AN ABSTRACT OF THE THESIS OF

Christoph F. Schauwecker for the degree of Master of Science in Wood Science presented on October 30, 2006.

Title: The Phytosanitation of Solid Wood Packaging Materials Using Wood Preservatives

Abstract approved:

Jeffrey J. Morrell

New species introductions have been associated with the movement of people for thousands of years. For instance, horses were introduced into North America by Spanish explorers, while pigs were introduced by the Polynesians into many Pacific islands long before the establishment of permanent European settlements. Both of these species introductions resulted in significant changes in both the biological and cultural composition of these places. However, during the past century, the amount of material transported by people has greatly increased due to the globalization of the economy and breakthroughs in material handling technology. These breakthroughs, such

as the steel shipping container and the wooden pallet, allow goods to be moved rapidly in a protected environment. This protected environment prevents goods from being damaged while in transit, but it has also improved the ability of pests to survive transit and resulted in invasive species introductions. The volume of goods being transported has steadily increased during the past 50 years and, thus, the number of species introductions has increased as well, resulting in a number of high profile pest introductions. For instance, the introduction of the Asian long horned beetle into Chicago and New York cost millions of dollars to eradicate and resulted in the loss of hardwood trees that previously lined the streets of these cities. In response to the Asian long-horned beetle and the introduction of the pine wood nematode into Europe, the Food and Agriculture Organization of the United Nations drafted and approved International Standard for Phytosanitary Measures Number 15. This measure recognizes solid wood packaging materials as an invasive species pathway and recommends sanitization through heat treatment or fumigation with methyl bromide. However, heat treatment or fumigation are imperfect mitigation tools. Thus, researchers continue to search for alternative methods.

Wood preservatives have long been formulated to prevent insect and fungal attack of wood products exposed to warm humid climates, but the ability of these chemicals to eliminate existing insect and fungal colonies has not been investigated. In this study, a number wood preservative systems were investigated to determine if these chemical formulations can be used for this application and to determine if wood preservatives, in general, are suited for use in phytosanitary applications.

A method for detecting the presence of wood boring insects through acoustic emissions was investigated. It was thought that a system of microphones and accelerometers could be used to detect wood boring insect presence and activity within a particular wood sample. These assumptions were based on earlier work conducted with termites. This system was to be used to determine if a sample contained wood boring insects prior to using it in the main study. However, acoustic emissions were not useful for this application, since feeding of the wood boring insects was sporadic, unlike termites which feed constantly.

Determining the feasibility of using wood preservatives in phytosanitary applications was addressed in three trials: the ability of preservatives to penetrate insect galleries, the ability of the insect larvae and pupae to penetrate a treated barrier and the ability of established wood boring insect populations to complete their life cycle under field conditions in wood pressure treated with preservatives

Preservatives completely penetrated between 80-100% of all insect galleries in western redcedar treated with ammoniacal copper quaternary compound, disodium octaborate tetrahydrate, or imidacloprid. However, barriers containing any of these three chemicals failed to prevent larvae from exiting the treated material, even in instances where the barrier was more than 6mm thick. The wood boring insects were unable to complete their life cycle under field conditions in pressure treated wood, while a large number of new house borer adults emerged from the untreated controls.

The wood preservatives investigated act more as insecticides than larvacides. However, vacuum pressure impregnation of solid wood packaging materials with the appropriate chemicals could provide lasting protection against invasive species introductions, allowing for the rapid, yet safe transportation of goods around the world.

The Phytosanitation of Solid Wood Packaging Materials Using Wood Preservatives

by
Christoph F. Schauwecker

A THESIS

submitted to

Oregon State University

in partial fulfillment of
the requirements for the
degree of

Master of Science

Presented October 30, 2006
Commencement June 2007

Master of Science thesis of Christoph F. Schauwecker presented on October 30, 2006.

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I understand that my thesis will become part of the permanent collection of Oregon State University libraries. My signature below authorizes release of my thesis to any reader upon request.

Christoph F. Schauwecker, Author

ACKNOWLEDGEMENTS

The author expresses sincere appreciation to the following individuals and organizations for their help during the completion of this work. Dr. Jeffrey J. Morrell has been a wealth of support during the planning, execution and writing process. Camille Freitag, Connie Love, Rand Sether and Milo Clauson have provided advice and assistance during the completion of the experimental phase. In addition, funding was provided by the United States Department of Agriculture under CSREES special grant number 37610-16287.

Interfor Pacific (Gilchrist, Oregon), graciously donated all of the ponderosa pine lumber used during this study, while the western redcedar lumber was supplied by East Forks Lumber Company of Coos Bay, Oregon. The author would also like to thank Royal Pacific located in McMinnville,Oregon, PacificWood Preserving Inc. of Sheridan, Oregon, and J.H. Baxter, LLC of Eugene, Oregon for donating retort time, allowing the author to treat samples using commercial treating facilities. Chemical Specialties Inc. (Charlotte, North Carolina) donated the imidacloprid containing compounds for this project and also treated all of the imidacloprid containing samples using a laboratory scale treating cylinder. In addition, treatment with ClearWood MW1 was donated by Jeld-Wen Inc., Klamath Falls, Oregon. Without the support of all of these corporate sponsors, this work would not have been possible.

The author would also like to thank Drs. Darrell Ross and Kimberly Wallin for their help with the entomology portion of this study. Dr. Michael Milota provided support through the use of his equipment and through being a member of the committee. In addition, Megan J. Moerdyk-Schauwecker aided in the proof reading of the preliminary manuscripts and supported the author over the duration of this project.

CONTRIBUTION OF AUTHORS

Dr. Jeffrey J. Morrell assisted with writing the chapters.

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INTRODUCTION

Background

During the past two hundred years, it is estimated that over 4,500 new species have been introduced into North America (NA) (Dubensky *et al,* 2001; Windle 1996; and United States Congress 1993), and more than four hundred of these species are known to feed on trees or shrubs (Haack 2003). Between 1906 and 1991, the 79 most damaging of the 4,500 introduced species caused documented losses exceeding $97 billion (Windle 1996).

The number of species introduced into the United States each year has steadily increased (Windle, 1996; United States Congress, 1993). The elevated number of species introductions is most likely due to a variety of factors including a rapidly expanding global economy, more rapid and efficient movement of materials around the globe and reduced trade barriers due to free trade agreements (Dubensky *et al,* 2001). Researchers have found that species travel the world using many different pathways. The most relevant materials to forest pests include fruits and vegetables, logs, chips, unseasoned lumber, solid wood packaging material (SWPM), and live plants (Dwinell, 2001a). SWPM is used in ninety-five percent of all international shipments (Dubensky *et al,* 2001), and its potential to harbor wood pests has attracted increasing attention. An estimated 88% of all scolytid interceptions made by APHIS during 1985 and 2001 were made on SWPM. This material is especially attractive because it remains biologically viable during its service life (Haack, 2003).

Since the mid 1980s, a number of different regional and national organizations have implemented phytosanitary standards to deal with SWPM. However, a coordinated global response to this invasive species pathway was not formulated until 2002 when the International Plant Protection Convention (IPPC) of the United Nations Food and Agriculture Organization (FAO) formulated International Standard for Phytosanitary Measures (ISPM) Number 15. This measure recognized SWPM as an invasive species pathway and suggested the use of heat treatment and fumigation with methyl bromide as methods to prevent invasive species introductions. Following the adoption of ISPM #15 by the FAO, a majority of large exporting and importing countries around the world have adopted it into their legal codes. However, research has shown that some fungi are heat tolerant allowing them to survive the temperatures specified in ISPM #15 (Morrell 1995). While methyl bromide is a well understood and widely used fumigant, it has also been shown to contribute to global warming by depleting the ozone layer. This chemical is scheduled to be phased out by 2015 under the Montreal Protocol (CSIRO 2001). In addition, these sanitation methods cannot be directly verified by importing countries, since neither heat treatment nor fumigation alter the wood. Likewise, re-infestation of SWPM is possible, since wood moisture content is not significantly reduced during the heat treatment process and methyl bromide is fleeting in nature.

Wood preservatives may be a promising alternative to heat treatment or fumigation. These chemicals have been specially formulated to control fungal growth on wood products exposed to decay prone environments. Wood preservatives also contain chemicals that prevent insect infestation. These attributes of wood preservatives would give lasting protection against invasive species introductions, and since wood preservatives alter the wood substrate, their use could be verified by importing countries. Further, the use of wood preservatives and the methods used to deploy them effectively are well understood and have been used since 1838 (Hunt and Garratt 1938). However, the ability of wood preservatives to eliminate established insect populations has not been investigated, nor has it been determined if adequate wood and gallery penetration can be achieved when these systems are applied to wood products above the fiber saturation point.

Objectives

The objectives of this project are:

1. Determine the ability of wood preservatives to penetrate frass packed insect galleries in refractory wood species.
2. Determine the ability of wood preservatives to prevent adult and larval penetration of a treated barrier.

3. Determine the ability of wood preservatives to control wood boring insects in field conditions
4. Develop a method of detecting wood boring insect larvae using acoustic emissions in order to aid sample selection.

Scope

In order to preserve species diversity, the introduction of unwanted species into new ecosystems must be prevented. SWPM is one path used by unwanted species to gain access into new ecosystems. In order to prevent this ISPM #15 has been ratified by the FAO and incorporated into national legal codes the world over. However, the treatment methods currently recommended within ISPM 15 are imperfect. During this study, wood preservatives were investigated to determine if they can be used for this application. Wood preservatives have some advantages over current treating methods, mainly because they provide long term protection and because they are verifiable by the importing country. However, wood preservatives have not been investigated to determine if they can destroy established wood boring insect populations within this material. Thus, this work was conducted to determine if wood preservatives could penetrate frass packed insect galleries, and to determine if larvae and adults penetrated a wood preservative treated barrier. In addition, a field trial was conducted to verify these results under conditions experienced by SWPM while in use. The conclusions drawn from this work will indicate if wood preservatives can be used in this application, and if currently available chemical formulations will be effective.

Thesis Structure and Units

This thesis is written in manuscript format. It has a comprehensive literature review (chapter 2) followed by a number of chapters describing each individual study. Chapters 3, 4 and 5, describe the wood larvae detection method, preservative penetration and the field exposure and were prepared as manuscripts suitable for publication in the Forest Products Journal. Chapter 6 is intended as part of a manuscript prepared for the 27th Annual Meeting of the Canadian Wood Preservers Association held in Vancouver, British Columbia, Canada on the 6th and 7th of November 2006. These chapters are followed by an overall conclusions chapter (chapter 7) and by a bibliography (chapter 8).

International Standard (SI) units are used through out this publication.

LITERATURE REVIEW

Introduction

Flora and fauna have traveled the world, following the travels of mankind. New species were established along ancient trade routes used by Europeans as they traveled from Europe to the Far East. Plants, animals, insects and fungi also followed Europeans as they explored the world. During the past two hundred years, it is estimated that over 4,500 new species have been introduced into North America (NA) (Dubensky *et al,* 2001; Windle 1996; and United States Congress 1993), and more than four hundred of these species are known to feed on trees or shrubs (Haack 2003). Between 1906 and 1991, the 79 most damaging of the 4,500 introduced species caused documented losses exceeding $97 billion (Windle 1996). More recently, the introduction of the Asian long horned beetle (*Anoplophora glabripennis*, ALB) from China to New York in 1996 and to Chicago in 1998 has resulted in an eradication program that has cost the United States federal government and state agencies $59 million dollars thus far (United States Department of Agriculture 2006). The introduction of the ALB and the pinewood nematode (*Burasaphelenchus xylophilus*, PWN) into Portugal has also received a great deal of media attention and rekindled interest in invasive species both in the scientific community and in the population at large.

The number of species introduced into the United States each year has steadily increased (Windle, 1996; United States Congress, 1993). The increasing number of species introductions are most likely due to a variety of factors including a rapidly expanding global economy, more rapid and efficient movement of materials around the globe and reduced trade barriers due to free trade agreements (Dubensky *et al,* 2001). Researchers have found that species travel the world using many different pathways. The most relevant materials to forest pests include fruits and vegetables, logs, chips, unseasoned lumber, solid wood packaging material (SWPM), and live plants. However, even non-related products such as cars and used machinery can harbor pests (Dwinell, 2001a and Bell, 2001).

SWPM is defined by the United States Department of Agriculture (USDA) Animal and Plant Health Inspection Service (APHIS) as "wood packing material other than loose wood packing materials used or for use with cargo to prevent damage, including but not limited to dunnage, crating, pallets, packing blocks, drums, cases, and skids" (United States Department of Agriculture 2006). SWPM is used in ninety-five percent of all international shipments (Dubensky *et al,* 2001), and its potential to harbor wood pests has attracted increasing attention. An estimated 88% of all scolytid interceptions made by APHIS between 1985 and 2001 were made on SWPM since it remains biologically viable during its service life (Haack, 2003).

Since the mid 1980s, a number of different regional and national organizations have implemented phytosanitary standards to deal with SWPM and other forest products. In 2002, the International Plant Protection

Convention (IPPC) of the United Nations Food and Agriculture Organization (FAO) formulated guidelines for regulating wood packaging material in international trade in an effort to increase the uniformity of these standards. Since the creation of the IPPC guidelines, national and regional governmental bodies have been incorporating these guidelines into their legal codes.

The IPPC guidelines state the reasons why SWPM and other forest products must be regulated and suggests methods for treating SWPM and other forest products. The methods suggested by the IPPC are fumigation, primarily with methyl bromide; heat treatment (HT); and drying the material as a form of HT.

The nature of SWPM and the reasons for the IPPC guidelines will be discussed as well as the recommended mitigation methods. A new mitigation method, low retention pressurized preservative treatment, will also be discussed as an alternative to HT and fumigation.

Solid Wood Packaging Material and Invasive Species

Prior to 1900, the United States federal government and others thought it would be advantageous to import different plant and animal species from around the world. One of the original purposes of the USDA was to promote the spread of introduced species (U.S. Department of Homeland Security, 2004). Forty years after the creation of the USDA, however, in 1912, the Plant Quarantine Act was passed by the U.S. Congress in response to the chestnut blight (*Chryphonectria parasitica*). Introduced into New York State in 1904, the blight rapidly destroyed the America Chestnut (*Castanea dentata*), a major source of income for Appalachian farmers at the time (The American Chestnut Foundation, 2004). The introduction of the Plant Quarantine Act in 1912, helped reduce the number of introduced species (Windle, 1996).

In the years that followed, the time required to cross large distances decreased, while the volume of materials moving from one location to another greatly increased. During the 1930s, the technology needed for mechanized material handling was developed. A corner stone of this technology was the wooden pallet. This device, constructed of rough cut, low grade lumber, is used to store and transport large amounts of material at one time. The full importance of the unit load material handling system was not discovered until World War II. The war effort and the reconstruction effort that followed required the movement of large amounts of materials quickly. The wooden pallet had many advantages: forklifts could be used to move pallets with ease, and the pallets were light, which reduced the weight of the packaging material involved. Pallets were also expendable, since they were inexpensive (Orr 1990). However, pallets also introduced a new way for species to travel the globe.

Formosan subterranean termites (*Coptetermes formosanus*) are an example of a species which was introduced into new environments by the rapid movement of materials. This termite species is extremely destructive and is found throughout southeastern Asia. Ships returning from the Pacific

Theater following World War II carried large amounts of wood material infested with these termites. During the unloading process, the termites were introduced into Louisiana, Texas, and the Carolinas. SWPM has also facilitated the further spread of the formosan termite to other parts of the southern United States (Laks, 2002).

The use of large, modern cargo ships and aircraft along with the advent of shipping containers has increased the likelihood of introducing exotic species with SWPM. In the past, SWPM dried during shipping or the time required to complete the voyage allowed the flora and fauna residing in the material to complete their life cycles and emerge. A majority of the time, the environment faced by the emerged species was not suitable for survival, resulting in their demise. Modern ships and aircraft reduce the time needed to move material and, thus, reduce the amount of drying. It also increases the chances that the flora and fauna found within the SWPM have not emerged. Both of these factors increase the likelihood of a species being inadvertently introduced into a new ecosystem. Shipping containers have also aided the movement of invasive species. These air and watertight containers increase the rate at which vessels can be loaded and unloaded. The containers are often lined with wooden panels, and wood is used as blocking, pallets, and in other forms within the containers. The material used for these purposes is often low grade and, thus has a great likelihood of containing insects and fungi. The shipping container protects these insects and diseases from the harsh environments experienced during shipping, resulting in the introduction of viable organisms into the importing country (McNamara and Kroeker, 2001; Dobensky *et al,* 2001). A combination of refrigerated containers and the rapid movement of material also allow for the importation of large quantities of perishable items (APL Limited, 2004), creating another pathway used by non-native species.

The rapid movement of cargo and the use of low-grade wood for packaging material have resulted in the introduction of the PWN into Portugal. In 1984, Scandinavian authorities found wood chips imported into Finland from NA were infested with PWN. This was of great concern to the authorities since the PWN is known to be the causal agent for pine wilt disease (Dwinell and Nickle, 1989). The PWN was introduced previously into Japan, South Korea, Taiwan, and the People's Republic of China with disastrous results. Researchers learned from the introduction of PWN into Japan that the species not native to NA were highly susceptible to this nematode, while species native to the PWN's home range were only attacked if the tree was already stressed (Linit, 2001). The discovery of the PWN in Finland resulted in swift legislative action by both the Scandinavian countries and Europe as a whole. By July of the following year, all Scandinavian countries had banned the importation of raw wood products from areas of the world known to have PWN. The European Plant Protection Organization (EPPO) likewise made the recommendation to its members in July 1985 that only kiln dried softwood lumber should be imported from NA and Southeast Asia (Dwinell and Nickle, 1989). The ban on the import of raw softwood products from NA into the European Union (EU) resulted in large economic losses for softwood lumber exporters as well as individuals exporting pulp chips to the Scandinavian countries. These actions resulted in the development of pasteurization processes to allow the import of green products. Unprocessed wood products once again came into the spotlight in 1999 during an extensive survey conducted throughout Europe. During this survey, the PWN was located in a small area (30km radius) of southern Portugal. The cause of the PWN outbreak has not been determined, but SWPM was believed to be the source and The Plant Quarantine Service of Finland confirmed these fears. The Finish agency found that 5% of all SWPM from NA or China tested was infested with PWN or its vector, the long horned beetle *Monochamus* spp. (McNamara and Kroeker, 2001; Dwinell and Nickle, 1989). In response to the outbreak, the EU passed an emergency measure for coniferous SWPM. The measure required that all coniferous SWPM be treated using heat, fumigation, or chemical pressure treatment. After treatment, the SWPM must be marked indicating compliance with the measure. This ruling went into effect in October of 2001 (USDA APHIS, 2003). Following the incidents associated with the PWN, other nations started to regulate the importation of non-manufactured wood products.

