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Phenolic Extractives and Natural Resistance of Wood

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1. Introduction

Wood is a natural organic material that consists mainly of two groups of organic compounds: carbohydrates (hemicelluloses and cellulose) and phenols (lignin), that correspond to (65-75%) and (20-30%), respectively (Pettersen 1984). The wood is also constituted of minor amounts of extraneous materials, mostly in the form of organic extractives (usually 4–10%) and inorganic minerals (ash), mainly calcium, potassium, and magnesium, besides manganese and silica.

Generally, wood has an elemental composition of about 50% carbon, 6% hydrogen, 43% oxygen, trace amounts of nitrogen and several metal ions.

Cellulose is a long-chain linear polymer exclusively constructed of β -1,4-linked D-glucose units which can appear as a highly crystalline material (Fan et al, 1982). Often 5000 to 15000 glucose rings are polymerized into a single cellulose molecule.

Hemicelluloses consist of relatively short heteropolymers consisting of the pentoses D-xylose and L-arabinose and the hexoses, D-glucose, D-mannose, D-galactose, D-rhamnose and their corresponding uronic acids. It is composed of only 500-3000 sugar units, and thus has a shorter chain than cellulose (Saka 1991)

Lignin, the third cell wall component, is an aromatic polymer synthesized from phenylpropanoid precursors (Adler 1977). It is a three-dimensional polymer formed of coniferyl, syringyl, and coumaryl alcohol units with many different types of linkages between the building blocks and by far the most complex of all natural polymers.

Extractives are chemical constituents residing in the lignocellulosic tissue that contains an higher diversity of organic compounds, for example triglycerides, steryl esters, fatty acids,

sterols, neutral compounds, such as fatty alcohols, sterols, phenolic compounds such as tannins (Fava et al, 2006), quinones (Carter et al, 1978; Ganapaty et al, 2004), flavonoids (Reyes-Chilpa et al, 1995; Ohmura et al, 2000; Chen et al, 2004; Morimoto et al, 2006; Sirmah et al, 2009), besides terpenoids (Kawaguchi et al, 1989; Chang et al, 2000; Watanabe et al, 2005) and alkaloids (Kawaguchi et al, 1989).

2. Extractives and natural resistance of wood

Cellulose is the major structural component of wood and also the major food of insects and decay fungi. Termites, like fungi, are important biological agents in the biodegradation of wood (Syofuna et al, 2012).

Extractives are low molecular weight compounds present in wood (Chang et al, 2001), also called secondary metabolites, and are indeed crucial for many important functional aspects of plant life. The relationship between extractives and natural durability of wood was first reported by Hawley et al (1924). The natural durability of wood is often related with its toxic extractive components (Scheffer and Cowling 1966; Carter et al, 1978; Hillis 1987; McDaniel 1992; Taylor 2006; Santana et al, 2010).

Heartwood extractives retard wood decay can protect the wood against decay organisms (Walker 1993, Hinterstoisser et al, 2000; Schultz and Nicholas 2002), but the natural durability is extremely complex and additional factors such as density of wood and lignin content, besides this dual fungicidal and antioxidant action, may be involved (Schultz and Nicholas 2002).

Several studies have shown that after removal of extractives, durable wood loses its natural resistance and makes them more susceptible to decay (Ohmura, 2000; Taylor et al, 2002; Oliveira et al, 2010). Several authors investigated the relationships between the wood properties and extractives (Carter et al, 1978; Schultz et al, 1990; Reyes-Chilpa et al, 1998; Chang et al, 1999; Morimoto et al, 2006).

One of the most limiting factors for the commercial utilization of wood is its low resistance to fungi and termites, especially in the semi-arid and sub-humid tropics. The biodegradation is supposed to be one of the major challenges to incur the heavy economic loss. Wood decay fungi and some species of termites are important and potent wood-destroying organisms attacking various components of the wood (Istek et al, 2005; Gonçalves and Oliveira 2006).

The largest group of fungi that degrades wood is the basidiomycetes and is divided into: white-rot, brown- rot and soft-rot fungi (Anke et al, 2006). Brown-rot fungi occurs most often in buildings, can degrade only structural carbohydrates (cellulose and hemicellulose), leaving lignin essentially undigested, whereas white-rot fungi utilize all wood constituents including both the carbohydrates and the lignin. Soft-rot fungi utilize preferably carbohydrates, but also

degrade lignin (Belie et al, 2000). They hydrolyze and assimilate as food the lignocellulose components by injecting enzymes into the wood cells (Erickson et al, 1990).

