laboratory procedure for effectively growing *A. pullulans* on fresh latex paints on wood axenically was developed to permit the direct testing of mildewcides in the laboratory (Zabel and Horner, 1981).

Potentially effective compounds are tested further in selected homes or extensive panel field tests. Painted wood panels are exposed in severe mildew hazard locations such as Miami, Florida or New Orleans, Louisiana. Vertical exposures of the panels facing north are generally used. The panels are mounted on offset racks to minimize washing of materials from one panel to the other. Ineffective mildewcides can generally be eliminated after a one-year exposure. Compounds that appear promising in the laboratory often prove ineffective in the field tests because they are exposed to a much broader microbial community.

Related degradative problems of microorganisms on surfaces

In addition to the mildews and molds that form on wood during its seasoning and structural uses (discussed in Chapter Fourteen), the growth of microorganisms on machine surfaces during paper manufacture and other industrial and home surfaces can cause serious biodeterioration problems. All these problems can be traced to conditions where adequate moisture and food substances (simple carbon compounds) accumulate. The offending fungi and bacteria involved are often the same opportunist

microorganisms that rapidly exploit any new wood resource. These additional problems are briefly mentioned and referenced here because of their close relationship with lumber molds and paint mildew.

Slime formation during pulp and paper manufacturing: The masses of bacteria and fungi that from on piping and machine surfaces during the pulping and papermaking processes are commonly known as "slime". Fragments of slime that loosen and get into the pulp suspension causes paper breaks, degrading holes or specks on the sheet, odors, production disruptions, and reductions in machine efficiency. Slime is caused by various species of bacteria and/or fungi that develop in recesses, corners, and other zones of low circulation in the system. Leathery and gelatinous slimes are often caused by bacteria. Semi-solid slimes are traceable to mixtures of bacteria and fungi. Stringy slimes are generally caused by fungi.

Slime formation varies considerably with season of the year, the kind of pulp, type of mill system, and mill practices. Slimes are generally most severe in the spring and fall when bacterial counts are highest in the intake water. General slime problems are more severe on groundwood or semi-mechanical than chemical pulps and sulfite than kraft mills. Slime problems also are more severe where large volumes of water are recirculated or where paper finishes include clays and starch. Many slime forming microorganisms are traceable to the incoming mill water and additives. Chlorination of intake water and bleaching agents used during pulping reduce slime formation and can minimize pipe plugging problems with iron bacteria, but chlorine must be used with caution since it can result in the formation of other compounds of concern. Control approaches for slime begin with good housekeeping practices, weekly cleanups and periodic flushing of the mill system with hot-toxicant detergent solutions. In cases where chlorination of intake water and good housekeeping are inadequate, the use of slimicides is necessary. Some current slimicides are the carbamates (disodium ethylenebisdithiocarbamate, sodium dimethyl dithiocarbamate), methylene bis-thiocyanate, 3,5-dimethyl-1,3,5-2H-tetrahydrothiadiazine-2-thione, and 1,2-dibromo-3 nitrilopropionamide. Slimicides must be effective at low retentions in paper products, be essentially non-toxic to higher organisms, and non-hazardous to fish and wildlife. Various companies and consultants listed in the pulp and paper journals generally determine and provide the specialized slimicide treatments necessary for a mill. These services involve a study of the mill system; determination of sampling points; periodic sampling of slurry and paper, cultural determinations of the frequency and type of microorganisms causing the slime; and decisions on the type of slimicide and concentration to use. A comprehensive

review of the history and controls of slime formation in pulp and paper mills was been assembled by Ross and Hollis (1976). Regular water sampling can be used to detect spikes in microbial activity before they become problematic. Alkaline pulping processes and closed mill systems that conserve fiber and energy favor the formation of anaerobic bacteria in white water tanks or storage chests, particularly where circulation is reduced. These problems become especially important when a plant has any temporary shutdown. Additional problems with the development of anaerobes are pipe corrosion, disagreeable odors, and in some cases, the production of explosive hydrogen and methane gases (Robichaud, 1991). Aeration by agitation, ventilation, and use of biocides are the recommended preventative measures.

Molds within the home: Growth of fungi (molds) on surfaces in poorly ventilated areas of homes where moisture accumulates are unsightly and can result in musty odors, allergies, and in some cases, disease. Molds also become problematic in buildings that become wet during construction, that flood or that develop plumbing leaks. Mold fungi can rapidly colonize surfaces. Although they tend to be more aggressive on cellulose products such as drywall, their presence on any surface is likely to elicit consumer concerns. A comprehensive review of the risk of molds on wood products can be found at Robbins and Morrell (2017)

Mold growth on inert materials: Some molds such as species of *Penicillium*, *Aspergillus*, and *Alternaria* grow on glass surfaces when there is a food base in cases of prolonged high humidity and may etch and damage the surface (Subramanian, 1983). This can be a serious problem on optical instruments in tropical zones. The growth of molds on electronic equipment under prolonged high humidity conditions can damage plastic materials and disrupt the circuitry. Favorable conditions for the growth of fungi and bacteria sometimes occur on the surfaces and filters in air-conditioning units. Allergies and some diseases are reported to be associated occasionally with air-conditioning and cooling units (Banaszak et al., 1970; Dondero et al., 1980) and their causes and controls have been reviewed by Ager and Tickner (1983).

Some research considerations

There is a never-ending search for more effective and lasting mildewcides that are non-toxic to mammals. A potentially useful procedure

of incorporating toxicants, chemically or by encapsulation into the binder or vehicle has been reported by Pittman and Lawyer (1982). Encapsulation could reduce leaching losses and deliver the toxicant only when needed at the time of microbial attack. Paint chemists and formulators have demonstrated remarkable skill in designing paints for a wide range of diverse purposes. The use of non-degradative ingredients and moisture control of films remain the basic keys to mildew control. Designing paint films to reduce the size and frequency of inner voids and to minimize free water entry from surfaces could be useful. Film designs might be sought to facilitate passage of water vapor out and repel free water ingress or to minimize surface detritus accumulations. Given the current state of knowledge on the factors affecting mildew development, it is probable that bio-resistant paints will be achieved when this property assumes a higher priority.

Summary

1. Fungi and bacteria are the principal biotic agents responsible for the destruction and disfigurement of paints.
2. Mildew, a major surface disfigurement is caused by the copious mycelial growth of pigmented hyphae on the surface.
3. Paints are suspensions of solid particles in a liquid, that react after application to a surface to form a durable continuous film over the surface. Paints are often grouped as those with the solids and other additives in an organic solvent (solvent-thinned paints) or water (waterborne paints).
4. Waterborne paints are generally more prone to mildew damage than solvent thinned paints.
5. *Aureobasidium pullulans* is the major mildew agent and occurs on paints worldwide in humid regions. Other principal mildew fungi are *Cladosporium* spp., *Alternaria* spp., and *Phoma glomerata*.
6. *A. pullulans* tolerates a wide range of growth conditions, resists dessication and actinic exposure, tenaciously adheres to substrates and appears to be well adapted to utilize detritus and simple carbon compounds that accumulate on paint surfaces when favorable moisture levels and temperatures occur.

7. Mildew development occurs on paints primarily in humid coastal and tropical regions. It develops more commonly on painted wood than metals or masonry. it occurs more commonly on the exterior of homes on shaded zones, north facing walls, or locations where shade trees or shrubbery restrict rapid drying of water on the paint surface.
8. Mildew develops inside the home principally on painted surfaces in bathrooms and window units due to condensation. They are also problematic in food-processing plants, paper mills, dairies and other industries where high humidities and air-borne detritus occur. Severe mildew and surface mold problems may occur on ceilings and walls and require specially treated paints and frequent washings to control.
9. Free moisture on the paint film appears to be the critical growth requisite for mildew development.
10. General control practices are to minimize moisture and detritus accumulation on paint films and to use paints treated with mildewcides where mildew hazards are severe. A wide range of proprietary compounds are available as mildewcides for various paint types and uses.
11. The design of paint films specifically to minimize microbial damage represents an area of potential promise and it is probable that bioresistant paints can be achieved when this property assumes a higher priority.
12. The growth of molds and bacteria on surfaces in pulp and paper mills results in the formation of slimes that can degrade the paper, produce odors, and reduce machine efficiencies. Related mold accumulations in air-conditioning and cooling units can cause allergies and some diseases.

References

Ager, B.P., Tickner, J.A., 1983. The control of microbiological hazards associated with air-conditioning and ventilation systems. Ann. Occup. Hyg. 27 (4), 341–358.

Ahmadjian, V., 1967. The Lichen Symbiosis. Blaisdell Publ. Co., Waltham, 152 pp.

American Society for Testing Materials (ASTM), 2016. Standard D3273. Standard test method for resistance to the growth of mold on the surface of interior coatings in an environmental chamber. ASTM Annual Book of Standards. ASTM International, West Conshocken, PA.

Baker, K.F., Cook, R.J., 1974. Biological Control of Plant Pathogens. WH Freeman, San Francisco, CA.

Banaszak, E.G., Thiede, W.H., Fink, J.N., 1970. Hypersensitivity pneumonitis due to contamination of an air conditioner. N. Engl. J. Med. 283 (6), 271–276.

Brand, B.G., Kemp, H.T., 1973. Mildew defacement of organic coatings. A review of literature describing the relationship between *Aureobasidium pullulans* and paint films. Fed. Soc. Paint. Technol. 132 pp.

Bravery, A.F., 1988. Biodeterioration of paint-a-state-of-the-art comment. In: Houghton, D.R., Smith, R.N., Eggins, H.O.W. (Eds.), Biodeterioration 7. Elsevier Applied Science, New York, NY.
Dondero, T.J., Rendtroff, R.C., Mallison, G.F., Weeks, R.M., Levy, J.S., Wong, E.W., et al., 1980. An outbreak of Legionnaire's disease associated with a contaminated air-conditioning cooling tower. N. Engl. J. Med. 30 (7), 365–370.
Drescher, R.F., 1958. Microbiology of paint films. IV. Am. Paint. J. March, 1–12.
Durrell, L.W., Goldsberry, K.L., 1970. Growth of *Aureobasidium pullulans* on plastic greenhouse roofing. Mycopathol. Mycol. Appl. 42, 193–196.
Eveleigh, D.E., 1961. The disfiguration of painted surfaces, with special reference to *Phoma violacea*. Ann. Appl. Biol. 49, 403–411.
Goll, M., Snyder, H.D., Birnbaum, H.A., 1952. New developments in the diagnosis of paint mildew. Off. Dig. 24, 149–152.
Hamburg, H.R., Morgans, W.H. (Eds.), 1979. Hess's Paint Film Defects. 3rd ed. Chapman and Hall, London.
Hill, D.O., April, G.C., April 1971. Influence of moisture transport on fungal growth. J. Paint Technol. 43 (560), 81–88.
Horvath, R.F., Esposito, M.M., 1979. Co-metabolism of paint by *Aureobasidium pullulans* BH-I-ATCC 34621. Curr. Microbiol. 2, 169–170.
Jakubowski, J.A., Gyuris, J., Simpson, S.L., 1983. Microbiology of modern coatings systems. J. Coat. Technol. 55 (705), 49–53.
Kaarik, A., 1974. Sapwood Staining Fungi. The International Research Group on Wood Preservation. Document No. IRG/WP/125, Stockholm, Sweden, 78 pp.
Klens, P.F., Leitner, G., Snyder, H.D., 1954. A field method for detecting mildew on paint. Am. Paint. J. 38 (39), 76–82.
Lambourne, R. (Ed.), 1987. Paint and Surface Coatings: Theory and Practice. Ellis Horwood Ltd, Chichester, U.K.
Martens, C.R., 1981. Waterborne Coatings. Van Nostrand Reinhold Co, New York, p. 316.
Morrell, J.J., Schneider, P.F., Williams, R.S., 2001. Protecting wood decks from biodegradation and weathering: evaluation of deck finish systems. For. Prod. J. 51 (11/12), 27–32.
Opperman, R.A., 1986. The anaerobic biodeterioration of paint. Biodeterioration 6, Proc. 6th Int. Biodet. Symp. CAB, International Mycology Institute, pp. 610–616.
Pittman, C.V., Lawyer, K.R., 1982. Preliminary evaluations of the biological activity of polymers with chemically bound biocides. J. Coating. Technol. 54 (690), 41–46.
Robbins, C., Morrell, J.J., 2017. Mold, Housing and Wood. Western Wood Products Association, Portland, OR.
Robichaud, W.T., 1991. Controlling anaerobic bacteria to improve product quality and mill safety. Tappi 74 (2), 149–153.
Ross, R.T., 1964. Microbiological deterioration of paint and painted surfaces. Dev. Ind. Microbiol. 6 (14), 149–163.
Ross, R.T., Hollis, C.G., 1976. Microbiological deterioration of pulp wood, paper, and paint. In: Miller, B.M., Litsky, W. (Eds.), Industrial Microbiology. McGraw-Hill Book Co, New York, pp. 309–354.
Rothwell, F.M., 1958. Microbiology of paint films. II. Isolation and identification of microflora on exterior oil paints. Off. Dig. 30, 368–379.
Schmidt, E.L., French, D.W., 1976. *Aureobasidium pullulans* on wood shingles. For. Prod. J. 26 (7), 34–37.
Schmitt, J.A., 1974. The microecology of mold growth. J. Paint Technol. 46, 59–64.
Smith, R.A., 1977. Fungi surviving in a latex paint film after surface disinfection. Dev. Ind. Microbiol. 18, 565–569.

Stirling, R., Morris, P.I., 2010a. Reducing depletion of western redcedar extractives from wood in service. In: Harrington, A. (Ed.), A Tale of Two Cedars. USDA Forest Service, Pacific Northwest Research Station Report PNW-GTR-828, Portland, Oregon.
Stirling, R., Morris, P.I., 2010b. Degradation of Carbon Based Preservatives by Black Stain Fungi. Document No. IRG/WP/10-30533. International Research Group on Wood Protection, Stockholm, Sweden.
Stirling, R., Morris, P.I., 2013a. Improved coating performance on wood treated with carbon-based preservatives and an ultraviolet/visible light protective coating. For. Prod. J. 63 (3/4), 95–100.
Stirling, R., Morris, P.I., 2013b. Perfromnce of Coatings on Wood Treated with Carbon-based Preservatives. Document No. IRG/WP/13-40638. International Research Group on Wood Protection, Stockholm, Sweden.
Subramanian, C.V., 1983. Hyphomycetes—Taxonomy and Biology. Academic Press, New York, p. 502.
Weismantel, G.D. (Ed.), 1980. Paint Handbook. McGraw-Hill Book Co, New York, pp.
Williams, R.S., 2010. Chapter 16: Finishing of wood. USDA Wood Handbook: Wood and an Engineering Material. USDA Forest Products Laboratory General Technical Report GTR-190, Madison, Wisconsin.
Winters, H., Isquith, IR., Goll, M., 1975. A study of the ecological succession in biodeterioration of vinyl acrylic paint film. Dev. Ind. Microbiol. 17, 167–171.
Zabel, R.A., Terracina, F., 1980. The role of *Aureobasidium pullulans* in the disfigurement of latex paint films. Dev. Ind. Microbiol. 21, 179–190.
Zabel, R.A., Horner, W.E., 1981. An accelerated laboratory procedure for growing *Aureobasidium pullulans* on fresh latex films. J. Coat. Technol. 53 (675), 33–37.

Further reading

American Wood Protection Association, 2017. Standard E24. Laboratory method for evaluating the mold resistance of wood and wood-based materials: mold chamber test. AWPA Book of Standards. AWPA, Birmingham, Alabama, pp. 463–467.
Ionu, M., 1988. The study of fungal contamination in the field of electronics, Biodeterioration, seventh ed. Elsevier Applied Science.
Ludwig, L.E., 1974. Formulation of mildew resistant coatings. J. Paint Technol. 46 (594), 31–39.
Williams, R.S., Feist, W.C., 1993. Durability of paint or solid-color stain applied to pre-weathered wood. Forest Prod. J. 43 (1), 8–14.

CHAPTER EIGHTEEN

Natural decay resistance (wood durability)

Contents

A first principle of decay prevention is the use of naturally durable or preservative-treated woods in those high hazardous decay conditions where the wood cannot be kept dry by structural design or handling practices. This chapter discusses the types and features of durable woods and the next chapter considers similar aspects of preservative-treated woods.

The benefits of using naturally durable woods have long been known (Graham, 1973). Phoenician boat builders routinely employed naturally durable Cedars of Lebanon or oaks and their overuse contributed to the decline of natural forests along the Mediterranean. These same builders clearly understood the differences between sapwood and heartwood, noting that sapwood was more susceptible to ravages of decay and marine borer attack. It is interesting to note that these builders utilized wood in a discriminatory manner similar to homeowners in the 20th century, but had no understanding of the nature of decay.

Although supplies of naturally durable wood are limited and could not meet the demand for all wood used in hazardous environments, interest in these species continues for many reasons. Decay resistance is an important tree property that has been neglected to date in most tree improvement programs that emphasize other attributes such as branch form, rate of growth and wood cell characteristics. Knowledge of the naturally toxic compounds in the durable woods may lead to the development of more

Wood Microbiology.
DOI: https://doi.org/10.1016/B978-0-12-819465-2.00018-8

effective wood preservatives and other wood protective chemicals. The use of naturally durable woods has also become an important option where there are concerns about the environmental safety of wood preservatives. This is especially important in organic farms, where the use of preservative treated wood is not allowed

The emphasis in this chapter will be on factors affecting decay. The term durability is used commonly in the lumber trade to designate decay resistance. Actually, wood durability is a broader term and reflects the resistance of wood to other deteriorating factors such as insects, marine borers, and weathering, as well as decay fungi. In this chapter we will limit our discussion to resistance to microbial, insect and marine borer attack.

An extensive literature has accumulated over the years on the many facets of decay resistance in woods. Many reports are of a practical type reporting service tests of various local species for fence post and local construction use. Brief reviews of decay resistance in commercial wood species are presented in both the U.S.D.A. (2010) and Panshin and deZeeuw, 1970). Comprehensive reviews of the topic, placing some emphasis on the nature and toxicity of the wood extractives have been assembled (Scheffer and Cowling, 1966; Hillis, 1987; Taylor et al., 2002).

Variations in decay resistance

Wood consists of several natural polymers and a wide range of cell-wall extractives that are primarily localized in the heartwood. Heartwood durability, as with any natural product, is characterized by wide variability both between species as well as between individual trees of the same species (Scheffer and Cowling, 1966). This variation reflects both the genetic potential of a tree and the environmental conditions under which the tree is grown. Heartwood durability of a species may vary dramatically. For example, there are differences exhibited between highly durable old-growth redwood and moderately durable second-growth timber of the same species (Clark and Scheffer, 1983; Ajuong et al., 2014). There is increasing evidence that the heartwood of second growth trees of some species is less durable than heartwood from older growth trees, although this is not always the case (Freitag and Morrell, 2001).

Species variations

Great variations in decay resistance occur among species ranging from a few months of service for some susceptible species to 40–50 years of service or more for a few highly durable species such as black locust, osage orange, or western red cedar in high decay hazard uses. The decay resistance ratings of the heartwoods of many commercial domestic woods based on extensive service records, post-farm experiments, and laboratory evaluations have been summarized (Scheffer and Cowling,1966; Scheffer and Morrell, 1998) (Table 18.1). The decay resistance rankings of many exotic species are also available (Scheffer and Cowling, 1966; Clark, 1969). Similar ratings are available in other countries. For example, Australian Standard AS5604 provides ratings for termite, lyctid beetle, decay and marine borer resistance for 250 species (Standards Australia, 2005), while the European Union has created standards for evaluating natural durability (European Union, 2016).

Table 18.1 Relative decay resistance of some commercial wood species commonly used in North America.[a,b]

Resistant and very resistant	Moderately resistant	Slightly or non-resistant
Baldcypress (old growth)	Baldcypress (new growth)	Alder
Catalpa	Douglas-fir	Ashes
Cedars	Honeylocust[d]	Aspens
Black Cherry	Western larch	Basswood
Arizona Cypress	Swamp Chestnut Oak	Beech
Juniper	Eastern white pine	Birches
Black locust[c]	Longleaf pine	Cottonwood
Red Mulberry[c]	Slash pine	Elms
Bur Oak	Tamarack	Hackberry
Chestnut Oak		Hemlock
Oregon White Oak		Hickories
Post Oak		Maples
White Oak		Red or Black Oak[d]
Osage-Orange[c]		Spruces
Redwood		Sweetgum[d]
Sassafrass		Sycamore
Black Walnut		Willows
Pacific Yew[c]		Yellow-poplar
Western Redcedar		

[a]Source: Scheffer and Cowling (1966).
[b]Common names are from Little (1979) which provides the scientific names.
[c]These woods have exceptionally high decay resistance.
[d]These species have shown a higher decay resistance than most of the other woods in their respective categories.

Stem position variations

The sapwood of almost all wood species is highly susceptible to decay regardless of durability status of the heartwood, but there are some exceptions. Sapwood in the transition zone between recently formed heartwood and innermost sapwood tends to be more decay resistant than recently formed sapwood in white oak. Sapwood in the vicinity of wounds where prior injuries have been walled off is more decay resistant than surrounding sapwood (Shigo, 1965). The inner stem tissues are somewhat more resistant to decay than newly formed sapwood in species where colored heartwood is not formed (spruces, fir, etc.). Samples of durable species such as northern white cedar, white oak, or black locust containing large proportions of sapwood are often sold in roundwood form for posts. In these cases, durable wood is limited to the heartwood portion of the product. A small cedar post consisting largely of sapwood will offer no more protection than a post of a decay susceptible species. Some sapwoods with large quantities of mineral deposits such as silica can be more resistant to insect or marine borer attack by virtue of their hardness.

In general, decay resistance increases from the cambium to the sapwood/heartwood interface (Fig. 18.1). In many species, durability is highest near the sapwood/heartwood interface and declines towards the pith (Zabel, 1948; Scheffer and Hopp, 1949; Scheffer et al., 1949; Gardner and Barton 1958; Gardner, 1960; Behr, 1974; Hillis, 1985). This decline is believed to reflect either biological detoxification, natural oxidation of heartwood extractives, or continued polymerization of extractives to produce less toxic compounds (Anderson et al., 1963). Microbial activity also may reduce heartwood durability with age (Jin et al., 1988). Decay resistance varies with stem height, with the most durable wood occurring near the base of the tree. Variations in the decay resistance of western redcedar appear to be well-correlated with distribution of the wood extractive thujaplicin (Nault, 1988). Durability reflects both the natural genetic potential of a tree and the environmental stresses to which the tree is subjected.