Legislation

Import and export of agricultural products has been regulated since the beginning of the twentieth century. It was recognized early on that the unregulated import of fruits and vegetables along with animals and animal products could lead to the introduction of unwanted species (Department of Homeland Security, 2004). The idea that most invasive species are associated with fresh agricultural products still holds true today. APHIS spends a majority of its time inspecting the vast numbers of shipments into the United States that contain fruits and vegetables or animal products. Since the early 1900s, APHIS has also recorded pest interceptions. Between 1985 and August 2001, 577,829 interceptions were made by the agency with an average of 36,882 interceptions per year. However, the agency currently inspects only two percent of all international shipments. Of the total number of interceptions between 1985 and 2000, 6,827 were scolytids or other wood boring insects (Haack, 2003). The importation of raw wood products as cargo has been regulated for some time both in the United States and around the world. Examples include the ban on the importation of raw wood materials into Europe, and similar restrictions put in place by NA, Australia and New Zealand. All of these restrictions are intended to prevent the importation of species that may damage the local forest ecosystems.

The idea that SWPM is also a pathway for the introduction of exotic species has only been recognized in the past ten years. The Australian

Quarantine and Inspection Service was the first to introduce legislation dealing with SWPM. The agency stated that it preferred shipments which did not contain any wood products, specifically any non-manufactured wood products such as SWPM (Australian Quarantine and Inspection Service 2004). Since then, other countries have followed suit. Most major importing and exporting countries currently have legislation in place dealing with SWPM. However, the rules imposed by different countries or regions vary greatly. Some countries have also criticized the rules imposed to deal with SWPM as an alternative to tariffs and other trade barriers. The General Agreement on Tariffs and Trade (GATT) prohibits these types of trade barriers. The issues surrounding phytosanitation as a trade barrier were discussed during the Uruguay Round of Negotiations in 1986. The result of these negotiations was The Agreement of the Application of Sanitary and Phytosanitary Measures which was signed in early 1994. This agreement gave authority to the IPPC to establish guidelines associated with phytosanitary measures and resulted in the establishment of international standards of phytosanitary measures. During the 2002 meeting of the IPPC, the organization ratified International Phytosanitary Measure 15 (IPSM #15), which addresses SWPM (Griffin, 2001; FAO, 2002).

The United States forest products industry is faced with a shrinking land base and increased environmental restrictions on harvest activities. This has led the industry to seek ways to increase yields. The industry has been doing this by looking at new technology and at offshore, fast growing, high-yield forest plantations for its raw material supply. This has led to the request by the forest products sector for permits to import large quantities of raw wood products into the continental United States. This could, in turn, lead to a greater risk of introducing destructive non-native species, especially since the areas from which the logs originate have similar climatic conditions to the continental United States (Dubensky *et al,* 2001). The United States federal government responded to the forest products industry requests by passing legislation in 1995 that required importers of raw wood products to obtain a permit from the federal government. These new regulations came under fire, however, from both environmental groups and trading partners, mainly in Europe. This resulted in delayed implementation of the new rules. The rules came into effect in 1999 and the United States was once again a net importer of forest products. In 1999, these imports totaled 18 billion board feet of softwood lumber or 35% of consumption (Dubensky *et al,* 2001; Hicks, 2001).

During 1996, the ALB was discovered in New York and two years later in Chicago. This beetle is a great threat to tree species of significant industrial and urban forestry importance. The pathway used by the ALB was investigated; researchers found that the beetle was imported into the United States in both instances within SWPM from China. In the years following the discovery of the ALB, nearly 59 million dollars were spent on eradication (Haugen and Iede, 2001; McNamara and Kroeker, 2001; Dwinell, 2001a; and

United States Department of Agriculture 2006). In addition, an interim rule was passed requiring heat treatment or fumigation of all SWPM originating or passing through China by amending section 319 of article 7, the same legislation originally introduced to prevent the unregulated importation of raw wood products (USDA APHIS, 2004). The number of scolytidae interceptions made by APHIS in packaging material originating from China following the introduction of this interim rule greatly decreased, proving the effectiveness of such legislation (Haack, 2003). U.S. legislation pertaining to the importation of SWPM with cargo was revised further in 2003 to reflect IPSM #15 by extending the restriction on the importation of SWPM to all countries except Canada (United States Department of Agriculture 2006).

Current Mitigation Methods for Solid Wood Packaging Material

IPSM #15 mandates that the material must be heat treated or fumigated prior to use in international trade. Once the material has been treated using either the fumigation or HT method, a symbol is applied to the material signifying that it fulfills the standards outlined in IPSM #15. The guidelines outlined by the IPPC also allow for the use of other methods to sanitize SWPM once they have been scientifically proven to be effective (FAO, 2002). Since 2002, when all members of FAO agreed to these guidelines, member nations have made them part of their legal codes. The universal requirements for all SWPM were deemed necessary due to the potential reuse of SWPM in other countries, making it difficult to determine origin. This argument persuaded the World

Trade Organization (WTO) and others that the regulations, and their suggested mitigation methods were necessary (McNamara and Kroeker, 2001).

Visual Inspection of Solid Wood Packaging Material

Visual inspections of SWPM are often performed by the material producer because it is not to the producer's advantage to utilize material that is severely decayed or infested with wood boring insects. Visual inspections can be used to detect severe cases of pest infestation. The method is inexpensive and requires minimal resources to complete. Visual inspection also has disadvantages. It is hard to detect the early stages of insect or fungal attack, since the entry holes for the insect larvae are small and fungal damage may not be visible on the surface (Roll-Hansen and Roll-Hansen, 1979; Wilcox, 1978). Inspection is hampered further by the low surface quality of most SWPM. Most SWPM is rough-cut, making it difficult for inspectors to quickly see entry holes or fungal damage. Reinspection of the material at its destination is also difficult since a majority of the SWPM surfaces are not visible to the inspector without unloading the cargo. Complete inspections of cargo are made at random by national plant protection agencies. However, the overall percentage of cargo inspected is small and increasing the number of these inspections would be costly. In addition, the space for such inspections is not available at most port facilities. Thus, visual inspection can be characterized as an important and low cost way of identifying severe fungal and insect infections, but is ineffective for detecting the early stages of fungal or insect attack (Morrell, 1995, 1996; Roll-Hansen and Roll-Hansen, 1979; Wilcox, 1978).

Debarking

Currently, Australian and New Zealand regulations require that all material imported be completely free from bark. Bark is attractive to many beetles and removing this material is one of the most effective ways to prevent beetle infestation of wood. Many beetles oviposite on the bark and the larvae, once hatched, tunnel further into the sapwood were the insect develops. Removing bark shortly after felling lowers the probability that eggs deposited by wood boring insects will develop and penetrate into the wood, where they will be more difficult to control. Debarking also removes the extremely rough surfaces that can carry other substances such as soil and other debris that harbor species normally not associated with wood (Morrell, 1995, 1996).

Completely debarking low quality logs typically used to manufacture SWPM can be difficult because these logs often contain large deformities and knots. Debarking equipment does not handle these defects well, resulting in residual bark on portions of the log and the resulting lumber. Debarking also does not eliminate insects and fungi already established within the log. Thus, debarking alone is not an effective means for preventing the introduction of the PWN or the ALB.

While bark removal is important for reducing the risk of insect attack, it also exposes the nutrient rich sapwood to fungal colonization. Some of the fungi growing on logs also use beetles as their vectors, increasing the risk of fungal dissemination in the destination country. Thus, the risk of introducing new species into an ecosystem cannot be completely eliminated by debarking. However, debarking remains a key part of any mitigation practice because removing a majority of the bark greatly reduces the probability of introducing exotic species at a relatively low cost (Morrell 1995, 1996,).

Heat Treatment

Heat has long been used to sterilize materials. In 1865, Louis Pasteur, a French chemist developed the pasteurization process. This process reduces microbial growth in liquids by heating them to 63°C for 30 minutes (Dwinell, 1996). During the 1930s, heating was investigated as a method for sterilizing wood prior to treatment with oil-based wood preservatives. Sterilizing the wood prior to treating was necessary to prevent the final product from deteriorating at the core where the preservative treatment had not penetrated the wood. Chidester used small samples of green loblolly pine (*Pinus taeda*) inoculated with different common wood decay fungi. Once the fungi had penetrated the core of the sample, Chidester heated the samples at different temperatures and for different lengths of time (Chidester, 1937). In 1939, Chidester reported that the temperature needed to kill fungi was highly time dependent and concluded that a core temperature of 76°C was adequate if this temperature was maintained for at least 30 minutes. Chidester found an inverse relationship between time and temperature and concluded that internal temperatures lower than 65°C were not practical because of the long times required to kill fungi (Chidester, 1939).

During 2001, the EU started to require HT or fumigation of all SWPM. The EU emergency requirements were aimed at preventing the introduction of the PWN and its vector (USDA APHIS, 2001). This HT standard required a minimum core temperature of 56°C for a minimum of 30 minutes, and was based on the results of a multinational study involving Canadian, European, and American scientists (Dwinell, 1997). These temperature and time requirements were later used in IPSM #15 for the HT standard (FAO, 2002). However, this temperature/time requirement does not eliminate all exotic species. Morrell (1995) compiled a list of species that can survive the HT schedule found in IPSM #15. The vast majority of the species found on the list are fungi that pose less of a risk than other organisms. Fungi are often restricted by environmental requirements, preventing them from becoming widespread pests (Hulme 1979). However, one thermotolerent species of concern is *Phellinus weirri* a root pathogen that can cause serious damage to forest ecosystems (Morrell, 1995).

The moisture content (MC) of wood during heating also affects the effectiveness of HT. Organisms in wood at moisture contents above the fiber saturation point (normally 28% MC, FSP) are more likely to be biologically active. The moist heat produced under these conditions results in the coagulation of proteins, disrupting critical cell functions and leading to cell death. Water is also a better heat conductor than air, lowering the time required to reach the necessary core temperature. The oxidation processes that are relied upon to kill organisms where the moisture content is below the FSP occur more slowly (Dwinell, 2001b). The risk of introducing exotic species in SWPM at lower MC's is lower; however, some species can tolerate these conditions. Thus, the type and condition of SWPM should be considered (Ormsby, 2001); however, neither the current wording of IPSM #15 nor the leading national legislative actions address this issue.

One major drawback to heating is that it is virtually impossible to verify that heat treatment has been carried out according to the IPPC standards, especially in material that is reused. The American Lumber Standard Committee, which is the governing body for HT of SWPM in the United States, has delegated oversight responsibilities for HT of SWPM to third party accreditation agencies. These third party agencies certify the HT process used by a SWPM producer and allow the production facility to use the international marking for heat treated SWPM. These marks, according to the American Lumber Standard Committee, must be applied, by stamping, branding, or labeling (American Lumber Standard Committee Incorporated, 2003). However, there is some concern that the markings applied in this manner may last longer than the phytosanitary effects of heat treatment.

Dwinell (1995) showed that heat-treated pine logs, placed in the field during the flight period of the pine sawyer beetle (*Monochamus sciuttolatus*) were re-infested with both the PWN and pine sawyers (Dwinell, 1995).

An alternative to heat treatment is the use of kiln-dried lumber for the manufacture of SWPM. This method of decontaminating forest products has been recognized by multiple national governmental bodies as well as by the IPPC (Australian Quarantine and Inspection Service 2004; FAO, 2002; American Lumber Standard Committee Incorporated, 2003). This practice reduces the moisture content of the wood below 20% and heats the core of the material above the temperature required for HT. The economic impacts of such a requirement vary greatly. Kiln drying softwood species to these requirements is economically viable, and indeed a large portion of the softwood lumber in use is treated to these standards (Clarke 2001). However, the costs of treating hardwood species to these requirements are much greater since the drying periods must be much longer to avoid degradation. The extensive use of hardwoods in SWPM make it difficult to require kiln drying for these materials (Clarke, 2001; Araman *et al,* 2003; McLeod III *et al,* 1991).

Although HT is an effective method for decontaminating different materials, the method also has some drawbacks. It is impossible to verify that heating has been completed, and the temperature and time currently suggested by the IPSM #15 may not be sufficient for neutralizing all the insects and fungi present in wood. However, requiring either higher temperatures than those currently suggested or lowering the moisture content of the SWPM would both have prohibitive economic impacts. These drawbacks are a major consideration by the IPPC and by national governments for implementation of global phytosanitation standards for SWPM.

Fumigation

Fumigants have been used for decades to control insects, fungi, and other potential pests in a variety of commodities. The ideal fumigant is toxic to flora and fauna under a wide range of climatic conditions. The chemical must also be non-explosive, easily dispersed, and able to rapidly penetrate the commodity being treated. The ideal fumigant must also not alter the material treated (Kenaga, 1957). These chemicals have been used in both international trade and in agriculture for over fifty years (Sarwar *et al,* 2003). Fumigants are routinely used to treat quarantined shipments at the port of arrival, to prevent decay in grain storage facilities or other agricultural products warehouses, and as remedial treatments for wood structures. Fumigants can even be used to prolong the life of utility poles (Morrell and Corden, 1986). The use of some fumigants to treat wood products intended for international trade has also been studied. The large body of literature supporting the effectiveness of these chemicals led the IPPC to recommend fumigation as the only alternative to HT for phytosantiation of SWPM. However, fumigants also have some drawbacks. Most are extremely dangerous to humans and some have very low ignition points, making them a fire hazard. This is especially problematic if the chemicals are being used while the material is in transit. Still other fumigant formulations are harmful to the environment.

Methyl bromide (MB), one of the most widely used and studied fumigants, was discovered in the 1930s. Prior to this time it had been used as a fire suppressant. This odorless chemical has a boiling point of 3.6°C and penetrates most materials quickly, yet has very low sorption. Fumigant sorption can affect commodity quality, making it important to be able to remove the chemical from the material quickly through aeration (Harris, 1963). These properties have led to the widespread use of MB in applications including soil, commodities such as grains, and structures (Kramer, 1992).

During the 1960s and 1970s, MB was investigated for controlling oak wilt caused by *Ceratocystis fagacearum*. This fungus caused oak death (*Quercus* spp.) throughout the southern and eastern United States. European countries banned the importation of all unprocessed oak products from the United States because of concerns about the presence of viable fungi in the material. MB was tested for its ability to kill this fungus in non-debarked logs. Logs were cut, close stacked, and covered with plastic tarpaulins; MB was then introduced at a concentration of 240 g/m^3 and this concentration was held for a period of days depending on the thickness of the logs or lumber. Viability tests of the oak wilt fungus after treatment showed that MB eradicated the fungus, allowing the logs to be sold internationally (Schmidt, 1982; Jones, 1963). Researchers also found an inverse relationship between temperature and the amount of MB needed to decontaminate logs and lumber. Material below the freezing point of water could not be adequately treated, even when large amounts of MB were introduced (Kenaga 1961, and Schmidt, 1983). The large body of evidence supporting the effectiveness of MB in oak and other commodities, led the IPPC to adopt MB as the only approved fumigant for use with SWPM (FAO, 2002).

High toxicity to humans, coupled with its lack of color or odor make MB difficult to handle (Harris, 1963). As a result, chloropicrin, another fumigant is usually added at low levels as an awareness agent. MB is also less effective when used to treat coniferous species above the FSP, yet much of the SWPM treated is above the FSP (Cross, 1991). Like HT, MB does not provide long term protection against re-infestation, since the chemical is rapidly dissipated after treatment (Harris, 1963). Verification of MB treatment by the importing nation is also impossible, since there is little or no MB in the SWPM days or weeks after treatment (Ruetze and Liese, 1985). MB has also been listed as an ozone depleting substance under the Montreal Accord. The Montreal Accord states that developed countries should have phase out the use of MB by January 2005 with the exception of pre-shipment and quarantine application, which are permitted until 2015 (CSIRO 2001).

The impending loss of MB as a quarantine tool has encouraged a search for replacements. This research has lead to some promising alternatives including. sulfuryl flouride (SF) and ethanedinitrile (EN). SF has been successfully used to decontaminate a variety of commodity products, both in the laboratory and in practice (Kenaga, 1957). SF has been found to be effective against a large variety of adult insects, but was less effective against eggs (Kenaga, 1957). Like MB, this chemical has little effect on most commonly fumigated materials in terms of color and odor (Kenaga, 1957). SF has been used as a space fumigant to eradicate termites and wood-boring insects in houses and has also been investigated for decontaminating oak logs for export (Dwinell, 2001c). The oak wilt fungus was killed throughout the sapwood, however, other microorganisms were not completely eradicated (Dwinell, 2001c). The drawbacks associated with SF are similar to those for MB. The gas is odorless, yet highly toxic; it does not perform well at low temperatures; and its use cannot be verified after fumigation (Kenaga, 1957).

The fumigant EN was developed in Australia and has been shown to eradicate insects, nematodes, fungi, and bacteria. Penetration and diffusion of EN is more rapid than MB and EN also has an odor (Ren 2001). Promising early results have led to commercialization as a space fumigant. Australia, and China are currently investigating this chemical for use as a quarantine fumigant to replace MB (CSIRO 2004). Drawbacks to EN include it’s extreme flammability (Matheson Tri-gas 2004); it ability to discolor some materials, especially plastics, and the lack of a long term track record (Sarwar *et al,* 2003).

