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B. Sc. (Hons.) Thesis
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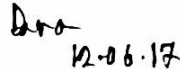
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DECLARATION

This is **Nabila Hasan Dana, Student ID- 120511**, hereby declare that the project thesis is the result of my own works and has not been submitted or accepted for any other degree at other institutions.

Nabila Hasan Dana

Date:

Dedicated

To

My Beloved Parents

ACKNOWLEDGEMENT

Abstract

This study assesses the decay resistance properties of bio- preservatives modified wood against decay fungi of the Basidiomycota. Small wood blocks from sapwood of *Anthrocephalaus chinensis* and *Bombax ceiba* were vacuum impregnated with neem leaf extract (NLE) and goran bark extract (GBE) and tested for decay resistance against the white rot fungi *Trametes versicolor*. The relationship of fungal species, extent of preservatives treatment (WPG) and mass loss (ML) induced by decay were examined. The ML caused by the decay fungi, however decreased with the increasing WPG in all of the wood species against all of the fungi tested. The increased decay resistance is attributed to the better impregnation of preservatives either in the wood cell wall or in the cell lumen providing a barrier to decay fungi. All the wood species modified with NLE and GBE enhanced the hydrophobic nature of wood.

Keywords: Bio- preservative, tannin, *Ceriops decandra* (Griff.) bark, *Azadiracta indica* (A. Juss) leaf extract, hot water extraction, decay test.

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CHAPTER ONE

Introduction

1.1 Background of the study

Wood is a renewable natural resource. From the ancient period, wood has a great demand in our day to day life. With the increasing population, the demand of wood is also increasing (Islam, 2006). Wood has also a great role in our industrial based economy, especially in the construction fields (Gonzalez-Laredo et al, 2015). Coming about the importance of wood products, the protection and utilization of wood has been practicing with the development of wood science (Freeman et al, 2003). Wood preservation is such kind of field for wood protection in wood science. The principle of the science and practice of wood preservation is to increase the service life of wood (Srivastava, 1969). Wood is an important building material. Like other building materials, wood deteriorates with the passing of time during service. Wood is deteriorated by the infestation of fungi, insects, marine borers and the action of fire (Srivastava, 1969).

Wood has some important natural properties such as, renewable resources, electrical insulation and sound absorption properties, high salvage value, low cost, low weight, low thermal conductivity, ease to work and high strength. Considering these properties, wood act as a valuable constructional material. Improvement of these natural properties by using modern technology, wood can be the earth most valuable constructional material. Wood is a biological product and for this reason, wood is liable to deteriorate unless it is properly preserved. The major issues of wood deterioration in service are decay due to fungal infestation, insects and marine organisms attack. Among these decay is one of the most important factor for the wood deterioration in service. Wood preservation is the art of preserving wood against the wood deteriorating agent (Srivastava, 1996). Wood preservation process requires some preserving agent for wood protection and these preserving agent called preservatives. Chemical and bio- preservatives are such type of preserving agents for wood preservation treatment. Chemical preservatives are of synthetic origin. Creosote, chromated copper arsenate (CCA), acid copper chromate (ACC), boric acid are the example of chemical preservatives. Bio- preservatives are of biological origin. Extract from various plant part (e.g. leaves, seed, and bark), cashew nut shell liquid, resin, tannin are the example of bio- preservatives.

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With the development of wood based industries, the use of wood preservatives is also increased adversely. Chromated copper arsenate (CCA), creosote, pentachlorophenol are some most common wood preservatives which are used by wood industries for expanding service of wood products for many decade. But from last few years environmentalist has restricted the use of these preservative concerning about the environment and health (Ashaduzzaman et al, 20013). On the other hand CCA, creosote, etc. have proven as effective wood preservative. However, completely removing such effective but toxic preservatives will take time (Schultz and Nicholas, 2007) and require considerable investment into the exploration of suitable sources of potent bio-active wood preservative.

Traditional wood preservation methods employ chemicals that are considered toxic and can adversely affect human health and the environment. Extending the service life of wood and wood products using natural compounds as bio-actives is proving to be an effective approach for wood protection from the prospective of human health and environmental protection (Laks, 1989; Freitag et al, 200; Schultz and Nicholas, 2002; Evans, 2003). Interest in the use of natural products as biocides is rapidly growing worldwide. Plant produces some aromatic and non- aromatic compounds and some of them are recognized as antifungal agents. Phenol, terpenoids, alkaloids, tannin are the products that have been extensively utilized in various applications (Geissman, 1963).

Extracts from plant leaves, especially *Azadiracta indica* leaves have showed highly effective result against wood decay fungi, like white rot and brown rot fungi and can potentially be developed into excellent organic preservatives (Lin et al, 2007; Yang and Clausen, 2007; Lotz, 1993). Bark from many trees, especially mangrove species are rich in antifungal agents, such as waxes, resins, tannin, etc. Tannin has been widely used as adhesives and also used as wood preservatives.

However, the application of plant based compounds for wood preservation became less attractive to the wood industrialist, because synthetic compounds were showed more effective result for wood protection. But now, there is a pressing need to replace synthetic compounds with bio-preservatives because of their toxicity to human health and detrimental impact on the environment. Derivatives from various plant parts, such as bark, wood, leaves, seeds and fruits, have been examined for their wood protection properties in many studies (Yang, 2009). The objective of this

study is to evaluate the potency of plant extracts as a bio- preservatives for better utilization of wood products.

1.2 Objectives of the Study

Population density is becoming higher and higher day by day. Wood is one of the Earth's most useful product. Wood is used both as fuel and constructional materials. The demand of wood products are also increasing with the increasing population demand. For this reason, preservation of wood is the best way to utilize wood properly and also to conserve the world forest. There are lots of preservative treatments had already been introduced for better utilization of wood products and so as the preservatives also introduced. So many chemical preservatives are used for preservative treatments. But now, those chemical compounds are declared as toxic compounds that are harmful for human health and the environment. Science and technology of wood are now moving towards the use of biological compounds for wood preservation. Extractives from plant parts are using for wood protection and proving themselves as effective compounds for wood preservation (Islam, 2006). The general objective of this study is to evaluate the effectiveness of plant extract as bio- preservatives for wood preservation. Specific objectives of this study are given below:

1. To extract *Azadiracta indica* leaf extracts and *Ceriops decandra* bark extracts
2. To assess the potentiality of these plant extracts as wood preservative to improve the decay resistance properties of less durable wood species.

