Evaluating the Effect of a Diluent Composed of Biodiesel and Diesel on the Efficacy of the Wood Preservative Creosote

by

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Abstract

The goal of this project was to evaluate the effect of a diluent as a co-solvent in the wood preservative creosote. The diluent was composed of 80% soybean biodiesel and 20% petroleum diesel. Three main experiments were completed to meet this goal. One was a fungal analysis to investigate the effect of the diluent on the efficacy of creosote against wood decay caused by white - rot and brown - rot wood decay fungi. No significant weight loss of the wood blocks occurred when the diluent was present. Another was to quantify the naphthalene present as an odor indicator in creosote by analyzing headspace gas. The results showed that there was a significant decrease when the diluent was added. The last was to investigate leaching of creosote components from treated wood. Results proved to meet environmental regulations. Overall, the results indicated that the diluent had no negative effects on creosote.

List of Abbreviations Used

ACA	Ammoniacal Copper Arsenate
ATCC	American Type Culture Collection
ATSDR	Agency for Toxic Substances and Disease Registry
AWPA	American Wood Protection Association
CA	Copper Azole
CA-B	Copper, Tebuconazole
CBA-A	Copper, Boric Acid, Tebuconazole
CCA	Chromated Copper Arsenate
CCME	Canadian Councils of Ministers of the Environment
CSA	Canadian Standard Association
GT	*Gloephyllum trabeum*
MCA	Micronized Copper Azole
NL	*Neolentinus lepideus*
NOISH	National Institute for Occupational Health and Safety
NPIC	National Pesticide Information Center
PAH	Polycyclic Aromatic Hydrocarbons
PCP	Pentachlorophenol
PP	*Postia placenta*
RBCA	Risk Based Corrective Action
RTA	Rail Tie Association
SH	*Stereum hirsutum*
TPH	Total Petroleum Hydrocarbons
TV	*Trametes versicolor*
USEPA	United States Environmental Protection Association
VOC	Volatile Organic Compound

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Chapter 1.0 Introduction

A trend towards improved environmental standards and sustainable products has forced industries to change their production methods to comply with new regulations. For example, in 2016, the provincial government of Alberta stated that their emissions goal for gaseous naphthalene was 3 $\mu g/m^3$ (Alberta Environment, 2016). Wood preservation companies have been striving to use more environmentally friendly production methods to provide sustainable products while still meeting their high standards in a competitive industry. Wood preservation companies are a large part of the economy. They produce utility poles, railway ties, bridge construction, building frames, decks, flooring and many other products (Bolin & Smith, 2013).Wood that has been treated with a wood preservative has a service life 6- 8 times longer than that of wood that is untreated. For instance, untreated railway crossties have a serviceable life of five years, whereas creosote treated ties could last over 30 years (Bolin & Smith, 2013).

Coal tar distillate is the official name for creosote in the United States and Canada (USEPA, 1999) and has been used as a wood preservative since 1948. It is a heavy-duty, oil borne preservative. Some pesticide products have creosote as an active ingredient to protect against termites, fungi, and other pests that can degrade or threaten the quality of wood products.

Though creosote is a very important wood preservative there are many environmental and health hazards associated with it. Long-term exposure could cause ulceration, potential cancer, and severe burning (ATSDR, 2002a). It is a restricted pesticide that can only be used in outdoor settings such as for rail ties or utility poles. Indoor usage is prohibited as well as usage around water and food supplies.

Creosote contains polycyclic aromatic hydrocarbons, otherwise known as PAH's. They generally have two or more aromatic rings and result from the incomplete combustion of coal (Simarro et al., 2013a) through either volcanic, geological or anthropogenic activities. They are formed specifically through cyclization. Light PAH's contain up to four benzene rings fused together, while more than four benzene rings fused together are considered heavy PAHs (Haritash & Kaushik, 2009). The United States Environmental Protection Agency (USEPA) has classified sixteen PAH's as priority pollutants which are toxic to the environment, potentially carcinogenic, and have an unpleasant and potent odor (USEPA, 1999). Creosote also contains phenolic compounds that are more soluble in water than the PAH's which are also considered hazardous chemical compounds (Environment Canada, 2013).

Due to the importance of creosote in the wood preservation industry, Stella-Jones Inc., has been funding research to determine how to make creosote a more environmentally friendly product. Through their research they have found that the addition of biodiesel as a co-solvent suppresses the potent odor of creosote. Since biodiesel is derived from plants

or animals it is environmentally friendly. To date, there have been no in-depth studies regarding biodiesel as a co-solvent in creosote.

Figure 1.1 shows the difference before and after creosote is applied to rail ties. It is evident from this image that the ties are completely treated with creosote. Emphasis can also be made on the fact that since these ties are going in the soil it is important to ensure that they will be as environmentally friendly as possible.

Figure 1.1: Photo of rail ties treated with creosote after preservative cycle, next to untreated ties at the Stella-Jones Inc., Truro Plant.

1.1 Objectives

The goal of this study was to investigate the effects of a diluent when added to creosote. The diluent was composed of 80% soybean biodiesel and 20% petroleum diesel. Such a diluent would only be beneficial as a co-solvent if the quality of creosote was not sacrificed. To reach this goal, several specific objectives were evaluated:

(i) investigate the use of the diluent as a co-solvent upon the efficacy of creosote against wood decay, caused by five specific wood decay fungi (including creosote tolerant fungi).

(ii) quantify the amount of naphthalene present in the headspace of creosote when the diluent was added as a co-solvent.

(iii) investigate leaching, to determine if regulations would be met if the diluent was added to creosote as a co-solvent.

Chapter 2.0 Literature Review

2.1 Wood

Wood is an organic compound that has fibrous and porous structural tissue (Pasangulapati et al., 2012). The xylem can be broken down into the heartwood and sapwood. The heartwood is the innermost layer of a tree, and it is surrounded by sapwood (Plomion et al., 2001). The structure of a tree can be broken down from exterior to interior as the outer bark, the inner bark, the vascular cambium, the sapwood, the heartwood, and the pith. A diagram depicting a cross section of a tree can be observed in Figure 2.1.

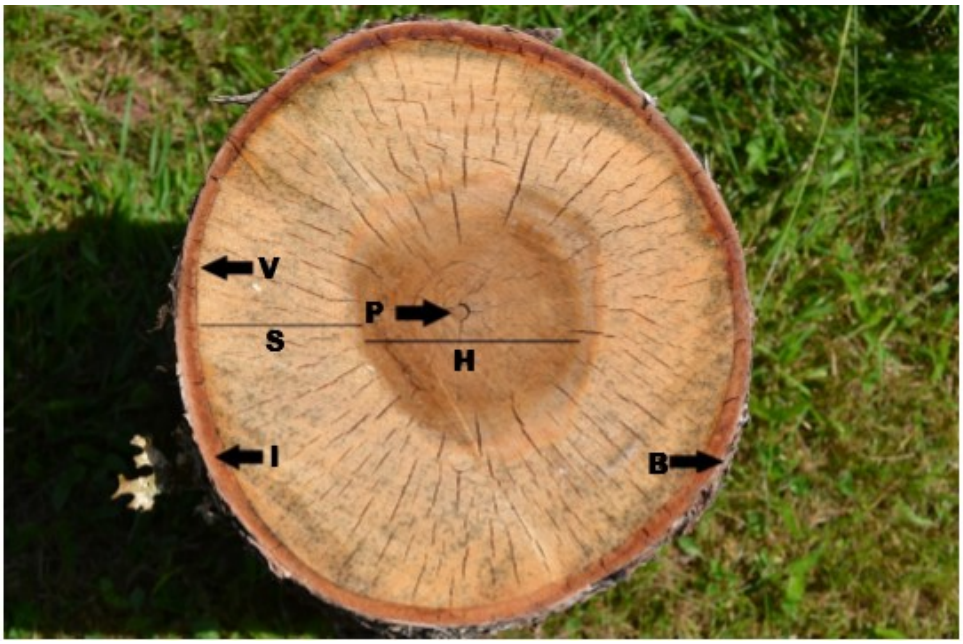

Figure 2.1: Cross-section of a tree illustrating the (B) outer bark, (I) inner bark, (V) vascular cambium, (S) sapwood, (H) heartwood, and (P) pith. Scale- tree diameter is 30 cm. Sourced From McKillop, (2014) with permission.

The outer bark protects the tree from harsh climate conditions, fungi and insects. The inner bark is referred to as the phloem and acts as a storage and transport area as it transports sugars and nutrients from the tree's leaves to various regions of the wood (Wiemann, 2010). The vascular cambium is capable of becoming bark or forming the xylem (wood) of the tree. It is also the separator of bark and wood. The separation is due to two cell layers where phloem cells are on the outside and xylem are on the inside layer (Plomion et al., 2001) The sapwood is the youngest growth layer. It is responsible for water and mineral transportation as well as starch storage (Plomion et al., 2001). Finally, the heartwood is the oldest growth layer of wood. It is formed from sapwood as it ages, and it is important for strength and support although the cells are no longer living (Wiemann, 2010). Eventually the heartwood becomes infiltrated by gums and resins which makes wood preservation difficult because the preservative cannot enter the wood cells (Wiemann, 2010).

2.2 Wood Decay

Wood decay is the biological process where energy is released into the soil because, lignin and cellulose are converted to carbon dioxide and water (Shortle & Dudzik, 2012). Due to the cellulose, hemicellulose, and lignin in the wood, wood decay fungi are a concern in the wood preservation industry (Morrell, 2011). Some preliminary precautions are taken in order to ensure that the wood receives as little fungal damage as possible. For instance, in order to improve fungi resistance, before the wood treatment

process has begun, workers may harvest the trees in colder months when insects are dormant and the fungi grows slower (Morrrell, 2011). There are three main types of fungal decay which include soft-rot, brown-rot, and white -rot. Fungi are classified by the way they break down wood. Soft-rot fungi are generally found in areas that are in or near aquatic environments. Brown rot and white rot fungi are generally classified in the phylum *Basidiomycota* (Riley et al., 2014).

Brown rot fungi break down cellulose and hemicellulose leaving a modified lignin that may crack and crumble over time (Riley et al., 2014). Brown rot decay is more dangerous than white rot decays because the interior of a tree could be rotted out before there are signs on the exterior; and it quickly loses strength (Shortle & Dudzik, 2012). Some species of Brown-Rot fungi include *Postia placenta, Neolentinus lepideus* (creosote tolerant), and *Gloephyllum trabeum.* White rot fungi break down cellulose, hemicellulose and lignin completely into carbon and water (Hori et al., 2013). Two examples of white rot fungi are *Trametes versicolor* and *Stereum hisutum.*

2.2.1 Brown-Rot Decay

Brown-rot fungi attack wood through hemicellulose degradation which begins with an initial cellulose attack by extracellular hydroxyl radicals; [-OH] from the Fenton reaction [$H_2O_2 + Fe_2+ + H+ \rightarrow H_2O + Fe_{3+} + -OH$] (Hori et al., 2013). As the hemicellulose components of the wood degrade, then support is lost leaving the cellulose vulnerable and it becomes hydrolyzed easily (Curling et al., 2002) .

2.2.2 White-Rot Decay

White rot fungi can either break down wood through simultaneous rot or selective rot. Simultaneous rot is a process where white-rot fungi will decompose each part of the cell wall at similar rates. In simultaneous rot, structural carbohydrates are removed at the same rate along with the lignin (Pandey & Pitman, 2003). The wood cell is penetrated by the microhyphae which cause holes in the secondary wall of the wood cell; the holes enlarge as decay continues. On the other hand, when selective rot occurs the lignin and hemicellulose are degraded first. The breakdown of lignin of hemicellulose results in the breakdown of the lamella (Pandey & Pitman, 2003). In order for this to occur, the hyphae invade the lumen of the wood cells and molecular substances are secreted by the hyphae. Ideal conditions for fungal growth and wood decay include moisture, warmth, and oxygen. Another major factor for wood decay are insects (Morrell, 2011).

2.2.3 Evaluation of Wood Decay

Standards have been set by the American Wood Protection Association (AWPA), the Canadian Standards Association (CSA), and several other organizations regarding analysis of decay fungi and its interaction with wood (AWPA, 2016). Along with these standards, researchers have also developed additional methods to analyze fungal growth on wood. For instance, Riley et al. (2014) conducted a study where brown and white rot fungi were examined. The overall objective of the study was to assess the generality of the brown and white rot classification paradigm. They assessed 33 different basidiomycetes using a principle component analysis. From their study they concluded

that a more nuanced system of categorization was required based upon improved understanding of wood decay. Bari et al. (2017) collected wood that had decayed and analysed the decay patterns. They found that at both laboratory conditions and natural conditions, simultaneous white rot fungi degradation occurred (Bari et al., 2017). Through many applications, wood products are prone to fungal decay, therefore using herbicides and insecticides is essential to extend their service life.

2.3 Wood Protection

Wood preservation has been attempted as far back as the ancient Greeks and Romans when they used olive oil to protect the wood used to build bridges (Richardson, 1993). There are several different ways to preserve wood such as brushing, spraying, dipping, and immersing the wood in baths of changing temperatures, or one of several different pressure treating cycles (Rabbi, et al., 2015). Brush and spray treatments are the simplest, requiring little of the complex machinery needed for other treatments at the expense of more labor being required. However, the lifespan is only slightly extended, a maximum being three years (Townsend, 2016). It was not until the nineteenth century during the industrial revolution when wood preservation became popular and scientists spent time developing wood treating methods involving heat, pressure, and vacuum applications.

There are three main steps to pressure treat wood, which consist of pre-treatment conditioning, pressure treatment and post-treatment conditioning. Moisture is removed from the peeled and sawn wood during pre-treatment because water acts as a physical barrier to the preservative (Environment Canada, 2013). The reduced moisture minimizes

leaching after treatment. The preservative treatment is either a full cell or empty cell process which refers to the way that the chemical interacts with the wood cells (Richardson, 1993). Post-treatment conditions include an expansion bath, steaming, accelerated fixation, and stabilization, which are used to minimize leaching and bleeding after the treatment process to remove excess preservatives (Wiemann, 2010).

2.3.1 Full Cell Treatment

Full cell pressure treatment was first invented in the ninetieth century by an American inventor, John Bethel (Bethel, 1838). It involves drawing air and water out of the wood cells with an initial vacuum to allow for the cell to be completely filled with preservative during the pressure period. This allows for very high net preservative retention. A diagram of a full cell cycle can be observed in Figure 2.2.

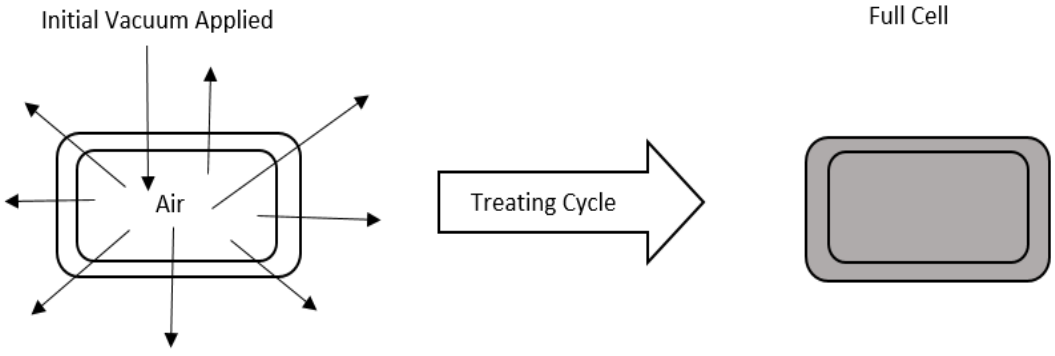

Figure 2.2: A diagram of the full cell process demonstrating the deposition of the preservative solution (modified after Hiziroglu, 2014)).

During the Bethel cycle, a vacuum is applied to the cylinder to remove air from the cells (Environment Canada, 2013). The vacuum is maintained while the cylinder is filled with preservative. Once the cylinder is full, pressure is applied and held for a period of time. After the pressure is released, the cylinder is emptied; and an additional vacuum is applied to remove excess preservative. (Environment Canada, 2013).

2.3.2 Empty Cell Treatment

Empty cell treatment involves an initial pressure which forces air into the cell. This process reduces the net retention of the preservative without sacrificing penetration (Hiziroglu, 2014). Due to the compressed air the preservative only penetrates the cell wall. The empty cell treatment is more common than the full cell treatment because it requires less preservative due to the air being forced into the cell.

There are two methods for the empty cell treatment known as the Rueping cycle and the Lowry cycle. The Rueping cycle was developed in 1902 and later modified into the Lowry cycle in 1906 (Lowry, 1906). The difference between them is that there is an initial pressure in the Rueping cycle which is absent in the Lowry cycle. A diagram of the empty cell cycle can be observed in Figure 2.3.

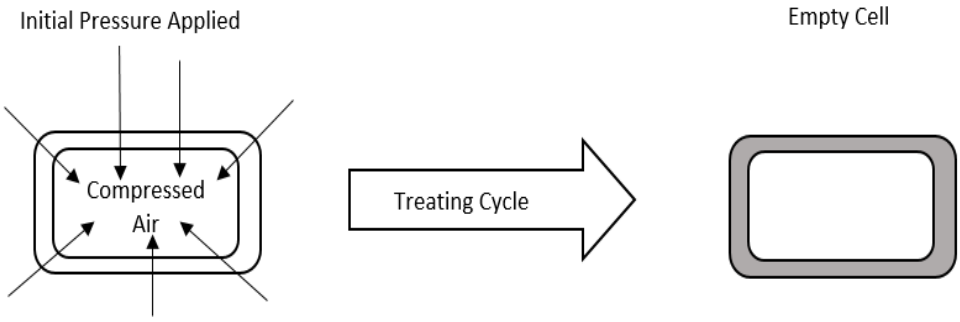

Figure 2.3: A diagram of the empty cell process demonstrating the deposition of the preservative solution (modified after Hiziroglu, 2014).

In an empty cell treatment cycle following the Rueping procedure the cylinder is pressurized before the preservative enters (absent in Lowry). After the initial pressurization the cylinder is filled while under pressure and remains under pressure for a given period of time (Environment Canada, 2013). After the pressure cycle is complete the preservative is drained from the cylinder and a post treatment vacuum is applied to prevent bleeding of the preservative from the wood (Wiemann, 2010).

2.4 Wood Preservatives

Wood preservatives are important to maximize the service life of wood when it is being used for industrial purposes which include but are not limited to railway ties, utility poles, and highway guards. Wood preservatives can be broken into two categories, oil-borne, and water-borne. Preservatives must follow specific standards set by government, and industrial standards in order to assure high quality and minimum risk.

