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Efficacy of microwave irradiation for phytosanitation of wood packing materials

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ABSTRACT

Wood packing materials (WPM) are important vectors of invasive xylophagous insects and pathogenic and decomposer wood fungi. The International Plant Protection Convention introduced the International Standards for Phytosanitary Measures No. 15 (ISPM No. 15) to regulate the development of treatments to sanitize WPM and prevent the introduction and movement of forest pests. Dielectric heating (e.g., microwave irradiation) has recently been included as an accepted treatment. In this study, the efficacy of microwave irradiation was tested on *Monochamus scutellatus* larvae and on four different pathogenic fungi, *Gremmeniella abietina*, *Heterobasidion annosum*, *Chondrostereum purpureum*, and *Mycosphaerella populorum*, five species of economic significance in Québec, in both jack pine and trembling aspen. We explored different temperature/time combinations on each species in order to accumulate data on the treatment. We irradiated *M. scutellatus* larvae at 56, 61, and 66 °C for 1–3 min and the four fungal species at 50, 55, 60, 65, 70, 75, and 90 °C for 0.5, 1, or 2 min. Fungi were tested at a wider range of temperatures to account for possible higher variation of resistance between species. We obtained 100% mortality in larvae treated at 56 °C for 2 min and at 61 °C for 1 min. The fungi species were much more resistant to the treatment. *G. abietina* was eliminated at 75 °C/0.5 min, *H. annosum* at 90 °C/1 min, *M. populorum* at 90 °C/2 min, and *C. purpureum* was still present at the highest temperature/time combination used. We demonstrated the capacity of microwave irradiation to kill the larvae with similar parameters as IPPC guidelines (60 °C for 1 min), though lethal temperatures for the fungi were very high. As the current ISPM No. 15 standard for microwave irradiation was insufficient to kill all tested fungal species, more work should be done on determining optimal combinations for the greatest number of species. Future studies should test a wider range of treatment times and expand trials to include more insect and fungal species to determine which temperature/time combination will allow us to keep both values as low as possible while assuring complete prevention of adult insect emergence and fungal re-growth.

1. Introduction

Invasive forest insect pests and plant pathogens cost an estimated $2.1 billion US each year in losses and damage within the United States (Pimentel et al., 2005). Wood boring beetles from the family Cerambycidae are among the most frequent and problematic insect groups which are regularly introduced through wood packing material (WPM) (Brockerhoff, 2009; Haack, 2006). Many serious pathogens are transported through dunnage, insect vectors, wood chips, and WPM (Mireku and Simpson, 2003). Large-scale use of microwave irradiation to eliminate wood boring insects and fungal pathogens inside WPM, including wood pallets, may become an important part of the overall effort to minimize the introduction of invasive pests across borders. The International Plant Protection Convention (IPPC) published the International Standards for Phytosanitary Measures No. 15 (ISPM No. 15) which set guidelines for the regulation of WPM in international trade in order to stem the accidental introduction of invasive pests (FAO, 2014). Signatory countries, such as Canada and the United-States, have

Abbreviations: IPPC, international plant protection convention; ISPM No. 15, international standards for phytosanitary measures no. 15; MC, moisture content; LT, lethal temperature.

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adopted these guidelines and inserted them into their own regulations for the control of out- and in-bound WPM (CBSA, 2012; USDA, 2004). Currently, the ISPM No. 15 standard recognizes several methods for the treatment of wood packing material including methyl bromide fumigation, heat treatment using conventional steam or dry kiln heat chamber and heat treatment using dielectric heating. Methyl bromide fumigation is regulated according to specific temperature and concentration combinations for a treatment period of 24 h, whereas steam and dry kiln heating require that wood core temperature be held at 56 °C for 30 min, a temperature/time combination determined initially to kill the pine wood nematode (*Bursaphelenchus xylophilus* [Steiner and Buhrer] Nickle), the causative agent of pine wilt disease (Ramsfield et al., 2010). Methyl bromide is however a short-lived gas which has been shown to contribute to the depletion of the ozone (WMO, 2006). The Montreal Protocol on Substances that deplete the ozone layer singled out methyl bromide and signatory countries are required to have completely phased out its use by 2013 (Madé, 2012; UNEP, 1994). Contracting parties to the IPPC are therefore encouraged to search for alternatives to fumigation (FAO, 2014). Conventional heating in kilns and with steam can only heat the wood core by transfer of energy from the outside-in and is more time consuming. Dielectric heating, such as occurs during microwave irradiation, affects water molecules more directly and can raise the inner temperature of wood much faster than conventional heating. Dielectric permittivity is a measure of how a medium is capable of being polarized by an electromagnetic field. The relative permittivity of water is very high whereas that of wood is much lower. Organisms within the wood are therefore affected more directly than they would be when treated with conventional heating. Several studies have tested the effects of microwave irradiation on the dielectric properties of wood to test the distribution of heat and the effect of moisture content, and have found that higher moisture contents lower the treatment's capacity to heat wood (Antti and Perré, 1999; Koubaa et al., 2008). The consensus is that higher percentages of moisture absorb more heat and lower the capacity of the microwaves to heat the wood. Heating efficiency also varies according to structural factors, such as the direction of the grain and the heterogeneity of the wood's interior (Antti and Perré, 1999). This does not however cause changes in the structural integrity of the wood and it has been suggested that moving the applicator or rotating the wood could remedy the situation (Antti and Perré, 1999; Fleming et al., 2005). Several studies have tested the efficacy of the treatment on insect pests within the wood for phytosanitation purposes and found it to be effective at temperatures ranging from 60 to 65 °C for much shorter time intervals (<5 min) than the 30 min suggested by the ISPM No. 15 for conventional heating (Fleming et al., 2003, 2004, 2005; Henin et al., 2008; Hoover et al., 2010; Nzokou et al., 2008). Many studies however did not focus on determining the optimal temperature/time combination and mostly tested temperatures for one treatment time or tested much higher times than are necessary to kill many wood pests.