Factors affecting durability

The wide variation in durability among and within species reflects a number of factors. As stated, extractive content plays the chief role in

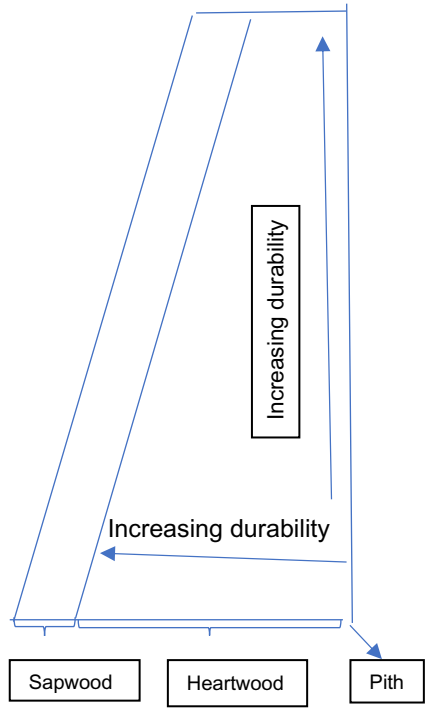


Figure 18.1 Effect of position in the tree on relative durability of the heartwood in decay resistant timber species.

durability and will be addressed at length later in this chapter. A number of other factors also influence durability.

Lignification: As discussed in Chapter Six, lignification was the important step in higher plant evolution that provided stiffness to stem tissues facilitating stem aerial development and protection against destruction by microorganisms. Lignin type and amount can have a significant influence on the rate and types of decay found in wood. Lignin content can vary widely between species, within individual portions of the same tree and within individual cell wall layers, rendering some portions of the wood, such as the primary cell wall and middle lamella, more resistant to microbial attack. Hardwood lignins are composed of both syringal and guaiacyl phenyl propane units, while coniferous lignin contains only guaiacyl residues. Hardwood lignins also differ from coniferous lignins in the types of linkages between phenyl propane units. Lignin plays an important role in decay resistance as evidenced by the increased susceptibility of partially delignified softwood timbers to soft-rot attack; (Morrell and Zabel, 1988; Zainal, 1976).

Lignin type, content and pattern of deposition appear to play critical roles in inception of soft-rot attack and relatively minor changes in lignin content can produce dramatic decreases in decay resistance. These effects suggest that the pattern of lignin deposition in the wood cell wall may be as critical as lignin type in determining natural resistance to some decay fungi.

Wood may be naturally durable for a number of reasons including the environment and organisms to which it is subjected. For example, many species exhibit extreme hardness or contain large quantities of silica or calcium carbonate (Taniguchi et al., 1986). These species are often resistant to insect or marine borer attack, which require that the animal chew into the hard wood (Bultman et al., 1977; Southwell and Bultman, 1971). Crystals may also alter moisture holding capacity of the wood, making it more difficult to wet, thereby limiting the microorganisms that can colonize the wood. Hardness creates similar difficulties for sawmills processing these species. While hardness represents one means of protection against insects or marine borers, the most common mechanisms of natural durability involve the production of toxic chemicals during heartwood formation.

Growth characteristics: In addition to chemical differences, some silvicultural factors may also influence durability. Many users have suggested that wood cut as the tree begins a growth flush and produces leaves or needles may contain higher levels of readily accessible carbohydrates, rendering the wood more susceptible to microbial attack. Wood harvested during dormant periods would be expected to contain lower levels of these compounds and should, therefore be less susceptible to attack. No evidence of this effect has been noted, although cutting during cooler dormant periods undoubtedly reduces the risk of staining and decay because conditions are less suitable for their growth. Similarly, limited studies of southern pine suggest that fertilization using high nitrogen content fertilizers can adversely influence resistance to disease and may affect durability. In theory, fertilization should increase tree growth, producing a wider band of decay susceptible sapwood. Furthermore, higher nitrogen levels are often correlated with increased susceptibility to fungal attack (Merrill and Cowling, 1965). Conversely, rapid growth might increase the levels of carbohydrates available for later synthesis of toxic extractives, although this effect has never been noted. It is generally difficult to relate carbohydrate levels in sapwood to subsequent heartwood durability (Taylor et al., 2007, 2008)

One important factor related to natural durability is the decay resistance of second-growth timber. Until recently, naturally durable species were primarily cut from old-growth forests; however, these stocks are nearly depleted in many regions or are protecting as cultural resources. As a result, we are beginning to harvest trees of naturally durable species from second-growth stands. Preliminary results suggest that the wood from second-growth tree of some species lacks the natural durability found in the old-growth trees. This effect has been noted with southern pines, bald cypress, Port Orford cedar, and, most recently, coastal redwood (Ajuong et al., 2014; Clark and Scheffer, 1983). Interestingly, this effect does not appear to occur with all species. For example, there is no significant difference in decay resistance of old-growth and second-growth Douglas-fir heartwood, a moderately decay-resistant species (Scheffer and Englerth, 1952) or western redcedar (Freitag and Morrell, 2001), a highly decay resistant species.

As we move to a managed, second-growth forest, we may find it necessary to alter our reliance on naturally durable woods, or identify methods for encouraging heartwood development in these woods at earlier points within a rotation age. Increased attention should be also paid to decay resistance as an important trait to be selected for and enhanced in tree breeding programs.

Miscellaneous factors: While many growth characteristics may affect durability, one factor that is poorly correlated with decay resistance is wood density. Many woods exhibit high density and are naturally durable, but other dense woods are rapidly degraded. As an example, white oak is dense and durable, while woods with similar density such as beech or maple, exhibit no natural resistance to decay fungi. Conversely, several light woods including western red cedar and coastal redwood are among our more durable species.

Once a tree is harvested and processed, handling procedures can also affect durability. For example, heat treatments of wet wood can either volatilize or denature wood extractives, decreasing natural durability (Scheffer and Eslyn, 1961). Exposure to gamma radiation can also adversely affect natural durability (Scheffer, 1963). Exposure to excessive wetting can lead to leaching of water-soluble extractives, also reducing natural durability (Johnson and Cserjesi, 1980). Although this is generally a slow process, exposures under certain conditions, such as excessively high acidity or under nutrient regimes that encourage the growth of detoxifying microbes, can also dramatically alter durability.

Decay resistance and heartwood formation

Natural durability is related primarily to the presence of toxic extractives which form and are deposited in the heartwood as the sapwood dies. As noted, sapwood has little or no natural durability.

As normal sapwood ages and is buried further into the tree, the parenchyma cells gradually die. These cells contain higher levels of extractives that form as carbohydrates in the parenchyma are transported from living cells to cells near the heartwood-sapwood transition zone where they are converted to a variety of phenolic compounds. The movement of carbon in this process remains poorly understood (Taylor et al., 2007) as does the nature of conversion to heartwood. Several researchers have suggested that toxic metabolites are transported to the sapwood/heartwood boundary and their accumulation to lethal levels induces cell death and heartwood formation. Alternatively, ethylene production has been shown to stimulate heartwood formation in cell tissue cultures. Further improvements of *in vitro* techniques for cell tissue culture would permit more detailed studies of heartwood formation.

The phenolic extractives produced as the parenchyma senesce diffuse into the surrounding tracheids, fibers, or vessels where they are absorbed into the wood cell walls. Some species produce an array of compounds toxic to fungi, insects, or marine borers, while phenolic extractive production is extremely limited or absent in others. These species are often considered to produce no heartwood, although large portions of the cells are no longer living. These species exhibit no noticeable heartwood durability (Table 18.1). Heartwoods are generally colored, but similar coloration can be produced by factors such as mineral stain or bacterial attack.

In this Chapter we will limit the discussion of heartwood extractives that contribute to durability to representative types of compounds in four major groups: polyphenols, terpenoids, tropolones, and tannins (Table 18.2). Heartwood extractives are covered in greater detail elsewhere (Scheffer and Cowling,1966; Hillis,1987; Taylor et al., 2002).

Polyphenols: Polyphenols include the stilbenes and flavonoids and are, by far, the most common heartwood extractives, occurring in the heartwood of nearly all species. Stilbenes are synthesized via the shikimic acid pathway (Kindl, 1985) and are common in the heartwood of *Pinus*, *Eucalyptus* and *Maclura* species (Hart, 1981). One stilbene, pinosylvin, and its monomethyl ether are the principal protective agents in pine

Table 18.2 Examples of heartwood extractives that inhibit fungal attack.[a]

Class of compound	Chemical name
Terpenoids	Carvacrol
	p-Methoxycarvacrol
	p-Methoxythymol
	Thymoquinone
	Sugiol
	Totarol
	Ferruginol
	Chamic acid, Chaminic acid
	1-Citronellic acid
Flavonoids	Quercetin
	Robinetin
	Taxifolin
	Dihydrobinetin Homoferreirin
	Ougenin
Stilbenes	Pinoslyvin monomethylether
	Pinosylvin dimethyl ether
	3,5,4′-Trihydroxystilbene
	2,3,4,5′-Tetrahydroxystilbene
	3,4,3′,5′-Pentahydroxystilbene
	4-Hydroxystilbene
Tropolones	Pterostilbene
	α, β, or y-thujaplicin
	α or β-thujaplicinol
	Pygemaein
	β-Dolabrin
	Nootkatin

[a]As summarized by Scheffer and Cowling (1966).

heartwoods. Isolated stilbenes are toxic to bacteria, fungi and insects, although their ability to protect wood from these agents varies (Hart, 1981; Hart and Hillis, 1974; Schultz et al, 1990). Flavonoids include many important phytoalexins (compounds produced by plants in response to microbial attack) (Grisebach, 1985) and heartwood extractives such as quercitin and taxifolin that are found in Douglas-fir (Kennedy, 1956).

Terpenoids: Terpenoids are derived from the condensation of C-5 isoprenoid units and can vary from relatively volatile isoprene monomers to the polymer, rubber (Croteau and Johnson, 1985). Monoterpenes such as pinene consist of two isoprene units. Terpenoids have created the large naval stores industry that still plays an important role in the forest industry. Turpentine is a mixture of terpenes and tall oil, an important by-product

of the Kraft pulping process consisting of mixtures of diterpenes and fatty acids. Terpenoids are synthesized by a variety of plant tissues, including the resin ducts in conifers. Highly resinous wood exhibits some natural durability. There is evidence that breeding for density can inadvertently result in woods with higher resin contents that then interfere with drying and preservative treatment.

Tropolones: Terpenoid compounds are important precursors for more toxic extractives, such as the tropolones. Of the heartwood extractives, the chemicals that have received the most attention for their decay resistance are the tropolones. These seven membered rings are specific to members of the Cupressaceae and include several isomers of thujaplicin. Tropolones are believed to derive from terpenoids that are in turn synthesized from acetyl CoA. The biochemistry of tropolones has not been studied in great detail, but monoterpenes such as carane and thujane are believed to give rise to two seven membered rings, thujic acid and B-thujaplicin, respectively.

Tannins: Tannins are important components of the wood and bark of many trees. Tannins from vegetables were originally used to cure animal skins because of their ability to precipitate and stabilize proteins in the skin. Tannins are proanthocyanidins that are highly complex polymers (Hillis, 1985) (Fig. 18.2). These compounds have been extensively studied in the tanning industry and have been explored as replacements for oil-derived components of adhesives. Tannins, because of their ability to precipitate proteins, have also been evaluated as potential wood preservatives; however, their water solubility and relatively low toxicity have limited application (Laks, 1987). The durability of oaks is believed to be associated with their high tannin content (Zabel, 1948). These compounds arise along synthetic pathways that are similar to those for lignin and stilbenes, but the systems are poorly understood.

Evaluating natural durability

In general, natural durability has been evaluated by exposing wood samples to the decay agents for various periods of time and rating the resultant degree of degradation. These tests are sometimes supplemented by extracting the wood and evaluating the toxicity of various fractions

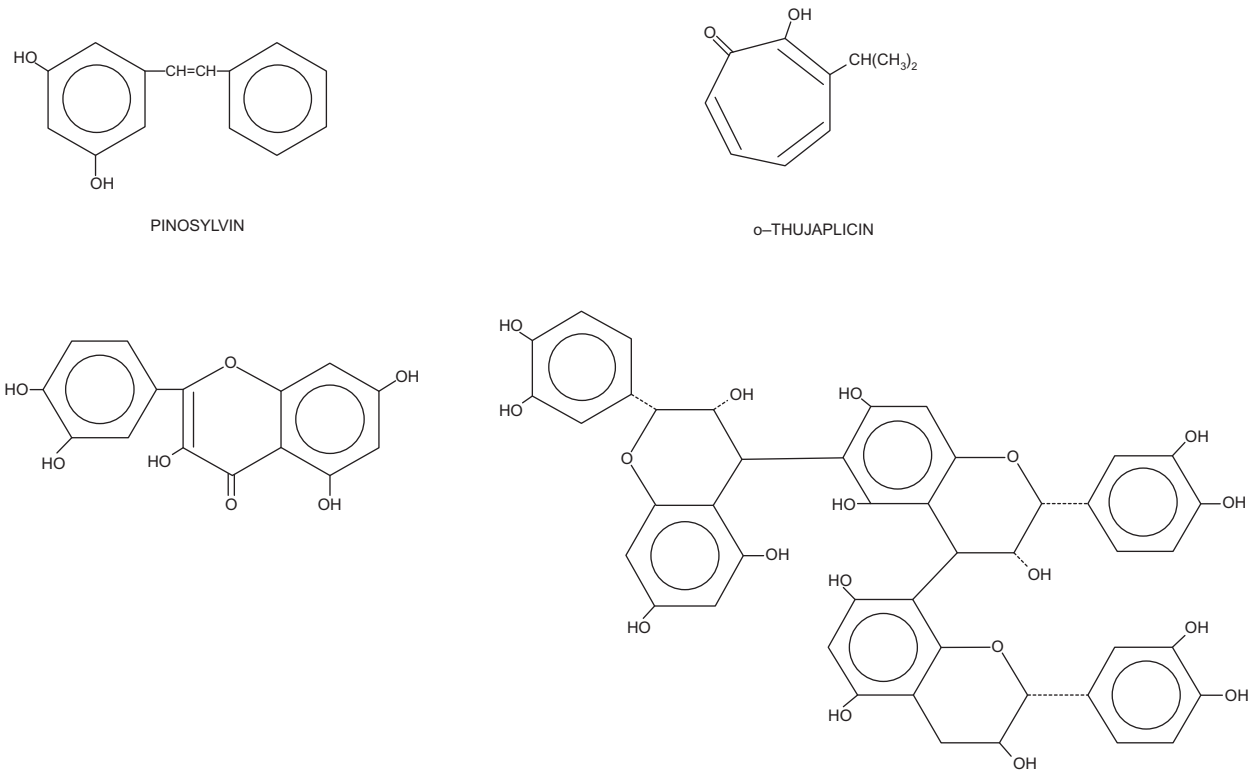


Figure 18.2 Examples of the structures for pinosylvin, α-thujaplicin, taxifolin and a proposed condensed tannin.

against specific organisms. Field trials (post farms) of natural durability were prevalent beginning in the 1920's, as scientists began to evaluate the properties of many local wood species as alternatives to chestnut and the cedars (Humphrey, 1916; White, 1922). These tests reflected a desire to identify woods with properties similar to existing naturally durable species. Hawley et al. (1924) first established the toxic nature of heartwood extractives and posed them as the reason for decay resistance.

Laboratory assays to evaluate natural durability began in the 1940s in an attempt to further explain the nature of durability and identify the toxic compounds (Anderson et al., 1963; Scheffer, 1957; Southam and Erlich, 1943; Kennedy, 1956; Zabel, 1948; Scheffer et al., 1949). In most cases, warm water, ethanol or other solvents were used to remove extractives from the wood. These extractives were then tested for activity against a variety of decay and non-decay fungi. In addition, these extracts were often tested for activity against common wood destroying insects (McDaniel, 1989). Most tests were performed in petri dishes or decay chambers using nutrient agar. While such tests provided a relative guide to chemical toxicity, they could not evaluate more subtle effects such as variations in deposition patterns of extractives in the wood or interactions between different extractives which must also play roles in natural wood durability. One of the best examples of the differences between laboratory and field trials can be found with coastal redwood (*Sequoia sempervirens*) which has a naturally durable heartwood. The extractives of this wood, however, are not highly toxic. Attempts to extract these materials from redwood and impregnate them into less durable timbers such as pine have met with little success. The results illustrate that durability is a function of many factors, likely including how the compounds are distributed in the wood cell walls.

A considerable amount of laboratory research has been performed to identify chemicals that could be synthesized as "natural preservatives" under the premise that such chemicals would be inherently safer and more effective than other biocides. In fact, many of the chemicals responsible for natural wood durability are as toxic or more toxic than existing wood preservatives. The major advantage of employing naturally durable woods over artificial wood preservation would be the elimination of the need for treating facilities to deliver chemical into the wood. Natural durability, however, can never completely replace the need for wood pressure treated with chemicals, since some hazards such as marine exposures are too severe for adequate performance of all but a few naturally

durable wood species. In many cases, naturally durable woods perform best out of direct soil contact.

Improved chemical assay methods using such techniques as radioisotope labeling, C-13 NMR spectroscopy, and ion magnetic spectroscopy have enhanced the study of natural wood durability, but methods for studying in situ deposition of toxic extractives are still lacking. The use of Near Infrared (NIR) or Fourier Transform Infrared (FTIR) Spectroscopy promise to further improve our understanding of extractives distribution, but they still require considerable interpretation. The use of tissue culture techniques may further our knowledge of the synthesis of heartwood extractives, but it will be difficult to study subsequent deposition processes using these methods. Developing an improved understanding of the nature and distribution of extractives related to durability could be especially useful for identifying new approaches to depositing chemicals in wood to protect against fungal and insect attack. This approach would be particularly intriguing because of evidence that the decay resisting extractives in some very durable woods are distributed in the wood in a manner more resistant to leaching than artificially applied fungicides.

Increasing concerns about the use of artificial wood preservatives may encourage increasing dependence upon natural durability for wood used in some locations such as water reservoirs, even under high decay hazards. They are also increasingly used in organic farm operations where preservative treated wood is explicitly banned. This increased demand comes at a time when supplies of durable species are declining and when concerns are being raised about decreased durability of second growth timber. These trends suggest that renewed efforts must be made to identify methods for improving the genetic capabilities to produce durable heartwood and to develop silvicultural practices that favor maximum production of this wood in the shortest period of time.

Summary

1. A first principal of decay prevention is the selection of naturally decay resistant or preservative-treated wood for uses where decay hazardous conditions cannot be avoided.

2. Decay resistance varies widely among species. Some species such as cypress and the cedars are very durable while others, such as beech and maple, are susceptible to decay.
3. Decay resistance in durable woods also varies with stem position. The sapwood of all wood species is generally susceptible to decay. Decay resistance is highest near the sapwood-heartwood interface and decreases toward the pith for many durable species. Also, durability generally decreases with stem height and the most durable wood occurs at the base of the trunk.
4. Durability also varies within a species and some genotypes are much more durable than others. Old-growth timber in durable species such as cypress and redwood is more decay resistant than second-growth timber.
5. The nature and amount of the toxic extractives in the heartwood appears to be the major factor affecting decay resistance. The primary toxic heartwood extractives are the polyphenols, terpenoids, tropolones, and tannins.
6. The use of durable woods as replacements for preservative-treated materials may increase in some environmentally sensitive areas. This suggests increased attention should be paid to the growing of decay resistant species in timber management.
7. An increased understanding of the mechanisms of natural decay resistance may lead to the development of improved treatment methods and lead to effective replacement wood preservatives.

References

Ajuong, E., Freitag, C., Morrell, J.J., 2014. Decay resistance and extractive content of second growth Port Orford cedar (*Chamaecyparis lawsoniana*) wood. Wood Fiber Sci. 46 (4), 502–509.

Anderson, A.B., Scheffer, T.C., Duncan, C.G., 1963. The chemistry of decay resistance and its decrease with heartwood aging in Incense Cedar (*Libocedrus decurrens* Torrey). Holzforschung 17 (1), 1–5.

Behr, E., 1974. Decay Resistance of Northern White Cedar from a Wet Site in Lower Michigan. Res. Rep. 229. Michigan State University Agric. Expt. Stat. East Lansing, Michigan.

Bultman, J.D., Beal, R.H., Ampong, F.F.K., 1977. Natural resistance of some tropical African woods to *Coptotermes formosanus* Shiraki. For. Prod. J. 29 (6), 45–51.

Clark, J.W., 1969. Natural Decay Resistance of Fifteen Exotic Woods Imported for Exterior Use. U.S.D.A. Forest Products Laboratory Research Paper FPL 103, Madison, WI, 5 pp.

Clark, J.W., Scheffer, T.C., 1983. Natural decay resistance of the heartwood of coast redwood *Sequoia sempervirens* (D. Don) Engl. For. Prod. J. 33 (5), 15–20.

Croteau, R., Johnson, M.A., 1985. Biosynthesis of terpenoid wood extractives. In: Higuchi, T. (Ed.), Biosynthesis and Biodegradation of Wood Components. Academic Press, Inc, New York, NY.
European Union, 2016. Standard EN-350. Durability of Wood and Wood-based Products: Testing and Classification of the Durability to Biological Agents of Wood and Wood-based Materials. European Union, Brussels, Belgium.
Freitag, C.M., Morrell, J.J., 2001. Durability of a changing western redcedar resource. Wood Fiber Sci. 33, 69–75.
Gardner, J.A.F., 1960. The distribution of dehydroquercitin in Douglas-fir and western larch. For. Prod. J. 10, 171–173.
Gardner, J.A.F., Barton, G.M., 1958. The extraneous components of western red cedar. For. Prod. J. 8, 189–192.
Graham, R.D., 1973. History of wood preservation. In: Nicholas, D.O. (Ed.), Wood Deterioration and its Prevention by Preservative Treatments. Syracuse University Press, Syracuse, NY.
Grisebach, H., 1985. Biosynthesis of flavonoids. Chapter 12. In: Higuchi, T. (Ed.), Biosynthesis and Biodegradation of Wood Components. Academic Press, Inc, New York.
Hart, J.H., 1981. Role of phytostilbenes in decay and disease resistance. Annu. Rev. Phytopathol. 19, 437–458.
Hart, J.H., Hillis, W.E., 1974. Inhibition of wood-rotting fungi by stilbenes and other polyphenols in *Eucalyptus sideroxylon*. Phytopathology 64, 939–948.
Hawley, L.F., Fleck, L.C., Richards, C.A., 1924. The relation between durability and chemical composition in wood. Ind. Eng. Chem. 16, 699–700.
Hillis, W.E., 1985. Biosynthesis of tannins. Chapter 13. In: Higuchi, T. (Ed.), Biosynthesis and Biodegradation of Wood Components. Academic Press, Inc, New York, NY.
Hillis, W.E., 1987. Heartwood and Tree Exudates. Springer-Verlag, New York.
Humphrey, C.J., 1916. Laboratory tests on the durability of American woods -I. Flask tests on conifers. Mycologia 8 (2), 80–93.
Jin, L., van der Kamp, B.S., Wilson, J., 1988. Biodegradation of thujaplicins in living western redcedar. Can. J. For. Res. 18, 782–786.
Johnson, E.L., Cserjesi, A.J., 1980. Weathering effect on thujaplicin concentration in western redcedar shakes. For. Prod. J. 30 (6), 52–53.
Kindl, H., 1985. Biosynthesis of stilbenes. In: Higuchi, T. (Ed.), Biosynthesis and Biodegradation of Wood components. Academic Press, Inc, New York, NY.
Kennedy, R.W., 1956. Fungicidal toxicity of certain extraneous components of Douglas-fir heartwood. For. Prod. J. 5, 80–84.
Laks, P.E., 1987. Flavonoid biocides: phytoalexin analogues from condensed tannins. Phytochemistry 26 (6), 1617–1621.
Little, E.L., 1979. Check List of Native and Naturalized Trees of the United States Including Alaska. U.S.D.A. Forest Service, Agriculture Handbook No. 541, Washington, DC, p. 375.
McDaniel, C.A., 1989. Major termiticidal components of Port-Orford-cedar, *Chamaecyparis lawsoniana* (A. Murr.) Parl. Mater. Org. 24 (1), 1–15.
Merrill, W.B., Cowling, E.B., 1965. Effect of variation in nitrogen content of wood on rate of decay. Phytopathology 55, 1067–1068.
Morrell, J.J., Zabel, R.A., 1988. The effects of partial lignin removal on soft rot capability of fungi isolated from preservative-treated southern pines. Mater. Org. 22 (3), 215–224.
Nault, J., 1988. Radial distribution of thujaplicins in old growth and second growth of western red cedar (*Thuja plicata* Donn). Wood Sci. Technol. 22, 73–80.
Panshin, A.J., deZeeuw, C., 1970. Textbook of Wood Technology. McGraw-Hill Book Co, New York.