Phosphine and other space fumigants have also been used to control insects and fungi in wood products. Phosphine has been shown to be an effective in-transit fumigant for wood chips; however, this technology has never been commercialized, possibly due to the highly flammable nature of the chemical (Leesch *et al*, 1989). Liquid fumigants that are used to treat large timbers in service have also been studied, but the time required for the chemicals to diffuse, and, thus become effective is prohibitive in relation to SWPM uses. Other drawbacks of these chemicals include disposal, their high mammalian toxicity, and the tendency to remain in the wood for long periods (Morrell and Corden, 1986).

Fumigants are a potent tool for the sanitation of SWPM. The chemicals are fast acting and generally do not alter the color or the mechanical properties of the material. However, this method of sanitizing SWPM has some of the same drawbacks as those associated with HT. None of the chemicals currently being considered provide long-term protection against re-invasion by fungi or insects. Fumigation of SWPM cannot be verified at the port of arrival, thus importing countries must rely upon a paper trail to verify treatment. MB, the fumigant with the longest track record and recommended by IPPC, is currently being phased out due to environmental concerns. While new fumigants are being developed, none are as effective as MB and most tend to be highly flammable. Finally, the high mammalian toxicity of all of these chemicals should encourage a search for less toxic mitigation methods.

Emerging Treatment Methods

In recognition of the problems associated with the current treating methods approved in IPSM#15, the IPPC allows for amendment of the guidelines to include new treatment methods (FAO 2002). A number of different treatments have been proposed including irradiation, microwave treatment, dip-diffusion methods and pressure treatment. All of these methods have advantages and disadvantages in comparison with the currently approved methods.

Irradiation

Gamma radiation (GR) has been successfully used to remove pests from a large number of materials, such as paper, pharmaceuticals, foods and other materials (Morrell, 1995, 1996; Keeney and Walkinshaw, 1990). Radiation has no effect on the mechanical or durability properties of dry wood (Scheffer, 1959; Kenaga and Cowling, 1959; and Becker and Burmester, 1962). Doses ranging from 73 to 130 krad are needed to eliminate the ambrosia beetle from wood; however, death is not instantaneous and it may be weeks before the insect perishes (Yoshida *et al,* 197 and 1975, and Schröder 2005). Only two to four krads are needed to sterilize the ambrosia beetle; however, sterilization does not prevent insects from vectoring fungi or nematodes from the SWPM into the surrounding environment (Morrell, 1996).

The amount of radiation tolerated by an insect is also highly dependent on the life cycle stage, with the amount of irradiation an insect can tolerate progressively increasing from egg to adult (Dwinell, 2001d). Eradicating fungi and nematodes through irradiation requires much higher doses than those needed to eliminate insects. A dose of 2.5 Mrad is recommended for fungi (Keeney and Walkinshaw, 1990). Doses of up to 1.2 Mrad are needed to eradicate the PWN (Dwinell, 2001d). Since radiation doses are time and source dependent, fungi or nematode mitigation requires either a longer time period or a stronger radiation source (Morrell, 1996).

Irradiation also does not solve the problem of being able to confirm treatment days or weeks after it has been completed, since the appearance of the wood is not altered (Morrell 1996). GR also does not appear to prevent the re-invasion of the material by insects or fungi following the treatment, thus similar precautions as those associated with HT and fumigation must be followed. There is also a knowledge gap associated with the radiation doses needed to eliminate insects and fungi in green material. It has been assumed that the doses required in green wood must be higher than those used in previous studies where dry wood specimens were utilized; however, organisms are more likely to be active in wet wood and are therefore more vulnerable to the treatment (Morrell, 1995). Additionally, a number of reports have emphasized the economic costs associated with irradiation and concluded that the high costs associated with constructing and operating large irradiation chambers make their use for a low value commodity such as SWPM cost prohibitive (Morrell, 1996, 1995; Dwinell, 2001d, Schröder 2005a). The cost of irradiating material has been estimated to be up to 60 times higher than fumigation (Cornwell, 1966).

Irradiation remains promising because it does not alter the wood properties and it has a proven track record. Further research must be done to reduce the cost of irradiating commodity products. Additionally, irradiation has the same problems of verification and prolonged protection that are associated with the methods currently approved for treating SWPM.

Microwave treatment

Microwave or radio frequency treatment of wood is an alternative form of traditional HT. Mortality of the PWN during radio frequency drying was mainly a function of wood temperature (Dwinell *et al,* 1994). Renewed interest in this technology has developed in response the ALB outbreaks in Illinois and New York. The major advantage of this technology over conventional HT is the speed at which the material can be heated. A 10 cm by 10 cm by 10 cm sample of aspen (*Populus tremuloides*) above the FSP requires 7380 seconds to reach 60°C at the center with conventional heat compared to as little as 120 seconds when a 900 watt microwave source is used (Fleming *et al,* 2003). Material 2.54 cm thick and 10 cm wide required only 30 seconds in a 900-watt microwave field to kill the ALB (Fleming *et al,* 2003). In later studies, Fleming and others found that a 2.45 GHZ microwave energy source could be used to eradicate cerambycid larvae (*Plectrodera scalator*) from red pine (*Pinus resinosa*) samples. Researchers are currently verifying these results and scaling-up the technology for commercial-size loads (Fleming *et al,* 2004). However, the promise of this technology for rapid production of phytosanitary SWPM is offset by some down sides. Heating rate is highly dependent on the moisture content of the material. Thus, it may take longer to heat drier material, since materials with different moisture contents are often intermixed in the same load all materials would have to be treated as if they were dry. Heating also originates below the surface of the material, resulting in cooler temperatures on the exterior of the SWPM, were many organisms are found. Microwave treatment has been shown to cause cracking of the heartwood, resulting in strength losses (Schröder 2005b). This method is no different from conventional HT in terms of the product it produces, meaning that it is impossible to verify treatment at the importing country and treatment does not give the SWPM long term protection.

Dip-diffusion methods

Dip-diffusion involves application of chemicals to the surface of the material by spraying, dipping or soaking the material in a solution containing one or more biocides to produce either a layer that prevents insect and fungal attack or that diffuses throughout the wood. Dip treatments also kill any insects and fungi found near the surface of the wood product, while inhibiting insect emergence. The depth of treatment varies widely, from less than 1mm in some species to complete penetration in others depending on the wood species, percentage of heartwood, MC and the chemical formulation. The depth of the treatment within the same piece of lumber also varies widely since the cross sections absorb more chemical than radial or tangential faces. Differences in the depth of the protective layer created by this method pose one of the challenges that must be overcome by researchers before dip diffusion is commercialized for SWPM treatment (Morrell, 2001a).

Dip-diffusion treatments have some distinct advantages including rapid treatment, low capital costs for equipment, the ability to be verifiable and the ability to provide protection that outlasts that provided by fumigation or HT. The chemicals used in this process are of concern, however, since a majority of used SWPM does not enter landfills, but instead is used for other purposes such as cooking fires. In the past, broadly toxic pesticides and fungicides have been used for dip treatments. Increased concerns relating to human and environmental health effects have encouraged the use of less broadly toxic chemicals. Some of the chemicals used in dip-diffusion include copper-8-quinolinolate, 3-iodo-2-propynyl butyl carbamate, didecyldimethyl ammonium chloride, and boron (Morrell, 2001a). Boron compounds are of greatest interest since they pose a low mammalian hazard, affect most insects and fungi and, most importantly, have the ability to diffuse deeply into many woods. Boron is also well understood. Borate diffusion can be achieved even in hard to treat species such as Douglas-Fir (*Pseudotsuga menziesii*) (Fowlie *et al,* 1988); however, the treatment must be done shortly after milling for the material to reach boric acid concentrations that will prevent decay (Carr, 1955). The same studies have also found that the time required for adequate diffusion to occur normally exceeds one month, which would be a problem when treating SWPM (Fowlie *et al,* 1988; Carr 1955; MacLean 1962). Boron solutions also tend to experience increased mold growth, however, this problem can be solved by combining boron with other elements (MacLean, 1962). An additional advantage of boron compounds is that the presence of the chemical can be detected using indicators, making it easy to verify the treatment. However, boron also has the drawback of being water-soluble and, thus, is susceptible to leaching if the SWPM is stored outside (Morrell, 2001a; MacLean, 1962; Fowlie *et al,* 1988).

Material treated by dip diffusion has two major advantages over the currently approved methods, it is verifiable, especially if a dye indicator is used in the treating solution, and it provides protection over an extended period of time. However, this method also has some drawbacks including lack of data showing that the treatment prevents emergence of established insects and fungi from the material. The use of chemicals also requires special care during disposal. For example, low temperature cooking fires may not completely combust insecticides and fungicides, thus making them available for human consumption (Morrell, 2001a). However, this problem can be overcome by choosing chemicals with low mammalian toxicity such as boron.

The risk of damaging the treated layer of wood is also a concern since loss of the protective barrier may allow renewed attack, while leaching may reduce the amount of protection. Despite these drawbacks, dip diffusion probably merits further testing as a SWPM mitigation tool.

Pressure treatment

Pressure treatment overcomes some of the problems associated with dip diffusion by forcing the treating chemical deeper into the wood. Pressure treatment has been in use since 1838 when the Bethel or full-cell process was patented. Since that time, numerous oil- and water-based chemicals have been employed to protect wood against decay using this process. Indeed, the pressure treatment process is well understood in regards to its ability to prevent decay in sound timbers (Hunt and Garratt, 1938). Pressure processes are used to treat nearly 360 million cubic meters of wood annually in NA, prolonging the service life of this material from a few years to decades (Morrell 2001b; Nicholas, 1973). During the past one hundred years, a majority of the research pertaining to this field in North America has been published in the annual proceedings of the American Wood Preservers Association (AWPA), which has resulted in standardized treating processes that are well understood and effective. The treating capacity of this industry is large, allowing it to rapidly treat packaging material used in international trade at a relatively low cost. As with dip diffusion treated material, the treatment would be verifiable by the importing country and the treatment would give the SWPM prolonged protection, reducing the risk of re-infestation following treatment. Pressure treatment has added benefits over dip diffusion treatment including uniformity and quality of treatment. The barrier created using the pressure method is generally much thicker, reducing the risk that the protective layer will be damaged to expose untreated wood. The thicker barrier also decreases the likelihood that an existing pest will exit the material in a viable state. In addition, pressure may force pesticides into the insect galleries eradicating them in situ (Morrell, 2001b). The treating process is also relatively rapid, reducing the amounts of inventory a SWPM producer would need to have on hand (Nicholas, 1973).

The pressure treatment process has considerable potential for mitigating pest risks on SWPM, but there are some knowledge gaps and disadvantages. There is little knowledge concerning the emergence of established insects or fungi from treated material. Members of the Buprestidae are known to continue development in pressure treated Douglas-fir poles and emerge through the treated shell. There is currently no evidence supporting the ability of this method to penetrate insect galleries, especially those that are tightly packed with frass. Pressure treatment tends to provide protection by placing high chemical retentions near the surface to provide decades of performance. This is unnecessary for SWPM since it does not come into ground contact, it is not exposed to highly decay prone environments and its service life is relatively short. As a result it may be possible to use less toxic chemicals at lower chemical retentions to reduce both costs and the potential environmental impacts of treated SWPM (Morrell, 2004).

Pressure treatment also has some drawbacks. It introduces potentially dangerous chemicals into the fuel wood supply chain. This problem can be addressed by using combustible chemicals with low mammalian toxicity. However, many of these chemicals have not been completely investigated for this application. In addition, the treatment envelope can still be damaged on pressure treated wood during dunnage production. The current solution to this problem is the application of diffusible preservatives to drilled and cut areas but this would be difficult in a production operation. Another major disadvantage of the pressure treatment process is that differences in permeability between species will result in different depths of treatment. This can be reduced by incising refractory species, however, incising reduces the strength of the material. Thus, incising may not be feasible for all SWPM applications. Inadequate protection in some species could allow for the continued movement of insects and fungi.

Pressure treatment of SWPM is an alternative to HT and fumigation with some distinct benefits; by virtue of deeper and longer lasting protection compared to the other proposed methods. However, some knowledge gaps must also be filled before this method can be applied to SWPM. These include determining chemical levels necessary to give adequate protection and assessing the ability of treatments to eradicate established insects and fungi.

Detection of Wood Boring Insect Larvae Using Acoustic Emission Detection Methods

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DETECTION OF WOOD BORING INSECT LARVAE USING ACOUSTIC EMISSION DETECTION METHODS

Abstract

Non-destructive methods to detect the presence of wood boring insects within solid wood packaging materials (SWPM) are being sought by both scientists and regulatory officials in order to prevent the introduction of unwanted species. Acoustic emissions can be used to detect the presence of termites within forest products through the use of sensitive microphones and accelerometers. The use of this technology provides a rapid, non-destructive method for detecting the presence of an infestation and a means of estimating its size. This same technology was applied to lumber with known wood boring insect larvae infestations to determine if the rasping noises produced by these organisms during feeding could be captured using either a microphone or an accelerometer. Work indicated that wood boring insect larvae did not feed continuously, and, thus, were difficult to detect using this technology. The results showed that acoustic methods were less suitable for detecting wood boring beetles than they were for assessing termite damage in wood products.

Introduction

Non-destructive methods for detecting insect larvae within logs, lumber and finished products are highly desirable. Such methods would allow researchers to monitor the feeding and movements of these insects in a non-intrusive manner. These same tools are also of interest to government inspectors seeking methods for rapid detection of invasive species found in various wood-based shipping materials. In addition, the forest products industry could use such methods to determine if wood inhabiting insects are present in their raw material stream.

A variety of studies have shown that acoustic emissions can be used to detect termites in wood products (Markin et al 2002, Schaffrahn et al 1993, Lemaster et al 1997, Fujii et al 1990, Noguchi et al 1991 and Thoms 2000). Similar approaches using acoustic monitoring have also been employed to detect insect larvae in agricultural products such as wheat or grapefruit (Webb and Landolt 1984, Cross and Thomas 1978, Webb et at 1988 and Hansen et al 1988). Thus, acoustic emissions may also be a useful method for the non-destructive detection and monitoring of wood boring insects.

Acoustic methods detect the rasping noises produced by the insects as they excavate passages within the medium in search of nourishment or shelter. The large number of termite workers found within one colony make foraging a continuous process, resulting in nearly continuous acoustic emissions. These emissions can be detected using microphones, or through the use of accelerometers. These signals are then amplified and displayed in the form of a wave for analysis. Similar techniques could be applied to wood boring insects since they also excavate passages within the wood. This study attempted to assess the potential for using acoustic emission detection technologies to detect the presence of wood boring insect larvae within solid sawn lumber.

Methods

A sound and vibration isolated environment was constructed using a concrete slab measuring 1.21 by 0.63 by 0.12 m thick and weighing approximately 225 kg. This cement block rested on four foam pads which were supported by a reinforced shipping crate. Sound isolation was achieved by lining a second shipping crate with soft egg carton foam and placing it over the cement slab. The support for the sound proofing structure was the same as that for the vibration isolating cement slab (Figure 1). Samples were inserted into the sampling chamber through a hatch located at the top of the upper shipping crate. The hatch was secured during monitoring with a screw to prevent acoustic pollution from penetrating the sampling environment.

Emissions from insect activity were collected using an omnidirectional microphone with a response frequency ranging from 30-15,000 Hz and a sensitivity of -68dB ± 3dB and a Kistler Instrument Corporation (Amherst New York) K-shear accelerometer type 8712A5M1 with a sensitivity of 1000mV/g and an frequency range of ± 5Hz. The accelerometer signal was amplified ten fold using a Kistler Instruments Company Piezotron Coupler type 5122 prior to signal processing using a personal computer. The signals generated from both the microphone and the accelerometer were processed using the integrated audio hardware found on an Intel motherboard utilizing an Intel

Pentium 3 processor with 998 MHz and 512 MB of RAM. The signal was captured and analyzed using GoldWave Shareware sound editing software (GoldWave Inc. St. John's, Newfoundland, Canada).

Figure 1. Interior of the acoustic sampling chamber with an insect infested board in place. The bottom of the sampling chamber consisted of cement to reduce ambient vibrations while the walls were lined with egg carton foam to dampen exterior noise.

The acoustic emissions detection system was tested using damp wood termites *(Zootermopsis angusticollis*) collected from an indigenous colony located in western Oregon. The insects were placed in a plastic storage container along with small, moistened wooden blocks that served as a food source. Testing commenced once the insects had started to inhabit the wooden blocks. The plastic storage container was placed on top of foam pads within the sampling chamber. An accelerometer was then screwed into the side of a termite infested block. The signal produced by the termites consuming the wood was recorded for forty-five minutes during daytime hours. The recorded signal was analyzed by removing background noise from the data using a high band pass filter with an initial cut off of 1000 Hz. The final signal was then stored in an uncompressed format in order to preserve the maximum amount of information.

The microphone tests were performed separately by affixing the microphone to the surface of a wooden block using putty. Prior to filtering, the signal strength was doubled using an integrated algorithm found within the software package. This allowed for better differentiation between ambient noises in the gathered data and those caused by the termites rasping on the wood. A high band pass filter with a 500 Hz initial cut off was then used to reduce the amount of ambient noise. Individual acoustic events could be detected, after the ambient noise had been removed, by listening to the recorded data and by examining the graphical output. The refined data were then stored in an uncompressed format.

Attempts to detect beetle larvae in wood were conducted using materials that had been naturally infested with wood boring insects. Three samples (230 to 300 mm wide by 25mm thick by 500mm long) were gathered fourteen days prior to this work from a sawmill located in central Oregon. All of the samples had visible beetle galleries on the board surfaces. The lumber was sprayed with water to maintain a wood moisture content well above the fiber saturation point in an effort to keep the larvae within the material viable. The accelerometer was screwed into the side of the sample, parallel to the grain. Once the accelerometer was in place, the sample was placed on top of two foam supports located at either end of the sample within the sampling chamber. The samples were then monitored for several forty-five minute sessions at various times of the day. Longer sampling periods were not feasible due to data collection limitations. The data were then analyzed and stored using the same procedures used for the termite studies.

Once data collection and analysis were complete, the samples were dissected into 10mm thick slices, perpendicular to the grain to verify the presence of insects within the material. The number of insect larvae detected during this process was recorded.