CHAPTER TWO

Literature Review

2.1 Wood Preservation

In wood science, wood preservation means the action of keeping wood in an unchanged form for a long time. Wood is biodegradable. Because of its natural origin and its organic nature it is an ecological habitat for a wide range of insect and fungi. These organisms fed on the food stored in the wood cell or on the cell wall materials causing degradation and loss strength. It is therefore, needed to protect wood from such destructive agent by treating wood with some preservatives (Shrivastava, 1997).

The treatment of wood with a mixture of chemicals to protect it from wood destroying organisms, like insects, fungi and marine borers is referred to as wood preservation (PNGS, 1989). Wood preservation in the widest sense implies the protection of wood against any factors that may damage it. Wood preservation is the action of improving wood's natural strength by treating them with chemicals that are toxic to the wood destroying organisms. It has made available the use of large number of species which previously had been used inferior only because of their less durability and gave only short service when they are exposed. In view of the limited availability of naturally durable species, it is therefore important to increase the service life of less durable wood with proper preservatives treatment (FAO, 1986).

2.2 Wood preservatives

The toxic chemicals that are applied to preserve wood are known as wood preservatives (Shrivastava, 1997). The term preservatives is initiated to include such chemicals that are used to protect wood against deterioration by decay, insects, marine borers, fire, weathering, water absorption and chemical action (AWPA, 1996).

Wood preservatives vary widely in origin, character, cost effectiveness and suitability for use under different conditions of service. The characteristics that required for an effective wood preservatives are: highly toxic toward wood destroying organism, non- leachable, ability to penetrate deeply into wood, non- corrosive to metals, colorless, odorless and paintable (FAO,

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1986). Depending on the origin preservatives may be of synthetic preservatives or bio-preservatives. Synthetic preservatives are of artificial origin and they are mostly chemical preservatives. Bio- origin preservatives are of bio- preservatives.

2.3 Wood Preservation Process

The operation by which a wood preservative is applied to or into the wood for preserving purpose is known as wood preservation process (Lahiry, 2001). The objective of wood preservation treatment is to introduce the preservative into the wood to prevent decay and insects attack and improve its service. Various treatment can be used depending on the wood species and the end use (FAO, 1986).

The preservation treatment may be grouped either in pressure process or non- pressure process. Non- pressure processes are carried out without the use of artificial pressure while pressure processes are in which the wood is placed in a cylinder and impregnated with preservative by applying pressure or vacuum.

Pressure Process

Wood is most often treated by immersing it in a preservative in a high-pressure apparatus and applying pressure to drive the preservative into the wood. Pressure processes differ in details, but the general principle is the same. Three pressure processes are commonly used: full cell, modified full cell, and empty cell.

Full cell process- The full-cell process is used when the retention of a maximum quantity of preservative is desired. It is a standard procedure for timbers to be treated with creosote when protection against marine borers is required. Water-borne preservatives may be applied by the full cell process if uniformity of penetration and retention is the primary concern. With waterborne preservatives, control over preservative retention is obtained by regulating the concentration of the treating solution.

Modified full cell process- The modified full-cell process is basically the same as the full-cell process except for the amount of initial vacuum and the occasional use of an extended final vacuum. The modified full-cell process uses lower levels of initial vacuum; the actual amount is

determined by the wood species, material size, and final retention desired. The modified full-cell process is commonly used for treatment of lumber with water-borne preservatives.

Empty Cell- The objective of the empty-cell process is to obtain deep penetration with a relatively low net retention of preservative. For treatment with oil preservatives, the empty-cell process should always be used if it will provide the desired retention. Two empty-cell processes, the Rueping and the Lowry, are commonly employed; both use the expansive force of compressed air to drive out part of the preservative absorbed during the pressure period.

Non-pressure Process

The numerous non-pressure processes differ widely in the penetration and retention levels of preservative attained, and consequently in the degree of protection they provide to the treated wood. Non-pressure methods, in general, consist of (a) surface application of preservatives by brief dipping, (b) soaking in preservative oils or steeping in solutions of waterborne preservatives, (c) diffusion processes with waterborne preservatives, (d) vacuum treatment, and (e) a variety of miscellaneous processes.

Vacuum process- In the vacuum process, wood products are enclosed in an airtight container from which air is removed with a vacuum pump. The container then is filled with the preservative without additional pressure and without the air re-entering. The partial removal of air from the wood, by the vacuum, followed by addition of the preservative creates a slight pressure that drives the preservative into the wood.

2.4 Wood Destroying Fungi

Both the sapwood and heartwood of most tree species are susceptible to decay. Decay fungi may grow in the interior of the wood or appear on wood surfaces. Wood decay fungi can be grouped into brown rot, white rot, and soft rot.

Brown rot- fungi that cause brown rot, are able to break down the cellulose component of wood for food, leaving a brow n residue of lignin. Brown-rotted wood can be greatly weakened even before decay can be seen. *Coniophora puteana* and *Postia placenta* are brown rot fungi.

White rot- white rot fungi, which break down both lignin and cellulose, have a bleaching effect, which may make the damaged wood appear whiter than normal. *Trametes versicolor* and *Pleurotus ostreatus* are white rot fungi.

Soft rot- soft rot fungi usually attack green wood with high moisture content (MC), causing a gradual softening from the surface inward that resembles brown rot. *Kretzschmaria deusta* is a soft rot fungi.

2.5 Commonly Used Chemical Wood Preservatives and Their Associated Risk to Human Health and the Environment

Creosote

Creosote is a distillate of coal tar and a heavy oily liquid. Creosote improves the weathering characteristics of wood, provides protection from insects and fungi, and promotes insolubility in water. It is used in railroad ties, large timbers, fence, posts, poles, and pilings.

Creosote may causes skin cancer in persons regularly exposed to it. Prolonged and repeated exposure may lead to dermatitis (inflammation of the skin). The liquid and vapors can irritate eyes and the respiratory tract. It may seep from wood into the environment and these may be a reason chronic toxicity in aquatic organism.

Chromated Copper Arsenate (CCA)

Chromated copper arsenate (CCA) is composed of the oxides of chromium, copper and arsenic. The copper serves as the primary fungicide; the arsenic serves as a fungicide and insecticide; chromium fixes the copper and arsenic in the wood. Used in fence posts, railroad ties, etc.

Primary concern with CCA treated wood is the arsenic. Arsenic is a highly toxic metal which can cause acute toxicity, chronic toxicity.

Pentachlorophenol

Pentachlorophenol is a synthetic substance and a water-repellent solutions containing chlorinated phenols. Pentachlorophenol is effective against many organisms, such as decay fungi, molds,

stains, and insects. Because pentachlorophenol is ineffective against marine borers, it is not recommended for the treatment of marine piles or timbers used in coastal waters. Pentachlorophenol is used industrially as a wood preservative for utility poles, railroad ties, and wharf pilings.