Scheffer, T.C., 1957. Decay resistance of western red cedar. J. For. 55, 434–442.
Scheffer, T.C., 1963. Effect of gamma radiation on decay resistance of wood. For. Prod. J. 13 (5), 268.
Scheffer, T.C., Hopp, H., 1949. Decay Resistance of Black Locust Heartwood. U.S. Dept. Agr. Tech. Bull., No. 984, Washington, D.C., 37 pp.
Scheffer, T.C., Englerth, G.H., 1952. Decay resistance of second-growth Douglas-fir. J. For. 50 (6), 439–442.
Scheffer, T.C., Eslyn, W.E., 1961. Effect of heat on decay resistance of wood. For. Prod. J. 11, 485–490.
Scheffer, T.C., Cowling, E.B., 1966. Natural resistance of wood to microbial deterioration. Annu. Rev. Phytopathol. 4, 147–170.
Scheffer, T.C., Englerth, G.H., Duncan, G.G., 1949. Decay resistance of seven native oaks. J. Agric. Res. 78, 129–152.
Scheffer, T.C., Morrell, J.J., 1998. Natural durability of wood: a worldwide checklist of species decay resistance. 22, Forest Research Laboratory, Oregon State University, Corvallis, Oregon, 40 p.
Schultz, T.P., Hubbard Jr., T.F., Jin, L.H., Fisher, T.H., Nicholas, D.D., 1990. Role of stilbenes in the natural durability of wood: fungicidal structure-activity relationships. Phytochemistry 29 (5), 1501–1507.
Shigo, A.L., 1965. The pattern of decays and discolorations in northern hardwoods. Phytopathology 55, 648–652.
Southam, C.M., Erlich, J., 1943. Effects of extract of western red cedar heartwood on certain wood-decaying fungi in culture. Phytopathology 33 (6), 517–524.
Southwell, C.R., Bultman, J.D., 1971. Marine borer resistance of untreated woods over long periods of immersion in tropical waters. Biotropica 3 (1), 81–107.
Standards Australia, 2005. AS 5604-2005: Timber-Natural Durability Ratings. Standards Australia, Melbourne, Australia.
Taylor, A.M., Gartner, B.L., Morrell, J.J., 2002. Heartwood formation and natural durability – a review. Wood Fiber Sci. 34 (4), 587–611.
Taylor, A.M., Brooks, J.R., Lachenbruch, B.L., Morrell, J.J., 2007. Radial patterns of carbon isotopes in the xylem extractives and cellulose of Douglas-fir. Tree Physiol. 27, 921–927.
Taylor, A.M., Brooks, J.R., Lachenbruch, B., Morrell, J.J., Voelker, S., 2008. Correlation of carbon isotope ratios in the cellulose and wood extractives of Douglas-fir. Dendrochronologia 26 (2), 125–131.
Taniguchi, T., Harada, H., Nakato, K., 1986. Characterization of inorganic deposits in tropical woods by a new method of isolation. J. Jpn. Wood Res. Soc. 32 (4), 227–233.
U.S.D.A., 2010. Wood Handbook: Wood as an Engineering Material. U.S. Forest Products Laboratory General Technical Report FPL-GTR-190, Madison, Wisconsin.
White, D.G., 1922. Relative durability of untreated native woods. South. Lumberm. (September) 2, 46–47.
Zabel, R.A., 1948. Variation in the Decay Resistance of White Oak. New York State College of Forestry, Syracuse, NY.
Zainal, A.S., 1976. The effect of a change in cell-wall constituents on decay of Scots pine by a soft rot fungus. Material und Organismen 11, 295–301.

CHAPTER NINETEEN

Chemical protection of wood (wood preservation)

Contents

While the proper handling and use of wood in well-designed structures can minimize damage from the biodeterioration agents, some uses require special chemical treatments to obtain economic service lives. This chapter reviews briefly the treatments and chemicals currently used to protect wood used under conditions suitable for aggressive decay development, placing special emphasis on fungal damage and environmental aspects. Two dated, but comprehensive treatments of this topic are also available (Nicholas, 1973; Hunt and Garrett, 1967).

Wood Microbiology.
DOI: https://doi.org/10.1016/B978-0-12-819465-2.00019-X

A brief early history

Preventing decay has long been a concern to wood users, preceding by hundreds of years, the discovery of fungi as the causal agents of decay. Wooden ships played a major role in the early explorations, discoveries, and development of world commerce. Even the earliest ship builders (circa 1000 B.C.) understood the need to use wood from durable species and there are reports of attempts to protect wood by daubing the surface with toxic oily extracts from cedar (Graham, 1973). A variety of surface barrier treatments were used to reduce the ravages of marine borers, but only copper sheathing proved effective. Unfortunately, nothing protected wood from decay in some uses and the search for effective treatments and new sources of durable woods accelerated in the 1800s. Durable wood was sorely needed, not only for new ships, but also to replace decaying wood in men-of-war of the feuding nations of Europe.

The huge fleets necessary to control the far-flung colonial empires of England, Spain, France and the Netherlands placed tremendous pressure on locating new supplies of wood for construction and repairs. Shortages of naturally durable timbers (e.g. oaks, cedars) forced builders to use less durable species that further exacerbated the decay problem. The shortage of durable woods stimulated a frantic search for effective methods to prolong wood service life. Of the many advances at this time, the use of creosote (a coal-tar distillate) by Moll around 1836 and the patenting of the full-cell process for pressure treating wood by Bethell in 1839 had major influences on the development of a wood preservation industry. The near simultaneous development of a treatment that could protect timber against decay and a method for moving into the wood created tremendous potential for reducing timber losses, but it was to be decades before these advances were fully utilized. It is also interesting to note that these advances preceded, by nearly 50 years, Hartig's discovery that fungi caused wood decay. The early history and major subsequent developments in wood preservation are available in several sources (Van Groenou, 1951; Graham, 1973).

The chemical protection of wood has developed into a major worldwide industry. In most countries, associations have developed among the wood scientists, preservative producers, and treaters of the region to develop specifications for preservatives, standards for treatments, and promote research. The American Wood Protection Association (AWPA) was

formed in 1904 in the United States for these purposes. It publishes an Annual Proceedings that is the principal source of information on wood preservation and new developments, nationwide. The AWPA has also developed and maintains a set of accepted standards for both preservatives and treatments that are widely used in the wood preservation industry. Similar groups, such as the Canadian Wood Preservation Association, and the Japan Wood Preserver's Association serve similar functions in other countries. The International Research Group on Wood Protection (IRGWP), formed in 1969, now plays a major international role in dissemination of information on the destructive agents of wood and stimulates collaborative research on wood protection.

Treatment choice and the biological hazard

Although methods for protecting wood have expanded and improved, the damage produced by wood-destroying agents and requirements for their control have remained relatively constant. Selection of preservatives and treating methods requires consideration of the type of product used, the degree of biotic hazard, the length of time the wood must be protected, and the financial merits of such treatments.

For example, wood subjected to high decay hazards such as in ground contact in tropical zones requires the most effective preservatives and treatments available. Wood subjected to repeat shock loads (for example, railroad ties) that cause rapid strength losses can generally be treated with lower levels of chemical, even though the decay hazard is high, since the wood will fail mechanically before serious decay damage occurs. Sawmill operators are primarily interested in preventing fungal and insect attack for relatively short periods during log storage and lumber seasoning until the wood dries below the fiber saturation point. Users of wood in cooling towers, other adverse decay conditions, or applications where termites are also a threat are concerned with protecting the wood for 30 or more years. It is important to stress that use requirements and environmental considerations generally dictate the type of wood preservative used and how it is applied.

Treatment results are normally expressed in terms of chemical loading and depth of preservative penetration. Chemical loadings for short term

protection such as prevention of mold and stain after sawing are normally expressed on the basis of solution absorbed per unit volume of wood. Typically, uptakes are expressed in gallons (liters) of chemical per thousand board feet (or cubic meters) of wood. These levels may also be expressed on a ppm basis for a shallow zone of the wood near the surface. Penetration of chemical into the wood is considered to be less important for short term protection. These processes typically deliver a shallow coating to the wood surface much in the same way as farmers spray the foliage of their crops to prevent various diseases.

Results for chemical treatments for longer term protection are normally expressed as weight of chemical per cubic foot of wood treated (lbs chemical/ft^3 or kg/m^3), weight of chemical per weight of wood (%mass/mass) or retention in an assay zone. Preservative penetration is essential for long-term performance and is usually measured visually when the treatments modify wood color, through the use of indicators specific for the treatment chemical or through surrogates added to the treating solution that can be visualized using indicators.

Treatments can be categorized in a variety of ways, but one simple approach is to look at the protective period required.

Short-term wood protection

Freshly cut wood is susceptible to invasion by decay and stain fungi until it dries below the FSP. While rapid processing and kiln drying can limit this damage, unforeseen delays can and do occur. The degradation of logs and unseasoned timber (pulpwood, chips, poles, posts, etc.) are major problems in areas where temperature and humidity conditions favor fungal growth or where the species being harvested contain high percentages of sapwood. These deterioration problems were considered in some detail in earlier chapters on log and chipwood storage (Chapter Thirteen) and sapstains and wood discolorations (Chapter Fourteen). They will be only briefly summarized here as they relate to other treatment methods and preservatives.

Most sawmills prevent stain and mold attack by either kiln drying or dipping/spraying freshly sawn high grade lumber with a fungicidal solution to coat the wood surface. This thin, prophylactic barrier prevents fungal spores from germinating on the wood surface and provides only short-term (5–6 months) protection. These chemicals can be applied using a variety of methods. Lumber pieces can be individually dipped in

the chemical, the surfaces can be sprayed as the material moves along the green chain or entire units can be immersed in the treatment solution. Each of these approaches has advantages and disadvantages. Dipping or immersion results in excellent chemical uptake on all surfaces, producing a well-treated surface barrier, but the process can be slow. Spray systems allow for more rapid treatment, but are prone to problems with clogged nozzles that results in incomplete surface treatment. These protective barriers can be thin since they are usually removed in the final planing process, but they must completely protect the wood surface to be effective.

The effectiveness of potential anti-sapstain chemicals was extensively evaluated on southern pine sapwood in the 1930s (Lindgren and Scheffer, 1939). Ethyl mercury phosphate and a sodium salt of pentachlorophenol (NaPenta) became the most commonly used chemicals for sapstain control. In the 1960, concerns about the safe use of mercury compounds left NaPenta as the mainstay of the sapstain control industry.

NaPenta had the advantage of low cost, broad-spectrum toxicity, and solubility in water for ease of application. These properties made it difficult for other chemicals to compete, but increasing environmental concerns about the safety of NaPenta led to the removal of this compound from the market and stimulated a search for alternative fungicides. The major concern with NaPenta was the presence of contaminants, known as dioxins. Many countries have since banned the use of NaPenta or the importation of lumber dipped in the chemical. This compound is no longer used in North America.

It is not within the scope of this chapter to outline all of the substitute chemicals currently being used, but several that merit attention include copper-8-quinolinolate, 2(thiocyanomethylthiobenzothiazole), a number of quaternary ammonium compounds, and 3-iodopropynyl butyl carbamate (Smith et al., 1985) (Table 19.1). In general, these compounds lack the broad-spectrum toxicity of NaPenta, but present much lower environmental risks. These compounds are often used in combinations to protect against the diverse array of fungi that can attack wood.

Although the chemicals used to prevent sapstain are changing, it is unlikely that we can completely divorce ourselves from their use during the air seasoning of high quality lumber. Increasing kiln capacity to eliminate air-seasoning is expensive and impractical for large-dimension material because of the long cycles required for complete drying. Delays in processing lead to the staining of green lumber prior to kiln-drying. In addition, many producers of kiln-dried lumber still apply some protective

Table 19.1 Chemical systems currently registered for protection of lumber against fungal stain and mold.

Abbreviation[a]	Active ingredient	CAS #[b]
Carbendazim	Methyl-2-benzimadazole carbamate	10605-21-7
Chlorothalonil	Tetrachloroisophthalonitrile	1897-45-6
Copper-8-quinolinolate	Bis(8-quinolinolato)copper	10380-28-6
TCMTB	2(Benzothiazolylthio)methylthiocyanate	21564-17-0
MBT	Methylene bisthiocyanate	6317-18-6
Propiconazole	1-[(2-(2,4-Dichlorophenyl)4-propyl-1, 3-dioxylan-2yl)methyl]-1,2,4-triazole	60207-90-1
Tebuconazole	1-(4-Chlorophenyl)-4,4 dimethyl-3-(1,2, 4-triazol-1-ylmethyl)-pentan-3-ol	107534-96-3
Cyproconazole		94361-06-5
Fenpropimorph	4[3-(4-*Tert*-butylphenyl)2-methylpropyl]-2, 6-dimethylmorpholine	91269-52-2
IPBC	3-iodo-2-propynyl butylcarbamate	55406-53-6
TBTO	Tri-butyltin oxide	
DDAC	Didecyldimethyl cocammonium chloride	7173-51-5
BAC	Alkyldimethylbenzyl ammonium chloride	68424-85-1
OPP	Sodium orthophenylphenate	132-27-4
Sulfone	Di-iodomethyl-p-tolylsulfone	20018-09-1
Isothiazolone	5-Chloro-2-methyl-4-isothiazolin-3-one 2-methyl-4-isothiazolin-3-one	
MBT	Methylene bisthiocyanate	6317-18-6
TCMTB	2(thiocyanatomethyl) thiobenzothiazole	21564-17-0
Cu-8	Copper-8-quinolinolate (oxine copper)	10380-28-6
CTL	Tetrachloroisophthalonitrile	1897-45-6
SMDC	Sodium methyldithiocarbamate	1965−8
Mercapt	2-Mercaptobenzothiazole	2492-26-4

[a]Abbreviations are for convenience in the next table.
[b]CAS = Chemical Abstract Services #. Primary manufacturer is listed; however, there may also be secondary manufacturers or distributors.

biocide to the surface to reduce the risk that mold or stain will develop due to inadvertent wetting during transport or storage. There remains a continued interest in the development of less toxic methods for limiting mold and stain, but it is challenging because of the range of organisms that can invade wet wood and the differences in wood species.

Long-term wood protection

While short-term decay and sapstain damage can be minimized by proper handling of wood and by dipping or spraying with fungicidal

solutions, preventing degradation for long periods requires more effective treatments.

Wood preservation is divided into non-pressure and pressure treatments (for more detailed references see Wilkinson, 1979; Levi, 1973; American Wood Protection Association, 2017; Hunt and Garrett, 1967; Nicholas, 1973).

Non-pressure treatments: Dipping, brushing, spraying, and soaking require minimal equipment and are often used by homeowners for treating limited amounts of wood or farmers who have a source of inexpensive timber. Brushing or spraying produces only a thin shell of preservative treatment that provides short-term protection to wood exposed out of ground contact, but is of minimal value for long-term protection needs or when the wood is exposed in soil (Morrell et al., 1999). The two most common uses of brush treatments are coating the cut ends of pressure-treated lumber to protect the untreated wood that exposed in cutting or the routine applicant of various deck coatings to protect the wood from ultraviolet light degradation and fungal attack (Morrell et al., 2001). These treatments penetrate only a short distance into the wood to provide temporary protection that can extend the useful life of the product.

As expected, dipping or soaking provides greater absorption of preservative solution deeper into the wood, particularly if the wood is dry. Manufacturers of wood door-frames and windows make extensive use of dip or low vacuum treatments to produce a protective barrier in above-ground uses where the wood is coated with a paint film. The species used are easily penetrated with liquids, particularly at the end-grain of joints where moisture absorption is high and the decay normally occurs. These materials are also usually coated or clad, further protecting the wood from wetting. Many farmers soak air-dry posts in oilborne preservatives for periods of 1–7 days prior to use (Morrell et al., 1999). The most common chemical for this purpose is copper naphthenate, which is available in either oil or water-soluble formulations.

Some wood users have also utilized this simple treatment method by dipping peeled, green posts first in a water-soluble preservative such as copper oxide and then with a second such as chromium trioxide. These compounds react in the wood to form a water insoluble precipitate. This process permits the wood to be treated while it is still green; however, chromium trioxide is no longer available for this purpose. The lack of chromium to help fix the copper results in rapid loss of copper to the surrounding

environment (Morrell et al., 2005). For this reason, this treatment system using water-soluble copper salts is no longer recommended.

One water borne treatment that can be used for protecting wood used in interior applications is boron dip diffusion. In this process, freshly cut lumber or poles are dipped for several minutes in a concentrated solution of borate and then solid piled to allow the boron to diffuse inward from the surface. Boron-treated material can be used in interior applications protected from wetting or it can be over-treated with heavy duty wood preservative for exterior use. The internal boron protects the wood from internal decay and has provided excellent protection to railway ties but is more difficult to use with larger timbers such as utility poles (Amburgey and Sanders, 2007, 2009; Cappellazzi et al., 2019).

While non-pressure treatments can improve wood service life, the high variation in preservative penetration and distribution along with the limited protection periods these treatments provide makes them less attractive where wood in costly structures must be exposed under adverse conditions.

Instead, the wood can be treated using vacuum treatments alone or in combination with pressure processes.

Pressure treatments: Pressure treatment with a heavy-duty preservative is virtually required where wood is used in high decay hazard conditions. When performed properly, pressure-treatment results in deeper, more uniform treatment of the wood. There are three basic pressure treatment processes.

The thermal process uses the natural development of small pressure differences inside the wood to force solution inward. In this process, dry wood is placed into a tank, preservative solution is added and heated to 150–230 °F (65.5–110 °C) for periods ranging from 16 to 48 hours. After the heating period, the preservative solution is withdrawn and a cooler preservative solution is pumped in. The cool solution results in a pressure differential in the wood that draws preservative inward, increasing uptake. The thermal process is used most extensively to treat western red-cedar and lodgepole pine posts. Because of the high temperature of the baths, the process is generally used with oil borne chemicals that are less likely to evaporate. The process is now also generally performed inside closed vessels because of concerns about the release of volatile organic compounds into the environment.

Vacuum treatments are primarily used to treat permeable woods for use in windows and door frames (fenestrations). Dry wood is placed inside

Figure 19.1 Commercial wood treatment cylinders (retorts) can range from 1.5 to 4 m in diameter by up to 42 m in length.

a sealed vessel and the air is withdrawn. Treatment solution is then added and the vacuum is released. The more permeable portions of the wood readily absorb the treatment. This process may be repeated to increase preservative uptake.

The two remaining pressure treatments, the full and empty cell processes, require more elaborate equipment and result in deeper, more uniform penetration than thermal or vacuum treatment (Fig. 19.1). The full cell or Bethell process, developed in 1839, represented a quantum leap in treatment technology. In this process, wood is placed in a treating cylinder (retort), a vacuum is drawn to remove as much air as possible from the wood, and the preservative is added to the retort. The pressure is gradually raised to a maximum of 150–200 psi (1050–1408 kPa) and held until gauges on the outside of the retort indicate that a sufficient amount of solution has been forced into the wood (Fig. 19.2). This level, called gauge retention, depends on the volume of wood being treated, as well as the retention and penetration values required by specifications for a given commodity. Once this target value has been achieved, the pressure is released and the preservative solution is withdrawn. At this point, the pressure release causes air in the wood to expand and force outward a certain amount of preservative. The amount of preservative released from the wood is called the kickback. Additional periods of heating in solution, steaming, and vacuum may also be used to remove surface deposits and reduce residual internal pressure inside the wood. These processes reduce subsequent bleeding of preservative once the wood is placed in service.

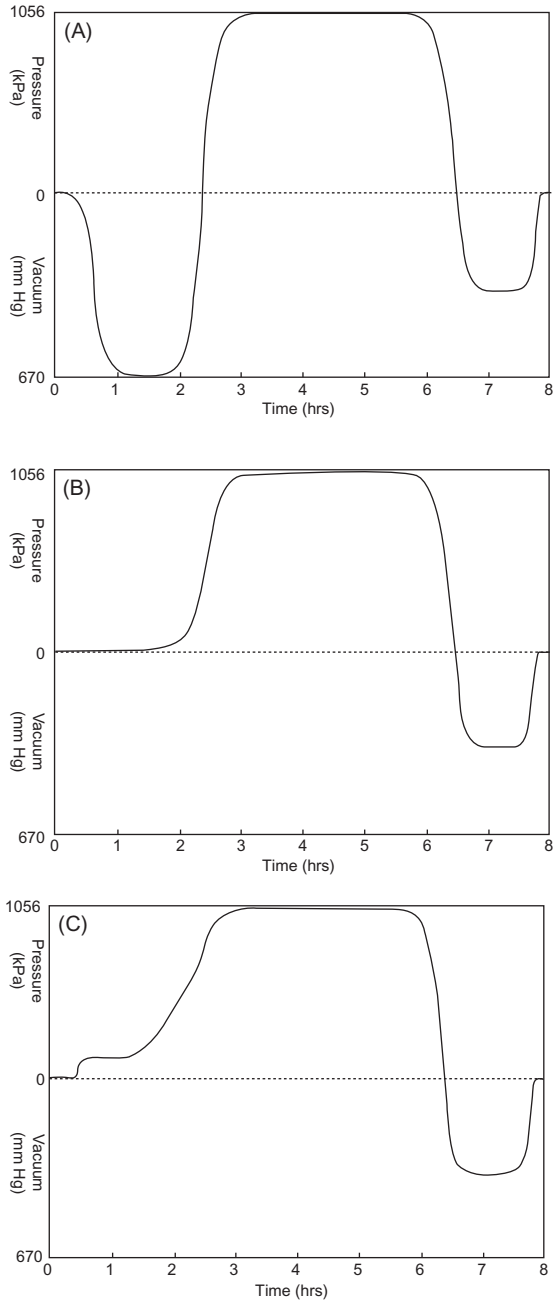


Figure 19.2 Pressure treatment processes include Full Cell (Bethel) (A) or Empty cell (Lowry and Rueping) (B and C) designed to deliver varying amounts of preservative into the wood.