2.4.1 Water-Borne Preservatives

Water-borne preservatives are soluble in water and protect wood against decay and insects (Prestemon, 1994). They are mainly used in residential settings because of their dry and paintable surface. They are not toxic to be around nor have a foul odor like other preservatives. Chromated Copper Arsenate (CCA) for instance was used as a wood preservative since the 1940's (USEPA, 2017). It is a mixture of chromium, copper, and arsenic and it is formulated as oxides and salts. It can be used above ground or immersed in either salt water or fresh water. It was used in residential settings until 2003 when it was discontinued for homeowner use. The main reason that CCA was withdrawn from common usage was because of the arsenic in the preservative which is highly toxic (NPIC, 2014).

Copper Azole (CA) is a water-borne wood preservative used to pressure treat products for most exterior residential applications, such as decks, patios, landscaping timbers, gazebos, residential fencing, walkways, boardwalks, and freshwater docks (USEPA, 2017). There are two types of CA available. The first is CBA-A which contains copper, boric acid, and tebuconazole. The second CA preservative is classified as CA-B which contains higher levels of tebuconazole, but it does not contain boric acid (NPIC, 2015).

Micronized Copper Azole (MCA) is a micronized copper-based wood preservative used in the pressure treatment of wood products for residential uses. It is specifically used above ground, or in contact with fresh water (NPIC, 2015). Micronized copper is coupled

with micronized tebuconazole and copper is suspended in the preservative for long-term protection. Research conducted by the EPA indicates that MCA is more environmentally friendly than CA because it leached from the wood at a much lower rate (USEPA, 2017).

2.4.2 Oil-Borne Preservatives

An Oil-Borne wood preservative is a solution of chemicals that are dissolved in a non-aqueous solution (Prestemon, 1994). The three most common oil borne wood preservatives are Copper Naphthenate, Pentachlorophenol, and Creosote. Copper Naphthenate is registered as a non-restricted pesticide. It can be applied by brush, dip, or pressure treatment (USEPA, 2017). It is effective at protecting wood against insect damage. Copper Naphthenate is an organometallic compound with a distinct green color. It can be produced in numerous ways such as mixing sodium naphthenate with a copper sulphate in an aqueous solution or a fusion method can be used where copper oxide and carbonate are dissolved into naphthenic acid when it is heated (USEPA, 2017).

Pentachlorophenol is an oil borne wood preservative. It is an organochlorine compound that is used as a pesticide and a disinfectant. It is created through the chlorination of phenol in the presence of a catalyst, at a temperature of 191°C (PubChem, 2018). When used to preserve wood, the wood can either be placed in a pressure vessel or it can simply be sprayed onto the wood, however studies have found that pressure treatment is more efficient (Stone, 2008). In 1987, pentachlorophenol was classified as a restricted product to industrial use only and it not to be used for residential or pesticide purposes. It is

considered a carcinogen and exposure can put individuals at risk for other health problems (ATSDR, 2001).

The third oil borne preservative is creosote. Creosote is used for commercial purposes only and it is not registered for residential use. It is a restricted pesticide that can only be used in outdoor settings such as for rail ties or utility poles. Indoor usage is prohibited as well as around water, and food supplies. Since the focus of this thesis is creosote, it will be looked at in greater detail than the other preservatives.

2.4.2.1 Creosote

The United Sates Environmental Protection Agency (USEPA) classifies coal tar distillate as creosote, which was originally discovered in 1834 by Friedrich Ferdinand Runge (Schorlemmer, 1885). Creosote is a black viscous liquid and it has a strong and unpleasant odour. It has an approximate distillation range of 200-400°C and is primarily composed of various phenols, cresols, and organic compounds (Melber et al., 2004). Similar to the other commonly used wood preservatives previously mentioned, creosote offers excellent wood protection, and has been a registered wood preservative since 1948 (Bolin & Smith, 2013).

One of the disadvantages of creosote production is the high variability in the product. The method of distillation used along with the origin of coal has an impact on the chemical composition of the creosote produced. Creosote is composed of thousands of chemicals; many of which are in quantities less than 1%. There are six major groups of chemicals of

which creosote is composed. The first group consists of aromatic hydrocarbons, including polycyclic aromatic hydrocarbons (PAHs) (Melber et al., 2004). PAHs could make up to approximately 70% of the creosote (Bedient et al., 1984). Phenanthrene, acenaphthene, fluorene, anthracene, and pyridine are the most abundant PAHs found in creosote (ATSDR, 2002b). The remaining five groups consist of tar acids/phenolics, tar bases/ Nitrogen containing heterocycles, aromatic amines, sulfur containing heterocycles, and oxygen containing heterocycles (Melber et al., 2004).

To properly treat wood with creosote there are guidelines set by organizations such as the AWPA and the CSA. For instance, according to the AWPA, the standards are as follows: The first is Standard P1/P13 (Standard for Creosote Preservative, whose preservative code is CR), which dictates that this type of creosote is a distillate derived from coal tar. More specifically, it is a distillate derived entirely from tar produced by the high temperature carbonization of bituminous coal (AWPA, 2016). The second AWPA standard is Standard P2 (Standard for Creosote Solution, whose preservative code is CR-S), which also dictates that the material must be a pure coal tar product derived entirely from tar produced by the carbonization of bituminous coal and may be either a coal tar distillate or a solution of coal tar in coal tar distillate. Finally, the third AWPA creosote standard is Standard P3 (Standard for Creosote-Petroleum Solution, whose preservative code is CR-PS), which is a mixture of coal tar distillate and petroleum oil (AWPA, 2016; CSA, 2015).

2.5 Railroad Ties

The primary purpose for creosote today is for preserving railway ties. A railway tie or switch tie is universally known as the rectangular support perpendicular to the rails on a railroad track. A traditional rail tie is made out of primarily hardwood, however in some parts of the world, concrete ties have been adopted and in more recent years a plastic composite (RTA, 2018). However, this trend is not favorable in North America due to economic and environmental purposes. In 2016 there were over 17 million rail ties installed across the US and over 2 million installed throughout Canada (RTA, 2018). To this day, approximately 4% of the wood used is softwood, while most rail ties are composed of hardwood from mid-western and eastern Hardwood forests. The species varies however, the general composition of rail ties is 50-60% oaks, and 40-50% mixed hardwoods (RTA, 2018). Softwoods are not used because they have a simpler basic suture compared to hardwoods. Softwoods only have two types of cells with little variation within. Hardwoods on the other hand have a larger structural capacity due to a higher number of basic cells and high variability within the cells (Wiedenhoeft, 2013.)

2.6 Polycyclic Aromatic Hydrocarbons

Polycyclic aromatic hydrocarbons (PAH) are organic compounds that only contain carbon and hydrogen in the form of aromatic rings. Some form linear arrangements, which are the least stable. The sixteen priority PAH's include: naphthalene, acenaphylene, acenaphthene, flourene, pentanthrene, anthracene, flouranthene, pyrene, flouranthene, pyrene, benz[a]anthracene, chrysene, benz[a]anthrene, benzo[b]flouro-anthene, benzo[k]fluoranthene, benzo[a]pyrene, dibenz[a,h]anthracene, benzo[a]pyrene,

benzo[g,h,i]perylene, indeno[1,2,3-cd]pyrene (Lerda, 2011). PAH's have been linked to various cardiovascular diseases and are classified as carcinogenic (ATSDR, 2002b). Specifically, they have been linked to cancers of the skin, lung, bladder, liver, and stomach (ATSDR, 2002b).

2.6.1 Naphthalene

Naphthalene is a solid, white volatile compound. It is the simplest form of a polycyclic aromatic hydrocarbon (PAH). It is insoluble in water, and it is denser than water. Air pollution standards currently state that the levels of naphthalene must be below 3µg m^{-3} (Alberta Environment, 2016). Naphthalene has a double benzene ring with five double bonds throughout the compound. The molecular weight of naphthalene is 128.73 g mol^{-1} and the chemical formula is $C_{10}H_8$ and is shown in Figure 2.4. It is difficult to break down due to its angular structure as well as resonance and electron induction.

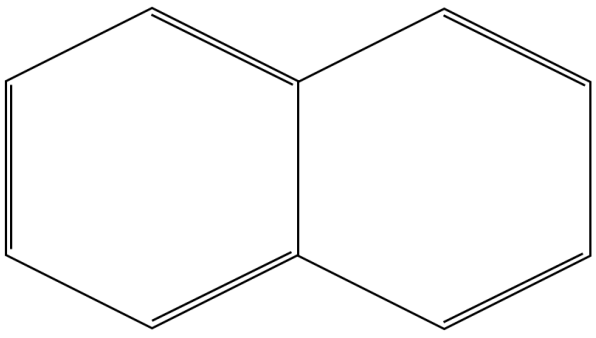

Figure 2.4: Chemical structure of naphthalene ($C_{10}H_8$) showing double benzene rings and double bonds.

Research has been done regarding biosafety of naphthalene existing in soil, such as a study by Simarro et al. (2013b). The objective of their study was to determine if it would be possible to biodegrade various PAH's including naphthalene, phenanthrene, and anthracene using a bacterial consortium. In theory, microorganisms used as a carbon source enriched microbial communities present in PAH-contaminated soils. The results showed that nutrients and soluble forms of nitrogen and iron favor biodegradation at neutral pH. Therefore, they found a way to significantly improve the biodegrading process of PAHs (Simarro et al., 2013b). In another study by (Uhlik et al., 2018) the use of fungi for bioremediation purposes was analyzed. The soil analyzed was contaminated with polychlorinated biphenyl- and poly-aromatic hydrocarbons. A mock community was made from bacterial strains. The results showed that the bacteria were capable of deriving carbon from naphthalene causing it to degrade, which proved the importance of the biodegradation process (Uhlik et al., 2018).

2.7 Olfactory Evaluation

The unpleasant odors from creosote are mainly from PAHs and adding biodiesel to creosote has been demonstrated to suppress odors. In order to evaluate potential risks and hazards associated with odors emitted from creosote it is essential to measure the levels of odors before and after the addition of odor suppressants. The olfactory system is capable of detecting odors from food and their influences. Smell is activated when the odious molecules come into contact with olfactory vesicles (Mainland et al., 2015). Since it is not possible to determine the quantity or even specifically identify compounds

through the olfactory system, then it is necessary to resort to other chemical quantification and identification methods.

Various methods may be applied to quantify naphthalene, a representative of PAHs in the headspace of creosote in a container. Naphthalene specifically was chosen due to its high volatility. Such methods include sorbent tubes, which are glass tubes with active charcoal that collect the PAH's as they pass through the tube. A Gas detector tube works in a similar way except they can read the amount of naphthalene directly after sampling without additional analysis required. Research has also been done using Summa Canisters, Gas Chromatography- Mass Spectrometry (GC-MS), and Headspace Gas Chromatography.

2.7.1 Sorbent Tubes

Sorbent tubes are beneficial for collecting Volatile Organic Compounds (VOC) such as naphthalene. Woolfenden, (2010) found that naphthalene was accurately analyzed with a radial diffusive sampler. In another study conducted by Phuphuakrat et al. (2010) , three absorbents were selected in order to evaluate the absorbent performance of light tars. The three absorbents were wood chips, activated carbon, and a synthetic porous cordierite. The results indicated that the activated carbon showed the best absorption performance out of the three absorbents.

2.7.2 Colorimetric Tubes

Gas detector tubes are employed as an economical and simple way to test dangerous airborne chemicals (Sensidyne, 2013). Gas detection has been used to deal with odor problems such as in a study by Phuphuakrat et al. (2010) where they used a gas detection tube due to the simplicity compared to a GC-MS. However, there are major drawbacks to using a gas detection tube. For instance, many times they prove to only be 20% effective. Furthermore, they are temperature sensitive, and they cannot distinct some chemicals from others due to a short portable column packed with a limited amount of adsorbents (Sensidyne, 2013).

2.7.3 Summa Canisters

Summa Canisters are closed cylinders with a vacuum. They work by attaching the canister to a Teflon tube, and then opening the vacuum valve. The vacuum causes air to go through the tube and into the canister so that the contaminates in air can be quantified. An experiment done by Aurell et al. (2011) analyzed open air sources to determine the emissions from open burning and open detonation. In order to sample the emissions, Summa Canisters were used for open area sampling. Naphthalene along with other PAH's was included in the chemicals analyzed. In a study, by Ochiai et al. (2002) two fused-silica-lined Summa Canister were compared for their recovery of 58 VOC's and then final analysis with a GC-MS. The results indicated that for practical use of both canisters, the relative humidity should be between 50% and 70%. Sin et al. (2001) developed a method where Summa Canisters were coupled with a GC-MS to analyze VOC's in ambient and indoor air. To do this, the stability of the $C_3 - C_{12}$ was monitored

over a course of four months. The results indicated that the method attempted was suitable to measure the air samples in ambient air and semi-confined parking lots.

2.7.4 Headspace Gas Chromatography (GC)

Gas chromatography is an analytical method that identifies different substances within a test sample. There are many advantages to use GC for compound analysis, including its ability to separate complex mixtures, to quantify analytes, and to determine trace levels of organic contamination. GC with a headspace has additional feature which allows to quantify the volatile compounds released to the gas phase from the solid or liquid samples that are sealed in headspace vials.

Previous studies of the use of a Headspace GC for analysis of naphthalene have been conducted. A study by Safarova et al. (2004) analyzed VOC's in waste water, and river waters. Headspace analysis was combined with GC-MS analyzed 53 VOC's including PAH's, chlorinated alkenes, and aromatic hydrocarbons. Another successful analysis of naphthalene using a headspace GC was conducted by Zwiener & Frimmel (1998). A headspace GC was used to analyze the PAH in groundwater. In this study, the objective was to determine the overall contamination level of the typical coal tar leachates. The results indicated that the well samples downgradient from the pollution had decreasing contamination levels. Another study by Eriksson et al. (2001) was completed to determine the hydrocarbons in creosote contaminated soil. To do this, headspace solid phase microextraction and GC-MS was used. The results indicated that the concentrations

for each compound were between 2 and 25 μg/g. The researchers also noted that at low temperatures naphthalene was still able to be detected on a headspace GC.

2.8 Leaching Analysis

Leaching is important to monitor in wood preservation due to the potential negative environmental impact that the preservatives could have on the surrounding ecosystems. Due to the hydroxyl groups' cellulose, lignin, and hemicellulose of which wood is composed, moisture and water are attracted to wood. The reaction of the hydroxyl groups with water cause a decomposition of the wood through swelling, shrinking, or mechanical failure over time (Wiemann, 2010). Furthermore, since creosote is a biocide, leaching could be disastrous to surrounding environments. Many leaching tests focus on water-borne preservatives, such as an accelerated leaching trial specified in the AWPA (AWPA, 2016). There are some experiments that can be applied to oil-borne preservatives. Natural rainfall experiments can also be done in order to have a realistic example of leaching in the environment such as an experiment conducted by Nauss (2001). Rainfall simulators are also used to simulate leaching due to a significant amount of rainfall over a short period of time.

Leaching tests are common to determine the leachability of chemicals such as PAH's from soils. An example of this is an experiment completed by Kim and Osaco, (2003), where the objective was to evaluate the leachability of hydrophobic organic pollutants (HOP). Soil samples were artificially contaminated with PAH's. The results indicated that the ionic strength of the chemical reduced leachability; and that neither the liquid-to-

solid ratio nor the pH influenced the leaching concentration. Another leaching experiment regarding PAH's was conducted by Liu et al. (2008). They used a standardized leaching test to evaluate PAH's in field conditions. A 70-day experiment was simulated to observe the release of PAH's. The test resulted in the PAH's degrading later in the leaching process. A study completed by Enell et al. (2004) was to determine the behavior of PAH's leaching from aged soil. The PAH's were monitored under saturated conditions and the results indicated that the PAH's would only be released from the soil when water was added.

Creosote has been tested for leaching in various rainfall simulations. Xiao et al. (2002) used rainfall simulators to determine the effect that the leaching of creosote had on the environment. Based upon their results, they concluded that there was variability when the flow rate and the temperature increased. A similar experiment involving creosote was completed by Becker, et al. (2001) regarding creosote where they used a method derived from German standard (DIN 38414). The results showed that PAH's as well as nitrogen heterocycles were leachable from creosote. Even though rainfall simulators have potential to be beneficial for environmental analysis, a recent study conducted by Lebow & Hirth, (2017) stated that current standardized methods were not suitable for in-service preserved wood. They compared alternate leaching methods such as E10-16 from the AWPA (2016) as well as methods that have been done under natural exposure. The results suggested that the small block test overestimated the results and the larger block test underestimated the results.

2.9 Biodiesel as an Odor Suppressant

Biodiesel has grown in popularity over the course of the 21st century. A global move has accepted it as a beneficial replacement of traditional diesel in an effort to move towards a more sustainable and environmentally friendly products (Meher et al., 2006). Biodiesel is a fuel composed of monoalkyl esters of long-chain fatty acids derived from renewable vegetable oils or animal fat, while meeting the requirements of ASTM D6751. Biodiesel is made through a process called transesterification. During transesterification, triglycerides react with methanol in the presence of an alkaline catalyst to generate a mixture of fatty acid mono-alkyl esters, otherwise known as fatty acid methyl ester (FAME) as well as a by-product, glycerol (Meher et al., 2006). A diagram of the transesterification process is illustrated in Figure 2.5.

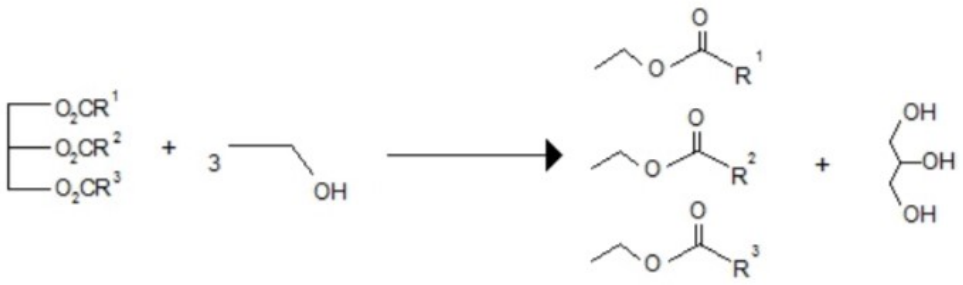

Figure 2.5: A simplified diagram of transesterification (Sourced from McKillop, 2014).