Pentachlorophenol is released to the air by evaporation from treated wood surfaces and factory waste disposal. It enters surface water and groundwater from factories, wood-treatment facilities, and hazardous waste sites. It also enters the soil as a result of spills, disposal at hazardous waste sites, and its use as a pesticide. These chemical compound move much with water and generally stick to soil particles than it evaporates. The compound can be present in fish or other species used for food. In air, soil, and surface water, pentachlorophenol lasts for hours to days. The compound is broken down in soil and surface water by microorganisms, and in air and surface water by sunlight, to other compounds, some of which may be harmful to humans. Pentachlorophenol is considered a probable human carcinogen.

2.6 Evaluation of Bio- preservatives

Wood degradation occurs due to microbial agencies and termites causing significant losses. Since the supply of wood is limited, it is necessary to protect the wood in service from biological deterioration. From time began, attempts were made by many workers to impart durability by treating the wood with natural and synthetic chemicals (Purushotham, 1970). The conventional wood preservatives although found to be very effective against wood destroying organisms, are said to cause environmental pollution and a few of them are hazardous to human beings (Fisher, 1968; Thompson, 1971; Onuorah, 2000). For this reason, there has been substantial global awareness to develop eco-friendly wood preservatives and those, which do not cause any bad effect on human health and the environment (Onuorah, 2000). There is a continuous search for bio-preservatives for wood preservation.

Green plants act as a source of innocuous fungicides or pesticides, which are non-toxic and easily biodegradable than synthetic chemicals. To develop eco-friendly wood preservatives, many studies have been conducted. Most of the reported work is on extractives from plant parts (Onuorah, 2000; Soni, 1975; Gupta and I.Dev, 1999). Neem (*Azadiracta indica*) possess a number

of toxic constituents exhibiting high toxicity against wood destroying microbes (Swathi *et al.*, 2004) and many mangrove species are good source of tannin, which has anti- fungal property.

One possible approach for developing new wood preservatives is to study the heartwood of naturally durable wood species, since an understood of the causes for natural durability might suggest alternative ways to protect limber. Consequently, we have studied the role which extractives, particularly stilbenols, play in the natural durability of heartwood (Schultzeral *et al.*, 1995). In continuing this reasoning, phenolic extractives have the ability to complex with metals, i.e., extractives are metal chelators. It was concluded that the combination of an organic biocide with metal chelating and/or antioxidant additives gives enhanced protection to wood against fungi as compared to the biocide alone and consequently, it may be possible to develop environmentally-benign wood preservative system based on this idea (Schultz and Nicholas, 2002).

Natural resource materials, like wood vinegar and bamboo vinegar are not used in wood industries, but used as folk medicine for skin diseases in Japan. Wood vinegar has been used in agricultural fields, food industries and in household cleaning. Anti- dermatophyte activity of phonolic compounds in wood vinegar was already demonstrated. To formulate the new environmental friendly wood preservative, scientist neutralized (up to pH 7) wood vinegar with NaOH and hot water extracts of *Pinus densiflora* and *Quercus serrate* saw dusts. This mixture could be a very good substitution for chemicals used to control sap staining fungi in the field (Velmurugan *et al.*, 2009).

The use of citrus waste as insecticide and/or fungicide has not been developed until now. Its activity is still in an initial stage of knowledge, but some studies published during the last years allow to foresee that essential oils, flavones and some phenolic compouds from citrus show some kind of biocide activity.

Neem (*Azadirachta indica*, A. Juss) is popularly known as village pharmacy, as all parts of this plant are used for several types of diseases since centuries. Extract of leaves exhibit the property of anti- bacterial, antifungal, antiviral (Tewari, 1992), anti- malarial (Chavan *et al.*, 1988), etc. Leaf extract also known to inhibit the growth of plant pathogens (Bhatnagar *et al.*, 1988; Kurucheve *et al.*, 1997) and wood decay fungi.

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Tannins are complex chemical substances derived from phenolic acids (sometimes called tannic acid). They are classified as phenolic compounds, which are found in many species of plants, especially in mangrove species such as *Rhizophora* spp. Tannin are found commonly in the bark of tres. *Ceriops decandra* bark is a common source of tannin. Tannins that stored in the bark of trees to protect the tree from being infected by bacteria or fungi. In this case, tannins precipitate out the enzymes and other protein exudates from bacteria and fungi thus not allowing these organisms to infect the tree. By using this properties, tannin can be a good eco- friendly wood preservative.

2.7 Evaluating New Wood Preservatives

Wood preservatives often need to provide protection from a wide range of wood-attacking organisms (fungi, insects, marine borers, and bacteria). Because they must protect wood in so many ways, and protect wood for a long time period, evaluating wood treatments requires numerous tests. Some of the most important tests are mentioned here, but they should be considered only as a minimum, and other tests are useful as well. Appendix A of the AWPA Standards provides detailed guidelines on the types of tests that may be needed to evaluate new wood preservatives.

The **laboratory leaching test** helps to evaluate how rapidly the treatment will be depleted. A treatment needs leach resistance to provide long-term protection. In this test small cubes of wood are immersed in water for 2 weeks.

The **laboratory decay test** is used to challenge the treated wood with certain fungal isolates that are known to aggressively degrade wood. It should be conducted with specimens that have been through the leaching test. The extent of decay in wood treated with the test preservative is compared to that of untreated wood and wood treated with an established preservative. This test can help to determine the treatment level needed to prevent decay.

Field stake evaluations are some of the most informative tests because they challenge the treated wood with a wide range of natural organisms under severe conditions. Stakes are placed into the soil in regions with a warm, wet climate (usually either the southeastern United States or Hawaii). At least two different sites are used to account for differences in soil properties and types of organ

-isms present. The extent of deterioration in wood treated with the test preservative is compared to that of untreated wood and wood treated with an established preservative.

Above-ground field exposures are useful for treatments that will be used to protect wood above ground. Although not as severe as field stake tests, above-ground tests do provide useful information on above-ground durability. Specimens are exposed to the weather in an area with a warm, wet climate (usually either the southeastern United States or Hawaii). The specimens are designed to trap moisture and create ideal conditions for above-ground decay. The extent of deterioration in wood treated with the test preservative is compared to that of untreated wood and wood treated with an established preservative.

Corrosion testing is used to determine the compatibility of the treatment with metal fasteners.

Treatability testing is used to evaluate the ability of a treatment to penetrate deeply into the wood. Shallow surface treatments rarely provide long-term protection because degrading organisms can still attack the interior of the wood.

Strength testing compares the mechanical properties of treated wood with matched, untreated specimens. Treatment chemicals or processes have the potential to damage the wood, making it weak or brittle.