The full cell process results in a maximum uptake, or retention, of preservative for a given depth of penetration and is used for treating marine piling to high retentions with creosote and for all wood with waterborne solutions. In the latter preservative type, the concentration of chemical in the water can be adjusted to achieve the desired chemical loading.

In the empty cell (Lowry or Rueping processes), no vacuum is used. The solution is added directly to the cylinder in the Lowry process and the pressure is raised and held until the desired amount of chemical is forced into the wood. The pressure is then released and air that was trapped in the wood expands outward, forcing excess preservative from the wood. The Lowry process traps more air in the wood, resulting in larger kickback of preservative at the end of the treatment cycle than a Bethell process. This extra kickback produces a lower retention for a given depth of penetration (Fig. 19.2). In the other empty cell process (Rueping Process), a low pressure (30 psi, 211 kPa) is applied before the solution is added to the cylinder, trapping additional air in the wood and increasing the amount of kickback. The empty-cell processes are used to treat utility poles and lumber with oilborne chemicals for terrestrial uses where a clean surface is desirable.

In addition to the treatments described, a number of variations have been developed to improve treatment with certain chemicals or wood species. One involves the application of preservative in liquefied petroleum gas (LPG) or methylene chloride. In the process, solution is forced into the wood by pressure, the excess solution is withdrawn and the pressure is released. Once the pressure decreases, the solvent volatilizes, leaving a clean paintable surface and the solvent can be recovered for reuse. This process was used for treatment of laminated timbers and utility poles with pentachlorophenol, but problems with surface decay and concerns about the safety of LPG during treatment have resulted in the discontinuation of this treatment (Lew and Wilcox, 1981).

In addition to the conventional processes, several other systems have been developed. The vapor boron process was developed simultaneously in the United Kingdom and New Zelaand. In this process, a vacuum is drawn over the wood and then trimethyl borate is introduced. The compound reacts with water in the wood to deposit the boron in the wood. This process has been used with composites, but it not currently used.

Supercritical fluid impregnation was first proposed in Japan in the 1980s. Supercritical fluids are materials that have been compressed beyond their critical pressure and heated above their critical temperature.

SCF's have properties that are similar to a gas in terms of the ability to move into wood, but can solubilize compounds at levels approaching conventional solvents. This allows them to penetrate through woods that normally resist treatment. SCF's are commercially used to impregnate spruce window materials in Denmark and continue to have great potential for treating a variety of other products. The initial costs of the treatment facilities have largely limited commercial use, but the fact that the process is non-swelling has tremendous potential for treating composite products.

The major wood preservatives

Numerous chemicals have been evaluated for their ability to protect wood, but only a select few have proven effective and are used in large quantities. These compounds can be divided conveniently into oil -and waterborne chemicals. The oil borne chemicals have the advantage of resistance to leaching and ease of penetration into the wood due to decreased surface tension. Most waterborne chemicals do not require costly solvents and leave the wood surface clean and paintable. The nature of the preservative carrier can have substantial effects on preservative performance (Arsenault, 1973). Carriers can reduce viscosity, enhance water repellency, or alter the bioactivity of the preservative. An example is a comparison of the use of pentachlorophenol (PCP) in liquified petroleum gas (LPG) or heavy oil. PCP has performed well in heavy oil, but LPG treatments with the same chemical are less effective and highly susceptible to surface decay (Lew and Wilcox, 1981). Similarly, the use of biodiesel to improve solvency of pentachlorophenol and reduce odors has no apparent negative effects on performance, but adding this same material to copper naphthenate has been shown to reduce decay resistance. Seemingly minor changes can have dramatic effects. Compositions or formulas for the major preservatives are indicated in Table 19.1 or Fig. 19.3.

Organic (oil borne) preservatives

The four oil borne preservatives most commonly used in the United States for pressure treatment of wood.

Creosote: Creosote is among our oldest, most reliable preservatives, and ushered in the era of effective wood protection. Creosote is a

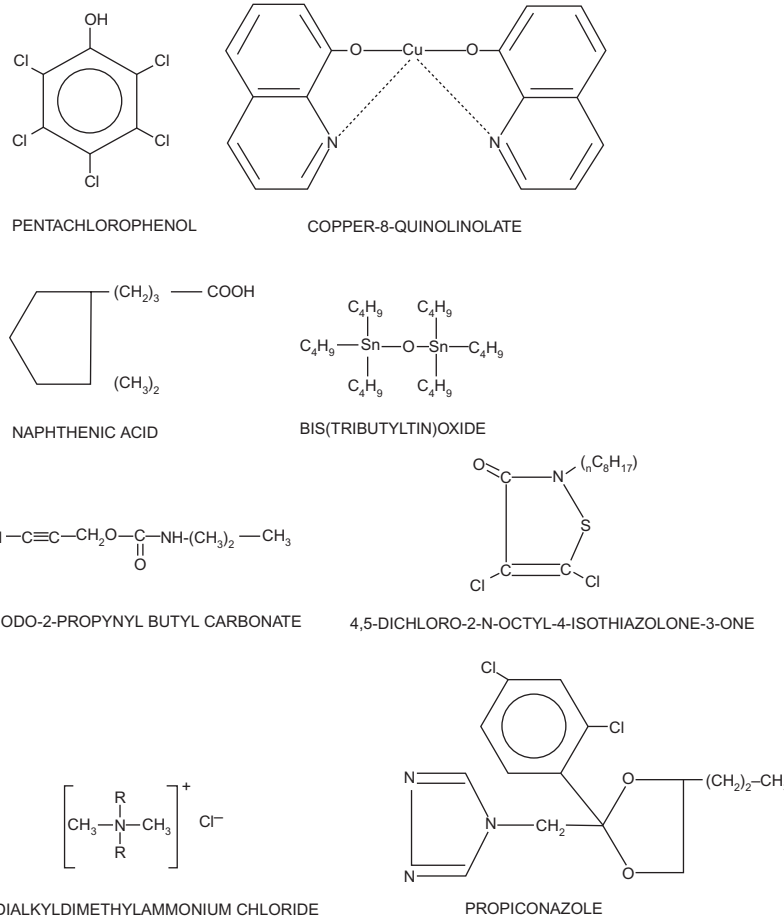


Figure 19.3 Chemical structures of some commonly used wood preservatives.

complex mixture containing over 200 polynuclear aromatic hydrocarbons (PAH) produced as a by-product of the high temperature carbonization (900−1175 °C) of coal to produce coke for the steel making process and from distillation of the tars and pitches formed (Fig. 19.4). The composition of creosote is variable, depending on the coal source and the distillation ranges used. The AWPA preservative standards specify boiling point ranges for various fractions rather than specific chemicals for this reason. The heavy oil fractions of creosote have proven to be the most effective toxic component. Because of its complexity, the toxic mode of creosote has never been fully determined; however, it protects wood against

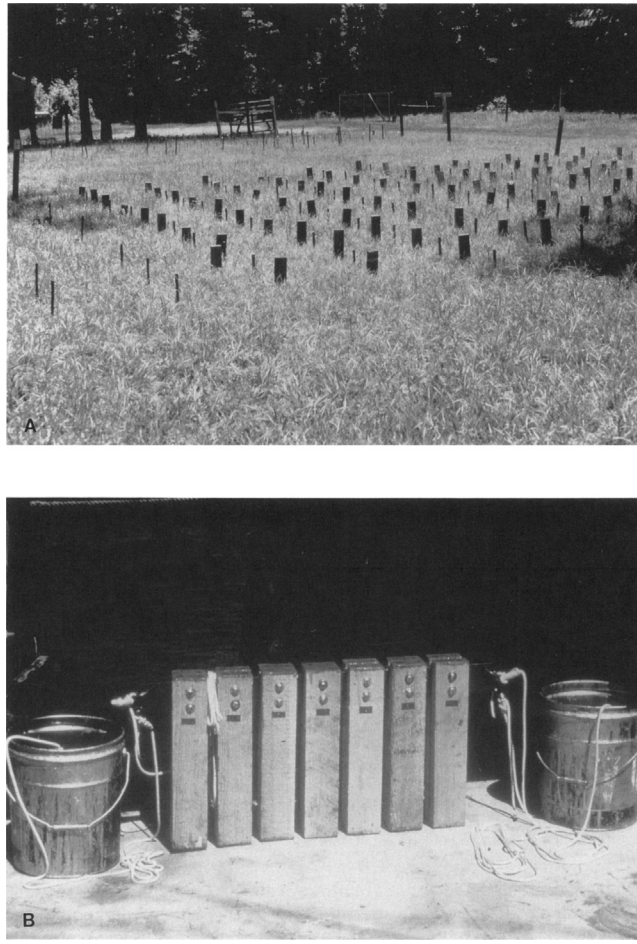


Figure 19.4 New preservatives are generally evaluated in small sapwood samples including (A) stakes partially buried in soil or (B) panels submerged in the ocean. In each case, the wood is regular inspected visually for evidence of damage. *Courtesy: Jed Cappellazzi.*

insects, fungi, and most marine borers (Webb, 1980). Commonly used retentions for wood uses in ground contact range from 72 to 192 kg/m^3, depending on severity of exposure. To put this in perspective, these levels represent adding 16 to almost 43% creosote by weight to an average softwood. No other preservative system is used at such high levels.

Creosote treated poles have lasted 80 years with no loss in effectiveness. Creosote was once among our most important wood preservatives, but recent emphasis on clean, dry wood products free of surface deposits

and concerns about the environmental safety of PAH's in general have relegated this preservative to applications where direct human contacts are minimal (Webb, 1987). Nevertheless, creosote is still used to protect 20–25% of the wood treated each year, primarily for utility poles, marine piling, and railroad ties (American Wood Protection Association, 2017). Well over 95% of railroad ties used in North America are treated with creosote.

Creosote has also been mixed with pentachlorophenol, copper naphthenate, naphthalene, and petroleum for specific uses. The majority of these blends have only been used on a limited basis. Creosote was blended with copper naphthenate in the 1940s, during the Second World War to extend creosote supplies, while pentachlorophenol was briefly added before it was discovered that the mixture was highly corrosive. Creosote/petroleum blends are widely used to protect railroad ties and bridge timbers, especially in tropical environments.

As a by-product of coke ovens, the creosote supply was dictated by the steel industry, which experienced wide fluctuations in production. As a result, periodic creosote shortages arose. In addition, the composition of creosote varied widely as some producers removed some of the more toxic fractions as distillate for other uses. At present, creosote specifications are tightly controlled, but the supply and quality variations stimulated a search for other preservatives. Creosote is a restricted-use pesticide and users of the chemical must be licensed by their respective states.

Pentachlorophenol: The search for a clean, effective alternative to creosote led to the development of pentachlorophenol (PCP), one of our first synthetic pesticides, in the 1930s (Rao, 1977). PCP is easily synthesized by successive chlorinations of phenol. It functions as a biocide by uncoupling oxidative phosphorylation. PCP, and its water-soluble sodium salt, exhibited broad-spectrum toxicity to fungi and insects, were relatively inexpensive, and could be supplied in large quantities. As a result, PCP rapidly replaced creosote in many applications especially for utility poles and bridge timbers. The synthesis of PCP carries with it the risk of inadvertently producing dioxins. Some dioxins are extremely hazardous. Concern over the presence of dioxins and related environmental problems led to reduced use of this chemical in some applications. The levels of dioxin in PCP have been markedly reduced by improved process control. PCP is primarily used to protect utility poles and industrial timbers and is only used in the U.S. and Canada where extensive testing has shown that

the benefits of use far out-weigh the risks. At present, PCP is a restricted use pesticide and users must be licensed by the appropriate state agencies.

Copper naphthenate: Copper naphthenate, a copper complex with naphthenic acids derived from the oil-refining process, has recently been promoted as a PCP replacement. This chemical, which is 20 times less toxic to humans than PCP has been used to protect industrial products for over 40 years. It has recently been more heavily used for railway bridges and ties. It has been used extensively for many years in the retail industry as a brush-on preservative for home and marine use. In cases where the green color of the preservative is objectionable, the less effective zinc salt is used.

Tributyltinoxide: TBTO was used extensively in Europe to protect window and door frames and in many marine paints as an anti-foulant for barnacles. In the United States, TBTO was primarily used for treating millwork, particularly where rapid removal of the solvent following treatment was desired. TBTO has not performed well in ground contact, where fungi have been shown to degrade this chemical. Reports about damage to shellfish beds from TBTO leaching from antifouling paints led to gradual discontinuation of use of this chemical for wood protection.

Copper-8-quinolinolate: This chemical, also known as Cu-8 or oxine copper, has the lowest mammalian toxicity of any wood preservative currently used for wood treatment. It is the only preservative approved for treatment of wood in direct food contact by the U.S. Food and Drug Administration. Cu-8 has been used to protect food crates and picnic tables, but its high cost and questions about its performance in direct ground contact have largely limited its use to specialty products.

Waterborne chemicals

Waterborne chemicals have been used in the United States for over one hundred years, beginning with mercuric chloride ($HgCl_2$) in the Kyanizing process (Hunt and Garrett, 1967). Water reduces solution costs and generally leaves wood surfaces clean and paintable. Another important advantage is the reduction in release of volatile hydrocarbons as atmospheric pollutants during the treatment process. Many of the waterborne systems use heavy metals and include chromated copper arsenate (CCA), ammoniacal copper zinc arsenate (ACZA), acid copper chromate (ACC), alkaline copper azole (CA), alkaline copper quaternary (ACQ) and micronized copper azole (MCA)

Chromated copper arsenate: CCA was developed in India in the 1930's and rapidly grew to become one of the most widely used preservatives for wood protection. In the United States, this wood preservative is formulated from chromium trioxide, copper oxide, and arsenic pentoxide. Copper and arsenic are excellent broad-spectrum fungicides. Arsenic is a competitive inhibitor for phosphorus in ATP synthesis and provides insect and marine borer protection. Chromium has strong affinity for and complexes with the lignin in wood to limit leaching. As a result, CCA was extensively used to protect lumber from decay fungi, insects, and most marine borers in residential and industrial applications. A number of formulations of CCA that varied in the proportions of the three metals were used in other countries.

Unlike oilborne chemicals that are deposited on the wood cell wall, but do not appear to be chemically bound, CCA is strongly fixed to the lignin component by the reduction of Cr (+6) to Cr (+3). This fixation, which is strongly influenced by wood pH and temperature, also appears to help retain arsenic and copper in the wood (Hartford, 1986).

In the last 1990's, CCA came under considerable pressure because it contained arsenic. There were a number of efforts to have CCA banned from use in residential applications. In addition, accidental burning of CCA treated wood in power plants in Florida resulting in the creation of heavy-metal contaminated ash generated additional negative publicity. The manufacturers voluntarily withdrew CCA from the market in the US for residential applications in 2003. It remains widely used for protecting wood used in industrial applications such as utility poles, marine piling and other applications where heavy-duty protection is required.

Ammoniacal copper arsenate (ACA)/Ammoniacal Copper Zinc Arsenate (ACZA): ACA was developed in the 1930s for impregnating difficult to treat or refractory wood species, such as Douglas-fir. Unlike CCA, which is an acidic system, ACA uses ammonia to solubilize or dissolve the copper. The ammonia improves the ability of the preservative penetrate into the wood through a combination of swelling the wood and dissolving extractives on the pits that impede flow. The result is deeper penetration into the wood. The absence of chromium in ACA means that it is not as strongly fixed to the wood. While some copper does react with the wood, the majority precipitates when the ammonia evolves from the wood.

ACA was replaced with ammoniacal copper zinc arsenate (ACZA) in the 1980s. The zinc reduced corrosion, but also resulted in slightly better

immobilization of the metals, reducing the potential for environmental contamination. ACZA tends to remain more closely bound to the wood than ACA and has fewer issues with corrosivity to metals used for attachments to the wood. ACZA has provided excellent long term protection (Johnson and Gutzmer, 1984; Lebow and Morrell, 1993, 1995; Wilcox, 1987).

Like PCP and creosote, CCA, ACA, and ACZA are restricted-use pesticides that can only be applied by certified applicators who have passed a state administered test on pesticide safety and usage.

Acid copper chromate (ACC): ACC has primarily been used to protect wood in cooling towers and other non-ground contact applications. Like CCA, the chromium in ACC reacts with the wood as well as the copper to help immobilize the metals. ACC sees limited use in North America, because it has been replaced by CCA and the alkaline copper compounds.

Chromated Zinc Chloride (CZC): Like ACC, CZC used chromium to help fix the zinc in the wood. Its predecessor, zinc chloride, was initially used to protect railroad ties and other timbers, but zinc chloride did not appreciably react with the wood and had a tendency to leach from the wood. As a result, it did not perform well in adverse environments. The excellent performance of the other waterborne chemica1s has sharply reduced demand for CZC and it is no longer used.

Alkaline copper compounds: Concerns about the presence of arsenic and chromium in CCA led to the development of ammonia based systems containing copper and a co-biocide to protect against organisms that were tolerant to copper. The two co-biocides were either quaternary ammonium compounds or triazoles. Unlike CCA, which is an acidic system, the alkaline copper compounds are similar to ACZA in that they initially used ammonia and later amines to solubilize the copper. The systems differ markedly in their interactions with the wood since there is no chromium to react with the wood and co-precipitate the copper and arsenic. The first of these systems was ammoniacal copper quaternary ammonium compound (ACQ). ACQ use was limited until CCA was withdrawn from the market, then its use skyrocketed. The ammonia in ACQ was a nuisance and was soon replaced with ethanol amine. Shortly thereafter, alkaline copper azoles (CA) were developed. These systems used copper with an azole as the co-biocide (either tebuconazole or propiconazole or mixtures of the two). These systems became the dominant preservatives used for residential applications in North America.

Micronized Copper systems: Micronized copper systems use very finely ground copper carbonate suspended in water containing co-biocides such as the quats or azoles. They are similar to the alkaline copper compounds in that they use copper with an organic co-biocide. There was great skepticism about the ability of the largely insoluble copper carbonate to provide wood protection, but longer-term tests have shown that a sufficient amount of copper solubilizes to create an effective barrier against fungal attack in properly treated wood. Micronized copper systems are now the predominant treatment for southern pine and other permeable wood species used in residential applications, but they are not suited for impregnation of more difficult to treat timbers because the particle sizes cannot pass through the pit membranes of these species.

Boron: Boron compounds are widely used outside North America for protecting wood from fungi and insects. These compounds diffuse with moisture and can completely penetrate heartwood. Borates are normally applied by dipping, followed by a 4–8 week wet storage period, but can also be pressure impregnated. There are a number of borates with differing degrees of water solubility. Sodium borates are more commonly used for pressure treatment because they can be easily dissolved in water. Zinc borate has much lower water solubility and is added to the mixture of wood chips and resins during manufacturing of oriented strandboard. Although borates are very safe, their application is limited by high susceptibility to leaching in most applications where preservative treatment is required. As a result, they are mostly used in interior applications with a high risk of termite attack. Boron treatments are widely used for protection against termites in Hawaii as well as for treatment of the sill-plates in houses in the Western U.S.

Other compounds: In addition to the above chemicals, several attempts have been made to develop waterborne formulations of PCP and creosote. Waterborne ammoniacal PCP formulations were developed in the 1980s, but tended to perform poorly because the PCP was poorly distributed in the wood and could not produce complete protection. It is no longer used. Similarly, copper naphthenate can be produced in a water-dispersed system and this preservative is seeing some use in agricultural applications. A pigment-emulsified creosote was developed in Australia. While this system has proven effective and easier to handle than traditional creosote, commercialization of this chemical has not occurred in North America (Krzyzewski, 1986; Watkins, 1977; Cookson and Greaves, 1986).

3-Iodo-2-propynyl butylcarbamate (IPBC) has long been used as an additive in paint films and it use as a wood preservative increased sharply when penta was classified as a restricted use pesticide. IPBC is still widely used for treatment of wood windows and doors as well as in various systems for limiting mold and stain on freshly cut lumber. However, it is not used in soil contact and performs best when protected from ultraviolet light.

Quaternary Ammonium Compounds: Quaternary ammonium compounds contain a central nitrogen molecule surrounding by chains of hydrocarbons of varying lengths. These materials are derived from the process of rending animal carcasses. There are a dizzying array of quaternary ammonium compounds, but the two most commonly used quats in wood protection are benzalkonium chloride (BAC) and didecyldimethyl ammonium chloride (DDAC). Quats were used in New Zealand as a stand-alone preservative in the 1980's, but performed poorly and are now generally only used in mixtures to limit the potential for degradation by resistant organisms.

Isothiazolinones are also widely used to provide protection against microbial contamination in various household items. They have been explored for wood preservation for over 3 decades, but are currently only used as an additive to limit mold growth on alkaline copper treated wood. However, one system 4,5-dichloro-2-*n*-octyl-4-isothiazolin-3-one (DCOI), has produced excellent long-term protection in soil contact and has been standardized for wood use in ground contact.

Non-biocidal treatments

While traditional biocides remain the primary approach to protecting wood where design alone cannot include fungi or insects, there has been a concerted effort to develop non-biocidal methods for protecting or modifying the wood. These approaches generally try to either fill the wood cells lumens with materials that prevent fungal attack or alter the wood chemistry to change wood/water relationships.

Bulking agents

A variety of compounds can be used to fill or bulk the wood cell lumens and potentially the capillary voids with materials that limit fungal access or oxygen diffusion.

Polyethylene glycol has long been used to stabilize wood either to retain structure of badly degraded wood or to limit checking and splitting. For example, the Swedish ship the Vasa, was recovered from the sediments of Stockholm Harbor after several hundred years of immersion. The badly degraded materials were sprayed with ethylene glycol to prevent the material from collapsing. Wood carvers and turners also soak their materials in glycols to limit splitting. Glycols can stabilize wood, but they are easily leached and not suitable for most applications.

Low molecular weight resins have also been explored for wood protection (Stamm and Seborg, 1943). The resins are impregnated into the wood using conventional treatment processes then cured by heating. While the resins do not appear to completely inhibit moisture sorption and the associated swelling and shrinking, they can sharply reduce the risk of decay. Mass loadings of 20–25% by weight are generally required for these treatments.

Furfurylation: Furfuryl alcohol is a lower molecular weight compound that is impregnated into the wood and then fixed by heating (Westin et al., 2004). As with resin impregnation, the process bulks the wood cells and requires 20–25% weight gains to be effective; however, the treated wood has performed well against fungi, marine borers and insects.