Due to the growing popularity of biodiesel, it has become more common and various avenues of research have been done into additional uses other than just a fuel source. For instance, Stella-Jones Inc., used biodiesel derived from animal fat as well as soybean biodiesel in an attempt to suppress the odor from creosote (Murray, 2016). During their research, they found that animal biodiesel was not beneficial as it had a potent odor on its own. However, the soybean biodiesel successfully supressed the odour of both creosote and pentachlorophenol. It has been suggested that this might be due to the Fatty Acid Methyl Esters (FAME). To further test this hypothesis, they added pure FAMEs to creosote and found that the odour of naphthalene was successfully supressed as tested in the lab (Murray, 2016).

Chapter 3.0 Experimental Methods and Materials

The goal of the present study was to determine whether adding a diluent to creosote would be beneficial. Specifically, regarding odour suppression, fungi resistance and leaching behavior. When the term diluent is used in this thesis, it is referring to a solution of 80% soybean biodiesel and 20% petroleum diesel. This blend of biodiesel/diesel was chosen because it is what the company uses currently.

There were three objectives to the present research. The first objective addressed in Section 3.2 was to determine if there was a significant difference in the efficacy of creosote when the diluent was added. This was addressed using two fungal decay studies. One was a wood block decay experiment where fungal decay of wood treated with creosote was measured. The other was an agar study which did not employ wood. The second objective addressed in Section 3.3 was to determine if the diluent would significantly reduce the quantity of naphthalene available in the creosote blend using headspace analysis. Finally, the third objective is addressed in Section 3.4 and was to quantify the amount of leaching that occurred with wood treated with creosote and diluent.

3.1 Pilot Plant Used for Wood Treatment

The pilot plant as shown in Figure 3.1, was designed as a miniature version of a full-scale wood pressure treatment system. The pilot plant consisted of a 4 L holding tank where the preservative was stored, heated, and then transferred to the pressure vessel by a piston

pump. The pump was also used to pressurize the pressure vessel. A pressure release valve and a pressure gauge were attached to a pipe from the pressure vessel, which dispensed the treating solution back into the storage tank, allowing the pressure to be reliably controlled. The main line had numerous drainage valves so that the system could easily be cleaned.

Figure 3.1: Pilot plant used to treat wood blocks as well as collect samples of liquids and gases.

The treatment cycle in the pilot plant was as follows: 30 min pressure treatment followed by a 30 min expansion bath. The creosote and diluent were mixed in a graduated cylinder before being poured into the holding tank of the pilot plant. After loading wood samples as well as the creosote/diluent into the pilot plant, the temperature was increased to 93.3°C (200 °F), to mimic full scale operation procedures. The temperature of the expansion bath was 98.9°C (200 °F). The treating cycle used a pressure period of 150 psi for 30 min, followed by a 30 min expansion bath. After the expansion bath, the treating cycle was complete, the preservative solution was drained.

3.1.1 Wood Species Evaluated

A series of 50 x 25 x 7.5 mm blocks were milled from hardwood samples of Rock Maple (*Acer saccharum*). Many of the American Wood Protection Association (AWPA) standards indicate the use of a non-durable coniferous wood such as Southern Yellow Pine (SYP), however, Rock Maple was chosen based upon its common use in the rail tie industry.

3.2 Fungal Decay Test

A method was developed to compare the difference in the fungal decay between blocks treated with creosote and blocks treated with creosote and diluent. The method was developed by using a combination of British Standard EN 113: (1997) Wood Preservatives-Testing Method for Determining the Effectiveness Against Wood Destroying Basidiomycetes- Determination of the Toxic Values and AWPA E10-16,

Laboratory Method for Evaluating the Decay Resistance of Wood-Based Materials Against Pure Basidiomycetes Cultures: Soil/Block Test (AWPA, 2016).

3.2.1 Selection of Test Fungi

All of the fungi species used for the decay trial were purchased through Cedarlane which is the Canadian distributer of the American Type Culture Collection (ATCC). The fungi species purchased were based on species listed in the AWPA and British Standard EN: 113. Five species of fungi were utilized for the study, three brown-rot fungi and two white-rot fungi. The brown rot fungi were *Postia placenta (ATCC 11538_TT)*, *Neolentinus lepideus (ATCC 12653)*, and *Gloephyllum trabeum (ATCC 11539)*. The white rot fungi chosen were *Stereum hirsutum (ATCC MYA-2819)* and *Trametes versicolor (ATCC 12679)*. *Gloephyllum trabeum* and *Postia placenta* were both chosen because of their common use in many experiments. *Trametes versicolor* was chosen for its rapid growth, *Neolentinus lepideus* was chosen for its tolerance to creosote and *Stereum hirsutum* was chosen because it was one of the recommended white rot fungi in the AWPA and because it's been included in previous research involving creosote.

3.2.2 Inoculation and Incubation of Microcosms

Postia placenta, Neolentinus lepideus, Stereum hirsutum, and *Trametes versicolor* were cultured according to ATCC Medium 200 (Yeast and Mold (YM) agar or YM broth). While *Gloephyllum trabeum* required ATCC Medium 337 (Potato Dextrose Yeast Agar).

3.2.2.1 Preparation of Medium

The YM agar was purchased from Fisher Scientific and made according to label instructions (41 g/L). Difco™ Potato Dextrose Agar was also purchased from Fisher Scientific and made according to label instructions (39 g/L). Fungal stock cultures were grown on the recommended agar and then transferred to an agar designed for this experiment. McKillop (2014), found that the best agar media for such an experiment was 4% malt, because it elicited the largest mass loss by weight when compared to other malt percentages. In addition to malt, the medium contained 2% of Difco™ Granulated Agar (Fisher Scientific).

Media was transferred from the media jar to 100 mm x 15 mm Sterile, Polystyrene petri dishes using a 25 mL Disposable Serological Pipet. The pipet was attached to a Fisherbrand Motorized Pipette. 20 mL of media were poured into each petri dish, as shown in Figure 3.2.

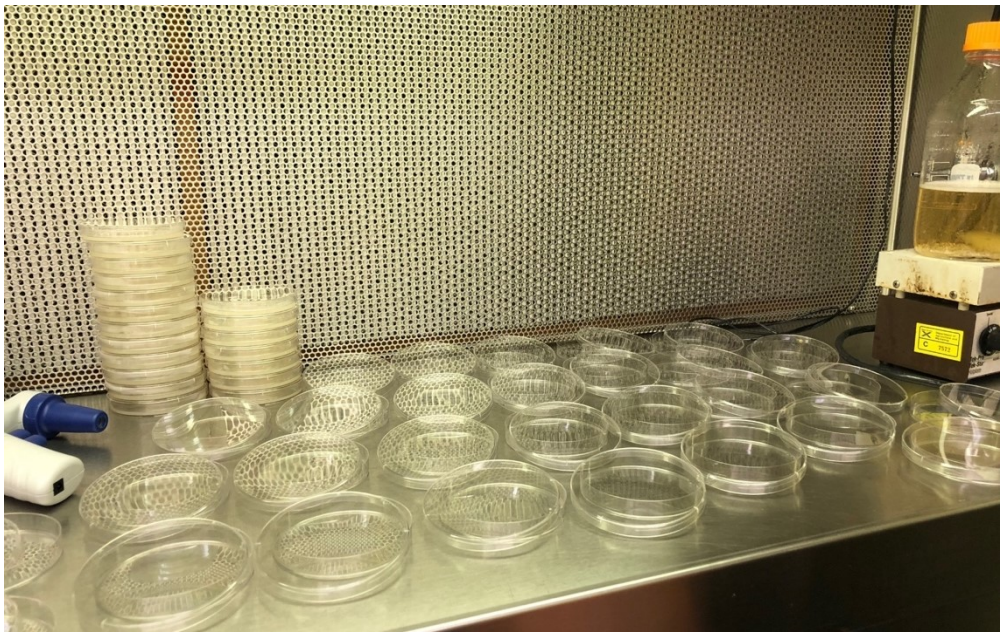

Figure 3.2 Preparation of petri dishes for decay fungi trial and agar plate trial using 4% malt agar.

3.2.2.2 Initiating Frozen Cultures

The fungi arrived from ATCC in test tubes, on dry ice. After arrival, the tubes were separated from the dry ice and left to warm for a few minutes. Then, under a laminar fume hood, the tubes were open, and the fungi were aseptically transferred from the tube to a petri dish with the proper agar (ATCC, 2013). This was done in replicates of five. The plates were placed in an incubator with a temperature of 25 °C. Once the fungi were established on the recommended agar, they were transferred to the 4 % malt agar for observation and continuous sub culturing at a temperature of 25 °C. After it was determined that the fungi were capable of survival on the malt extract agar, preparations for the experiment began.

3.2.3 Block Preparation

Rock Maple sapwood test blocks were milled so that they would fit in an averaged sized Petri dish. The block size was 50 x 25 x 7.5 mm ± 5 %, as seen in Figure 3.3. Section 3.1 outlined the treatment cycle used to treat the blocks. In each treatment cycle, 60 blocks were used. These test blocks were treated with preservative and then exposed to select fungi decay for a 16-week period. A group of control blocks were set aside, and the remaining test blocks were pressure treated with one of the following three preservative treatments: 100% Creosote; 70% Creosote, 30% Diluent; or 50% Creosote, 50% Diluent.

Figure 3.3: 50 x 25 x 7.5 mm Rock Maple wood block used for fungi trial against a 15 cm ruler.

The creosote was a P1/P3 Canadian Origin Creosote, with a lot batch number of 1682P1 provided by Ruetgers Canada Inc. located in Hamilton Ontario. Canadian Clean Fuels located in of Brampton Ontario provided a blend of B80 ULSD # 2 Clear which was a blend of 80% soybean biodiesel, and 20% diesel that had a ticket number of SJ006. Ten replicates were used for each treatment. After the wood was removed from the pilot plant, the spent preservative solution was drained and stored in a cold, dark environment and later analyzed on a headspace GC (Section 3.3.5). In order for the blocks to be used for the decay trial there were a series of preparation steps post preservative treatment that occurred.

The blocks were first placed in an oven for one week at a temperature of 60 °C. The purpose of this was to stabilize the moisture content. After the blocks were removed from the oven they were placed in a conditioning chamber with a temperature range of 20-30 °C ± 2 with a relative humidity of 25-75%. They remained in the conditioning chamber for a total of 21 days. After conditioning, the blocks were weighed and then placed in bags and then vacuum sealed shut. The package was sent to Nordion in Laval QC and sterilized by gamma rays at 29.7 kGy and 32.2 kGy on October 6, 2018. The Irradiation lot number was 727602.

Upon their return, the blocks were aseptically weighed and placed on the agar plate with the fungi cultures as seen in Figure 3.4. Once the petri dishes were labelled, they were placed in an incubator at 25 °C ± 1. In order to ensure that the culture transfer would be successful, they were given time to reach the size of a quarter before the wood blocks

were added to the dish. Since the fungi had different growth rates, they could not be transferred on the same day, the transfer period took 5 days. Once the wood blocks were placed in contact with the fungi, they were evaluated periodically, and observations were recorded.

Figure 3.4 Sterile environment where the wood blocks were weighed and aseptically transferred to petri dish with fungi.

3.2.4. Determination of Preservative Retention

Preservative retentions are outlined in the Standard U1-16 of the AWPA (2016). The minimum retention level for maple wood treated with creosote was 160 kg/m^3. The calculation to determine the preservative retention of the cubes was taken from AWPA Standard E10-16 and is as follows:

$$\text{Preservative Retention } \left(\frac{\text{kg}}{\text{m}^3}\right) = \left(\frac{\text{GC}}{\text{V}}\right) * 10$$

In this equation:

- G represents the amount of preservative absorbed by the sapwood block (final weight of the preservative treated cube minus the initial weight of the cube in grams)
- C represents the amount of preservative (in grams) in 100 g of treating solution
- V represents the volume of the cube in cubic centimeters

  *this is a correction factor used in conversion to the final unit of kg/m^3

3.2.5 Measuring Decay

At the end of the 16-week incubation period, the blocks were removed from the incubator, and all visible mycelia were carefully removed by scraping the fungal mat off the wood block with a razor blade, and then brushing it with a small brush. The blocks were then weighed, and placed in an oven for 7 days at a temperature of 60 °C. They

were placed in the oven to stabilize the moisture content. After they were removed from the oven, they were re-weighed and placed in a conditioning chamber for 21 days. After the 21-day period was complete, the blocks were removed and weighed for the final time. At all points during the experiment, the blocks were weighed to the nearest 0.01g. After the experiment was complete, the weight loss percent was calculated using an equation provided by the AWPA (2016) as seen below.

$$\text{Weight loss, percent} = \frac{100(T_3 - T_4)}{T_3}$$

In this equation:

- T_3 is the weight of the wood before it was placed on the agar with the fungi
- T_4 is the final weight of the wood block after conditioning or oven

3.2.6 Agar Study

An agar study was the second fungal experiment for testing the efficacy of creosote. The experiment was set up similar to that reported by Guillén et al. (2009). This experiment had the same preparation requirements as described in Section 3.2.2 except that wood was not involved. There were 5 replications per fungi. Blank paper disks were dipped in three different blends of creosote and placed on the agar plates equidistant from the fungi after they had grown to the size of a quarter. The plates underwent 16-week incubation period with a temperature of 25 °C. Once removed, observations were recorded. The

preservative solutions used included: 100% creosote; 70% creosote/ 30% diluent; and 50% creosote/ 50% creosote.

3.3 Odor Suppression

Quantification of naphthalene in the headspace of creosote is important for odor suppression analysis. The goal was to quantify naphthalene in the headspace of creosote and then compare it to the quantity of naphthalene in the headspace of a creosote/ diluent blend. Various methods were attempted to meet this goal. Hardwood blocks were prepared and treated in the pilot plant as stated in Section 3.1. The dimensions of wood samples were 25 cm x 15 cm x 15 cm as shown in Figure 3.5. Treatment solutions were prepared by mixing creosote and the diluent at various concentrations. The combinations of creosote and diluent were as follows:1) 100% Creosote, 2) 50% Creosote, 50% Diluent, and 3) 70% Creosote, 30% Diluent.

Figure 3.5: 25 cm x 15 cm x 15 cm block of wood used in pilot plant to simulate full scale production against 15 cm ruler.

3.3.1 Sorbent Tubes

The wood blocks were treated as described in Section 3.1. When the pilot plant was finished with the pressure stage, the treating solution in the pressure vessel was drained for 4 minutes to create a headspace. This headspace was used for air sample collection. The remaining solution went through a 30-minute expansion bath. After the expansion bath, a sorbent tube was attached to a valve on the top of the pressure vessel, and then the valve was opened and approximately 20 L of air was pumped through the tube using a Dräger pump. An example of a sorbent tube with a Dräger pump can be seen in Figure 3.6. Afterwards, the tubes were sealed and sent to Agat Laboratories in Dartmouth NS for the analysis of PAHs using NOISH 5515 (NOISH, 1994).

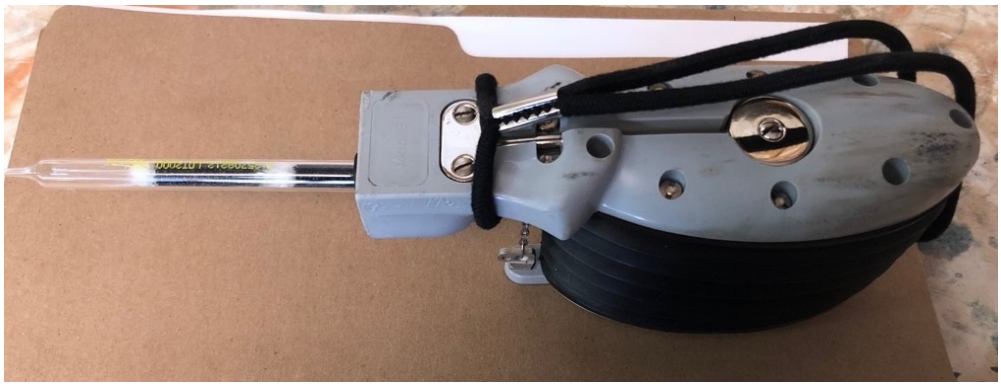

Figure 3.6: Charcoal Sorbent Tube attached to a Dräger Pump to collect air samples of creosote.

3.3.2 Summa Canister

Wood samples were treated in the pilot plant following the cycle described in Section 3.1. When the treatment was complete, the preservative was drained out of the pilot plant for 4 minutes in order to create a headspace for sampling. After the headspace was created the valves were closed and the creosote underwent a 30 min expansion bath. After the expansion bath a Summa Canister was attached to a valve on the top of the pressure cylinder using a Teflon tube. The Summa Canister was opened, and its vacuum drew gas into the canister. Then the Summa Canister was closed and detached from the pilot plant. When the preservative was drained from the pilot plant it was collected for liquid sample analysis. An example of the Summa Canisters used for this experiment is shown in Figure 3.7. The Summa Canisters and the liquid samples were sent to Agat Laboratories in Dartmouth NS and Maxxam Analytics in Bedford NS for analyzing naphthalene content using US Environmental Protection Agency (USEPA) method 5021A (USEPA, 2003).

Figure 3.7: Summa Canister used to collect creosote gas samples for PAH analysis.

In addition to sampling directly from the pilot plant, an indirect method was also implemented. After treatment the creosote/diluent was drained into a 10 L bucket and then a 10-minute expansion bath was applied. After the expansion bath an air sample was collected using a Summa Canister using the same method as above. The purpose for the transfer was to determine if there was enough headspace in the pilot plant when the sample was being collected. The decrease in expansion bath was due to the accelerated cooling rate of the preservative in the bucket compared to that in the pilot plant. It was more important to sample hot preservative as opposed to a 30 min expansion bath because of the volatility of the naphthalene being tested. The sample was collected using a Teflon tube and a valve as seen in Figure 3.8.

Figure 3.8: Valve attached to the lid of the 10 L bucket to collect samples using a Summa Canister.

3.3.3 Gas Detection Tubes

Isobutyl Acetate tubes provided by Sensidyne were used to collect air samples in the same manner as the sorbent tubes except that instead of being sent to an analytical laboratory to be analyzed, the concentration of naphthalene could be determined within minutes after sampling. Colorimetric tubes were used as directed by their user manual (Sensidyne, 2013). Figure 3.9 shows a Colorimetric tube used as well as the pump. Air samples were taken from both the pilot plant and the 10 L bucket as noted in Section 3.3.2. Colorimetric tubes were also used in a laboratory setting in order to determine their detection limit and sensitivity. In trial runs, 10 mL, 25 mL, 50 mL, and 100 mL volumetric flasks was used. 1 mL of creosote was added to the volumetric flask and left for 24 hours for the chemicals to reach equilibrium. After the 24-hour period, the colorimetric tubes were used to collect an air sample, and then read shortly after.