Results and Discussion

Termites were readily detected in woody materials using the instrumentation described. Individual acoustic events could be detected by examining the audible or graphical output produced by the software package. The data gathered using the accelerometer clearly differentiated ambient noise and vibration levels from those produced by the termites (Figure 2). The signal produced by the termites could be isolated by visually examining the graphical output of the data; events corresponding to termite rasping were characterized by spikes in the graphical display that far exceeded the spikes from ambient noises. These spikes in the data were verified as relating to termite activity by listening for rasping noises in the audio portion of the data file. Following data analysis of the microphone data, acoustic signals produced by the termites could also be audibly differentiated from the ambient noise. However, acoustic events produced by the termites and recorded by the microphone did not produce the large spikes in the visual rendering of the data that characterized such events detected using the accelerometer (Figure 3).

Although the test boards contained four to seven beetle larvae per board (Table 1), the accelerometer data collected from each board showed no consistent spikes that might suggest larval mining (Figure 4). While the larvae detected in the boards were alive, their activities in the wood differed markedly from termite workers. While termite workers feed nearly constantly, beetle larvae appear to feed more sporadically. This behavior makes it difficult to accurately detect beetle presence over short time periods. The use of long time periods might be useful, but this would reduce the value of this technique for rapid insect detection.

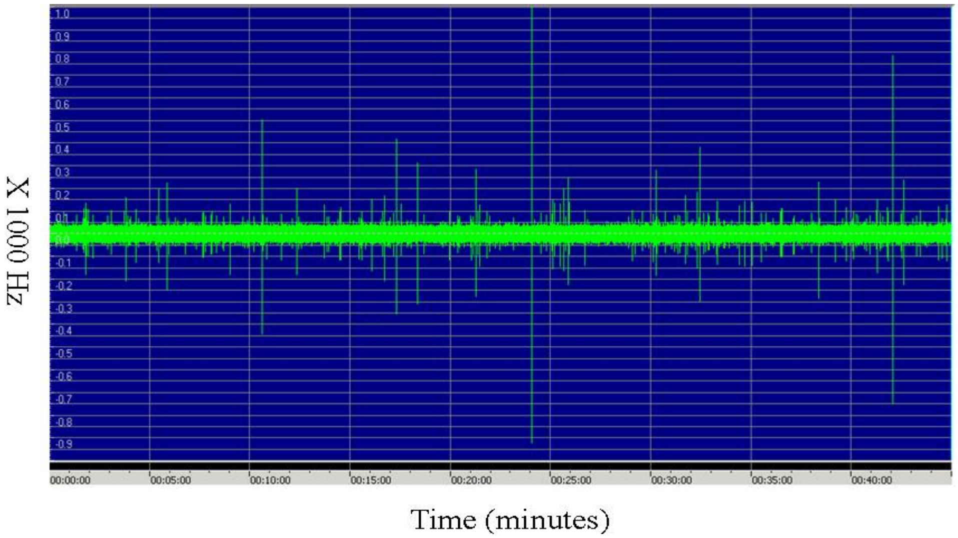

Figure 2. Accelerometer output from a ponderosa pine (*Pinus ponderosa*) sapwood block containing dampwood termite workers. Spikes correspond with rasping noises in the audio portion of the data. These rasping noises are associated with individual termite mining events.

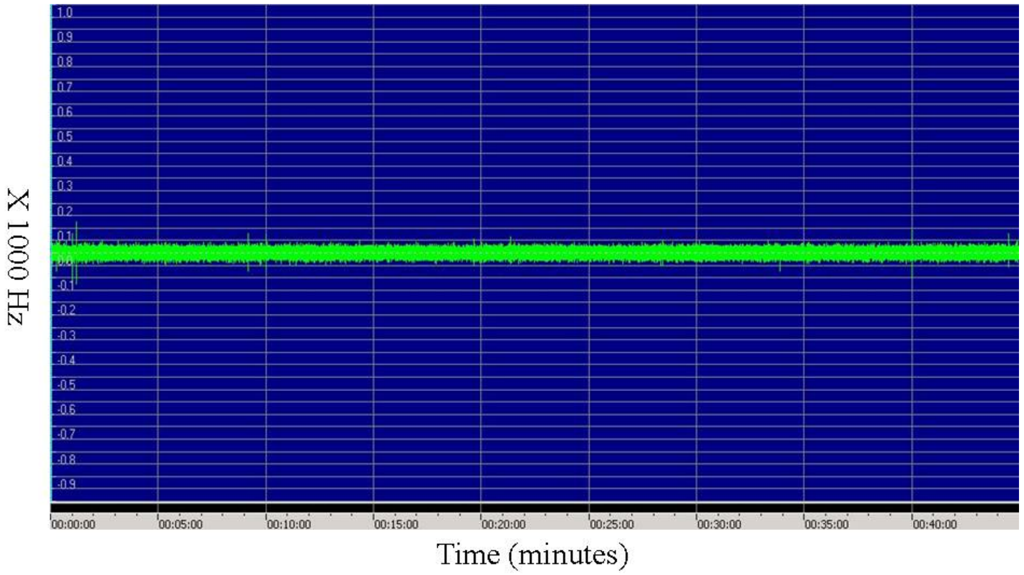

Figure 3. Microphone output from a ponderosa pine sapwood block infested with dampwood termites. The absence of large spikes illustrates the reduced sensitivity of this detection method.

Sample #	Width (m)	Number of insect larvae found
1	0.23	5
2	0.30	4
3	0.25	7

Table 1. Number of larvae, as determined by dissection, present in each of the three test boards used to assess acoustic detection.

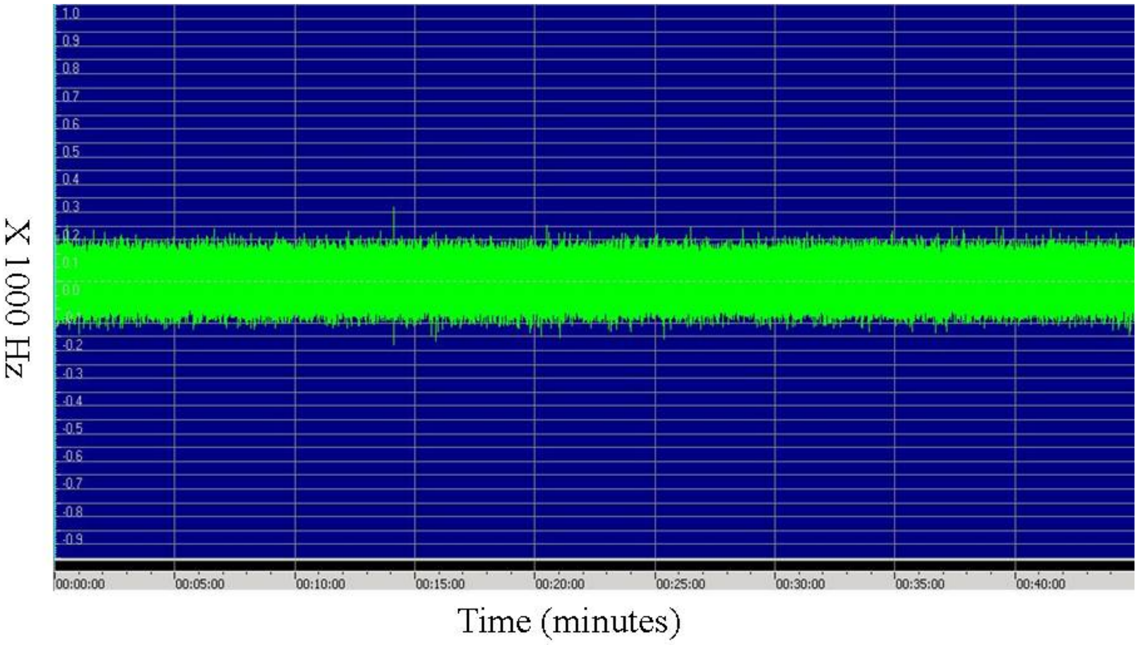

Figure 4. Accelerometer output from a naturally beetle infested board (sample number 3) which contained seven beetle larvae. The output shows no evidence of insect related activity.

Conclusions

The results support the findings of other researchers that termite activity in woody material can be detected using acoustic emissions. However, the feeding behavior of wood boring beetle larvae differed greatly from that of termites in being more sporadic. Thus, acoustic monitoring of woody materials is not a reliable method for detecting the presence of the insect larvae within the materials, unless very long test times are used. These long times would limit the ability to test many samples, limiting use of this method as a pest detection tool.

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Penetration of Insect Galleries in Western Redcedar by Water Borne Wood Preservatives Using Pressure Treatment Processes

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PENETRATION OF INSECT GALLERIES IN WESTERN REDCEDAR BY WATER BORNE WOOD PRESERVATIVES USING PRESSURE TREATMENT PROCESSES

Abstract

Heat treatment and fumigation are currently the only two recognized methods of sanitizing solid wood packaging material under International Phytosanitary Measure #15. However, wood preservatives may be able to achieve the same level of protection against invasive species introductions, since they are specially formulated to prevent fungal and insect attack in wood products. These preservatives and the vacuum pressure method of applying them are well understood, but the ability of these chemicals to penetrate existing insect galleries has not been investigated. The ability of four wood preservatives to penetrate frass-packed galleries in refractory western redcedar was investigated. ClearWood MW1 penetrated 100% of the insect galleries found within the samples, while disodium octaborate tetrahydrate penetrated 92% of the galleries and ammoniacal copper quaternary compound penetrated 87.9% of the galleries. An experimental compound consisting of 30 parts per million (ppm) imidacloprid, 300ppm 2-n-octyl-4-isothiazolone-3-one and 100ppm green dye penetrated 90.7% of the galleries. These results indicate that wood preservatives applied using the vacuum pressure method can penetrate a majority of the insect galleries found even in refractory species. However, complete penetration cannot be guaranteed. Thus, the

treatments must also provide an effective barrier to prevent wood boring insect adults and larvae from exiting the material.

Introduction

The ability of different wood preservative systems to penetrate various timber species is well understood, with research dating back to the early twentieth century (AWPA 1917; Allerton and Allardyce 1911; Weiss 1912; MacLean 1926, 1927, 1928, 1952; Weaver and Levi 1979; Kumar and Morrell 1989; Lebow et al 2005). These findings have led the American Wood Preserver's Association (AWPA) to prescribe minimum preservative penetration depths for wood exposed under varying environmental conditions (AWPA 2004b). Historically, however, the ability of a wood preservative solution to penetrate insect galleries has received little attention. This lack of information reflects a tendency to avoid treating materials with insect damage. Treated wood is a value-added product normally used in areas where its strength properties and aesthetic qualities are important. However, data pertaining to the ability of chemicals to penetrate insect galleries is important where preservative treatments are used to eliminate beetles and other insects from wood in phytosanitary applications. Insect larvae in wood can be problematic because some species tend to pack their galleries with frass that can limit ingress of chemicals and allow the larvae to survive the treatment. The ability of the preservative to penetrate the frass-packed insect galleries is vital since these pathways allow for direct contact between the larvae and the biocides used to control them.

Preservative movement through insect galleries is especially important in refractory species since the diffusion and capillary action processes that help achieve chemical penetration in more permeable woods may be insufficient to penetrate insect galleries located deep within a sample. This study was initiated in order to assess the ability of different wood treating systems to penetrate insect galleries in western redcedar (*Thuja plicata* Donn ex D. Don*)*, a refractory species, utilizing commercial treating conditions (McLean 1952).

Methods

Western redcedar heartwood planks were collected in their green condition from a sawmill located in Coos Bay, Oregon. The selection criterion for the material was the presence of insect galleries caused by *Trachykele blondeli* Marseul, a Buprestidae that invades the heartwood of living redcedar from British Columbia through California and New Mexico (Furniss and Carolin 1977). These insects tend to pack their galleries with frass, which limits fluid ingress. The boards were cut into 150mm wide by 50mm thick by 300mm long pieces that were end sealed with a marine epoxy. Four groups of 30 samples were then randomly selected from the prepared boards. The first group was treated in the green condition using ammoniacal copper quaternary compound (ACQ) to a target retention of 4.0 kg/m^3 utilizing a commercial treating facility, while the second group was treated in the green condition with Timbor Industrial (disodium octaborate tetrahydrate, U.S. Borax Inc, Valencia California) at a commercial treating facility to a target retention of 6.72kg/m^3 of boron oxide. Treating parameters for both treatments can be found in Table 1.

The samples were returned to Oregon State University and the end sealant was cut off. After drying, each sample was cut lengthwise into 10mm wide strips (Figure 5). These smaller strips were labeled with the sample number and a location code, allowing for reassembly of the samples at a later time. This material was then oven dried according to American Society for Testing and Materials (ASTM) Standard D4442-92 (ASTM 1997) to limit further movement of the treating chemicals. Following drying, the cut surfaces on the thirty ACQ surfaces were sprayed with Chrome Azurol S following AWPA Standard Method A3-00 for determining penetration of copper containing preservatives (AWPA 2004a). The presence of copper was indicated by a dark blue color. Each sample was photographed using a five-mega pixel digital camera under natural light conditions, minimizing shadows. These images were used to assess preservative penetration in beetle galleries.

Figure 5. Cutting diagram for the penetration samples showing the widest face of the treated cedar block and the location of the cuts to produce 10mm thick strips. The dissection of the samples in this manner allowed for the assessment of preservative penetration into the insect galleries through the sample.

The penetration measurement process was repeated for the boron samples with the following changes. The samples were sanded with an orbital sander utilizing 100 grit sand paper in order to remove any borates that had been smeared across the surface of the samples during sawing (the boron indicator is extremely sensitive). The presence of boron was then determined according to AWPA Standard Method A3-00 for determining penetration of boron-containing preservatives and fire retardants (AWPA 2004a). This indicator turns red in the presence of boron.

Two additional treatments were evaluated for their ability to penetrate insect galleries because of their low toxicity to non-target organisms. ClearWood MW1 (Chemical Specialties inc. Charlotte North Carolina) contains both tebuconazole and imidacloprid and is used to treat millwork. However it is heated to temperatures in excess of 56 °C, meaning that materials treated with this chemical would meet current heat treatment requirements outlined in International Phytosanitary Measure 15 (Food and Agriculture Association of the United Nations 2002). Heating the treating solution is necessary for solution stability and also increases chemical penetration into the wood. The samples were treated in a commercial treating plant and then cut into 10mm wide strips without drying, since there is no chemical indicator to determine the presence of this chemical. Sections were photographed using an 8 mega pixel digital camera under natural daylight conditions to reduce the effects of shadows. Areas that were not treated (i.e. appearing dry) were later highlighted through the addition of a black line using Adobe Photoshop version 8.0.

The remaining system evaluated for this study was an experimental treating solution containing 30 parts per million (ppm) imidacloprid, 300ppm 2-

n-octyl-4-isothiazolone-3-one and 100ppm green dye (Sensient Colors Inc St. Louse Missouri, Alizarine Cyanine Green G-AF). The dye served as an indicator for the presence of the other chemicals in the samples. Treating parameters can be found in Table 2. The treated samples were dried and then cut into 10mm thick slices as described for the ACQ and borate treated samples. The presence of the green dye was used to determine treatment depth. The samples were photographed using an 8 mega-pixel digital camera under natural light conditions, and then the resolution was enhanced by intensifying the green color range using the select color range feature in Adobe Photoshop version 8.0 (Adobe, 2003).

Table 2. Process conditions used to treat western redcedar with ACQ, boron, or the experimental formulation.

Treatment	Treating solution temperature (°C)	Vacuum cycle		Pressure	
		Vacuum achieved (kpa)	Duration (s)	Pressure achieved (kpa)	Duration (s)
ACQ	29	50.8	600	965.2	6300
Borate	27	84.6	600	1103.2	1200
Experimental formulation	22	86.4	600	1241.1	1800

The number of insect galleries visible on each 50 x 300mm face was counted along with the number of galleries that were completely penetrated by the treating chemical. These values were summed for the 15 sections from each piece. Samples that did not contain insect galleries were removed from the data set used for the statistical analysis but were included when determining the thickness of the treated shell. These penetration values were then subjected to an ANOVA comparison using the PROC MIXED function in SAS version 9.0 (SAS Institute, $\alpha = 0.05$). The results were also used to calculate an overall mean for the number of insect galleries penetrated across all chemical treatments.

Results and Discussion

The treated shell on all of the ACQ treated samples was very shallow, confirming the refractory nature of the material. The penetration depth was much less than the 10mm penetration required under AWPA Standard U1-04 (AWPA 2004b). Incising which is required by the AWPA for treatment of many refractory species, would have helped improve treatment, but would also increase the cost of treating SWPM and reduce the strength of the final product. Despite the lack of incising, the treating process did completely penetrated 88% of the insect galleries found in the 18 samples with beetle galleries (insect attack in the remaining 12 samples was limited to within 10mm of the surface of the blocks, thus these blocks were not used in this analysis, Table 3). There was no evidence that the copper component of ACQ diffused away from the treated galleries into the surrounding material (Figure 6). Thus, the presence of insect galleries greatly increase the overall preservative coverage through the depth of the sample but the increase was defined by the insect galleries. The lack of diffusion into the surrounding wood combined with the lack of a thick treated shell might allow insect larvae that survived the initial treatment to complete their life cycle and emerge from the material by avoiding intermittent treated pockets, pupating and then penetrating the thin layer of treated material found at the surface, increasing the risk of an invasive species introduction.

Table 3. Percent of galleries in western redcedar boards penetrated by ACQ, borate or tebuconazole / imidacloprid based preservative systems.

Treatment	Average (standard deviation)	Range	Lower 95% confidence limit	Upper 95% confidence limit
ACQ	87.9 (11.0)	75.7-100	81.5	94.4
Borate	92.9 (11.4)	69.5-100	87.6	98.2
Experimental treatment	90.7 (22.7)	57.1-100	84.9	96.6
ClearWood MW1	100 (0)	100-100	94.1	100

Figure 6. Example of ACQ penetration in western redcedar with *T. blondeli* galleries. The black color was caused by a reaction between Chrome Azurol S and the copper component of ACQ indicating that a majority of the galleries were penetrated by the preservative. Note that the chemical did not diffuse away from the galleries.