Dimethyloldihydroxyethelenurea (DMDHEU): is another system that is impregnated into the wood using conventional pressure processes, then heated to react or "fix" the materials (Militz, 1993). This system has also performed well against fungi and insects, but is not yet widely used (Militz et al., 2011).

Chemical modification

While bulking can protect wood, it may not completely alter the wood/moisture relationships. As a result, the wood may still swell and shrink with moisture changes and this can lead to checking and deformation. Chemical modification alters basic wood/moisture interactions, usually by modifying or removing hydroxyls on the hemicellulose and cellulose (Hill, 2006; Rowell, 2005,2006).

Acetylation: This process reacts acetic anhydride with the hydroxyls in the wood. The resulting reaction blocks the hydroxyls, making the wood less hygroscopic. Acetylation is not new- having been developed in the early part of the 20th Century, but it was not economical (Tarkow et al., 1946). The process requires very permeable wood species (radiata pine or red alder are commonly used) and the treatment effectively increases the wood mass by 18%–20%, but the process is effective against fungi, insects, and marine borers. While more expensive than conventional preservative treatments, it is attracting attention as architects and builders seek non-biocidal treatments.

Thermal Modification: Exposure of wood to elevated temperature can result in gradual degradation of the polymers, notably hemicelluloses. Thermal modification was developed in the 1940s as a means for altering the color of the wood (Stamm, 1959; Stamm et al., 1946). Light woods could be darkened to appear similar to more valuable darker woods such as black walnut. The process also altered the wood/moisture relationships, resulting in products that were less hygroscopic. Thermal modification has been extensively studied and there are a variety of processes being used. In general, the results have been mixed. While reduced hygroscopicity can slow water ingress in locations where drying is possible (such as above ground locations- for example siding on a house), decay can still progress. In addition, the wood remains highly susceptible to termite attack, making it unsuitable for many areas of the world. Thermal modification likely has niches where it will perform, but others where the changes in wood chemistry are insufficient to overcome the inherent susceptibility of the wood to biological attack.

While many wood modification methods were originally developed in North America, they have emerged more completely in Europe as increasing restrictions on biocide uses and a general public reaction against chemicals have encouraged the development of alternative wood protection strategies.

Natural biocides

Many plants produce defensive chemicals that limit attack by fungi, insects, and marine borers. Some of these are in the heartwood,

while others are produced in seeds, leaves and other plant parts. These chemicals represent millions of years of co-evolution by plants to produce chemicals toxic or repellent to pathogens that might feed on them.

Scientists have long coveted the chance to use natural products to protect timber and there is an extensive literature exploring the potential for delivering extracts from one plant or timber into a less durable material. In general, these efforts have failed to produce the results desired. These failures may be because the distribution in the original plant could not be replicated or the extracts were not universally effective against all possible agents of decay. In some cases, the extracts could not be easily delivered because of the solvents required. For example, attempts to use cinnamaldehyde from leaves to protect wood against stain and decay fungi were very successful when ethanol was used as the carrier, but attempts to produce water soluble or miscible solutions resulted in loss of activity.

One other aspect of using natural extracts is toxicity. There is an inherent belief that natural products are somehow safer, but many of the extractives in timber are effective because they are highly toxic. Attempts to use these products as timber protectants will invariably require testing that will likely show that compounds pose hazards in regular use. Despite these limitations, understanding the nature of extractive activity could provide models for creating effective compounds that lack the inherent toxicity of the parent molecules.

Supplemental preservative treatments

Initial preservative treatments can provide excellent long-term protection against fungi, insect or marine borer attack, but there are several instances where some form of supplemental treatment is necessary. Over time, the initial preservative treatment will slowly migrate from the wood, eventually reaching the point where the remaining chemical level is too low to inhibit fungal or insect attack. The depleted surface then becomes susceptible to surface decay, generally by soft rot fungi. Alternatively, checks can develop as the wood seasons in service. These checks can penetrate beyond the depth of the original treatment, allowing fungi and insects access to the untreated wood interior where they produce internal decay.

A number of chemicals have been developed to provide supplemental or remedial protection to wood in service. These chemicals can be divided into two broad areas, those protecting against surface degradation and those that can penetrate further into the wood to control internal decay.

External treatments: External preservatives are applied as liquids or pastes to enhance the residual protection. Liquid preservatives are most often applied when cuts or borings are made in preservative treated wood. The preservative provides a shallow barrier against damage. A variety of chemicals can be used for this purpose, although 2% copper naphthenate is most commonly used. Treatment of cuts or drill holes in treated wood is required within the North American treatment standards.

Pastes are generally used to protect the groundline zone of large wood structures from soft-rot attack. These formulations often include an oil-soluble chemical to provide surface protection and a water-soluble component that penetrates for a short distance into the wood to inhibit further fungal attack. Groundline treatment chemicals have included pentachlorophenol, copper naphthenate, creosote, sodium dichromate, sodium fluoride, and sodium octaborate tetrahydrate. Environmental concerns have resulted in a gradual shift to pastes containing copper naphthenate, copper carbonate or sodium borates. These treatments are extensively used in the electric utility industry to provide supplemental protection to the groundline zone of wood poles, particularly with southern pine or western redcedar. They are often applied when a structure is 20–25 years old and are then regularly reapplied at 10–15 year intervals to provide continuous protection.

Internal Treatments: The initial preservative treatment is largely confined to the sapwood. As a result, species with thin sapwood shells tend to have a high percentage of untreated heartwood. Checks that open as poles season in service expose this untreated wood to fungal and insect attack. This poses a major challenge because this wood is inherently resistant to fluid movement; however, a number of systems have been developed that move either by liquid or gaseous diffusion.

Internal treatments include void treatments, fumigants and water diffusible rods. Void treatments are applied by drilling holes into wood voids and pouring or forcing a set quantity of chemical into the void. A variety of compounds have been used as void treatments including creosote, pentachlorophenol, chlordane, chlorpyrifos, sodium fluoride, and potassium dichromate. Regulatory changes have resulted in a sharp decrease in the materials used for this purpose. Void treatments currently include oilborne preservatives such as copper naphthenate, water diffusible borates and

insecticides. Void treatments are presumed to function by producing a barrier in the void to prevent further insect or fungal attack, but their effectiveness has never been fully proven.

Fumigants are agricultural chemicals that are applied as liquids through steep angled holes drilled into the wood. The holes are plugged and the chemical volatilizes to move throughout the wood as a gas. Fumigant movement through wood up to 4 m from the point of application has been reported (Helsing et al., 1984). Four fumigants are registered for decay control; chloropicrin (96% trichloronitromethane), metham sodium (32.1% sodium n-methyldithiocarbamate), methylisothiocyanate (96% MITC in an aluminum tube) and dazomet (3,5-Dimethyl-1,3,5-thiadiazinane-2-thione). Fumigants are highly effective fungicides that rapidly diffuse through the wood to eliminate most decay fungi within one year after application and provide protection against renewed invasion for periods ranging from 7 to 20 years (Zabel et al., 1982; Helsing et al., 1984; Morrell, 1989). Despite their excellent performance, fumigants are only effective in ground contact when there is an existing preservative shell to protect the wood surface (Morrell et al., 1986). Fumigants are widely used in North America to protect wood utility poles, bridge timbers, laminated beams, and marine piling.

Fumigants are highly effective, but their volatility and high toxicity have led some users to question their safety. Borates are an alternative to fumigant treatment. Boron can be dispersed in glycol and applied to treatment holes much in the same as the fumigants. The boron can then diffuse outward from the hole. Boron can also be heated to a molten state and then formed into water soluble, glass-like rods for application. The rods are inserted in the same holes used for fumigant application. Once applied, moisture in the wood solubilizes the boron, which moves across and down through the wood. Although not yet widely used in North America, fused borate rods are widely used in Europe for controlling decay in above ground structures. They tend to take a little longer to reach effective levels in the wood, but once there, remain for many years after treatment.

Non-chemical methods for improving wood performance

Wood varies widely in its degree of treatability. For example, sapwood is generally treatable while heartwood, because of the high

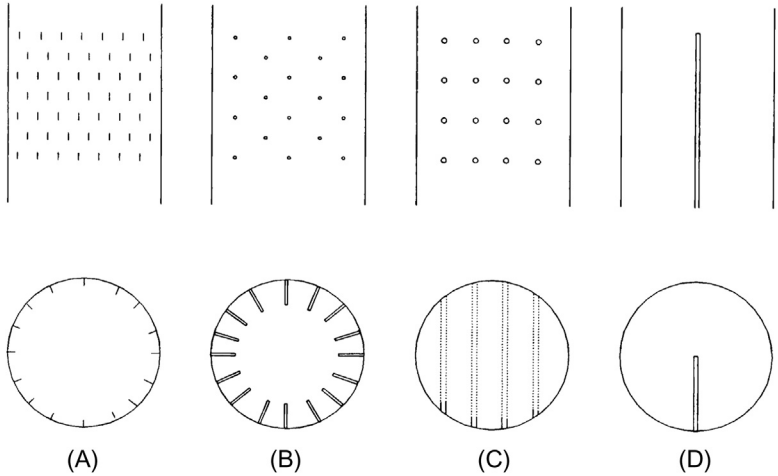


Figure 19.5 (A–D) Kerfing, incising, radial drilling and through boring are all used to improve preservative penetration into refractory (difficult to treat) species.

percentage of aspirated pits, tyloses, or extractives is difficult to treat. In addition, geographic source has a marked effect on the treatability of certain woods. For example, Douglas-fir from the coastal regions of the pacific Northwest is far easier to treat than the same species grown further inland. As a result of the treatment variables, a number of preparative steps have been developed to improve the treatment of wood, and, thus, enhance performance (Graham et al., 1969; Helsing and Graham, 1976; Morrell, 2012) (Fig. 19.5).

Incising uses steel teeth to punch numerous small holes into the wood surface, increasing the amount of end-grain exposed to the preservative solution, improving the depth and uniformity of preservative treatment. Incising is required for treatment of many thin sapwood species.

Through-boring and radial drilling are also used to increase the amount of cross section exposed to treatment in decay sensitive zones and are extensively used to improve treatment in the groundline zone of electric utility poles (Graham et al, 1969). Through boring results in nearly complete preservative impregnation of the drilled area, virtually eliminating the risk of decay in that zone. Radial drilling results in a deep shell of treatment sufficient to support the pole even if the core decays.

Kerfing is a different approach to improving the performance of large timbers and poles. A saw kerf is cut to the pith center, normally from the pole base to about the groundline zone and acts to relieve normal wood

drying stresses. The kerf opens and closes seasonally as the pole wets and dries, but the risk of decay in this zone is reduced because the kerf is preservative treated, sharply reducing the risk of fungal attack (Helsing and Graham, 1976). However, kerfing does not prevent decay fungi from invading through checks further up the pole.

In addition to specific pretreatments, simple processes including drying the wood prior to treatment, cutting the wood to length and drilling all holes prior to treatment can ensure that the envelope of treatment remains intact. These processes are covered in detail elsewhere (Nicholas, 1973; Hunt and Garrett, 1967; Wilkinson, 1979).

Environmental considerations

Within the last decade or so, environmental considerations have become a major preoccupation for commercial wood treaters and major users of treated wood products. This is particularly true of users of PCP, inorganic arsenicals, and creosote, since these chemicals are now restricted-use pesticides. In 1977, the US EPA listed these 3 chemicals for potential revocation of their pesticide registrations. This process, called "rebuttable presumption against registration and reregistration" (RPAR), reviews supporting data for each pesticide provided by industry and the major users to determine if the benefits of registration outweigh the risk of continued chemical usage. In this process, the 3 chemicals were declared restricted-use pesticides and users must now be licensed by the state. In addition, the wastes generated during the production of these chemicals and the subsequent treatment process are now more heavily regulated (Wise, 1986). The latter development has resulted in considerable expense to established treaters who must control existing waste problems while dealing with environmental contamination that resulted from previous handling practices. This damage means that some treating plants would qualify as EPA toxic waste sites if they were closed. Some plants estimate that complying with environmental regulations costs them about 1 million dollars per year.

The increased environmental restrictions have stimulated a search for safer, more environmentally acceptable wood preservatives. This effort was spearheaded by the Electric Power Research Institute (EPRI) through

research at Michigan Technological University's Institute of Wood Research (Preston, 1986; Preston et al., 1983). Unfortunately, the process of preservative development takes many years and none of the potential replacement chemicals identified in these tests are widely used in industrial applications. The immediate need to substitute safer chemicals has also stimulated renewed interest in previously identified chemicals such as copper naphthenate, Cu-8, and TBTO. Of these, copper naphthenate is the most widely used, but even it is still only used to a limited extent.

Looming problems are the safe disposal of used treated wood products, soil contamination in sensitive sites and leaching of chemical into surface waters. As a result, chemicals that are currently acceptable may find themselves in the same position as PCP, creosote or inorganic arsenicals. This possibility emphasizes the need to continue developing and testing new preservatives.

Wood preservative development and testing

Unlike agricultural chemicals, which need only protect a plant from attack by a limited number of pathogens for a short time, preservatives must protect wood against a wide array of organisms for periods of 30 years or more. Therefore, the process for developing a new wood preservative is considerably more involved and time consuming.

Since field trials of a new preservative would require decades, a number of accelerated, short-term tests have been developed as a guide to long term performance (Behr, 1973; Baines, 1984; Fahlstrom, 1975; Halabisky and Ifju, 1968; Duncan, 1958; McKaig, 1986; Smith, 1967). An interesting history of the developments in the evaluation of wood preservatives and of accelerated tests was assembled by Colley (1953).

Petri plate tests: Most chemicals are initially tested for their ability to control decay fungi in petri plate tests (Richards, 1923). This test method varies widely between laboratories. It can include exposing the test fungus to a media containing known concentrations of the test chemical, or soaking filter paper in the test chemical and placing this on the surface of a previously inoculated plate. The presence of the fungus and its growth rate are used as a measure of chemical effectiveness. These results are then compared with results of similar tests with accepted preservatives.

While this method provides a relative measure of toxicity, the growth of fungi on artificial media is markedly different from growth in wood. For example, the addition of excess sugar can sometimes allow a fungus to overcome the protective value of some preservatives. In addition, some chemicals that have low water solubility or those that are strongly fixed to wood cannot migrate into the media to inhibit the test fungus (Dost and Scheffer, 1983). As a result, these chemicals will not perform well in Petri dish tests, even though they may be very effective wood preservatives. Also, the short-term toxicity of volatiles can give misleading information on the long-term activity of a compound. Despite these short-comings, Petri dish tests provide a simple method for rapidly screening a range of potential preservative candidates.

Soil block test: A second, and more common method for testing preservative involves the soil-block test developed initially by Leutritz (1946) and later modified and improved (Duncan, 1953, 1958; American Wood Protection Association, 2017). In this method, a block (typically a 19 mm cube) is treated with the test chemical at a given solution strength to produce a desired target retention. After weighing to determine the amount of chemical absorbed, the blocks are dried, reweighed, and exposed to one of 4 selected test fungi (*Rhodonia placenta*, *Trametes versicolor*, *Neolentinus* (*Lentinus*) *lepideus*, or *Gloeophyllum trabeum*) which were previously inoculated onto wood wafers on top of a sterile moist soil bed in a bottle. The blocks are exposed to the test fungus for 3–6 months, depending on the fungus, then removed from the bottles, oven-dried and reweighed. This final weight is compared to the initial treated dry weight to determine the weight loss due to fungal exposure. The fungi used in this test were selected because of their known tolerances to specific toxins; *R. placenta* has a high tolerance to copper, *T. versicolor* is tolerant to arsenic, *G. trabeum* is tolerant to PCP, and *N. lepideus* is resistant to creosote. All but *T. versicolor* are brown rots, reflecting the higher incidence of this decay type in structural decay. The results following exposure of the treated blocks are compared to those obtained following exposure of untreated blocks to the same test fungi and similarly treated blocks not exposed to the test fungi. Blocks treated with effective levels of a known wood preservative are included for comparison. The chemical retention in the block is plotted against the wood weight loss and the point where weight loss due to test variables intersects with weight loss from fungal attack is termed the threshold (Fig. 19.6). This level can then be compared to thresholds for known chemicals and can be used to develop

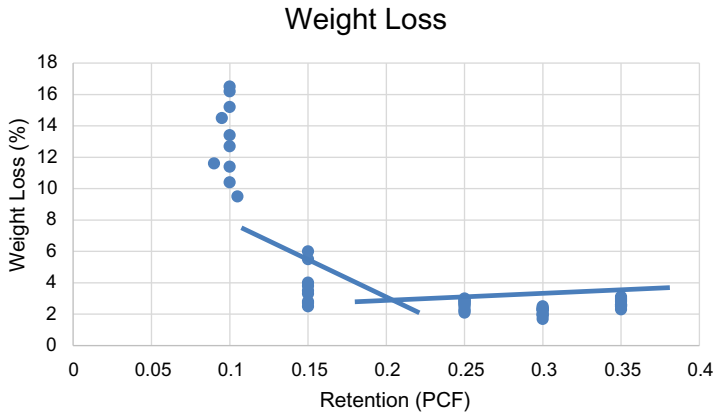


Figure 19.6 The relationship between chemical loading and wood weight loss when exposed to decay fungi in a soil block test can be used to calculate a threshold for limiting fungal attack. The intersection of the two lines is the approximate threshold for protection against attack by this test fungus as described in AWPA Standard E10.

target retentions for field tests. Smaller blocks can be used to shorten the test time, but care must be taken since chemical losses will tend to increase sharply with increasing wood surface to volume ratios.

Information on the stability (or resistance to leaching) of the preservative being tested can also be gained by exposing a matching set of test blocks to various leaching and weathering exposures. There are many variations on the soil block test. The European decay block test (EN113) uses malt extract agar in place of the soil, but essentially functions on the same premise; using weight loss as a measure of protection. There remain, however, some questions concerning the potential effects of sugars present in the media on decay rates.

Soil block tests provide a relative measure of preservative performance against white and brown fungi, but does not test the resistance to soft rot organisms which are known to be more resistant to wood preservatives. At present, there is no standard test for resistance to soft rot attack, although the use of soil or vermiculite burial tests which increase the wood moisture content to levels typical for soft rot attack appear most popular (Nilsson, 1973; Zabel et al., 1985). Soil block tests provide a relative guide to decay resistance, but this method cannot adequately represent the associations between various fungi in a natural environment.

The disadvantage of the soil block test is that it takes 3–6 months to complete. At the end of the test, mass losses in some blocks will reach

60–70% for brown rot fungi and well over that figure for an aggressive white rot fungus. While useful, we know that wood properties decrease sharply at much lower levels of decay suggesting that other methods for assessing the effects of decay might be more useful. Nicholas has performed extensive studies using longitudinal or radial compression on small wood blocks to predict decay rates and has also explored small scale bending tests for this purpose (Janzen and Nicholas, 2002; Li et al., 2006, 2009; Nicholas and Jin, 1996; Nicholas et al., 1991). While these methods are extremely sensitive and provide more rapid results and have been standardized AWPA Standards E22 and E23, they are only sparingly used (American Wood Protection Association, 2017).

Decay or Fungal cellars: The fungal cellar is another accelerated method for testing fungal associations without establishing field tests in tropical countries. In this method, small wood stakes ($12 \times 25 \times 150$ mm long) are treated to selected retentions with the test chemical and exposed in a soil bed which is usually situated in a greenhouse. The temperature in this facility is usually maintained at or above 28 °C and the soil moisture levels are kept high. The soil conditions can be manipulated by adding organic matter and the moisture conditions are often maintained at high levels to foster the development of soft rot attack. Most Fungal Cellars create conditions simulating tropical conditions and can produce rapid degradation within one year (Preston, 1986).

Laboratory Termite tests: In addition to fungal attack, preservatives must protect against insects. Most chemicals are tested against termites since they cause the most widespread insect-related wood damage. Termite protection can be evaluated in the field or under laboratory conditions. In a AWPA Standard E1 laboratory test, a set number of termite workers is placed in a chamber with the pre-weighed, treated wood for a given length of time (30 days) (American Wood Protection Association, 2017). In some cases, the termite workers are given a choice of untreated or treated wood. At the conclusion of the test, the number of living workers are counted, the block is rated for degree of damage, and the wood is reweighed to determine the weight loss due to termite attack. These results are compared to similar tests using untreated wood or wood treated with accepted wood preservatives. These tests can also be performed with certain beetles. In general, laboratory insect evaluations are difficult and there are many questions concerning the validity of these procedures.

Figure 19.7 A section of a large field installation of treated pole-diameter posts evaluating various wood species and treatment methods. The test site is located near Corvallis, Oregon.

Field stake tests: One of the first tests used to evaluate new chemicals to be used in soil contact is the stake test. In the U.S., the American Wood Protection Association Standard E7 typically uses 19 by 19 mm by 450 mm long sapwood stakes of an easily treated species such as southern pine that are treated to a given loading of the test chemical as well as a standard reference preservative (Fig. 19.7). European methods use slightly larger stakes (25 by 50 by 500 mm long) of Scots pine sapwood (*Pinus sylvestris*) while older trials by the U.S. Forest Products Laboratory used nominal 2 in by 4 in. (50 by 100 mm) lumber. The stakes are buried to half their length in soil at a field test site. Most preservative development efforts involve multiple test sites with varying degrees of fungal and termite attack.

At each inspection, the stakes are pulled from the ground and visually rated from 10 (sound) to 0 (destroyed) for both termite attack and fungal deterioration. The results of the test chemicals are compared to those obtained with other stakes without treatment or treated with an accepted wood preservative. In this method, the performance of stakes treated with lower retention of the test chemical can be used as indicators of future performance of the more heavily treated test chemicals (Colley, 1984; Hartford and Colley, 1984). Stake tests can last from 3 to 20 years depending on the test site and chemical retentions used. The most extensive stake tests in the United States are operated by the U.S. Forest Products Laboratory in Gulfport, Louisiana and evaluate a variety of

pressure, dip and brush-on treatment (Blew, 1948; Crawford et al., 2002). Many companies also operate field test sites and one of largest is at the Austin Carey Forest operated by the University of Florida near Gainesville.

Wood assembly tests: A variety of tests have been developed to evaluate the performance of preservatives when wood is not in direct ground contact. These tests simulate door frames or joinery, where the decay hazard is far lower than in ground contact. For decades, the most common above ground test used an L-joint design with a tight, water-trapping joint to accelerate decay. The L-joint test uses mortise and tenon pieces to create a water trapping joint that simulates a window frame. These L-joints are exposed out of ground contact. Painting the joint and then cracking it to create a pathway for water to enter further accelerates the decay process.