Figure 3.9: Colorimetric tube attached to pump to collect air samples of creosote.

3.3.4 Perkin Elmer Clarus 680 GC coupled to a Clarus SQ8 MS

A Perkin Elmer Clarus 680 Gas Chromatographer (GC) coupled to a Clarus SQ8 Mass Spectrometer (MS) was used for quantifying the amount of naphthalene in the headspace of creosote mixtures. The GC-MS had an Rxi-5 ms column (30 m × 0.25 mm × 0.25 um), in which case a low-polarity phase was used. The method used to analyze these samples was developed with assistance of a technician from Perkin Elmer. The GC injection port was operated at 250 °C in splitless mode, and 1 mL/min helium was used as a carrier gas. The injection volume was 1 μL. The initial oven temperature was set at 50 °C and held for 2 mins, then increased to 250 °C at a rate of 20 °C/min and held for 3 mins, giving a total run time of 15 mins. In terms of MS settings, an electron impact (EI) source with electron energy of 70 eV was used. The source and transfer line temperatures were 150 °C and 200 °C, respectively. Data were acquired in 45-400 m/z scan mode, and 3 mins solvent delay was applied to protect the MS.

Samples were prepared as follows: 10 mL of creosote was quantitatively transferred to a 20 mL vial and sealed. The vial was then placed in a hot water bath at a set temperature of 90 °C. The sample sat in the bath for 30 min and then a gas tight syringe was used to collect 1mL of headspace from the 20 mL vial. After the gas sample was collected, it was inserted in a 1.5 mL vial containing 0.5 mL of acetone. The tip of the syringe was placed at the bottom of the acetone and the gas was bubbled through. The acetone samples were

then analyzed using the GC-MS method noted above. The concentration of naphthalene was determined against an established calibration curve.

3.3.5 Bruker Scion 456-GC configured for Headspace Analysis

A Bruker Scion 456-GC configured for headspace analysis with a TQ Mass Spec was used for the equilibrium headspace analysis of creosote and diluent blends. The GC used can be seen in Figure 3.10. The GC had a Bruker BR-5 ms FS, that was 60 meters with a 0.25mm ID and a 0.25 µm df column. A combiPAL autosampler was used where the injection mode was GC Headspace with a 1 mL heated syringe. The syringe temperature was 70.0 °C and the agitator temperature was 80.0 °C with a speed of 500 rpm. It had a cycle of 5 seconds on and 2 seconds off. The incubation time of the sample was 45 minutes, the plunger fill speed was 500 µL/sec and the plunger inject speed was 500 µL/sec with a post injection time of 0.5 sec and a syringe flush time of 10 sec.

The total run time of the GC was 1 hour, 2 minutes and 30 seconds. The injection volume was 300 µL with a source temperature and a transfer line temperature of 300 °C. The front-end injector had a temperature of 200 °C with a hold time of 45 min. The total time was 45 min. The middle injector had a pressure of 10 psi, a hold time of 20min and a total time of 20min. Finally, the column oven was 50°C with a stabilization time of 0.5 min.

When the GC had a temperature of 75.0 °C the rate time was 0.0 °C /min with a hold time of 2.00 min. When it was 200.0°C the rate was 10°C /min with a hold time of 0.00

min and a total time of 14.50 min. When the temperature was 295.0 °C the rate was 2°C /min with a hold of 0.00 min and a total time of 62.00 min. When the GC temperature was 320.0°C, it had a rate of 50°C /min and a hold of 0.00 with a total run time of 62.50 min. The Bruker Scion 456-GC configured for headspace can be seen in Figure 3.10.

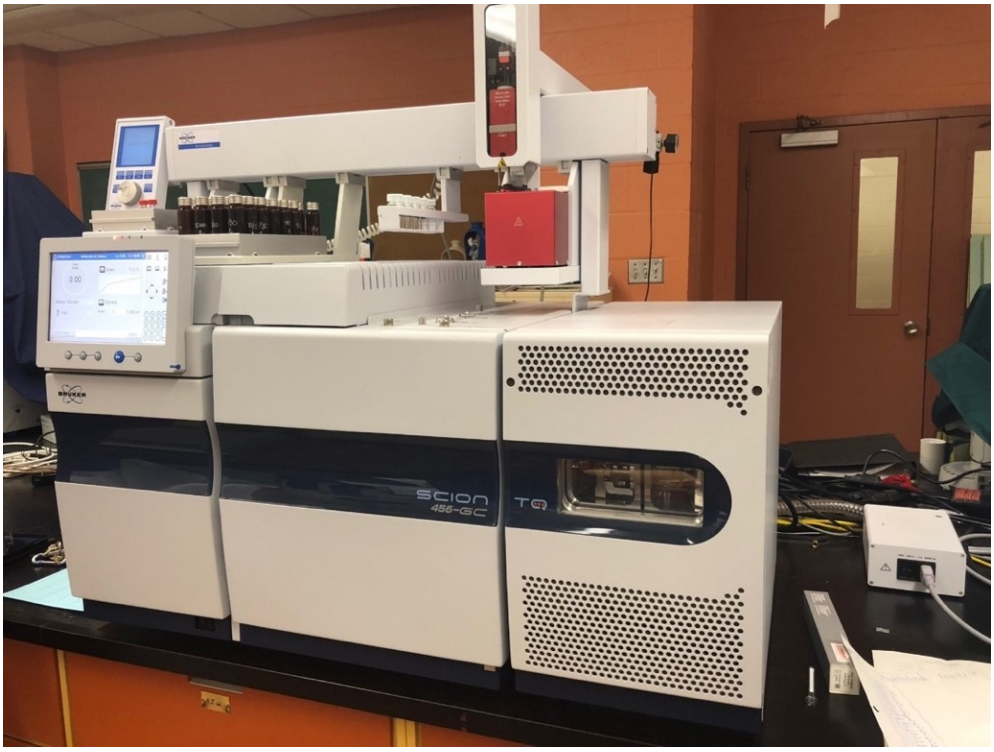

Figure 3.10: Bruker Scion 456-GC preparing to sample naphthalene concentrations in headspace.

3.3.5.1 Standard Curve Development

Analysis of naphthalene levels in various creosote blends was completed using a modification to the method 5021A Volatile Organic Compounds in various sample matrices using equilibrium headspace analysis (USEPA, 2003). To prepare the standard calibration curve, solid naphthalene of analytical grade (99%) was purchased from Sigma-Aldrich as well as HPLC grade P-xylene. To make a stock solution, 0.0500 g of naphthalene was added to a 50 mL amber volumetric flask followed by adding 50 mL of p-xylene. In order to confirm that the solution was properly dissolved, twenty-five inversions were completed. The stock solution was labeled and placed in a fume hood. Through experimentation it was determined that a suitable naphthalene concentration range was between 10 mg/L and 60 mg/L. Three standard curves were made with the following mg/L concentrations: 10 mg/L, 20 mg/L, 30 mg/L, 40 mg/L, 50 mg/L, and 60 mg/L; where the naphthalene concentrations were plotted on the X axis and the peak areas were plotted on the Y axis. The results showed that a logarithmic equation was suitable for the curve. The finalized average curve can be observed in Figure 3.11 below.

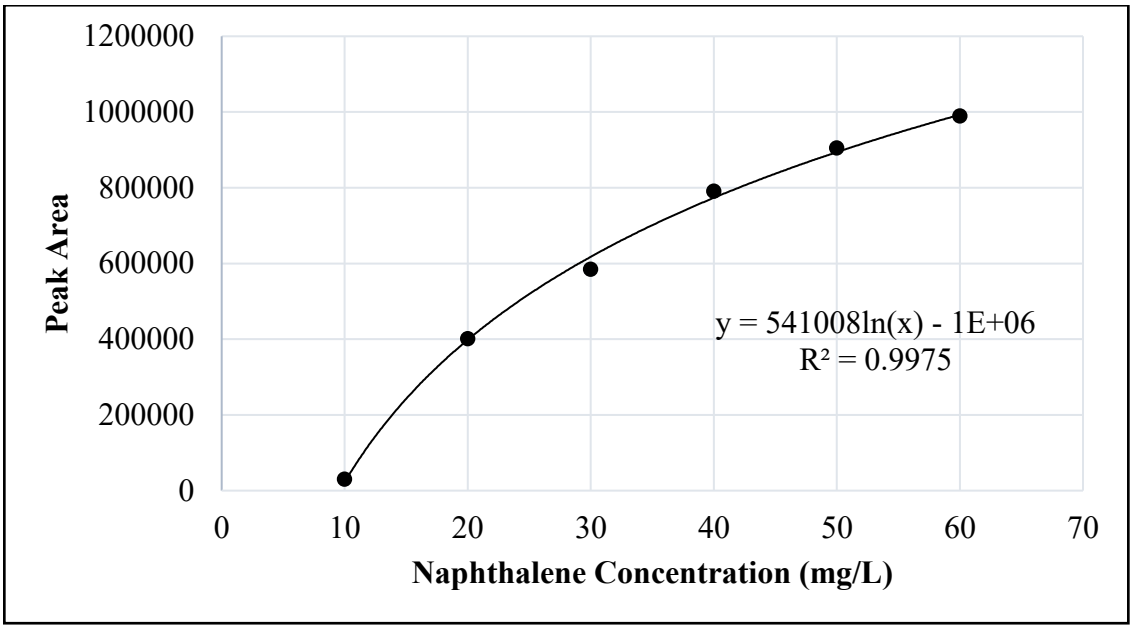

Figure 3.11: Standard Curve of Peak Area versus Naphthalene Concentrations (mg/L) where; $R^2 = 0.9975$ and the equation y = 541008 ln(x) – 1E + 06.

Based on Figure 3.11, 10 mg/L - 60 mg/L naphthalene concentration was an acceptable range for a standard curve. The equation for the curve was y = 541008ln(x) -1E+06. The R^2 value was 0.9975. It is important to recognize that these values represent the headspace of a liquid sample with a known concentration of naphthalene. The naphthalene that is in the headspace generates a GC-MS response as a peak area count. In this case, the naphthalene in the headspace is being quantified based on a known quantity in the p-xylene. Since a gas sample of naphthalene was not used to create the standard curve the exact concentration of naphthalene in gaseous phase is unknown.

3.3.5.2 Sample Preparation

After the standard curve was finalized, a series of steps were used to assure that the samples prepared would minimize variability and maximize accuracy. In order for samples to fit on the standard curve they had to go through a dilution series. To begin 2.5 grams of creosote solution were weighed and transferred into a 25-mL volumetric flask. P-xylene was added to the flask to bring it to a volume of 25 mL. The solution was mixed and then 2.5 mL of creosote/p-xylene was transferred to another volumetric flask where P-xylene was added to bring the flask to volume, and the cycle repeated. A diagram of this process is illustrated in Figure 3.12. After doing various trials on the GC, a 2000-fold dilution factor was chosen to be used for all samples, allowing all samples to fall in the concentration range of the standard curve. An example of the dilution series used can be observed in Figure 3.13.

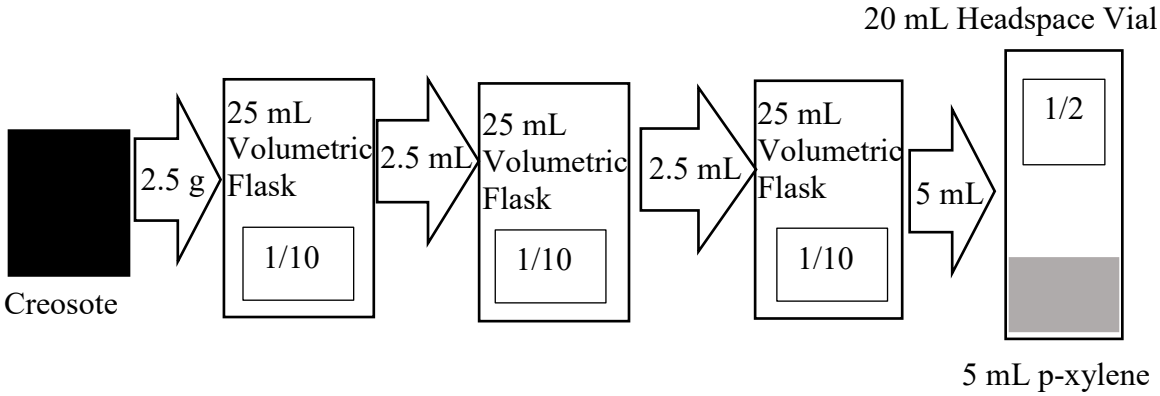

Figure 3.12: Diagram of series dilution used to properly dilute the sample with a dilution factor of 2000. Total dilution = 1/10 *1/10 * 1/10 * 1/2 = 1/2000.

3.3.5.3 Sample Analysis

Due to the variability observed while preparing the standard curve, it was anticipated that the same variability would occur when the samples were analyzed. Therefore, seven samples of each creosote blend were prepared using p-xylene. These blends included 50% creosote, 70% creosote, and 100% creosote. Five samples of pure diluent (80% biodiesel + 20% diesel) were also made using the same dilution factor of 2000. This was done to ensure that there was no naphthalene in the diluent blend. Pure p-xylene was also analyzed using the GC, as well as six standards, 2 x 10 mg/L, 2 x 40 mg/L, and 2 x 60 mg/L. The purpose of running standards along with the liquid samples was to ensure that the standard curve was accurate, as they acted as a reference point.

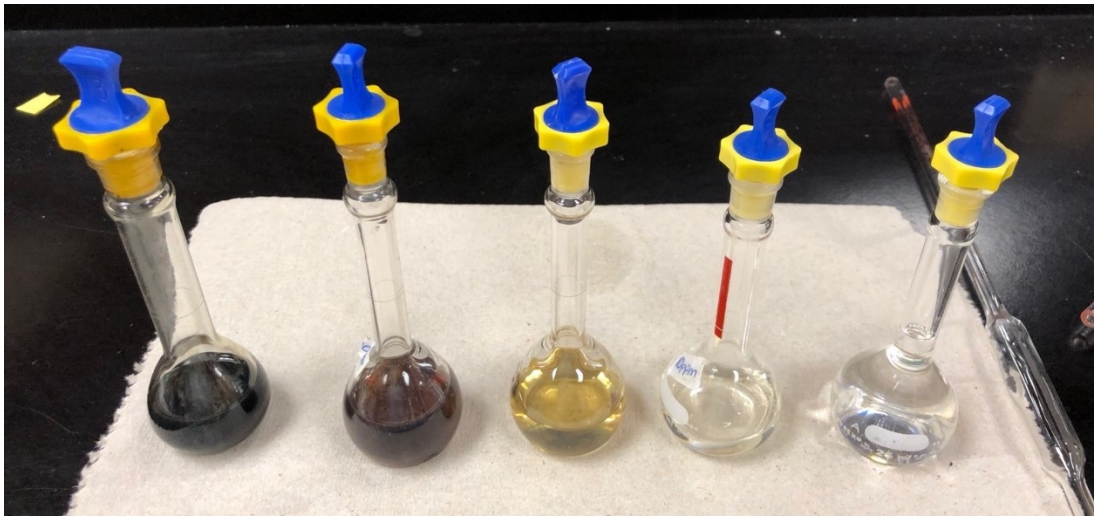

Figure 3.13: Example of dilution series where creosote/diluent is diluted with p-xylene.

3.4 Leaching Analysis

The purpose of the leaching experiment was to determine the amount of leaching associated with a given amount of rainfall in a controlled environment. Rainfall simulation is important for wood preservation because it gives an indication how much preservative would leach from the wood into the environment. It is also important when testing a new preservative to ensure that it is beneficial for long term use. Figure 3.14 is an example of the rail ties used for the leaching analysis.

Figure 3.14: Rail ties used for rainfall simulation before they were cut to dimensions 100.3 cm x 15.24 cm x 20.32 cm.

3.4.1 Wood Preparation for Leaching

The size of a standard rail tie is 243.84 cm x 15.24 cm x 20.32 cm. However, for the rail tie to fit in the rainfall simulator for the leaching experiment, the tie was cut to dimensions of 100.3cm x 15.24cm x 20.32cm. Figure 3.15 is an outline of the rail ties used for the experiment. The shaded boxes indicate sides that were omitted in rainfall coverage calculation.

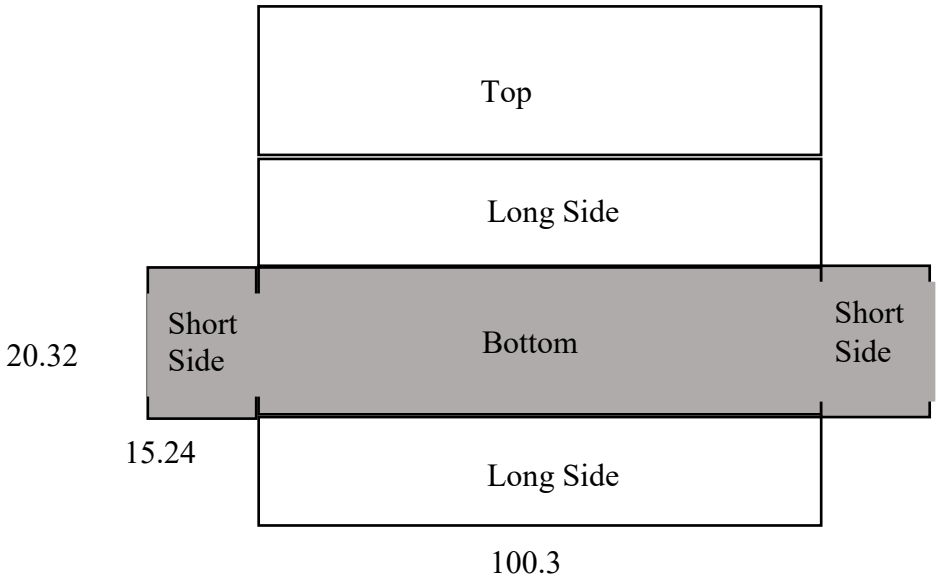

Figure 3.15: Box design of rail tie with dimensions 100.3cm x 20.32cm x 15.24 cm after being cut for the experiment.

To determine the total area exposed to rainfall, each exposed side of the wood was calculated and then added together. The surface area of the top or bottom was 2038.10 cm^2, the long side was 1528.5 cm^2 and the short side was 309.67 cm^2. Since the rainfall did not reach the bottom of the rail tie, it was not included in the calculation. The short sides were also omitted because they were both sealed with Paraffin Wax, so it was impossible for creosote to leach from them. Therefore, the total area that the creosote leached from the wood was from the top and both long sides, in which case the total surface area exposed to water was 5094.5 cm^2 = 0.50945 m^2.

