SOME TECHNOLOGICAL WOOD PROPERTIES OF *ALBIZIA MALACOPHYLLA*

BY

KIMAIYO SYLVESTER KIBET

(B.Sc., Moi University)

A THESIS SUBMITTED IN PARTIAL FULFILLMENT OF THE REQUIREMENTS FOR THE AWARD OF DEGREE OF MASTER OF SCIENCE IN WOOD SCIENCE AND TECHNOLOGY (PRESERVATION OPTION)

DEPARTMENT OF FORESTRY AND WOOD SCIENCE

SCHOOL OF NATURAL RESOURCE MANAGEMENT

UNIVERSITY OF ELDORET, KENYA

JULY, 2013

DECLARATION

Declaration by the Candidate

This thesis is my original work and has not been presented for a degree in any other University. No part of this thesis may be reproduced without the prior written permission of author and/or University of Eldoret.

Name	Sign	Date
Kimaiyo Sylvester Kibet	…………………	……………………

Declaration by Supervisors

This thesis has been submitted for examination with our approval as university Supervisors.

Name	Sign	Date
Dr. Peter K. Sirmah	…………….	…………............

Senior lecturer,

Department of Forestry and Wood Science,

University of Eldoret, Kenya.

Name	Sign	Date
Dr. Francis G. Mburu	………………	………………

Head/Senior lecturer,

Department of Forestry and Wood Science,

University of Eldoret, Kenya.

DEDICATION

I dedicate this thesis to my parents Mr. and Mrs. Peter Chelimo for their love, prayers, financial support and encouragements. May God richly bless you!

ABSTRACT

Studies were carried out on wood dimensional stability, density, ash content, natural durability and anatomy of the lesser known *Albizia malacophylla* so as to improve its utilization potential thus reducing increasing scarcity in hardwood timber supply in Kenya. Wood specimens were exposed to hydrated copper (II) sulphate in a desiccator and dimensional changes evaluated. Wood density was based on oven dry method while ash was generated in a muffle furnace at 525 °C and measured. Extractive content was based on soxhlet extraction using different solvents at different extraction cycles. Natural wood durability against fungi was based on a soil bed test in a laboratory and termite test in the field using AWPA: E7-1993. Thin wood sections were examined for anatomical features using Leica DMLB microscope fitted with EC3 type camera. Wood extracts were subjected to FTIR and GC-MS analysis using standard laboratory procedures. Data was analyzed using statistica Version 7 for windows. Pair wise comparison for dimensional stability and density was done using two sample independent t-tests to test for equality of means while two-way ANOVA was used to test significant difference in means of percentage mass loss between specimens extracted using different solvents and different decay periods. *A. malacophylla* heartwood is dimensionally stable (5.5%) with less dimensional stable sapwood (9.6%). There was a significant difference ($p = 0.000049$) between the dimensional stability of heartwood and sapwood. Wood density ranged from 0.84 g/cm^3 in sapwood to 1.01 g/cm^3 in heartwood while extractive content ranged from 2.35% to 9.71% in heartwood and 2.27% to 4.6% in sapwood. *A. malacophylla* wood is durable against fungi 8.1% mass loss and very durable against termites, mass loss< 5% reported after 6 months exposure. Results revealed that there was a significant interaction ($p < 0.05$) between exposure period and the treatments. Heartwood has thin-thick walled wood fibers, exclusively solitary vessels rich in gums and other deposits consistent with the found natural durability. Prismatic crystals were seen as long chains and all ray cells are procumbent. Extractives were found to slow down the growth of fungi under laboratory conditions explaining the observed natural durability. FTIR and GC-MS analysis of crude heartwood extractives indicated presence of aldehydes, ketones carbonyl compounds, esters, aromatic, carboxylic acids and aliphatic carbonyl compounds. Put together, these results indicate that wood from *A. malacophylla* is a source of valuable biomolecules useful in medical, cosmetic and wood industry. The wood can be a feasible material in construction, furniture and fencing industry where durability is important. Understanding such properties of *A. malacophylla* provides a basis for a variety of uses both in indoor and outdoor purposes. The research recommends further studies to isolate and identify specific extractive compounds that are responsible for natural durability of *A. malacophylla* wood.

TABLE OF CONTENTS

DECLARATION ii

DEDICATION iii

ABSTRACT iv

TABLE OF CONTENTS v

LIST OF TABLES xi

LIST OF FIGURES xii

LIST OF PLATES xiv

ACRONYMS, ABBREVIATIONS AND SYMBOLS xv

ACKNOWLEDGEMENTS xvii

CHAPTER ONE 1

1.0 INTRODUCTION 1

1.1 Problem statement and justification 3

1.2 Objectives 4

CHAPTER TWO 5

2.0 LITERATURE REVIEW 5

2.1 Botanical description of *Albizia malacophylla* 5

2.2 Wood extractives 6

2.2.1 Meaning of extractives 6

2.2.2 Uses and influence of extractives on wood properties 8

2.3 Classification of wood extractives 9

2.3.1 Tannins 9

2.3.2 Isoprenoids 10

2.3.3 Flavanoids 13

2.3.4 Stilbenes 14

2.3.5 Quinones...16

2.3.6 Lignans, neolignans and related compounds...17

2.4 Natural durability and the role of extractives...18

2.5 Wood destroying microorganisms 19

2.5.1 Bacteria...19

2.5.2 Fungi...20

2.5.2.1 White rot fungi 21

2.5.2.2 Brown rot fungi 22

2.5.2.3 Soft rot fungi 22

2.5.2.4 Molds (Non wood decaying fungi) 23

2.6 Methods of detecting and measuring wood decay 24

2.6.1 Visual Observation..24

2.6.2 Mass Loss 25

2.6.3 Mechanical Properties..26

2.6.4 Permeability 27

2.6.5 Immunodiagnosis 27

2.6.6 NIR Analysis 28

2.6.7 Soil bed test 28

2.7 Wood destroying insects 29

2.7.1 Termites 29

2.7.2 Other insects 32

2.8 Marine borers 33

2.9 Wood physical properties 35

2.9.1 Moisture content 35

2.9.2 Wood density and specific gravity 35

2.9.3 Dimensional stability 37

2.10 Methods of extractive removal from wood 38

2.10.1 Soxhlet extraction 38

2.10.2 fexIKA method of extraction 40

2.10.3 Accelerated solvent extraction 41

2.10.4 Subcritical water extraction 45

CHAPTER THREE 47

3.0 MATERIALS AND METHODS 47

3.1 Tree sampling 47

3.1.1 *Albizia malacophylla* 47

3.1.2 *Pinus patula* 47

3.2 Wood dimensional stability 48

3.2.1 Specimen preparation 48

3.2.2 Evaluation of dimensional stability 48

3.3 Wood density 49

3.3.1 Specimen preparation 49

3.3.2 Wood density determination 49

3.4 Wood ash content 49

3.4.1 Specimen preparation 49

3.4.2 Evaluation of ash 50

3.5 Anatomical description of heartwood 51

3.5.1 Preparation of wood specimens and sectioning 51

3.5.2 Preparation of permanent slides 51

3.6 Evaluation of extractives in wood 52

3.6.1 Wood chips preparation 52

3.6.2 Soxhlet extraction 52

3.6.3 Cold soaking extraction 53

3.6.4 Evaluation of total extractives 54

3.7 Assessment of wood decay against fungi in a soil-bed tests 55

3.7.1 Preparation of test specimens 55

3.7.2 Soil bed preparation 56

3.7.3 Planting of test specimens in the soil bed 57

3.7.4 Assessment of wood decay 58

3.8 Assessment of wood durability against termites in a field test 58

3.8.1 Preparation of wood specimens 58

3.8.2 Test site preparation 59

3.8.3 Exposure of test specimens 59

3.9 Contribution of heartwood extractives to natural durability 60

3.10 Characterization of heartwood extractives 61

3.10.1 FTIR analysis 61

3.10.2 GC-MS analysis 61

3.11 Data analysis 62

CHAPTER FOUR 63

4.0 RESULTS AND DISCUSSION 63

4.1 Wood density and dimensional stability of *A. malacophylla* 63

4.2 Amount of wood ash 65

4.3 Wood anatomical characteristics 66

4.3.1 Gross wood features 66

4.3.2 Microscopic features 66

4.4 Determination of extractive content 69

4.4.1 Amount of wood extractives by soxhlet extraction 69

4.4.2 Amount of wood extractives by cold soaking extraction method 71

4.5 Natural wood durability tests 72

4.5.1 against fungi in a soil bed test 72

4.5.2 Growth inhibition test 75

4.5.3 against termites in a field test 81

4.6 FTIR and GC-MS analysis of heartwood extractives 83

CHAPTER FIVE 92

5.0. CONCLUSIONS AND RECOMMENDATIONS 92

5.1. Conclusions 92

5.2 Recommendations 94

5.2.1 From the study 94

5.2.2 For further research 94

REFERENCES 95

APPENDIX 110

LIST OF TABLES

Table 1: Density and dimensional stability of *A. malacophylla* wood................................63

Table 2. Mean percent yield of extractives from *A. malcophylla* wood. 69

Table 3: Soil bed moisture content...73

LIST OF FIGURES

Figure 1: Structures of typical condensed and hydrolysable taninns 10

Figure 2: Chemical structure of typical steroids from R: alkyl group 12

Figure 3: Chemical structure of flavanoid family 14

Figure 4: Chemical structure of stilbenes 15

Figure 5: Chemical structure of Quinones 16

Figure 6: Chemical structure of lignans 17

Figure 7: Extraction cycle of the *fex*IKA method 40

Figure 8: Diagram showing accelerated solvent extraction system 42

Figure 9:Comparison between extractive content of *A. malacophylla* from different solvents 71

Figure10: Percentage mass loss of solvent extracted and non-extracted wood specimens of in a fungal soil-bed test. 73

Figure 11: Percentage growth inhibition of *A. malacophylla* heartwood extractives from different solvents… 76

Figure 12: Effect of hexane heartwood extractives of *A. malacophylla* on the growth of fungi 77

Figure 13: Effect of acetone heartwood extractives on the growth of fungi 78

Figure 14: Effect of *A. malacophylla* heartwood water extract on fungal growth 79

Figure 15: Mass loss of wood specimens exposed to termites..82

Figure 16: FTIR spectrum of dichloromethane extractives from *A. malacophylla* heartwood..84

Figure 17: FTIR spectrum for hexane extractives from *A. malacophylla* heartwood.........85

Figure 18: FTIR Spectrum of acetone extractives from *A. malacophylla* heartwood.........86

Figure 19: FTIR spectrum of water extractives from *A. malacophylla* heartwood.............87

Figure 20 (a-f): Mass spectra of acetone extract from *A. malacophylla* heartwood at different retention time...90

Figure 21: Mass spectrum of water extract from *A. malacophylla* heartwood 91

LIST OF PLATES

Plate 1: A mature *Albizia malacophylla* tree...6

Plate 2: Soxhlet extractor 39

Plate 3: Preparation of Wood chips from *A. Malacophylla* heartwood and sapwood.......52

Plate 4: Extraction of *A. Malacophylla* wood chips by cold soaking method 54

Plate 5: Rotary evaporator...55

Plate 6: Soil bed test..57

Plate 7: Termite field test layout at Cheptebo 59

Plate 8: Gross wood features of *A. malacophylla* (a) sapwood and heartwood delimitation, (b) halfed portion of the stem...66

Plate 9: Photomicrographs of *A. malacophylla* heartwood (A) tangential section ×10 (B) tangential section × 20 and (C) Cross section, (D) showing vessels (E) radial section ×20 and (F) radial section ×10…………………………… 68

Plate 10: Fungal growth on potato dextrose agar treated to different concentrations of heartwood extractives of *A. malacophylla* 80

Plate 11: Extent of termite attack on wood specimens exposed in the field for six months..... 81

ACRONYMS, ABBREVIATIONS AND SYMBOLS

%	Percentage
° C	Degrees Celsius
ε	Dielectric constant
mg	Milligram
µg	Microgram
g	Gram
µm	Micrometer
w	Width
h	Height
t	Tangential
r	Radial
l	Longitudinal
CO_2	Carbon dioxide
eV	Electron volt
° F	Degrees Fahrenheit
MPa	Mega Pascals
CCA	Copper chrome arsenate
ASE	Accelerated solvent extraction
ANOVA	Analysis of variance
AWPA	American Wood Protection Association
ESE	Enhanced solvent extraction
FSP	Fiber saturation point

FTIR	Fourier Transform Infrared Spectroscopy
GC-MS	Gas chromatography/ mass spectroscopy
KBr	Potassium Bromide
KEFRI	Kenya Forestry Research Institute
MC	Moisture content
NSIT	National Institute of Standards and Technology
NIR	Near infrared
PCP	Pentachlorophenol
PDA	Potato dextrose agar
PFE	Pressurized fluid extraction
PLE	Pressurized liquid extraction
RH	Relative humidity
SFE	Supercritical fluid extraction
SWE	Subcritical water extraction
TAPPI	Technical Association for the Pulp and Paper Industry

ACKNOWLEDGEMENTS

I would like to express my heartfelt appreciation to my supervisors Dr. Peter K. Sirmah and Dr. Francis G. Mburu for their invaluable input to this work. Their wise advice and encouragement gave me a reason to move on even when things were hard. I wish to thank Dr. Peter K. Sirmah for his concern, help and consistent guidance right from the beginning of this research work. I would equally wish to thank Dr. Francis G. Mburu for the help and the generosity he extended to me during my research work.

Many thanks go to Mr. Solomon Itubo of Malaba County Council for his assistance during identification and sampling of the tree species. I also thank Mr. Reuben Shanda together with the wood anatomy laboratory staff of KEFRI, for their assistance offered during the preparation of permanent slides for photomicrography. Special thanks are due to Mr. Richard Nyagwachi of Biological Sciences Laboratory, University of Eldoret for his assistance during isolation and the culture of fungi.

I am very grateful for the help offered by the staff of Forestry and Wood Science Department, particularly Mr. Joseph Maranga, Mr. Robert Koech, Mrs. Sarah Andati and Mr. Geoffrey Munene who assisted me when I was setting up my experiments. I am thankful to my colleagues Mr. Godfrey Ali and M/s Beatrice Kinyua for their assistance during collection of data. Special thanks go to my sister Ruth Jemutai and my brothers Timothy Kipkurgat, Nehemiah Kipchirchir, Hezekiah Kiprotich and Amos Kiprop who have always kept on encouraging me to move on.

To all my friends who assisted me in one way or another and whom I have not mentioned here, I say thank you all. Finally I thank the almighty God for His unfailing love and for giving me good health.

CHAPTER ONE

1.0 INTRODUCTION

Albizia malacophylla is a lesser known tropical hardwood species characterized by a rough, pale brown grey bark and large irregular flat scales (Eggeling and Dale, 1951; World Conservation Monitoring Center, 1998). Generally; it grows to about 4.5 metres tall and sometimes attains a height of 9 metres (Eggeling and Dale, 1951). It is restricted to the expanse African wood grassland and savannah. In Kenya, it is mainly found in Teso, Bungoma County. Preliminary survey indicated that its wood is mainly used for fencing posts, charcoal production, firewood, as well as for construction purposes. The tree is reported to lose large part of its habitat to agriculture and human settlement (World Conservation Monitoring Center, 1998). In spite of its utilization potential, *A. malacophylla* is still considered as a non-commercial species whose properties are not well known.

Understanding technological wood properties such as durability can provide information on the likely uses of forest resources as well as important predictions for the serviceable life of products made from them (Gambetta *et al.,* 2004). In many instances, more than one technological property of wood is crucial to the end product use (Forest Products Laboratory, 1999). In selecting a wood species for a product, texture, grain pattern and color may be assessed alongside other properties like dimensional stability, machinability or decay resistance (Forest Products Laboratory, 1999).

Most tropical hardwoods possess good natural durability and are resistant to decay and insect attack (Williams *et al.,* 2001).Teak (*Tectona grandis* L.f) has a good natural durability and for many years it has been used for outdoor purposes (Williams *et al.,* 2001). It is dimensionally stable and very resistant to surface checking and end-grain splitting (Williams *et al.,* 2001). *Albizia adianthifolia*, *A. feruginea* and *A. zygia* have moderately to very durable heartwood, coarse texture, low shrinkage and little movements (Kukachka, 1969). At 12% moisture content, *A. feruginea* has an average density of 705.45kg/m^3 while *A. adianthifolia* and *A. zygia* have average density of 561.55kg/m^3 (Kukachka, 1969). It has been reported that timber from these species should be suitably used for joinery and carpentry (Kukachka, 1969).

Evaluation of natural durability of wood leads to more confidence in wooden structures and it increases competitiveness (Acker *et al.,* 2003). The most important property of wood as a building material is its biological durability (Calonego *et al.,* 2010). Anatomical features such as aspiration of bordered pits, to a great extent, results in reduced permeability while high levels of gums limit the penetration of water (Schubert *et al.,* 2011, Gerardin *et al.,* 2004).

Extractives contribute to wood color, fragrance, durability and at the same time, influences pulping, drying adhesion, hygroscopicicity and acoustic properties (Umezawa, 2001; Baeza and Freer, 2001). Flavanoids in wood contribute to diversity in wood colorations and have significant effect on its durability (Sirmah *et al.,* 2011).

Wood density is closely associated with many wood properties (Githiomi and Kariuki, 2010). Where actual data is not available, density can give a wide indication of mechanical strength, shrinkage, heating value of wood and other properties (Githiomi and Kariuki, 2010). The quality of wood as a building material depends mainly on its density (Kollmann and Côté, 1984). Cracking and twisting, shrinkage and swelling, causing significant sensible problems in wood utilization for building and construction are attributed to undesirable dimensional changes (Sun *et al.,* 2010). Calorific value is an important energy parameter for the selection of fuel wood for energy plantation (Kulla and Obi, 2005). This study focuses on wood properties of the lesser known *A. malacophylla* with a view of finding ways of improving its utilization.

1.1 Problem statement and justification

There is rising demand on timber products in Kenya because of massive constructions, improper utilization of wood and high rate of population growth (Mburu *et al.,* 2008; Marshall *et al.,* 1994). The over dependence on timber resource in Kenya and other developing countries has posed a major challenge such as an increasing scarcity in hardwood timber supply (Sirmah *et al*., 2008; Mburu *et al.,* 2007; Marshall *et al.,* 1994). Projections show that by the year 2020, the demand for poles will be at 2.7 million m^3 per annum (KFMP, 1994). The nature of the forest resource that will be used to meet this demand is changing from trees with established history of durable performance, to small trees such as *Albizia malacophylla* whose properties are not well known (Taylor *et al.,* 2006).

Reliable and up-to date information on the situation of forest resources such as wood products is essential to support decision making for policies and programmes in forestry and sustainable development (FRA, 2010). The research aimed at understanding the wood density, dimensional stability ash content, anatomical features, heartwood natural durability and quantity of extractives in heartwood and sapwood, contribution of extractives to natural durability and chemical characterization of heartwood extractives of the lesser known *Albizia malacophylla*.

1.2 Objectives

The general objective of the study is to understand technological wood properties of *Albizia malacophylla* in relation to its utilization potential.

The specific objectives are:

1. To determine dimensional stability, density and ash content of the wood.
2. To carry out anatomical description of its heartwood.
3. To determine its sapwood and heartwood extractive contents.
4. To determine its heartwood natural durability against fungi and termites.
5. To characterize its heartwood extractives

CHAPTER TWO

2.0 LITERATURE REVIEW

2.1 Botanical description of *Albizia malacophylla*

The genus *Albizia* represents a large number of species widely distributed throughout the tropics of the world (Kukachka, 1969). In Africa the genus is represented by about 30 species, but many of them are relatively small trees (Kukakchka, 1969).

Albizia malacophylla is a savannah tree which attains a height of 4.5-9.0 meters tall at maturity. It is mainly found in wooded grasslands, and thickets with an altitude range of 850m-1680m above sea level. The tree is known as *Ekutata* in Ateso, a common language in Western Kenya and Eastern Uganda. Botanically, it is considered as pioneer species hence does not grow in tropical forests with a closed canopy (Eggeling and Dale, 1951). It is characterized by a grey-black, rough bark that is scaling raggedly (Eggeling and Dale, 1951). The new leaves are pale bright green in color while young branchlets are relatively hairy (Eggeling and Dale, 1951). The leaves are classified as bipinnate with 3-6 pair pinnae and 6-11 pairs of medium large leaflets all about equal in size to 3cm long. It has hemispherical, white and sweet smelling flowers. The seeds are round and flat (Eggeling and Dale, 1951).

Plate 1: A mature *Albizia malacophylla* tree (Author, 2011)

2.2 Wood extractives

2.2.1 Meaning of extractives

According to Umezawa (2001), wood extractives are the constituents of wood with different chemical composition and are extracted from wood with neutral organic solvents and cold or hot water. Some extractives are obtained as exudates from wounded trees (Umezawa, 2001; Tsoumis 1991). Certain inorganic wood materials that are not components of the cell wall such as calcium salts and silica inclusions are at times called extractives even though they are not soluble in the above solvents.

Thus, all inorganic material can be considered as extractives (Umezawa, 2001; Tsoumis, 1991). Extractives are produced when the living ray cells in the inner zone of the sapwood die to form the non-living heartwood. Most of the extractives are found in the heartwood, the presence of some forming the basis of general darkening of this portion of the tree (Kollmann and Côté 1984; Scheffer and Morrell, 1998). Extractives can be removed from wood by extraction using different solvents with different polarities (Soon and Chang, 2012; Baeza and Freer, 2001). Thus, extractives can be divided into lipophilic and hydrophilic compounds depending on the type of solvents used in the extraction stage. Those extractives that are extracted with low polar organic solvents such as dichloromethane and hexane include mainly resin acids, fatty acids, triglycerides, sterile esters and diterphenyl alcohols are classified as lipophylic extractives (Soon and Chiang, 2012; Dorado *et al,* 2001). On the other hand, oligolignans, lignans, phenolic compounds as well as hydrophilic salts, sugars and starch are mainly extracted with polar solvents such as water, methanol, ethanol or acetone and they are grouped as hydrophilic (Soon and Chiang, 2012).

The extractives are sometimes classified as saponifiables and unsaponifiable according to the influence caused on the pulping process (Baeza and Freer, 2001). Saponifiables fatty acids, resin acids, some steryl esters, and glycerides are considered to be compounds that form soluble soaps under alkaline conditions. Unsaponifiables (waxes, some steryl esters, diterpene alcohol and aldehydes, sterols, triterpene alcohols, and fatty alcohols) do not

form soaps and have a tendency to deposit and cause pitch problems (Baeza and Freer, 2001).

2.2.2 Uses and influence of extractives on wood properties

Wood properties such as color, fragrance, strength and durability are influenced by extractives (Umezawa, 2001; Baeza and Freer, 2001; Browning, 1975). Extractives also contribute to the technical and economic pitch problems in the pulping processes (Umezawa, 2001; Baeza and Freer, 2001). Furthermore, extractives affect wood adhesion, hygroscopic and acoustic properties (Umezawa, 2001).

Some extractives are attributed to specific biological properties. Taxol found in *taxus spp*. in very small quantities is a promising drug against cancer (Umezawa, 2001). Similarly, an aryltertrlin lignin podophyllotoxin although it is associated with severe gastrointestinal toxicity, has a strong antitumor and antimitotic activity (Umezawa 2001). Flavanoids have been reported to reduce blood-lipid and glucose thus enhancing human immunity. In addition flavanoids are natural antioxidants capable of scavenging free superoxide radical and anti-aging agents (Ghasemzadeh *et al.,* 2011; Rao *et al.,* 2003). Tannic acid has been described as having antimutagenic, anticarcinogenic and antioxidant activities (Lopez *et al.,* 1999).

2.3 Classification of wood extractives

2.3.1 Tannins

Tannins are phenolic compounds that are soluble in water with molecular weight between 500 and 3000 (Umezawa, 2001). They are compounds of high astringency, able to precipitate alkaloids, gelatin, and other proteins. Polyphenols are plant secondary metabolytes consisting of hydrolyzable and condensable forms (Reed, 1995; Lopez *et al.,* 1999; Umezawa, 2001; Hernes and Hedges, 2004). Esters of an aliphatic polyol and phenolic acids can be hydrolyzed into smaller components and hence are the hydrolysable tannins while oligomers and polymers of polyhydroxyflavan-3-01 units are the condensed tannins. Even though hydrolyzable and condensed tannins are different in structure, they have similar number of phenolic units and thus commonly called plant polyphenols (Umezawa, 2001). In many woody plants tannins are spread widely in wood, bark, needles and leaves (Min *et al.,* 2008; Hernes and Hedges, 2004; Umezawa, 2001).

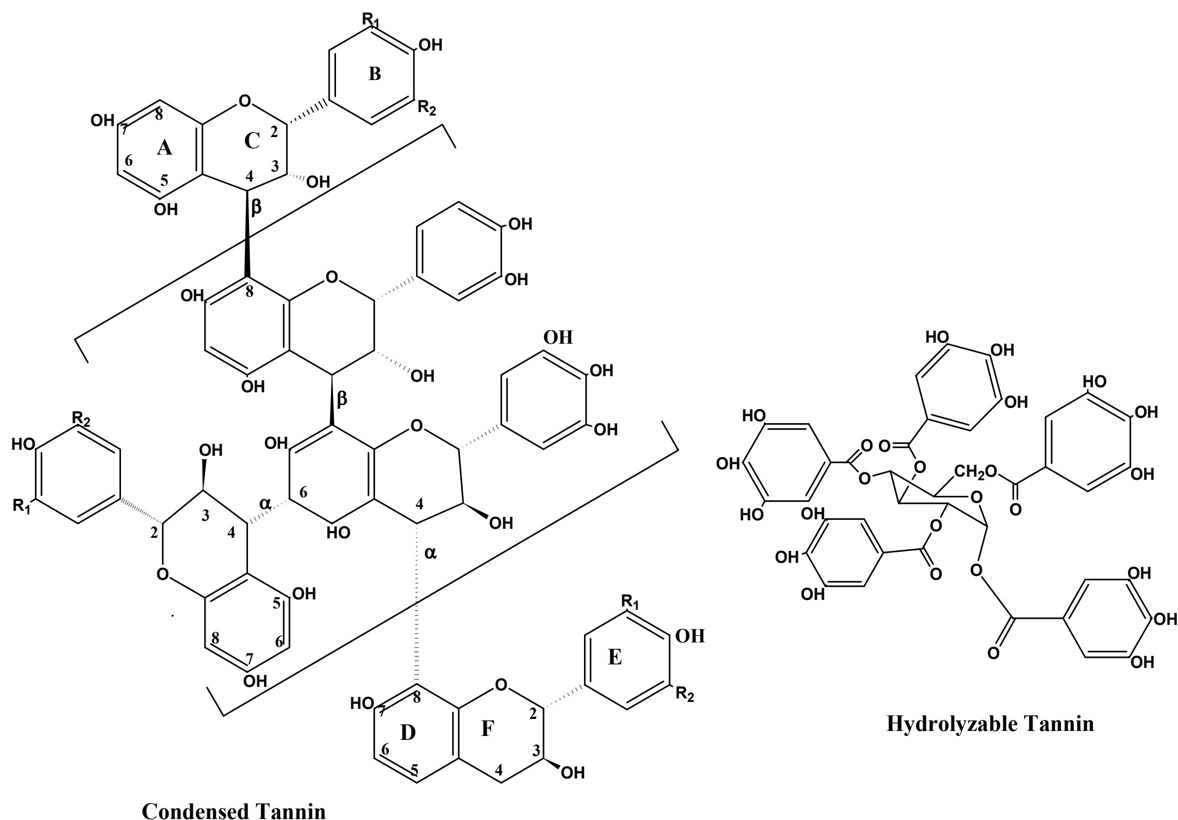


Figure 1: Structures of typical condensed and hydrolysable taninns (Hernes and Hedges, 2004)

2.3.2 Isoprenoids

Isoprenoids are compounds that are made up of isoprene (C_5H_8) units linked linearly or cyclically. In plants, isoprenoids range from essential and somewhat universal primary metabolites, such as sterols, carotenoids, quinones, and hormones, to more distinctive and occasionally species-specific secondary metabolites that may serve as plant defense and communication (Kirby and Keasling, 2009).