Termites cause significant losses to annual and perennial crops and damage to wooden components in buildings (Verma 2010). Damage caused by subterranean termites, *Nasutitermes*, *Coptotermes* and *Reticulitermes* historically has been a concern of researchers worldwide. Korb (2007) estimated annual damage caused by termites at about U.S. \$50 billion worldwide. In the city of Sao Paulo, Brazil, alone, a 20-year loss of \$3.5 billion was incurred (Lelis, 1994).

The concentration of extractives varies among species, between individual trees of the same species and within a single tree. Some of these extractives render the heartwood unpalatable to wood destroying organisms. Factors affecting wood consumption by termites and fungi are numerous and complexly related. The amount however can vary from season to season even in the same tissue or are restricted in certain wood species (Taylor et al, 2006).

Several woods contain extractives which are toxic or deterrent for termites, bacteria and fungi resistance (Maranhão 2013; Taylor et al, 2006). Termite resistance of wood is a function of heartwood extractive variability while individual extractives inhibit fungal growth (Neya et al, 2004; Arango et al, 2006).

Biological deterioration of wood is of concern to the timber industry due to the economic losses caused to wood in service or in storage. Fungi, insects, termites, marine borers and bacteria are the principal wood biodegraders. They attack different components of wood at different rates giving rise to a particular pattern of damage (Sirmah 2009). Degradation is influenced by environmental conditions of the wood; whether in storage or in use. The degraded wood material is returned into the soil to enhance its fertility (Silva et al, 2007).

The proposal of this study is to demonstrate the importance of phenolic compounds in natural resistance of wood biodegradation. We collected information of the most representative phenolic compounds (flavonoids, stilbenes, quinones and tannins) found in wood, responsible for resistance of some wood species to bio-degraders (Toshiaki 2001; Windeisen et al, 2002).

3. Flavonoids

Flavonoids are secondary metabolites that occur naturally in all plant families (Harbone 1973). Widely distributed in all parts of plants, these compounds afford protection against ultraviolet radiation, pathogens, and herbivores (Harbone and Willians 2000). The general structure includes a C₁₅ (C₆-C₃-C₆) skeleton joined to a chroman ring (benzopyran moiety), classified into flavanones, flavones, chalcones, dihydroflavonols, flavonols, aurones, flavan-3-ols, flavan-3,4-diols, anthocyanidins, isoflavonoids, and neoflavonoids. Some examples of each class of flavonoids are described in figure 1.

Figure 1. Classification of flavonoids

Flavonoids have an important effect on the durability of wood (Chang et al, 2001; Wang et al, 2004). Accord to Schultz and Nicholas (2000) flavonoids protect heartwood against fungal colonization by a dual function: fungicidal activity and being excellent free radical scavengers (antioxidants). Flavonoids are natural antioxidants and have received attention due to their role in the neutralization or scavenging of free radicals (Gupta and Prakash 2009). Pietarinen (2006) showed that the radical scavenging activity is particularly important because both white-rot and brown-rot fungi are believed to use radicals to disrupt cell walls.

The heartwood of *Lonchocarpus castilloi* Standley (Leguminosae) is highly resistant to attack by the dry wood termites *Cryptotermes brevis* (Walker) (Isoptera: Kalotermitidae). Two flavonoid isolated from the heartwood of this plant, castillen D and castillen E (Figure 2), that presented feeding deterrent activity to *C. brevis* (Reves-Chilpa et al, 1995).

Figure 2. Structure of castillen E and castillen D

Ohmura et al (2000) reported that flavonoids present in *Larix leptolepis* (Pinaceae) wood, principally taxifolin and aromadedrin, showed strong feeding deterrent activities against the subterranean termite, *Coptotermes formosanus* Shiraki (*Isoptera*: Rhinotermitidae) and suggested that some flavonoids such as quercetin and taxifolin (Figure 3) might be useful for termite control agents considering their abundance in plants.

Figure 3. Structure of quercetin and taxifolin

The heartwood of *Acacia auriculiformis* (Leguminosae) has been shown to contain a number of different flavonoids and proanthocyanidins content (Sarai et al, 1980; Barry et al, 2005). According to Schultz et al (1995) the durability of *Acacia* species was attributed the presence of dihydromorin and aromadedrin (Figure 4).

Figure 4. Structure of dihydromorin and aromadedrin

From heartwood of *Morus mesozygia* (Moraceae), besides dihydromorin, were isolated morin and pinobanksin (Figure 5), but the resistance against wood destroying basidiomycetes, *Coriolus versicolor*, *Lentinus squarrosulus* and *Poria* spp. was related to the presence of dihydromorin (Toirambe Bamoninga and Ouattara, 2008).