Full size rail ties were treated with a 70% creosote, 30% diluent blend in an industrial scale plant. Full sized ties were used to best replicate real-world service conditions as seen in Figure 3.14. They were treated in a full-scale cylinder along with ties that were being treated for sale. After the ties were treated, they were left in a dry room until they were cut to the dimensions 100.3 cm x 15.24 cm x 20.32 cm, so they would properly fit in the rainfall simulators. After the ties were cut, the ends were sealed with Paraffin Wax as seen in Figure 3.16 to prevent leaching from the cross-section that was cut.

Figure 3.16: Creosote/ diluent treated rail tie after it was measured, cut, and then sealed with Paraffin Wax.

3.4.2 Outline of Rainfall Mechanism

The rainfall simulators used were constructed out of two plastic animal feed troughs, a pump and nozzles for dispersing water over the wood sample as seen in Figure 3.17. This allows for several years of environmental exposure to be re-created in a short period of time. In addition, the use of a closed loop system prevented water contamination. There were eight rainfall simulations completed using six rainfall simulators. The rainfall simulators that were reused were cleaned between trials.

Figure 3.17: Rainfall simulator where the rail tie is sitting above the pool of water. In which case the water is being pumped through the PVC pipes and raining onto the wood sample.

The method used was that reported by Lebow et al. (2003). It was used for analyzing the leaching of CCA treated wood, however creosote was the preservative tested in this study. In the experiment, Lebow et al. (2003) simulated rainfall for 9 hours a day, for a total of 5 days with a flow rate of 3.0 mm/hour. In the present experiment, the wood was exposed for 9 hours a day, for 5 days, with a flow rate of 4.3 L/min. A faster flow rate was more appropriate as it simulated how the preservative would react after being exposed to a significant amount of rainfall within a short period of time.

To obtain wood cores, a hollowed-out drill bit was drilled into the rail tie, and then the sample was removed using an extractor. Both water samples and wood samples were

collected and sent to Maxxam Analytics in Bedford NS to be analyzed for semi-volatile organics by GC-MS using EPA Method 8270D (USEPA, 2014). They were also analyzed for hydrocarbons using Atlantic RBCA Hydrocarbons (RBCA, 2016).

3.5 Statistical Design and Analysis of Experiments

In all of the statistical analysis the statistical software used was SAS 9.4 and Minitab 2017. Each experiment used a different statistical analysis, however a 95% confidence interval ($\alpha = 0.05$) was consistent.

The fungal decay experiment in Section 3.2 was analyzed as a 2-factor factorial. The factors were the preservative concentration at four levels and the species of fungi at five levels. In addition, concentration of creosote was also compared through multiple means comparison using Tukey's test. Normality and constant variance were met through the Central Limit Theorem and independence was met through the design of the experiment. The percent weight loss of the blocks was calculated after the blocks were removed from the oven and again, after they were removed from the conditioning chamber. Statistical analysis was completed for each percent weight loss calculated.

The headspace analysis presented in Section 3.3 was analyzed using multiple means comparison with Tukey's test. Normality and constant variance were both met therefore the Central Limit theorem was not necessary. No transformations were required.

The Leaching analysis presented in Section 3.4 was analysed used a two-sample-t test in Minitab. Since wood cores were taken before and after the experiment a two-sample t test was completed to determine if the data were significantly different. Fifteen PAH's were compared as well as total petroleum hydrocarbons (TPH's) to determine if there was a significant difference in the concentrations before and after leaching.

Chapter 4.0 Results and Discussion

The goal of the present study was to determine whether adding a diluent to creosote would affect its properties, specifically regarding odour suppression, fungal control and leaching behavior. When the term diluent is used hereon, it is referring to a solution of 80% soybean biodiesel and 20% diesel. When a percent creosote value is given, it refers to the amount of creosote present and the balance would be the diluent. For example, 70% creosote refers to a system containing 30% diluent (as defined above) and 70% creosote.

The three main objectives will correspond to the Experimental Material and Methods presented in Chapter 3.0. The results of the first objective, which was to determine whether there would be a significant difference in the efficacy of creosote when the diluent was added; was addressed using two fungal studies. The first was a wood block decay experiment where fungal decay of wood treated with creosote was measured. The other was an agar study which did not employ wood. The experimental results are presented in Section 4.1. The study addressing the second objective is in Section 4.2, which determines if the diluent would significantly reduce the quantity of naphthalene available in the headspace of creosote. Finally, the study targeting the third objective presented in Section 4.3 has quantified the amount of leaching that occurred with wood treated with creosote and diluent.

4.1 Fungal Decay Test

This experiment was designed to determine whether or not a biodiesel/diesel diluent would affect the efficacy of creosote in preventing fungal decay. Three brown rot decay fungi and two white rot decay fungi were selected to compare the efficacy of various concentrations of creosote and diluent. The treatments included a control without any preservatives added, 50% creosote/50% diluent, 70% creosote/ 30% diluent, and 100% creosote. The brown rot decay fungi were *Postia placenta*, *Neolentinus lepideus*, and *Gloephyllum trabeum*. The white rot decay fungi chosen were *Stereum hirsutum* and *Trametes versicolor*. The wood used was Rock Maple since the majority of rail ties used across North America are made from hardwood. The method used was modified after E10-16 of the AWPA (2016) Laboratory method for Evaluating the Decay Resistance of Wood-Based Materials Against Pure Basidiomyete Cultures: Soils/ Block test; and British Standard EN 113 Test Method for Determining the Protective Effectiveness Against Wood Destroying Basidiomycetes (1997).

Data collected were the percent loss of wood mass after it had been in contact with the decay fungi for a 16-week incubation period. Mass of the wood samples was measured twice, the first after oven noted as post oven drying and the second after conditioning noted as post conditioning.

4.1.1 Measuring Decay: Post Oven

The first of the measuring decay analysis was after the wood blocks were removed from the oven. At this point the experiment was complete, and the blocks were placed in the oven to dry post the 16-week incubation period. A two-factor factorial was employed to determine whether the diluent in the creosote or the fungal species had a significant effect on the weight loss. A Least Means (LS) test was conducted on the data to determine which treatments were significantly different from the control. The LS means do not represent the true mean, for the true mean, refer to the descriptive statistics. The true mean is designed for use when the design is balanced, however, when the design is unbalanced, the LS means are required. The LS means are adjusted for other effects in the model and estimate the means for a balanced population (SAS, 2008). Following the two-factor factorial, Tukey's test was also completed within each fungal species to determine if there was a significant difference based upon mean percent weight loss of the wood. All statistics were completed where $\alpha = 0.05$, with a confidence interval of 95%. The descriptive statistics of the post oven analysis as well as the two-factor factorial and the multiple means comparison are listed in Table 4.1 to 4.3.

Table 4.1. Descriptive statistics of weight loss of wood blocks; post oven drying.

Fungi Species *	Creosote Concentration (%)	Weight (%)	Standard Deviation	Sample Size
GT	0	13.56	4.64	10
GT	50	6.03	0.69	10
GT	70	6.18	0.23	10
GT	100	6.18	0.48	10
NL	0	10.93	1.76	10
NL	50	6.61	0.58	10
NL	70	6.09	0.77	10
NL	100	6.14	0.39	10
PP	0	9.97	3.57	10
PP	50	6.01	0.23	10
PP	70	6.30	3.05	8
PP	100	6.25	0.31	10
SH	0	5.72	0.71	9
SH	50	6.33	1.04	10
SH	70	6.19	0.26	10
SH	100	5.94	1.08	9
TV	0	19.11	4.50	10
TV	50	7.35	1.50	9
TV	70	7.29	0.76	10
TV	100	7.06	0.45	10

*Fungi Abbreviations are as follows: *Postia placenta* (PP), *Neolentinus lepideus* (NL), and *Gloephyllum trabeum* (GT). The white rot decay fungi chosen were *Stereum hirsutum* (SH) and *Trametes versicolor* (TV).

Table 4.2. Two-factor factorial analysis on weight loss of wood block post oven drying where the factors are fungi species and creosote concentration.

Fungi Species*	Creosote Concentration (%)	Weight Loss (%) LS Means **	LS Means Multiple Means Comparison ***
GT	0	13.55	B
GT	50	6.03	E
GT	70	5.99	E
GT	100	5.86	E
NL	0	10.93	BC
NL	50	6.47	E
NL	70	5.95	E
NL	100	6.14	E
PP	0	9.97	CD
PP	50	5.94	E
PP	70	5.73	E
PP	100	6.25	E
SH	0	5.61	E
SH	50	6.06	E
SH	70	6.15	E
SH	100	5.73	E
TV	0	19.10	A
TV	50	5.93	E
TV	70	6.81	E
TV	100	7.06	DE

*Fungi Abbreviations are as follows: *Postia placenta* (PP), *Neolentinus lepideus* (NL), and *Gloephyllum trabeum* (GT). The white rot decay fungi chosen were *Stereum hirsutum* (SH) and *Trametes versicolor* (TV).

**LS Means is not the same as the true mean, for true mean, refer to the descriptive statistics

***Data with the same letter are not significantly different at $\alpha = 0.05$.

Table 4.3. Multiple means comparison with Tukey's test of wood block weights post oven. *

Fungi Species**	Creosote Concentration (%)	Sample Size	Tukey's Test ***
GT	0	9	A
GT	50	10	B
GT	70	10	B
GT	100	10	B
NL	0	10	A
NL	50	10	B
NL	70	9	B
NL	100	10	B
PP	0	10	A
PP	50	10	B
PP	70	9	B
PP	100	10	B
SH	0	9	A
SH	50	10	A
SH	70	10	A
SH	100	10	A
TV	0	10	A
TV	50	10	B
TV	70	10	B
TV	100	10	B

*Each fungus was tested separately

**Fungi Abbreviations are as follows: *Postia placenta* (PP), *Neolentinus lepideus* (NL), and *Gloephyllum trabeum* (GT). The white rot decay fungi chosen were *Stereum hirsutum* (SH) and *Trametes versicolor* (TV).

***Data with the same letter are not significantly different at $\alpha = 0.05$.

The two-factor-factorial indicated that the species of fungi nor the concentration of creosote had a significant effect on the results except for the control blocks which were expected to be significantly different from the others. Tukey's test confirmed that there was no significant difference in the weight loss of wood blocks treated with 100% creosote, 70% creosote or 50% creosote as shown in Table 4.3. Based upon the results of the Post Oven fungal decay analysis, it can be concluded that the diluent did not have a significant effect on the efficacy of the creosote to prevent decay of the wood blocks.

4.1.2 Measuring Decay: Post Conditioning

Both the AWPA (2016) and the British Standards (1997) indicate that the blocks be weighed after the conditioning period. To compare the results of the conditioning period to the results of the post oven analysis, a two-factor factorial was employed to determine whether the diluent in the creosote or the fungal species had a significant effect on the weight loss of wood samples. A Least Means (LS) test was conducted to determine which treatments were significantly different from the control. Once again, the least mean does not represent the mean of the data, for the true mean refer to descriptive statistics. Following the two-factor factorial, a multiple means comparison using Tukey's test was also conducted within each fungal species tested to determine if there was a significant difference based upon mean percent weight loss of the wood. All statistics were completed where $\alpha = 0.05$. The descriptive statistics of the post conditioning analysis as well as the two-factor factorial and the multiple means comparison can be observed in Table 4.4-4.6.

Table 4.4. Descriptive statistics of weight loss of wood blocks; post conditioning.

Fungi Species *	Creosote Concentration (%)	Weight (%)	Standard Deviation	Sample Size
GT	0	12.13	4.67	10
GT	50	5.86	0.73	10
GT	70	5.99	0.21	10
GT	100	5.96	0.41	10
NL	0	9.71	1.79	10
NL	50	6.42	0.47	10
NL	70	5.95	0.88	10
NL	100	6.05	0.305	10
PP	0	7.33	3.57	10
PP	50	5.94	0.27	10
PP	70	6.28	2.99	8
PP	100	6.27	0.19	10
SH	0	4.35	1.01	9
SH	50	6.029	0.19	10
SH	70	6.15	0.34	10
SH	100	6.07	1.04	9
TV	0	17.27	4.15	10
TV	50	6.90	1.45	9
TV	70	6.81	0.61	10
TV	100	6.75	0.43	10

*Fungi Abbreviations are as follows: *Postia placenta* (PP), *Neolentinus lepideus* (NL), and *Gloephyllum trabeum* (GT). The white rot decay fungi chosen were *Stereum hirsutum* (SH) and *Trametes versicolor* (TV).

Table 4.5. Two-factor factorial analysis on weight loss of wood blocks post conditioning where the factors are fungi species and creosote concentration.

Fungi Species*	Creosote Concentration (%)	Weight Loss (%) LS Means **	LS Means Multiple Means Comparison ***
GT	0	12.12	B
GT	50	5.86	D
GT	70	5.99	D
GT	100	5.97	D
NL	0	9.71	BC
NL	50	6.47	D
NL	70	5.95	D
NL	100	6.05	D
PP	0	7.11	CD
PP	50	6.005	D
PP	70	6.36	D
PP	100	6.27	D
SH	0	4.23	D
SH	50	6.06	D
SH	70	6.12	D
SH	100	6.07	D
TV	0	17.26	A
TV	50	5.93	D
TV	70	6.81	D
TV	100	6.74	D

*Fungi Abbreviations are as follows: *Postia placenta* (PP), *Neolentinus lepideus* (NL), and *Gloephyllum trabeum* (GT). The white rot decay fungi chosen were *Stereum hirsutum* (SH) and *Trametes versicolor* (TV).

**LS Means is not the same as the true mean, for true mean, refer to the descriptive statistics

***Data with the same letter are not significantly different at $\alpha = 0.05$.

Table 4.6. Multiple means comparison with Tukey's test of wood block weights post conditioning. *

Fungi Species **	Creosote Concentration (%)	Sample Size	Tukey's Test***
GT	0	10	A
GT	50	10	B
GT	70	10	B
GT	100	10	B
NL	0	10	A
NL	50	10	B
NL	70	9	B
NL	100	10	B
PP	0	9	A
PP	50	10	A
PP	70	8	A
PP	100	10	A
SH	0	8	A
SH	50	10	A
SH	70	10	A
SH	100	9	A
TV	0	10	A
TV	50	8	B
TV	70	10	B
TV	100	10	B

*Each fungus was tested separately

**Fungi Abbreviations are as follows: *Postia placenta* (PP), *Neolentinus lepideus* (NL), and *Gloephyllum trabeum* (GT). The white rot decay fungi chosen were *Stereum hirsutum* (SH) and *Trametes versicolor* (TV).

***Data with the same letter are not significantly different at $\alpha = 0.05$.

There was a significant difference in weight loss between control blocks and the creosote treated blocks in the two-factor factorial analysis where $\alpha = 0.05$. However, there was no significant difference in weight loss among blocks treated with 100% creosote, 70% creosote, or 50% creosote. This was also confirmed through Tukey's test. Based upon the results of the post conditioning fundal decay analysis, the diluent used did not have a significant effect on the efficacy of creosote to prevent decay of the wood blocks.

4.1.3 Agar Study

The test fungi were also exposed to creosote treatments using agar plates, without wood present. The purpose of this experiment was to observe the interactions between the fungi and the preservative blends. The plates were incubated for a 16-week period similar to the wood blocks and then removed for observation. Since it is difficult to observe the zones of inhibition and the equal growth throughout, Figure 4.1 is an example of the zone of inhibition as well as what is meant when the fungi are said to have grown equally across the plate. In Figure 4.1 the zone of inhibition is very visible for the 70% creosote disc which is referring to the small circle around the creosote dipped disc. There is also equal fungi growth throughout the plate evident with some concentrated growth around the 50% creosote and 50% diluent. The fungi in Figure 4.1 is *N. lepideus* and the growth is not surprising because it is known as a creosote resistant or tolerant fungus. Figure 4.2 to Figure 4.6 represent the fungi plates examined in this observational analysis.

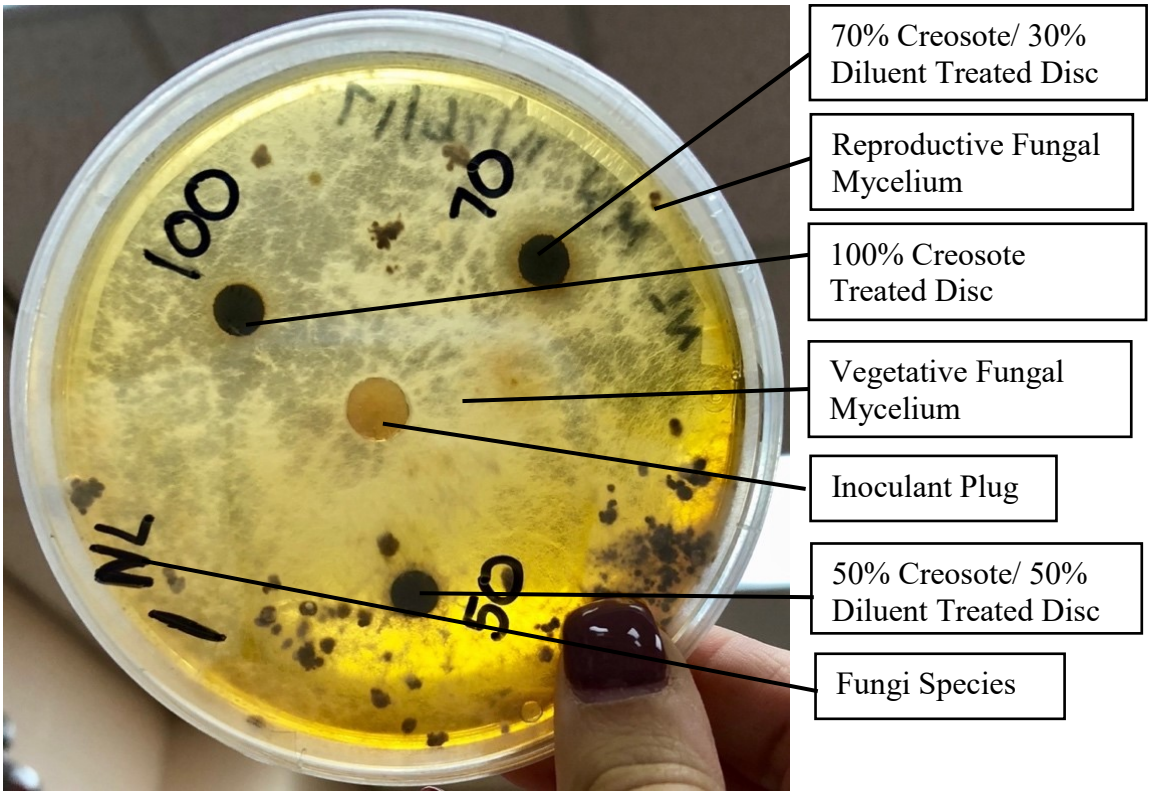

Figure 4.1: Close-up image of the fungus *N. lepideus* to fully explain the terms used in the description of the plates.