Generally, isoprenoids are of three classes namely: terpenoids, steroids, and tropolones. The chemistry of each class has been developed separately despite their close association in biosynthesis (Umezawa, 2001). Among the plant secondary metabolites, isoprenoids represent the largest, highly diverse group with many functions in plant primary and secondary metabolism and are present in vast number of plants including woody plants (Kirby and Keasling 2009; Umezawa, 2001).

Terpenoids are made of isoprene (C_5) units. Depending on the number of the constituent isoprene units, terpenoids are separated into monoterpenes (C_{10}), sesquiterpenes(C_{15}), diterpenes (C_{20}), sesterterpenes(C_{25}), triterpenes(C_{30}), tetraterpenes(C_{40}) and polyterpenes (C_{50}) (Kirby and Keasling, 2009; Umezawa, 2001). Normally wood contains considerable amounts of volatile or essential oils which can easily be separated from wood by steam distillation. These volatile oils and the resin acids are considered as terpenoids. The composition of volatile monoterpenes and sesqueterpenes are the main causes of characteristic odors associated with fresh wood (Umezawa, 2001; Browning, 1975).

Terpenes can formally be considered as condensation products of two or several isoprene (2 – methylbutanidiene) molecules resulting in dimmers and higher oligmers with the elementary formula ($C_{10}H_{16})_n$ (Browning, 1975).

Steroids are compounds with cyclopentanoperhydrophenanthrene skeleton. They are derived from squalene (Umezawa, 2001). β-Sitosterol, is widely distributed strand in the plant kingdom. Examples of steroids are shown in figure 2.

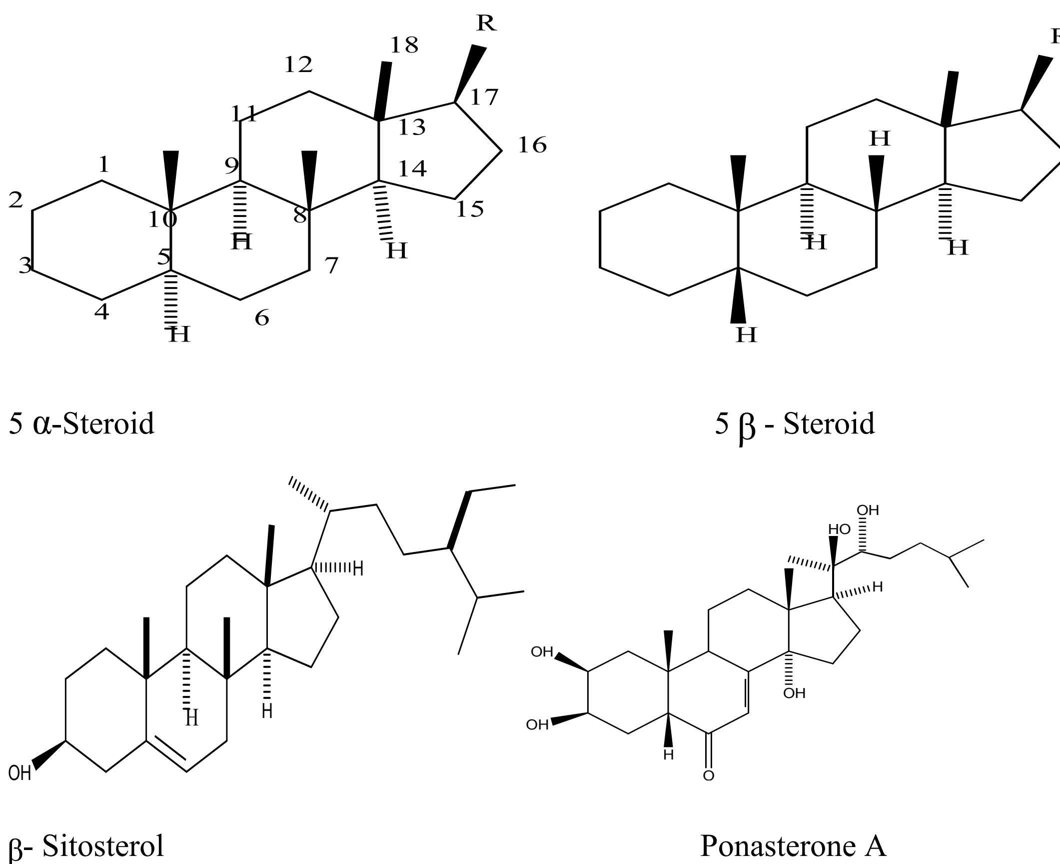


Figure 2: Chemical structure of typical steroids from R: alkyl group (Umezawa, 2001)

Tropolones are nonbenzenoid aromatic compounds having a seven-membered enolone structure, and they are distinctive in many decay resistant conifers such as cedar (Umezawa, 2001). Tropolones are made up of 10 or 15 carbon atoms and they have been considered as mevalonate origin, which is a subclass of isoprenoids (Umezawa, 2001).

2.3.3 Flavanoids

Flavonoids are large family of polyphenolic compounds that are synthesized by plants possessing a common phenylbenzopyrone structure (C6-C3-C6) (Ghasemzadesh *et al.,* 2011; Ren *et al.,* 2003; Umezawa *et al.,* 2001). In woody tree species, flavanoids accumulate in bark, leaves and heartwoods whereas smaller amounts are found in the sap wood and seeds. Flavanoids are further found in the flowers, fruits and roots of many plants (Sirmah *et al.* 2011; Umezawa 2001). They are classified according to the saturation level and opening of the central pyran ring and according to the position of the substitutes present on the parent molecules, mainly into flavanones, flavones, chalcones, dihdroflavonols (flavanonols), flavonols, aurones, flavan-3-ols (catechins), flavan-3, 4-diols (leucoanthocyanidins), anthocyanidins, isoflavonoids, and neoflavonoids (Ren *et al.,* 2003; Narayana *et al.,* 2001; Umezawa, 2001). It has been reported that flavanoids protect plants from ultraviolet induced injury and prevent fungal colonization of heartwood (Sirmah *et al.,* 2011).

Examples of flavanoids are illustrated in figure 3;

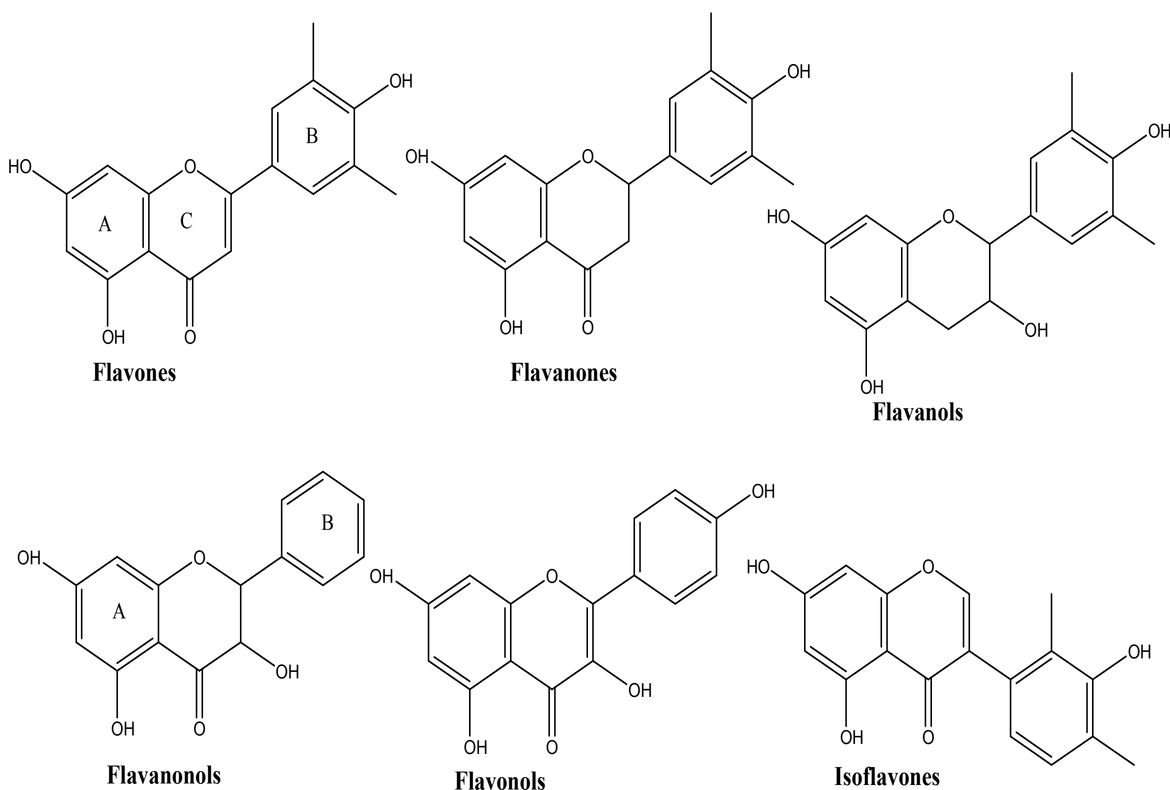


Figure 3: Chemical structure of flavanoid family (Ren *et al.,* 2003)

2.3.4 Stilbenes

Stilbenes refer to compounds that have 1, 2-diphenylethene structure, but currently the newly discovered bibenzyls and phenanthrenes which are composed of C_6-C_2-C_6 skeleton have also been grouped as stilbenes (Umezawa, 2001). The 1, 2-diphenylthylene derivatives possess a conjugated double bond system and are thus, very reactive compounds.

Stilbenes are elaborated from CoA esters of cinnamic acids. Their biosynthesis is similar to that of flavanoids and they occur in Pinaceae, Moraceae, Lecuminosae, among other tree families (Umezawa, 2001).

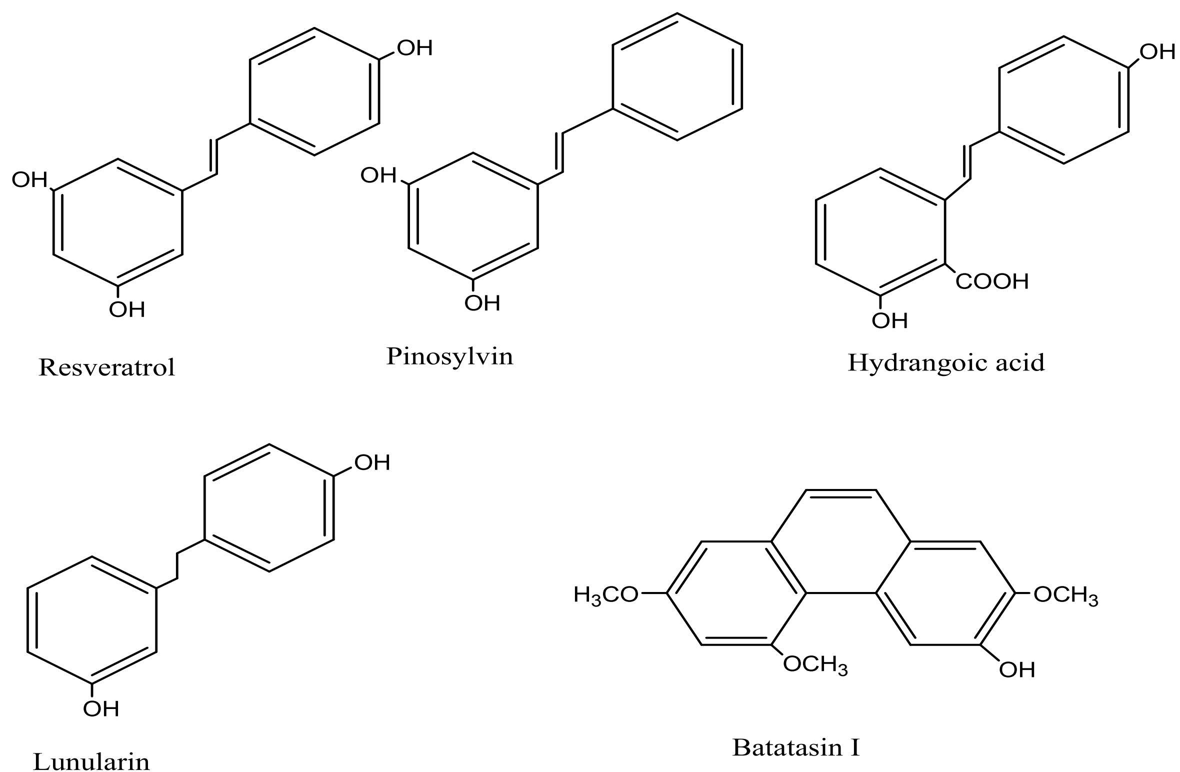


Figure 4: Chemical structure of stilbenes (Umezawa, 2001)

2.3.5 Quinones

A number of quinones are present in many plant families, and most of them are benzoquinones, naphthoquinones, or anthraquinones. (Umezawa, 2001). Majority of the quinones in nature are *p*-quinones although o-quinones are also present.

They are biosynthesized through different pathways, such as the shikimate, the mevalonate, and the acetate-malonate (polyketide) pathways (Umezawa, 2001). They are pigments and they possess various biological activities. For example juglone, which is found in walnut (*juglans nigra*), is well known as allelochemical and it is a skin irritant. Technoquinone and its interrelated compounds possess strong anti-termitic activity while mansonone and its congener cause allergies (Umezawa, 2001).

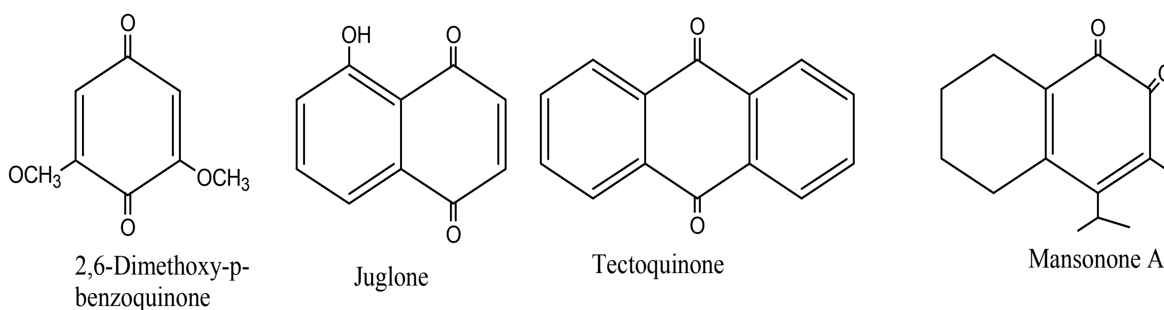


Figure 5: Chemical structure of Quinones (Umezawa, 2001)

2.3.6 Lignans, neolignans and related compounds

Lignans and neolignans are phenylpropanoids present in numerous plants which include hardwoods, softwoods and medicinal plants (Umezawa, 2001). Phenylpropanoid dimer with the phenylpropane unit linked by the central carbon (C8) of their side chain is grouped as a lignan otherwise they are classified as neolignans (Umezawa, 2001). Lignans usually occur as glycosides and they are classified into many subgroups: dibenzylebutanes, dibenzocyclooctadienes, dibenzylbutyrolactones, furans, furofurans, ariltetralins, and arylnaphtalenes (Umezawa, 2001).

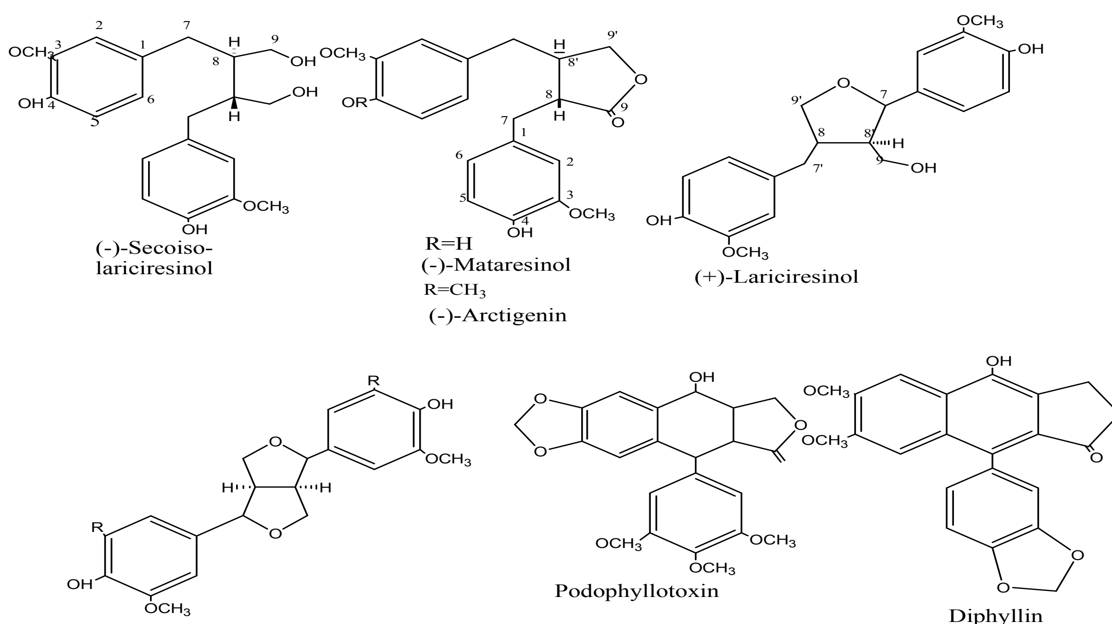


Figure 6: Chemical structure of lignans (Umezawa, 2001)

2.4 Natural durability and the role of extractives

Wood is a natural organic and renewable material that is subjected to biological degradation by different organisms (Vetter *et al.,* 2009; Mohareb *et al.,* 2010). The natural durability of wood indicates its ability to resist the attack of foreign organisms (Panshin and de Zeew, 1980). It is commonly measured by exposing wood to decay fungi under accelerated conditions but wood may also be tested for resistance to termites, marine borers and beetles (Taylor *et al.,* 2002). The natural durability of wood against fungi as well as resistance to insects and marine borers varies widely (Scheffer and Morrel, 1998). It is important that the wood species be evaluated against hazards to which they will be subjected so as to confirm performance under intended exposure conditions (Scheffer and Morrel, 1998). Sapwood has low durability but heartwood of some species can be very resistant to biodeterioriation. Heartwood has greater natural durability in comparison with sapwood of the same species due to the presence of a wide variety of toxic extraneous materials such as essential oils, tannins, and phenolic substances in the heartwood (Panshin and de Zeew; 1980; Baeza and Freer, 2001). Natural durability varies between and within the heartwoods of individual trees (Taylor *et al.,* 2002).

Each wood species possess different physical and chemical properties affecting their durability (Susi *et al.,* 2011). The most important property of wood as a building material is its biological durability (Calonego *et al.,* 2010).

The sapwood of all indigenous species, even those in which the heartwood is highly durable, is vulnerable to attack by biological agents. This is explained by its lack of extractives in sufficient quantity or toxicity to inhibit the growth of micro organisms.

Furthermore, the presence of reserve food in the parenchyma cells of sapwood may increase its susceptibility to decay and particularly to bacterial and fungal staining (Panshin and de Zeew, 1980).

Other factors such as lower moisture content, low rate of diffusion and the blocking of cell cavities by gums, resins, and tyloses in the vessels and tylosoids in the resin canals of heartwood explain its greater durability (Schubert *et al.,* 2011; Panshin and de Zeew, 1980).

2.5 Wood destroying microorganisms

2.5.1 Bacteria

Bacteria are often associated with fungi since their presence in wood has been observed to condition wood in favor of attack by fungi (Nicholas, 1973). It has been known that bacteria can destroy pure cellulose, but can also seriously damage lignified tissues when the conditions are favorable (Nicholas, 1973). For bacteria to cause wood damage, moisture content should be high and in most cases higher than can be tolerated by most fungi. Bacteria build up in logs that are ponded for a long period of time, in veneer bolts of susceptible woods kept for longer time in holding tanks or a water spray, untreated wood in wet or damp ground, wetter wood structures if cooling towers and some even invade living trees (Nicholas, 1973). The sour smell of logs and green timber is an indication of presence of bacteria (Nicholas, 1973; Forest Products Laboratory, 1999).

Wood attacked by bacteria is abnormally permeable because the bacteria damage the pit membrane forming passageways from cell to cell.

Such wood with abnormal absorptivity of the sapwood have a tendency of acquiring excess amount of preservatives, adhesives, paint and pickup excessive moisture (Nicholas, 1973; Forest Products Laboratory 1999). Normally bacteria have little effect on wood properties except prolonged infestation by some species which results in severe degradation of the wood cell walls causing weakening ((Nicholas, 1973; Forest Products Laboratory 1999).

Like some fungi, bacteria may inactivate or destroy preservatives. They have the ability to detoxify creosote and high tolerance of CCA, pentachlorophenol (PCP) and tributyltin, although it is thought that the damage to well-treated wood must be very small (Nicholas, 1973). The most common bacteria in wood products are traced to species of *Bacillus, Aerobacter* and *Psedomonas* all of which are inhabitants of soil and water (Nicholas, 1973).

2.5.2 Fungi

Fungi cause wood decay, mold and most sapwood stains (Forest Products Labratory, 1999). Wood decaying fungus requires; oxygen, favorable temperatures, water and food (Chirra, 1995).

They normally occur when the wood moisture content exceeds 30%, favorable temperature range between 50° and 90 °F, an adequate supply of oxygen and an appropriate source of energy and nutrients (Chirra, 1995). Under favorable growth conditions, decay-producing fungi may attack either heartwood or sapwood in most wood species (Forest Products Laboratory, 1999).

The ability of the decay fungi to degrade wood varies among fungal species and chemical properties and structural features of the wood (Silva *et al.,* 2007). Wood-destroying fungi feed on living or dead wood and they are unique because of their ability to metabolize lignified cell wall substances thereby seriously reducing the strength of wood (Roll, 2003; Blanchette; 1991; Scheffer and Cowling, 1966). Wood-staining fungi and molds, utilizes the contents of xylem parenchyma cells and mostly discolor the wood (Scheffer and Cowling 1966).

Brown rot, white rot and soft rot are types of wood decay that are generally recognized (Powell *et al.,* 2001; Kollmann and Côté, 1984; Scheffer and Cowling 1966). Brown rot and white rot are caused by basidiomycete fungi while soft rot is caused by ascomycotina and deutromycota (Ghosh *et al.,* 2008; Scheffer and Cowling, 1966). Differences among these major types of deterioration are reflections primarily of differences in enzymatic capacities of the causal organisms (Scheffer and Cowling, 1966).

2.5.2.1 White rot fungi

The mycelia of the organisms can penetrate the cell cavity, and release ligninolytic enzymes to decompose all major wood components cellulose, hemicelluloses and lignin xylon to a white sponge-like and soft substance (Gao *et al.*, 2010; Worrall *et al.,* 1997). White rot produce three types of extracellular enzymes often referred to as lignin modifying enzymes (Gao *et al.*, 2010; Bending *et al*, 2002). They are lignin peroxidase (LiP), manganesedependent peroxidase (MnP) and laccase (Lac). Since white rot fungi cannot utilize lignin as an energy source, it is assumed that they degrade it for access to

cellulose in the cell wall (Gao *et al.*, 2010). Wood affected by white rot normally does not crack across the grain and will only shrink and collapse when severely degraded. The strength of the infested wood decreases gradually until it becomes spongy to the touch and stringy when broken.

2.5.2.2 Brown rot fungi

Brown rot fungi is one of the most common and destructive types of decay in wooden structures (Diouf *et al.,* 2002). It primarily decays the cellulose and hemicelluloses in wood but lignin, the substance which holds the cells together is only degraded to a limited extent (Ann-Christine, 1996; Worrall *et al.,* 1997). Wood affected by brown rot is usually dry and fragile, readily crumbles into cubes because of longitudinal and transverse cracks tending to crack across the grain. At initial stage of decay, even before any external evidence of decay can be seen brown rot-fungi cause extensive changes in wood cell wall structure causing rapid decline in strength properties (Ann-Christine, 1996, Diouf *et al.,* 2002). Brown rot fungi are different from other cellulolytic fungi because they lack enzyme activities needed for the enzymatic degradation of crystalline cellulose (Ann-Christine, 1996; Silva *et al.,* 2007).

2.5.2.3 Soft rot fungi

Soft rot is the most recently described type of wood decay and has proven difficult to define and differentiate from other decays (Worrall *et al.,* 1997). It attacks wood mainly near the surface. Their hyphae exclusively can develop within the secondary walls of

wood cells, where they enzymatically form chains of cylindrical cavities parallel with the microfibrils (Scheffer and Cowling, 1966).

They degrade only the cellulose and hemicelluloses, and typically occur in wood of high moisture content and high nitrogen content. Soft rot fungi look like brown rot. They are most commonly found in rotting window frames, wet floor boards and fence posts, etc., where nitrogen is recruited from soil or from atmospheric contamination. Some of these fungi are common decomposers of cellulose in soil and they are the least specialized of the wood decaying fungi. Soft rot may degrade all cell constituents but usually prefers carbohydrates, especially in hardwoods (Worrall *et al.,* 1997).

Soft-rot fungi have a greater tolerance for low oxygen concentration than basidiomycete decay fungi, and as a result they can decay wood that is saturated with water (Nicholas, 2003).

2.5.2.4 Molds (Non wood decaying fungi)

Mold and fungus stains occur mostly in sapwood, where the fungi obtain food primarily from the contents of xylem parenchyma cells (Scheffer and Cowling, 1966). Mold fungi discolor the wood by developing pigmented spores on the surface as well as causing shallow discolorations within the wood (Scheffer and Cowling, 1966). Staining fungi usually discolor wood both on the surface and internally and cause significant economic loss to wood industries worldwide (Scheffer and Cowling, 1966; Velmurugan *et al.,* 2009). In the case of blue stain or sap stain the discoloration is caused by pigmented hyphae that grow in the wood.

Other staining fungi discolor wood by secreting chromogenic substances (Scheffer and Cowling, 1966). Both mold and fungus stains are caused by ascomycetes and fungi imperfecti. A number of species of staining fungi grow in living trees. When exposed for long period to highly favorable conditions, most molds and staining fungi can cause soft rot (Scheffer and Cowling, 1966).