The extent of decay in these tests is evaluated in a manner similar to that used in stake testing, although the test takes longer to produce substantial decay. To overcome this problem, some laboratories have removed small samples from test members at regular intervals to determine changes in wood permeability related to fungal attack or have cultured the wood to determine if fungal colonization has occurred. This allows the early effects of fungal attack (increased permeability) to be detected before advanced decay becomes visible. A more common approach to accelerated testing is to expose materials under more aggressive tropical or sub-tropical conditions. Most above ground preservatives for the North American market are evaluated near Hilo, Hawaii, which has year-round warm temperatures and receives almost 5 m of rainfall per year. Decay of untreated pine specimens occurs within 18–24 months under these conditions.

Changes in wood preservative use have also encouraged the development of an array of alternative non-soil contact tests. The ground proximity test exposes small blocks (19 by 50 by 125 mm long) on concrete blocks under a permeable shade cloth that allows rainfall to enter, but reduces ultraviolet light damage. The samples are visually assessed for decay on a regular basis. This test is most aggressive in warm, wet climates.

Similarly, the ground proximity termite test exposes similar-sized blocks to potential termite attack under a cover that prevents wetting and simulates a dry crawl space. Termite tests tend to be specifically designed to take advantage of the behavior of a given species. For example,

the ground proximity test works best with Formosan termite who tend to more aggressively explore upwards for wood.

Samples are also exposed in various sandwich or stack tests that create avenues for moisture entry and trapping. The variety of test methodologies reflects the imagination of the researchers, who are all trying to create realistic accelerated exposures

Marine borer tests: While most testing methodology involves protecting wood from fungi and insects, some chemicals may also be promising marine borer control agents. Testing these chemicals against terrestrial decay agents provides little information on marine performance. Although most marine testing is done in the ocean, potential marine biocides can also be tested under controlled laboratory conditions to determine the toxicity to certain marine borers. This task is difficult due to the need to maintain marine conditions. Ocean testing involves the exposure of small wood samples treated to selected retentions of the test chemical at sites where marine borer hazards have been previously documented (American Wood Protection Association, 2017; Roe and Hochman, 1959; Johnson and Gutzmer, 1984). The specimens are attached to racks that can be easily retrieved for inspection. At each inspection, the specimens are rated for the degree and type of marine borer attack. Where appropriate, the specimens can be x-rayed to detect internal shipworm attack. Although the AWPA standard for the testing of wood preservatives in the marine environment list 2 specimen sizes, a variety of test dimensions are employed ranging from thin (6 mm thick) coupons to full-size test piling. Very small specimens are easily handled and produce rapid results, but they may not truly represent exposure conditions because they expose a large surface area relative to the wood volume and expose higher amounts of end grain that will lose chemical more rapidly. The use of larger panels (19 by 76 by 460 mm long) can overcome many of the limitations of small sample size and are most commonly used for marine tests. Large specimens are difficult to handle, expensive to install, require diving inspections that may be less accurate and take longer to test.

Potential wood preservatives must also be evaluated for possible environmental hazards. Ideally, the preservative should have low mammalian toxicity, be safe to handle, be non-toxic to fish and be easily disposed. In fact, most chemicals do not fit the ideal profile, but proper testing can identify the potential risks and allow registration of the chemical to be based

upon sound scientific knowledge, and not conjecture or public pressure. It is not within the scope of this chapter to discuss environmental testing.

Preservative resistance (tolerance)

There are large variations in the capacity of many microorganisms to resist or tolerate exposure to toxicants. Resistance and tolerance to toxicants are used synonymously in the literature. The development of resistance by bacteria to antibiotics (e.g. penicillin, streptomycin) and insects or fungi to pesticides (e.g. D.D.T. and benomyl) is a serious problem in medicine and agriculture, respectively.

Resistance arises both by selection among the various genotypes comprising a species and by occasional mutations. Favorable mutations can rapidly accumulate in bacterial and fungal populations that produce huge numbers of propagules that have short life cycles. Resistance most commonly occurs with heavy usage of chemicals with a single site for toxic action or in cases where the threshold between treatment levels and toxic levels to the microorganism is narrow. Resistance is less common in the older inorganic pesticides that have toxic effects at multiple sites and where the treatments of surfaces or inert materials (wood) permit the usage of higher concentrations. While most wood preservatives can protect wood for many years against a wide array of fungi, insects, and marine borers, a limited number of organisms have either high tolerances to some of these chemicals (probably because they are already resistant to similar compounds in nature) or in a few cases, evolved the ability to degrade the compound. Some of the reported tolerances of several decay and other common wood-inhabiting fungi to wood preservatives or their toxic components are listed in Table 19.2. Information on the tolerances of fungi to chemicals has several important uses. Tolerant fungi are often selected to increase the rigor of tests for evaluating new wood preservatives or treatments. Selection of a preservative and its retention for a wood product in some special uses must also consider the common decay fungi and their tolerances. A potentially valuable use of the tolerant fungi is the degradation of the preservatives in toxic sites (Madhosingh, 1961; Duncan and Deverall, 1964; Cserjesi, 1967). Such fungi are promising candidates for disposal of wastes generated during the synthesis and

Table 19.2 Examples of organisms with tolerance to preservatives.

Preservative	Fungus	Role in wood	References
Creosote	*Neolentinus lepideus*	Brown rot	Zabel (1954)
	Irpex lacteus	White rot	Cowling (1957)
	Antrodia radiculosa	Brown rot	Hudson (1952)
	Hormconis resinae	Stainer	Marsden (1954)
	Paecilomyces variotii	Soft rot	Kerner-Gang (1976)
Pentachlorophenol	*Gloeophyllum trabeum*	Brown rot	Zabel (1954)
	Irpex lacteus	White rot	Duncan (1953)
	Chaetomium globosum	Soft rot	Savory (1955)
	Cephaloascus fragrans	Stainer	Cserjesi (1967)
	Meruliporia incrassata	Brown rot	DaCosta and Kerruish (1964)
Copper naphthenate	*Meruliporia incrassata*	Brown rot	Cowling (1957)
	Rhodonia placenta	Brown rot	Zabel (1954)
	Fomitopsis cajanderi	Brown rot	Cowling (1957)
	Wulfiporia cocos	Brown rot	Cowling (1957)
Chromated copper arsenate	*Meruliporia incrassata*	Brown rot	Cowling (1957)
	Phialophora malorum	Soft rot	Daniel and Nilsson (1988)
Arsenic	*Gloeophyllum trabeum*	Brown rot	Cartwright and Findlay (1958)
Chromated zinc chloride	*Coniophora puteana*	Brown rot	Cartwright and Findlay (1958)
Sodium fluoride	*Trichoderma* spp.	Mold	Verrall (1949)
Phenyl mercury oleate	*Xylobolus frustulatus*	White rot	Cowling (1957)
Mercury	*Penicillium cyclopium*	Mold	Brown (1953)

application of these chemicals and could provide an environmentally sound approach for hazardous waste disposal (see Chapter Twenty). The methods by which many microbes detoxify chemicals remain poorly understood; however, a brief discussion of several of the detoxification agents can provide a guide to these processes.

Decomposition of creosote: Creosote is a highly complex mixture of organic chemicals, but several organisms have developed the ability to utilize this mixture as a sole carbon source, including the marine bacterium, *Pseudomonas creosotensis* (O'Neill et al., 1960; Drisko and O'Neill, 1966;

Stranks and Hulme, 1975; Seesman et al., 1977), and the imperfect fungus, *Hormocomis* (*Cladosporium*) *resinae*. Almost a third of the fungi colonizing creosoted wood have the ability to degrade creosote (Kerner-Gang, 1976).

One fungus, *H. resinae*, is the most commonly isolated fungus from creosoted southern pine utility poles (Zabel et al., 1982) and can be grown on creosote as a sole carbon source (Christensen et al., 1942; Marsden, 1954). Although this species commonly colonizes creosoted wood, it does not cause wood degradation and its effect on longevity of the treated wood remains uncertain. A number of other fungi exhibit tolerance to creosote, including the basidiomycete, *Neolentinus lepideus* , which is used in the soil block test. The soft rot fungus, *Paecilomyces variotii* and the yeast *Candida albicans* have also been shown to be capable of degrading creosote (Kerner-Gang, 1976).

The ability of microorganisms to degrade creosote has been employed in land farming schemes to detoxify waste products produced during the wood-treating process (Wise, 1986). Since creosote is virtually removed from the soil, this method offers considerable advantages over disposal in toxic waste sites. In the latter instance, the waste is only stored, and can be returned to the original producer at a later date.

Decomposition of pentachlorophenol: This chemical is a potent inhibitor of microbial oxidative phosphorylation, but a number of organisms have developed the ability to decompose PCP in soil. Savory (1955) was among the first to suggest that soft rot fungi were more resistant to PCP than basidiomycetes, comparing *Chaetomium globosum* and *Trametes versicolor*, respectively. Exposure of PCP treated blocks to *Trichoderma* sp has also been shown to increase weight losses caused by *Gloeophyllum trabeum*, suggesting some preservative modification or interaction between the two fungi (Duncan and Deverall, 1964). Similarly, *T. viride* and *Coniophora puteana* removed approximately 63% of the original PCP in treated red pine blocks (Unligil, 1968); however, other studies suggest that soil mineralization or immobilization of preservative might also account for some of these losses (Luetritz, 1965).

Several species, including *Trichoderma harzianum* and *T. virgatum* have been shown to degrade PCP, generally by methylation of PCP to pentachloroanisole (Cserjesi, 1967; Cserjesi and Johnson, 1972). The latter compound is much less toxic than PCP and a number of other species were subsequently shown to utilize this method of PCP detoxification (Stranks and Hulme, 1975).

In addition to fungi, bacteria may also play a role in PCP detoxification in soil contact, and a number of isolates have been shown to use PCP as a sole carbon source (Chu and Kirsch, 1972). In some cases, the normal soil microflora can significantly reduce the levels of labeled PCP in as little as 24 hours (Kirsch and Etzel, 1973).

The use of bacterial degradation has been proposed as one method for eliminating PCP from contaminated soil (Edgehill and Finn, 1983; Pignatello et al., 1983). Several systems are currently available for this process (Wise, 1986).

As expected in such a complex system, the environmental conditions can play an important role in the degree and rate of decomposition. Several studies suggest that the addition of exogenous nutrients can increase the preservative resistance of some fungi (Schmidt and Ziemer, 1976), but may slow degradation by others (Bumpus et al., 1985). Thus, each system must be carefully analyzed before attempts are made to use biological degradation to remove toxic wastes.

Decomposition of inorganic arsenicals: Unlike creosote and penta, the inorganic arsenicals such as CCA pose a different problem for organisms attempting to utilize treated wood. Generally, these chemicals have strong interactions with the wood and may not be as accessible to microbial activity. Furthermore, the lack of carbon in these formulations makes these compounds less useful as an energy source. Since the organism cannot use these chemicals directly as an energy source, it must develop methods for inactivating them or enhance alternative physiologic pathways that avoid the toxicant. The former method requires far less "effort" and is the more common method for detoxifying the inorganic arsenicals.

The literature on inorganic arsenical resistance by wood decay fungi is limited (DaCosta, 1972; Madhosingh, 1961; Chou et al., 1973; Young, 1961). Examination of CCA treated pine blocks colonized by *Rhodonia placenta*, a copper tolerant fungus, indicated that the hyphae absorbed an average of 3–4% of the total metal ions on a mycelial dry weight basis in a pattern that closely followed the original concentrations of copper, chromium or arsenic. In some instances, no copper or arsenic could be detected in the wood cell wall, suggesting complete removal by the test fungi (Chou et al., 1973). Some fungi are inherently copper tolerant. For example, *Wulfiporia cocos* is a naturally copper tolerant basidiomycete that immobilizes copper by over-production of oxalic acid that reacts to form less toxic copper oxalate (Archer et al., 1990; Morrell, 1991).

In general, the strong wood/chemical interactions coupled with low energy value of the inorganic arsenicals probably limits the number of organisms capable of degrading these compounds, although a number have or have developed tolerances to high levels.

Summary

1. The early use of creosote (by Moll in 1836) and the development of pressure treatments (by Bethell around 1839) for the preservation of wood preceded, by nearly 50 years, Hartig's discovery that fungi caused wood decay.
2. Critical steps in selecting the preservative and treatment type necessary for a wood product are estimating the decay hazard, judging the length of time the wood must be protected, and weighing the relative financial merits of various treatments.
3. Short-term treatments, involving the dipping or spraying fungicides onto lumber or log surfaces, are available to protect logs and lumber from sapstains and early decay during storage or lumber seasoning.
4. Long-term treatments are necessary for wood used when conditions conducive to decay prevail. These treatments are often subdivided into non-pressure and pressure treatments. The non-pressure treatments include dipping, brushing, spraying, and soaking the wood in a preservative. These methods provide only a thin shell of treated wood and provide useful protection to wood exposed above ground to intermittent wetting. Most surface treatments are of minimal value to wood in ground contact and not recommended. Pressure treatments are necessary to protect wood exposed to decay conditions in costly structures.
5. Pressure treatments use combinations of pressure, vacuum, and sometimes elevated temperatures to force the preservative solutions deep into the wood. Long-term protection requires that the penetration exceed the depth of subsequent check formation.
6. The major wood preservatives are oilborne: creosote, pentachlorophenol, copper naphthenate, copper-8-quinolinolate, and water soluble: alkaline copper compounds, micronized copper azoles, chromated copper arsenate, borates, and ammoniacal copper zinc arsenate.

7. Remedial treatments such as groundline pastes, void treatments or fumigants are useful for arresting decay that occurs on treated wood in service.
8. Environmental considerations have led the EPA to place creosote, pentachlorophenol, and arsenical compounds in a restricted use category. This has resulted in a major search for new wood preservatives.
9. Wood preservatives are tested and compared with other preservatives by petri plate, soil block tests and the exposure of treated stakes in decay cellars, field stake tests, or wood assembly tests.
10. Tolerance or resistance to wood preservatives exists among some common decay fungi. Resistant strains of decay fungi are generally used as test organisms in the evaluation of new wood preservatives. Information on the tolerant decay fungi associated with a specific wood application is useful in decisions on what preservative to use and the retention level.
11. Some tolerant or resistant fungi are able to degrade the preservative. This group may eventually provide an economical and environmentally acceptable way to dispose of some toxic wastes.
12. Major research needs are the development of effective, economical, and environmentally acceptable wood preservatives and exploring biological control possibilities to insure the future protection of the enormous amounts of wood that must be used under conditions conducive to decay.

References

Amburgey, T.L., Sanders, M.G., 2009. Tie dual treatments with Timbor and creosote or copper naphthenate. Crossties Nov/Dec 20–21.

Amburgey, T.L., Sanders, M.G., 2007. Borate pre-treated crossties... a 20-year field test. Proc. Am. Wood Prot. Assoc. 103, 109–111.

American Wood Protection Association. 2017. Annual Book of Standards. AWPA, Birmingham, Alabama.

Archer, K., van der Waals, J., Hedley, M., 1990. The comparative performance of "copper naphthenate" formulations in laboratory decay tests. Proc. Am. Wood Preserv. Assoc. 86, 78–95.

Arsenault, R.D., 1973. Factors influencing effectiveness of preservative systems. In: Nicholas, D.D. (Ed.), Wood Deterioration and its Prevention by Preservative Treatments. Syracuse University Press, Syracuse, NY, pp. 121–278.

Baines, E.F., 1984. Preservative evaluation using a soil bed and static bending strength to measure performance. Proc. Am. Wood Preserv. Assoc. 80, 67–79.

Behr, E.A., 1973. Decay test methods. In: Nicholas, D.D. (Ed.), Wood Deterioration and its Prevention y Preservative Treatments. Syracuse University Press, Syracuse, N.Y, pp. 217–246.

Blew, J.O., 1948. Comparison of wood preservatives in stake tests. Proc. Am. Wood Preserv. Assoc. 43, 88–119.
Brown, F.L., 1953. Mercury tolerant *Penicillia* causing discoloration in Northern White Pine Lumber. J. For. Products Res. Soc. 3 (4), 67–69.
Bumpus, J.A., Tien, M., Wright, D., Aust, S.D., 1985. Oxidation of persistent environmental pollutants by a white rot fungus. Science 228, 1434–1436.
Cappellazzi, J., Konkler, M.J., Morrell, J.J., 2019. Boron Dual-Treatments for Douglas-fir Utility Poles: Tracking Boron Migration Over Time. Document No. IRG/WP/19-3073. International Research Group on Wood Protection Stockholm, Sweden.
Cartwright St., K.G., Findlay, W.P.K., 1958. Decay of timber and its prevention. Her Majesty's Stationary Office, London.
Colley, R.H., 1953. The evaluation of wood preservatives. Bell Syst. Tech. J. 32, 120-169 and 425 505.
Chou, C.K., Chandler, J.A., Preston, R.D., 1973. Uptake of metal toxicants by fungal hyphae colonizing CCA-impregnated wood. Wood Sci. Technol. 7, 206–211.
Christensen, C.M., Kaufert, K.H., Schmitz, H., Allison, J.L., 1942. *Hormodendrum resinae* (L) an inhabitant of wood impregnated with creosote and coal tar. Am. J. Bot. 29, 542–558.
Chu, J.P., Kirsch, E.J., 1972. Metabolism of pentachlorophenol by an axenic bacterial culture. Appl. Microbiol. 23, 1033–1035.
Colley, R.H., 1984. What is log probability index. Proc. Am. Wood Preserv. Assoc. 80 (247), 256.
Cowling, E.B., 1957. The relative preservative tolerances of 18 wood-destroying fungi. For. Prod. J. 7 (10), 355–359.
Cookson, L.J., Greaves, H., 1986. Comparative bioassays of two high temperature and two low temperature-derived creosotes. Holzforschung 40 (1), 59–64.
Crawford, D.M., Woodward, B.M., Hatfield, C.A., 2002. Comparison of Wood Preservatives in Stake Tests—2000 Progress Report. U.S.D.A. Forest Service Forest Products Laboratory Research Note FPL-RN-02, Madison, WI, p. 120.
Cserjesi, A.J., 1967. The adaptation of fungi to pentachlorophenol and its degradation. Can. J. Microbiol. 13 (3), 1243–1249.
Cserjesi, A.J., Johnson, E.L., 1972. Methylation of pentachlorophenol by *Trichoderma virgatum*. Can. J. Microbiol. 18 (1), 45–49.
DaCosta, E.W.B., Kerruish, R.M., 1964. Tolerance of *Poria* sp. to copper based wood preservatives. For. Prod. J. 14, 106–112.
DaCosta, E.W.B., 1972. Variation in the toxicity of arsenic compounds to microorganisms and the suppression of the inhibitory effects by phosphate. Appl. Microbiol. 23, 46–53.
Daniel, G.F., Nilsson, T., 1988. Studies on preservative tolerant species. Int. Biodeterior. 24, 327–335.
Dost, W.A., Scheffer, T.C., 1983. *Aspergillus* bioassay of wood treated with waterborne arsenical salts. Mater. Org. 18 (2), 135–140.
Drisko, R.W., O'Neill, T.B., 1966. Microbial metabolism of creosote. For. Prod. J. 16, 31–34.
Duncan, C.G., 1953. Soil-Block and Agar-Block Techniques for Evaluating Oil-Type Preservatives: Creosote, Copper Napththenate and Pentachlorophenol. Division of Forest Pathology, Bureau of Pl ant Industry, Forest Products Laboratory, Madison, Wis, Special Release No. 37.
Duncan, C.G., 1958. Studies of Methodology of Soil-Block Testing. U.S. Forest Products Laboratory Report 2119. Forest Products Laboratory, Madison, WI.
Duncan, C.G., Deverall, F.J., 1964. Degradation of wood preservatives by fungi. Appl. Microbiol. 12 (1), 57–62.

Edgehill, R.U., Finn, R.K., 1983. Microbial treatment of soil to remove pentachlorophenol. Appl. Environ. Microbiol. 45 (3), 1122–1125.
Fahlstrom, G.B., 1975. A small stake test procedure for accelerated evaluation of wood preservatives. Proc. Am. Wood Preserv. Assoc. 71, 216–220.
Graham, R.D., Miller, D.J., Kunesh, R.H., 1969. Pressure treatment and strength of deeply perforated Pacific Coast Douglas-fir poles. Proc. Am. Wood Preserv. Assoc. 65, 234–242.
Halabisky, D.O., Ifju, G., 1968. Use of respirometry for fast and accurate evaluation of wood preservatives. Proc. Am. Wood Preserv. Assoc. 64, 215–223.
Hartford, W.H., 1986. The practical chemistry of CCA in service. Proc. Am. Wood Preserv. Assoc. 82, 28–43.
Hartford, W.H., Colley, R.H., 1984. The rationale of preservative evaluation by field testing and mathematical modeling. Proc. Am. Wood Preserv. Assoc. 80, 257–269.
Helsing, G.G., Graham, R.D., 1976. Saw kerfs reduce checking and prevent internal decay in pressure-treated Douglas-fir poles. Holzforschung 30 (6), 184–186.
Helsing, G.G., Morrell, J., Graham, R.D., 1984. Evaluations of fumigants for control of internal decay in pressure-treated Douglas-fir poles and piles. Holzforschung 38 (5), 277–280.
Hill, C., 2006. Wood Modification: Chemical, Thermal and Other Processes. Wiley Series in Renewable Resources. J. Wiley & Sons, Ltd.
Hudson, M.S., 1952. *Poria radiculosa*, a creosote tolerant organism. For. Prod. J. 2 (2), 73–74.
Graham, R.D., 1973. History of wood preservation. In: Nicholas, D.O. (Ed.), Wood Deterioration and its Prevention by Preservative Treatments. Syracuse University Press, Syracuse, N.Y, pp. 1–30.
Hunt, G.M., Garrett, G.A., 1967. Wood Preservation, third ed. McGraw-Hill, New York, N.Y.
Janzen, S., Nicholas, D.D., 2002. Use of Transverse Compression Properties as a Measurement of Wood Deterioration. Part 1. Effect of White Rot on Yellow Poplar. International Research Group on Wood Protection Document No. IRG/WP/02-40239, Stockholm, Sweden.
Johnson, B.R., Gutzmer, D.I., 1984. Marine Exposure of Preservative-Treated Small Wood Panels. USDA Forest Products Laboratory Research Note FPL-0248.
Kerner-Gang, W., 1976. Effects of micro-organisms on creosote. Intl. Symp. Berlin-Dahlem. Mat. Holz. 3, 319–330.
Kirsch, L.J., Etzel, J.E., 1973. Microbial decomposition of pentachlorophenol. J. Water Pollut. Control. Federat. 45, 359–363.
Krzyzewski, J., 1986. Formulating an arsenical creosote system—possible wood preservative. Proc. Am. Wood Preserv. Assoc. 82.
Lebow, S.T., Morrell, J.J., 1993. Pressure treatment of sitka spruce lumber with ammoniacal copper zinc arsenate or chromated copper arsenate. For. Prod. J. 43 (10), 41–44.
Lebow, S.T., Morrell, J.J., 1995. Interactions of ammoniacal copper zinc arsenate (ACZA) with Douglas-fir. Wood Fiber Sci. 27, 105–118.
Leutritz Jr., J., 1946. A wood-soil contact culture technique for laboratory study of wood-destroying fungi, wood decay, and wood preservation. Bell Syst. Tech. J. 25, 102–135.
Levi, M.P., 1973. Control methods. In: Nicholas, D.O. (Ed.), Wood Deterioration and its Prevention by Preservative Treatment. Syracuse University Press, Syracuse, N.Y., pp. 183–216.
Lew, J.D., Wilcox, W.W., 1981. The role of selected dueteromycetes in the soft-rot of wood treated with pentachlorophenol. Wood Fiber 13 (4), 252–264.