CHAPTER THREE

Materials and Methods

3.1 Goran Bark Collection and preservative formation

The bark of goran (*Ceriops decandra* Griff.) was collected from the Sundarbans West Forest Division which is situated approximately between 22°24'- 22°26' north and 89°23'- 89°22' east. The average age of the trees was around 8- 9 years. The bark was collected from the main stem of the trees, usually from the bottom part of the main stem, usually below the breast height (1.3 meter). After collecting the bark, nuisance from collected bark was removed by hand peeking, then washed in clean water to remove mud, sand, etc. and to get clean bark. Then the clean bark were chopped to get smaller size and sun dried for two days. These dried particles then blended to get powder form of bark. Then these bark powder were screened by using a sieve having 200 micro mm^2 pore openings to remove larger particles.

Goran (*Ceriops decandra*) bark extracts were extracted followed by a hot water extraction method. The powdered bark were mixed with distill water at a ratio of 7.5:1 (v/w) to get a solution, rather than the standard ratio of 5:1 (v/w). Then the solution was cooked in a cooking pan with lid for 3 hours at a temperature of 80±5°C. The cooked solution was cooled down at room temperature and filtered with a 100 micro mm^2 openings sieve. The extract solution was then evaporated in a horizontal laminar flow chamber to get solid content of the extract. The solution was stored in a refrigerator at a temperature of -4°C for farther applications.

3.2 Neem Leaf Collection and Preservative Formation

The neem (*Azadiracta indica* A. Juss) leaf was collected from the Khulna University campus. The mature leaves were collected from the living trees of *A. indica* and the age of the trees was around 6- 7 years. The collected leaves were washed with clean water to clean the leaves, then sun dried for one day. The sun dried leaves were ground in blender to powdered it and the powder was screened with a 200 micro mm^2 sieve openings to get even size leaf particles.

Extraction of *A. indica* leaf extract was done by hot water extraction method as followed for the extraction of goran (*Ceriops decandra*) bark extracts.
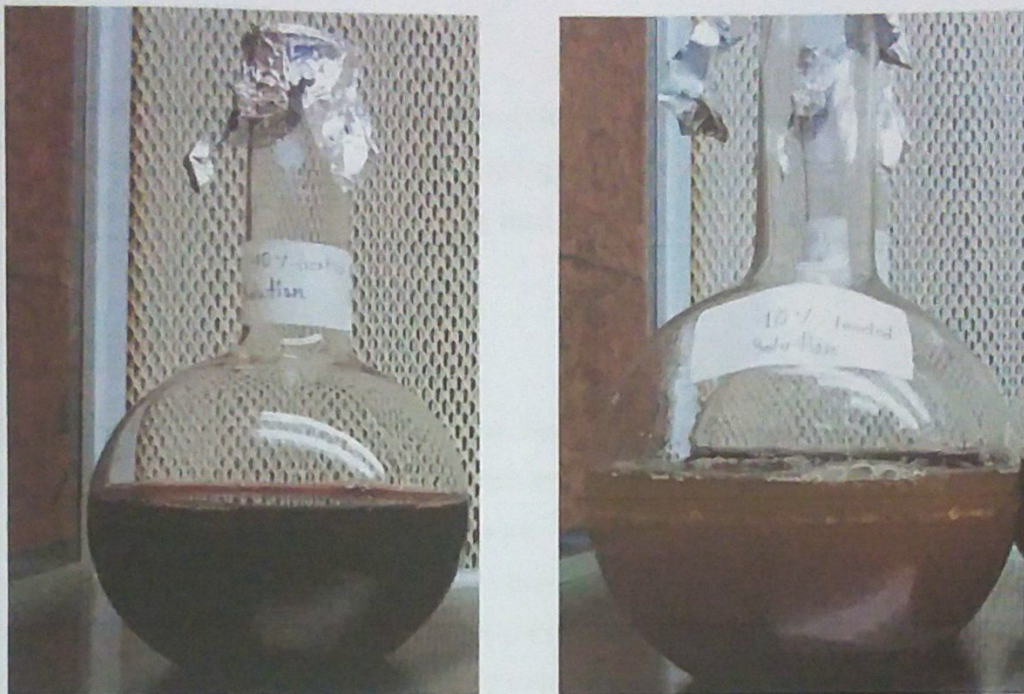
Figure 1: Goran bark and neem leaf extract

3.3 Collection and Preparation of Wood Samples

Wood samples of kadam (*Anthrocephalaus chinensis* Lam.) and shimul (*Bombax ceiba* L.) were collected from local sawmill. Wood samples were collected from the sap portion of a felled log for both of the species. Sapwood samples of *A. chinensis* and *B. ceiba* were collected from a felled log having straight bole which was knot, sap stain, spiral grain and other defects free. Wood samples were collected in a smaller size wood plank from the sawmill. Sapwood planks were collected usually above the breast height (1.3 meter) of the log.

Wood planks were sawn into smaller size wood blocks by using vertical band saw in the Wood Workshop of Forestry and Wood Technology Discipline. The wood block dimensions were of 20 × 20 × 5 mm (radial × tangential × longitudinal). The edges of the wood blocks were sanded for batter finishing. The sanded wood blocks were oven dried at 103±2 °C for 24 hours to get moisture

content (MC%) about 4%. Then the dry wood blocks were stored in air tight jar for later applications.

3.4 Collection Wood Decaying Fungi

Wood decaying white rot fungi *Trametes versicolor* were used in this treatment. The white rot fungi *T. versicolor* were collected from the Pathology laboratory of Forestry and wood Technology Discipline. The cultures of *T. versicolor* were maintained on 2% malt agar medium at 22°C and 65% relative humidity (RH).

3.5 Preservatives Impregnation

Vacuum pressure impregnation method was used to impregnate the aqueous solution of preservatives into wood blocks. Oven dried treated wood blocks were vacuum pressure impregnated with aqueous solution of *C. decandra* bark extract and *A. indica* leaf extract of 0%, 10%, 20% and 40% solute content to achieve different amounts of weight percent gain (WPG). Vacuum desiccator was used to impregnate the preservatives at a -0.9 bar vacuum for 20 minutes. The wood blocks were kept in the impregnating solution for 2 hours for intake of *C. decandra* bark extract and *A. indica* leaf extract solution into wood samples. Impregnation was done separately for different species wood blocks. The impregnated wood blocks were then weighed after wiping out the excess solution from the wood blocks. Curing of wood blocks were then done in an oven at 103±2°C for 24 hours. After 24 hours, cured wood blocks were cooled and reweighed again to calculate weight percent gain (WPG). Weight percent of treated wood blocks were calculated followed by the standard formula of weight percent gain, which is:

$$WPG\ (\%) = [\frac{(W_1 - W_0)}{W_0}] \times 100$$

Where, W_1 = Oven dry weight of treated wood

W_0 = Oven dry weight of un-treated wood

Figure 2: Vacuum treatment and preservative retention

Figure 3: Wood samples after preservative treatment

3.6 Wood Decay Test

Treated and un- treated wood blocks were exposed to white rot (*T. versicolor*) fungi to test their decay resistance properties. For this treatment, *T. versicolor* fungi were grown on 60ml of 4% malt agar medium in 500ml squat glass jars at 22°C and 65% RH for 2 weeks prior to the exposer of wood blocks of both species. After covering the malt agar medium with mycelia mat, wood blocks were exposed to decay for 12 weeks. Treated and un- treated (control) wood blocks were exposed to decay in squat jars separately. For each treatment, 6 replicates (n= 6) were exposed to decay for both of the species and as for the control too.