2.6 Methods of detecting and measuring wood decay

Presently, lack of accurate, rapid, non-destructive methods for detecting and quantifying wood decay is a major deterrent to wood preservation research (Nicholas and Crawford, 2003; Stephan *et al.,* 2000). It will be necessary to develop improved methods for measuring the extent of wood decay in test samples before accelerated test methods can become a reality (Nicholas and Crawford, 2003; Clausen, 1997). Some techniques are designed to detect viable decay fungi while others detect residual nonviable fungal metabolizes (Clausen, 1997) and all of them require some specific skill for interpretation (Clausen, 1997).

2.6.1 Visual Observation

At present, visual observation is the method normally used to evaluate outdoor wood decay test specimens (Nicholas and Crawford, 2003; Clausen, 1997), however was not applied to the current study. This method is subjective, rather than quantitative, does not detect early decay and consequently does not provide the information needed for accelerated testing (Nicholas and Crawford, 2003).

2.6.2 Mass Loss

During the process of decay, fungi utilize wood as a carbon source and produce CO_2 and water as by-products leading to gradual wood mass loss (Nicholas and Crawford, 2003; Clausen, 1997).

Mass loss can be measured at specific times or gradually by determining the weight of test specimens and comparing this with the original weight of the specimens. This method is widely used in laboratory soil block decay tests and provides reasonably good results.

Limitations of this method are associated with difficulty in making adjustments for variation in wood moisture content, loss of wood preservatives and inability to make adjustments for the weight gain due to colonization by the fungus (Nicholas and Crawford, 2003; Stephan *et al.,* 2000).

Because of these problems, the use of mass loss as a method for detecting decay is essentially limited to laboratory soil block tests. When used for this application, the main weakness of this test is its inability to detect the early stages of decay (Nicholas and Crawford, 2003; Acker *et al.,* 2003). It is generally admitted that mass losses of less than 2 to 3% are statistically not significant because of the inherent inconsistency of this method (Nicholas and Crawford, 2003; Acker *et al.,* 2003). Even small mass losses in the early stages of decay are associated with significant strength losses, especially when the wood is colonized by brown-rot fungi (Nicholas and Crawford, 2003; Acker *et al.,* 2003). For example, it has been shown that the compression strength of wood has been reduced

by at least 20% by the time a mass loss of 2 to 3% has been attained (Nicholas and Crawford, 2003).

2.6.3 Mechanical Properties

The mechanical properties of wood are adversely affected by wood decay fungi (Nicholas and Crawford, 2003; Clausen, 1997). The continuous measurement of selected strength properties could be used to monitor the wood decay process.

There are many properties that can be measured but the most suitable for detecting and measuring wood decay in laboratory and field tests are compression, bending, and torsion strength (Nicholas and Crawford, 2003; Machek *et al.,* 2001). Recent studies have shown that the extent of wood decay can be quantified by measuring the compression strength of wood wafers in the radial direction. This method is based on the comparative compression strength at the 5% level of end matched wafers, one of which is exposed to the fungus in a soil block test and the other serving as a non-exposed control. The use of thin wafers (5 mm in the longitudinal direction) and compression strength analysis has made it possible to carry out tests in approximately 6 weeks, compared to approximately 16 weeks necessary for the standard soil block test utilizing mass loss (Nicholas and Crawford, 2003; Machek *et al.,* 2001; Sexton *et al.,* 1993).

Other studies have indicated that it may be advantageous to use modulus of elasticity (MOE) rather than 5% compression strength as a measure of wood decay since use of MOE results in less variability among samples and appears to provide a better estimate of

toxic threshold values (Nicholas and Crawford, 2003; Machek *et al.,* 2001). The torsion strength of wood is often used to measure the shear strength. Torsion shear might be used to measure the extent of decay in test specimens exposed to wood decay fungi (Nicholas and Crawford, 2003; Stephan *et al.,* 2000). Wood toughness is another property suitable for assessing early stages of fungal attack (Sexton *et al.,* 1993).

2.6.4 Permeability

The measurement of permeability could be used to detect incipient decay (Nicholas and Crawford, 2003; Sexton *et al.,* 1993). This could be done by measuring the uptake of decalin when the samples are dipped in this low viscosity liquid (Nicholas and Crawford, 2003; Machek *et al.,* 2001). This method has not been very successful because of great variability of wood permeability but if better methods of measuring the permeability of wood could be developed then this method might have some potential (Nicholas and Crawford, 2003 Clausen, 1997).

2.6.5 Immunodiagnosis

This method is very sensitive and has the potential to detect incipient stages of decay in aboveground test specimens (Nicholas and Crawford, 2003; Machek *et al.,* 2001). It entails use of polyclonal and monoclonal antibodies to detect the presence of wood decay fungi (Clausen, 1997). One disadvantage of this method is the inability to quantify the progressive development of wood decay (Nicholas and Crawford, 2003). As a result, it may be necessary to combine this method with methods such as bending stiffness for better evaluation of test specimens (Nicholas and Crawford, 2003; Clausen, 1997).

2.6.6 NIR Analysis

Near infrared (NIR) spectroscopy has been used recently for many applications that involve rapid analysis of various substrates (Nicholas and Crawford, 2003; Clausen, 1997). NIR may be useful in detecting and quantifying biodeterioration of wood and it has been established that there is a good correlation between the NIR analysis and mass loss of wood decayed by brown-rot fungi (Nicholas and Crawford, 2003; Clausen, 1997). There also appears to be a good relationship between NIR analysis and changes in compression strength of brown-rotted wood (Nicholas and Crawford, 2003; Clausen, 1997).

Up to now, only wood degraded by brown rot fungi has been investigated and additional studies with wood decayed by white-rot and soft-rot fungi are needed since these microorganisms are also associated with the wood degradation process (Nicholas and Crawford, 2003). It is also not known whether this method can detect the early stages of decay, so additional work is also needed in this area. The availability of lightweight, portable NIR instruments and the fact that these instruments can be fitted with fiber optic probes makes this potential method of decay detection particularly attractive (Nicholas and Crawford, 2003; Stephan *et al.,* 2000).

2.6.7 Soil bed test

In the evaluation of new wood preservatives, the soil bed test is normally used to provide initial information on the efficacy against wood decay fungi (Stephan *et al.,* 2000;

Nicholas and Crawford, 2003; Machek *et al.,* 2001). Both white-rot and brown-rot fungi are generally included in this evaluation.

The toxic threshold value is determined in this test by using test specimens that are treated with varying levels of the preservative in question (Nicholas and Crawford, 2003). In order to have a valid test, about 12 weeks exposure for brown-rot fungi is required. However, when softwoods are tested against white-rot fungi the exposure time need to be increased in order to achieve sufficient decay in the untreated specimens (Acker *et al.,* 2003; Nicholas and Crawford, 2003).

2.7 Wood destroying insects

The class insecta is a large and diverse group of organisms which has invaded both terrestrial and aquatic habitats (Nicholas, 1973). It is taxonomically subdivided into 26 orders of which 6 are important wood destroying insects and include: Isoptera (termites), Hemiptera (bugs), Coleoptera (beetles), Lepidoptera (moths and Butterflies), Diptera (flies) and Hymenoptera (bees, wasps and ants). Most of the economically important wood destroying insects is of the orders Coleoptera, Hymenoptera and Isoptera (Nicholas, 1973).

2.7.1 Termites

Termites belong to the class isoptera. They are soft bodied, pale in color, with mouth parts for biting and chewing and utilizing cellulose as food source (Verma *et al.,* 2009). They live in large colonies consisting of reproductive forms, sterile workers, soldiers and

immature individuals. The king and the queen are pigmented and fully developed winged adults whereas the sterile castes, the workers and the soldiers, are wingless and do not have eyes. The queen lays approximately 3000 eggs and lives up to 25 years (Verma *et al*., 2009). Workers are responsible for building the shelter tubes and collect food to feed the young and other members of the colony while the Soldier termites guard the colony and its occupants (Verma *et al.,* 2009). There are over 2800 described species of termites, with about 185 considered to be pests. Those that belong to the families Kalotermitidae (Neotermes), Hodotermitidae (Hodotermes, Anacanthotermes), Termitidae (Amitermes, Ancistrotermes,Cornitermes,Macrotermes,Microcerotermes, Microtermes, Odontotermes, Procornitermes, and Syntermes) and Rhinotermitidae (Coptotermes, Heterotermes, and Psammotermes) are known to cause great loss in agriculture (Verma *et al.,* 2009). Termites depend wholly on either dead or living wood, or the woody tissue of plants, decayed or not decayed (Verma *et al.,* 2009). Termites are useful in recycling woody and other plant material. They help to aerate soil. They are able to decompose cellulose, the main component of wood. Termites are very many in tropical and subtropical environments and they play a key role in the functioning of the ecosystem (Jouquet *et al.,* 2011; Verma *et al.,* 2009).

Studies by Jouquet *et al.,* (2005) found that *Odontotermes and Ancistrotermes* termites had strong influence on the microbial structure of soil bacterial and fungal communities in the open savanna. Subterranean termites are considered economic pests worldwide because they destroy wood and wooden products, building materials, forests, and other commercial products and hence they are recognized as one of the most serious problems

in wood utilization (Lin *et al.,* 2009; Verma *et al.,* 2009; Scholz *et al.,* 2010; Sonowal and Gogoi, 2010). For instance, annual combined damage from Eastern and Formosan subterranean termites exceeds 2 billion US dollars in the United States (Clausen and Yang, 2007). Termites of the genus *Coptotermes* have been shown to occupy healthy living trees with rotten cores in Amazonian rainforests (Apolinario and Martius, 2004).

Some woods are resistant to termite attack due to the presence of some active component as part of their natural defense (Verma *et al.,* 2009). Extractives in termite-resistant woods and other plants are known to affect subterranean termites. The exact mechanism(s) by which extractives repel, deter or kill termites is unclear, but some researchers have assumed that extractives have some termite toxicity and/or repellency properties (Ragon *et al.,* 2008). Relative to commercial insecticides, the activity of most natural extractives against termites is generally low. Extractives often have antioxidant properties whose effect on termites may be due to coincidence, evolutionary design, or unknown factors (Ragon *et al.,* 2008). Antioxidants may interfere with lignocellulosic digestion by the termites and their symbiotic microbes thus, termites will avoid wood that contains some negligible level or more of antioxidant compounds, whether the antioxidants are natural extractives or man-made, and toxic or gentle (Ragon *et al.,* 2008). It may not be exclusively the amount and toxicity of extractives in a heartwood sample that gives heartwood some termite resistance, but the extractives' double toxicity and antioxidant properties. In addition to toxicity and antioxidant activity extractives may have other properties, that could also affect termites, or two or more factors could act together, perhaps synergistically (Ragon *et al.,* 2008). Silva *et al.,* (2007) found that

heartwood of *C. echinata* (Brazil wood) was resistant to the attack of the dry-wood termite (*Cryptotermes brevis*).

2.7.2 Other insects

The powder post beetles (*Lyctus*) of the family Lyctidae constitute one of the most economically important groups of wood destroying insect and are surpassed in significance only by termites (Nicholas, 1973).

Lyctus beetles are extremely serious pests of dry, seasoned wood and often are associated to broad-leaved deciduous trees but heartwood is not attacked. The pests obtain food mainly from the starch and reserve food materials stored in the ray parenchyma. The larvae bore small tunnels approximately 1/16″ in diameter and as the insect attack repeatedly, the wood is reduced into a fine flourlike powder (Nicholas, 1973).

Bostirichid beetles the family bostrichidae attack mainly hardwoods in the same manner as *Lyctus*. An example is *xylobiops basilare* which is capable of attacking freshly cut and partially seasoned hardwood species (Nicholas, 1973).

The beetles of the family *Anobiidae* commonly referred to as ‘’death watch beetles’’ damage wood in a similar manner to that caused by *Lyctus.* However, depending on the species the anobiid powder post beetles attack hardwoods and softwood species as well as heartwood and sapwood. The anobiids have a life cycle of about 2 to 3 years and require wood moisture content of about 15% or more for viable attack (Nicholas, 1973; Forest Products Laboratory, 1999).Vulnerability to anobiid invasion can be avoided by

reducing the moisture content of wood through improved ventilation and the sensible use of insulation and vapor barriers (Nicholas, 1973; Forest Products Laboratory, 1999). When choosing hardwood lumber for building or manufacturing purposes, any evidence of powder-post invasion should not be ignored, because the beetles may continue to be active long after the wood is put to use (Forest Products Laboratory, 1999).

The pinhole insects commonly referred to as ambrosia beetles or bark beetles consists of two families, scolytidae and Platypodidae. They are named ambrosia beetles because they cultivate and feed on fungi which they introduce into their covered passages which are usually excavated across the grain of the wood and are kept clean of boring dust. The beetles derive less or no nourishment from the wood itself. Damage by ambrosia beetles can be prevented in freshly sawn lumber by dipping the product in an approved chemical solution (Forest Products Laboratory, 1999).

Beetles in the family Bostrichidae and weevils in the family Curculionidae are associated with wood moisture contents favorable for wood-infesting fungi because they may benefit nutritionally from the fungi (Forest Products Laboratory, 1999). Therefore, the same procedures for protecting wood against wood decay fungi apply to protection against attack by these insects (Forest Products Laboratory, 1999).

2.8 Marine borers

Marine borers are animals that attack wood in marine environment and include those marine invertebrates which bore into and damage timber in saline water (Nicholas, 1973).

They are widely distributed throughout the world and mainly destructive in warm water regions (Nicholas, 1973).

Borer infestation occurs immediately there is enough supply of oxygen. Wood that has been attacked by marine borers may or may not show visible damage (Nicholas, 1973). Limnoria, burrow below the surface of the wood and produce a lacework that is readily noticeable while teredinids or shipworms enter through a tiny hole on the surface and bore into the wood as they grow and they can destroy the interior load without any visible damage on the surface, causing sudden failures of piers (Nicholas, 1973).

The animal organisms responsible for the biological deterioration of wooden structures are grouped into two, which are the phylum Mollusca and the phylum Crustacean (Nicholas, 1973). The phylum Mollusca are responsible for the wood destruction in the marine environment, *Teredo* and *Bankia* of the family teredinidae and the pholads *Martesia* and *Xylophaga* being the particular genera involved. They are oyster or clamlike in appearance in their larval stages and they metamorphose into wormlike animals as they bore into the wood (Nicholas, 1973). The Crustaceans borers look like a saw bug. The genus *Limnoria* is the most common wood borers of this class and they burrow below the surface of the wood and form a series of tunnels (Nicholas, 1973).

All woods tested for resistance to marine boring organisms have been damaged to some extent and most resistant woods have shown variable resistance in different harbors (Nicholas, 1973). It has been reported that timbers with high content of silica are more resistant to marine borers than woods of low silica content (Nicholas, 1973). Some

woods, however, such as Greenheart and *Lignum vitae* have low silica content and their resistance has been attributed to the presence of alkaloid content (Nicholas, 1973).

The service life of wooden structure immersed in marine environment depend on the time of immersion, types and abundant of borer species and the environmental conditions (Singh and Sasekumar, 1996). For example deterioration of wooden structures is greater in tropical than in temperate waters (Sirmah *et al.,* 2009).

2.9 Wood physical properties

2.9.1 Moisture content

Moisture content of wood is defined as the weight of water in wood expressed as a fraction, generally a percentage, of the weight of oven dry wood (Forest Products Laboratory, 1999). Wood properties such as weight, shrinkage, strength, depend on its moisture content. Moisture content (mc) in trees can range from about 30% to more than 200% of the weight of wood substance (Forest Products Laboratory, 1999). Sapwood usually has higher moisture content than heartwood in softwoods while in hardwoods, the difference in moisture content between heartwood and sapwood depends on the species (Forest Products Laboratory 1999).Variability of moisture content exists even within individual boards cut from the same tree (Forest Products Laboratory 999).

2.9.2 Wood density and specific gravity

Density is the mass per unit volume. It is a measure of the quantity of cell wall material contained in a specific volume of a piece of wood (Grabner *et al.,* 2005; Usta, 2003;

Kollmann and Côté, 1984). It is generally assumed that the quality of wood as a building material depends mainly on its density (Kollmann and Côté, 1984).

There is a close correlation between mechanical properties, hardness, abrasion resistance, and heat value of wood on the one hand and density on the other (Panshin and De Zeew, 1980; Kollmann and Côté, 1984; Grabner *et al.,* 2005). Since density is influenced to a large extent by the moisture content of the wood, comparisons of density figures can only be made at the same moisture content (Kollmann and Côté, 1984; Usta, 2003). Therefore wood density is expressed as green, oven dry or air dry depending on the moisture content (Usta, 2003).

Wood is a porous material composed of cell wall substances and cavities containing air and extractives (Kolmmann and Côté 1984; Tsoumis, 1991). Without cavities and intercellular spaces the relative density of the cell wall materials is basically constant for all timbers with a specific gravity of 1.53 on an oven dry mass and volume basis (Usta, 2003). The specific gravity of 1.53 is an ideal physical value for a lignified cellulosic cell wall which is completely nonporous (Usta, 2003). However, wood is not comprised of 100% cell wall material because it contains air spaces in the cell lumens (Usta, 2003).

There are several methods of measuring density; the simplest is by weighing and finding its volume. If the specimen has regular dimensions and no cracks one can measure its length, width and thickness to calculate volume. For specimens of irregular shape, an immersion method is suitable. The method is suitable for determining the density of

pulpwood. The specimens should have the approximate shape of a disc with a diameter of 12 cm (4 ¾″) and a width of 2.5 cm (1″) (Kollmann and Côté, 1984). Another method of quickly determining the density of woods with a density lower than 1 g/cm^3 consist of measuring the proportion of a test specimen (1 sq. inch in cross section and 10″ long, marked into ten equal divisions of 1″) that is submerged when the specimen is floated upright in a cylinder filled with water.

An exact determination of density may be carried out by immersion in mercury (Kollmann and Côté, 1984). The use of radiation methods has in recent years become prominent for the measurement of material quality. According to Kollmann and Côté, (1984) the application of radiation methods (X-rays, beta rays and gamma radiation) have gained importance to wood technologist for rapid, accurate and non destructive physical measurements of wood properties such as moisture content and density.

2.9.3 Dimensional stability

Wood dimensional stability is the ability to resist changes in its dimensions under certain conditions. Wood is composed of three polymers namely cellulose, hemicelluloses and lignin that contain hydroxyl groups which attract moisture through hydrogen bonding (Sun *et al.,* 2010; Panday *et al.,* 2010; Ozmen, 2007). The changes in dimensions of wood are caused by variation in relative humidity (RH) when its moisture content is below fiber saturation point (FSP) (Sun *et al.,* 2010; Hernandez, 2007). Below fiber saturation, increase in moisture content cause fiber expansion where as above FSP the wood is dimensionally stable because water exists as free water in the void structure of

wood and fiber dimensions cannot be further changed by increasing moisture content (Forest Products Laboratory,1999; Ozmen, 2007).

Dry wood undergoes small changes in dimension with normal changes in relative humidity (RH), where a more humid air cause slight swelling while drier air cause slight shrinkage (Forest Products Laboratory, 1999). These dimensional changes that accompany shrinkage and swelling of wood are the major sources of both visual and structural problems in furniture industry (Ozmen, 2007; Zhang, 2003). Dimensional changes lead to cracking and twisting, shrinkage and swelling, and cause troubles in wood utilized for building and construction. In addition, too much moisture can result in fungal attack hence changes in color and rate of degradation.

To obtain long-term service life, wood must be well protected from dimensional changes (Sun *et al.,* 2010; Forest Products Laboratory, 1999). Dimensional stabilization refers to mechanical or chemical treatments or modifications that reduce the tendency of wood to shrink or swell with its associated changes in moisture content (Kollmann and Côté, 1984).

2.10 Methods of extractive removal from wood

2.10.1 Soxhlet extraction

Soxhlet extraction is the classical and one of the oldest methods that was first described in 1879. It is a versatile method that can be used to separate a single gram to many grams

with almost 100% recovery (Chemo, 2003). A solid specimen is put in a porous container (thimble) and condensed solvent is allowed to extract continuously (Chemo, 2003).

The basic components of soxhlet extractor include condenser for cooling solvent vapor, thimble (Porous container) which holds the solid specimen, allowing the condensed solvent to saturate and pass through thus extracting active material (Chemo, 2003). It also contains a distilling pot which holds the solvent pool and act as a reservoir for the concentrated material (Chemo, 2003).

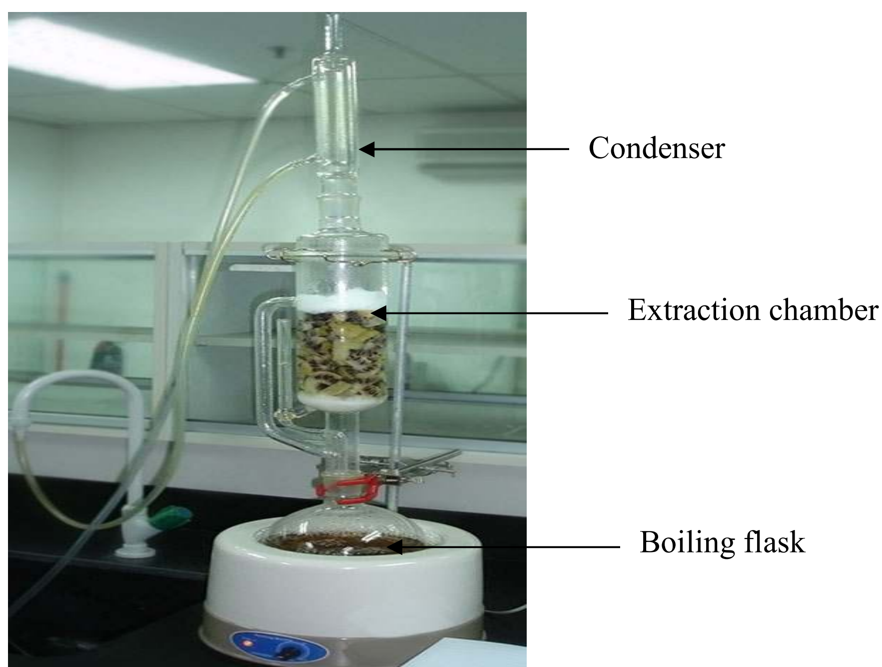


Plate 2: Soxhlet extractor (http://www.wikipedia.org/wiki/soxhlet_extractor)

2.10.2 fexIKA method of extraction

*fex*IKA is an advanced extraction method that Schwanninger and Hinterstoisser (2002) compared with the classical soxhlet extraction. The method operates in four phases of extraction cycle as shown in figure 7 (Schwanninger and Hinterstoisser, 2002)

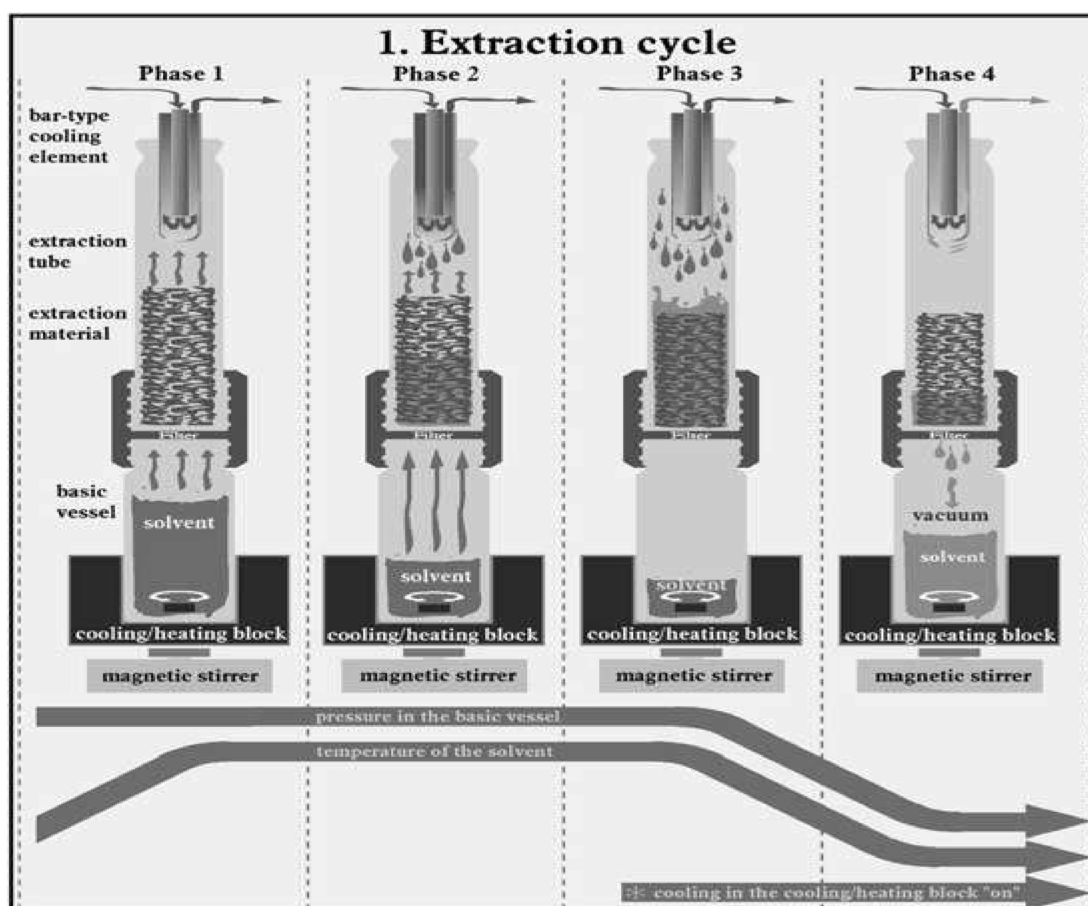


Figure 7: Extraction cycle of the *fex*IKA method (Schwanninger and Hinterstoisser, 2002).

In phase 1, the extraction material is fed into a prepared extraction tube. The basic vessel is filled with the solvent, a magnetic stirring rod is introduced and the basic vessel is

mounted with the extraction tube. The process is started after the experimental conditions (temperatures, number of cycles, filtration time) have been entered into the controller program (Schwanninger and Hinterstoisser, 2002).

In phase 2, the vapor of the boiling solvent penetrates the membrane filter, the extraction material and condenses on the bar-type cooling element. This successive continuous stream of solvent vapor serves to heat up and vigorously fluidize the extraction material/solvent mixture at boiling temperature. This fluidized bed technique makes extraction principally effective (Schwanninger and Hinterstoisser, 2002).

In phase 3 heating is switched off after the elapse of the heating period and stirring is continued. After the valve has been opened, the cooling liquid is directed through the cooling/heating block. This results in rapid cooling of the block, the basic vessel and its contents (Schwanninger and Hinterstoisser, 2002).

In phase 4 This cooling off generates a vacuum in the basic vessel and the resulting pressure difference with reference to the atmospheric pressure transmits the extractive solution through filter into the basic vessel. This cycle may be repeated any number of times. Therefore the procedure may be customized for all extraction conditions (Schwanninger and Hinterstoisser, 2002).

2.10.3 Accelerated solvent extraction

Accelerated solvent extraction (ASE) also known as Pressurized Fluid Extraction (PFE), Pressurized Liquid Extraction (PLE) or Enhanced Solvent Extraction (ESE) was first

described in 1995 (Giergielewicz-Mozajska *et al.,* 2001; Ramos *et al.,* 2002). The method has been shown to be valuable and a superior alternative to conventional methods such as soxhlet and soxtec or ultrasonic a method mainly for the separation of micro pollutants from (semi-) solid environmental media (Ramos *et al.,* 2002).