The treated shell found on the borate treated samples was slightly thicker than that found on the ACQ treated material. However, the treated shell on these samples was still less then 10mm thick. Of the 27 samples that contained insect galleries, 93% of the insect galleries in these boards were completely penetrated by the borate treating solution (Table 3). This treatment also differed markedly from the ACQ treatment in that the borate diffused away from the initially treated insect gallery into the surrounding wood, greatly increasing preservative coverage (Figure 7). This diffusion process led to a variety of different levels of preservative coverage ranging from complete coverage of the cross section to narrow longitudinal streaks away from a penetrated gallery. The greatest coverage was seen in samples that contained more than 4 insect galleries, while the lowest amount of additional preservative coverage was seen in samples were the galleries only penetrated a short distance into the sample interior.

Figure 7. Example of boron penetration of western redcedar with *T. blondeli* galleries. The red areas indicate the presence of boron after an indicator has been applied. A majority of the insect galleries were penetrated by the chemical and chemical diffused away from the penetrated galleries in the longitudinal direction, increasing preservative coverage.

ClearWood MW1 was unique among the treatments investigated in that it penetrated the complete cross-section in 20 of the 30 samples treated and completely penetrated all of the insect galleries (Table 3 and Figure 8). The heated treating solution was most likely the cause for this increased penetration.

Figure 8. Example of penetration of the ClearWood MW1 treatment in western redcedar showing that chemical penetrated the entire cross section in a majority of the samples regardless of the presence of insect galleries. Areas within the black lines were not penetrated with ClearWood MW1.

The green dye amended treatment containing 30 parts per million (ppm) imidacloprid, and 300 ppm 2-n-octyl-4-isothiazolone-3-one produced a treated shell approximately equivalent to the ACQ treatment, but more insect galleries were penetrated with the ACQ treatment (Table 3). This treatment also displayed the greatest penetration variability of the four treatments with penetration ranging from 57 to 100% of the galleries (standard deviation of 23%). The increased variability may reflect the use of multiple treating charges although the treatment time and pressures were the same for each charge. Some diffusion away from the treated insect galleries was also noted;

as evidenced by preservative streaks in the longitudinal direction. However, diffusion in the tangential and radial direction was limited (Figure 9).

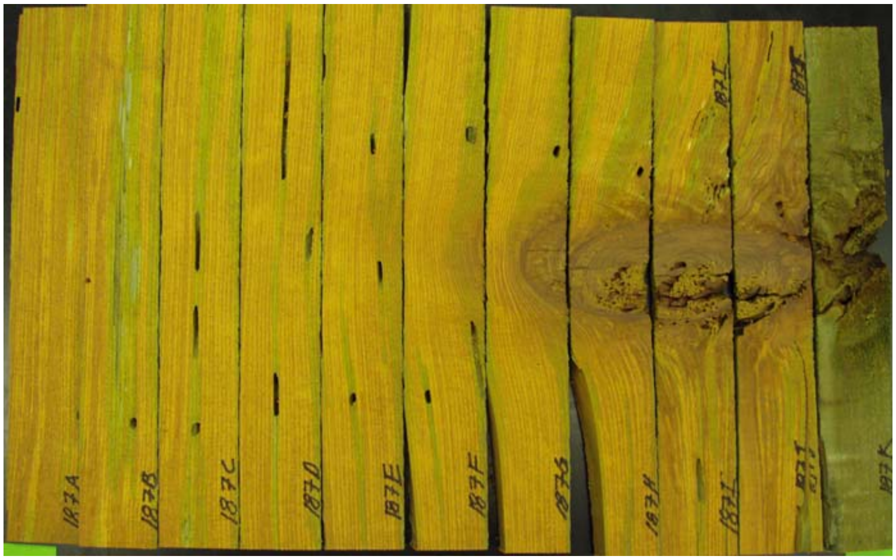

Figure 9. Example of penetration of an experimental treatment in western redcedar showing that a majority of *T. blondeli* galleries were penetrated, as indicated by the green color surrounding the black insect galleries. The green streaks moving away from the insect galleries suggest that some diffusion occurred in the longitudinal direction.

ClearWood MW1 was used as the reference level for the ANOVA analysis because of the high degree of preservative penetration (Table 4). There were significant statistical differences in the percentage of the galleries completely penetrated between the ClearWood MW1, and ACQ (p-value 0.007, with 85 degrees of freedom (D.F.)). Gallery penetration was significantly greater in ClearWood MW1 than the experimental treatment (p-value 0.0291, 85 D. F.). There was no significant difference between penetration in the ClearWood MW1 and boron treatments, nor did penetration differ significantly between ACQ treated material and the experimental treatment.

Table 4. ANOVA results comparing penetration of ACQ, borate and experimental treatments vs. ClearWood MW1 treatment in insect galleries in western redcedar lumber.

Treatment	Standard Error	Degrees of Freedom	T statistic	P-value
Intercept	0.0295	85	33.92	<0.0001
ACQ	0.0439	85	-2.74	0.0074
Borate	0.0397	85	-1.78	0.0790
Experimental treatment	0.0417	85	-2.22	0.0291

Conclusion

The four waterborne treating solutions investigated in this study completely penetrated a majority of the insect galleries found in western redcedar. However, not all galleries were completely penetrated. Statistically significant differences in the treating efficiency between some of the treating systems were found. The ACQ wood preservative system and the experimental treatment containing 30 parts per million (ppm) imidacloprid, with 300 ppm 2-n-octyl-4-isothiazolone-3-one were less likely to completely penetrate insect galleries than the other two treatments. The treated shell created through the use of the ACQ, boron or the experimental treating system would not meet AWPA requirements of 10mm of penetration for lumber, although the actual depth of the preservative penetration required to inhibit larval movement or adult emergence remains unknown. The ClearWood MW1 system achieved more than 10mm of penetration in all samples, probably due to the use of heated treating solution.

Water borne wood preservatives penetrated a majority of tightly packed insect galleries in western redcedar. However, further studies are needed to investigate the effectiveness of other treating solutions containing specific insecticidal or larvacidal agents. In addition, studies focusing on the minimum thickness of treated material needed to prevent the emergence of adult insects should also be conducted.

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The Ability of Wood Preservatives to Prevent Invasive Species Introductions in Field Conditions

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THE ABILITY OF WOOD PRESERVATIVES TO PREVENT INVASIVE SPECIES INTRODUCTIONS IN FIELD CONDITIONS

Abstract

Current methods of sanitizing solid wood packaging materials before employing them in international trade are imperfect, since they cannot be verified by the importing country and they do not provide lasting protection against new insect and fungi attack. Wood preservatives, applied using the pressure processes, may provide an alternative to heat treatment or fumigation. Pressure treating solid wood packaging material provides long-term protection and the treatment is verifiable by the importing country. Properties of the chemicals employed are well understood, but their ability to eliminate established wood boring insects has not been investigated. Ponderosa Pine samples infested with *Arhopalus productus* were treated with disodium octaborate tetrahydrate, ammoniacal copper quaternary compound or an experimental treating solution containing 30 parts per million (ppm) imidacloprid, 300ppm 2-n-octyl-4-isothiazolone-3-one and 100ppm green dye. The samples were then monitored for beetle activity every four weeks for one year. Wood preservatives prevented adult insect emergence from treated wood, but larval activity was not controlled. The results suggest that wood preservatives can be used for phytosanitary applications since the predominant invasive species threat associated with wood boring insects is linked to the adult beetle. However, current chemical formulations should be

amended in order to prevent the introduction of invasive species through larval movement.

Introduction

The risk of introducing invasive species through solid wood packaging materials (SWPM) infested with wood inhabiting beetles is of great concern to the United Nations Food and Agriculture Organization (FAO). The FAO currently recommends heat treatment or fumigation to mitigate this risk; however, neither of these treatment methods provides protection against later reinfestation. In addition, neither of these mitigation methods are independently verifiable by the importing countries. There is a continued need for mitigation treatments that are broadly effective at eliminating established pests, prevent reintroductions of pests and whose presence can be verified during the entire life cycle of the material. Pressure treatment with preservatives may represent an alternative approach to SWPM mitigation. This process would produce an external barrier to repel pests and residual chemical loadings could be easily verified. However, questions remain about the ability of these treatments to eliminate established pests or to inhibit subsequent emergence.

The performance of barriers against emergence of established pests has received minimal attention. However, surface barriers produced by wood preservatives have long been assessed for their ability to resist termite attack. A majority of this work has focused on subterranean termites, since these species are more destructive to structures. Barriers of chromated copper arsenate (CCA), copper naphthenate, ammoniacal copper zinc arsenate (ACZA), ammmoniacal copper quaternary compound (ACQ), and pentachlorophenol along with different borate chemistries prevent termite attack (Kard 2003; Myles 1994; Yamaguchi 2001; Quarles 1992; Mankowski and Morrell 1993; Preston et al 1996). Imidacloprid also produces a highly effective barrier against *Cryptotermes brevis* (Potter and Hillery 2002).

Previous studies have shown that wood can be sprayed or dipped in different borate chemistries to control the wood-infesting beetles *Hemicoelus gibbicollis* and *Lyctus brunneus* (Williams and Mauldin 1986; Williams and Amburgey 1987; Suomi and Akre 1992). Boric acid applied in this manner effectively controlled these insects at concentrations of 0.20% boric acid equivalent (BAE) or greater. The probable mode of action was starvation due to a loss of gut symbionts. These studies evaluated non-beetle infested wood and were not concerned about the ability of the treatments to penetrate into existing insect galleries.

While boron treatments controlled *Hemicoelus gibbicollis* and *Lyctus brunneus*, these beetles do not pack their galleries with frass but instead reduce the wood to a fine power that is easily penetrated by treating chemicals (Eberling 2002). However, other wood boring insects tightly pack their frass in the galleries as they mine both the heartwood and the sapwood. This behavior is exemplified by the new house borer (*Arhopalus productus*), which is commonly found in fire-killed, insect-killed or wind-thrown western softwoods (Eaton and Lyon 1954). Another species that exhibits this behavior is the red oak borer (*Enaphalodes refulus*), which is known to form a tightly packed frass plug prior to over-wintering in a heartwood gallery (Fierke et al 2005). Similar behavior has been associated with the eucalyptus longhorned borer (*Phoracantha semipunctata*) which is considered a serious introduced pest in a large portion of the world (Bybee et al 2004). These types of wood boring insects are difficult to control using dip or spray on treatments since the tightly packed frass limits bulk fluid movement into the gallery, while the low permeability of the heartwood prevents the treating solution from reaching high chemical concentrations in close proximity to larvae found deep within the lumber. This is a critical concern because low grade packing materials are often produced from either salvaged material or the core of the log where the probability of beetle infestation is much greater.

The penetration of infested materials is further hampered by tree responses to the insect attack. Pits in cells in the sapwood adjacent to larval galleries can aspirate, to prevent desiccation. This pit aspiration also hinders preservative movement, preventing complete penetration of the wood (Haygreen and Bowyer 1996). Softwoods further respond to tree injury by developing traumatic resin canals to wall off the attacked portion of the tree. This highly viscous material may also prevent the ingress of wood preservatives into the insect galleries (Haygreen and Bowyer 1996).

Neumann and Minko (1981) noted that large amounts of resin were produced by radiata pine (*Pinus radiata*) in response to attack by the sirex wood wasp (*Sirex noctilio*). The first indication of attack by the sirex wood wasp is the appearance of resin droplets on the bark of the tree (Ciesla 2003). Sirex is an excellent example of a pest that poses a great threat to North American coniferous forests (Ciesla 2003).

Hardwoods often respond to insect attack by producing tyloses around the area attacked, effectively walling off the attacked portion of the stem. Hardwoods may also extrude gums into the vessels, resulting in complete or partial blockage of the vessel elements essential for preservative penetration (Haygreen and Bowyer 1996).

Developing data on the ability of various systems to penetrate into beetle galleries and the surrounding wood as well as the ability of these treatments to inhibit later beetle emergence will be critical for allowing pressure treatments to be used for mitigation. The objective of this research was to determine if established wood boring insect populations found in lumber cut from fire killed ponderosa pine logs (*Pinus ponderosa*) from central Oregon could complete their life cycle after the material had been treated with one of three different wood preservative chemicals.

Methods

Preliminary trial to determine dimensions

In order to determine how many boards were needed for the main tests, a preliminary trial was performed on fifteen 41mm thick by 1 m long ponderosa pine boards collected from a sawmill located in central Oregon. The lumber was cut from logs salvaged from a large forest fire that had occurred two years earlier. Materials from the same mill and the same timber sale were used throughout this study. The width of the boards varied from 92mm to 304mm, but the material could be placed into one of two categories: less than or greater than 200mm wide. The materials had a large number of insect galleries present on at least one board face. All of the material was dissected by cutting a series of 10mm thick slices across the grain; any insect larvae that were bisected during this process were recorded. The resulting data was used to determine how many 1m long samples would be required to obtain a sample size of 30 boards containing at least two insect larvae per sample.

Sample collection and preparation

The sample size determination study indicated that sixty, 1m long by at least 200mm wide samples would be required for each treatment. The material was collected in 2.5m long sections from the mill, and then cut to 1m long samples that were randomly assigned to one of the four treatments (three treatment types and one control group). The 0.5m long section remaining from each board was dissected to verify larval activity. A total of 300 samples were allocated to four treatment groups. Eighty samples were treated for each of the three treatment groups, while 60 samples were set-aside for the control group. The samples were stickered with six mm thick stickers to increase bulk chemical flow during the treatment process and the end grain was sealed using a marine epoxy to retard penetration and to simulate longer pieces of lumber. The packets were then banded together and sent to the different treating facilities.

The treating process

Three chemical treatments were evaluated. All materials were treated while the wood was wet to simulate what would likely happen in a commercial operation. One set of 80 samples was treated with ammoniacal copper quaternary (ACQ) compound to a target retention of 6.4 kg/m^3, while another 80 samples were treated with disodium octaborate tetrahydrate (DOT) to a target retention of 7.0 kg/m^3. These treatments were applied in commercial treating plants located in Western Oregon, and each was treated in a single charge and in the green condition (moisture content over the fiber saturation point). The third chemical investigated was an experimental treatment containing 30ppm imidacloprid, 300ppm 2-n-octyl-4-isothiazolone-3-one, and 100ppm green dye (Sensient Colors Inc. Milwaukee Wisconsin). The dye was added as an indicator of chemical penetration. However, the experimental treating cylinder could not treat all 80 samples in a single charge. Thus, the samples were treated in five charges, but the treating parameters for each charge were the same (Table 5). All treatments were preformed at ambient temperatures (22-29°C).

Table 5. Treating parameters for each of the chemicals investigated.

Treatment	Treating solution temperature (°C)	Vacuum cycle		Pressure	
		Vacuum achieved (kpa)	Duration (min)	Pressure achieved (kpa)	Duration (min)
ACQ	29	- 81	120	1000	900
Borate	27	- 84.6	10	1103.2	20
Experimental formulation	22	- 86.4	10	1241.1	30

Data collection

After treatment, the marine epoxy was cut from the sample ends and replaced by a wax coating containing 15ppm of imidacloprid to retard beetle emergence. The wax coating was also applied to the ends of the control samples. All existing galleries on the surfaces of the samples were then marked using spray paint so that new beetle activity could be distinguished. During this process, ten samples were randomly selected from each of the three chemical treatments for chemical analysis. The remaining material was numbered and stickered (6mm spacing) to allow air movement and placed inside screened enclosure boxes. The samples were then exposed outdoors in western Oregon. After one month, all of the samples including the controls, were inspected for evidence of new gallery formation on the surface of the material (these new galleries were recorded and marked using a different color of spray paint). An additional ten samples were randomly removed from the three treatment groups for chemical analysis to determine if additional preservative migration had occured. The remaining 60 samples from each treatment group were then inspected at 4 week intervals for evidence of beetle activity during the remainder of the study. In addition, any other observations concerning the appearance of the samples such as mold growth or the development of seasoning checks were noted. The samples were then returned to the enclosure in the same order that they were removed. This process was repeated monthly for one year.

Climatic data was also collected for each 4-week period from a weather station located near Corvallis, Oregon (44° 38” 03’ / 123° 11” 24’; latitude / longitude) and operated by the Oregon State University College of Oceanic and Atmospheric Sciences. The maximum and minimum daily temperatures were collected along with the amount of precipitation. Temperature data was averaged for each 4-week period, while the total amount of precipitation was also noted. This information was used, along with the other observations collected during the inspections to understand larval and insect behavior. Degree days, which are normally used to describe insect development, were not used due to a lack of basic knowledge concerning the developmental rates of *Arhopalus productus* the beetle that was most prevalent in the test material (Furniss and Carolin 1977).

Chemical analysis

Three 10mm thick cross sections were cut at random locations along each of the 20 ACQ or DOT treated boards sampled immediately after treatment. An additional 10 borate treated boards were similarly sampled one month after treatment to measure potential diffusion of this water soluble treatment. The ACQ and imidacloprid treated sections were oven dried at 103°C and 80°C respectively, then the cross sections were photographed for later measurement of preservative distribution in the cross section as well as to estimate the number of internal insect galleries penetrated by the treating chemical. The percent of galleries completely penetrated was then calculated for each treatment. The presence of boron was determined by oven drying the 60 cross sections, sanding the cut surface to remove any boron carried by the saw blade, and spraying with boron indicator in accordance with AWPA Standard A3-00 method 1. A color change from yellow to red indicated the presence of boron (AWPA 2004b).

Two 15 mm thick assay zones were removed from each cross section, one from the narrow and the other from the wide face of the original board. The cuts were made to remove 15mm in each direction from the corners prior to grinding since this material is normally treated more thoroughly than the remainder of the material and, might bias the results. The assay zones were then oven dried (103°C for ACQ and DOT and at 80°C for imidacloprid samples) and ground to pass a 30 mesh screen. The ACQ treated material was analyzed for copper oxide content using a Spectro Titan X-ray Fluorescence Analyzer, according to AWPA Standard A11-93 (AWPA 2004c). The borate samples were analyzed using AWPA standard A2-98 Method 16 (AWPA 2004a). Briefly, weighed (1-2 gram) samples of ground wood were extracted by adding 75-100 ml of water to the sample and heating at 100°C for thirty minutes. The filtrate was then removed and analyzed for boron content using a spectrophotometer after adding Azomethine-H and a buffer solution. Boron levels were calculated by comparison with similar solutions with known amounts of boron.