After 12 weeks wood blocks were taken away from the mycelia mat and reweighed. Prior to reweight, wood blocks were wiped to remove surface mycelia. Then wood blocks were oven dried at 103±2°C for 24 hours. Oven dried wood blocks were then cooled and reweighed to determine the mass loss and decay moisture content. These were calculated by using standard formula (WSE, 2013) and expressed as percentage weight loss (WL %) and decay moisture content:

$$WL \% = \left[\frac{(W_1 - D_o)}{W_1}\right] \times 100$$

Where, W_1= treated oven dry weight of wood (before 12 weeks)

D_o= decayed oven dry weight of wood with fungi attack (after 12 weeks)

Figure 4: Wood sample in *T. versicolor* cultured media

Figure 5: Wood sample after 12 weeks

3.7 Statisticall Analysis

All the data, obtained during the 3.7 laboratory tests for assessing the potentiality of *C. decandra* bark extract and *A. indica* leaf extract as wood preservatives to improve the decay resistance properties of wood sample were analyzed by using Microsoft Office Excel 2013 and SAS software. ANOVA (Analysis of Variance) and LSD (Least Significant Difference) were performed at 5% significance level.

CHAPTER FOUR

Results and Discussions

4.1 Results

4.1. a Weight Percent Gain (WPG%)

Results of neem (*A. indica*) leaf extract (NLE) impregnation of two different species *A. chinensis* and *B. ceiba* are *A. chinensis* 2.58, 4.55, 6.65 and *B. Ceiba* 1.94, 4.42, 6.44 for 10%, 20% and 40% solutions which are present in here as mean weight percent gain (WEP%) of the treated wood blocks based on the dry weight after curing and oven- dry weight recorded prior to treatment. Treatment of wood blocks of two different species with NLE was done in four different concentrations 0% (control), 10%, 20% and 40%.

From the annalysis, it has been found that higher weight percent gain (WPG%) of neem leaf extract was in *A. chinensis* 6.56% and lower weight percent gain was in *B. ceiba* 1.94%. The highest weight percent gain was found in 40% neem leaf extract treated wood blocks of two different species *A. chinensis* 6.56% and *B. ceiba* 6.44%.

Statistical analysis showed that, there was no significant difference between two species treated with 20% and 40% neem leaf extract. But there was a different between two species treated with 10% NLE. From the analysis, there was significant difference between the treatments 10%, 20% and 40% NLE (P= 0.05). The graphical presentation of the treatments and WPG% is showed in **Figure 6.**

The mean values of weight percent gain (WPG%) of goran (*C.decandra*) bark extract treated wood blocks of two different species are *A. chinensis* 2.55, 4.60, 5.67 and *B. Ceiba* 2.78, 4.72, 5.79 for 10%, 20% and 40% solutions. The values are presented for different concentrations of goran bark extract solution (GBE) 0% (control), 10%, 20% and 40%.

From the analysis, it has been found that higher weight percent gain (WGP%) of gorn bark extract was in *B. ceiba* 5.79% and lower in *A. chinensis* 2.55%. The highest weight percent gain was found in 40% Goran bark extract treated wood blocks of two different species *B. ceiba* 5.79% and *A. chinensis* 5.67%.

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Statistical analysis showed that, there was no significant difference between WEG (%) two species wood blocks treated with 10%, 20% and 40% goran bark extract. But there was significant difference between the treatments (P= 0.05). The graphical presentation is figured in **Figure 7**.
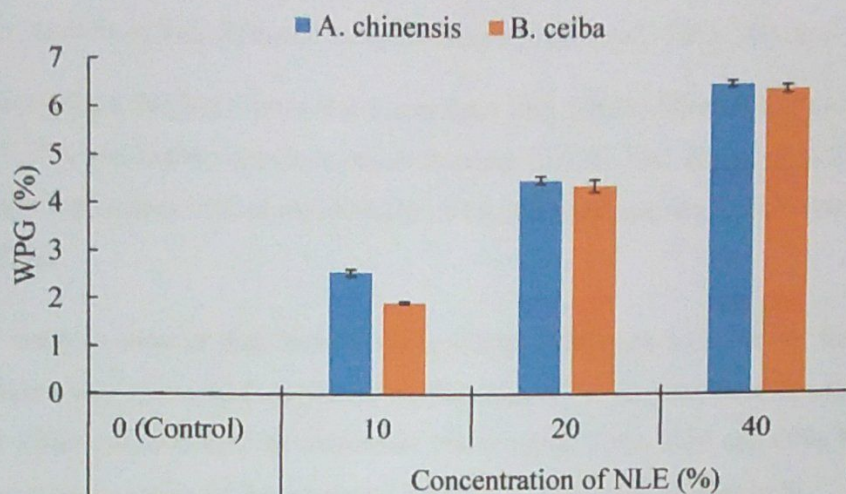

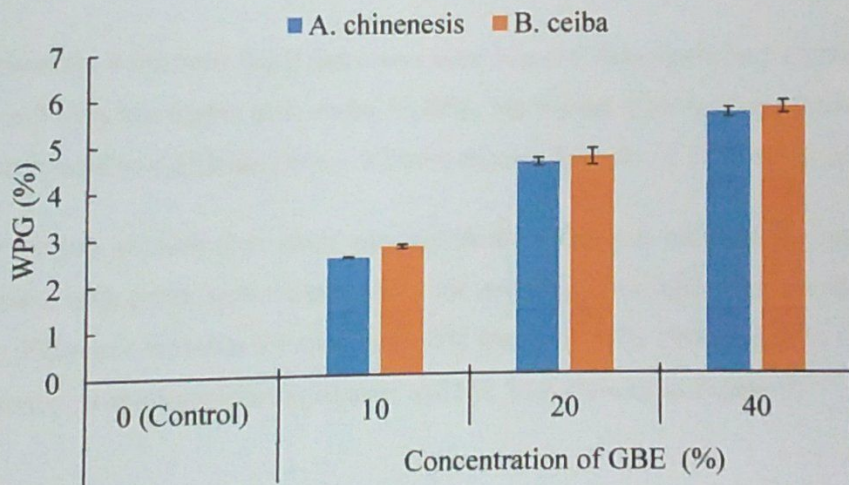**Figure 6:** Weight percent gain (WPG%) of neem leaf extract (NLE) for two different wood species