ASE (figure 8) consists of a stainless-steel cell where the specimen is placed at selected temperature and pressure during extraction, heaters that are electronically controlled, pumps for solvent delivery and a vial used for collecting the liquid extract(Ramos *et al.,* 2002).

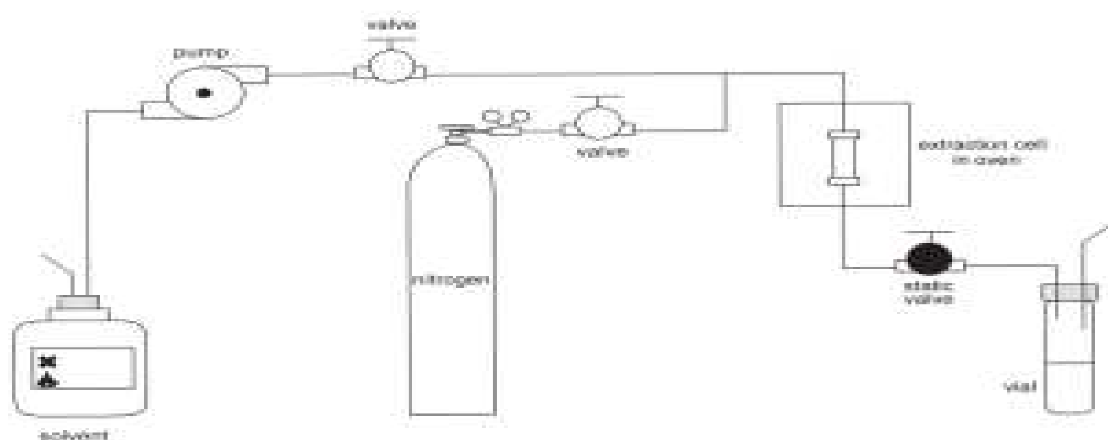


Figure 8: Diagram showing accelerated solvent extraction system (Giergielewicz-Mozajska *et al.,* 2001)

Until recently (2002), dionex (Sunnyvale, CA, USA) ASE 200 was the only PLE system available commercially (Ramos *et al.,* 2002). With this system a temperature of up to 200 °C and pressures up to 21MPa in extraction cells of 1, 5, 22 or 33ml can be reached and

up to 24 specimens can be put in carrousel and the extracts collected in 26 vials of 40 or 60 ml since four extra vials are for rinsing (Ramos *et al.,* 2002).

ASE 300 can be used for larger specimens whereby 12 specimens can be placed in the carrousel. In this model the same temperatures as with ASE 200 can be reached but only for pressures up to 10MPa. The model has cells of 34, 66 and 100 ml and 12 collection vials (plus one rinsing vial) of 250 ml (Ramos *et al.,* 2002).

In ASE system, the process of extraction is performed at temperatures that exceed the boiling point of a solvent thus the pressure inside the extraction cell must be kept high so as to keep the solvent in a liquid state and help to force the solvent into the matrix pores (Zhao *et al.,* 2012; Ramos *et al.,* 2002; Giergielewicz-Mozajska *et al.,* 2001). The extraction process consists of desorption from a solid particle, diffusion through the solvent located inside a particle pore, and transfer to the bulk of the flowing fluid steps. Each step depends on many factors, which can be varied through temperature and pressure modification (Giergielewicz-Mozajska *et al.,* 2001).

The specimen in the extraction cell is extracted with an organic solvent at a temperature that range from room temperature to 200 °C and a relatively high pressures ranging from 4 to 20 MPa (Ramos *et al.,* 2002). Raising the temperature increases the diffusion rates, the solubility of the analytes, but decreases the viscosity and surface tension of the solvents (Giergielewicz-Mozajska *et al.,* 2001; Ramos *et al.,* 2002). These changes improve the contact of the extraction which can be achieved more rapidly and with less solvent use compared with classical methods (Ramos *et al.,* 2002).

The ASE system can use a broad range of solvents except those having an auto ignition of 40 to 200 °C such as carbon disulfide, diethyl ether and 1,4 dioxane. Generally, strong acids and bases should not be used as solvents because they are corrosive (Giergielewicz-Mozajska *et al.,* 2001). The ASE system provides an opportunity for the use of a wide range of solvents, even those that are not effective in conventional methods. However, the use of solvents that are not used in conventional methods may lead to solubilization of matrix components, which would otherwise remain insoluble under conditions of conventional extraction (Giergielewicz-Mozajska *et al.,* 2001).

The extraction process in a typical ASE method consists of several stages. A weighed specimen is placed in the extraction cell and sometimes copper is also added to the cell to remove sulfur. The cell can be operated in a pre heat or pre fill mode. In the preheat mode, the oven is first heated up to the suitable temperature and then the cell is loaded into the oven. The solvent is introduced into the cell after a given time and the extraction process begins. In the pre fill mode, the cell is filled with the solvent then it is loaded into the oven. This mode allows the removal of interstitial air and prevents degradation of compounds that oxidize easily and hence recommended for thermally labile compounds (Giergielewicz-Mozajska *et al.,* 2001).

The process of extraction can be performed in a static or dynamic mode. The static process begins with heating the cell with the specimen to a suitable temperature during the equilibration time, lasting about 5 minutes and then it is followed by static extraction. The analytes are isolated from the sample under static conditions. If low recoveries are

obtained in a single stage, the process can be repeated many times. The dynamic mode of ASE can be compared with high-performance liquid Chromatography done at elevated temperatures, where the column packing is replaced by the specimen. Although this improves mass transfer, this type of extraction is hardly used mainly due to higher solvents consumed compared with static process (Giergielewicz-Mozajska *et al.,* 2001).

2.10.4 Subcritical water extraction

Subcritical water extraction (SWE), also called hot water extraction, pressurized (hot) water extraction or superheated water extraction, is an emerging method based on the use of water as an extraction solvent at temperatures between 100 and 374 °C (critical point of water, 374 °C and 22 MPa) and at a pressure high enough to keep it in liquid form (Ramos *et al.,* 2002). Under these conditions, the polarity of water can easily be lowered by increasing the temperature. Surface tension and viscosity of water is also reduced when the temperature is increased at moderate pressure, leading to improved solubility of the analytes in this solvent (Ramos *et al.,* 2002).

Provided water remains in liquid state, pressure has limited control on its solvent characteristics. An increase in pressure from 0.1 to 10MPa yields increase of dielectric constant (ε) of only 0.37 (Ramos *et al.,* 2002). SWE consists of two pumps, one for deoxygenated water and one for the selected organic solvent, an oven containing a stainless steel heating coil and the extraction cell, a stainless steel cooling coil and a vial for extract collection (Ramos *et al.,* 2002).

After filling the extraction cell with the specimen and placing it in the oven, the extraction typically begins with pumping both the water and the organic solvent at their selected flow-rates until the pressure selected for the SWE is reached. Extraction begins once the oven reaches the selected temperature for the SWE experiments (Ramos *et al.,* 2002). The water is heated by passing it through a heating coil before reaching the stainless-steel extraction cell and is constantly pumped through the specimen during extraction. The hot water containing the analytes is mixed with the organic solvents through a T-piece placed in the oven at the outlet of the cell (Ramos *et al.,* 2002). Before collecting in a vial, the mixture is passed through a cooling coil where the temperature decreases rapidly. Water becomes a polar solvent and the less polar analytes previously dissolved in SWE are partitioned to the less polar solvent preventing adsorption to the tubing (Ramos *et al.,* 2002). The organic layer is removed and concentrated after separation before further cleaning up and analysis (Ramos *et al.,* 2002).

Two types of continuous SWE are possible in addition to static SWE; namely the regular extraction, where the analytes are eluted at a selected temperature and pressure, and a sequential extraction. Programming the temperature may lead to selective removal of compound classes of different polarity (Ramos *et al.,* 2002).

It has been reported that the SWE has greater potential than SFE with CO_2 for selective analyte extraction because of the wide range of polarities that can be generated by changing the temperature of the subcritical (Ramos *et al.,* 2002).

CHAPTER THREE

3.0 MATERIALS AND METHODS

3.1 Tree sampling

3.1.1 Albizia malacophylla

From a household plantation in Teso Western Kenya, four mature *Albizia malacophylla* were randomly selected. The trees were felled, quarter sawn into boards measuring 1m long × 0.1m wide × 0.1m thick and transported to the Wood Science laboratory, University of Eldoret within twenty four hours. In the laboratory, boards free of knots and with no observable infection by mold, stain or wood rot fungi were sorted and air dried for a period of 14 days. The boards were then sawn into appropriate sizes for each experiment.

3.1.2 Pinus patula

Four 16-year old *Pinus patula* trees were randomly selected from Kaptagat forest plantation in Elgeyo Marakwet County, Kenya. The trees were felled and quarter sawn into boards measuring 1m × 0.1m × 0.1m (l × r × t) then transported to Wood Science Laboratory, University of Eldoret. They were then air conditioned for a further period of two weeks to lower wood moisture. Clear sapwood boards were then identified, and sawn into small appropriate specimens as per each experiment.

3.2 Wood dimensional stability

3.2.1 Specimen preparation

Forty specimens of both heartwood and sapwood measuring 20mm × 20mm × 20mm were cut. The dimensions of all the specimens were measured to the nearest 0.01mm using vernier caliper. Sixteen heartwood specimens were soxhlet extracted for 12 hrs at 6-10 cycles per hour, four using each of the different solvents; hexane, dichloromethane, acetone and water while four were not extracted. Similar procedure was applied for sapwood specimens and all other tests. The purpose of extraction was to test whether extractives influences wood dimensional stability.

3.2.2 Evaluation of dimensional stability

Volumes (V_0) of air dried wood specimens (extracted or not) were calculated prior to exposing them in desiccators containing hydrated copper (II) sulphate solution. The specimens were weighed after every 48 hours until constant mass was reached, indicating that they have attained maximum moisture content. The dimensions of the specimens were measured again and wet volume (V_1) determined. Swelling coefficient was calculated using the formula as shown in equation 1;

$$\% \text{ Swelling coefficient} = \frac{V_1 - V_0}{V_0} \quad \text{.. Equation 1}$$

The experiment was replicated four times.

3.3 Wood density

3.3.1 Specimen preparation

From heartwood and sapwood of *A. malacophylla*, fifty specimens were cut and measured using vernier caliper into dimensions of 20mm × 20mm × 20mm. Twenty heartwood specimens were soxhlet extracted for 12 hours at 6-10 cycles per hour, five each with each of the solvents; hexane, dichloromethane, acetone and water while five were not extracted. The same procedure was followed in extracting sapwood specimens.

3.3.2 Wood density determination

The density D (g/cm^3) was determined by first calculating the volume V (cm^3) of wood specimens that were previously extracted or not. The specimens were then weighed to the nearest 0.001g to find mass M (g) and the volume calculated using the formula as shown in equation 2;

$$\text{Density} = \frac{M}{V} \qquad \text{equation 2}$$

The experiment was replicated five times.

3.4 Wood ash content

3.4.1 Specimen preparation

Ash content was determined according to TAPPI T 211 (Ash in wood, pulp and paper board: combustion at 525 ° C - unpublished TAPPI standard).

Empty crucibles were carefully cleaned marked and placed in the muffle furnace at 525 ± 25 °C for 30-60 minutes, removed and cooled in the desiccator and weighed to the nearest 0.1 mg and the weight recorded.

This step was repeated until the weight of the crucible varied by less than 0.3mg from the previous weight. This final weight was recorded as the crucible tare weight.

3.4.2 Evaluation of ash

1.0g test specimens (wood chips of both sapwood and heartwood), previously dried at 103 ± 2 °C were weighed into the tared crucible and placed into the muffle furnace at about 100 °C.

The furnace temperature was then raised slowly to 525 ± 25 °C so that the specimens carbonized without flaming. After complete combustion of the specimens as indicated by the absence of black particles, the crucible was removed from the furnace its cover was replaced, and then placed in a desiccator containing anhydrous alumina to facilitate its cooling to room temperature.

The crucible with the ash was weighed to the nearest 0.1 mg. This process was repeated until the weight of the ash remained constant ± 0.2 mg. Ash content was calculated as shown in equation 3.

$$\text{Ash content } (\%) = \frac{A}{B} \times 100 \qquad \text{Equation 3}$$

Where; A = Weight of ash, (g) and B = Weight of test specimen, moisture free (g)

3.5 Anatomical description of heartwood

3.5.1 Preparation of wood specimens and sectioning

Heartwood specimens of *A. malacophylla* were cut into cubes measuring 1cm×1cm×1cm. The specimens were marked with a pencil then softened by boiling in water for one hour. Thin sections (of between 5-20µm) for microscopic examination were prepared by use of sliding microtome.

3.5.2 Preparation of permanent slides

The thin wood sections were stained for 20 minutes in safranin then washed in running water until the water became colorless. The sections were then dehydrated by first rinsing in 30% alcohol for 20 minutes, followed by 50% alcohol for 15 minutes then in 70% alcohol for 10 minutes. Rinsing was continued by further soaking in 85% alcohol for 7 minutes, 95% alcohol for 3minutes, and two times in absolute (100%) alcohol for 3 minutes each. The sections were finally rinsed two times in xylene for 3 minutes.

Dehydrated sections were then mounted on a Dpx mountant. Each section was mounted on a separate slide, covered with cover slips and labeled accordingly. Microscopic observation was then made at ×10, ×20 and ×40 magnifications and photomicrographs taken using Leica DMLB microscope fitted with EC3 camera.

3.6 Evaluation of extractives in wood

3.6.1 Wood chips preparation

Heartwood and sapwood boards of *A. Malacophylla* were separately reduced to chips (approximately 0.1mm diameter) in a wood lathe machine (plate 3). The chips were then separately dried at 60 °C to constant mass before solvent extraction.

Plate 3: Preparation of wood chips from *A. malacophylla* heartwood and sapwood (Author, 2011)

3.6.2 Soxhlet extraction

Both batch and series extraction was performed in a soxhlet extractor. In the batch extraction, 5g of wood chips (both heartwood and sapwood) were extracted separately using 150ml of hexane dichloromethane, acetone and water for 12 hours at a rate of 6-10 cycles per hour. Each experiment was replicated three times.

In series extraction, (5g of wood chips) was extracted, initially in less polar solvent (hexane) under the previous conditions (12hrs and 6-10 cycles per hour) then followed by dichloromethane, acetone and finally water. At each stage of extraction, percentage yield of extractives was evaluated.

3.6.3 Cold soaking extraction

10g of wood chips were separately soaked in bottles containing 150ml of different solvents (hexane, water, acetone and dichloromethane) for 3, 10 and 14 days. Soaking was carried out at room temperature (22 °C to 25 °C) and bottles shaken regularly. After each extraction period, the chips were sieved; the mixture (extract plus the solvent) poured into a round bottomed flask. Solvent was then evaporated to dryness in a rotary evaporator and the amount of extractives determined as follows;

$$(\%)\text{Extractive} = \frac{W_T - W_F}{W_D} \times 100 \quad \text{.......... Equation 4}$$

Where W_T = total weight of the flask plus the extract, W_F =Weight of empty flask and W_D= weight of dry chips before extraction.

The experiment was replicated three times.

Plate 4: Extraction of *A. Malacophylla* wood chips by cold soaking method (Author, 2012)

3.6.4 Evaluation of total extractives

Before extraction was started, the distilling pot (soxhlet round bottomed flask) was cleaned, oven dried and its weight determined (W_F). After extraction, the solvent was evaporated to dryness using rotary evaporator, then the flask plus the extract weight (W_T). The percentage yield of the extract was determined gravimetrically in relation to the mass of the dry chips (W_D) using the formula shown in equation 5

$$(\%)\ \text{Extractive} = \frac{W_T - W_F}{W_D} \times 100 \qquad \text{Equation 5}$$

Plate 5: Rotary evaporator (Author, 2012)

3.7 Assessment of wood decay against fungi in a soil-bed tests

This study was based on a modified AWPA: E23-07 (American Wood Protection Association) test standard.

3.7.1 Preparation of test specimens

Thirty two specimens of *A. malacophylla* heartwood measuring 50mm × 10mm × 5mm, (l × r × t) were soxhlet extracted in each of the following solvents; hexane, dichloromethane, acetone and water solvents while thirty two were not extracted. Thirty two specimens of *Pinus patula* sapwood and not solvent extracted were cut to the same

dimensions as above and served as controls. All the specimens were conditioned at 60 °C in an oven to a constant mass before they were subjected to decay test.

3.7.2 Soil bed preparation

Containers measuring 250mm × 150mm× 170mm (l × w × h) were filled with different types of unsterile soil layers (bottom to top) as follows; 20mm of gravel, 20mm of sand and 130mm of forest soil.

The forest soil was collected from Kaptagat Forest in Elgeyo Marakwet County, Kenya. Soil moisture was determined by randomly removing 3g of the soil, its wet weight recorded and then oven dried until constant mass is attained giving oven dry weight. The moisture content was then determined directly from wet and oven dry weights using the formula shown in equation 6:

$$\text{Soil mc } (\%) = \frac{S_w - S_d}{S_d} \times 100 \quad \text{......Equation 6}$$

Where, mc is moisture content, S_w is mass of wet soil and S_d is the oven dry mass of the soil.

The moisture content of the soil was thereafter maintained by weighing the containers weekly and sufficient water added to bring the weight of the container back to the original weight that gave the required moisture content.

3.7.3 Planting of test specimens in the soil bed

Un-extracted, extracted heartwood specimens of *A. malacophylla* and *Pinus patula* controls were exposed to the unsterile soil-bed under room temperature (22 °C to 25 °C) and controlled moisture that optimize wood decay of about 70% moisture content.

The specimens were randomly distributed 20mm apart, labeled for identification purposes and then buried so that approximately 1/3 of each piece protruded above the soil bed as shown in Plate 6.

Wood specimen

Plate 6: Soil bed test (Author, 2012)

3.7.4 Assessment of wood decay

Wood decay was assessed by evaluating the mass losses once every four weeks for 32 weeks. During each inspection a replicate set of 4 specimens was removed from the soil-bed cleaned from any mycelium and soil which adhered to the surface. The specimens were then oven-dried at 102 ± 3 °C for 48 hours and the mass loss was determined gravimetrically. The mass loss of each test specimen was expressed as a percentage of the original mass before exposure using the formula shown in equation 7;

$$\text{Mass loss, \%} = \frac{W_0 - W_1}{W_0} \times 100 \quad \text{.. Equation 7}$$

Where, W_o = weight before exposure to decay and W_1 = weight after exposure to decay

3.8 Assessment of wood durability against termites in a field test

Wood decay resistance against termites was evaluated according to American Wood Protection Association, AWPA: E7-1993 standard method of determining resistance to subterranean termites.

3.8.1 Preparation of wood specimens

Soxhlet extraction was done on ninety six heartwood specimens of *A. malacophylla* measuring 100mm × 10mm × 5mm (l× r × t), twenty four in each of the following solvents; dichloromethane, hexane, acetone, and water. In addition to this, there were twenty four non solvent extracted heartwood specimens and twenty four *Pinus patula* sapwood were prepared totaling one hundred and forty four specimens. Before exposure, all the specimens were dried in an oven at 60 °C to nearly constant mass.

3.8.2 Test site preparation

Termite nests were arbitrarily selected at Cheptebo, Kerio valley in Elgeyo Marakwet County, Kenya. Before the specimens were exposed to the termites, the place was cleared of plant life and any other cellulosic material.

3.8.3 Exposure of test specimens

The test specimens were exposed to termite attack in a randomized design at a distance of 30 cm from the termite nest with two thirds of their lengths buried in the soil and a spacing of 20cm apart as shown in plate 7;

Plate 7: Termite field test layout at Cheptebo (Author, 2012)

Every one month, four specimens per treatment were removed gently from the soil to minimize soil disturbance, and soil on the surface of the specimens brushed off.

The specimens were then dried until they attained constant weights after which the extent of attack was evaluated by mass loss using the formula shown in equation 8

$$\% \text{ Mass loss} = \frac{W_i - W_f}{W_i} \times 100 \quad \text{……………………………………Equation 8}$$

Where W_f = final weight after termite attack and W_i = initial weight before exposure.

3.9 Contribution of heartwood extractives to natural durability

To understand how heartwood extractives of *A. malacophylla* contribute to resistance of wood to decay, fungal mycelia were grown in 9-cm petri dishes filled with 20 ml of potato dextrose agar (PDA) medium containing 0, 50 and 250 ppm of extractives. Control dishes were not treated with the extractives. Plates were inoculated at the center by placing a10 mm diameter fungi cut from the edge of an initially growing colony of fungus on potato dextrose agar medium.

The cultures were left incubated at room temperature for a period of 9 days. Fungal growth was evaluated daily by measuring two vertical diameters of the colony. Each experiment was replicated three times. Growth was expressed as a percentage of the available space for growth using the formula shown in equation 9.

$$\text{Growth inhibition, \%} = 100 \times \left(1 - \frac{d_1}{d_0}\right)$$ ……………Equation 9 (Gerardin *et al*., 2004)

Where d_0=diameter of the untreated culture (i.e. the control) and d_1= diameter of the culture with the extracts

3.10 Characterization of heartwood extractives

3.10.1 FTIR analysis

1µg of extract was mixed with a spatula end full of KBr, grinded evenly in a mortor and pressed into disks. This was then examined in FTIR wavelength ranges of between 4000-500 cm^{-1}

3.10.2 GC-MS analysis

1 µl of the wood extractive was injected into konik mass spectrum at konik 4 RGC 4000B (GCMS). The operating conditions were as follows: The GC was equipped with a TRB-5 capillary column 15mm×0.25mm (ID) × 0.25mm composed of 5% diphenyl, 95% dimethyl polysiloxane. The oven temperature was programmed from 60 °C with an increase of 8 ^{0}C/min to 150 °C and held at this temperature for 2 minutes followed by a 10 °C/min temperature rise to 250 °C where again the temperature was held for 4 minutes.Helium was used as the carrier gas with a constant flow rate of 1ml/min. The injection was operated at 200 °C, mass spectra was obtained at 70 eV scanning over the 40-50 m/z, the range employing a scan time of 0.5 seconds with a dwell time of 1215 µseconds. Interpretation of the chromatograph was carried out by comparing the

fragmentation pattern corresponding to the individual peak of interest with that present in NSIT database/ library. Peak areas were used to express the percentage composition of the individual components identified from the various extracts.

3.11 Data analysis

Data was analyzed using statistica Version 7 for windows. The soxhlet extraction from the specimen using various solvents were analyzed by using Analysis of variance (ANOVA), while pair wise comparison for dimensional stability and density was used to test for equality of means using two sample independent t-test. The percentage mass loss of the different specimens was compared at different decay period for the various treatments using two-way ANOVA. The analysis assumes all means are from the same population.

CHAPTER FOUR

4.0 RESULTS AND DISCUSSION

4.1 Wood density and dimensional stability of *A. malacophylla*

Table 1 shows wood density and dimensional stability of *A. malacophylla* and effects of solvent extraction on these properties.

Table 1: Density and dimensional stability of *A. malacophylla* wood (Author, 2012)

Specimen	*Density (g/cm³)		Dimensional stability (%)	
	Heartwood ± Sd	Sapwood ±Sd	Heartwood ±Sd	Sapwood ±Sd
Un-extracted	1.01 ± 0.11	0.84 ± 0.21	5.52±0.50	9.6±0.80
Hexane	0.95 ± 0.09	0.81 ± 0.10	5.53±0.44	9.61±0.32
Dichloromethane	0.94 ± 0.08	0.79 ±0.13	5.58±0.12	9.65±0.26
Acetone	0.91 ± 0.13	0.76 ± 0.07	6.21±0.25	9.98±0.11
Water	0.89 ± 0.06	0.75± 0.13	6.40± 0.09	10.14±0.20

*Density at 12% moisture content

A. malacophylla heartwood has a density of about (1.01 g/cm³) in comparison with a sapwood density of (0.84g/cm³). Reported wood density of other *Albizia* species range

from 561.55kg/m^3 to 705.45 kg/m^3(Kukakchka, 1969). Mohmod *et al., (*1993) and Zhang, (2003) have reported that wood density is proportional to high wood strength.

Thus, the high density reported suggests that *A. malacophylla* wood is very strong and durable. The density of both heartwood and sapwood decreased proportionately after solvent extraction and removal of extractives. In heartwood, the highest drop in density was observed in water extracted specimens (mean difference of about 0.13g/cm^3) while hexane extracted specimens showed the least drop in density (mean difference of about 0.07 g/cm^3). Similar trend was observed in sapwood specimens. The results showed that presence of extractives influences wood density. Two sample independent t-test showed that density of water extracted heartwood specimens were significantly different from the un-extracted ($p=0.034132$) while those specimens that were extracted using hexane, dichloromethane and acetone were not significantly different ($p>0.05$). On the other hand, there was no significant difference in density of solvent extracted and un-extracted sapwood specimens ($p>0.05$). Studies by Singleton *et al.,* (2003) on effects of extractives on wood density of western Hemlock observed that indeed removal of extractives reduces the wood density.

A. malacophylla heartwood is dimensionally stable (5.52%) with less dimensionally stable sapwood (9.60%). The results indicated a significant difference between the dimensional stability of the heartwood and sapwood ($p = 0.000049$). Literature studies have attributed the dimensional stability of heartwood from other wood species to the presence of high extractive content (Kose and Taylor, 2012). To test whether extractives

influences dimensional stability, wood specimens were extracted using different solvents. Results showed that extracting wood using hexane had virtually no effect on the dimensional stability (5.53%). Two-sample t-test indicated that un-extracted heart wood and hexane or dichloromethane extracted heartwood were not significantly different ($p > 0.05$). However, extracting wood using water and acetone had significant effect on dimensional stability ($p < 0.05$).

It can be concluded from the study that more polar solvents such as water and acetone extraction has more influence on dimensional stability of *A. malacophylla.* This is in agreement with literature studies showing that polar solvents remove extractives located in the cell wall. Such extractives are known to bind to the polymeric cell wall by means of multiple hydrogen bonds making the wood dimensionally stable (Royer *et al.,* 2010). The high dimensional stability of *A. malacophylla* reported, indicates strong hydrophic nature of this wood and may be one of the factors contributing to its natural resistance. This is in agreement with observation by Neya *et al.,* (2004) who reported strong hydrophobic nature on *Burkea africana* wood.

4.2 Amount of wood ash

Wood ash content generated ranged from 2.5% in sapwood to 3.1% in heartwood. The amount of ash is quite high but within the range found in some tropical woods (Browning, 1975).

4.3 Wood anatomical characteristics

4.3.1 Gross wood features

The sapwood of *A. malacophylla* is white to yellow in color, about 5cm wide and clearly distinct from the dark brown heartwood (Plate 8). The wood is hard, compact with a coarse texture, wavy grains and characteristic chocking smell when fresh. The growth rings are distinct and the wood is difficult to split longitudinally.