Figure 5. Structure of morin and pinobanksin

According to Sirmah et al (2009) the durability of *Prosopis juliflora* wood (Leguminosae) was assigned to (–)-mesquitol (Figure 6), but Pizzo et al (2011) related that (–)-mesquitol alone cannot be considered the single most important factor in determining the durability of the *Prosopis* species. Laboratory tests indicated that the heartwood of *P. juliflora* was resistance against to both white- and brown-rot fungi (Sirmah 2009).

Figure 6. Structure of mesquitol

The antifeedant activity of some flavonoids against the subterranean termite *Coptotermes formosanus* Shiraki was examined with no-choice tests and two-choice tests (Ohmura et al, 2000). The structure-activity relationships of these flavonoids (Figure 7) were evaluated and it was found that flavonoids containing hydroxyl groups at C-5 and C-7 in A-rings showed high antifeedant activity. Furthermore, the presence of a carbonyl group at C-4 in the pyran rings of the compounds was necessary for the occurrence of high activity. 3-hydroxyflavones and 3-hydroxyflavanones with 3', 4'- dihydroxylated B-rings exhibited higher activity than those with 4'-hydroxylated B-rings.

Figure 7. Flavonoids and antifeedant activity against the subterranean termite *C. formosanus*

The antifeedant activities of pterocarpanes isolated from the heartwood of *Pterocarpus macrocarpus* Kruz. (Leguminosae) were evaluated against the subterranean termite, *Reticulitermes speratus* Kolbe (Isoptera: Rhinotermitidae). Three isolated pterocarpanes, (-)-homopterocarpin, (-)-pterocarpin, and (-)-hydroxyhomopterocarpin were tested (Figure 8). The most active antifeedant against *R. speratus* was (-)-homopterocarpin. However, all pterocarpanes showed antifeedant activity against *R. speratus* (Morimoto et al, 2006).

Figure 8. Structure of homopterocarpin, pterocarpin and hydroxyhomopterocarpin

From the heartwood of *Dalbergia latifolia* (Leguminosae) were isolated and identified as active against termites and fungi, the neoflavonoids, latifolin, dalbergiphenol, and 4-methoxydalbergione (Figure 9).

Figure 9. Structure of latifolin, dalbergiphenol, and 4-methoxydalbergione

With respect to activity against *Trametes versicolor*, a white-rot basidiomycete, latifolin and 4-methoxydalbergione showed activity. Dalbergiphenol exhibited relatively high antifungal activity against the brown-rot basidiomycete, *Fomitopsis palustris* (Sekine et al, 2009).

Latifolin showed high termiticidal activity and termite-antifeedant against *Reticulitermes speratus* (Kolbe). Dalbergiphenol and 4-methoxydalbergione exhibited moderate termite-antifeedant activity (Sekine et al, 2009).

The structure-activity relationships of latifolin (Figure 10) and its derivatives were analyzed to check if there was a correlation between antitermitic and antifungal activity. It was found that the termite mortality in response to the derivatives 2'-O-methylatifolin, latifolin dimethyl ether, and latifolin diacetate increased 2-fold compared to latifolin. No difference was presented in mortality of termites in the presence of 5-O-methylatifolin and latifolin. The results indicate that the phenolic hydroxyl group at C-5 of the A ring provides antitermitic activities.

$R_1 = \text{OH}$, $R_2 = \text{OH}$	latifolin
$R_1 = \text{OH}$, $R_2 = \text{OMe}$	2'-O-methylatifolin
$R_1 = \text{OMe}$, $R_2 = \text{OH}$	5-O-methylatifolin
$R_1 = \text{OMe}$, $R_2 = \text{OMe}$	latifolin dimethyl ether
$R_1 = \text{OAc}$, $R_2 = \text{OAc}$	latifolin diacetate

Figure 10. Structure of latifolin and its derivatives

With respect to antifungal activity of these compounds, it was found that all compounds presented less activity against white- and brown-rot fungi than latifolin. In addition, both C-5 and C-2' phenolic hydroxyl groups in the A and B rings have antifungal activity against white- and brown-rot fungi. In conclusion, the bioactivity of latifolin depends upon the position of phenolic hydroxyl groups (Sekine et al, 2009).

The heartwood of *Dalbergia congestiflora* Pittier (Leguminosae) tree presented natural resistance to fungal attack. The antifungal effect of various extracts from the *D. congestiflora* heartwood was evaluated against *Trametes versicolor* fungus (Martínez-Sotres et al, 2012). The major component of hexane extract that caused 100% growth inhibition from tested fungi was characterized as (-)-Medicarpin (Figure 11). Medicarpin also isolated from heartwood of *Platymiscium yucatanum* (Leguminosae) was identified active against *T. versicolor* (Reyes-Chilpa et al, 1998).