Li, G., Nicholas, D.D., Schultz, T.P., 2009. Evaluation of Variables That Affect Dynamic MOE in Wood Decay Studies. International Research Group on Wood Protection Document No. IRG/WP/09-20409, Stockholm, Sweden.
Li, G., Nicholas, D., Schultz, T., 2006. Effect of Decay on the Proportional Limit of Thin Wood Samples Stressed in the Bending Model. International Research Group on Wood Protection Document No. IRG/WP/06-20334, Stockholm, Sweden.
Lindgren, R.A., Scheffer, T.C., 1939. Effect of blue stain on the penetration of liquids into air-dry southern pine. Proc. Am. Wood Preserv. Assoc. 35, 325–336.
Luetritz, J., 1965. Biodegradability of pentachlorophenol as a possible source of depletion from treated wood. For. Prod. J. 15, 269–272.
Madhosingh, C., 1961. Tolerance of some fungi to a water-soluble preservative and its components. For. Prod. J. 11, 20–22.
Marsden, D.H., 1954. Studies on the creosote fungus, *Hormodendrum resinae*. Mycologia 46, 181–183.
McKaig, P.A., 1986. Factors Affecting Decay Rates in a Fungus Cellar II. International Research Group on Wood Preservation IRG/WP/2259, Stockholm, Sweden.
Militz, H., 1993. Treatment of timber with dimethylol resins to improve their dimensional stability and durability. Wood Sci. Technol. 27, 347–355.
Militz, H., Schaffert, S., Peters, B., Fitzgerald, C.J., 2011. Termite resistance of DMDHEU treated wood. Wood Sci. Technol. 45, 547–557.
Morrell, J.J., 2012. Wood Pole Maintenance Manual: 2012 Edition. Research Contribution 51. Forest Research Laboratory, Oregon State University, Corvallis, OR, 46 pages.
Morrell, J.J., 1991. Copper tolerant fungi: a brief review on their effects and distribution. Proc. Am. Wood Preserv. Assoc. 87, 265–270.
Morrell, J.J., 1989. The Use of Fumigants for Controlling Decay of Wood: A Review of their Efficacy and Safety. International Research Group on Wood Preservation IRG/WP/3525, Stockholm, Sweden.
Morrell, J.J., Freitag, C.M., Chen, H., 2005. Sequential treatments with fluoride and copper: effects of solution concentration and dipping time on treatment. For. Prod. J. 55 (7/8), 57–62.
Morrell, J.J., Miller, D.J., Schneider, P.F., 1999. Service Life of Treated and Untreated Fence Posts. 1996 Post-farm Report. Research Contribution 26. Forest Research Laboratory, Oregon State University, Corvallis, Oregon, 24 p.
Morrell, J.J., Schneider, P.F., Williams, R.S., 2001. Protecting wood decks from biodegradation and weathering: evaluation of deck finish systems. For. Prod. J. 51 (11/12), 27–32.
Morrell, J.J., Smith, S.M., Newbill, M.A., Graham, R.D., 1986. Reducing internal and external decay of untreated Douglas-fir poles: a field test. For. Prod. J. 36 (4), 47–52.
Nicholas, D.D. (Ed.), 1973. Wood deterioration and its prevention by preservative treatments (2 volumes). Syracuse University Press, Syracuse, NY.
Nicholas, D.D., Jin, Z., 1996. Use of Compression Strength Loss for Measuring Decay in a Soil Block Test. International Research Group on Wood Protection Document No. IRG/WP/96-20083, Stockholm, Sweden.
Nilsson, T., 1973. Studies on wood degradation and cellulolytic activity of microfungi. Stud. For. Suecica 104, Stockholm, 40pp.
O'Neill, T.B., Drisko, R.W., Hochman, H., 1960. A Creosote Tolerant Marine Bacterium. U.S. Naval Civil Engineering Laboratory Technical Note N-398, Port Hueneme, CA.
Pignatello, J.J., Martinson, M.M., Steiert, J.G., Carlson, R.E., Crawford, R.L., 1983. Biodegradation and photolysis of pentachlorophenol in artificial freshwater systems. Appl. Env. Microbiol. 46 (5), 1024–1031.

Preston, A.F., 1986. Development of new wood preservatives: the how and the what. In: Proceedings The Wood Pole Conference. Portland, Oregon, pp. 78–97.
Preston, A.F., McKaig, P., Walcheski, P.J., 1983. The use of laboratory, fungus cellar, and field tests in the development of wood preservatives. Proc. Am. Wood Preserv. Assoc. 79, 207–212.
Rao, K.R. (Ed.), 1977. Pentachlorophenol. Plenum Publishing Corp., New York.
Richards, C.A., 1923. Methods of testing relative toxicity at wood preservatives. Proc. Am. Wood Preserv. Assoc. 19, 127–130.
Roe, T., Hochman, H., 1959. Use of Small Panels in Screening Potential Wood Preservatives. Symposium on Treated Wood for Marine Use. Special Technical Publication 275. American Society for Testing Materials, Philadelphia, PA.
Rowell, R.M., 2005. Chapter 14. Chemical modification of wood. In: Rowell, R.M. (Ed.), Handbook of Wood Chemistry and Composites. CRC Press, Boca Raton, Florida, pp. 381–420.
Rowell, R.M., 2006. Chemical modification of wood: a short review. Wood Mater. Sci. Eng. 1, 29–33.
Savory, J.G., 1955. The role of microfungi in decomposition of wood. Rec. Ann. Conv. BWPA 1955.
Schmidt, O., Ziemer, B., 1976. Hemmstaff-Toleranz von Pilzen. Material und Organismen 11 (3), 215–230.
Seesman, P.A., Colwell, R.R., Zachary, A., 1977. Biodegradation of creosote/naphthalene treated wood in the marine environment. Proc. Am. Wood Preserv. Assoc. 73, 54–60.
Smith, R.S., 1967. Carbon dioxide evolution as a measure of attack by fungi and its application to testing wood preservatives and sapstain preventatives. Ann. Appl. Biol. 59, 473–479.
Smith, R.S., Byrne, A., Cserjesi, A.J., 1985. New Fungicidal Formulations Protect Canadian Lumber. Special Publication No. SP-25. Forintek Canada Corp., Vancouver, B.C.
Stamm, A.J., 1959. Dimensional stabilization of wood by thermal reactions and formaldehyde cross-linking. Tappi 42 (1), 39–44.
Stamm, A.J., Burr, H.K., Kline, A.A., 1946. Heat stabilized wood. Ind. Eng. Chem. 38 (6), 630–637.
Stamm, A.J., Seborg, R.M., 1943. Resin-treated Laminated Compressed Wood (Compreg). U.S. Forest Service, Forest Products Laboratory Report 1381, Madison, WI.
Stranks, D.W., Hulme, M.A., 1975. The mechanism of biodegradation of wood preservatives. Org. Holz. 3, 345–353.
Tarkow, H., Stamm, A.J., Erickson, E.C.O., 1946. Acetylated Wood. U.S. Forest Service, Forest Products Laboratory Report 1593, Madison, WI.
Unligil, H.H., 1968. Depletion of pentachlorophenol by fungi. For. Prod. J. 18, 45–50.
Van Groenou, H.B. (Ed.), 1951. Wood preservation during the last 50 years. AW Sijthoff, Leiden, Netherlands.
Verrall, A.F., 1949. Some molds on wood favored by certain toxicants. J. Agric. Res. 78 (12), 695–703.
Watkins, J.B., 1977. Timber preservation. Australian Patent No. 514897.
Webb, D.A., 1987. Creosote-Recent developments and environmental considerations. Proc. Am. Wood Preserv. Assoc. 83, 11–18.
Webb, D.A., 1980. Creosote, its biodegradation and environmental effects. Proc. Am. Wood Preserv. Assoc. 76, 65–69.

Westin, M., Lande, S., Schneider, M., 2004. Wood Furfurylation and Properties of Furfurylated Wood. International Research Group on Wood Protection Document No. IRG-WP/04-40289, Stockholm, Sweden.
Wilcox, W.W., 1987. Effectiveness of ammoniacal copper zinc arsenate wood preservative as indicated by a soil-block analysis. For. Prod. J. 37 (7/8), 62–63.
Wilkinson, J.G., 1979. Industrial Timber Preservation. Associated Business Press, London, United Kingdom, p. 532.
Nicholas, D.D., Wilson, J.A., Vasishth, R., 1991. Effect of Soft Rot Decay on the Static Bending Strength of Wood. International Research Group on Wood Protection Document No. IRG/WP/2361, Stockholm, Sweden.
Wise, E.D., 1986. RCRA surface impoundment options for creosote and pentachlorophenol wood treating facilities. Proc. Am. Wood Preserv. Assoc. 82, 13–19.
Young, G.Y., 1961. Copper Tolerance of Some Wood-Rotting Fungi. USDA Forest Products Laboratory Report 2223, Madison, Wisconsin.
Zabel, R.A., 1954. Variations in preservative tolerance of wood-destroying fungi. For. Prod. Res. Soc. 4, 166–169.
Zabel, R.A., Lombard, F.F., Wang, C.J.K., Terracina, F., 1985. Fungi associated with decay in treated southern pine utility poles in the eastern United States. Wood Fiber Sci. 17 (1), 75–91.
Zabel, R.A., Wang, C.J.K., Terracina, F.C., 1982. The fungal associates, detection and fumigant control of decay in treated southern pine poles. Report EL-2768. Electric Power Research Institute, Palo Alto, CA.

Further reading

Dekker, J., 1976. Acquired resistance to fungicides. Ann. Rev. Phytopathol. 14, 405–428.
Eslyn, W.E., 1976. Wood preservative degradation by marine bacteria. Proceedings of the 3rd International biodegradation Symposium. Applied Sciences Publishers, Ltd., London.
Esteves, B.M., Pereira, H.M., 2009. Wood modification by heat: a review. Bioresources 4 (1), 370–404.
Gjovik, L.R., Gutzmer, D.I., 1989. Comparison of Wood Preservatives in Stake Tests (1987 Progress Report). USDA Forest Products Laboratory. Research Note FPL-RN-02, Madison, Wisconsin.
Graham, R.D., Corden, M.E., 1980. Controlling Biological Deterioration of Wood with Volatile Chemicals. Final Report EL-1480, Research Project 212-1. Electric Power Research Institute.
Hartford, W.H., 1973. Chemical and physical properties of wood preservatives and wood-preservative systems. In: Nicholas, D.O. (Ed.), Wood Deterioration and its Prevention by Preservative Treatment. Syracuse University Press, Syracuse, N.Y, pp. 1–120.
Hirt, R.R., 1949. An isolate of *Poria xantha* on media containing copper. Phytopathology 39, 31–36.
Scheffer, T.C., 1964. Biological observations of significance for improved preservative treatment. Holzforschung 18 (3), 88–94.
Stanlake, G.J., Finn, R.K., 1982. Isolation and characterization of a pentachlorophenol degrading bacterium. Appl. Environ. Microbiol. 44 (6), 1421–1427.
Wang, C.J.K., Terracina, F.C., Zabel, R.A., 1989. Fumigant Effectiveness in Creosote and Penta Treated Southern Pine Poles. Final Report Electric Power Research Institute Report No. EPRI EL-6197, Palo Alto, CA.

CHAPTER TWENTY

Some trends in wood microbiology research and a new emphasis (biotechnology)

Contents

Like many of the biological sciences, wood microbiology is in a period of rapid change. Powerful new analytical tools, techniques and advances in other disciplines have opened up new insights and understandings on how wood-inhabiting microorganisms damage wood and how fungi function.

Wood colonizing microorganisms are generally considered to be pests requiring control, but these same agents are important in the recycling of carbon and nitrogen. Without this natural decomposition, large quantities of these organic elements would be permanently lost and we would soon be inundated in piles of debris.

Special attention has been placed on understanding the decay process as a major biologic event, considering the microecological factors affecting the colonization of wood by sequences of wood-inhabiting microorganisms, and considering the environmental impacts and alternatives to the traditional chemical approaches to decay control (Eriksson et al., 1990). Research continues to explore the potential useful roles fungi may play in the wood-using industries as effective biopulping or biobleaching agents,

Wood Microbiology.
DOI: https://doi.org/10.1016/B978-0-12-819465-2.00020-6

as efficient degraders or biotransformers of wastes and toxicants, or biological control agents against decay and stain fungi. Recent advances in biotechnology now offer the possibility of modifying fungal genotypes to carry out some of these roles more effectively.

In this final chapter, we first review some important societal changes affecting future wood supplies and then summarize some of the major research needs and opportunities that may lead to more effective and environmentally acceptable strategies for prevention and control of decays and discolorations in wood products. We close with a section on how biotechnology may impact wood using industries and make fungi useful, beneficial agents.

Changes in forests and wood supplies

We are currently in a period of changing wood supplies worldwide. Countries such as New Zealand, Chile, Canada and Russia have emerged as major exporters of timber. Many other countries, including the United States, are net timber importers. As the population continues to expand, the wise, efficient use of wood will become increasingly important. Wood is a major renewable industrial material for construction and serves as an important fuel source in many regions. Wood accounts for 25% of the value of the major industrial materials in the United States and demand is projected to increase by 25% by 2030 (National Research Council, 1990).

Changing views on forest values and shifts from timber production to multiple use management schemes that favor protection, recreation, and watershed uses in many areas will further reduce the timber base in a period of increasing demand for wood. By and large, old-growth native forests are increasingly protected on public lands, placing increasing emphasis on the management of plantation resources on private land. This process has led to the emergence of Chile and New Zealand as major timber exporters of non-native plantation species such as radiata pine and eucalyptus species. Log diameters will also decrease, leading to an increased proportion of juvenile wood in the resource creating challenges for those seeking to use timber. This material will be increasingly employed in modified wood products, such as particle board and laminated stock.

Reduced timber availability will place added emphasis on improved utilization of existing resources for a variety of applications, some of which remain to be developed. The eventual decline in petrochemical resources will place further pressure on the wood resource which remains our most important renewable fiber and chemical resource. Microorganisms can be employed in a number of processes to protect this resource from deterioration or to enhance recovery of various products from the material.

Fungal damage

Since many major industrial uses of wood involve its modification, understanding how fungi decay wood has great potential for developing more efficient processes and minimizing decay and stain damage.

Information on the chemical, physical and ultrastructural aspects of decay and the physiology of fungi are both basic to developing new decay control approaches as alternatives to current poisoning (chemical) treatments. Examples of potentially rewarding topics are:

a) Characterizing the small, diffusible oxidizing agent in brown rots that is responsible for the rapid depolymerization of crystalline cellulose so that its production could be blocked for wood protection or exploited for activities to make wood more nutritionally valuable.

b) Determining methods that disrupt the extracellular matrix surrounding the hyphal tips of decayers to potentially block enzyme transfer and the decay process.

c) Developing chemicals that inhibit cell wall synthesis of decay fungi. In essence, the triazoles already do this by inhibiting ergosterol synthesis.

d) Identifying critical biochemical pathways in protein synthesis by fungi that can be disrupted.

Fungi occupy an interesting space in that, as heterotrophs, they have some physiological activities similar to those of animals and this can make them difficult to control because any toxicants can also affect higher life forms. Conversely, differences in fungal cell wall structure and extracellular digestion processes make them vulnerable to other control strategies.

Many fungi can decay wood, however, only a few types have been studied thoroughly. New, novel methods by which fungi digest lignocellulose alone or in concert with other organisms may await discovery. There are thousands of decay fungi, yet our knowledge of the chemistry of decay rests on the study of only a few dozen species.

Many, if not most, laboratory studies of fungi are conducted in pure cultures. In nature, many microorganisms are involved sequentially in the colonization and alteration of wood during the decay process. Studying the microecological aspects of the decay process may reveal cohorts of organisms that more efficiently decay wood or sequences of organisms and interactions which suggest potent new biological control agents or ways to accelerate decay.

Studies of the types and sequences of fungi involved during wood colonization and decay also may reveal effective ways to modify wood to impart attractive colors or patterns or improve properties such as penetrability. Emerging high throughput sequencing techniques can provide enormous detail on the organisms present in a substrate and community ecology techniques can allow us to establish fungal relationships. These techniques permit detection of previously unculturable organisms and researchers are struggling to understand the roles of so many organisms in the decay process. This discovery process offers exciting new possibilities for developing a better understanding of the microbial ecology of decaying wood.

Wood as substrate for mushroom production

The ability of fungi to colonize wood and produce reproductive structures has been exploited for centuries in Asia for the production of mushrooms like Shiitake (*Lentinulus edodes*) and the Oyster mushroom (*Pluerotus ostreatus*) (Leatham, 1982; Zadrazil, 1974). These white rot fungi are inoculated into freshly-cut hardwood logs, that are incubated in cool, moist environments for up to two years (Leatham, 1982; San Antonio, 1981). Once they begin fruiting, the fungus produces mushrooms for one to two years before it exhausts the nutrients in the log. At that point there is little value left in the log, although there have been some suggestions to use this material as animal feed, since the fungi have solubilized the wood polymers to make them more digestible (Kirk, 1983).

Although the majority of Shiitake and other wood related mushrooms are produced in Asia, the need to develop markets for under-utilized hardwoods has stimulated the development of Shiitake production in the United States. The recent emphasis on health consciousness and the growth of the

gourmet food industry have also stimulated the development of a U.S. market. Recent studies in New Zealand suggest that high yields of Shiitake mushrooms can also be attained using coniferous sawdust.

While the use of logs to produce mushrooms will never replace more conventional uses, the process converts relatively low value material into a high value product with a ready market. As with any commodity, the producer must always contend with biological problems that include drying of the logs and contamination by competing microorganisms. The latter problem can be particular vexing, since certain Shiitake strains exhibit tolerance to competing species such as *Trichoderma*; however, this resistance can be overcome and producers must continually screen for new, resistant strains. Conversely, the problems of *Trichoderma* inhibition of Basidiomycetes can be exploited to prevent wood decay. There is a continuing need for research to develop more value-added products and mushroom production easily matches that need.

Use of fungal mycelium to produce packaging

We live in a disposable world where packaging is often used only once before being discarded, thereby filling our limited landfill space. Most of this packing, including plastics and Styrofoam is produced from non-renewable petroleum. Developing alternative renewable packaging material could reduce this impact. A number of fungi have been grow on grain molds to create foam like materials that can used several times then composted. While still expensive and not widely used, this product illustrates the potential for positively using the attributes of decay fungi.

Microbial generation of feedstocks

The carbohydrate polymers in wood represent a tremendous feed source, but they are largely inaccessible to most ruminants. The use of microorganisms to convert wood into a more accessible substrate for animal consumption or the use of fungi to degrade lignin to produce valuable by-products have both been discussed, but little research has been

conducted in these areas (Kirk, 1983). Similarly, fermentation of glucose and other hexoses to ethanol is commercially feasible using spent sulfite liquor as the substrate (Eriksson, 1990). This potential has also been exploited in biofuels research where hardwoods are grown on short rotations and the resulting wood is acid treated prior to enzymatic activity to degrade the hemicelluloses to produce ethanol. These strategies have some potential, but are largely limited by their high production costs compared with petroleum based materials. The brown rot fungi would appear to be ideally poised for these applications, since they can readily depolymerize cellulose and modify the lignin to make it more accessible to fungal enzymes. The use of a controlled brown rot pretreatment could make wood more usable for ruminants. Unfortunately, such pretreatments have received little attention (Kirk and Shimada, 1985).

A second application of this technology would involve the modification of lignin-based preparations by brown rot fungi to produce glues or resins. Most currently produced resins are petroleum-based and their availability could be sharply reduced in future years. Brown rot fungi produce free-radicals that initially depolymerize lignin and leave a demethylated residual. In this process, they create reactive sites that, if controllable, could be used to develop new types of polymers. Substitution of biomodified lignin for even a portion of the currently used resin would result in significant savings, while conserving a non-renewable material.

The use of fungi to produce chemical feedstocks from wood or wood waste has also been proposed; however, the cost of production coupled with the availability of less expensive substitutes largely limits research in this area. Renewed oil shortages would undoubtedly alter the economics, making biological production of feedstocks more feasible.

Biological control of fungal stain and decay

While most attempts to utilize wood inhabiting fungi have involved their ability to degrade wood, some organisms have the potential to protect wood from attack by other fungi. The use of fungi to protect wood from decay was advanced in the early 1960s by a number of researchers (Kallio, 1971, Hulme and Shields, 1972; Nelson, 1969; Ricard and Bollen, 1967) based upon findings in control of agricultural pathogens

(Baker and Cook, 1974). In the biological control scheme, a control agent is introduced into the wood either through a freshly cut stump, by spraying the surface with microorganisms, or by inoculation into a hole drilled into the wood. In practice, control of established pathogens is extremely difficult and most strategies seek to prevent colonization or protect the host from attack. Thus, biological protection is a more appropriate term for this strategy. The control agent then colonizes the wood to eliminate any existing decay fungi and prevent subsequent fungal invasion. The use of bioprotection is predicated on two critical points; the fungus must be capable of completely colonizing the substrate without damaging the wood and the protection must be long-term. These two requirements have severely limited the list of potential bioprotectants (Preston et al., 1982; Freitag et al., 1991).

The first bioprotection of wood decay fungi in forest products was reported by Ricard and Bollen (1967), using *Scytalidium* sp. to inhibit *Antrodia carbonica* in Douglas-fir poles. Although the effectiveness of this agent was debated, the findings stimulated additional research. More recently, a bioprotection formulation, Binab AB, has been marketed in Europe for protecting Scots Pine (Ricard, 1976; Bruce and King, 1986; Morris et al., 1984). This wood species appears to be colonized by a limited number of fungi including *Neolentinus lepideus* , a brown rot fungus that is particularly sensitive to the bioprotectants. Where this fungus dominates, the biocontrol agents should perform well, but there is considerable debate concerning the long term effectiveness of this formulation (Morris et al., 1984; Morris and Dickinson, 1981; Bruce and King, 1983; Bruce et al., 1983). Recent tests on southern pine and Douglas-fir suggest that this bioprotectant cannot completely control the numerous decay fungi associated with these species (Morrell and Sexton, 1990). In addition, the agent was unable to completely eliminate decay fungi already established in the wood, nor did it perform well against white rot fungi. Brown rot fungi are an important component of many decay systems, but recent studies have shown that white rot fungi are far more common in coniferous woods than previously thought (Zabel et al., 1982; Graham and Corden, 1980; Eslyn, 1970). While agriculture can deal with a small percentage of incomplete protection (as yield loss), the presence of small amounts of decay which can subsequently enlarge to destroy additional wood cannot be tolerated in a large wood structure. As a result, bioprotection does not appear to be feasible without the use of supplemental treatments that alter the ecology of the wood to favor growth or activity

of the bioprotectant. For example, chemical pretreatments to eliminate any competing fungi may provide an edge to the bioprotectant, which is applied some years after chemical treatment.