The ISPM No. 15 standard was recently modified to include dielectric heating and requires that wood not exceeding 20 cm in its smallest dimension be heated at 60 °C for 1 continuous min with a maximum period of 30 min to reach the required temperature in the entire volume of the wood (FAO, 2014). As this is a relatively new treatment, the present study has the objective of increasing the amount of data available by testing microwave irradiation as a viable phytosanitary treatment in northeastern North America. We selected larvae from a common species of Cerambycidae, *Monochamus scutellatus* (Say), which is native to North America, as well as four pathogenic fungi, *Gremmeniella abietina* (Lagerberg) Morelet, *Heterobasidion annosum* (Fr.) Bref., *Chondrostereum purpureum* (Pers.) Pouzar and *Mycosphaerella populorum* G.E. Thompson.

M. scutellatus lays its eggs beneath the bark of weak and dying conifers (Peddle, 2000). The larvae bore deep into the wood and are often the cause of economic losses in the lumber industry as they attack cut logs in fields and lumberyards, reducing the quality of the wood. Though *M. scutellatus* has not yet been shown to be so, many species in the *Monochamus* genus (*M. carolinensis* [Olivier], *M. altematus* Hope, and *M. salruarius* [Gebler]) are vectors of the pine wood nematode which has caused heavy damages in pine forests in Japan, China, Korea and Taiwan (Fielding and Evans, 1996; Kosaka et al., 2001; Shi et al., 2012; Suzuki, 2002; Zhao et al., 2007) and in Portugal and Spain (Abelleira et al., 2012; Robertson et al., 2011).

Pathogenic fungi capable of killing trees are of great economic significance. Epidemics of Dutch elm disease (Dedic and Zlatanovic, 2001) (*Ophiostoma ulmi*; *Ophiostoma novo-ulmi* [Brasier]) and sudden oak death (*Phytophthora ramorum* [Werres et al., 2001]) are examples of the risks posed by non-indigenous fungi. The criteria in ISPM No. 15 are well adapted to xylophagous beetles and the pine wood nematode but have been shown to be insufficient for several fungi species (Ramsfield et al., 2010). More data shoud therefore be collected to adequetely adjust accepted treatment norms. The four fungi species used in this study were selected based on their presence in the northeastern North America and on their likeliness of selecting hosts commonly used in wood pallet production in this region. In addition, they are minor pests in forests and plantations of certain regions and have all been reported as being found outside North America, meaning that their transport via imported and exported wood is a likely occurrence. Of the four, only species of the *Heterobasidion* genus have been found to have successfully invaded new areas without the help of an insect vector (Gonthier et al., 2012, 2004), though this does not preclude that the others are not capable. *H. annosum* is an economically significant conifer root and butt rot fungus in the Northern Hemisphere (Asiegbu et al., 2005; Lygis et al., 2004). *C. purpureum* is the causal agent of silver leaf disease on many fruit trees and scrubs (Beever, 1970) and is used as bioherbicide against unwanted non-indigenous trees (Pitt et al., 1999; Ramsfield et al., 1996). *G. abietina* is the causal agent of scleroderris canker and attacks mainly pine in North America and sometimes spruce (EPPO, 2009; Wallace, 2012). Finally, *M. populorum* is the causal agent of septoria canker and attacks all species of *Populus* in North America as well as some exotic and hybrid poplars (EPPO, 1980). The accumulation of data on fungal resistance to microwave irradiation will help establish important criteria for effective treatment in case of newly discovered outbreaks.

This study's objective was to determine minimum microwave irradiation combinations of temperature and time required to eliminate cerambycid larvae (*M. scutellatus*) and four different fungal wood pathogens (*G. abietina*, *H. annosum*, *C. purpureum*, and *M. populorum*), all indigenous or established in northeastern North America, in industrial grade wood such as would be found in wood pallets and other types of wood packing materials. Based on the results from previous studies on the heat treatment of wood-boring insect larvae (Fleming et al., 2003, 2004, 2005; Henin et al., 2008; Hoover et al., 2010; Nzokou et al., 2008; Ramsfield et al., 2010) and of fungal pathogens (Ramsfield et al., 2010; Tubajika et al., 2007), we hypothesized that microwave treatments at temperatures of 56 °C or higher at 2 min would be sufficient to kill all larvae and at temperatures of 70 °C or higher would be sufficient to provoke mortality and inhibit re-growth in all fungi species, given the generally higher resistance of fungi to harsh conditions. We also expect that the higher porosity of jack pine than trembling aspen will allow the microwaves to heat more effectively the organisms inside and therefore decrease the temperature level required to kill them in the softwood blocks versus in the hardwood blocks.