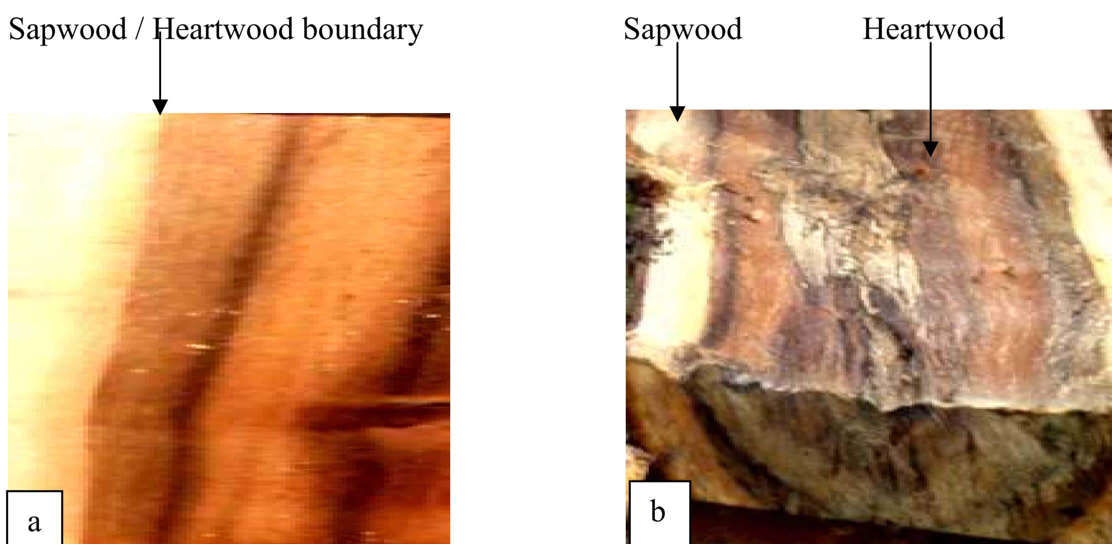


Plate 8: Gross wood features of *A. malacophylla* (a) sapwood and heartwood delimitation, (b) halfed portion of the stem (Author, 2012)

4.3.2 Microscopic features

Plate 9 is a photomicrograph of transverse, tangential and radial sections of *A. malacophylla* heartwood. The vessels are exclusively solitary and the fibers are thin-to-thick walled. The axial parenchyma showed vascicentric distribution pattern under the

microscope. Gums and other deposits are present in the vessels. Apetorgbor (2007) reported similar microscopic features in *Albizia zyga.* Tangential observation showed rays of 1 to 3 cells wide, two cells per parenchyma strands and presence of prismatic crystals in long chains. Radially, it was observed that all ray cells are procumbent. The prismatic crystals appeared in radial alignment in the heartwood and as short chains in sapwood.

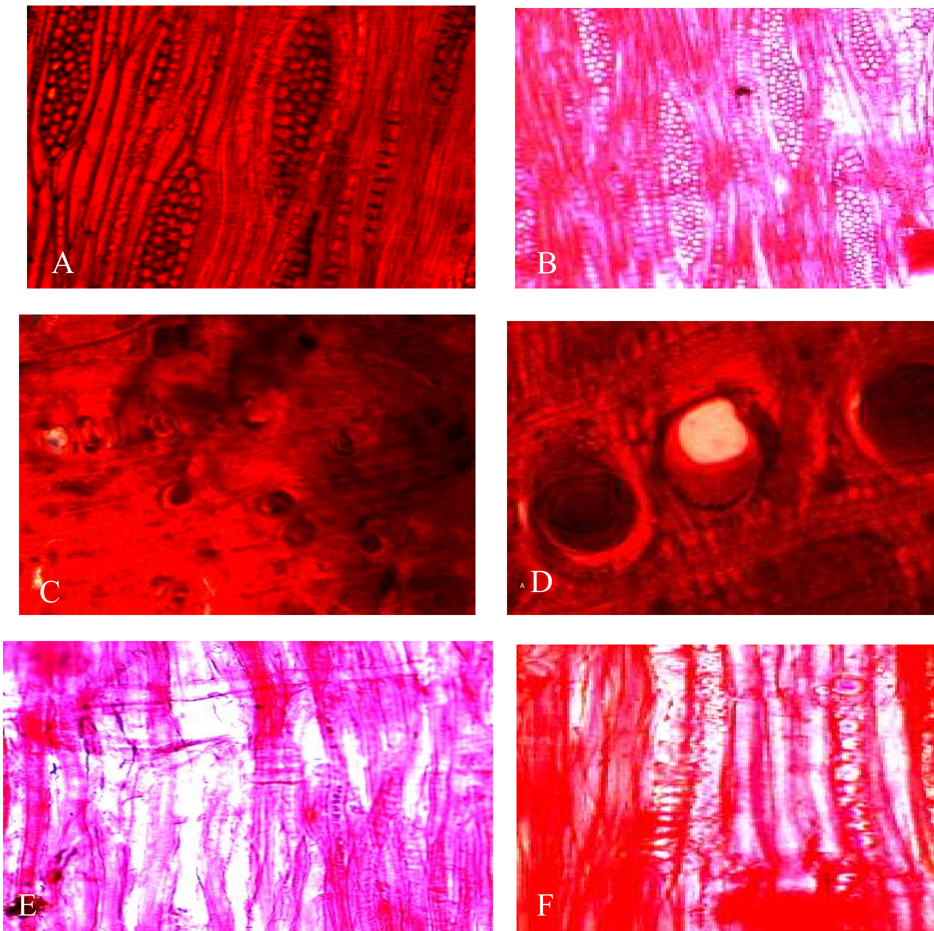


Plate 9: Photomicrograph of *A. malacophylla* heartwood (A) tangential section ×10 (B) tangential section ×20 and (C) cross section. (D) Showing vessels (E) radial section ×20 and (F) radial section × 10 (Author, 2012)

Presence of gums and other deposits in the heartwood vessels and thick-walled fibres are features consistent with good natural durability and strength respectively (Junior *et al.,* 2009; Mohmod *et al.,* 1993).

4.4 Determination of extractive content

4.4.1 Amount of wood extractives by soxhlet extraction

The mean percentage yield of extractives in the sapwood and heartwood of *A. malacophylla* by soxhlet extraction are presented in table 2:

Table 2. Mean percent yield of extractives from *A. malcophylla* wood (Author, 2012)

Solvent	Batch extraction (%)		Series extraction (%)	
	Heartwood ±Sd	Sapwood ±Sd	Heartwood ±Sd	Sapwood ±Sd
Hexane	2.36± 0.08	2.27± 0.13	2.35±0.09	2.20± 0.25
Dichloromethane	4.24±0.64	2.68±0.31	3.16±0.12	2.81±0.18
Acetone	5.23±0.71	3.38±0.82	4.62±0.41	3.12±0.56
Water	9.71±0.5	3.95±1.2	9.62±0.5	4.6±0.31

Sd=Standard deviation

Batch extraction of heartwood using hexane gave a relatively small amount of extractives (2.36%) which increased up to 9.71% yield in water extraction. Similar trend of extractive yield was observed in sapwood with a reported yield of 3.95% in water extract. Generally, *A.malacophylla* heartwood has a higher quantity of extractives than sapwood which are significantly different ($p = 0.000347$). This is in agreement with literature

information reporting that high levels of extractives are located in heartwood (Kollmann and Côtè, 1984; Sirmah *et al.,* 2009; Hashemi and Latiberi, 2011).

Hexane extractions showed similar amount of extractives both in heartwood and sapwood that were not significantly different (P>0.05). The results also revealed that the effect of the process used for extraction (batch or series) was not significant (p>0.05). Studies by Golpyegani *et al.,* (2012), on variability of extractives by different solvents on white mulberry (*Morus alba* L.) found similar results.

When a more polar solvent e.g. dichloromethane was used in series extraction it removed a relatively small amount of extractives suggesting that some had already been removed by the less polar solvent e.g. hexane. The highest quantity of heartwood extractives in series extraction was recorded in water extract (9.62%) followed by acetone (4.62%). Generally percentage yield of extractives increased with increasing solvent polarity and are in agreement with observation reported by Sirmah *et al.,* 2008 and Mburu *et al.*, 2007 implying most extractives are soluble in water and alcohols. Total yield of extractive from this species is high as is expected of tropical hardwoods (Umezawa, 2001; Tsoumis, 1991). From the perspective of natural durability where extractives contribute to wood durability (Gerardin *et al.,* 2004; Mburu *et al.*, 2007; Sirmah *et al.,* 2009), this study suggest that heartwood would be suitably used in areas where durability is vital. However, the timber may not be suitable in pulp and paper industry where extractives cause processing problems (Umezawa, 2001; Baeza and Freer, 2001; Gutierrez *et al.,* 1998). Studies by Grabner *et al.*, (2005) on effects of heartwood extractives on

mechanical properties of larch found that the mechanical properties increase with increase in extractives. Thus, the presence of high amount of heartwood extractive suggests that *A. malacophylla* could be used in areas where strength is desirable such as railway sleepers.

4.4.2 Amount of wood extractives by cold soaking extraction method

The mean percentage yield of extractives in sapwood and heartwood of *A. malacophylla* by cold soaking extraction is presented in Figure 9

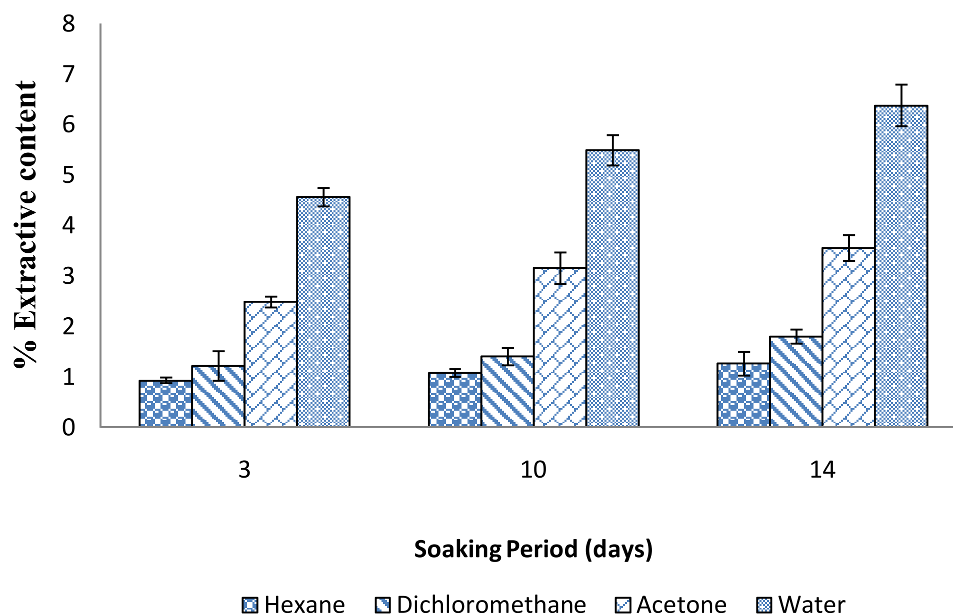


Figure 9: Comparison between extractive content of *A. malacophylla* from different solvents (Author, 2012)

The results indicated that the amount of extractives were significantly influenced by the soaking period and the type of solvent used ($p<0.05$). This implies that the percentage yield of extractives increased as soaking period increased as shown in figure 9. Similar observations have been reported in literature (Albulescu *et al.,* 2004). For instance, the amount of extractives after 3 days was about 0.93% when hexane was used as the solvent and increased to 1.26% when soaking period was increased to 14 days. Similar trend of observations were made on the percentage yield of extractives when other solvents were used.

The percentage yield of extractives was also different for the different solvents used. Water removed the highest amount (6.38%) of extractives when the chips were soaked for 14 days while hexane removed the least amount (1.26%) for the same duration. Similar observations have been reported in literature from other wood species (Yamamoto and Hang, 1988). The two way ANOVA revealed a significant interaction effect of soaking days and solvent used ($P<0.05$).

4.5 Natural wood durability tests

4.5.1 against fungi in a soil bed test

The soil bed moisture content was maintained at ≈70% throughout the test by weighing the containers weekly and adding sufficient water to bring the containers to original weight that brought the appropriate moisture content (Table 3).

Table 3: Soil bed moisture content (Author, 2012)

Soil specimen	% moisture content
1	71.4
2	70.1
3	68.5
4	69.9
5	72.2
Average	**70.4**

Natural durability of *A. malacophylla* against fungal decay was tested by exposing extracted and non-extracted heartwood specimens to unsterile soil-bed for 32 weeks. The wood mass losses due to fungal degradation in the soil bed are reported in figure 10.

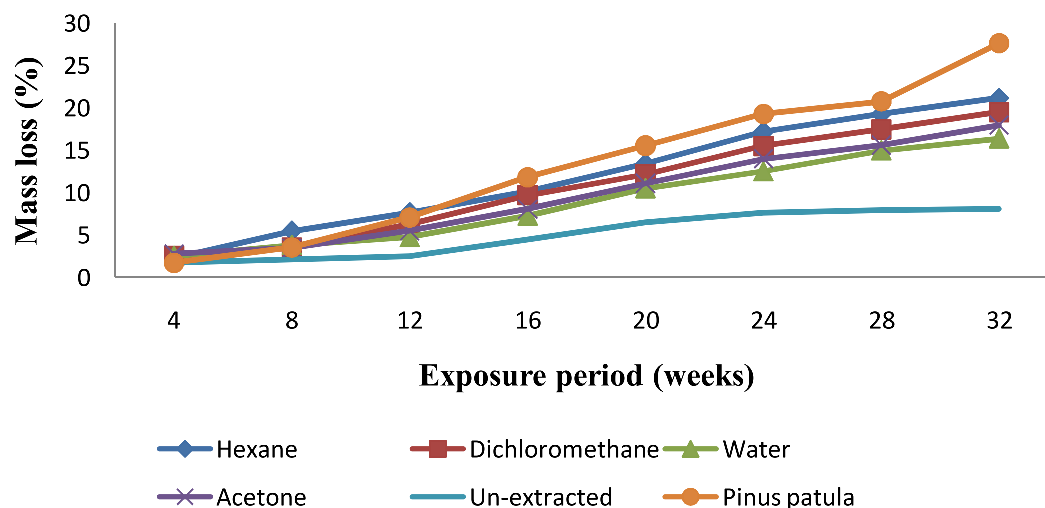


Figure10: Percentage mass loss of solvent extracted and non-extracted wood specimens in a soil-bed test (Author, 2012)

Pinus patula, a perishable wood species, was similarly exposed as the control. The first 4 weeks of exposure showed little variability in decay with percentage mass loss on all test specimens of about 2.3%. This unclear decay pattern during initial period of exposure in soil bed test has been reported (Acker *et al.,* 2003). During the next eight weeks, mass loss on solvent extracted specimens increased tremendously in comparison with the un-extracted specimens retaining the least mass loss average of 2.55%. Thereafter the mass loss increased immensely on the *Pinus patula* and the solvent extracted wood specimens. Towards the 24th week of exposure *Pinus patula* specimens had a mass loss of 19.32%, hexane (17.27%), dichloromethane extracted (15.55%) and acetone extracted (14%) while water extracted had mass loss of 12.56%. In all the test specimens, mass losses due to fungal decay increased with exposure period and there was significant difference amongst the treatments (p=0.000). This is in agreement with observation made by Acker *et al*., (2003). It has been reported that mass loss in softwoods is generally low in soil bed test (Acker *et al.,* 2003; Machek *et al.,* 2001). Therefore, for mass loss assessment criterion, it is recommended that softwoods be exposed for a period of 32 weeks while hardwoods require about 16 or 24 weeks exposure period (Acker *et al.,* 2003).

From the results it is clear that extractives play an important role in the natural durability as indicated by the lowest mass loss of the un-extracted wood specimens (8.1%) after 32 weeks exposure in soil bed. In their study on the decay resistance of Southern Asian timbers in sand block tests, Takahashi and Kishima (1973), observed that the non extracted specimens were more resistant to decay compared to extracted specimens.

The results further revealed that the interaction effect between the exposure period and the treatments significantly contributed to the percentage mass loss of the wood specimens (p=0.000)

Generally, the mass loss increased with decreasing solvent polarity implying that the extractives that are soluble in low polar solvents are mostly responsible for the decay resistance of *A. malacophylla.*

4.5.2 Growth inhibition test

To test whether heartwood extractives contribute to fungal growth inhibition, a fungus was isolated from soil-bed test experiment and cultured in a potato dextrose agar medium inoculated with 0, 50 and 250ppm of extractives. Fungal mycelium growth was measured daily from the centre of the Petri dish and growth inhibition evaluated at the end of the experiment. Figure 11 shows percentage growth inhibition of hexane, acetone and water extracts of *A. malacophylla* heartwood at different concentration level wood

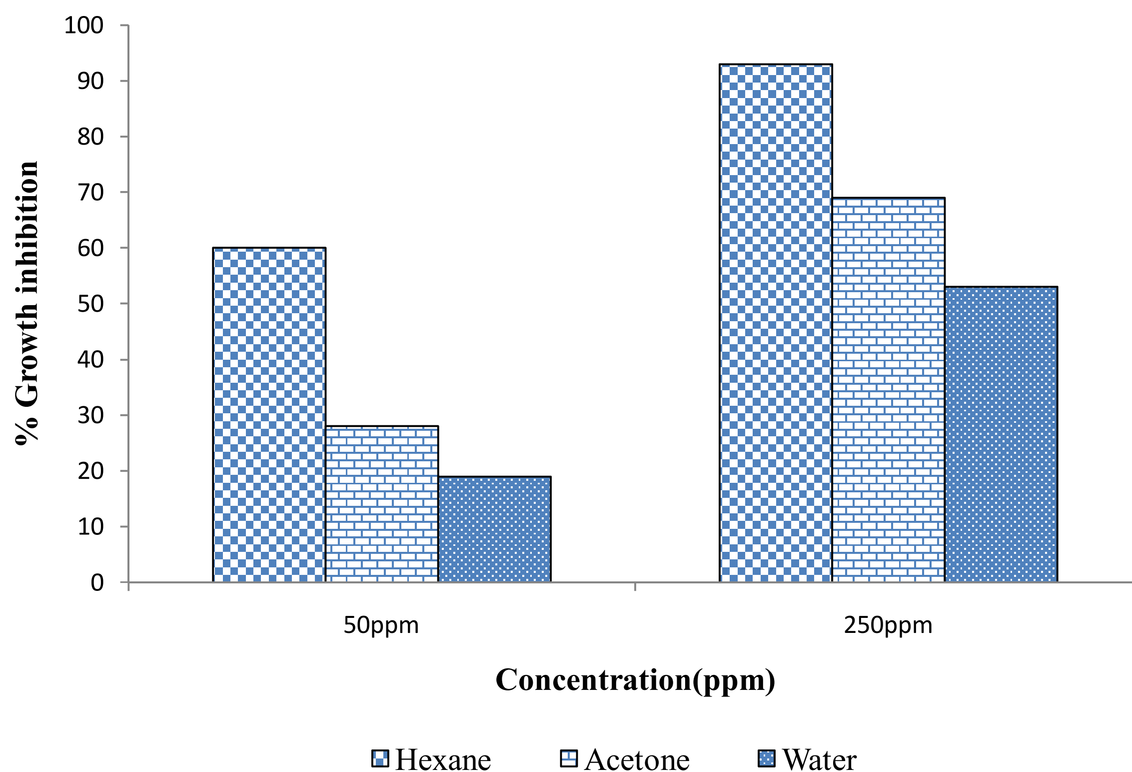


Figure 11: Percentage growth inhibition of *A. malacophylla* heartwood extractives from different solvents (Author, 2012)

Hexane extractives gave the highest inhibition activity against growth of fungi at 50 and 250 ppm concentrations (60% and 93% respectively). Acetonic extract fungal growth inhibition was 28% at 50ppm and 69% at 250ppm concentration level at the same time. Water extracts showed the least inhibition activity of 19% at 50ppm and 53% at 250ppm. Literature studies have shown similar growth inhibition properties of heartwood extractives of other wood species (Neya *et al.,* 2004; Mburu *et al.,* 2007; Sirmah *et al.,* 2008).

From the results it can be concluded that heartwood extractives of *A. malacophylla* inhibits the growth of fungi and the inihibition activity is dependent on the type of solvent used for extraction. It can also be concluded that hexane extract has the highest growth inhibition activity even at low concetration in comparison with water and acetone extractives.The development of fungi on potato dextrose agar treated with different concentration of hexane, acetone and water extracts as a function of time was measured daily.

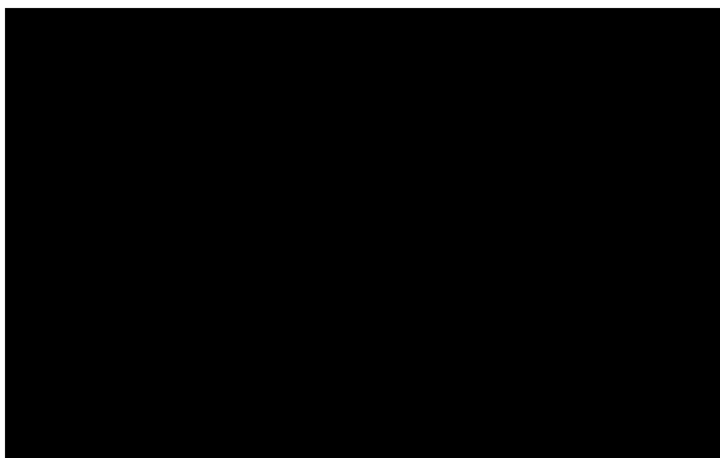


Figure 12: Effect of hexane heartwood extractives of *A. malacophylla* on the growth of fungi (Author, 2012)

Fungi started growing on the potato dextrose agar (PDA) medium treated to 50 and 250 ppm hexane extract after 2and 4 days respectively (Figure 12). The experiment was stopped when fungal growth in the control dishes (0 ppm) attained 90mm diameter.

During this period, dishes with 50mm and 250mm hexane extract had attained mean diameter growth of 20mm and 6mm respectively. Similar trend of fungal activity was observed in acetonic and water extracts as illustrated in figures 13 and 14 respectively

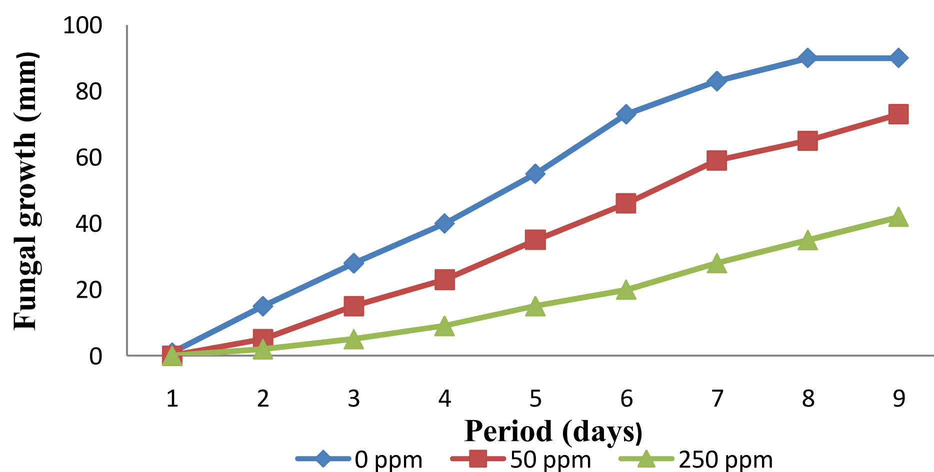


Figure 13: Effect of acetone heartwood extractives on the growth of fungi (Author, 2012)

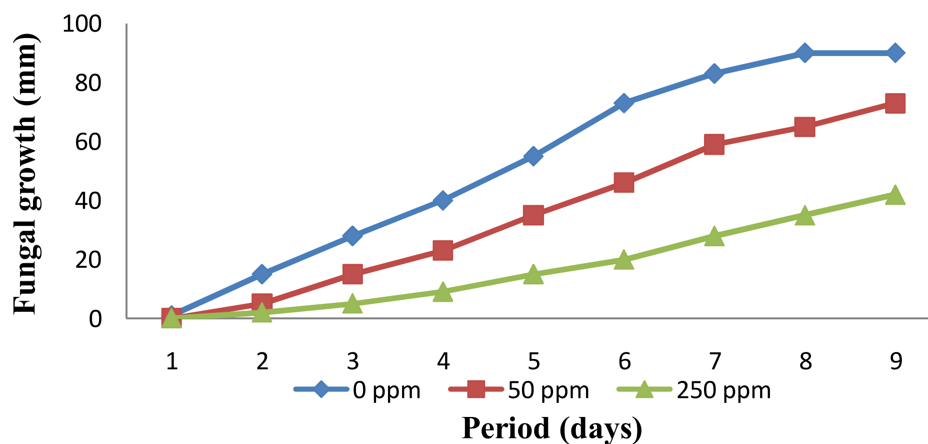
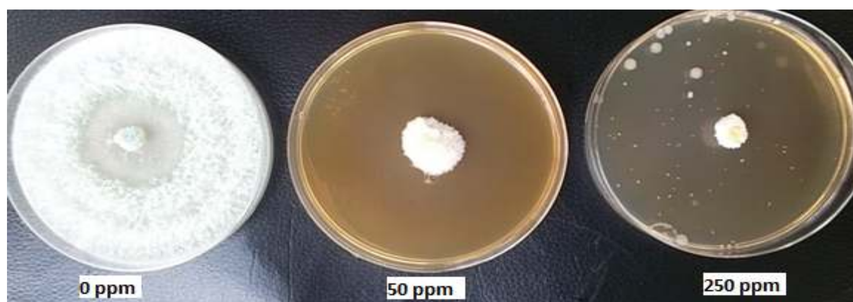


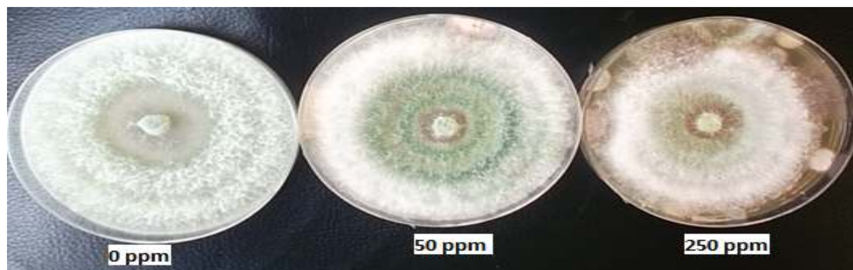
Figure 14: Effect of *A. malacophylla* heartwood water extract on fungal growth (Author, 2012)

It is concluded that hexane, water and acetone extracts possess fungistatic properties with hexane showing greater activity even at low concentration. Plate 10 (a, b and c) shows the activity and the behavior of fungi during the growing period.



a. Hexane extract increasing extract concentration

b. Acetone extract increasing extract concentration



c. Water extract increasing extract concentration

Plate 10: Fungal growth on potato dextrose agar treated to different concentrations of heartwood extractives of *A. malacophylla* (Author, 2012)

The results indicated that the extracts inhibited the growth of fungi at different concentration levels (Plate 10 a, b, c). Hexane extract was more effective as growth inhibitor even at low concentration level when compared with acetone and water extracts.

In general it can be concluded that heartwood extractives of *A. malacophylla* restricts the growth of fungus and thus, contribute to reported wood natural durability. Similar observations have been made on extracts of other wood species (Mohareb *et al.,* 2010; Sirmah *et al.,* 2008; Mburu *et al.,* 2007; Martinez-inigo *et al.,* 1999).