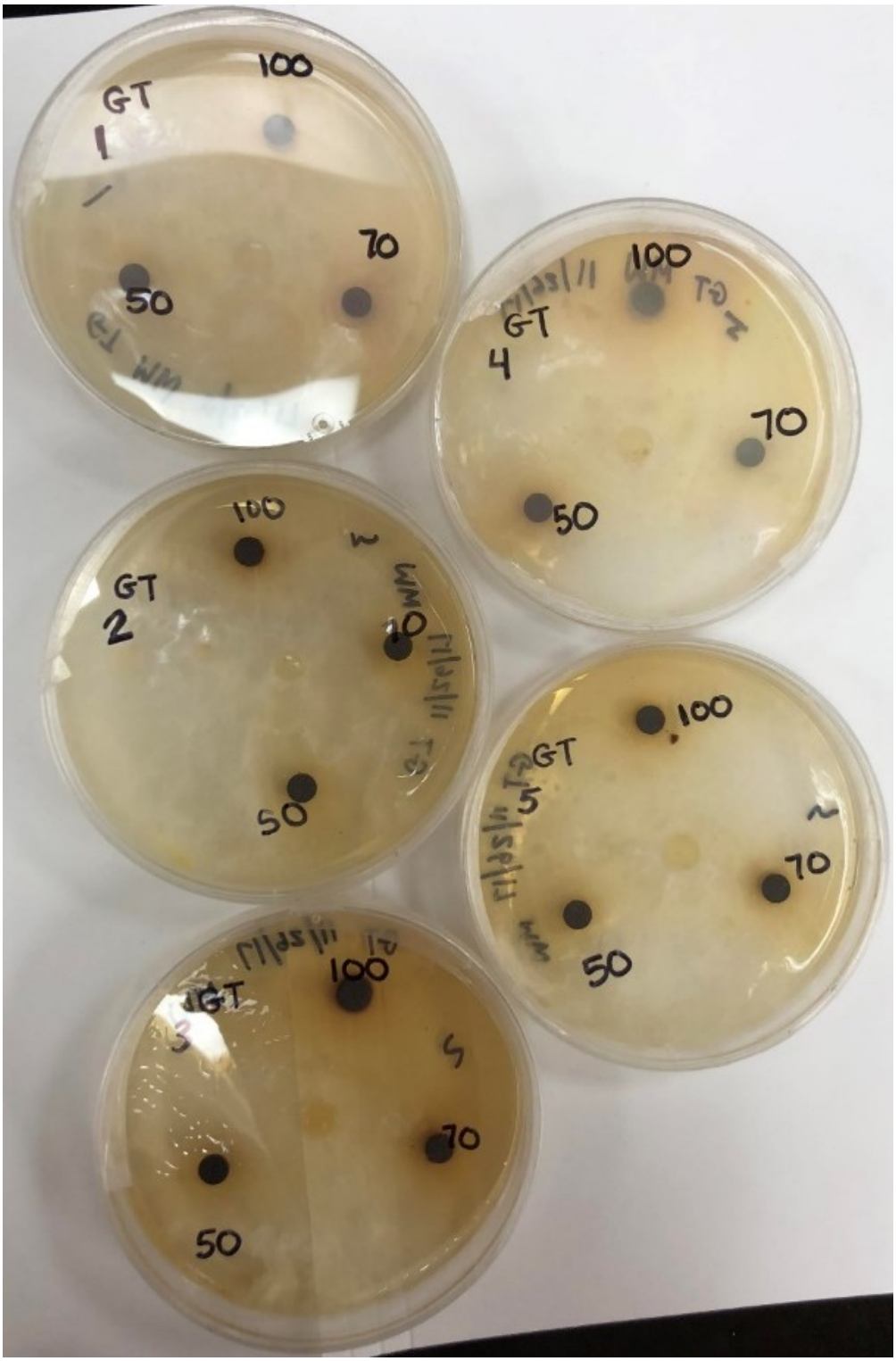

Figure 4.2: Blank Discs with 100%, 70% and 50% creosote discs on agar with *G. trabeum* in center.

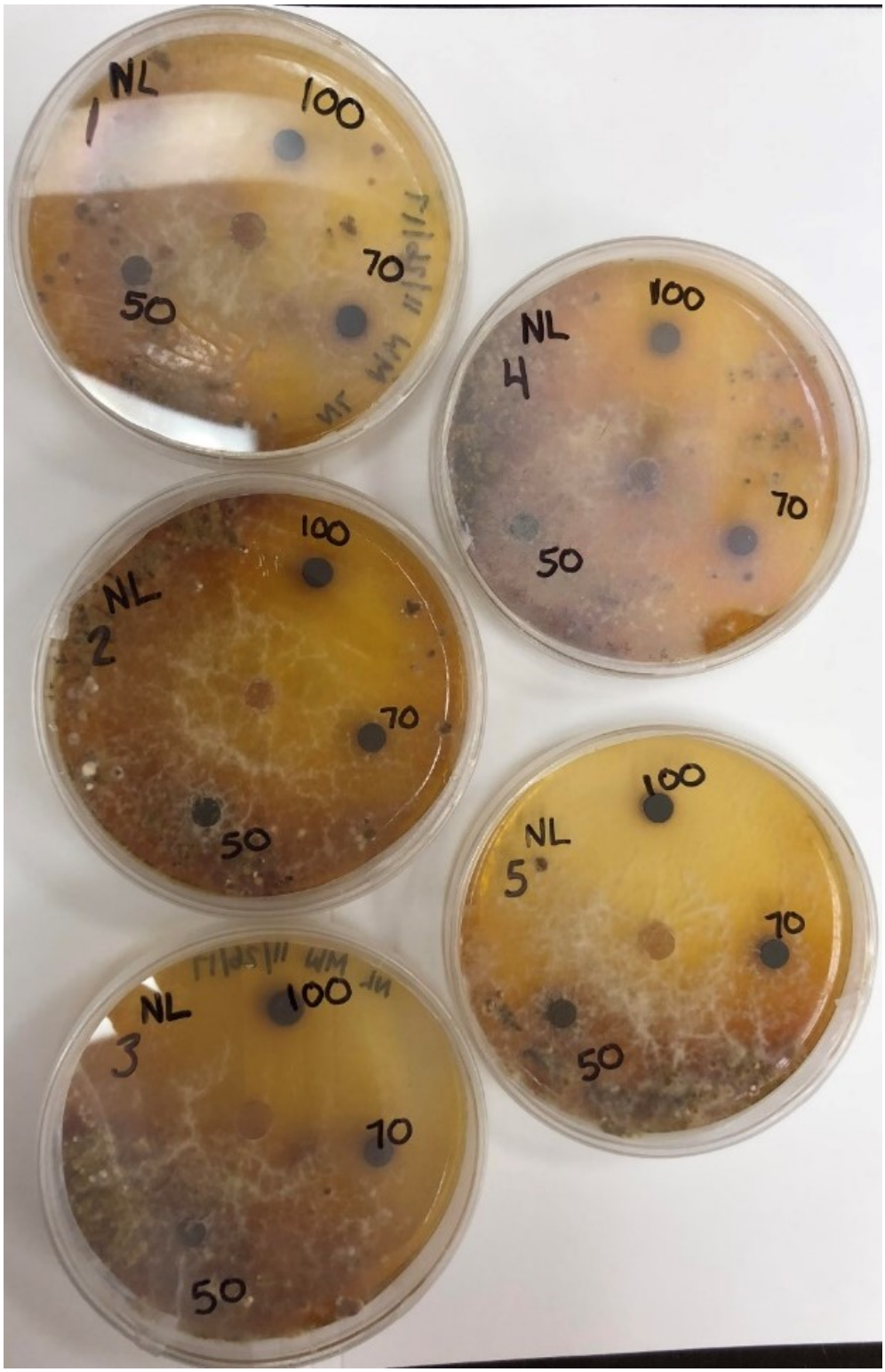

Figure 4.3: Blank Discs with 100%, 70% and 50% creosote discs on agar with *N. lepideus* in center.

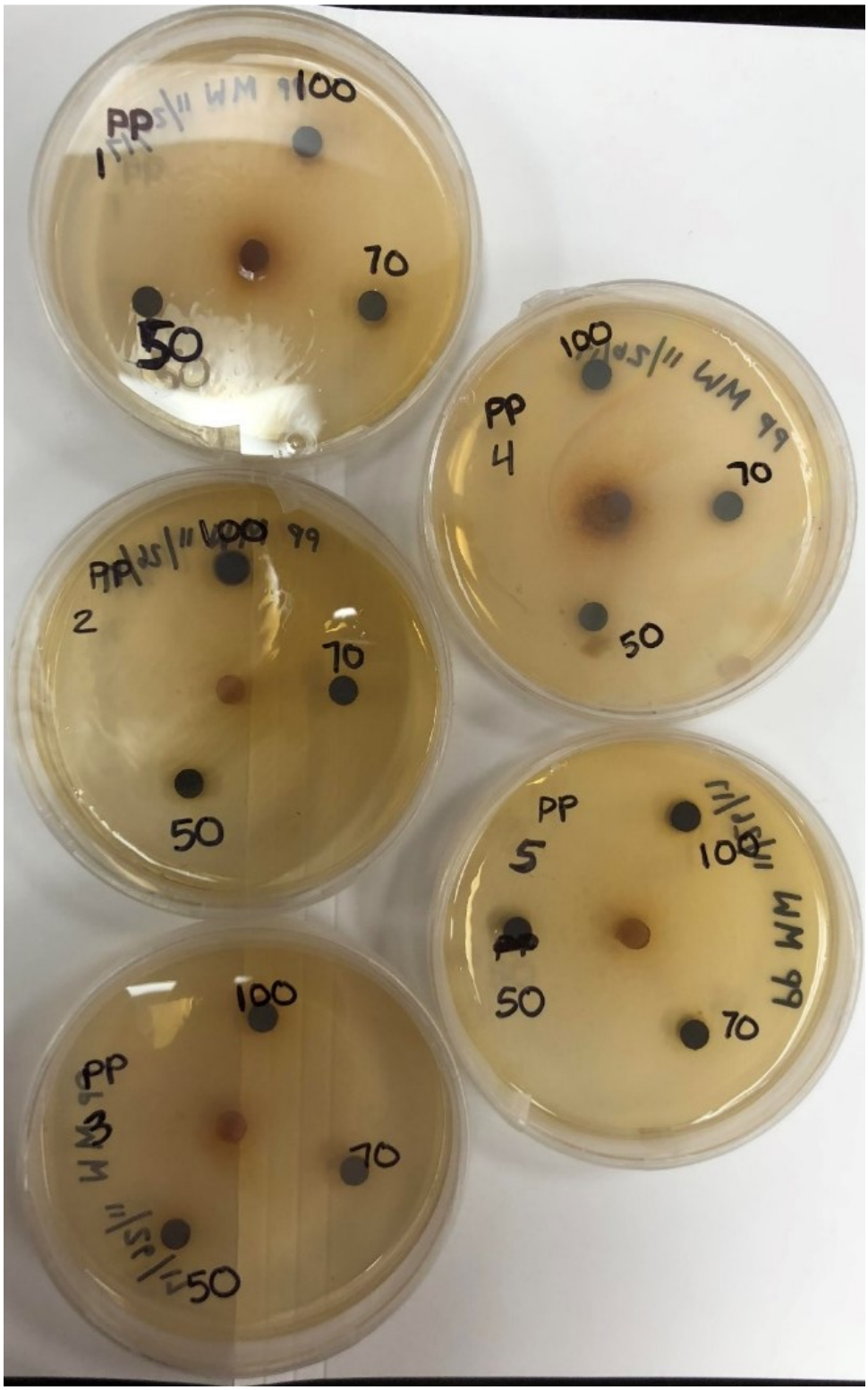

Figure 4.4: Blank Discs with 100%, 70% and 50% creosote discs on agar with *P. placenta* in center.

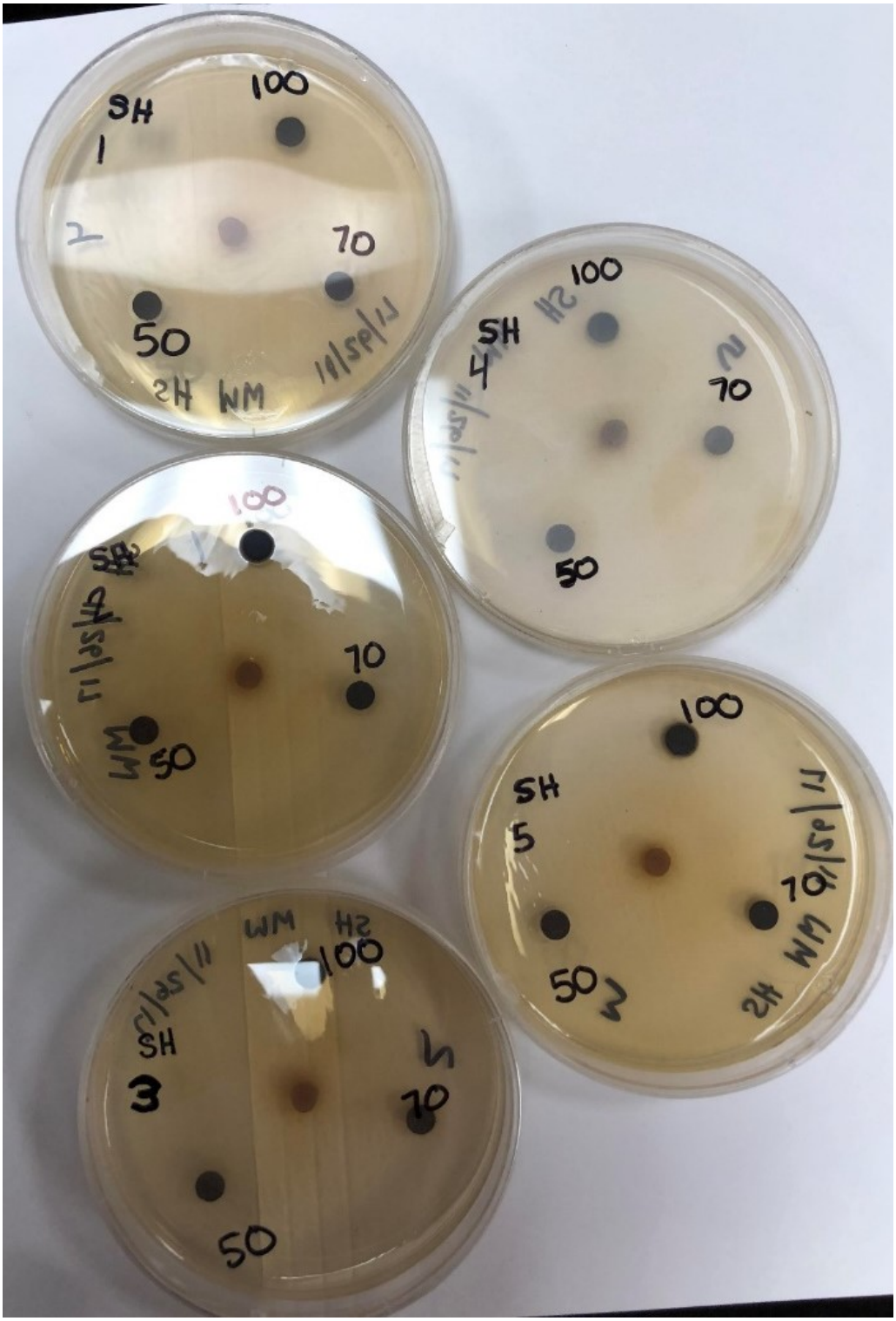

Figure 4.5: Blank Discs with 100%, 70% and 50% creosote discs on agar with *S. hirsutum* in center.

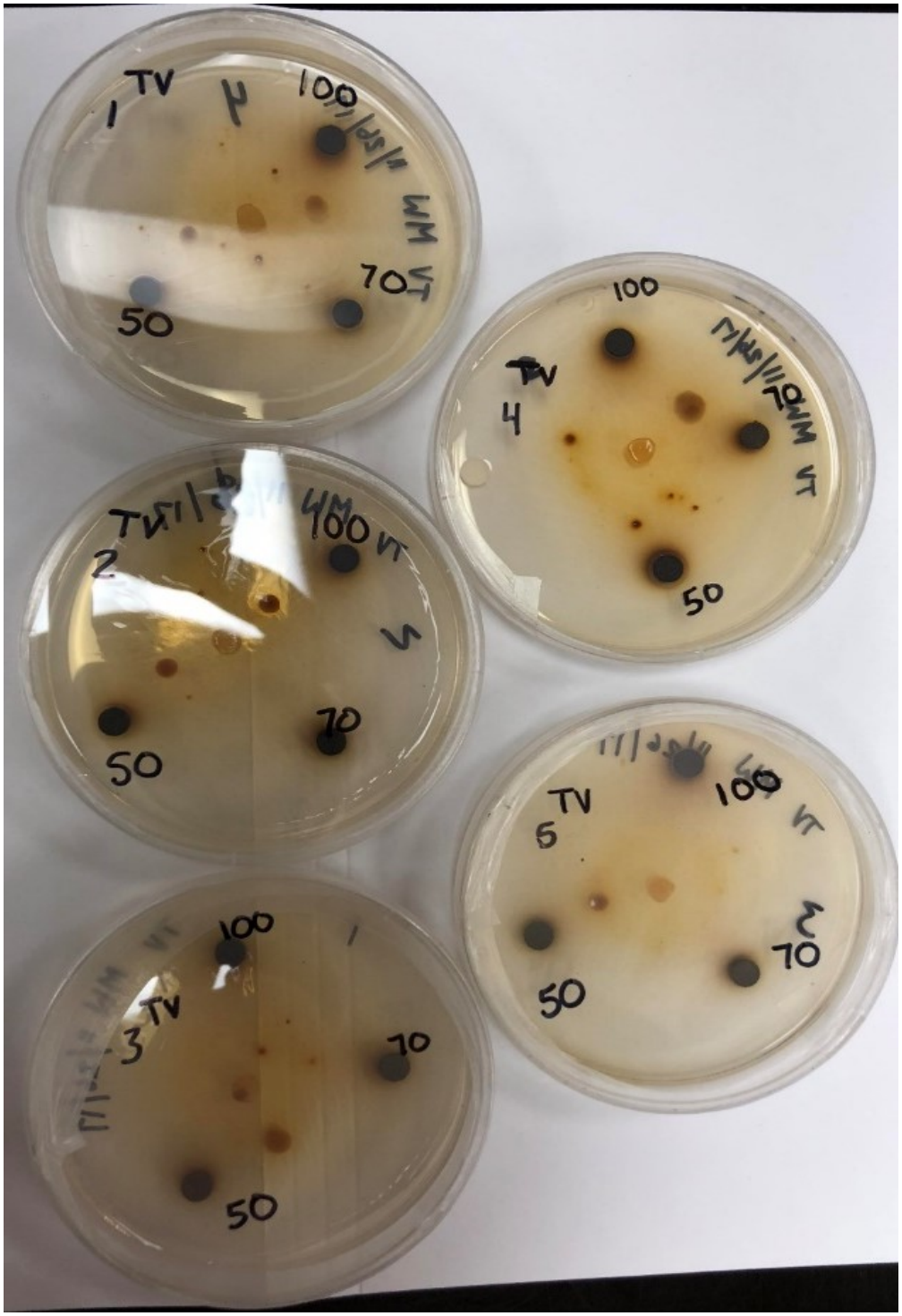

Figure 4.6. Blank Discs with 100%, 70% and 50% creosote discs on agar with *T. versicolor* in center.