The samples treated with the experimental treating solution were analyzed for imidacloprid content by weighing 0.62-0.74g samples of wood flour and extracting this material in 25ml of methanol in screw cap bottles while sonicating for 3 hours. An aliquot (10µl) of the extract was then removed from the screw cap bottle through a 0.2 µm syringe-filter. The filtered extract was then analyzed by high performance liquid chromatography (HPLC, Table 6). Standard solutions containing 1-10ppm of imidacloprid were analyzed and peak area was used to develop a standard curve. This calibration curve was then used to determine the amount of imidacloprid in the unknown samples (Jin and Walcheski 2006). Isothiazolone was not analyzed in this experiment because the test was primarily insect driven, while isothiazolone was primarily added to provide fungal protection.

Table 6. HPLC conditions used to analyze imidacloprid in wood extracts

Column	Inertsil ODS-3 C-18 150x 4.6 mm, 3µm
Mobile phase A	30% acetonitrile/ 70% water applied to column from minute 0 to 8 and 11 to 25
Mobile phase B	90% acetonitrile/ 10% water applied to column from minute 8 to 11
Detector	Ultra-violet light, 270nm
Flow rate	1mL/min
Injection volume	10µL
Data collection time	8 min
Imidacloprid retention time	5min

Data analysis

The monthly emergence data from the sixty samples per treatment were analyzed using an ANOVA analysis, where the untreated control group was used as a reference level. Significant differences (α=0.05) between treatments are denoted by letters above the bar, with the bar representing the average number of new insect holes for any given time period and treatment.

Chemical analyses for ACQ and borate were examined to determine if they met the current AWPA standards for ground contact applications. Chemical analysis from the narrow and wide faces were compared to determine if significant (α =0.05) differences existed between these two assay zones using ANOVA. In addition, the percentage of completely penetrated galleries bisected in each section was calculated and compared using an analysis of variance.

Results

Sample size determination

A total of 51 insect larvae were found in the fifteen 1m long boards initially examined to estimate the replication needed for the main study. A disproportionate number of insect larvae were found in the material wider than 200mm (Table 7). This material had an average of 5 larvae per board, with 60% of the samples containing 2 or more insect larvae. This was most likely caused by a lower surface area to volume ratio, an important feature since many larvae found in the narrower material had been killed during the milling process. These results indicated that a sample size of 60 boards per treatment would be needed for the remainder of the study. This sample size allowed for a 10% buffer in obtaining 30 samples containing at least 2 larvae per board. In addition, this sample size was consistent with the 60 samples used by Williams and Mauldin (1986) to investigate the ability of borate to mitigate lyctid beetle infestations in *Virola* lumber from Brazil.

Table 7. Number of larvae found within each of fifteen 1m long by 41mm thick pine boards of varying widths.

Width (mm)	Number of larvae found
92	1
146	0
146	0
190	0
197	0
222	14
240	0
240	8
241	0
247	1
248	11
249	0
250	3
305	5
305	8
250	3

Chemical penetration and retention

All three treatments completely penetrated the ponderosa pine sapwood; however, only a thin treated shell, approximately 3mm thick, was produced around the heartwood portion of the samples. In addition, all galleries were completely treated, regardless of whether they were found in the heartwood or sapwood. The thickness of the treated shell in the borate samples did not visibly increase one month after treatment, nor was there evidence of significant borate diffusion away from the treated insect galleries. Examples of typical penetration patterns for all three treatments can be found in Figures 10-12.

Figure 10. Example of copper penetration (green areas) in cross sections cut from unseasoned ponderosa pine containing active beetle infestations and pressure treated with ACQ.

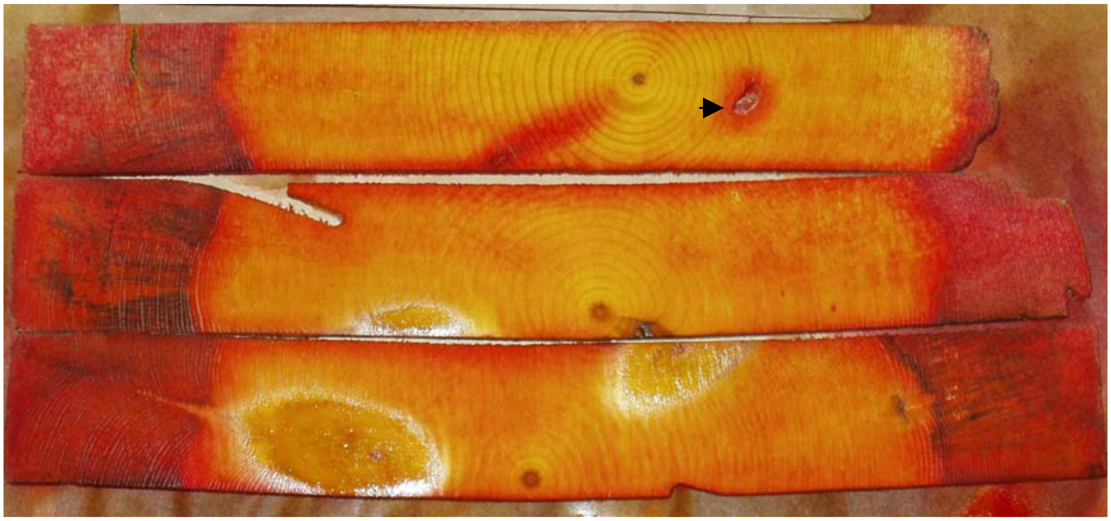

Figure 11. Example of boron penetration (red areas) in cross sections cut from unseasoned ponderosa pine containing active beetle infestations and treated with disodium octaborate tetrahydrate. Note the treated insect gallery, surrounded by untreated heartwood, in the first cross section (arrow).

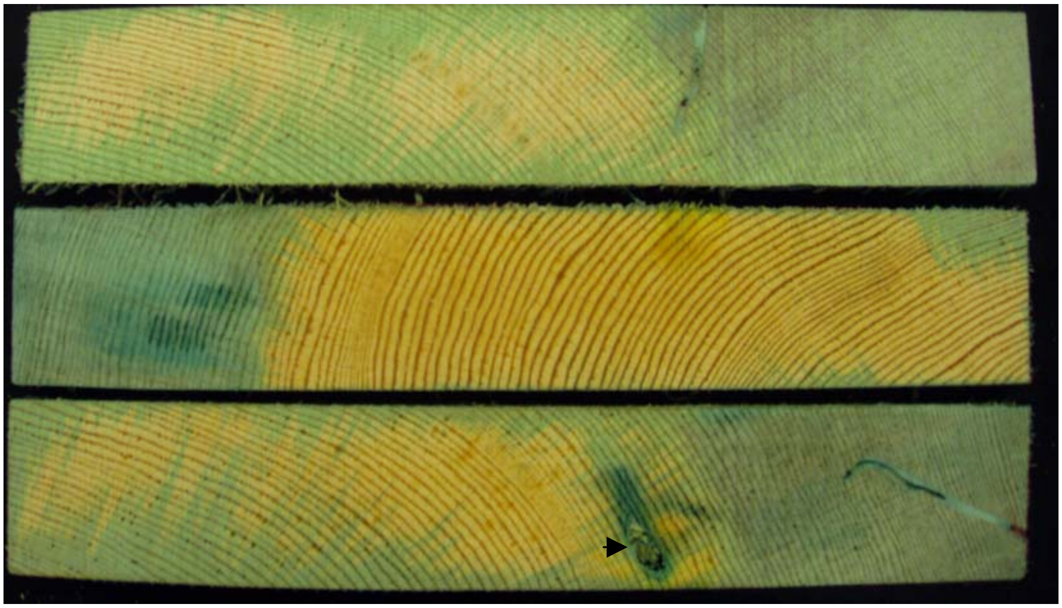

Figure 12. Example of green dye penetration representative of imidacloprid distribution (green areas) in cross sections cut from unseasoned beetle infested ponderosa pine lumber pressure treated with an imidacloprid/ dye mixture. Note the frass filled gallery in the third cross section surrounded by untreated heartwood (arrow).

Copper oxide retentions in the assay zone from the ACQ treated boards averaged 8.07 kg/m^3 (7.08 and 9.05 lower and upper 95% confidence limits), easily exceeding the targeted 6.4 kg/m^3 retention specified for ground contact exposure (AWPA 2004d). ANOVA analysis using PROC MIXED showed that retention did not differ significantly between the side and the top faces of the boards (P-value of 0.405 F-statistic of 0.70, with 1 degree of freedom in the numerator and 50 in the denominator). The ANOVA analysis of the borate retentions indicated that a one month diffusion period did not significantly affect retention (P-value of 0.38 with an F statistic of 0.77, 1 degree of freedom in the numerator and 106 degrees of freedom in the denominator); however, a significant difference was noted in retentions between the narrow and wide faces at α level of 0.05 (P-value of 0.0292, F statistic of 4.89, 1 degree of freedom in the numerator and 106 in the denominator, Table 8). Chemical retentions differed by over 10%. The side retention was 9.13 kg/m^3, exceeding the target retention of 7 kg/m^3 (lower 95% confidence limit of 6.37 and an upper confidence limit of 11.89 kg/m^3), while the wide face retentions averaged 5.70 kg/m^3 (95% confidence interval of 2.96 and 8.43 kg/m^3) and did not meet the target retention. These differences in chemical retention can be explained by differences in wood anatomy. The sides of the boards contained larger amounts of more easily treated sapwood, while the wide face of the boards contained more difficult to treat heartwood. The aspirated pits in this heartwood most likely accounted for the differences in chemical retention.

Imidacloprid retentions did not differ significantly between the side and wide faces of the boards (P value of 0.601, F-statistic of 0.28 with 1 degree of freedom in the numerator and 52 degrees of freedom in the denominator). The mean imidacloprid concentration for all materials tested was 0.0150 kg/m^3 with a 95% confidence interval from 0.124-0.0176 kg/m^3. The relatively small variation in retention may be a result of treating the materials in a laboratory environment under more controlled conditions.

Table 8. Differences in borate retentions in beetle infested ponderosa pine lumber sampled on the side or wide faces along with mean retentions for ACQ and imidacloprid in all samples.

Treatment	Sample Orientation	Number of samples analyzed	Mean Retention (SE) kg/m^3	95% confidence interval
Borate	Wide side	60	5.70 (0.1.07)	2.986-8.44
Borate	Narrow side	60	9.13 (1.09)	6.36 – 11.88
ACQ	All samples	60	8.06 (0.45)	7.17-8.96
Imidacloprid	All Samples	60	0.0150 (0.0013)	0.124-0.0176

Insect emergence data

Insect emergence data was considered by season for the purposes of this study. Fall months were defined as the 4 week periods ending on September 15th, October 15th and November 15th, while Winter included the 4 week periods ending on December 15th, January 15th and February 15th. Spring was defined as the three 4 week periods ending on March 15th, April 15th and May 15th. Observations made on June 15th, July 15th and August 15th were considered the summer season. All of the data pertaining to new insect galleries detected are summarized by month in Figure 11.

There was a considerable amount of beetle activity in control samples in the Fall, but this activity was entirely larval in nature and no adult emergence holes were noted on the treated samples. The number of new galleries found on the surface declined as the temperatures cooled. One active subterranean termite (Isoptera Rhinotermitidae *Reticuloternes hesperus*) colony was present in the untreated samples and an adult new house borer was also collected from this material, but no other adult beetles were captured. Ten new galleries were found on the surface of the borate treated material in Fall; however, none of these new galleries were adult exit holes. Adult exit holes are distinctly oval in nature, and lacked frass packing, as described by Eaton and Lyon (1954) and observed in the laboratory for a new house borer pupae placed in a ponderosa pine block. Similarly, 4 new larval galleries were found on the surface of the ACQ treated material. A distinct ammonia odor was also noted in association with the ACQ treated material, but it is unclear what effect, if any, that this odor had on beetle activity. By November 15th, average daytime temperatures had dropped below 10°C and insect activity had ceased. This coincides with findings by Keena (2002) investigating the effects of temperature on *Anoplophora glabripennis* (Coleoptera: Cerambycidae). The material treated with imidacloprid was not in test during this time period due to delays in the treating process.

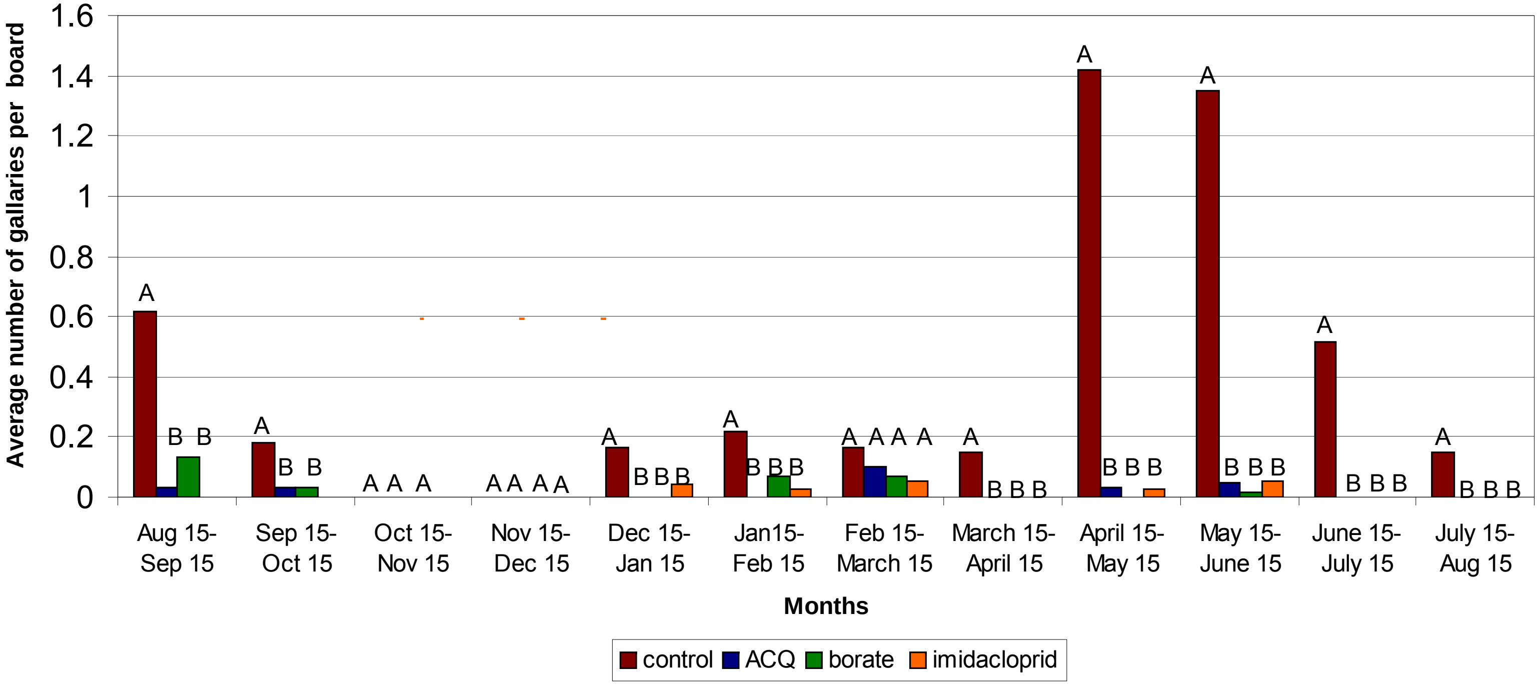

Figure 13. Average number of new larvae or adult holes detected per month on the surfaces of beetle infested pine samples treated with ACQ, borates, or imidacloprid. Imidacloprid treated materials were not included during the first three time periods. Bars with the same letters do not differ statistically significantly from one another (α =0.05).

Insect activity in the winter was only observed when average high temperatures exceeded 10°C. All insect activity observed during this time period was a result of larvae coming to the surface and then re-entering the material. No adult insects were collected and no new adult emergence holes were identified. The large amount of rain received during this period was conducive to fungal growth and chemical leaching. Basidiomycetous fruiting bodies were found on one of the control samples, while substantial mold growth was noted in the top five layers of the borate treated samples. These layers received a majority of the moisture from the rainfall. The dye incorporated with the borate and imidacloprid treatments also leached considerably, causing these materials to lose most of its blue-green color. Once temperatures exceeded 10°C in mid-January and February, larval activity on the surface of the untreated material far exceeded that seen in the treated material. Destructive examination of the 500mm long untreated scraps retained from the sample preparation showed that two or more larvae were present in 50% of the samples. The larvae moved slowly when exposed, but became more active at temperatures in excess of 25°C.

The remainder of the 500mm long untreated scraps were destructively sampled during the spring. The wood had been stored under a roof but experienced outside temperatures and relative humilities during the winter months to simulate the low moisture content conditions experienced by larvae found in the center of the treated and untreated stacks of lumber. Over 50%

of the samples contained 2 or more active larvae despite the cold temperatures and low wood moisture contents. The rate of larval activity generally increased as temperatures warmed later in the season. In addition, one new house borer pupae was removed from this material in early May.

The spring months were associated with increased insect activity in the samples; however, activity was largely confined to the untreated control samples. All larval activity observed on March 15th and April 15th was associated with larvae coming to the surface and reentering the material. Average daytime highs during this time period were 12 and 17°C, respectively. This activity was similar to that observed between January and February 15th. Larval activity significantly increased in the untreated control samples during the period ending on May 15th. Eighty five new observations of beetle activity were made over this time period and 21 of these were adult exit holes. However, only two new galleries were detected on the surfaces of the ACQ and imidacloprid treated material and neither of these holes was caused by an exiting adult. No new larval galleries or adult exit holes were found on the surfaces of the borate treated material. Increased insect activity likely reflected the significantly higher average daytime high temperatures seen during this time period. Mold growth that was present throughout the winter on the borate treated materials had receded as the wood dried. The heaviest amounts of mold growth were centered on old frass-packed larval galleries, suggesting that fungal spores were transported by the larvae or that the fungi were using partially modified wood components found in the frass. The resulting fungal colonies may aid the larvae in the conversion of cellulose into simple sugars. However, the increased presence of free sugars in the partially digested frass also favored mold growth.