Figure 7: Weight percent gain (WPG%) of goran bark extract (GBE) of two different wood species

4.1. b Mass Loss (ML%)

The meam mass loss (%) of NLE treated decayed wood blocks of two different species after 12 weeks are *A. chinensis* 15.09, 12.33, 10.75, 9.77 and *B. ceiba* 16.93, 13.65, 12.80, 12.27 for control (0%), 10%, 20% and 40% solutions. Treatment of wood blocks of two different species with NLE was done in four different concentrations 0% (control), 10%, 20% and 40%.

From the analysis, it has been found that lower mass loss (ML%) of neem leaf extract was in *A. chinensis* 9.77% and higher mass loss was in *B. ceiba* 16.93%. The lowest mass loss was found in 40% neem leaf extract treated wood blocks of two different species *A. chinensis* 9.77% and *B. ceiba* 12.27%.

Statistical analysis showed that, there was significant difference between the mass loss of two species treated with neem leaf extract. From the analysis, it has also been found that there was significant difference between the treatments 0% (control) 10%, 20% and 40% NLE (P= 0.05). The graphical presentation of the treatments and ML % is showed in **Figure 8**.

The meam mass loss (%) of goran bark extract (GBE) treated decayed wood blocks of two different species after 12 weeks are *A. chinensis* 15.09, 12.52, 1.29, 9.29 and *B. ceiba* 16.93, 13.20, 12.32, 12.28 for control (0%), 10%, 20% and 40% solutions. The values are presented for different concentrations of goran bark extract solution (GBE) 0% (control), 10%, 20% and 40%.

From the analysis, it has been found that lower mass loss (ML%) of gorn bark extract was in 5.79% *A. chinensis* 9.29% and higher in *B. ceiba* 16.93%. The lowest mass loss was found in 40% goran bark extract treated wood blocks of two different species *B. ceiba* 12.28% and *A. chinensis* 9.29%.

Statistical analysis showed that, there was significant difference between the mass loss of two species treated with goran bark extract. From the analysis, it has also been found that there was significant difference between the treatments 0% (control) 10%, 20% and 40% GBE (P= 0.05). The graphical presentation of the treatments and ML % is showed in **Figure 9**.
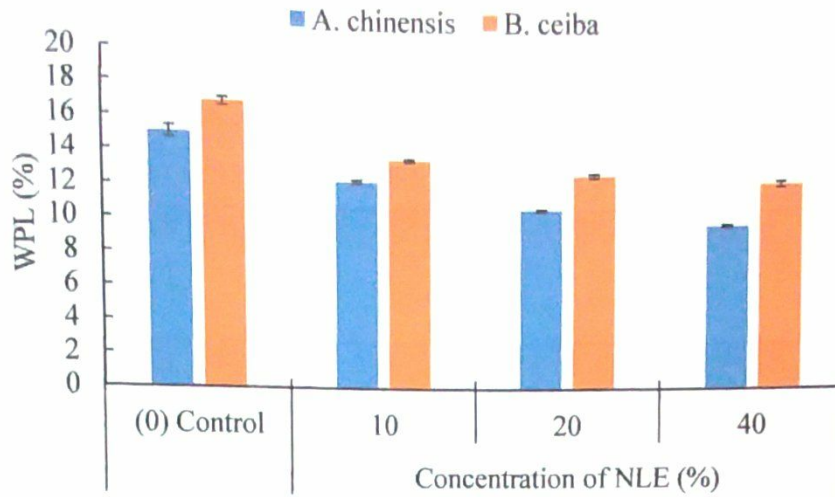
Figure 8: Mass loss (ML%) of Neem leaf extract (NLE) of two different wood species
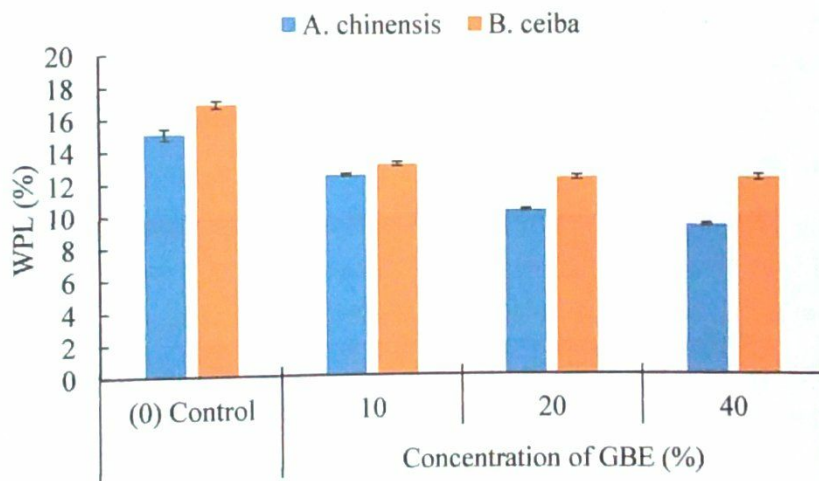

Figure 9: Mass loss (ML%) of Goran bark extract (GBE) for two different wood species

4.2 Discussion

The average weight percent gain (%) of neem leaf extract (NLE) treated wood blocks of two different species are *A. chinensis* 2.58, 4.55, 6.65 and *B. Ceiba* 1.94, 4.42, 6.44 for 10%, 20% and 40% solutions. The meam mass loss (%) of NLE treated decayed wood blocks of two different species after 12 weeks are *A. chinensis* 15.09, 12.33, 10.75, 9.77 and *B. ceiba* 16.93, 13.65, 12.80, 12.27 for control (0%), 10%, 20% and 40% solutions.

The average weight percent gain (%) of goran bark extract (GBE) treated wood blocks of two different species are *A. chinensis* 2.55, 4.60, 5.67 and *B. Ceiba* 2.78, 4.72, 5.79 for 10%, 20% and 40% solutions. The meam mass loss (%) of GBE treated decayed wood blocks of two different species after 12 weeks are *A. chinensis* 15.09, 12.52, 1.29, 9.29 and *B. ceiba* 16.93, 13.20, 12.32, 12.28 for control (0%), 10%, 20% and 40% solutions.