Observations for *G. trabeum* are shown in Figure 4.2. There were similar small zones of inhibition around each preservative dipped disc, indicating that the diluent did not affect the interaction that the fungi had with the preservative. The response of *N. lepideus* can be observed in Figure 4.3. The fungi interacted with the 50% creosote disc, however it did not react with the 70% creosote disc or the 100% creosote disc. Observations of the *P. placenta* decay fungi can be seen in Figure 4.4. The fungi had a small zone of inhibition around each disc. Since there were no observational differences, it is acceptable to state that either 50%, 70% or 100% creosote is effectively resistant to the decay fungi *P. placenta*. Figure 4.5 shows the results from *S. hirsutum* decay fungi which is a slow growing fungus. It did not interact with any of the discs. Furthermore, the fungus grew equally away from all of the discs indicating that it did not favour one preservative treatment compared to another. The fungus *T. versicolor* is known for its rapid growth and can be observed in Figure 4.6. It is evident that there was no significant growth in any particular direction on the fungi plates. There were zones of inhibition around each of the creosote treatments which indicated that regardless of the concentration of diluent added, the creosote was able to prevent fungal decay.

4.1.4 Discussion of Fungal Decay Studies

In all the experiments above, the presence of biodiesel in the diluent had no significant effect on the efficacy of creosote as a wood preservative. Based upon the results of the fungi decay trials in Sections 4.1.1 and 4.1.2 there was no significant difference in weight loss of wood blocks treated with different creosote/diluent concentrations except the control which is the wood block without any preservative. It is important to note in this

work that all of the fungi had a coefficient of variation between 20-30% which is good for a biological experiment. The method is a screening test which is typically used to determine the resistance of wood-based materials to decay by selected fungi under controlled laboratory conditions. This method specifies minimum control mass losses which were not met in the present experiment. However, the objective of the experiment was not to determine a minimum retention, but to compare the resistance that creosote treated blocks had against fungi compared to blocks that had been treated with a creosote/ diluent blend. Since the method was only used to determine fungi species and setting up the experiment there were many variations from the method in the AWPA (2016) as well as the British Standards (1997). One of the main variations was the species of wood used. The AWPA (2016) used Southern Yellow Pine which is a softwood and easily breaks down. However, Rock Maple was used for this experiment which is a hardwood and is more durable than softwoods. Additionally, brown-rot fungi generally colonize softwoods and white-rot fungi are generally found on hardwoods (Schmidt, 2006; Ibach, 2013). Another major difference between this experiment and E10-16 was that a pilot plant was used to pressure treat wood blocks in this experiment and in the E10-16 standard the blocks are treated in a laboratory setting.

There were no significant differences observed when the creosote blocks were compared to one another at various creosote concentrations. Observations in Section 4.1.3 demonstrated that the addition of biodiesel did not significantly impact the efficacy of creosote against the five fungi species tested in this study. Specifically, the fungus *S. hirsutum* was noted to have grown the slowest out of all of the species examined. *N.*

lepideus, G. trabeum, and *P. placenta* are in the top 10 most prevalent fungi found on softwoods (Zabel & Morrell, 1992). They also found that *G. trabeum* and *T. versicolor* are the most common on hardwoods. *S. hirsutum* was not on either list and it generally attacks spruce trees according to Schmidt (2006). Based on these findings, it is understandable why *S. hirsutum* did not perform as well as the other fungi species in the decay trial. It is also important to note that according to the AWPA (2016) *N. lepideus* is a creosote tolerant fungus, since the preservative is a combination of creosote, biodiesel, and diesel, there were other contributing factors as to why the fungus still broke down the wood.

To date, there have been no additional studies done involving a diluent of biodiesel/ diesel used as a co -solvent in creosote. Similar research is also very limited, including a study on pentachlorophenol degradation in the presence of biodiesel conducted by Langroodi et al. (2012) and an evaluation on the efficacy of copper naphthenate using biodiesel as a carrier performed by McKillop (2014). The objective of the study by Langroodi et al. (2012) was to evaluate the comparative decay resistance of wood treated with PCP/diesel/KB3 carrier and a modified diesel/biodiesel mixture. A 2-year efficacy study using an accelerated soil contact decay test was conducted to compare diesel/KB3 with the diesel/biodiesel blend using Southern Yellow Pine. The results indicated that there was no difference in wood mechanics between diesel/KB3 and diesel/biodiesel which was measured in terms of modulus of elasticity (Langroodi et al., 2012). Furthermore, in another study done by Langroodi et al. (2011), a 6-month study was done to determine the bioremediation in diesel versus biodiesel in soil. The results indicated

that the co-metabolic effect of biodiesel on micro-organisms could accelerate degradation of PCP in treated wood after disposal. In another study petroleum products were substituted for biodiesel when used as a co-solvent for pentachlorophenol. A decay trial was completed using the two fungi *G. trabeum* and *N. lepideus*. The results indicated that the biodiesel caused an increased protection against the fungi (Ge et al., 2012).

In a counter study against biodiesel being added as a co- solvent, Morrell & Freitag (2011) did a laboratory soil block test to address the concerns that the biodiesel as a co-solvent would have a negative impact on the biocide performance. The study indicated that biodiesel should not be used with Copper Naphthenate. On that note, an MSc thesis completed by McKillop (2014) focused on biodiesel being added as a co-solvent to copper naphthenate. In her research, she worked on the efficacy of the preservative, copper naphthenate in the presence of biodiesel, following the standard test E10 regulated in the AWPA and the British standard. She concluded that there was no significant difference in mean mass loss when biodiesel was added as a co-solvent. The mass loss of wood products using copper naphthenate was similar to that of CCA whether biodiesel was added or not. It is also important to note that an analysis of 100% biodiesel was also done by McKillop (2014) and Pennie (2012). In both studies blocks treated with 100% biodiesel followed the AWPA E10 standard. The results indicated that in the presence of biodiesel, there was no significant wood block decay, but the fungi were heavily concentrated on the block because they were attacking the biodiesel itself.

Based on the results in our research, it can be concluded that addition of a diluent of 80% soybean biodiesel and 20% diesel to creosote did not affect the efficacy of creosote. This result is consistent with the observations reported by Langroodi et al. (2011) and (2012) as well as McKillop (2014).

4.2 Odor Suppression

The objective of this section of the project was to determine if there was a significant difference in the amount of naphthalene present in the headspace of creosote solutions when various concentrations of diluent were added. The diluent consisted of 80% soybean biodiesel and 20% diesel. Various methods were employed in order to determine the most effective way of quantifying naphthalene in the headspace of creosote which included Sorbent Tubes, Colorimetric Tubes, Summa Canisters, GC-MS and headspace GC in order to observe odor reduction.

4.2.1 Sorbent Tubes

Wood samples were treated in the pilot plant as described in Section 3.1. Sorbent tubes were used on three trials of 100% creosote. After the samples were collected the tubes were sent to Agat Laboratories to determine the quantity of naphthalene collected. The results indicated that Sample 1 had a naphthalene concentration of 10.9 (µg/tube), Sample 2 had a concentration of 67.4 (µg/tube), and Sample 3 had a concentration of 13.8 (µg/tube). The results showed that Sample 1 and Sample 3 were similar, whereas Sample 2 was higher. These results are unreasonable because all three samples collected were from trails of 100% creosote therefore the results should be the same. The

difference in results is likely due to the to the fact that the exact amount of air that went through the tube could not be precisely controlled when the sampling was taken from an enclosed cylinder. Since it was difficult to control the volume of air that went through the tube it is difficult to compare one naphthalene concentration to another.

Sorbent tubes are traditionally used for environmental monitoring to test air quality of an industrial plant. In such cases the amount of air is not limited to an enclosed container so the amount of air passing through the tube is accountable as long as the pump is reasonably accurate. Furthermore, according to the National Institute for Occupational Health and Safety NIOSH (1994) naphthalene testing requires a volume of between 100-200 L and has a range of 19 – 83 mg/m^3. Therefore, this kind of method is clearly not designed for an enclosed container and better for industrial workplace settings. Based upon the lack of precision with the sampling device, no further testing was conducted using charcoal tubes and thus an alternative method, Summa Canisters were attempted.

4.2.2 Summa Canister

Summa Canisters are widely used for collecting gaseous samples due to their quick and easy sampling method. They were used in this project to determine if they would be a suitable method for quantifying naphthalene in the headspace of creosote. The results of the samples collected are summarized in Table 4.7.

Table 4.7. Naphthalene concentrations (mg/m^3) from Summa Canister results analyzed by Maxxam Analytic and Agat Laboratories. *

Creosote Concentration	Rep number	Naphthalene Concentration (mg/m^3)	Analytical company	Date Sampled
100	Rep 1	59	Agat Labs	Oct 6 2016
100	Rep 2	82	Agat Labs	Oct 6 2016
100	Rep 3	77	Agat Labs	Oct 6 2016
100	Rep 1	94	Agat Labs	24-Nov-16
100	Rep 2	330	Agat Labs	24-Nov-16
100	Rep3	86	Agat Labs	24-Nov-16
75	Rep 1	130	Agat Labs	Dec 6 2016
75	Rep 2	200	Agat Labs	Dec 6 2017
75	Rep 3	250	Agat Labs	Dec 6 2018
75	Rep 1	110	Agat Labs	Dec 8 2016
75	Rep 2	230	Agat Labs	Dec 8 2016
75	Rep 3	1300	Agat Labs	Dec 8 2016
100	Sample 1 Rep 1	130	Agat Labs	Feb 15 2017
100	Sample 1 Rep 2	74	Agat Labs	Feb 15 2017
100	Sample 1 Rep 3	80	Agat Labs	Feb 15 2017
100	Sample 2 Rep 1	18	Agat Labs	Feb 15 2017
100	Sample 2 Rep 2	21	Agat Labs	Feb 15 2017
100	Sample 2 Rep 3	29	Agat Labs	Feb 15 2017
50	Rep 1	507	Maxxam Analytic	June 6 2017
100	Rep 1	560	Maxxam Analytic	June 6 2017
100	Rep 2	312	Maxxam Analytic	June 6 2017

* Statistics were not completed due to inconsistency in this method.

From Table 4.7 it is evident that there is high variability within the results obtained using the Summa Canisters. For instance, the range of naphthalene concentration detected in 100% creosote was from 18- 560 mg/m^3, while the range detected in 75% creosote was 110 – 1300 mg/m^3. These results did not reflect the true concentrations of naphthalene in different trials, and therefore Summa Canister testing is not a reliable method for use in this project. Similar to using sorbent tubes, the challenge was accurately sampling the same amount of air. Variations in sampling were introduced when the headspace in the

pilot plant varied from one trial to another. Since the system was kept closed when the pressure vessel was being emptied for four minutes it was not possible to determine if the same amount of headspace available for each trial. Furthermore, the volume of air required for the Summa Canister may have been much higher than the amount of headspace available in the pilot plant. Temperature was also an influential factor, which affected the volatility of naphthalene, and thus influenced the naphthalene concentration in air.

Supporting evidence was obtained regarding the use of Summa Canisters to quantify naphthalene. At a Battelle Conference on Remediation of Chlorinated and Recalcitrant Compounds held in Palm Springs California from April 8-12, 2018, an insightful talk with Kesler (2018) occurred. During the conversation, he recommended that Summa Canisters not be used when quantifying the concentration of naphthalene. His company found that PAH's tend to stick to the sides of the Summa Canisters and were difficult, sometimes impossible to remove during cleaning. The theory was confirmed by McHugh (2018) who found significant variability in his results and assumed it to be from the use of Summa Canisters. This is because summa Canisters are reused and if cleaning is incomplete then a false positive could be found. The analysis suggests that this is specifically an issue for vapour samples because generally liquid samples are collected in a container that is later disposed of.

4.2.3 Colorimetric Tubes

In theory, the colorimetric tubes were perfect for such an experiment. In comparison to the other methods attempted, they were made specifically to pick up naphthalene levels in the air. The amount of air that went through the tube was controlled every time due to the pump that came with the tubes. When testing naphthalene directly from the pilot plant, the indicator colour would turn brown. This meant that the tube was overloaded and could not take an accurate reading. Samples were also taken after the spent creosote that was poured into a bucket as explained in Section 3.3.2. To overcome this issue, a high dilution factor was applied in a laboratory setting. For example, in a laboratory setting, the indicator worked well for a sample of 100% creosote. However, when a blend of creosote and biodiesel was tested, the indicator turned blue, which meant there was another reaction and/or adsorption occurring in the tube. After reading more into the problem, it was found that the indicator could not work when fatty acids were present in the sample (Sensidyne, 2013). Since biodiesel is comprised of a mixture of free fatty acids and fatty acid esters, the tube could not be used to determine the amount of naphthalene in a blend of creosote and biodiesel/ diesel diluent.

4.2.5 Perkin Elmer Clarus 680 GC coupled to a Clarus SQ8 MS

Another attempt to quantify naphthalene in the headspace of creosote was to use a GC-MS. For this analysis, the results were analysed in replicates of either two or three. A table of the compiled results can be observed in Table 4.8.

Table 4.8. Naphthalene concentrations (mg/L) obtained from Perkin Elmer Clarus 680 GC coupled to a Clarus SQ8 MS.*

Concentration of Creosote	Duration of Run (min)	Naphthalene (mg/L)	Date sampled
50 rep 1	30	0.56	June 16 2017
50 rep 2	30	2.95	June 16 2017
50 rep 3	30	3.06	June 16 2017
100 rep 1	30	13.28	June 16 2017
100 rep 2	30	8.09	June 16 2017
100 rep 3	60	1.84	June 16 2017
50 rep 1	60	1.17	June 16 2017
50 rep 2	60	1.61	June 16 2017
50 rep 1	90	6.56	June 16 2017
50 rep 2	90	3.28	June 16 2017
100 rep 1	60	9.14	June 16 2017
100 rep 2	60	28.66	June 16 2017
100 rep1	90	52.03	June 16 2017
100 rep 2	90	22.70	June 16 2017
100 rep 1	30 splitless	6.08	June 20 2017
100 rep 2	30 splitless	2.12	June 20 2017
100 rep 1	60 splitless	0.73	June 20 2017
100 rep 2	60 splitless	18.69	June 20 2017
100 rep 1	90 splitless	63.69	June 20 2017
100 rep 2	90 splitless	15.04	June 20 2017
100 rep 1	30 split	0.99	June 20 2017
100 rep 2	30 split	1.03	June 20 2017
100 rep 1	60 split	0.77	June 20 2017
100 rep 2	60 split	1.85	June 20 2017
100 rep 1	90 split	24.77	June 20 2017
100 rep 1	90 split	0.86	June 20 2017

* Statistics were not completed due to inconsistency in this method.

The results in Table 4.8 show that it was difficult to have consistency when analysing the naphthalene on the GC-MS. For instance, on June 20, 2017 a concentration of 100% creosote was analyzed with 2 reps. The first rep resulted in a 24.77 mg/L yield while the second rep resulted in a 0.863 mg/L yield. Due to the large variation in the results, the method proved to be unreliable. One of the largest contributing factors to this

unreliability was the method used for sample collection. As described in Section 3.3.4, the creosote and diluent mixtures (100%, 70%, 50% creosote) were sealed in a container and heated to a pre-set temperature. The air samples were then collected by an air tight syringe and then the sample was injected and dissolved in acetone solvent. The naphthalene content in acetone was determined by a GC-MS. Each step in this method contributed to experimental error; particularly collecting an air sample using an air tight syringe and then transferring it to an acetone solvent.

Since the GC-MS did not have promising results, a headspace GC was used. The headspace GC was chosen because it allowed for the air sample to be analyzed through an autosampler which significantly decreased error. The following section focuses on using a headspace method in order to quantify the naphthalene in the headspace of creosote.

4.2.6 Bruker Scion 456-GC configured for Headspace Analysis

The final method for this series of experiments was to collect liquid preservative samples from the pilot plant and measure the naphthalene concentration in the headspace of liquid samples using a headspace GC. Stored creosote was used that was collected after the wood blocks were treated for the wood block analysis. The liquid mixtures analyzed were 100% creosote; 70% creosote/30% diluent; and 50% creosote/50% diluent. Each creosote/diluent sample was diluted with p-xylene through a series of dilution, with a dilution factor of 2000. To view the standard curve, refer to Figure 3.11. The results, descriptive statistics and the ANOVA analysis are listed in Table 4.9.

Table 4.9. Descriptive statistics and multiple means comparison of the naphthalene concentration (mg/L) in the headspace of creosote using a Bruker Scion 456-GC configured for Headspace. **

Creosote Concentration (%)	N*	Mean (mg/L)	St. Dev	Multiple Means Comparison
100	5	72634	6796	A
70	7	48668	5405	B
50	6	45377	7558	B

*Sample sizes may differ due to outliers
**P-xylene and biodiesel were also tested to ensure that there was no naphthalene present.

The results showed that there was a 33% difference between 100% creosote and 70% creosote and a 37.5% decrease between the 100% creosote and the 50% creosote. These results indicate that the decrease in the quantity of naphthalene found in the headspace of creosote was mainly from dilution factor. Previous research suggests that when biodiesel is added to creosote, a significant odor reduction occurs. This can be confirmed through the authors sampling experience. When she was collecting samples, the 100% creosote solutions had a foul, and potent odor in which case a ventilation mask was required for sample collection. However, when biodiesel was added to the creosote solution the odor significantly reduced and a mask was not required during sample collation.