Insect activity peaked during the summer. In addition to a high level of larval activity in the control boards, a large number of adult exit holes were also detected in this material (Figure 14). During the period ending on June 15th, 81 new insect galleries were detected on the control samples and 20 of these were adult exit holes, characterized by their round nature and lack of frass packing. However, no actual adults were collected during this inspection. No adult exit holes were found in the treated samples and the number of new insect galleries found on the surface of this material had also greatly decreased. The lack of adult insect emergence from the treated material could be attributed to the insecticide nature of the wood preservatives investigated. These findings were associated with an average high temperature of 24°C and an average low temperature of 11°C. The inspections conducted during the weeks of July 15th and August 15th found no new insect activity associated with the treated materials, while the number of new adult emergence holes in the control samples peaked during the month of July, when two adult insects were found. These insects were positively identified as new house borers. In addition, the appearance of seasoning checks on the surface of the samples was also noted during this inspection.

Checking was limited to the upper 5 layers of the stacks and was most severe in the control samples; however, some seasoning checks were also present in the treated material. During the August 15th inspection, the presence of seasoning checks had spread through out the samples, including some internal checking. New adult emergence holes were also found in the control samples during this inspection; however, the number had greatly decreased, as had the over all occurrences of new galleries on the surface of the material. This decline may be due to a lack of moisture in the material.

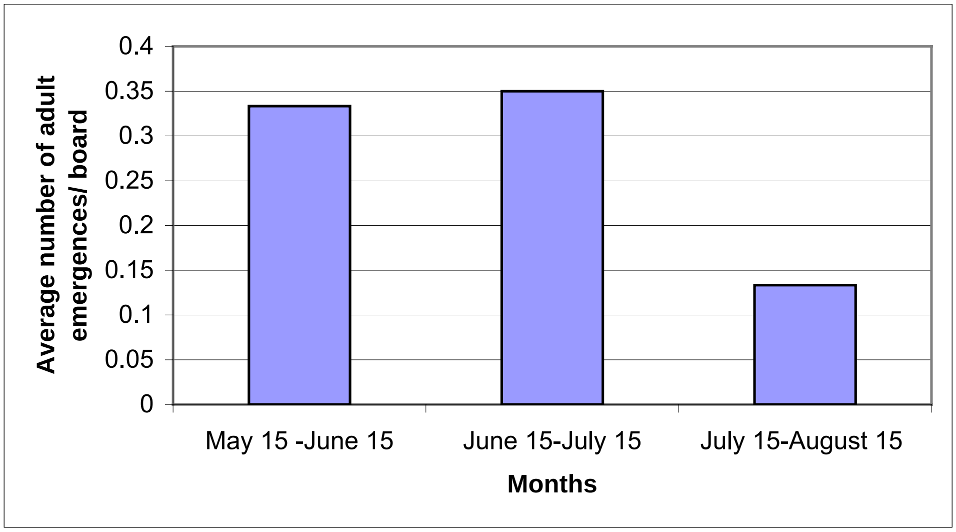

Figure14. Average number of new adult insect emergence holes found in 60 untreated ponderosa pine control samples.

Conclusions

Ponderosa pine lumber with active wood boring insect populations can be treated with conventional wood preservative to retention levels that will prevent the emergence of adults. None of the wood preservatives investigated prevented immature insects from continuing to survive within the wood. The periodic surfacing of larvae is a significant concern since this can result in the infestation of non-treated wood products stored on or in close proximity to the treated solid wood packaging material. These larvae might then successfully complete their life cycles and emerge from the wood as adults, resulting in an invasive species introduction. One problem with interpreting these results is the lack of data regarding larval behavior in different wood species. Thus, further behavioral studies on wood boring insects in other wood species are recommended. Additional work focused on developing effective, yet environmentally friendly larvacides for inclusion with current wood preservatives is also suggested.

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Ability of Preservative Treated Barriers to Prevent the Emergence of Established Wood Boring Insect Larvae From Solid Wood Packaging Materials

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For Submission to the Proceeding of the 27th Annual Meeting of the Canadian Wood Preservers Association
November 7th and 8th 2006
Vancouver British Columbia Canada

ABILITY OF PRESERVATIVE TREATED BARRIERS TO PREVENT THE EMERGENCE OF ESTABLISHED WOOD BORING INSECT LARVAE FROM SOLID WOOD PACKAGING MATERIALS

Abstract

Wood preservatives can penetrate a majority of frass packed insect galleries even in refractory species, but complete penetration cannot be guaranteed. Thus, it is important that an effective barrier can be established around the timbers used to transport goods in international trade in order to prevent invasive species introductions. In this study, ammoniacal copper quaternary compound, disodium octaborate tetrahydrate and an experimental imidacloprid formulation were investigated for this application. These chemicals, did not inhibit larval penetration since they lack larvacidal properties. However the approach developed during this study may be useful for other work related to evaluating chemical formulations for invasive species prevention.

Introduction

The mitigation of pests in solid wood packaging materials (SWPM) has become a major international concern as countries seek to prevent the unintentional importation of invasive species established within these materials. While fumigation and heat treatment are recommended for mitigation, each has drawbacks and there is a continued search for better methods for dealing with these materials. One alternative approach is pressure treatment with preservatives. This process offers a simple, verifiable mitigation approach. Despite its long use for wood protection, there is little data on the ability of pressure treatment to eliminate established beetle populations.

Preliminary studies have shown that treatment with waterborne wood preservatives using commercially available processes completely penetrated frass-packed insect galleries in refractory western redcedar (*Thuja plicata*) with 88% to 100% efficiency. Incomplete treatment could allow some larvae to survive the initial treating process. These larvae are of phytosanitary concern because they could pupate and emerge from the material as adults, resulting in invasive species introductions. Ideally, the preservative envelope surrounding the untreated core of a board will be deep enough and sufficiently active to either inhibit or kill larvae or pupating adults. However, there is little data on the ability of these treatments to limit egress of insects that survive treatment.

Wood preservatives have long been used to prevent the establishment of fungi and insect populations in uninfested lumber. This is preferably achieved by completely penetrating the wood with a chemical that acts as a fungicide and insecticide. However, this is not possible in all species, including many of the economically important species found along the West Coast of North America. An alternative method of preventing fungal and insect attack is establishing a treated envelope around the untreated material, but incising is often required to establish an envelope thick enough to prevent fungal and insect attack in this material (Morrell and Winandy 1987). Choi et al. (2003) found that fungal spores were able to penetrate this treated envelope through surface checks in above ground applications; however, the process was extremely slow. They found that mobile portions of the wood preservatives produced a very thin shell of protection over the newly exposed wood within the check, hampering fungal attack. The treated shell can also deter subterranean termite attack in structural lumber, and a variety of preservatives have been investigated for this purpose. Amine copper quaternary compound (ACQ) and chromated copper arsenate (CCA) prevented termite attack at low retentions (4.0 and 2.1 kg/m^3 respectively), while disodium octaborate tetrahydrate (DOT) required higher loadings to prevent termite attack (Preston et al 1996). Similarly, imidacloprid has been shown to prevent termite attack (Potter and Hillery 2002) as well as destruction of ash trees (*Fraxinus spp.*) by the emerald ash borer (*Agrilus planipennis* Fairmaire). Imidacloprid in the ash leaves also led to adult mortality (Wang et al 2001). Imidacloprid has also been studied for disinfecting ash logs to allow for their transport to processing centers outside of the quarantine areas established in the Midwest (Nzokou et al 2006). Dipping green logs in imidacloprid for four hours prevented adult insects from emerging through the bark of the logs. However, the study also found substantial evidence that larvae survived this treatment despite dwelling in shallow galleries that extended no more then 100mm into the sapwood (Nzokou et al 2006).

While studies suggest that external barriers will limit entry and egress by various wood inhabiting insects, preliminary tests on materials naturally infested with wood boring insects, notably the new house borer *Arhopalus productus*, showed that some beetles could survive initial pressure treatment. This was determined through a field trial where larvae were later found to be able to move to the surface of treated wood. However, the effects of treatment on the development of the immature *A. productus* (Cerambycidae Coleoptera) was unclear.

Little is known about the behavior of the new house borer, because a majority of this insect’s life cycle is completed deep within the wood, often well into the heartwood of the tree. These deep galleries are often tightly packed with frass (Eaton and Lyon 1954), increasing the difficulty of reaching these insects with wood preservatives or fumigants. In addition, the larvae reach a large size prior to pupation and have energy reserves in the form of fat bodies that may allow them to avoid digesting preservative treated materials, thus providing a worst-case scenario for phytosanitary treatments. These attributes, along with the widespread occurrence of the new house borer in fire killed western conifers, makes it an ideal model species to assess the effects of preservative treatments on wood boring insects. Thus, this species was used to assess the ability of larvae and adults to bore through a simulated treated wood envelope (STE), and to determine if the larvae completed development after being exposed to treated materials.

Methods

One hundred fifty Ponderosa pine (*Pinus ponderosa*) boards (41mm thick by at least 200mm wide by 2.5 m long) were collected from a sawmill in Central Oregon. These samples were cut from logs harvested in a salvage logging operation following a forest fire two years earlier; and contained extensive beetle galleries. One 0.5m long sample was cut from the center of each board and used for this study, while the remainder of the material was used to study field emergence (Chapter 4). The samples were initially kept wet, but were then allowed to equilibrate to ambient moisture conditions and stored under cover until needed for this study.

Larvae extraction and insect rearing

Insects were extracted from the boards over a 6 month period from late fall to early spring by splitting the wood into small fragments using an axe and a splitting mall. Some of the 0.5m long samples were also cut into shorter segments when knots were present to ease the extraction process. The insect larvae became visible on the surface as the wood was split into smaller units, at which point they were collected by hand. In some cases, however, the insect larvae fell out of the exposed galleries after splitting.

The extracted larvae were placed on a synthetic diet described by McMorran (1965). This diet was modified by eliminating the 10ml of water per

100ml of diet called for in the original recipe and by doubling the amount of nutrient agar. In addition, the diet was modified by adding 25 cm^3 of 30 mesh ponderosa pine wood flour per 100ml of media (Gardiner 1970). A 10mm thick layer of the media was then poured into 100mm diameter petri dishes. Once the media had hardened, a depression approximately as long and wide as the larvae was cut into the surface of the media. One larva was then placed in each petri dish and a layer of solidified media was placed on top of it. The larvae were then incubated in a dark chamber at 32°C and 65% relative humidity. The larvae were observed on a weekly basis and were moved to a new petri dish if the original dish was contaminated by fungi or the media lost an excessive amount of moisture.

Exposure of larvae and pupae to different wood preservatives

Two mm thick ponderosa pine sapwood veneers were cut into 10mm wide by 100mm long strips. Groups of 100 strips were vacuum treated with ACQ or disodium octaborate tetrahydrate (DOT) to target concentrations of 1.2 kg/m^3 or 0.4 kg/m^3, respectively. A third set of 100 veneer strips were pressure treated using an experimental treating system containing 30ppm imidacloprid, 300ppm 2-n-octyl-4-isothiazolone-3-one and 100ppm green dye (Sensient Colors Inc. Milwaukee Wisconsin) in a laboratory scale treating cylinder. An additional 100 veneer strips were used as untreated controls. Five veneer strips from each treatment were oven dried and then ground into 30 mesh wood flour for later analysis.

Ground wood from the ACQ treated material was analyzed for copper oxide content by x-ray fluorescence spectroscopy using a Spectro Titan Analyzer (Spectro Titan Instruments Austin Texas) according to AWPA standard A11-93. The results were used to estimate the average retention for all samples treated with ACQ (AWPA 2004b).

The DOT treated samples were analyzed using AWPA standard A2-98 Method 16 (AWPA 2004a). Briefly, 1-2gof ground wood was extracted by adding 75-100 ml of water to the sample and heating at 100°C for thirty minutes. The filtrate was then collected and analyzed for borate content using a spectrophotometer after adding Azomethine-H and a buffer solution. The resulting borate concentrations were then used to estimate the amount of chemical deposited in each of the 100 treated strips.

Samples treated with the imidacloprid based treating solution were analyzed by placing 0.62-0.74g of wood flour in 25ml of methanol in a screw cap bottle and sonicating for 3 hours in an ultrasonic bath. The samples were allowed to settle over night, then an aliquot (10μl) of the filtrate was withdrawn into a disposable syringe through a 22μ syringe filter and analyzed using high performance liquid chromatography (HPLC). Standards ranging from 1-70ppm imidacloprid dissolved in methanol were prepared and analyzed by HPLC (for HPLC conditions see Table 9). A 2ppm standard was also run after every fifth sample to ensure proper calibration. The area under each peak was calculated and a linear equation of peak area verses concentration was computed using the data. This calibration curve was then used to determine the amount of imidacloprid in the unknown samples (Jin and Walcheski 2006). Chemical concentrations in these samples were used to estimate imidacloprid retentions for the remainder of the veneer strips.

Table 9. HPLC conditions used to analyze imidacloprid in methanol extracts.

Parameter	Condition
Column	Inertsil ODS-3 C-18 150x 4.6 mm, 3μm
Mobile phase A	27.5% acetonitrile, 22.5% ammonium phosphate buffer (1.30g ammonium phosphate dibasic dissolved in 1l of HPLC water) 49% HPLC water and 1% methanol. Applied to column during minutes 0 to 8 and 11 to 25
Mobile phase B	90% acetonitrile/ 10% water applied to column during minutes 8 to 11
Detector	Ultra-violet light, 270nm
Flow rate	1mL/min
Injection volume	10μL
Data collection time	8 min
Imidacloprid retention time	5min

The veneer strips were used to create simulated treatment envelopes surrounding untreated material, one veneer strip was oriented so that it spanned the center of an empty petri plate. A second veneer strip treated with the same chemical was placed in the plate 5-15mm away from first veneer strip, depending on the size of the larvae used. Once the two veneer strips were placed in the petri dish, the space between the veneers was filled with artificial diet to simulate untreated wood. Artificial diet was also placed to a depth of 10mm on the outside of the enclosure. A space was cut into the diet between the veneers to accommodate the larvae. The larvae was then introduced into the space between the two veneers so that it was parallel with the veneers and the plate was incubated in the dark at 32°C. Larval position was observed every 48 hours for 20 days. The time required for larvae to penetrate through the veneer and the time required until larval death occurred were of particular interest along with any other behavioral changes. Initial trials assessed single veneers, but subsequent trials used several veneers to simulate thicker treatment barriers. Each treatment and barrier thickness was assessed on a minimum of 3 larvae. In addition, 4 larvae were exposed to artificial diet, modified with 0.5ml of chlorpyrifos, a known contact larvicide, per 100ml of media to determine if the diet composition affected efficacy. These larvae were monitored on a 24 hour basis until death occurred.

A limited number of pupae were also exposed to the treated veneers. The pupae were placed inside holes drilled tangentially into ponderosa pine blocks. The holes were slightly larger than the pupae diameter but still small enough that an emerging adult would be forced to excavate wood to move. The hole was drilled 43mm deep in green ponderosa pine sapwood blocks (48 by 42 by 75mm long) high cut from the same material used to collect the larvae. The remaining 5 sides of the block were then covered with treated 2mm thick veneer strips. Care was taken during this process to ensure that the insect pupae were not exposed to adhesive and that no gaps existed between the untreated block and the veneer. The hole was plugged using a cork stopper with a small groove cut into it to allow for gas exchange after the pupae had been placed into the chamber. The sides of the block that had been enclosed with the veneer were then covered with a moisture barrier to limit desiccation and the samples were incubated at 32°C. The blocks were then observed for emerging adults over a 6 week period.

Results and Discussion

One hundred sixty three larvae were recovered in the extraction process, however, only 137 were extracted successfully. The remaining 26 larvae were damaged during the extraction process, primarily due to puncture wounds inflicted by wood splinters associated with ingrown knots. The larvae found during this process varied greatly in size and developmental stage. However, more lower in-star larvae were found early in this study. A majority of the larvae removed later in the test had progressed into the later in-star stages, using larvae size as a guide. The activity level of the larvae also varied from fall to early spring. Larvae removed during the warmer periods in late fall and early to mid-spring were extremely active upon removal from the wood substrate, while little larval activity was observed immediately after removal during the colder winter months. However, larval activity increased greatly once they were exposed to temperatures in excess of 25°C for two to three hours, and resembled that of the larvae removed during warmer periods.

Larvae propagation using a modified artificial diet

Early in this study, some larvae, especially lower in-star larvae, died, due to excess moisture present on the surface of the media. To reduce moisture, the 10 ml of distilled water per 100 ml of media called for in the original formulation was eliminated and the nutrient agar content was doubled along with the amount of dry powdered xylem tissue. This resulted in a very thick medium containing very little free water. These changes allowed almost any size larvae to be successfully propagated, with a majority of the larvae transitioning from one in-star stage to the next while held under laboratory conditions. However, very small larvae (less then 5mm long) that were assumed to be in the first or second in-star stages could not be propagated successfully using this method and, thus, were not utilized during the remainder of the study. This led to the loss of an additional 31 specimens. Once the diet had been modified, ten larvae of various sizes were incubated in the dark at 32°C for four weeks to determine the short term effects of the diet on the larvae. These larvae exhibited no negative responses after this time, and this formulation was used for the remainder of the study.

Exposing larvae to simulated treated envelopes

All larvae were extracted and placed on the artificial diet for two weeks prior to exposure to the veneers to determine if they responded favorably to the diet. All of the larvae responded favorably, with larvae consuming an average of 15.7 cm^3 of the diet after two weeks (Figure 15). Five larvae each were then exposed to 2mm thick wafers containing ACQ, DOT, imidacloprid or untreated veneers. Chemical retentions for ACQ and DOT were slightly above the targeted level (Table 10), while the imidacloprid retentions were nearly twice the targeted amount. This over-treatment resulted from re-treatment after the material initially failed to meet targeted retention levels.