One area where bioprotection may have the immediate application is the prevention of fungal stains. Stains are normally limited by prophylactic chemical treatments, but increasing environmental concerns have led to a search for safer methods of stain control. Biological protection may be especially appropriate for this application since the protective period is relatively short and the protection can be delivered by surface colonization of the bioprotection agent. Preliminary testing suggested that bioprotectants limited fungal staining, but variations in performance were noted (Benko, 1988; Seifert et al., 1988). As discussed in an earlier chapter, research has also explored the use of pigmentless *Ophiostoma* species that colonize the wood ahead of the native stain fungi, thereby inhibiting discoloration. These fungi have only been used to a limited extent but illustrate the vast potential for exploiting the existing microbial ecosystem to limit fungal attack (Berendt et al., 1995).

While biological protection remains a promising strategy, it has largely remained experimental because it cannot provide a consistent degree of protection. However, biological controls are widely used in agriculture. There are a number of differences in approach that make this possible. The most important is how the users perceive risk and performance. In timber protection, failure to prevent decay could result in loss of a structure, injury and possibly death. Failure of a biological control to prevent disease of a crop will lead to loss of yield. While important from an economic perspective, the risks of using biological protection in agriculture are much lower and therefore more easily accepted.

Technology in pulp and paper

The use of microbes in the field of pulp and paper technology has received extensive study, probably because the net gains from their application are so easily measured (Kirk et al., 1983). The pulp and paper industry is much more process oriented than other forest products industries and is therefore more willing to consider new technologies. Furthermore, the economies of scale in a large paper mill make small returns on investments

attractive because of the total mill capacity. Incorporation of biotechnology has been proposed from the treatment of wood chips all the way through to the treatment of pulping effluents.

The use of fungi that selectively utilize lignin prior to chemical or mechanical pulping processes received tremendous attention as paper companies sought to reduce their chemical and energy costs while increasing pulp yields (Kirk, 1983; Kirk et al., 1983; Kirk and Shimada, 1985). These savings can be particularly significant in thermomechanical pulping, where small decreases in wood weight due to fungal attack can substantially decrease energy consumption during the pulping process (Kirk, 1985). Since mechanical pulping is an energy intensive process, these savings can become significant. In this process, a delicate balance must be maintained between lignin modification and carbohydrate utilization which decreases both paper strength and yield; however, some studies have considered incorporating residual fungal mycelium in the pulp to increase yields. Long fungal pre-treatment times (approaching 18 days) and other logistical hurdles have largely precluded commercial use of this technology.

While biological pretreatment has tremendous potential in the pulp and paper industry, there are no commercially viable processes in use. In general, the process of delignification requires the addition of energy either by the degradation of cellulose or hemicellulose, or by the addition of sugars (Kirk et al., 1984). At present, the degree of control required for stable commercial pretreatments has not yet been determined, although new advances in understanding fungal mediated lignin degradation may create new avenues for using these technologies. Most of these advances have been performed on a relatively limited number of white rot fungi. For example, most of the studies in the United States concentrated on *Phanerochaete chrysosporium* or *Trametes versicolor* and only 25–30 of the thousand or more white rot species have been researched to any extent (Kirk and Shimada, 1985). This lack of depth suggests that more efficient species for bioconversion processes remain undiscovered.

In addition to the few species involved, a number of substantial hurdles loom before the successful implementation of bioconversion strategies, including the difficulty of scaling up from a basic laboratory process to a large-scale commercial endeavor, the need to prevent contamination of the mixture by competing fungi, and the slowness of delignification in conifers. While all of these present a challenge, continued research may create new opportunities for exploiting these capabilities.

A second application of biotechnology to the pulp and paper industry addresses concerns about dioxin by-products produced during chlorine bleaching processes. Some dioxins are among the most toxic human-made compounds and strict limits have been placed on their production. For decades, paper mills routinely used chlorine bleaches to brighten their pulps, producing dioxins as by-products. Dioxins are extremely difficult to breakdown and this posed problems since most paper mills use and discharge copious amounts of water into nearby surface waters. Strict limits on dioxin content encouraged a search for alternative bleaching strategies. Ozone and a variety of other treatments have been substituted, but biobleaching has also been proposed. Biobleaching has shown promise in hardwood pulps, where the combination of reduced lignin content and the white color of the fungal hyphae in the pulp resulted in decreased Kappa number and increased brightness.

Biodecolorization is a third area where fungi, particularly white rotters, immobilized on filter media, could be used to remove pigmented compounds from pulp mill effluent.

Other potential areas for process improvement within the pulp and paper industry include pretreatments of hemicellulose for ethanol products, the use of microfungi for improving strength properties of paper, the use of brown rot fungi to create highly reactive, modified lignins that could be used to produce modified polymers, or the use of enzymes within paper mills to reduce the buildup of slime within pipes (Eriksson, 1990). These provide just a few of many opportunities for utilizing microorganisms in the pulp and paper industry.

Another new area in the bioconversion field is the use of white rot fungi to solubilize coal. This approach could make coal easier to handle without diminishing energy content. Studies indicated that Leonardite coal could be solubilized by exposure to cell free fractions of *Trametes versicolor*, a common white rot fungus (Pyne et al., 1987; Cohen et al., 1987), but the process was not commercialized.

Biotechnology in chemical waste management

Each year, millions of pounds of chemicals are hauled to hazardous waste sites where they are catalogued and stored. These chemicals remain

the responsibility of the producer until they are destroyed. Some of these chemicals can be burned in specially licensed incinerators, but the costs are high. Unfortunately, the technology to safely and economically eliminate toxic wastes remains elusive. The ability of white rot fungi and some bacteria to decompose complex structures such as lignin may provide one alternative to incineration or continued hazardous waste storage.

The ability of some fungi to detoxify preservatives is well known (Duncan and Deverall, 1964; Cserjesi, 1967; Chu and Kirsch, 1972; Seesman et al., 1977; Stranks and Hulme, 1973), but only recently have these same organisms been evaluated for their ability to completely degrade toxic wastes (Brown et al., 1987; Stanlake and Finn, 1982; Hammel et al., 1986). In addition, many natural microbial populations become more tolerant when continuously exposed to low levels of the toxin and this trait has been exploited to develop chemically resistant microbial populations (Edgehill and Finn, 1983). Recent results have indicated that bioelimination can result in the conversion of such highly toxic wastes as DDT to carbon dioxide (Bumpus and Aust, 1987; Bumpus et al., 1985).

While the results have been encouraging, and biological agents have been used in a number of soil farming schemes to purify contaminated soil (Dean-Ross, 1987), there are many details that need to be resolved before bioelimination becomes routinely used. At present, the rates of elimination are very slow, and the chemicals tested still remain in the solution at low levels (i.e. complete elimination has not been achieved). In addition, care must be taken to ensure that the wastes are completely converted to carbon dioxide, not just converted to less toxic intermediates. Regulators must also decide the appropriate levels of residual toxin that can remain in the material. In some instances, incomplete detoxification cannot be tolerated although the levels required for site remediation continue to change.

In general, laboratory bioelimination studies have concentrated on the use of pure cultures of white rot fungi and mixed cultures of bacteria in the presence of low levels of relatively pure chemicals. Many bacteria are capable of utilizing complex waste products either as sole carbon sources, or in conjunction with externally app1ied sugars. The effectiveness of these organisms when challenged by higher levels of combinations of chemicals often found in hazardous waste sites remains unclear.

In practice, bioremediation agents must compete with an existing microbial flora which may also be degrading the compounds present. The

ecology of bioremediation sites appears to vary widely and, as a result, is poorly understood. Most commercial remediation efforts employ the microbia1 flora already present at the site and attempts to stimulate the process by adding exogenous nutrients or oxygen. Further improvements in bioremediation will most likely depend on the development of more detailed information on the characteristics of specific sites, the organisms present in chemically contaminated soils and the physiology of these organisms in relation to chemical decomposition.

Spalting of wood

One intriguing use of fungi is to purposely discolor wood. Fungi produce a variety of pigments and induce a range of color changes as they degrade wood. Some of these colors as well as the zone lines produced by some white rot have long found a market in the art world. Spalting is the term used to describe the array of colors and patterns produced by these fungi and this wood has long been used in various art pieces. Fungi produce a range of colors in the wood including red, green and yellow that have been used in a style called intarsia. Formerly, collectors searched through downed trees in the woods for evidence of this damage, but recently, processes have been developed to use specific fungi to either create zone lines in wood or to grow fungi in culture so that the pigments can be extracted and used to dye other materials (Robinson et al., 2014). Spalting markedly increases the value of many wood pieces. This process has also been explored for producing natural dyes that might serve as replacements for synthetic aniline dyes as a means for reducing water pollution (Weber et al., 2014).

The future

It is apparent that there are numerous potential uses for wood inhabiting fungi within our current technology, including biopulping, bioelimination, biomass production, spalting, and biocontrol; however, the technologies to fully implement most of these approaches still appear to

be some years away. Never the less, the potential savings through reduced energy consumption, decreased environmental hazards, increased wood utilization, longer service life or added value for many forest products indicate that research outlays in this area are well worth the investment.

One area not addressed in this topic has been biotechnology or the modification of existing organisms to improve certain physiological characteristics, such as lignin degrading capability (Alic and Gold, 1985; Tien et al., 1987). While this technology is well-developed in other fields, it is relatively new to the study of wood deterioration. However, improving delignification capabilities while restraining degradation of the carbohydrates could increase the prospects for biopulping. Enhanced lignin degrading capabilities could also be used to improve bioremediation efforts. Some of these improvements could be accomplished through careful strain selection given the wide ranges of degradative capability of a single fungal species. However, gene modification and insertion techniques could result in much more rapid improvements. There is considerable debate about the dangers of releasing modified organisms into the natural environment and it would appear that the most likely path for rapid implementation of biotechnology in wood products would involve the selection of existing strains of fungi to accomplish the desired task. As we become more sophisticated in our approach to the manipulation of microbes, the use of altered microbes may then be contemplated.

As these technologies are developed, the information developed concerning the physiology and ecology of the wood inhabiting fungi will provide an improved understanding of the roles of these fungi in the degradation process and should ultimately lead to further applications of these fungi in industrial processes.

Summary

1. Wood degrading fungi may have many potential uses for improving wood utilization in a time of changing forest resources.
2. High value mushrooms can be grown on logs of underutilized hardwood species.
3. Some fungi may be useful for improving the digestibility of wood components for ruminants

4. Specific organisms can be used to inhibit stain or decay fungi.
5. White rot fungi have excellent potential for modifying lignin in wood prior to pulping to reduce energy consumption. These same fungi may also be used for pulp bleaching and decolorization of process wastewater.
6. White rot fungi may also be used for detoxifying certain hazardous wastes and, in combination with other organisms, represent an important component in remediation of hazardous waste sites.
7. Some fungi can be exploited for their ability to color wood to produce attractive zone lines, thereby marketing increasing the value of art products
8. The number of wood degrading organisms screened for potential users is limited. More useful capabilities may exist in the large number of less thoroughly researched fungi.

References

Alic, M., Gold, M.H., 1985. Genetic recombination in the lignin degrading basidiomycete *Phanerochaete chyrsosporium*. Appl. Environ. Microbiol. 50 (1), 27–30.
Baker, R., Cook, R.J., 1974. Biological Control of Plant Pathogen. Freeman Press, San Francisco, CA, 433 p.
Benko, R., 1988. Bacteria as Possible Organisms for Biological Control of Blue Stain. International Research Group on Wood Preservation Document No. IRG/WP/1339, Stockholm, Sweden.
Berendt, C.J., Blanchette, R.A., Farrell, R.L., 1995. Biological control of blue-stain fungi in wood. Phytopathology 85, 92–97.
Brown Jr., J.F., Bedard, D.L., Brennan, M.J., Carnahan, J.C., Feng, H., et al., 1987. Polychlorinated biphenyl dechlorination in aquatic sediments. Science 236, 709–712.
Bruce, A., King, B., 1983. Biological control of wood decay by *Lentinus lepideus* (Fr) produced by *Scytalidium* and *Trichoderma* residues. Mater. Org. 18 (3), 171–181.
Bruce, A., King, B., 1986. Biological control of decay in creosote treated distribution poles. Mater. Org. 21 (3), 165–179.
Bruce, A., Austin, W.J., King, B., 1983. Control of growth of *Lentinus lepideus* by volatiles from *Trichoderma*. Trans. Br. Mycol. Soc. 82 (3), 423–428.
Bumpus, J.A., Aust, S.D., 1987. Biodegradation of DOT (1,1,1-trichloro-2,2-bis-(4-chlorophenyl) ethane) by the white rot fungus *Phanerochaete chyrsosporium*. Appl. Environ. Microbiol. 53 (9), 2001–2008.
Bumpus, J.A., Tien, M., Wright, D., Aust, S.D., 1985. Oxidation of persistent environmental pollutants by a white rot fungus. Science 228, 1434–1436.
Cohen, M.S., Bowers, W.C., Aronson, H., Gray Jr., L.T., 1987. Cell-free solubilization of coal by *Polyporus versicolor*. Appl. Environ. Microbiol. 53 (12), 2840–2843.
Cserjesi, A.J., 1967. The adaptation of fungi to pentachlorophenol and its biodegradation. Can. J. Bot. 13, 1243–1249.
Chu, J.P., Kirsch, E.J., 1972. Metabolism of pentachlorophenol by an axenic bacterial culture. Appl. Microbiol. 23, 1033–1035.
Dean-Ross, D., 1987. Biodegradation of toxic wastes. ASM N. 53 (9), 490–492.
Duncan, C.G., Deverall, F.J., 1964. Degradation of wood preservatives by fungi. Appl. Microbiol. 12 (1), 57–62.
Edgehill, R.U., Finn, R.K., 1983. Microbial treatment of soil to remove pentachlorophenol. Appl. Environ. Microbiol. 45 (3), 1122–1125.

Eriksson, K.-E.L., 1990. Biotechnology in the pulp and paper industry. Wood Sci. Technol. 24, 79–101.
Eriksson, K.-L.L., Blanchette, R.A., Ander, P., 1990. Microbial and Enzymatic Degradation of Wood and Wood Components. Springer-Verlag, New York.
Eslyn, W.E., 1970. Utility pole decay. Part II. Basidiomycetes associated with decay in poles. Wood Sci. Technol. 4, 97–103.
Freitag, M., Morrell, J.J., Bruce, A., 1991. Biological protection of wood: status and prospects. Biodeterior. Abstr. 5 (1), 1–13.
Graham, R.D., Corden, M.E., 1980. Controlling Biological Deterioration of Wood With Volatile Chemicals. Final Report Project 212-1. Electric Power Research Institute, Palo Alto, CA.
Hammel, K.E., Kalyanaraman, B., Kirk, T.K., 1986. Oxidation of polycyclic aromatic hydrocarbons and dibenzo (p) dioxins by *Phanerochaete chrysosporium* lignase. J. Biol. Chem. 261 (36), 16948–16952.
Hulme, M.A., Shields, J.K., 1972. Interactions between fungi in wood blocks. Can. J. Bot. 50 (6), 1421–1427.
Kallio, T., 1971. Protection of spruce stump against *Fomes annosus* (Fr.) Cooke by some wood inhabiting fungi. Acta For. Fennica 117, 19p.
Kirk, T.K., 1983. Degradation and conversion of lignocellulose. In: Smith, J.E., Berry, D. R., Kristiansen, B. (Eds.), The Filamentous Fungi. Volume 4. Fungal Technology. Edward Arnold, London, pp. 266–295.
Kirk, T.K., 1985. Biological decomposition of solid wood. In: Rowell, R.M. (Ed.), The Chemistry of Solid Wood. Advances in Chemistry Series 207. American Chemical Society, Washington, D.C.
Kirk, T.K., Shimada, M., 1985. Lignin biodegradation: the microorganisms involved and the physiology and biochemistry of degradation by white rot fungi. Chapter 21. In: Higuchi, T. (Ed.), Biosynthesis and Biodegradation of Wood Components. Academic Press, Inc, New York, 679 p.
Kirk, T.K., Jeffries, T.W., Leatham, G.F., 1983. Biotechnology: applications and implications for the pulp and paper industry. Tappi 66 (5), 45–51.
Kirk, T.K., Tien, M., Faison, B.D., 1984. Biochemistry of the oxidation of lignin by *Phanerochaete chrysosporium*. Biotech. Adv. 2, 183–199.
Leatham, G.F., 1982. Cultivation of shiitake, the Japanese forest mushroom on logs: a potential industry for the United States. For. Prod. J. 32, 29–35.
Morrell, J.J., Sexton, C.M., 1990. Evaluation of a biocontrol agent for controlling basidiomycete attack of Douglas-fir and southern pine. Wood Fiber Sci. 22, 10–21.
Morris, P.I., Dickinson, D.J., 1981. Laboratory Studies on the Antagonistic Properties of Scyta1idium spp. to Basidiomycetes with Regard to Biological Control. International Research Group on Wood Preservation IRG/WP/1130, Stockholm.
Morris, P.I., Dickinson, D.J., Levy, J.F., 1984. The Nature and Control of Decay in Creosoted Electricity Poles. Record Annual Convention British Wood Preserver's Association.
National Research Council, 1990. Forestry Research: A Mandate for Change. National Academy Press, Washington, D.C.
Nelson, E.E., 1969. Occurrence of fungi antagonistic to *Poria weirii* in a Douglas-fir fir forest soil in western Oregon. For. Sci. 15 (1), 49–54.
Preston, A.F., Erbisch, F.H., Kramm, K.R., Lund, A.E., 1982. Developments in the use of biological control for wood preservation. Proc. Am. Wood Preserv. Assoc. 78, 53–61.
Pyne Jr., J.W., Stewart, D.L., Frederickson, J., Wilson, B.W., 1987. Solubilization of Leonardite by an extracellular fraction from *Coriolus versicolor*. Appl. Environ. Microbiol. 53 (12), 2844–2848.

Ricard, J.L., 1976. Biological control of decay in standing creosote-treated poles. J. Inst. Wood Sci. 7 (4), 6–9.
Ricard, J.L., Bollen, W.B., 1967. Inhibition of *Poria carbonica* by *Scyta1idium* sp., an imperfect fungus isolated from Douglas-fir poles. Can. J. Bot. 46, 643–647.
Robinson, S.C., Hinsch, E., Weber, G., Leipus, K., Cerney, D., 2014. Wood coloration through pressure treatment: the potential of extracted colorants from spalting fungi as a replacement for woodworker's aniline dyes. Materials 7, 5427–5437.
San Antonio, J.P., 1981. Cultivation of the shiitake mushroom. Hortic. Sci. 16, 151–156.
Seesman, P.A., Colwell, R.R., Zachary, A., 1977. Biodegradation of creosote/naphthalene treated wood in the marine environment. Proc. Am. Wood Preserv. Assoc. 73, 54–60.
Seifert, K.A., Breuil, C., Rossignol, L., Best, M., Saddler, J.H., 1988. Screening of microorganisms with the potential for biological control of sapstain on unseasoned lumber. Mater. Org. 23, 91–95.
Stanlake, G.J., Finn, R.K., 1982. Isolation and characterization of a pentachlorophenol degrading bacterium. Appl. Environ. Microbiol. 44 (6), 1421–1427.
Stranks, D.W., Hulme, M.A., 1973. The mechanisms of biodegradation of wood preservatives. Org. Holz 3, 345–353.
Tien, M., Kersten, P.J., Kirk, T.K., 1987. Selection and improvement of lignin degrading microorganisms: potential strategy based upon lignin model amino acid adducts. Appl. Environ. Microbiol. 53 (2), 242–245.
Weber, G., Chen, H.-L., Hinsch, E., Freitas, S., Robinson, S.C., 2014. Pigment extraction from the wood staining fungi *Chlorociboria aeruginosa*, *S. cuboideum*, and *S. ganodermphthorum* show potential for use as textile dyes. Color. Tecnol. 130, 445–452.
Zabel, R.A., Wang, C.J.K., Terracina, F.E., 1982. The Fungal Associates, Detectiono, and Fumigant Control of Decay in Treated Southern Pine Poles. Report EL 2768. Electric Power Research Institute, Palo Alto, CA, 80p.
Zadrazil, F., 1974. The ecology and industrial production of *Pluerotus ostreatus*, *Pluerotus florida*, *Pluerotus cornucopia*, or *Pluerotus eryngii*. Mush. Sci. 9, 621–652.

Author Index

Note: Page numbers followed by "*f*" and "*t*" refer to figures and tables, respectively.

C

G

H

I

J

K

L

M

N

O

P

Q

R

S

T

Subject Index

Note: Page numbers followed by "*f*" and "*t*" refer to figures and tables, respectively.

C

D

E

F

G

H

I

N

O

P

Q

R

S

T

U

V

W

X

Z

WOOD MICROBIOLOGY

Decay and Its Prevention

Robert A. Zabel & Jeffrey J. Morrell

SECOND EDITION

It is estimated that approximately 10% of the timber cut in the world is used to replace timber that has decayed in service. This represents an astounding loss of material. Educating timber users on wood deterioration and its prevention can help reduce the magnitude of these losses, thereby helping conserve forests.

Wood Microbiology, second edition, presents the latest advances in wood decay and its prevention. Coverage includes classification of fungi and bacteria, factors affecting growth and survival, fungal metabolism, and wood chemistry. There are also chapters that focus on the anatomical aspects, chemical changes, and ultrastructural effects of wood decay. Additionally, this book discusses major issues associated with wood decay, detecting decay, and how to take protective action against it.

This is a one-stop reference resource for wood scientists, wood processing and preserving professionals, foresters and forest pathologists, as well as students of forestry, and wood science and technology courses. It is authored by two leading experts with over 80 years of experience working with timber durability.

- Provides updated taxonomy and classification of decay groups
- Presents detailed descriptions of anatomical, chemical, and ultrastructural aspects of wood decay
- Includes discussions on major issues associated with decay, how to detect decay and preventative measures

About the authors

Jeffrey Morrell is the Director at the National Centre of Timber Durability and Design Life, University of the Sunshine Coast, Brisbane, Australia. He has over 40 years of experience working with timber durability, including 35 years at Oregon State University teaching and supervising the durability program.

Robert A. Zabel was a former Professor at SUNY College of Environmental Science and Forestry (ESF). Sadly now deceased, he was the major contributor to the first edition of Wood Microbiology. Prof. Zabel taught at ESF for over 40 years, and this revised edition is based upon notes from his course.

ISBN 978-0-12-819465-2

9 780128 194652

ELSEVIER

ACADEMIC PRESS

An imprint of Elsevier

elsevier.com/books-and-journals