Figure 11. Structure of medicarpin

4. Quinones

Various types of quinones (benzoquinones, naphthoquinones, or anthraquinones) occur in many plant families (Toshiaki 2001). The above mentioned classification of quinones is described in Figure 12. Termite resistant woods are said to contain allelochemicals such as quinones that possess natural repellent and toxic properties (Carter et al, 1978; Scheffrahn 1991; Ganapy et al, 2004; Dungani et al, 2012).

Figure 12. Classification of quinones

The heartwood of *Tectona grandis* L. f. (Lamiaceae) contains a large amount of quinones that possess considerable influence on the natural durability of teak wood. The naphthoquinone, 4', 5'-dihydroxyepiisocatalponol (Figure 13) plays a key role in the resistance of teak against

fungi attack. In-vitro bioassays indicated that this compound acted as a fungicide against the White-rot fungi *Trametes versicolor* (Niamké et al, 2012). Tectoquinone (Figure 13), an anthraquinone, presented strong antitermitic activity and is assumed to be at the origin of the resistance of teak wood to termites (Haupt et al, 2003; Kokutse et al, 2006). According to Wolcott (1955) this substance is highly repellent to the dry-wood termite *Cryptotermes brevis* (Walker) and Sandermann and Dietrichs (1957) demonstrated its toxicity to subterranean termite *Reticulitermes flavipes*.

Figure 13. Structure of 4', 5'-dihydroxyepiisocatalponol and tectoquinone

Castillo and Rossini (2010) isolated naphthoquinones from heartwood of *Catalpa bignonioides* (Bignoniaceae) that showed activity against the termite *Reticulitermes flavipes*. The most abundant and active termiticidal compounds were catalponol and catalponone (Figure 14).

Figure 14. Structure of catalponol and catalponone

From heartwood of *Tabebuia impetiginosa* (Bignoniaceae) were isolated naphthoquinones, mainly lapachol (Figure 15), that showed no repellent activity to *Reticulitermes* termites but it was repellent to two other termites, *Microcerotermes crassus* (Isoptera: Termitidae) and *Kaloterms flavicollis* (Isoptera: Kalotermitidae) (Becker et al, 1972).

Figure 15. Structure of lapachol

The naphthoquinone, 7-methyljuglone (Figure 16) was isolated and identified as termicidal constituent of heartwood of *Diospyros virginiana* L. (Ebenaceae). Its dimer, isodiospyrin possess also termicidal activity against *Reticulirmes flavipes*, but to a lesser extent (Carter et al, 1978).

Figure 16. Structure of 7-methyljuglone and isodiospyrin

5. Stilbenes

Stilbenes are compounds possessing the 1,2-diphenylethene structure, as well as bibenzyls and phenanthrenes, which are composed of $C_6-C_2-C_6$ skeleton. Stilbenes derivatives of 1,2-diphenylethylene, process a conjugated double bond system. There are two isomeric forms of 1,2-diphenylethylene: *trans*-stilbene and *cis*-stilbene, and the chemical structure of these two stilbenes are shown in Figure 17.

Figure 17. The chemical structure of stilbenes

Hydroxylated *trans*-stilbene has an important role in heartwood durability, especially for a resistance to fungal decay. The durability and resistance to decay by *Pinus sylvestris* (Pinaceae) is due to pinosylvins (Figure 18). Pinosylvins present in the heartwood of *Pinus* species is formed as a response to external stress such as fungal infections or UV light. The 2, 4, 3', 5'-tetra and 3, 4, 5, 3', 5'-pentahydroxystilbenes are responsible for wood resistance against Brown-rot and whit-rot fungi (Schultz et al, 1995).

Figure 18. The chemical structure of pinosylvin and derivatives

From the heartwood of *Bagassa guianensis* (Moraceae) was isolated moracins including others polyphenols such as flavonoids and stilbenoids (Figure 19), that presented activity against *Pycnoporus sanguineus*, a white-rot fungus. Possible synergism between compounds have been hypothesized (Royer et al, 2012).

Figure 19. The chemical structure polyphenols from *B. guianensis*

6. Tannins

Tannins constitute a distinctive and unique group of higher plant metabolites. They presented polyphenolic character and relatively large molecular size (from 500 to >20,000). They are thought by some to constitute one of the most important groups of higher plant defensive secondary metabolites (Haslam 1989).

The designation of tannin includes compounds of two distinct chemical groups: hydrolysable tannins (Figure 20) and condensed tannins (Figure 21).