4.5.3 against termites in a field test

Plate 11 shows the physical appearance of wood specimens that were subjected to termite attack in the field for a period of six months.

A. malacophlla **(Un-extracted)** | ***A. malacophylla*** **(Solvent extracted)** | ***Pinus patula*** **(Control)**

Plate 11: Extent of termite attack on wood specimens exposed in the field for six months (Author, 2012)

Least attack by termites on all wood specimens was observed during the first three months of exposure. On the fourth month termite activities increased and this coincided with the onset of wet weather conditions.

Figure 15 shows the percentage mass loss of wood specimens that were exposed to termite attack for six months.

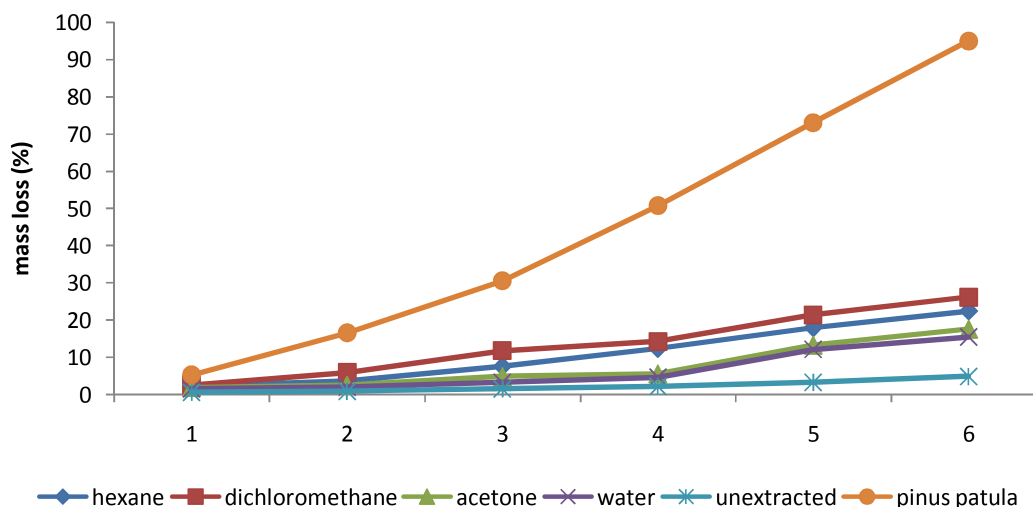


Figure 15: Mass loss of wood specimens exposed to termites (Author, 2012)

The highest mean mass loss was observed in heartwood specimens extracted with dichloromethane (26.1%) followed by those that were extracted by hexane solvent (22.4%). The un-extracted specimens showed the least mass loss of (4.8%) while the *Pinus patula* controls had mass loss of 95% after 6 months of exposure.

From the results, it can be deduced that extractives of *A. malacophylla* heartwood contribute to termite resistance.

The extractives soluble in less polar dichloromethane solvent contribute mostly to the natural durability of *A. malacophylla* wood. Heartwood specimens that were extracted with highly polar water solvent were least attacked by termites (15.4%).

It can be predicted that water would have extracted polar substances such as sugars and other soluble carbohydrates thus making the specimens less desirable to termite attack.

Studies by Lukmandaru (2011) on the variability in the natural resistance of teakwood and its relationship with wood extractive content found that the lower the methanol (highly polar solvent) extractive content, the less severe the mass loss. This was attributed to the removal of polar components that would have induced the termite activity in attacking and degrading the teakwood. In general, it is concluded that indeed, heartwood extractives of *A. malacophylla* especially those that are soluble in the less polar dichloromethane solvent contribute to the natural resistance to termite attack.

4.6 FTIR and GC-MS analysis of heartwood extractives

Studies were carried out to understand the main functional groups present in the *A. malacophylla* heartwood extracts. FTIR and GC-MS analysis was carried out on these extracts. Figure 16 shows an FTIR spectrum of dichloromethane heartwood extract of *A. malacophylla*.

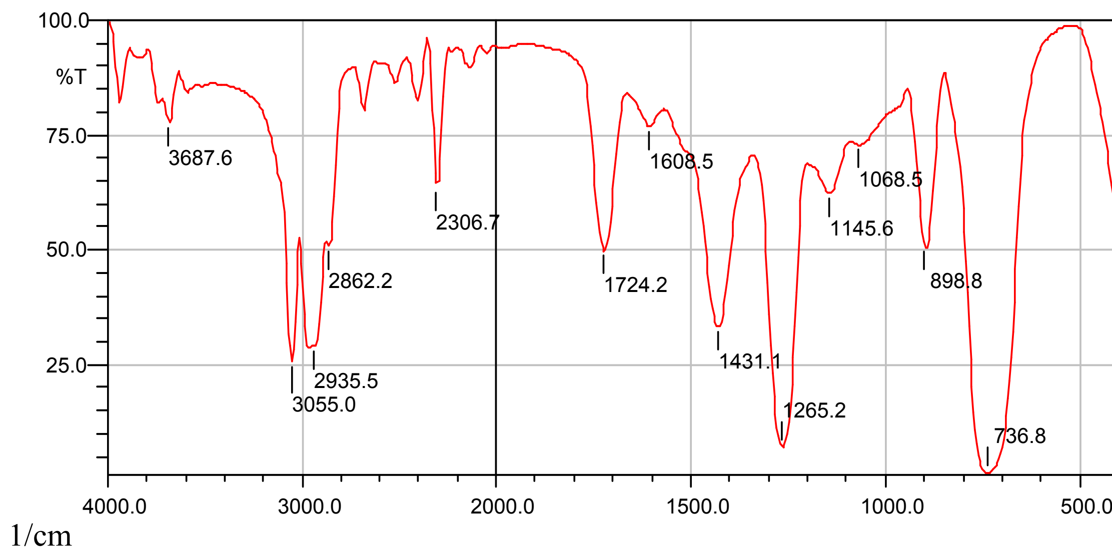


Figure 16: FTIR spectrum of dichloromethane extractives from *A. malacophylla* heartwood (Author, 2012)

The spectrum indicates an OH stretch at 3055 cm^{-1} and a strong aliphatic stretch at 2935 cm^{-1}. The C=C group absorption is indicated by vibrations at 3687 cm^{-1}, typical of either an alkene or an aromatic compound, confirmed by strong vibrations at 1431 cm^{-1} and 1608 cm^{-1} and C-H out of plane bending at 898 cm^{-1}. The strong carbonyl (C=O) absorption is evident at 1724 cm^{-1}, corroborated by C-H absorption at 2862 cm^{-1}. This indicates presence of aldehydes, ketones, carboxylic acids, esters or amines. The strong phenyl ring substitution band is evident at 736 cm. Figure 17 shows FTIR of hexane heartwood extract of *A. malacophylla* extractives

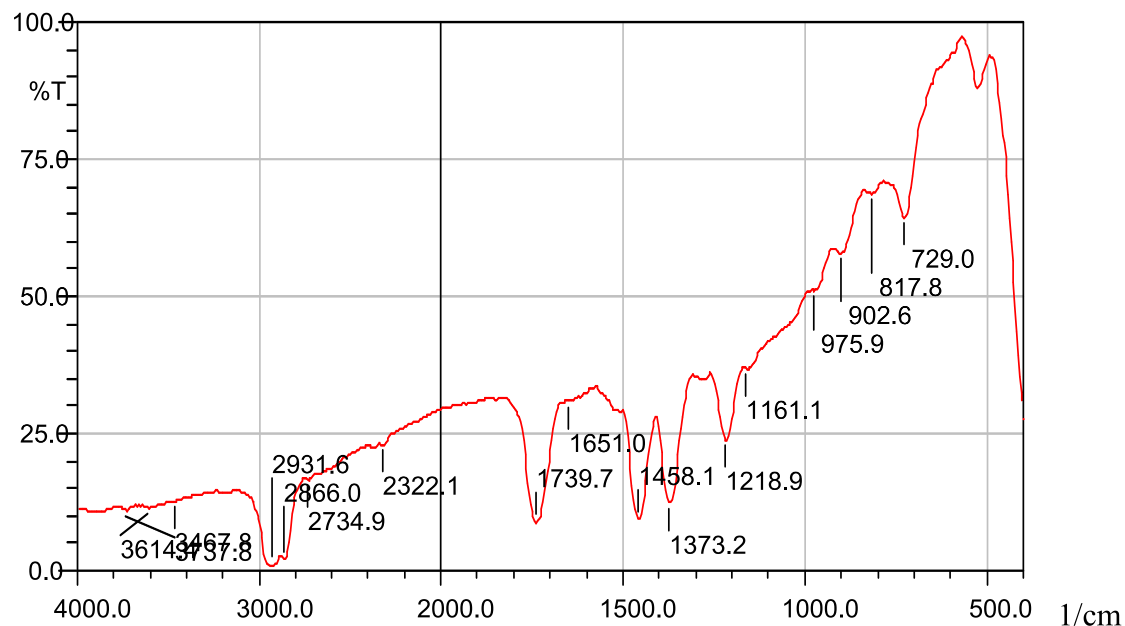


Figure 17: FTIR spectrum for hexane extractives from *A. malacophylla* heartwood (Author, 2012)

The spectrum indicated C=C group absorption at 34614.4cm^{-1}, 3467cm^{-1} and 3737cm^{-1} and alkene vibrations at 1651.0cm^{-1}. There was C-O group absorption at 1161.1 cm^{-1} and 1218.9cm^{-1}. Vibrations are not prominent but were well represented indicating presence of aldehydes, ketones esters amines and carboxylic acids.

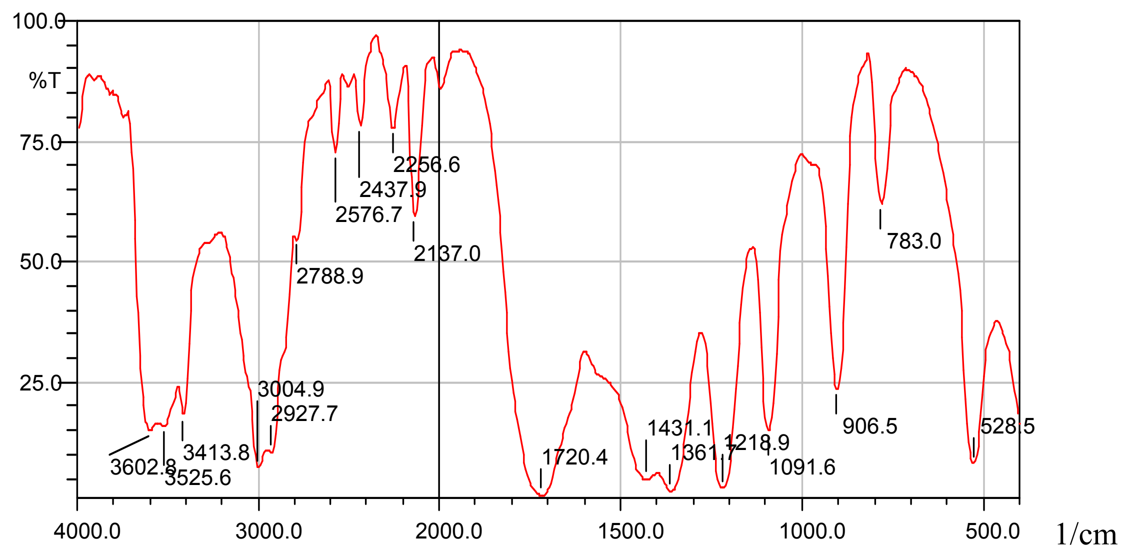


Figure 18: FTIR Spectrum of acetone extractives from *A. malacophylla* heartwood (Author, 2012)

The spectrum indicated a C=C group absorption at3525.6 cm^{-1} and 3625.6 cm^{-1}. The absorption at 2927.7 cm^{-1} is attributed to the presence of aliphatic hydrogen while carbonyl group absorption at 1720.4cm^{-1} is confirmed to be an aldehyde at 2788.9cm^{-1}. Presence of alcohol was indicated by spectrum absorption at 3525.6cm^{-1} and 3602.8cm^{-1}.

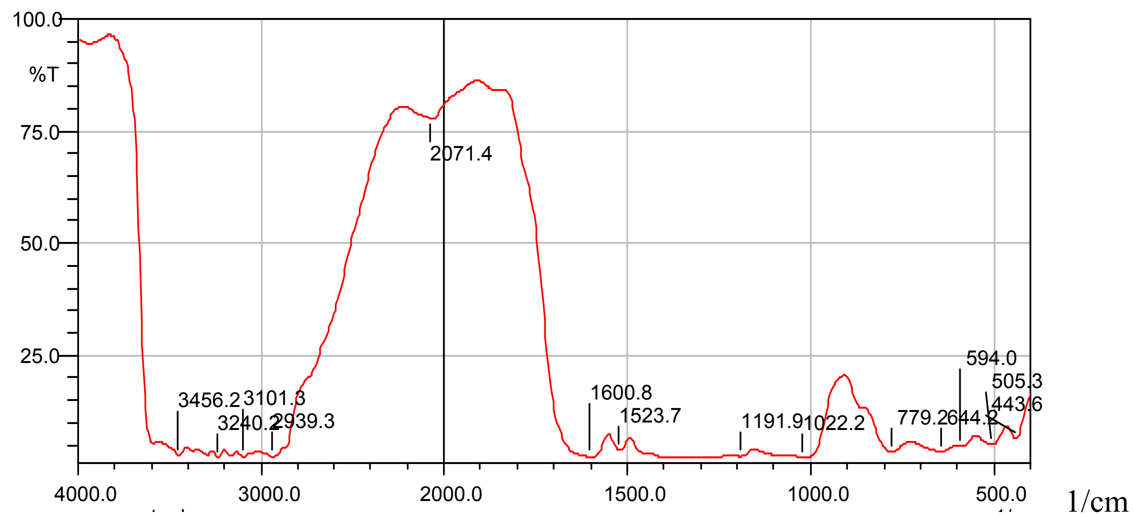
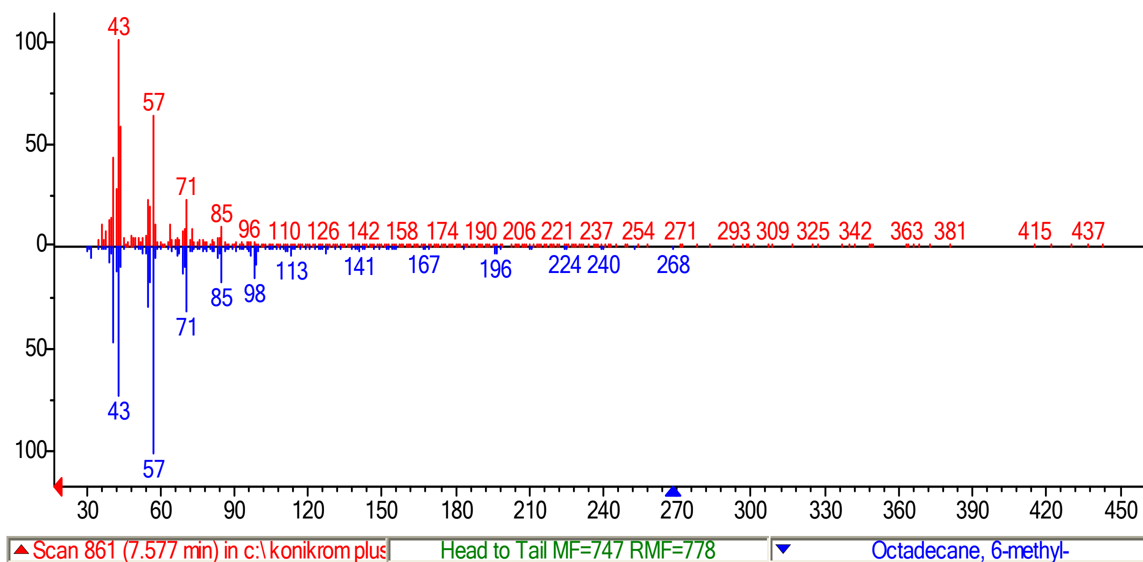


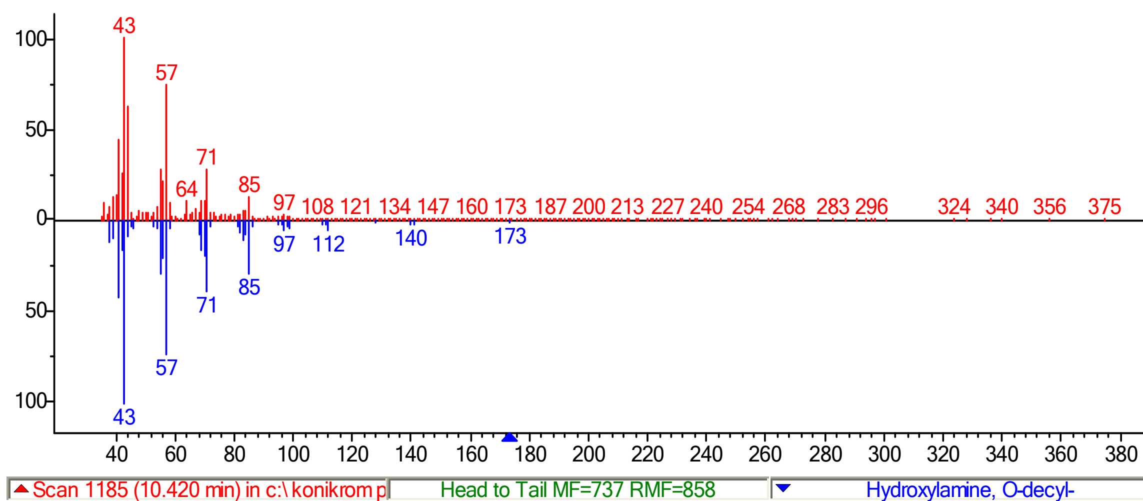
Figure 19: FTIR spectrum of water extractives from A. malacophylla heartwood(Author, 2012)

C=C group absorption at 3101.3cm^{-1} and an aromatic vibration is indicated at 1523.7cm^{-1} absorption band and C-O group absorption at 1191.9cm^{-1}.

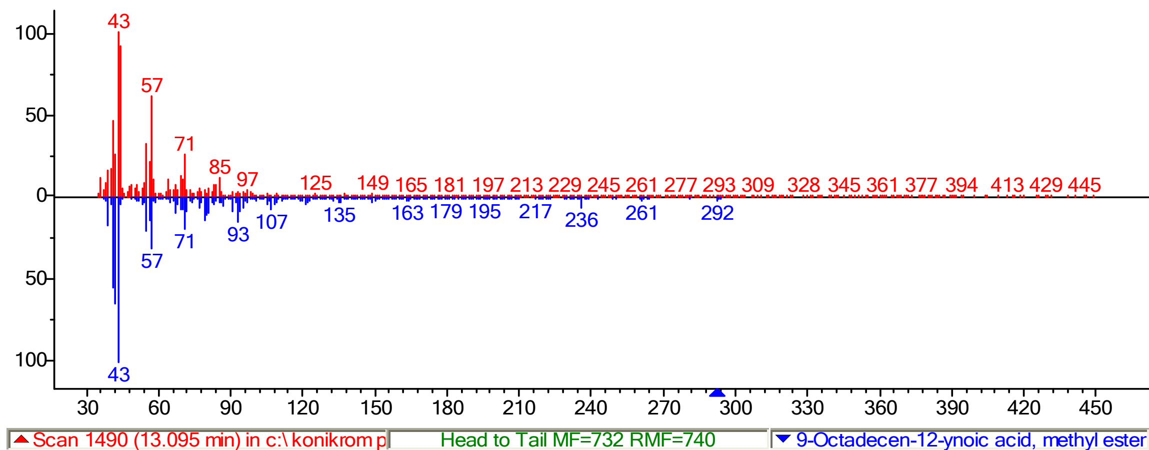
Figure 20 (a-f) shows mass spectra of acetone heartwood extractives of *A. malacophylla* at different retention times.



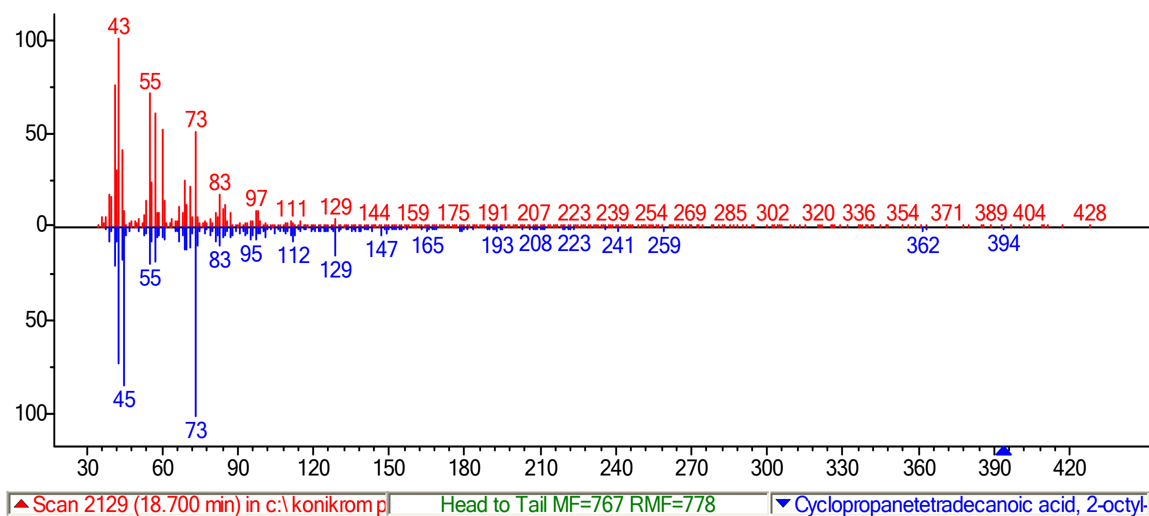
a. Mass spectrum at 7.577 minutes retention time



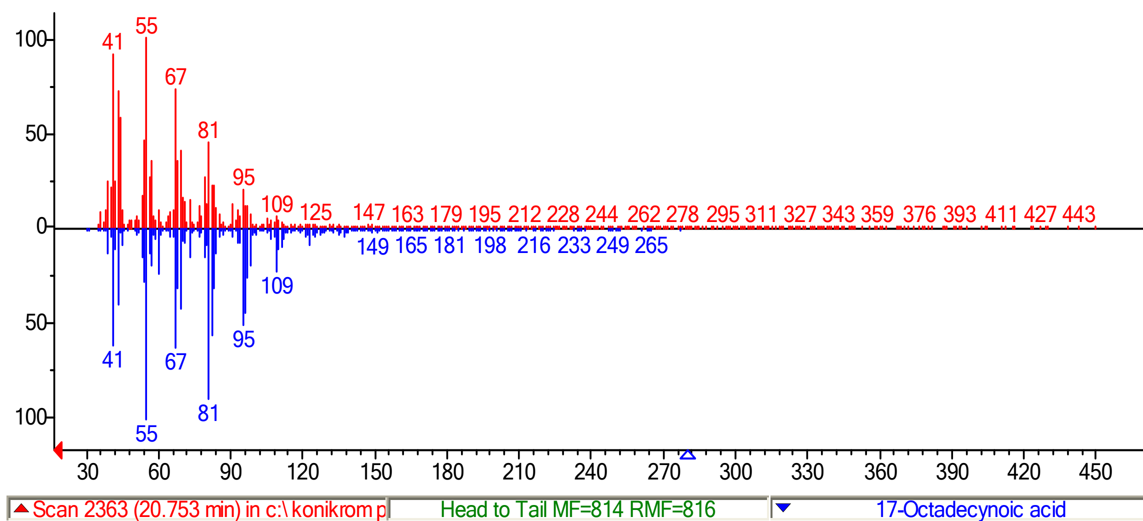
b. Mass spectrum at 10.420 minutes retention time



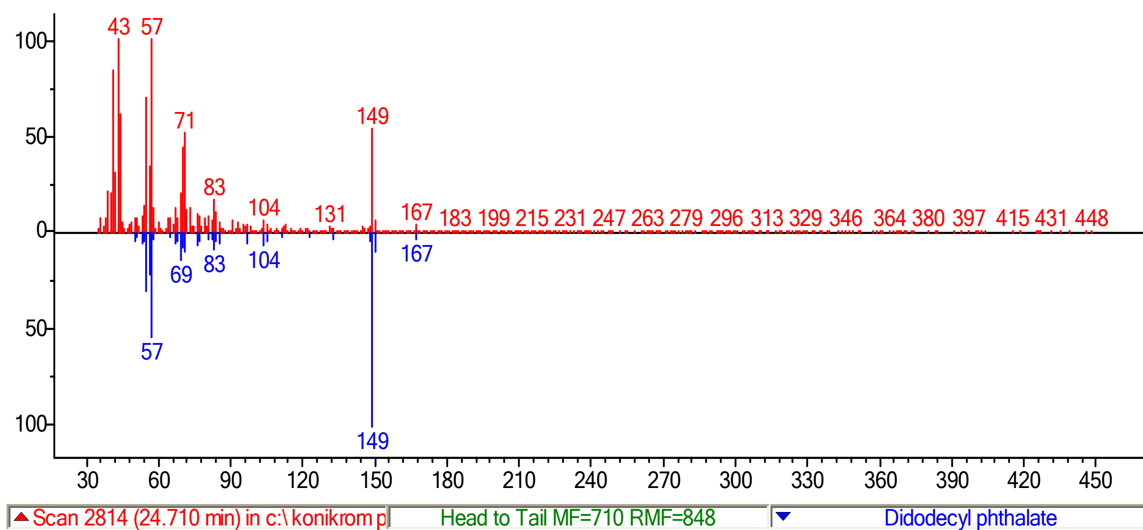
c. Mass spectrum at 13.095 minutes retention time



d. Mass spectrum at 18.700 minutes retention time



e. Mass spectrum 20.753 minutes retention time



f. Mass spectra at 24.710 minutes retention time

Figure 20 (a-f): Mass spectra of acetone extract from *A. malacophylla* heartwood at different retention time (Author, 2012)

NIST library showed presence of carboxylic acid groups (decanoic, octadecanoic, and tetradecanoic acids), at the different retention times tested. Similar groups of compounds were also reported in water extracts as shown in figure 21.