In this study, it has been found that higher the weight percent gain lower the mass loss of treated wood blocks. Ashaduzzaman *et al*. (2013) found the similar result by using cashew nut shell liquid (CNSL). It is evident that limited preservatives treatment at any WPG tested against the selected white rot decay fungi failed to meet the requirements for establishing a threshold (which corresponds to the point at which no weight loss occurred), a ML of less than 3% employed in the EN113 protocol (Eaton and Hale, 1993). Many studies have reviled that alcoholic extraction shows better results against wood decay fungi than hot water extraction. The marked activity of the alcoholic extracts may be attributed due to the presence of a large quantity of phenolic, terpenoid and trace amount of flavonoid in them (*Dhyani et al*., 2005). Hot water extraction may isolate a large quantity of carbohydrate which may acted as a nutrient to the fungi (Dhyani and Tripathi, 2005). Dhyani and Tripathi, (2006) found that 4.5% alcoholic extracts record a mass loss of about 18-19% when subjected against white rot (*Trametes versicolor*). In this study, it has recorded 9.29-9.77% mass loss for 40% hot water extraction as compared to the size of the tested wood samples. From this result it can be assumed that NLE and GBE can be a potential source of protecting wood from fungal deterioration.

CHAPTER FIVE

Conclusion

5. Conclusion

Wood is important as construction material and important of tree to protect environment is unique. To continue these two objectives, it is essential to be alternative to the preservation of wood. On the other hand, there is an increasing environmental concerns related to the use of synthetic wood preservatives. So, evaluation of new eco- friendly, low cost and more effective natural wood preservatives is essential. Recently, herbal extracts that are not harmful to the environment, have been showed to be effective natural preservatives.

There is a huge source of natural preservatives in plant community. Among them *A. indica* leaf extract and *C. decandra* bark extract are the one. About 35% of the goran bark is water soluble tannin. This NLE and GBE can be extracted by simple hot water extraction method. According to the results of this study, NLE and GBE mixed with aqueous solution can be good alternative to traditional synthetic preservatives. In this study, highest weight percent gain (%) was obtained from 40% NLE and GBE treated wood blocks and lowest mass loss (%) was found from 40% NEL and GBE treated wood blocks. Higher the weight percent (%), lower the mass loss (%). This study also reviled that, these extractives may be used as bio-preservative for wood used in indoor purposes. However, further studies on field stake test is needed to recommend these two extracts as good bio-preservatives for wood preservation.

Reference

Ashaduzzaman, M., Hale, M. D., Tverezovskiy, V. and Ormondroyd, G. A. (2013). *Effect of bio-resin from cashew nut shell liquid (CNSL) on decay resistance properties of wood.* The International Research Group in Wood Protection. Document no. IRG/WP 13-40649 Stockholm.

Bhatnagar, D. and S. P. McCormick. 1988. *The inhibitory effect of neem (Azadirachta indica A. Juss) leaf extracts on Aflatoxin synthesis in Aspergillus parasiticus.* J. American oil Chemist society, 65: 1166-1168.

Chavan, S.R. and S. T. Nikam (1988). *Investigation of alkanes from Neem leaves and their mosquito larvicidal activity.* Pesticides, 22 (7): 32-33.

Evans, P. (2003*) Emerging technologies in wood protection.* For Pord J 53: 14-22.

European standard EN 113 (1996) *Wood preservatives – Method of test for determining the protective effectiveness against wood destroying basidiomycetes- Determination of the toxic values.* European Committee for Standardization (CEN), Brussels, Belgium.

FAO, 1986. *Wood Preservation Manual.* FAO Forestry Paper 76. FAO, Rome, 1- 135pp.

Fisher, C. W. 1968. *Effluent Handling for Wood Preserving Plants.* Environment, Health and Safety Section, Research Department, Koppers Co., Inc, Pittsburg, pa; In Mimeo.

Freitag, M., Morrell, J. J., and Bruce, A. (1991) *Biological protection of wood: status and prospects.* Biodecterior Abstr 5:1-13.

Geissman, T. A. (1963) *Flavonoid compounds, tannin, lignin and related compounds.* In: Florkin, M. and Stotz, E.H. (eds) Pyrrole pigment, isoprenoid compounds and phenolic plant constituents, vol 9. Elsevier, New York, p265.

González-Laredo, R.F., M. Rosales-Castro, N.E. Rocha-Guzmán, J.A. Gallegos-Infante, M.R. Moreno-Jiménez and J.J. Karchesy. (2015). *Wood preservation using natural products.* Madera y Bosques 21(Núm. esp): 63-76.

Gupta, P and I.Dev, 1999. *Studies on the fungal toxicity of Sal (Shorea robusta) heartwood extractives.* J. Timb. Dev.Assoc. (Inadi0. 45 (1-2): 939.

Islam, M. M. 2006. *Preservative treatment of Mango (Mangifera indica) and Raintree (Albizia saman) wood with neem (Azaderacta indica) extracts as eco-friendly wood preservative and evaluation of its efficacy.* M. Sc. Thesis. Forestry and Wood technology Discipline, Khulna University, Khulna, Bangladesh. 50pp.

Kurucheve, V., G.J. Ezhilalnad, J. Jayraj (1997). *Screening of higher plants for fungi toxicity against Rhizoctonia solani in vitro.* Indian phytopathology, 50(2): 235-241.

Lahiry, A. K. 2001. *Applied Wood Preservation.* Magnum opus, Dhaka, 342- 344pp.

Laks, P. E. (1989) *Wood preservation- looking ahead.* Construct Specif 42:60-69.

Lin, C. Y., Wu, C.L. and Chang, S.T. (2007) *Evaluation the potency of cinnamaldehyde as a natural wood preservative.* The International Research Group on Wood Protection. Document no. IRG/WP 07-30444 Stockholm.

Onuorah, E. O. 2000. *Short communication: The wood preservative potential of heartwood Extracts of Milicia excelsa and Erythrophleum suaveolens.* Bio- resources Technology, 75: 171-173.

Purushotham, A. 1970. *Protection of pulpwood (timber, bamboo) from deterioration due to biological agencies (fungi and insects, etc.) during transit and storage.* J. Timb. Dev. Assoc. India, 16 (2): 51-53.

Schultz, T. P. and Nicholas, D.D. (2007). *Totally organic wood preservative for exterior residential applications.* In: Barnes, H.M. (ed) Wood protection 2006. Forest Products Society, Madison, pp 289- 294.

Shrivestava, M. B. 1997. *Wood Technology.* Vikas PublicationHouse Pvt. Ltd. New Delhi.