Figure 20. Structure of hydrolysable tannins

Hydrolysable tannins are molecules with a polyol (D-glucose) as a central core. The hydroxyl groups of these carbohydrates are partially or totally esterified with phenolic groups like gallic acid (gallotannins) or ellagic acid (ellagitannins). Hydrolysable tannins are usually present in low amounts in plants.

Condensed tannins are probably the most ubiquitous of all plant phenolics, and presented exceptional concentrations in the barks and heartwoods of a variety of tree species. They are oligomers or polymers of flavonoid units (flavan-3-ol) linked by carbon-carbon bonds not susceptible to cleavage by hydrolysis (Sirmah 2009).

Condensed tannins are natural preservatives and antifungal agents, found in high concentrations in the bark and wood of some tree species (Zucker 1983). Most plant-pathogenic fungi excrete extracellular enzymes such as cellulases and lignases, involved in the invasion and spread of the pathogen. Condensed tannins most likely act as inhibitors of these enzymes by complexing, blocking their action (Peter et al, 2008). For this reason, extract from various woods and barks rich in tannin have been used as adhesives and wood preservatives for a long time (Brandt 1952; Plomely 1966; Mitchell and Sleeter 1980; Pizzi and Merlin 1981; Laks et al, 1988; Lotz and Hollaway 1988; Toussaint 1997; Thevenon 1999).

Figure 21. Structure of condensed tannins

7. Conclusions

The protection of wood against biodeterioration is related to its chemical composition, mainly due to the accumulation of extractives in the heartwood. Wood extractives are nonstructural wood components that play a major role in the susceptibility of wood against wood decay organisms. The attack of these organisms in general can be prevented with synthetic organic and inorganic preservatives; however, such products are very harmful to human health and the environment. Several studies have considered that, it is possible the application of wood extractives as natural preservatives. The main components of wood extractives that confers natural resistance against biodeterioration agents are, tannins, flavonoids, quinones and stilbenes.

- Frequently, condensed tannin can be obtained inexpensively by extracting the bark materials with hot water solvent and has been used as preservatives for a long time.
- Flavonoids exhibit antifungal activity as well as feeding deterrent activities against subterranean termites.
- Quinones – possess natural repellent and toxic properties, mainly against termites.
- Stilbenes has an important role in heartwood durability, especially for a resistance to fungal decay.

The characteristics of all wood species are described in Table 1.

Scientific name	Familie name	Common name	Resistance	Origin
<i>Acacia auriculiformis</i>	Leguminosae	Australian wattle	Durable wood (Ashaduzzaman et al, 2011)	Australia, Indonesia, Papua New Guinea
<i>Bagassa guianensis</i>	Moraceae	Tatajuba	Very resistant (Rover et al. 2012)	Guianas and Brazil
<i>Catalpa bignonioides</i>	Bignoniaceae	Common Catalpa Indian Bean	Highly decay resistant heartwood (Muñoz-Mingarro et al, 2006)	North America
<i>Dalbergia congestifolia</i> Pittier	Leguminosae	Rosewood	Resistant wood (Martínez-sotres et al, 2012)	Central America
<i>Dalbergia latifolia</i>	Leguminosae	Indian rosewood	Resistant wood (Lemmens, 2008)	Asia
<i>Diospyros virginiana</i>	Ebenaceae	Common persimmon	-	Africa, Asia
<i>Larix leptolepis</i>	Pinaceae	Japanece larch	resistant (Schaffer and Morrell 1998)	Japan
<i>Lonchocarpus castilloi</i>	Leguminosae	Black cabbage bark	very resistant (Schaffer and Morrell 1998)	Latin America
<i>Morus mesozygia</i>	Moraceae	Mulberry	Non-resistant (Schaffer and Morrell 1998)	Africa
<i>Pinus sylvestris</i>	Pinaceae	Redwood, Scots pine	Non-resistant (Schaffer and Morrell 1998)	Europe, Asia
<i>Platymiscium yucatanum</i>	Leguminosae	Granadillo	very resistant (Schaffer and Morrell 1998)	Latin America
<i>Prosopis juliflora</i>	Leguminosae	Mesquite, algarroba	Resistant (Ramos et al, 2006)	South and Central America
<i>Pterocarpus macrocarpus</i>	Leguminosae	Burma padauk	very resistant (Schaffer and Morrell 1998)	Native to Thailand and Myanmar
<i>Tabebuia impetiginosa</i>		Brazil wood	Very resistant (Paes et al, 2005)	Latin America
<i>Tectona grandis</i> L. f.	Lamiaceae	teak	Very resistant (Kokutse et al, 2006)	Native to southern Asia

Table 1. List of wood species with their family, common names, resistance and distribution.

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