There are a number of possible reasons why this headspace method may not have been suitable for such an analysis. The main reason is that naphthalene may not have been a proper representative. It was assumed that naphthalene would be a beneficial marker as the base chemical because environmental assessments analyze naphthalene content on a

regular basis. Naphthalene may be a contributor to odor, but the overall reaction may be more complex than just focusing on naphthalene alone. Previous research has shown a reduction in the odor of various wood preservatives. For instance, Fortin et al (2002) conducted a study regarding pentachlorophenol and odor suppression. The study concluded that based upon the airborne substances, C4-, C5-, and C6- alkylbenzene isomers contributed to odor perceived by humans. Based on the current results it is evident that naphthalene is not a beneficial base chemical to be used as the marker and the occurring process is more complex than can be demonstrated in this thesis. In order to further comprehend the complexity of the chemical reaction, analysis of all of the PAH's is required.

4.3 Leaching Test Using Rainfall Simulators

Eight rainfall simulators were used in the leaching experiment. The total volume available in each simulator was 378 L. In order to ensure that the wood was above the water level and was not submerged during the experiment, 55 L of water was added at the beginning. The water was recirculated through the rainfall simulators at a rate of 4.13 L/min. The simulators were run for 9 hours a day for 5 days (totaling 45 hours), and the total rainfall that fell on the wood was 11, 151 L.

The following catchment equation is used to convert mm of rainfall to L of rainfall based upon the surface area of location being rained on. (U.S. Army, 2013). Catchment Area (m^2) * Annual Rainfall (mm) = Total Rainfall on Catch Area (L). Abbotsford, BC has one of the highest rainfall rates in Canada with an annual average of 1538 mm (Osborn,

2018). Therefore, using the catchment equation above, if the area of wood exposure in this experiment which is 0.50945 m^2 (see section 3.4.1) is simulated in this experiment then the total rainfall would be 783.53 L. In order to determine how many years of rainfall was simulated for the experiment based on the quantity of rainfall given above, the total rainfall simulated in this experiment would be divided by the amount of rainfall in L compared to the annual rainfall in Abbotsford, BC. Therefore, the experiment simulated 14.2 years of rainfall total.

$$4.13 \frac{\text{L}}{\text{min}} \text{ x } \frac{60\text{min}}{1\text{ hour}} \text{ X } 45 \text{ hours} = 11{,}151 \text{ L}$$

$$0.50945\text{m}^2 \text{ x } 1538\text{mm} = 783.53 \text{ L}$$

$$11{,}151 \text{ L} \div 783.53 \text{ L} = 14.2 \text{ years of rainfall.}$$

4.3.1 Leaching of PAHs and TPHs from Rail Tie

Wood core samples were collected before and after rainfall simulation. Water samples were also collected before and after sampling. The purpose of collecting water samples before the experiment was to confirm that there were no initial polyaromatic hydrocarbons (PAH's) or total petroleum hydrocarbons (TPH) in the water samples before the experiment. The PAH's included in this analysis were: Acenaphthene, Acenaphthylene, Anthracene, Benzo(a)anthracene, Benzo(a)pyrene, Benzo(b)fluoranthene, Benzo(g,h,i) perylene, Benzo(k)fluoranthene, Chrysene

Dibenz(a,h)anthracene, Fluoranthene, Fluorene, Indeno(1,2,3-cd) pyrene, Naphthalene, Phenanthrene, and Pyrene. The concentrations are summarized in Table 4.10 and 4.11. The TPH's analyzed in this experiment included: Benzene, Toluene, Ethylbenzene, Total Xylenes, C6 - C10 (less BTEX), and C10-C32 Hydrocarbons. Once again, the concentrations are summarized in Tables 4.10 and 4.11.

From each rail tie, twelve core samples were taken (three core samples from each long side). After the core samples were collected they were complied and ground in a coffee grinder and sent away for analysis of PAH and TPH. The results are presented in Table 4.10. A two-sample-t test was done for the data before and after leaching. For the PAH concentrations, the t-value was -0.97 and the p-value was 0.350. The T-value of the TPH concentration was 0.71 and the p-value was 0.488. Based on the results from the two-sample t test there was no significant difference between the concentration of PAH or TPH before and after the leaching experiment at $\alpha = 0.05$.

Table 4.10: Concentrations (mg/kg) of PAH and TPH in wood cores before and after leaching.

Sample	Concentration (mg/kg)	Standard Deviation	Sample Size
PAH Before	39088	10171	8
PAH After	34583	8311	8
TPH Before	8396	1902	8
TPH After	7699	2006	8

Water samples were collected in duplicate for each rail tie and were analysed for PAH and TPH content with results shown in Table 4.11. Two two-sample-t tests were

completed between Samples 1 and 2 of the PAH and TPH concentrations in order to confirm that there was no significant difference. The t-value of the PAH concentrations was -0.41 and the p-value was 0.690. The T-value of the TPH concentrations was -.10 and the p-value was 0.942. The statistical analysis indicated that there was no significant difference between Sample 1 or Sample 2 for either the PAH or TPH concentrations. Experimental results showed that there was no significant difference between Sample 1 and Sample 2 at $\alpha = 0.05$.

Table 4.11: Concentrations (mg/kg) of PAH and TPH in water samples collected in duplicate after leaching.

Sample	Average Concentration (mg/kg)	Standard Deviation	Sample Size
PAH Sample 1	8.206	2.720	8
PAH Sample 2	8.756	2.674	8
TPH Sample 1	57.28	16.94	8
TPH Sample 2	58.09	16.39	8

4.3.2 Discussion of Leaching Experiment

The standard set by the Canadian Council of Ministers of the Environment (CCME) Environmental quality guidelines for PAH's states that the permissible exposure limit for PAH's in freshwater sediment is 7.38 mg/kg of dry weight (CCME, 1999). As observed from Table 4.11, the average PAH concentration in the water was 8.206 mg/kg which is higher than the standard stated above. However, since the experiment was simulated for a period of approximately 14.2 years, the overall leaching would theoretically be lower than 7.38 mg/kg. Furthermore, the RBCA (2016) states that the acceptable limit of TPH

in sediment ecological screening levels of the protection of freshwater and marine aquaculture life should be a maximum of 500 mg/kg. Based on the results from Table 4.12 it is evident that the values are much lower than this which indicates that the standard was met.

It is important to acknowledge that the leaching experiment was simulating a long period of time, in which case the TPH would not leach into the environment at once but over a period of 14.2 years. There has not been a significant amount of research completed regarding the addition of biodiesel to creosote as a co-solvent. A study was completed by Ge et al. (2012) where conventional petroleum biodiesel was replaced with biodiesel as a co-solvent for pentachlorophenol. A leaching test was conducted, and the results indicated that biodiesel performed as well as the conventional petroleum-based co-solvents. In summary, rail ties treated with a creosote/diluent preservative leach at rates that meet environmental standards in Canada.

Chapter 5.0 Summary of Experiments

This project was broken into three major sections which were: fungal analysis, odor suppression and leaching. The results of the fungi analysis were able to confirm that with the addition of a diluent composed of 80% soybean biodiesel and 20% petroleum diesel there was no significant difference in the percent weight loss of rock maple. The results showed significant differences between the control blocks and the creosote treated blocks with the exception of *S. hirsutum*. *S. hirsutum* generally attacks spruce trees, not Rock Maple which was the wood species used in this experiment. Another discovery was that *N. lepideus*, which is a creosote resistant fungus according to the AWPA (2016), did not attack wood blocks treated with creosote at a higher rate compared to the other fungi analysed. The second fungi analysis confirmed that the fungi did not show any significant favoring towards the discs that were treated with creosote and diluents.

The second section of the project aimed at evaluating the concentration of naphthalene in the headspace of creosote. In this section various methods were attempted in order to quantify naphthalene., A method using a GC with headspace was successful. The samples tested using a GC with headspace consistently exhibited a reduction in naphthalene concentration based upon the dilution factor of the diluent added to creosote.

Finally, the leaching analysis which simulated rainfall on eight rail ties using eight rainfall simulators, over a simulated period of 14.2 years was performed. Wood cores that were collected from the rail ties before and after the experiment did not show a

significant loss in the concentration of PAH's of TPH's. Furthermore, water samples collected and analyzed for PAH's and TPH's would have been within the standard if the full timeline of the experiment were considered.

Chapter 6.0 Conclusions

The goal of this research was to evaluate whether the addition of a diluent composed of 80% soybean biodiesel and 20% petroleum diesel would act as a beneficial co-solvent for the wood preservative creosote. Based on the results from the experiment completed, several conclusions can be made.

The first objective of the projected was to investigate the use of the diluent as a co-solvent upon the efficacy of creosote against wood decay, caused by five specific wood decay fungi (including creosote tolerant fungi). The results concluded that the addition of the diluent as a co-solvent did not have a negative effect on the protective ability of creosote.

This research could benefit from further exploration in each section analyzed. The fungi analysis would be more insightful if fungi species specifically known to attack Rock Maple were compared in the future research. It could also benefit from a longer fungi trial to ensure that the minimum retention levels were met.

The second objective of this experiment was to quantify the naphthalene concentration present in the headspace of creosote when the diluent was added as a co-solvent. The results indicated that there was a significant decrease in the concentration of naphthalene emitted into the environment when the diluent was added. However, additional research should focus on odor suppression where an analysis is completed using all of the PAHs.

Additional experiments should also focus on sense tests where various concentrations of creosote and diluent are presented to volunteers who determine if the solution has an odor or not.

The final objective was to investigate leaching in order to determine if regulations would be met if the diluent was added to creosote as a co-solvent. The results indicated that through simulating long-term rainfall in a short period of time, environmental regulations were met according to Canadian guidelines. Further research would compare a blend of creosote/diluent with a blend of creosote. Additionally, collecting samples throughout the experiment, such as on an hourly or daily basis, would be more beneficial for further research as oppose to sampling before and after the experiment.

Based upon the results presented in this thesis, the recommended blend of creosote/ diluent for Stella-Jones Inc., is 70% creosote/ 30% diluent for three main reasons. Firstly, the leaching analysis, where 70% creosote/30% diluent and the results met environmental regulations. Secondly, based upon observations in the fungi analysis the blend of 70% creosote/ 30% diluent was the most beneficial against fungi because it acted similar to the 100% creosote and it did not have additional fungi growth like the 50% creosote/ 50% diluent. Thirdly, there was a significant decrease in the concentration of naphthalene emitted into the environment when the diluent was added to creosote.

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Appendix (A) Fungi Decay

Table A.1: Raw data of post oven percent weight loss of Rock Maple blocks plated with *Trametes versicolor*

Control	100	50	70
20.38	6.65	6.45	5.85
14.88	6.82	6.04	6.62
16.38	7.36	6.37	7.41
15.14	7.40	7.20	7.50
16.11	7.36	6.30	6.43
18.66	6.82	5.99	5.93
15.81	6.60	1.89	7.33
20.10	7.50	10.67*	6.67
26.95	6.40	6.77	7.29
26.68	7.70	6.45	7.15

*Indicates values that were removed because they were outliers according to statistical software

Table A.2 Raw data of post oven percent weight loss of Rock Maple blocks plated with *Stereum hirsutum*

Control	100	50	70
7.05	5.59	6.07	6.35
5.97	6.16	6.09	6.13
5.02	3.79	5.78	5.95
5.35	8.02	5.84	5.39
4.97	5.24	6.07	6.18
4.70	5.83	5.89	6.34
5.80	4.21	5.88	6.46
5.33	5.46	6.32	6.15
6.30	5.97	6.31	6.61
N/A**	7.04	6.39	5.93

*Indicates values that were removed because they were outliers according to statistical software

**There is a value missing in the control because the plate was contaminated and could not be used.

Table A.3 Raw data of post oven percent weight loss of Rock Maple blocks plated with *Gloephyllum trabeum*

Control	100	50	70
11.31	5.60	5.78	5.70
10.60	5.94	6.13	6.11
12.13	5.73	5.84	5.91
9.70	4.54	4.94	5.98
13.82	5.94	6.16	5.98
17.79	7.59	7.67	6.29
10.72	5.63	5.60	6.40
24.71*	5.98	5.97	5.85
10.18	5.83	5.90	5.78
14.62	5.85	6.34	5.92

*Indicates values that were removed because they were outliers according to statistical software

Table A.4 Raw data of post oven percent weight loss of Rock Maple blocks plated with *Postia placenta*

Control	100	50	70
7.09	5.95	5.67	0.71
9.81	6.19	6.29	11.85*
11.24	6.07	6.12	6.39
7.03	6.53	5.90	6.21
16.77*	6.06	5.66	6.26
6.27	6.87	6.17	5.98
9.28	6.35	6.02	6.72
7.95	5.87	6.16	6.55
14.30	6.53	5.99	6.72
-7.90*	6.09	5.45	6.11

*Indicates values that were removed because they were outliers according to statistical software

Table A.5 Raw data of post oven percent weight loss of Rock Maple blocks plated with *Neolentinus lepideus*

Control	100	50	70
11.05	6.38	6.89	6.45
12.90	6.36	6.40	7.17
13.12	5.69	7.17	6.17
11.26	6.22	6.64	6.11
9.07	5.90	5.90	6.21
8.34	6.68	5.87	3.73*
9.10	6.07	6.54	6.27
10.64	6.72	7.06	5.86
10.54	5.73	6.08	5.59
13.27	5.69	6.16	5.99

*Indicates values that were removed because they were outliers according to statistical software

Table A.6 Raw data of Post Conditioning percent weight loss of Rock Maple blocks plated with *Trametes versicolor*

Control	100	50	70
18.92	6.37	6.45	5.85
13.29	6.66	6.04	6.62
14.754	6.95	6.37	7.42
13.43	7.01	7.10	7.50
14.42	6.92	6.30	6.43
16.89	6.57	5.99	5.93
14.10	6.22	1.89*	7.33
18.46	6.91	10.67*	6.67
23.35	6.26	6.77	7.29
24.98	7.63	6.39	7.15

*Indicates values that were removed because they were outliers according to statistical software

Table A.7 Raw data of Post Conditioning percent weight loss of Rock Maple blocks plated with *Stereum hirsutum*

Control	100	50	70
6.62*	5.67	6.07	6.13
4.36	5.96	6.10	5.95
3.67	3.73*	5.78	5.39
N/A	8.11	5.84	6.18
3.96	5.52	6.07	6.34
3.67	5.88	5.89	6.46
3.33	4.52	5.88	6.15
4.31	5.48	6.33	6.61
3.44	6.37	6.31	5.93
4.79	7.16	6.39	6.13

*Indicates values that were removed because they were outliers according to statistical software

Table A.8 Raw data of Post Conditioning percent weight loss of Rock Maple blocks plated with *Gloephyllum trabeum*

Control	100	50	70
10.02	5.95	5.60	5.70
9.11	6.05	5.94	6.11
10.69	5.28	5.73	5.91
8.23	6.31	4.54	5.99
12.21	5.76	5.93	5.98
16.46	5.79	7.59*	6.29
9.21	6.19	5.63	6.40
23.31*	6.29	5.98	5.85
8.74	6.48	5.83	5.78
13.30	5.64	5.85	5.92

* Statistical outliers and were not included in the final analysis.

Table A.9 Raw data of Post Conditioning percent weight loss of Rock Maple blocks plated with *Postia placenta*

Control	100	50	70
5.53	6.22	6.29	0.71*
8.05	6.30	6.12	11.85*
9.60	6.03	5.90	6.39
5.51	6.38	5.66	6.21
15.45*	6.21	6.17	6.26
4.42	6.62	6.01	5.98
7.64	6.47	6.16	6.72
6.23	6.07	5.99	6.55
3.57	6.39	5.45	6.72
5.13	6.04	6.29	6.11

* Statistical outliers and were not included in the final analysis.

Table A.10 Raw data of Post Conditioning percent weight loss of Rock Maple blocks plated with *Neolentinus lepideus*

Control	100	50	70
9.68	6.14	6.89	6.45
11.83	6.40	6.40	7.17
11.98	5.92	7.18	6.17
9.75	5.56	6.65	6.11
7.97	6.25	5.90	6.21
7.03	5.81	5.87	3.73*
7.89	6.60	6.54	6.27
9.63	5.92	7.06	5.86
9.29	6.11	6.08	5.59
12.11	5.89	6.16	5.99

* Statistical outliers and were not included in the final analysis.

Appendix (B) Headspace Analysis

Table B.1 Raw Data of the Naphthalene Concentration (mg/L) in the headspace of Creosote before dilution calculation

Percent Creosote (%)	Naphthalene Concentration (mg/L)
50	29.58
50	24.02
50	21.61
50*	0*
50	19.23
50	19.80
50	21.89
70	24.67
70	23.06
70	27.82
70	28.01
70	23.68
70	22.07
70	21.03
100	38.69
100	34.99
100	37.78
100	30.97
100*	106.31*
100	39.15
100*	68.45*

* Statistical outliers and were not included in the final analysis.

Appendix (C) Leaching

C.1. Total Polyromantic Hydrocarbons (PAH) in Wood Cores Before and After Leaching

Tie	Total PAH Before (mg/kg)	Total PAH After (mg/kg)
1	39152	32456
2	39325	34901
3	38469	34711
4	34919	31295
5	54789	48781
6	51847	43999
7	27534	26665
8	26308	23855

C.2. Total Petroleum Hydrocarbons (TPH) in Wood Cores Before and After Leaching

Tie	Total TPH Before (mg/kg)	Total TPH After (mg/kg)
1	7608	8038
2	7424	7054
3	7803	7807
4	6859	7056
5	10242	11563
6	12355	9171
7	7692	5403
8	7184	5498

C.3. Total Polyromantic Hydrocarbons (PAH) in Water After Leaching Experiment.

Rail Tie	Total PAH Sample 1 (mg/kg)	Total PAH Sample 2 (mg/kg)
1	7.59	8.04
2	11.49	11.34
3	5.69	4.18
4	12.08	11.70
5	5.69	9.23
6	7.23	10.46
7	10.38	5.65
8	5.50	9.45

C.4. Total Petroleum Hydrocarbons (TPH) in Water After Leaching Experiment.

Rail Tie	TPH Sample 1 (mg/kg)	TPH Sample 2 (mg/kg)
1	54.42	63.72
2	78.63	63.62
3	35.12	32.52
4	77.73	79.63
5	60.33	67.83
6	64.40	65.63
7	33.51	34.72
8	54.12	57.03