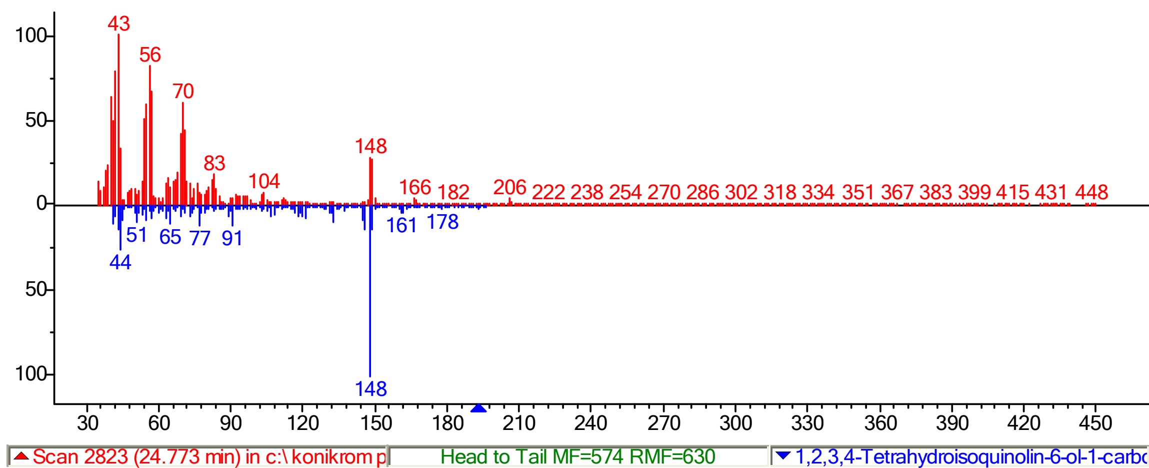


Figure 21: Mass spectrum of water extract from A. malacophylla heartwood (Author, 2012)

At 24.773 minutes, the NIST library indicated presence of 1,2,3,4-Tetraydroisoquinolin-6-ol-1-carboxylic acid at probability of 56.6%.

CHAPTER FIVE

5.0. CONCLUSIONS AND RECOMMENDATIONS

5.1. Conclusions

The wood density of *A. malacophylla* ranged from 1.01g/cm^3 in heartwood to 0.86g/cm^3 in sapwood. The heartwood is dimensionally stable (5.52% swelling coefficient) with a less dimensionally stable sapwood (9.6% swelling coefficient), indicating its strong hydrophobic nature. Both the wood dimensional stability and density was lowered by solvent removal of extractives. Wood ash content ranged from 2.5% in sapwood to 3.1% in heartwood.

Anatomical study of *A. malacophylla* showed that the sapwood is white to yellow in color, and quite distinct from the dark brown heartwood. Microscopic observation revealed thin to thick walled fibers and presence of gums and other deposits in the vessels. The vessels are exclusively solitary. The wood rays are 1 to 3 cells wide with two cells per parenchyma strand. All ray cells are procumbent. The prismatic crystals appeared as long chains in the tangential section.

Albizia malacophylla heartwood contains high amount of extractives (≈ 9.71%). Most of the extractives are soluble in the polar water solvent (9.71%) while small amount are soluble in a non polar hexane solvent (2.36%).

Wood natural durability against decay fungi in soil bed test showed that *A. malacophylla* is durable as indicated by mass loss of 8.1% after 32 weeks of exposure.

The natural durability was lowered by solvent removal of extractives. The mass loss increased with decreasing solvent polarity indicating that the extractives that are soluble in low polar solvents contribute most to the decay resistance. Potato dextrose agar medium test indicated that heartwood extractives inhibit the growth of fungi to some extent at different levels of concentration. Hexane extracts were effective as growth inhibitors even at low concentration levels in comparison with water and acetone extracts.

Natural durability tests against termite attack in the field showed that *A. malcophylla* heartwood is naturally durable, (4.8%) mass loss. The natural durability was lowered by solvent removal of extractives. Dichloromethane extracted wood specimens were more susceptible to termite attack as indicated by the higher mass loss (21.2%) in comparison with those extracted with water and acetone (mass loss of 16.4% and 17.9% respectively). Natural durability of *A. malacophylla* heartwood may be attributed to its high density, dimensional stability, anatomical features and high extractive content.

FTIR and GC-MS analysis of crude heartwood extractives indicated the presence of compounds such as aldehydes, ketones carboxylic acids and esters, aromatic, aliphatic and carbonyl compounds.

In summary, *A. malacophylla* wood could be suitably used in areas where durability is important such as in outdoor work, furniture industry, railway sleepers and bridge construction because of its strong hydrophobicity and color properties.

5.2 Recommendations

Based on the outcome of this study recommendations are made as follows:

5.2.1 from the study

1. *A. malacophylla* wood could be suitable for use in making furniture due to its strong hydrophobic nature and colour properties.

2. The wood from *A. malacophylla* is suitable for outdoor purposes because of the confirmed natural durability.

5.2.2 for further research

1. Further studies on bonding, treatability and mechanical properties of *A. Malacophylla* wood.

2. Further work to isolate and identify specific extractive compounds that are responsible for natural durability of *A. malacophylla* wood.

REFERENCES

Acker J., Stevens M., Carey J., Siera-Alvarez R., Militz H., Bayon I., Kleist G., Peek R., (2003). Biological durability of wood in relation to end-use. *Holz als Roh- Und Werkstoff, 35-45.*

Albulescu M., Alexa N., Cojan C., (2004). *Calendula officinalis* flowers, source of extracts with antioxidant activity. *Series Chemistry, 13(2):169-176.*

Ann-Christine R., (1996). Decay Mechanism of brown rot Fungi. Technical Research Centre of Finland, VTT Publications 268. 67p.

Apetorgbor, M., (2007). *Albizia zyga* (DC) J. F. Macbor [internet] Record from protabase. Loupe, D., Oteng-Amoaka A. and Brink M. (editors). PROTA (Plant Resources of Tropical Africa /Resource Vegetales de l'Afrique Tropicale), Wageningen, Netherlands http://database.prota.org/search html accessed 4th June 2012.

Apolinario F. and Martius C., (2004). Ecological role of termites (Insecta, Isoptera) in tree trunks in Central Amazonian rainforest. *Forest Ecology and Management, 194:23-28.*

AWPA: E7-93. Standard method of evaluating wood preservatives by field tests with stakes. 1993.

AWPA: E23-07. Accelerated method of evaluating wood preservatives in soil contact.2008.

Baeza J. and Freer J., (2001). "Chemical characterization of wood and its components." In wood and Cellulosic Chemistry Ed Marcel Dekker, Inc. Newyork.

Bending G., Friloux M., Walker A., (2002). Degradation of contrasting pesticides by white rot fungi and its relationship with lignolytic potential. *FEMS microbiology letters, 212:59-63*

Blanchette (1991). Delignifigation by wood decay fungi. *Annual Review of Pathology 29:381-403.*

Browning B., (1975). *The Chemistry of Wood*. Robert E Kreiger Publishing Company. New York.

Calonego F., Severo E., Furtado E., (2010). Decay resistance of thermally modified *Eucaliptus grandis* wood at 140 °C, 160 °C, 200 °C and 220 °C. *Bioresource technology, 101:9391-9394.*

Chemo (2003). Beginners guide to soxhlet extraction. Anabolicminds.com

Chirra F., (1995). Ohio pesticide application training. In: wood preservation student work book. The Ohio State University Extension. Product Code 821-813.

Clausen C., (1997). Immunological detection of wood decay fungi- An overview of techniques developed from 1986 to the present. International *Biodeterioration and Biodegration 39(2-3):133-143.*

Clausen C. and Yang V., (2007). Protecting wood from mould, decay, and termites with multicomponent biocide systems. *International Biodeterioration & Biodegradation, 59:20-24.*

Dorado J., Beek T., Clausen F., Sierra-Alvarez R., (2001). Degradation of lipophylic wood extractive constituents in *Pinus Sylvestris* by white-rot fungi *Bjerkandera sp.* and *Trametes versicolor*. *Wood Science and Technology, 35:117-125*.

Diouf P., Delbarre N., Perrin D., Gerrardin P., Rapin C., Jacquot J., Gekaye E., (2002). Influence of tropolonce on *Poria placenta* wood degradation. *Applied and Environmental Microbiology, 68(9): 4377-4382.*

Eggeling J., and Dale R., (1951). *The indigenous trees of Uganda protectorate*. Robert Maclehose and Company limited. The University Press Glasgow.

Forest Products Laboratory, (1999). Wood handbook—Wood as an engineering material. Gen. Tech. Rep. FPL–GTR–113. Madison, WI: U.S. Department of Agriculture, Forest Service, Forest Products Laboratory.

FRA, (2010). Global Forest Resources Assessment Country Report, Kenya. Food and Agricultural Organization of the United Nations. Forestry Department. Rome.

Gambetta A., Susco D., Zanuttini R., (2004). Determination of the natural durability of larch wood (*Larix decidua* Mill.) from the Western Italian Alps. *Holzforschunng, vol. 58, pp. 678-681.*

Gao D., Du l. Yang J., Wu W., Liang H., (2010). A critical review of the application of white rot fungus to environmental pollution control. *Critical Review in Biotechnology, 30(1):70-77.*

Gerardin P., Neya B., Dumarcay S., Petrissans M., Mohamed S., Huber F.,(2004). Contribution of gums to natural durability of *Prosopis africana* heartwood. *Holzforschung, 68: 39-44*.

Ghasemzadeh A., Jaafer H., Rhamat A., (2011). Effects of solvent type on phenolics and flavanoids content and antioxidant activities in two varieties of young ginger (Zingiber officinale roscoe) extracts. *Journal of Medicinal Plant Research 5(7):1147-1154.*

Ghosh S., Militz H., Mai C. (2008). Decay resistance of treated wood with functionalized commercial solutions. *Bioresources 3(4) 1303-1314*.

Giergielewicz-Mozjska H., Da browski L., Namiesnik J., (2001). Accelerated Solvent Extraction (ASE) in the Analysis of Environmental Solid Samples - Some Aspects of Theory and Practice. *Critical Review in Analytical Chemistry 31(3):149-165.*

Githiomi J., Kariuki J., (2010). Wood basic density of *Eucaliptus grandis* from plantations in Central Rift Valley, Kenya: variation with age, height level and between sapwood and heartwood. *Journal of Tropical Forest Science, 22(3): 187-195*.

Grabner M., Muller U., Gierlinger N., Wimmer R., (2005). Effects of heartwood extractives on mechanical properties of Larch. *IAWA Journal, 26(2): 211-220*.

Grabner M., Wimmer R., Gierlinger N., Evans R., Downes G., (2005). Heartwood extractives in larch and effects on X-ray densitometry. *Can. J. For. Res.35:2781-2786.*

Golpayegani A., Bremand I., Gril J., thevenon M., Arnould O., Pourtahmosi K., (2012). Effects of extraction on dynamic mechanical properties of white mulberry (*Morus alba* L). *Journal of Wood Science 58:153-162*

Gutierrez A., Rio J., Gonzalez-vila F., Romero J., (1998). Variation in the composition of wood extractives from *Eucalyptus globulus* during seasoning. *Journal of Wood Chemistry and Technology, 184(4), 439-446.*

Hashemi S., and Latibari A., (2011). Evaluation and Identification of Walnut heartwood extractives for protection of poplar wood. *Bioresource 6(1):152-163*.

Hernandez R., (2007). Swelling properties of hardwoods as affected by their extraneous substances, wood density, and interlocking grain. *Wood and Fiber Science, 39(1):146-158.*

Hernes P. and Hedges J., (2004). Tannin signatures of barks, needles, leaves, cones, and wood at the molecular level. *Geochimica et Cosmochimica Acta, 68(6):1293-1307.*

Images, http://www.wikipedia.org/wiki/soxhlet_extractor accessed 7th June 2012

Jouquet P., Ranjard L., Lepage M., Lata J., (2005). Incidence of fungus-growing termites (Isoptera, Marcrotermitinae) on the structure of soil Microbial communities. *Soil Biology and Biochemistry, 37:1852-1859.*

Jouguet P., Traore S., Choosai C., Hartmann C., Bignell D., (2011). Influence of termites on ecosystem functioning. Ecosystem services provided by termites. European *Journal of Soil Biology xxx: 1-8.*

Junior E., Terziev N., Daniel G., (2009). Wood anatomy of three lesser known species from Mozambique. *IAWA Journal, 30(3):277-291*

Kirby and Keasling J., (2009). Biosynthesis of plant isoprenoids: Perspectives for microbial engineering. *Annu. Rev. Plant Biol., 60:335-355.*

Kenya Forest Master Plan, (KMFP) (1994). Development Programmes, Ministry of Environment and Natural Resources. Government of Kenya.

Kollmann, F. and Côté, W. (1984). Principles of Wood Science and Technology, vol.1. springer Verlag: Germany.

Kose C., Taylor A., (2012). Evaluation of mold and termite resistance of included sapwood in Eastern red cedar. *Wood and Fiber Science, 44(3):1-6.*

Kukachka B., (1969). Properties of Imported Tropical Woods. US Department of Agriculture, Forest Service. Forest Product Laboratory.

Kulla D., and Obi A., (2005). Calorific values of wood species of Nigerian origin. *Journal of Applied Science and Technology (JAST), 10 (1&2):66-71*

Lin L., Chen Y., Wang S., Tsai M., (2009). Leachability, metal corrosion and termite resistance of wood treated with copper-based preservative. *International Biodegradation and Biodeterioration, 63:533-538.*

Lopez G., Schulman H., Hermes-Lima., (1999). Polyphenol tannic acid inhibits hydroxyl radical formation from Fenton reaction by complexing ferrous ions. *Biochimica Acta 1472:142-152.*

Lukmandaru G., (2011). Variability in the natural termite resistance of plantation teak wood and its relations with wood extractive content and color properties. *Journal of Forestry Research 8(1):17-31*

Machek L., Militz H., Sierra-Alvarez R., (2001). The use of an acoustic technique to assess wood decay in laboratory soil-bed tests. *Wood Science and Technology, 34:467-472.*

Marshall N., Jenkins M., (1994). Species in danger. Hard times, Hardwood Indigenous Timber and the timber trade in Kenya. Traffic international, Cambridge. United Kingdom.

Martinez-iningo M., Immerzeel P., Gutierrez A., Rio J., Sierra-Alvarez R., (1999). Biodegradability of extractives in sapwood and heartwood from scots pine by sapstain and white rot fungi. *Holzforschung, 53:247-252.*

Mburu F., Dumarcay S., Bocquet J., Petrissans M., Gerardin P., (2008). Effect of chemical modifications caused by heat treatment on mechanical properties of *Grevillia robusta* wood. *Polymer Degradation and Stability, 93:401-405*

Mburu F., Dumarcy S., Huber F., Petricians M., Gerardin P., (2007). Evaluation of thermally modified *Grevillea robusta* heartwood as an alternative to shortage of Wood Resource in Kenya. Characterization of physichochemical properties and improvement of bioresistance. *Bioresource Technology, 98:3478-3486*.

Mburu F., Dumarcay S., Gerardin P., (2007). Evidence of fungicidal and termicidal properties of *Prunus africana heartwood* extractives. *Holzforschung, 6 :323-325*.

Min R., Pinchak E., Merkel R., Walker S., Tomita G., Anderson C., (2008). Comparative antimicrobial activity of tannin extracts from perennial plants on mastitis pathogens. *Scientific Research and Essay, 3(2):066-073.*

Mohareb A., Sirmah P., Desharanais L., Dumarcy S., Petrissans M., Gerardin P., (2010). Effects of extractives on conferred and natural durability of *Cupressus lusitanica* heartwood. *Ann. For. Sci., 67(5):504*

Mohmod A., Amin A., Kasim J., Jusuh M., (1993). Effect of anatomical characteristics on the physical and mechanical properties on *Bambusa blumeana*. *Journal of Tropical Forest Science 6(2):159-170.*

Narayana K., Reddy M., Chaluvadi M., Krishna D. (2001). Bioflavanoids classification, pharmacological, biochemical effects and therapeutic potential. *Indian Journal of Pharmacology, 33:2-6*

Neya B., Hakkou M., Petrissans M., Gerardin P., (2004). On the durability of *Burkea africana* heartwood : Evidence of biocidal and hydrophobic properties responsible for durability. *Ann. For. Sci. 61:277-282*.

Nicholas D., (1973). Wood deterioriation and its prevention by preservative treatments. Syracuse University Press : New York.

Nicholas D.and Crawford D., (2003). Concepts in the development of new accelerated test methods or Wood decay. American Chemical Society. US department of Agriculture, madison.

Ozmen N., (2007). Dimensional stabilization of fast growing forest species by acetylation. *Journal of Applied Sciences, 7(5) :710-714*

Panday K., Hughes M., Vuorinen T., (2010). Dimensional stability, UV resistance and static mechanical properties of scots pine chemically modified with Alkylene epoxides. *Bioresources, 5(2): 595-615.*

Panshin, A. and De Zeeuw C. (1980). *Text book of Wood Technology 4th ed*. Macgraw Hill Book Company: USA

Powell K., Pedley S., Daniel G., Corfeid M., (2001). Ultrastructural observations of microbial succession and decay of wood buried at a Bronze Archeological site. *International biodeterioration & degradation, 47:165-173*.

Ragon K., Nicholas D., Schultz T., (2008). Termite –Resistant Heartwood: The Effect of Non-Biocidal Antioxidant properties of Extractives (Isoptera:Rhinotermitidae). *Sociobiology, 52(1)*

Ramos l., Kristenson E., Brinkman U., (2002). Current use of pressurized liquid extraction and subcritical water extraction in environmental analysis. *Journal of Chromatography A, 975: 3-29.*

Rao R., Tiwari A., Kumar U., Reddy V., Ali A.,Rao J., (2003). Novel 3-0-Acyl Mesquitol analoques as free radical scavengers and enzyme inhibitors: Synthesis, Biological evaluation and structure-activity relationship. *Bioorganic and Medicinal Chemistry Letters 13:2777-2780.*

Reed J. (1995). Nutrition toxicology of tannins and related polyphenols in forage legumes. *J Anim Sci, 73:1516-1528.*

Ren W., Quao Z., Wang H., Zhang L.(2003). Flavanoids: promising anticancer agents. *Medicinal Research Reviews, 23(4):519-534*.

Roll, D., (2003). Wood Preservation, category 4b: study guide for commercial applicators.

Royer M., Garcia-Perez M., Duque-Fernandez A, Diouf P., Stevanovic T., Pouliot R. (2010). Antioxidant, toxicological and antiproliferative properties of Canadian polyphenolic extracts on normal and psoriatic keratonicytes. *J Ethnophamacol 132(1):251-258*

Schwanninger M. and Hinterstoisser B., (2002). Comparison of classical wood extraction method using a soxhlet apparatus with an advanced extraction method. *Holz als Roh Und Werkstoff 60:343-346.*

Scheffer T. and Cowling E., (1966). Natural resistance of wood to microbial deterioration. *Annul. Rev. Phytopathol, 4:147-168.*

Scheffer T. and Morrell J., (1998). Natural Durability of Wood. A World wide checklist of species. Forest Research Laboratory, Oregon State University. Research contribution. 22. 58p

Scholz G., Militz H., Gascon-Garrido P., Ibiza-Palacios M., Oliver-Villanueva J., Peters B., Fitzgerald C., (2010). Improved resistance of wood by wax impregnation. *International Biodeterioration & Biodegradation, 64:688-693*.

Schubert M., Volkmer T., Lehringer C., Schwarze F., (2011). Resistance of bio-ionised Wood treated with Wood preservatives to blue-stain and wood-decay fungi. International *Biodeterioration and Biodegradation, 65, 108-115.*

Sexton C., Corden M., Morrel J., (1993). Assessing fungal decay of wood by small scale toughness tests. *Wood and Fiber Science, 25(4):375-383.*

Silva C., Monteiro M., Brazolin S., Lobez G., Richter A., Broga M., (2007). Biodeterioration of Brazilwood *Caesalpinia echinata* Lam.(Leguminosae-Caesalpiniodae) by rot fungi and termites. *International Biodeterioration & Biodegradation, 60 :285-292*.

Simsek H., baysal E., Peker H., (2010). Some mechanical properties and decay resistance of wood impregnated with environmentally-friendly borates. *Construction and Building Materials, 24: 2279-2284.*

Sirmah P., Durmacy S., Masson E., Gerardin P., (2009). Unusual amount of (-)- mosquitol from the heartwood of *Prosopis Juliflora*. *Natural Products Research 23(2):183-189.*

Sirmah P., Mburu F., Laych K., Dumarcy S., Gerardin P. (2011). Potential antioxidant compound from Different parts of *Prosopis juliflora*. *Journal of Tropical Forest Science 23(2) 187-195.*

Sirmah P., Mburu F., Muisu F., Wahungu G., Waweru C., (2009). Incidence of marine borer attack at different depths at Mtongwe Jetty Pontoon Mombasa, Kenya. *Afr. J. Ecol. 47, 693-698.*

Sirmah P., Muisu F., Mburu F., Dumarcay S., Gerardin P., (2008). Evaluation of *Prosopis juliflora* as an Alternative to Wood Shortage in Kenya. *Bois Et Foret Des Tropiques* N^0 *298(4).*

Singelton R., DeBell D., Gartner B., (2003). Effects of extraction on wood density of western Hemlock (Tsuga heterophylla (Raf.) Sarg) *Wood and Fiber Science, 35(3): 363-369.*

Singh H. and Sasekumar A., (1996). Wooden Panel Deterioration by Tropical Marine Wood borers. *Estuarine, Coastal and Shelf Science, 4 :755-769.*

Sonowal J. and Gogoi, (2010). Dimensional stability, thermal degradation and termite resistance studies of chemically treated wood. *International Journal of Chemistry, 2(2)*

Soon L. and Chiang L., (2012). Influence of differrent extraction solvents on lipophylic extractives of Acacia hybrid in Different wood portions. *Asian Journal of Applied Sciences: 107-116.*

Susi P., Aktunganov G., Himanen J., Korpela T., (2011). Biological control of wood decay against fungal infections. *Journal of Environmental Management, 92:1681-1689*.

Stephan I., Goller S., Rudolph D., (2000). Improvements of monitoring the effects of soil organisms on wood in ground contact. *Holz als- Und Werkstoff, 58:115-119.*

Sun Q., Yu H., Liu Y., Li J., Lu Y., Hunt J., (2010). Improvement of water resistance and dimensional stability of wood through titanium dioxide coating. *Holzforschung, 64: 757-761.*

Takahashi M. and Kishima T., (1973). Decay resistance of 65 South East Asian timber specimens in accelerated laboratory tests. *Tonan Ajia Kenkyu, 10 :524-541.*

TAPPI Standard, T 211 om-07 (draft). Ash in Wood, Pulp, Paper and Paperboard : Combustion at 525 ° C. Un published TAPPI Standard.

Taylor A., Gartner B., Morrel J., (2002). Heartwood formation and natural durability-review. *Wood and Fiber Science, 34(4) pp. 587-611*.

Taylor A., Gartner B., Morrel J., Tsunoda K., (2006). Effects of heartwood extractive fraction from *Thuja plicata* and *Chamaecyparis nootkatensis* on wood degradation by termites or fungi. *J of Wood Sci, 52:147-153*.

Tsoumis, G. (1991). Science and Technology of Wood. Structure, Properties, Utilization. Champan and Hall: New York.

Umezawa T., (2001). 'Chemistry of Extractives'. In Wood and Cellulosic Chemistry. Ed Marcel Dekker, Inc. Newyork, pp 213-241.

Usta I., (2003). Comparative study of wood density by specific amount of void volume (porosity). *Turk J Agric For, 2:1-6.*

Velmurugan N., Chun S., Han S., Lee Y., (2009). Characterization of Chikusaku-eki and Mokusaku-eki and its inhibitory effect on sapstaining fungal growth in laboratory scale. *International Journal of Environmental Science and Technology, 6(1):13-22*

Verma M., Sharma S., Prasad R., (2009). Biological alternatives for termite contrlol : A review. *International Biodeterioration and Biodegradation 6 : 959-972*

Vetter L., Stevens M., Acker J., (2009). Fungal decay and derability of organosilicon-treated wood. *International Biodeterioration and Biodegradation, 63:130-134*.

Williams R., Miller R., Gangstad J., (2001). Characteristics of ten tropical hardwoods from certified forests in Bolivia, Part 1 weathering characteristics and dimensional change. *Wood and Fiber Science pp. 618-626*.

World Conservation Monitoring Centre, (1998). *Albizia malacophylla* var malcophylla. In IUCN Red List of Threatened species.Version 2012.2< www.IUCNredlist.org.> Accessed 4th June 2012

Worrall J., Anagnost S., Zabel R., (1997). Comparison of wood decay among diverse lignocolous fungi. *Mycologia, 89(2): 199-219*

Zhang S., (2003). Wood quality attributes and their impact on wood utilization. Unedited paper submitted to the World Forestry Congress, Quebec City, Canada.

Zhao L., He Y., Deng X., Yang G., Li W., Liang J., Tang Q., (2012). Response surface modelling and optimization of accelerated solvent extraction of four lignans from fructus schisandrae. *Molecules 17 : 3618-3629.*

APPENDIX

PUBLICATION

Wood dimensional stability and extractives as reasons for termite and fungal resistance of the lesser known Albizia malacophylla *Kenyan wood species*

Sylvester Kibet, Peter Sirmah, Francis Mburu & Fred Muisu

Journal of the Indian Academy of Wood Science

ISSN 0972-172X

J Indian Acad Wood Sci
DOI 10.1007/s13196-013-0092-9

Your article is protected by copyright and all rights are held exclusively by Indian Academy of Wood Science. This e-offprint is for personal use only and shall not be self-archived in electronic repositories. If you wish to self-archive your article, please use the accepted manuscript version for posting on your own website. You may further deposit the accepted manuscript version in any repository, provided it is only made publicly available 12 months after official publication or later and provided acknowledgement is given to the original source of publication and a link is inserted to the published article on Springer's website. The link must be accompanied by the following text: "The final publication is available at link.springer.com".

Springer

Author's personal copy

J Indian Acad Wood Sci
DOI 10.1007/s13196-013-0092-9

ORIGINAL ARTICLE

Wood dimensional stability and extractives as reasons for termite and fungal resistance of the lesser known *Albizia malacophylla* Kenyan wood species

Sylvester Kibet · Peter Sirmah · Francis Mburu · Fred Muisu

Received: 19 December 2012 / Accepted: 22 April 2013
© Indian Academy of Wood Science 2013

Abstract Studies were carried out on termite and fungal resistance of the lesser known *Albizia malacophylla* Kenyan wood species. In addition wood dimensional stability, amount and chemical nature of heartwood extractives was also evaluated. Wood resistance against white rot tropical fungi was based on a laboratory soil bed test and subterranean termite resistance in the field according to American Wood Protection Association: E7-93 (1993) standard. Wood extractives were subjected to infra-red analysis using standard laboratory procedures. *Albizia malacophylla* heartwood is dimensionally stable (5.5 %) with a less dimensionally stable sapwood (9.6 %). Heartwood extractive content is high (9.7 %) in comparison to sapwood (4.6 %). *Albizia malacophylla* heartwood is resistant to fungi (8.1 %) mass loss and very resistant to termites (4.8 %) mass loss reported after 6 months exposure. Removal of extractives significantly lowered heartwood dimensional stability, termite and fungus resistance. Heartwood extractives were able to inhibit the growth of fungi under laboratory sterile conditions. Infra-red analysis of crude heartwood extractives indicated presence of aldehydes, ketones carbonyl compounds, esters, aromatic, carboxylic acids and aliphatic carbonyl compounds. Put together, the nature, amount of heartwood extractives and wood dimensional stability are at the origin of the found termite and fungus resistance of *A. malacophylla* wood.