Healthy larvae required approximately eight days (four 48 hour periods) to penetrate the 2mm thick ponderosa pine veneers when the envelope was untreated or treated with ACQ or DOT (Figure 16) and none of the larvae died after contacting either the treated or the untreated veneers. The behavior displayed by the larvae in contact with an imidacloprid treated veneer differed substantially from that found with the other treatments. One larva required less then 48 hours to emerge from between the treated veneers. The behavior of this larva and others that had penetrated the imidacloprid barrier later became extremely aggressive. Two of these larvae excavated their way through the plastic petri dish and into the surrounding container, while one larva exited through the plastic petri dish and then tunneled into some soft plastic based material used to support the petri plates. In contrast, another larva perished after 12 days without exiting through the veneer, while the remaining larva survived until the end of the study, but did not exit through the STE. Neither of these larvae differed significantly in size or appearance from the other larvae prior to the commencement of the study.

Table 10. Average chemical retentions for the 2mm thick veneers used to construct simulated treated envelopes.

Treatment	Targeted Retentions (kg/m^3)	Actual Retention[a1]
ACQ	1.2	1.31 (0.25)
DOT	0.4	0.426 (0.134)
Imidacloprid	0.0115	0.0222 (0.0025)

[a1] Achieved values represent means of 5 replicates while values in parenthesis represent one standard deviation (kg/m^3)

Figure 15. A new house borer larvae consuming the artificial diet two weeks after removal from a ponderosa pine block.

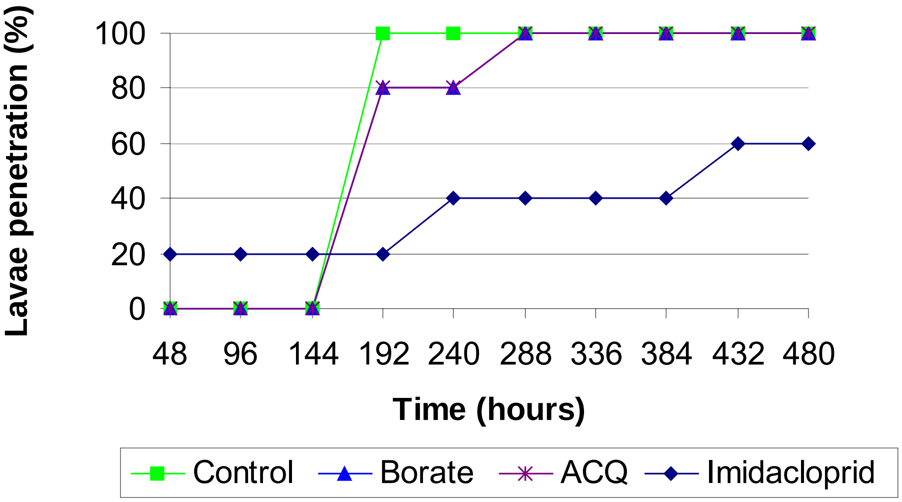

Figure 16. Percent of larvae boring through a 2mm thick untreated or preservative treated veneer at 48 hour intervals after introduction. Observations were made on five larvae per treatment .

Three larvae exposed to untreated veneers and three exposed to DOT treated veneers required less then 48 hours to tunnel through 4mm of veneer, while two larvae required less then 48 hours to penetrate a 4mm thick ACQ treated shell (Figure 17). These data suggest that penetration time was not linearly correlated to barrier thickness. One larva exposed to the ACQ treatment perished 8 days after insertion without penetrating the veneer; however, cause of death could not be established due to excessive mold growth.

Larvae required an additional 144 hours to penetrate the imidacloprid treated veneers. This was significantly longer than the time required to bore through the other treatments, including the 2mm veneers. The highly aggressive behavior describe earlier in association with larvae exposed to imidacloprid was repeated. As in the previous study, one larva survived the entire 20 day trial without exiting through the imidacloprid treated envelope. Once again, no cause could be established for this behavior.

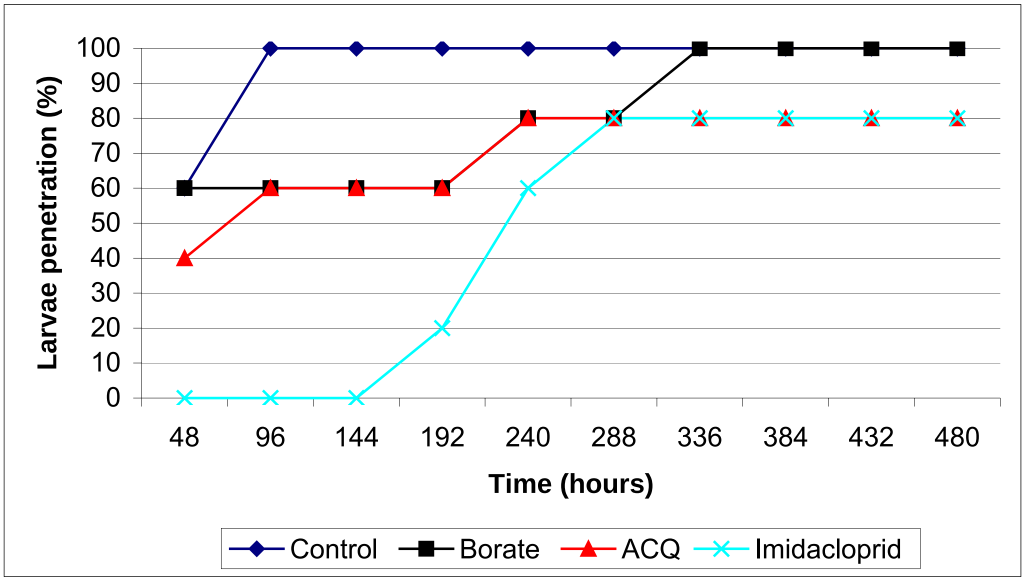

Figure 17. Amount of time required for insect larvae to penetrate a 4mm thick simulated treated envelope.

The barrier trial was repeated once more using a 6mm thick barrier treated with either imidacloprid or ACQ (Figure 18). Limited larval availability allowed for the use of only 4 and 3 replicates, respectively. The thicker barrier did not significantly increase the time required for the larvae to penetrate through the wood. The average time required for penetration was 4 days for the ACQ treated material and 8 days for larvae exposed to the imidacloprid treated veneers. The aggressive behavior expressed by the larvae exposed to the imidacloprid based treatment was also noted. Imidacloprid exposed larva, like the others, survived until the end of the 20-day period. These larva were then placed on a fresh petri plate that did not contain any insecticides or larvacides, where they later pupated. These results indicated that exposure to the treated barrier had little or no permanent effect on the larvae.

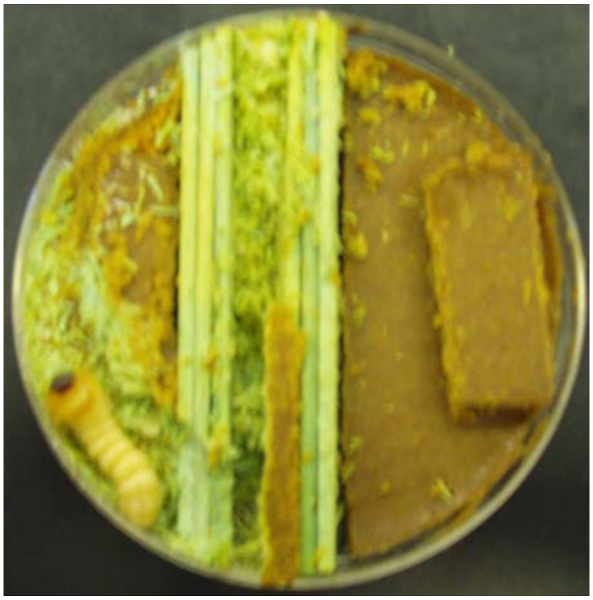

Figure 18. Petri dish containing larva that has penetrated a 6mm thick imidacloprid treated barrier 8 days after being introduced into this environment.

Incorporation of a larvacide and imidacloprid into the diet

While the larvae were clearly capable of penetrating the treated barriers, there remained a question about whether the media had affected the chemical sensitivity of the larvae. For example elevated media pH might alter either biocidal activity or the ability of the organism to sorb the chemical. Larvae grown in media amended with 10,000 ppm of imidacloprid were unaffected, continued to feed heavily and bored through the plates consuming, on average, 23.5 cm^3 of the diet. These larval also expressed the same aggressive behavior as described earlier, with one of the larva exiting the petri dish by boring a hole through the lid. Media was also amended with 0.5ml of chlorpyrifos, a contact insecticide, per 100 ml of diet to determine if proven larvacides would be effective in this environment. All of the larvae exposed under these conditions died within 48 hours. However, chlorpyrifos cannot be used as a chemical to treat SWPM due to its broadly toxic nature. The inability of the imidacloprid to kill the larvae was thought to have been associated with the pH of the media. Thus the pH of the diet was determined by testing the diet before the agar solidified. This investigation found that the diet had a pH of 5.70, which is well within the tolerances for imidacloprid. Thus, media pH did not reduce effectiveness.

Microscopic examination of frass from the simulate treated envelope study

The lack of apparent effects of the treated barrier on larvae activity could have been due to avoidance, whereby the larvae excavated but did not ingest the wood. Frass (0.2g) produced by one larva from the 4 mm thick barrier treatment was examined under a dissecting microscope. The shape of the frass and the co-mingling of wood fibers with components of the artificial diet was of particular interest since co-mingled materials and wood fiber with rounded edges would suggest that the wood had been digested by the insect, while frass with sharp edges and loose wood particles would suggest that the larva had merely removed the wood so they could pass through the area, perhaps in search of more palatable material. Likewise, the surface texture was of interest since it would also yield evidence suggesting digestion of the wood rather then the mere removal. Frass removed from petri dishes containing imidacloprid, ACQ, or DOT treated veneers contained wood particles with intact cell structure, sharp edges, and no evidence that wood particles were intermixed with the artificial diet. These results suggest that this material was not digested by the larva. The material appeared to have been mined and moved aside so that the insect could reach more desirable materials such as the artificial diet (Figure 19). In contrast, wood in frass produced by larvae exposed to untreated ponderosa pine had rounded edges suggesting digestion rather then the removal of the material and was co-mingled with remnants of the diet (Figure 20).

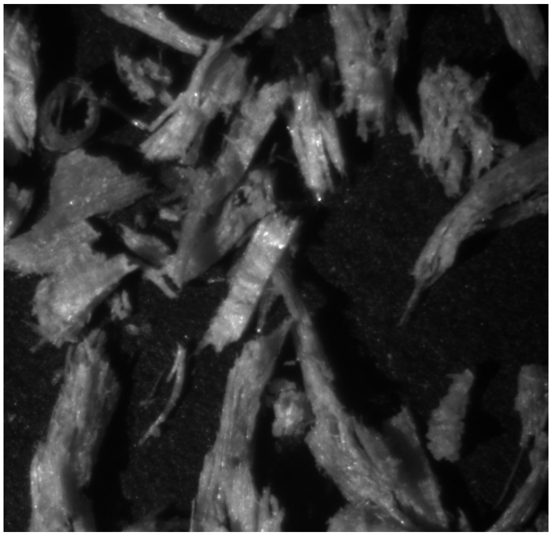

Figure 19. Micrograph of frass (10x) produced by a larva in contact with a 4mm thick imidacloprid treated veneer barrier showing wood with non-digested, sharp edged materials.

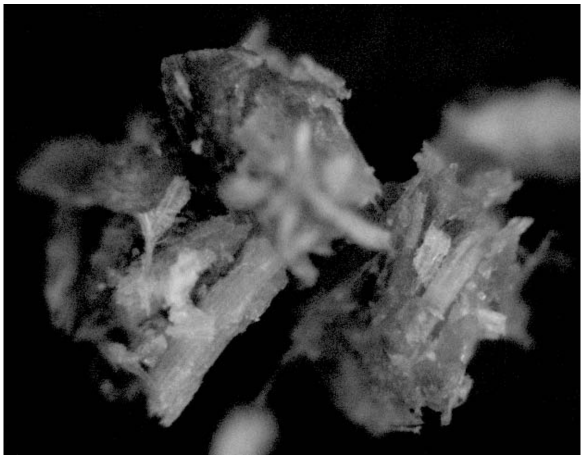

Figure 20. Micrograph (6x) of frass produced by a larva in contact with a 4mm thick ponderosa pine veneer barrier with rounded edges on the individual particles suggestive of digestion of the wood.

Exposure of pupae to a simulated treated envelope

Only three pupae developed from the original 35 larvae, the remaining larvae perished during the rearing process and were rapidly overgrown by fungi. The death of these larvae was probably due to laboratory conditions that were suitable for larval survival, but were not attuned to the nutritional needs required to pupate. One pupae completed its life cycle and emerged as an adult from an untreated ponderosa pine block within 4 days, the second pupae reached the adult stage, and produced a substantial amount of frass before dying, while the third pupae never reached the adult stage. A fourth pupae recovered directly from the ponderosa pine boards was placed inside a ponderosa pine block covered with 2mm thick treated imidacloprid veneer. This pupa reached the adult stage and began to excavate a passage; however, the adult never reached the treated shell, and was found dead within the gallery. While, the results were inconclusive, they indicate that placing pupae within wood blocks cut from the same species as the host material may be an acceptable way to test the ability of these organisms to penetrate a treated shell.

Implications

The results suggest that the current formulations of ACQ, DOT and the experimental treating solution containing imidacloprid cannot control established wood boring insects within SWPM when the larvae cannot be reached directly during the treating process. Larvae could potentially exit the treated material and enter adjacent non-treated woody materials where the insect could complete its life cycle and emerge as an adult. These adults could result in an invasive species introduction. However, field trials containing materials treated with ACQ, DOT or imidacloprid indicate that adult emergence can be prevented (Chapter 5). In addition, limited trials conducted using chlorpyrifos suggest that chemical impregnation can limit emergence of these types of insects in SWPM, even in situations were the chemical does not directly contact the larvae during the treating process.

A method for determining the effectiveness of a particular phytosanitary method was also developed during the course of this study. This method allows for the evaluation of different treatments under more controlled laboratory conditions and would be useful for investigating treatments that do not come into direct contact the pests during the application process. The method is also useful because it allows the larvae to be observed as they interact with the treatment as a part of their normal foraging activity.

Additional Research Needs

One of the drawbacks of the current study was the lack of detailed information on the biology of the target organisms. This made it difficult to assess treatment effects on the larvae. Improving our understanding of the developmental stages of the target insects could allow researchers to better assess the implications of the barrier penetration behavior. The Petri dish assays, while time consuming, appear to provide a simple method for assessing this behavior; however, considerably more larvae and other beetle species need to be examined.

The method could also be used to assess the ability of the larvae to ingest treated materials both by analyzing frass and by destructively sampling the larvae to assess metabolic activity and physiologic response.

Further research is also recommended to develop more target surface treatments that are both toxic to the insects and repellent. In addition, a better understanding of the chemical concentration needed for effective protection is needed. At the same time, identifying new treatment chemicals must also be pursued. The majority of wood treated worldwide is impregnated with heavy metals that would be unacceptable for solid wood packing since much of this material is burned at the end of its useful life. Creating heavy metal ash as well as the potential for airborne contaminants would pose unacceptable health risks. The identification of more effective organic molecules could mitigate these environmental and health risks.

Conclusions

New house borer larvae were clearly capable of excavating through veneers treated with three common biocides, although they appeared to avoid digestion of the resulting particles. The results suggest that larvae can survive treatment and could penetrate through a treated shell to move into adjacent untreated wood. The results suggest that more effective treatments must be developed to limit this larval migration.

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CONCLUSIONS

Wood preservatives applied in a commercial treating facility penetrated the vast majority of insect galleries found within refractatory wood. However, complete penetration could not be achieved and a thick chemical impregnated envelope could not be developed. Since complete penetration could not be accomplished, laboratory trials were conducted to determine if a preservative treated barrier could prevent the emergence of both immature and mature insects from the wood. Laboratory studies found that barrier thickness did not influence the ability of the larvae to exit the material. This finding was confirmed through a field trial which indicated that three wood preservatives had little affect on the ability of larvae to penetrate the wood surface. However, no adults emerged from the treated samples, indicating that current wood preservatives acted as insecticides rather then larvicides. This data suggests that wood preservative systems can be used as a mitigation tool for SWPM since adult insect emergence is prevented. However, the chemical formulations should be altered to increase their effectiveness against the larval stages of wood boring insects.

Additional Research Needed

The results highlight a number of areas were additional research will be required in order to prevent accidental species introductions. The biology and behavior of many wood boring insects remains poorly understood. The lack of data concerning the feeding patterns of these insects and the processes used by the larvae to digest cellulose hamper efforts intended to introduce larvicides into these organisms through ingestion. In addition, basic life cycle data, such as the number of instars and degree days required for pupae development are essential in any control effort. However, this data is not available for many wood-boring insects. Other data, pertaining to the permeability of the larvae skin would also be helpful for the development of more effective contact larvicides. In order to conduct this research, however, non-destructive methods must be developed to observe the larvae in situ.

Research pertaining to the development of certifiable sanitation systems for SWPM will also be required. These systems must be effective against the broad range of insects and fungi that may be present in or on SWPM, while being specific enough to prevent the accidental poisoning of non-target organisms. The development of these systems is further complicated by the fact that most SWPM is used for other purposes once it has completed its service life. Two of the uses of greatest concern to those developing treating systems are the use of this material as fuel in low temperature cooking fires and for house construction. Both of these uses can potentially lead to the long-term exposure of humans to treatment chemicals and, in the case of fires, to their combustion components as the material is burned.

Current mitigation methods have already greatly reduced the number of invasive species introductions. Thus, if an interdisciplinary approach is used to answer these research questions and provide a greater understanding of both the ecological and biological functions of wood boring insects and potential methods of preventing their spread, the invasive species threat associated with international trade, resulting from the use SWPM (a renewable resource), can be sharply limited. The new preservative systems resulting from such efforts could also prevent the accidental poisoning of non-target organisms in other applications where treated lumber is used.

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