Soni, P.L. 1975. *The chemistry of extractives of Dalbergia sissoo, Part- I, The occurrence of 3, 5 Dihydroxy-trans-stilbene in the heartwood.* J. Ind. Acad. Wood Science. 6 (2): 57-58.

Swathi Dhyani, Sadhana Tripathi and Indradev, 2004. *Preliminary screening of neem (Azadirachta indica) Leaf extractives against poria monticola- A wood destroying fungus*. J. Ind. Acad. Wood Sci., (N.S.) 1 (1 & 2): 103-112.

Tewari, D.N. 1992. *Monograph on Neem*. International book distributer, Dehradun.

Thompson, W. S. 1971. *Contribution of the wood preserving industry to water pollution. In: Proceeding of conference on pollution abetment and control in the wood industry*. W. S. Thompson (ed.) Mississippi Forest Products laboratory. Mississippi state college: 50-75.

Yang, D. Q. (2009) *Potential utilization of plant and fungal extracts for wood protection*. For Prod J 59:97- 103.

Laboratory Test Result

Table 1: Mean weight percent gain (WPG%) of Neem leaf extract (NLE) after the impregnation for two different wood species

Mean WPG (%) of NLE		
Concentrations (%)	**Species**	
	Anthrocephalaus chinensis	*Bombax ceiba*
Control	0	0
10	2.58 (±0.17)	1.94 (±0.07)
20	4.55 (±0.19)	4.42 (±0.32)
40	6.56 (±0.17)	6.44 (±0.23)

Table 2: Mean weight percent gain (WPG%) of Goran bark extract (GBE) after the impregnation for two different wood species

Mean WPG% of GBE		
Concentrations (%)	**Species**	
	Anthrocephalaus chinensis	*Bombax ceiba*
Control	0	0
10	2.55 (±0.02)	2.78 (±0.10)
20	4.60 (±0.22)	4.72 (±0.44)
40	5.67 (±0.25)	5.79 (±0.35)

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Table 3: Mass loss (ML%) of Neem leaf extract (NLE) treated decayed wood species after 12 weeks

Mean ML (%) of NLE		
Concentrations (%)	Species	
	Anthrocephalaus chinensis	*Bombax ceiba*
Control	15.09 (±0.83)	16.93 (±0.56)
10	12.33 (±0.24)	13.65 (±0.20)
20	10.75 (±0.18)	12.80 (±0.28)
40	9.77 (±0.16)	12.27 (±0.43)

Table 4: Mass loss (ML%) of Goran bark extract (GBE) treated decayed wood species after 12 weeks

Mean ML% of GBE		
Concentrations (%)	Species	
	Anthrocephalaus chinensis	*Bombax ceiba*
Control	15.09 (±0.83)	16.93 (±0.56)
10	12.52 (±0.18)	13.20 (±0.34)
20	10.29 (±0.20)	12.32 (±0.37)
40	9.29 (±0.21)	12.28 (±0.43)

Statistical Analysis Result

ANOVA for NLE for Weight percentage gain (%)

Dependent Variable: WPG

Source	DF	Sum of Squares	F Value	Pr > F
Treatment	7	284.79166918	1073.04	0.0001
Error	40	1.51660398		
Corrected Total	47	286.30827316		

LSD for NLE for WPG%
P= 0.05, df= 40 MSE= 0.037915
Critical Value of T= 2.02
Least Significant Difference= 0.2272

Means with the same letter are not significantly different.

Treatment Grouping	Mean	N	Treatment
A	6.5591	6	T4
A	6.4437	6	T8
B	4.5468	6	T3
B	4.4246	6	T7
C	2.5762	6	T2
D	1.9440	6	T6
E	0.0000	6	T1
E	0.0000	6	T5

ANOVA for GBE for Weight percentage gain (%)

Dependent Variable: WPG

Source	DF	Sum of Squares	F Value	Pr > F
Treatment	7	228.68606796	499.70	0.0001
Error	40	2.61514825		
Corrected Total	47	231.30121621		

LSD for GBE for WPG%
P= 0.05, df= 40 MSE= 0.065379
Critical Value of T= 2.02
Least Significant Difference= 0.2984

Means with the same letter are not significantly different.

Treatment Grouping	Mean	N	TRTMENT
A	5.7860	6	T8
A	5.6704	6	T4
B	4.7190	6	T7
B	4.6041	6	T3
C	2.7766	6	T6
C	2.5533	6	T2
D	0.0000	6	T1
D	0.0000	6	T5

ANOVA for NLE for Weight loss (%)

Dependent Variable: WL

Source	DF	Sum of Squares	F Value	Pr > F
Treatment	7	220.58314795	148.56	0.0001
Error	40	8.48441517		
Corrected Total	47	229.06756312		

LSD for NLE for WL%
P= 0.05, df= 40 MSE= 0.21211
Critical Value of T= 2.02
Least Significant Difference= 0.5374

Means with the same letter are not significantly different.

Treatment Grouping	Mean	N	TRTMENT
A	16.9297	6	T5
B	15.0855	6	T1
C	13.6525	6	T6
D	12.7976	6	T7
D	12.3308	6	T2
D	12.2698	6	T8
E	10.7455	6	T3
F	9.7652	6	T4

ANOVA for GBE for Weight loss (%)

Dependent Variable: WL

Source	DF	Sum of Squares	F Value	Pr > F
Treatment	7	249.39932399	153.55	0.0001
Error	40	9.28107181		
Corrected Total	47	258.68039580		

LSD for GBE for WL%
p= 0.05 df= 40 MSE= 0.232027
Critical Value of T= 2.02
Least Significant Difference= 0.5621

Means with the same letter are not significantly different.

Treatment Grouping	Mean	N	TRTMENT
A	16.9297	6	T5
B	15.0855	6	T1
C	13.1957	6	T6
D	12.5241	6	T2
D	12.3243	6	T7
D	12.2808	6	T8
E	10.2907	6	T3
F	9.2941	6	T4

Abbreviations of treatments (T1, T2,…..etc.)

Treatment	Concentration	Species
T1	(0) Control	A. chinensis
T2	10 NLE	A. chinensis
T3	20 NLE	A. chinensis
T4	40 NLE	A. chinensis
T5	(0)Control	B. ceiba
T6	10 NLE	B. ceiba
T7	20 NLE	B. ceiba
T8	40 NLE	B. ceiba
Treatment	Concentration	Species
T1	(0) Control	A. chinensis
T2	10 GBE	A. chinensis
T3	20 GBE	A. chinensis
T4	40 GBE	A. chinensis
T5	(0)Control	B. ceiba
T6	10 GBE	B. ceiba
T7	20 GBE	B. ceiba
T8	40 GBE	B. ceiba