Keywords *Albizia malacophylla* · Wood resistance · Fungi · Termites · Kenya

S. Kibet · P. Sirmah (✉) · F. Mburu · F. Muisu
Department of Forestry and Wood Science, Chepkoilel University College, P.O. Box 1125, Eldoret, Kenya
e-mail: sirmahkipkosgei110@hotmail.com

Introduction

Albizia malacophylla is a lesser known tropical hardwood species, characterized by a rough, pale brown grey bark with large irregular flat scales and attains a height of about 9 m at maturity (Eggeling and Dale 1951). The tree has lost large part of its expanse African wooded grassland due to overexploitation hence is currently endangered and facing threats of extinction (World Conservation Monitoring Centre 1998). In Kenya the remaining *A. malacophylla* is restricted to Teso, Bungoma County. Preliminary survey indicated that its wood is preferred for fencing posts, general construction, charcoal production and firewood over other local species, yet its wood properties are not well known. Wood properties of scarce and endangered species lead to its conservation through cultivation in plantations and possibly expand its utilization as a commercial wood (Silva et al. 2007).

Indeed, other tropical hardwoods such as Teak (*Tectona grandis*) possess good natural durability against decay and insect attack, are dimensionally stable and very resistant to surface checking and end-grain splitting (Williams et al. 2001). *Albizia adianthifolia*, *A. feruginea* and *A. zygia* have moderate to very durable heartwood, course texture, low shrinkage and little movements hence suitable for joinery and carpentry purposes (Kukakcha 1969). It is therefore important to carry out wood durability tests for better and professional utilization of this forest produce.

Evaluating reasons of natural durability of wood leads to more competitiveness and confidence as a building material (Acker et al. 2003; Calonego et al. 2010). Aspiration of bordered pits results in reduced permeability while high levels of gums limit the penetration of water (Schubert et al. 2011; Gerardin et al. 2004). Extractives contribute to wood color, fragrance, durability, pulping, drying adhesion, hygroscopicicity and

Author's personal copy

acoustic properties (Umezawa 2001; Baeza and Freer 2001). Flavanoids in wood contributes to diversity in wood colorations and have significant effect on its durability (Sirmah et al. 2011). Wood density is closely associated with its mechanical strength, shrinkage and heating value (Githiomi and Kariuki 2010). Cracking, twisting, shrinkage and swelling are attributed to undesirable dimensional changes (Sun et al. 2010).

This study reports termite and fungal resistance of *A. malacophylla* wood and some reasons that contribute to the found resistance.

Materials and methods

Wood procurement

From a household plantation in Teso Western Kenya, mature *A. malacophylla* were randomly selected, felled and quarter sawn into boards 1 m long × 0.1 m wide × 0.1 m thick. Defect free boards were sorted and air dried until they attained moisture content of <20 % before sawing further into appropriate sizes for termite, fungal and dimensional stability tests. Mature *Pinus patula* a perishable Kenyan species was similarly acquired for comparison purposes from Kaptagat forest plantation in Elgeyo Marakwet County, Kenya.

Wood resistance against fungi in a soil bed tests

Wood specimen preparation

This study was based on a modified AWPA: E23-07 (2008) (American Wood Protection Association) test standard. One hundred and sixty (160) heartwood specimens of *A. malacophylla*, 50 mm × 10 mm × 5 mm, (l × r × t) were prepared. To understand effects of extractives on wood resistance, thirty-two specimens were soxhlet extracted separately in 150 ml of hexane, dichloromethane, acetone and water for 48 h at a rate of 6–10 cycles per hour. Thirty-two specimens of *P. patula* sapwood not solvent extracted were cut to the same dimensions as above. All the wood specimens were then conditioned at 60 °C in an oven to a constant mass (w_o) before they were subjected to fungi in a soil bed test. Solvent residue was evaporated to dryness and extractive yield expressed as percentage of initial weight of wood specimens (w_o).

Soil bed preparation and exposure of wood specimens

Plastic containers 250 mm × 150 mm × 170 mm (l, w, h) were filled with unsterile soil layers (bottom to top) as follows: 20 mm of gravel, 20 mm of sand and 130 mm of forest soil. The forest soil was collected from Kaptagat Forest. Un-extracted, solvent extracted heartwood specimens of *A. malacophylla* and *P. patula* were exposed to the unsterile soil bed under room temperature (22–28 °C) and controlled soil moisture of about 70 % that is reported to optimize fungal wood decay. Soil beds were weighed weekly and sufficient water added to bring the weight of the container back to the original weight that gave the required moisture content. The specimens were labeled for identification purposes then buried randomly 20 mm apart so that ~1/3 length of each specimen protruded above the soil bed level.

Fungal wood decay resistance assessment

Fungal wood decay was assessed by evaluating the mass losses once every 4 weeks for 32 weeks. During each inspection a replicate set of 4 specimens was removed from the soil bed cleaned from any mycelium and soil particles which adhered to the surface. The specimens were then oven-dried at 102 ± 3 °C for 48 h and the mass loss evaluated as a percentage of the original mass before exposure using the formula below:

$$\text{Mass loss } \% = \frac{w_0 - w_1}{w_0} \times 100$$

where w_1 and w_0 is the weight of test specimen after and before exposure to fungal decay respectively.

Wood resistance against termites in a field test

Wood specimen preparation

This study was based on a modified American Wood Protection Association, AWPA: E7-93 (1993) method of determining resistance to subterranean termites. One hundred and twenty (120) heartwood specimens of *A. malacophylla*, 100 mm × 10 mm × 5 mm (l, r, t) were prepared. To understand effects of extractives on termites wood resistance, twenty-four specimens were soxhlet extracted separately in 150 ml of hexane, dichloromethane, acetone and water for 48 h at a rate of 6–10 cycles per hour. In addition twenty-four non solvent extracted heartwood specimens and twenty-four *P. patula* sapwood were cut to the same dimensions as above. All the wood specimens were conditioned at 60 °C in an oven to a constant mass (w_f) before they were exposed to termites.

Termite nest preparation, exposure and attack evaluation

Termite nests were arbitrarily selected at Cheptebo, Kerio Valley in Elgeyo Marakwet County, Kenya and the test site cleared of plant life and any other cellulosic material. The test specimens were exposed to termites in a randomized

Author's personal copy

design at a distance of 30 cm from the termite nest with two-thirds of their lengths buried in the soil and a spacing of 20 cm apart. Every month, four replicate specimens were gently removed from the soil and soil particles on the surface of the specimens brushed off. The specimens were then dried until they attained constant weights (w_i) after which the extent of attack was evaluated by mass loss using the following formula:

% mass loss $= \frac{w_t}{w_i} \times 100\%$, where, w_t and w_i is the weight of test specimen before and after termite attack respectively.

Effects of heartwood extractives on fungal growth

Fungal mycelia initially isolated from attacked wood samples in the soil bed test described previously, were grown in 9-cm petri dishes filled with 20 ml of potato dextrose agar (PDA) medium containing none (control), 50 and 250 ppm of *A. malacophylla* heartwood extractives. Petri dishes were inoculated at the center by placing a 10 mm diameter fungi cut from the edge of an initially growing colony of fungus on PDA medium. The cultures were incubated in a sterile chamber at 22–24 °C for a period of 9 days. Fungal growth was evaluated daily by measuring two vertical diameters of the colony.

Each experiment was replicated three times. Growth was expressed as a percentage of the available space for growth according to Gerardin et al. 2004 as follows.

$$\text{Growth inhibition}\,\% = 100\left(1 - {}^{d_1}/_{d_0}\right)$$

where d_o and d_1 is the diameter of the untreated culture and culture with the extracts respectively.

Effects of extractives on wood dimensional stability

Twenty specimens from *A. malacophylla* heartwood and sapwood measuring 20 mm × 20 mm × 20 mm (dimensions measured to the nearest 0.01 mm) were cut. Sixteen heartwood specimens were soxhlet extracted, four in each of the solvents (hexane, dichloromethane, acetone and water as previously described) while four were not extracted. Similar procedure was applied for sapwood specimens. The purpose of extraction was to test whether extractives influences wood dimensional stability. All wood specimens solvent extracted or not were oven-dried at 60 °C to constant weights and initial volumes (V_0) evaluated. All specimens were then exposed to hydrated copper (II) sulphate solution. The specimens were weighed after every 48 h until constant mass was reached, indicating that they have attained maximum moisture content. The dimensions of the specimens were measured again and wet volume (V_1) determined. Swelling coefficient was calculated using the formula below:

$$\text{Swelling}\,(\%) = \frac{V_1 - V_0}{V_0} \times 100$$

The experiment was replicated three times.

FTIR analysis of heartwood extractives

1 µg of extract was mixed with a spatula end full of KBr, ground evenly in a mortor and pressed into disks. This was then examined in an FTIR between wavelength number ranges of 500–4,000 cm^{-1}.

Data analysis

Data was analyzed using Statistica version 7 for windows and analysis of variance (ANOVA). Pair wise comparison for dimensional stability was used to test for equality of means using two sample independent *t* tests. The analysis assumes that all the means comes from the same population.

Results and discussions

Wood dimensional stability

Figure 1 shown below reports the effects of solvent extraction of *A. malacophylla* wood on extractive yield and its dimensional stability.

A. malacophylla heartwood contains high amount of extractives (9.7 %) and is dimensionally stable (5.52 %) than sapwood, indicating strong hydrophobic nature of this wood species. Literature studies have attributed the dimensional stability of heartwood and high extractive content to wood natural durability (Kose and Taylor 2012; Neya et al. 2004)). Heartwood and sapwood dimensional

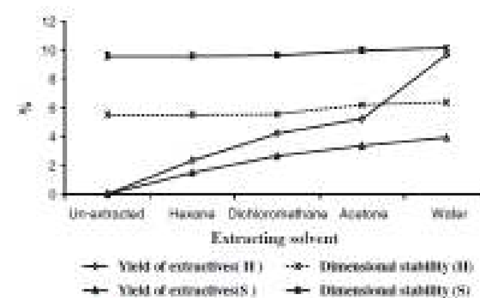


Fig. 1 Relationships of *A. malacophylla* heartwood (*H*) and sapwood (*S*) extractives and its dimensional stability

Author's personal copy

stability is significantly lowered ($p < 0.05$) through removal of extractives by different solvent extraction. The more polar solvents such as water and acetone extraction have more influence on both dimensional stability and extractive removal than the less polar hexane and dichloromethane ($p < 0.05$). This observation is in agreement with literature studies showing that polar solvents remove extractives located in the cell wall (Royer et al. 2010). Such extractives are known to bind to the polymeric cell wall by means of multiple hydrogen bonds making the wood dimensionally stable (Royer et al. 2010) and may be one of the factors contributing to its natural resistance.

Natural wood durability tests against fungi

Solvent extracted (water, hexane, dichloromethane, acetone, and unextracted *A. malacophylla* heartwood specimens were exposed alongside *P. patula* (a perishable species) to fungi in unsterile soil bed for a period of 32 weeks. The wood mass losses due to fungal degradation in the soil bed are reported in Fig. 2 below:

The first 4 weeks of exposure showed little variability in decay with percentage mass loss on all test specimens of about 2.3 %. This unclear decay pattern during initial period of wood exposure in soil bed test has been reported for other species (Acker et al. 2003). In the next 8 weeks, mass loss on solvent extracted specimens increased tremendously in comparison with the un-extracted specimens which still retained the least mass loss average of 2.6 %. Thereafter the mass loss increased immensely on the *P. patula* and the solvent extracted *A. malacophylla* wood specimens. In all the test specimens, mass losses due to fungal decay increased with exposure period with a significant difference amongst the treatments ($p < 0.05$). This is in agreement with observation that mass loss in softwoods is generally low in soil bed test (Acker et al. 2003; Machek et al. 2001). In our current experiment removal of extractives lead to significantly higher mass losses ($p < 0.05$), suggesting reduced decay resistance. It is therefore clear that *A. malacophylla* extractives contribute to wood natural resistance against fungi. Indeed, in their study on the decay resistance of Southern Asian timbers in sand block tests, Takahashi and Kishima (1973), observed that the non extracted specimens were more resistant to decay compared to solvent extracted specimens. Generally, the mass loss increased with decreasing solvent polarity implying that extractives soluble in low polar solvents such as hexane are mostly responsible for fungal decay resistance of *A. malacophylla*.

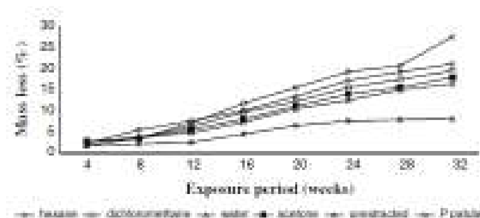


Fig. 2 Percentage mass losses of wood specimens in the fungal soil-bed

Fungal growth inhibition by *A. malacophylla* extractives

Unidentified tropical white rot fungus from the soil bed test experiment was cultured in PDA medium and inoculated with 0, 50 and 250 ppm of extractives. Fungal mycelium growth was measured daily from the centre of the petri dish and growth inhibition evaluated at the end of the experiment.

Figure 3 below shows percentage growth inhibition of hexane, acetone and water extracts of *A. malacophylla* heartwood at different concentration level.

Hexane extractives gave the highest inhibition activity against growth of fungi at 50 and 250 ppm concentrations in comparison with acetonic and water extracts at the same level of concentration. Literature studies have reported similar growth inhibition properties of heartwood extractives of other tropical wood species (Neya et al. 2004; Mburu et al. 2007; Sirmah et al. 2008). The development of fungi on PDA treated with different concentration of hexane extracts as a function of time was measured daily. The results are presented in Fig. 4a and b.

Fungi started growing on the PDA medium treated to 50 and 250 ppm hexane extract after 2 and 4 days respectively. Similar trend of fungal activity was observed in acetonic and water extracts (results not shown), suggesting fungistatic properties of *A. malacophylla* heartwood

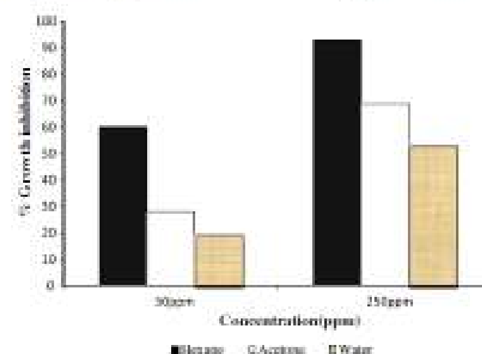


Fig. 3 Percentage growth inhibition of *A. malacophylla* heartwood extractives

Author's personal copy

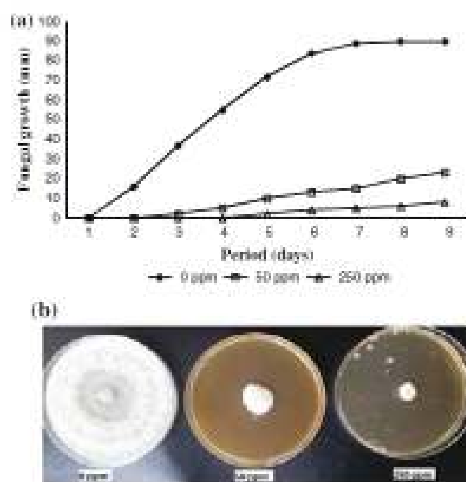


Fig. 4 Fungal growth on potato dextrose agar treated to different concentrations of hexane extractives. **a** Fungal growth as function of time in increasing hexane extract concentration. **b** Appearance of fungi after 9 days in increasing hexane extract concentration

extractives even at low concentrations. This observation is similar for most tropical hardwood species reported in literature (Neya et al. 2004; Mburu et al. 2007; Sirmah et al. 2008). Indeed it can be inferred that heartwood extractives of *A. malacophylla* restricts the growth of fungus thus contributing to the reported wood natural durability. Similar observations have been made on extracts of other wood species (Mohareb et al. 2010; Sirmah et al. 2008; Mburu et al. 2007; Martinez-iningo et al. 1999).

Natural wood durability tests against termites

Figure 5a below shows the physical appearance of wood specimens that were subjected to termite attack in the field for a period of 6 months while Fig. b presents the % mass loss of wood specimens over the same period of time.

The highest mean mass loss was observed in heartwood specimens extracted with dichloromethane (26.1 %) followed by those that were extracted by hexane solvent (22.4 %). The un-extracted specimens showed the least mass loss of (4.8 %) while the *P. patula* controls were severely attacked and had mass loss of 95 % after 6 months of exposure. Heartwood specimens extracted

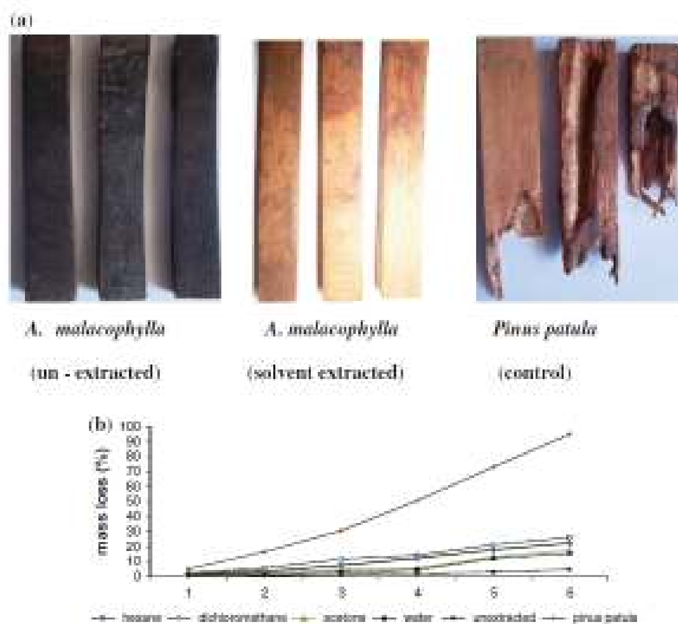


Fig. 5 **a** Extent of termite attack and **b** mass loss of wood specimens exposed to termites

Author's personal copy

with highly polar water solvent were least attacked by termites (15.4 %). It can be predicted that water would have removed polar extracts such as sugars and other soluble carbohydrates thus making the specimens less desirable to termite attack. Studies by Lukmandaru (2011) on the variability in the natural resistance of teakwood and its relationship with wood extractive content found that the lower the extractive content, the less severe the mass loss to termites. This was attributed to the removal of polar components that would have induced the termite activity in attacking and degrading the teakwood. In general, heartwood extractives of *A. malacophylla* especially those that are soluble in the less polar dichloromethane solvent contribute most to the natural resistance to termite attack.

FTIR analysis of heartwood extractives

FTIR Studies were carried out to understand the main functional groups present in the *A. malacophylla* heartwood extracts. Figure 6 below is an FTIR spectrum of dichloromethane heartwood extract.

The spectrum indicates an OH stretch at 3,055 cm^{-1} and a strong aliphatic stretch at 2,935 cm^{-1}. The C=C group absorption is indicated by vibrations at 3,687 cm^{-1}, typical of either an alkene or an aromatic compound, confirmed by strong vibrations at 1,431 and 1,608 cm^{-1} and C–H out of plane bending at 898 cm^{-1}. The strong carbonyl (C=O) absorption is evident at 1,724 cm^{-1}, corroborated by C–H absorption at 2,862 cm^{-1}. This indicates presence of aldehydes, ketones, carboxylic acids, esters or amines. The strong phenyl ring substitution band is evident at 736 cm^{-1}. Figure 7 below is an FTIR spectrum of acetonic heartwood extract.

The spectrum indicated a C=C group absorption at 3,525.6 and 3,625.6 cm^{-1}. The absorption at 2,927.7 cm^{-1} is attributed to the presence of aliphatic hydrogen while carbonyl group absorption at 1,720.4 cm^{-1} is confirmed to be an aldehyde at 2,788.9 cm^{-1}. Presence of alcohol was indicated by spectrum absorption at 3,525.6 and 3,602.8 cm^{-1}.

Conclusions

A. malacophylla heartwood is dimensionally stable (5.52 %) with a less dimensionally stable sapwood (9.6 %), indicating its strong hydrophobic nature. Heartwood contains high amount of extractives (9.7 %) in comparison to

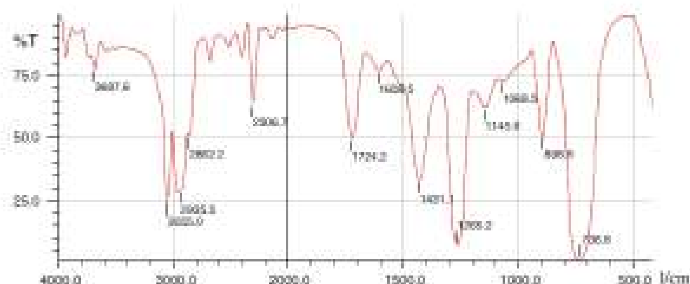


Fig. 6 FTIR spectrum of dichloromethane extractives

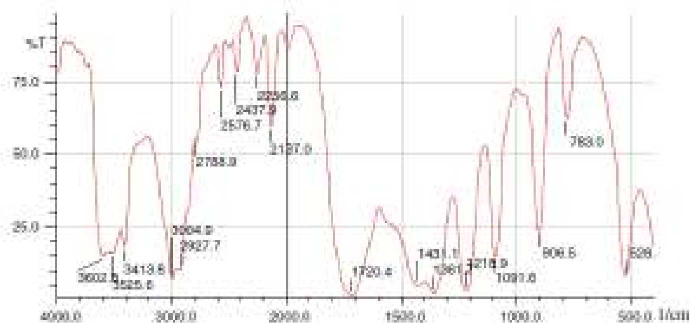


Fig. 7 FTIR Spectrum of acetone extractives

Author's personal copy

sapwood (4.6 %). Most of these extractives are soluble in the polar water solvent (9.7 %) while small amount are soluble in a non polar hexane solvent (2.36 %). Removal of extractives by solvent extraction makes this wood less dimensionally stable, less hydrophobic and prone to degradation by fungi and termites. *A. malacophylla* wood is durable against decay fungi in soil bed (8.1 %) mass loss and very resistant to termites in field (4.8 %) mass loss but in both cases resistance is lowered by solvent removal of extractives. Heartwood extractives inhibit the growth of fungi to some extent at different levels of concentration. Hexane extracts are effective as fungal growth inhibitors even at low concentration levels in comparison with water and acetone extracts. Infrared analysis indicated presence of aldehydes, ketones carbonyl compounds, esters, aromatic, carboxylic acids and aliphatic carbonyl compounds in the different extracts. Put together, the nature, amount of heartwood extractives, wood dimensional stability and its strong hydrophobicity are at the origin of the found termite and fungus resistance of *A. malacophylla* wood.

References

Acker J, Stevens M, Carey J, Sierra-Alvarez R, Militz H, Bayon I, Kleist G, Peek R (2003) Biological durability of wood in relation to end-use. Holz als Roh-Und Werkstoff 61:35–45

AWPA: E23-07 (2008) Accelerated method of evaluating wood preservatives in soil contact 2008

AWPA: E7-93 (1993) Standard method of evaluating wood preservatives by field tests with stakes 1993

Baeza J, Freer J (2001) Chemical characterization of wood and its components. In: David NS, Shiraishi N (eds) Wood and cellulosic chemistry, 2nd edn. Marcel Dekker Inc, New York, pp 275–384

Calonego F, Severo E, Furtado E (2010) Decay resistance of thermally modified *Eucalyptus grandis* wood at 140 °C, 160 °C, 200 °C and 220 °C. Bioresour Technol 101:9391–9394

Eggeling J, Dale R (1951) The indigenous trees of Uganda protectorate. Robert Maclehose and Company limited. The University Press, Glasgow

Gerardin P, Neya B, Dumarcay S, Petrissans M, Mohamed S, Huber F (2004) Contribution of gums to natural durability of *Prosopis africana* heartwood. Holzforschung 68:39–44

Githiomi J, Kariuki J (2010) Wood basic density of *Eucalyptus grandis* from plantations in Central Rift Valley, Kenya: variation with age, height level and between sapwood and heartwood. J Trop For Sci 22(3):187–195

Kose C, Taylor A (2012) Evaluation of mold and termite resistance of included sapwood in Eastern red cedar. Wood Fiber Sci 44(3):1–6

Kukachka B (1969) Properties of imported tropical woods. US Department of Agriculture, Forest Service. Forest Product Laboratory

Lukmandaru G (2011) Variability in the natural termite resistance of plantation teak wood and its relations with wood extractive content and color properties. J For Res 8(1):17–31

Machek L, Militz H, Sierra-Alvarez R (2001) The use of an acoustic technique to assess wood decay in laboratory soil-bed tests. Wood Sci Technol 34:467–472

Martinez-inigo M, immerzeel P, Gutierez A, Rio J, Sierra-Alvarez R (1999) Biodegradability of extractives in sapwood and heartwood from scots pine by sapstain and white rot fungi. Holzforschung 53:247–252

Mburu F, Dumarcy S, Huber F, Petricians M, Gerardin P (2007) Evaluation of thermally modified *Grevillea robusta* heartwood as an alternative to shortage of wood resource in Kenya. Characterization of physicochemical properties and improvement of bioresistance. Bioresour Technol 98:3478–3486

Mohareb A, Sirmah P, Desharnais L, Dumarcy S, Petrissans M, Gerardin P (2010) Effects of extractives on conferred and natural durability of *Cupressus lusitanica* heartwood. Ann For Sci 67:504

Neya B, Hakkou M, Petrissans M, Gerardin P (2004) On the durability of *Burkea africana* heartwood: evidence of biocidal and hydrophobic properties responsible for durability. Ann For Sci 61:277–282

Royer M, Garcia-Perez M, Duque-Fernandez A, Diouf P, Stevanovic T, Pouliot R (2010) Antioxidant, toxicological and antiproliferative properties of Canadian polyphenolic extracts on normal and psoriatic keratinocytes. J Ethnopharmacol 132(1):251–258

Schubert M, Volkmer T, Lehringer C, Schwarze F (2011) Resistance of bio-ionised wood treated with wood preservatives to blue-stain and wood-decay fungi. Int Biodeterior Biodegrad 65:108–115

Silva C, Monteiro M, Brazolin S, Lobez G, Richter A, Broga M (2007) Biodeterioration of Brazilwood *Caesalpinia echinata* Lam. (Leguminosae-Caesalpinioideae) by rot fungi and termites. Int Biodeterior Biodegrad 60:285–292

Sirmah P, Muisu F, Mburu F, Dumarcay S, Gerardin P (2008) Evaluation of *Prosopis juliflora* as an alternative to wood shortage in Kenya. Bois et Foret des Tropiques 298(4):25–35

Sirmah P, Mburu F, Laych K, Dumarcy S, Gerardin P (2011) Potential antioxidant compound from different parts of *Prosopis juliflora*. J Trop For Sci 23(2):187–195

Sun Q, Yu H, Li J, Lu Y, Hunt J (2010) Improvement of water resistance and dimensional stability of wood through titanium dioxide coating. Holzforschung 64:757–761

Takahashi M, Kishima T (1973) Decay resistance of 65 South East Asian timber specimens in accelerated laboratory tests. Tonan Ajia Kenkyu 10:524–541

Umezawa T (2001) Chemistry of Extractives. In: David NS, Shiraishi N (eds) Wood and cellulosic chemistry, 2nd edn. Marcel Dekker Inc, New York, pp 213–241

Williams R, Miller R, Gangstad J (2001) Characteristics of ten tropical hardwoods from certified forests in Bolivia, part 1 weathering characteristics and dimensional change. Wood Fiber Sci 33:618–626

World Conservation Monitoring Centre (1998) *Albizia malacophylla* var malcophylla. In: IUCN red list of threatened species, version 2012.2. http://www.IUCNredlist.org. Accessed 12 Nov 2012