Antifungal Activities of the Inner and Outer Barks of the *Swietenia mahagoni* (L) Jacq (Mahogani)

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Abstract

Swietenia mahagoni (L) Jacq or Mahogany bark has been reported to show some antifungal activities. The present work therefore focused on investigation of its possible activity against certain fungi responsible for some human infections. Thus 70% EtOH extract of inner and outer barks of mahogany were fractionated into EtOAc, n-BuOH and aqueous fractions and their activities on two dandruff and three athlete's foot fungal strains screened by agar well diffusion method. The EtOAc fraction for inner bark showed pronounced activities, viz. 25-20 mm on the dandruff fungi and 18-15 mm against the athlete's foot fungi and is selected for further study. The bark of S. mahogoni was collected from East Dagon Township, Yangon Region. The plant sample was identified at Department of Botany, University of Yangon. Preliminary phytochemical tests revealed the presence of alkaloids, α-amino acids, anthraquinones, carbohydrates, flavonoids, glycosides, phenolic compounds, reducing sugars, saponins, starch and tannins in these samples. In addition, some nutritional values such as moisture (11.98 %), ash (7.01 %), protein (2.50%), fiber (29.34 %), fat (0.50 %) and carbohydrate (48.67 %) were determined by AOAC methods. The relative abundance of some elements by ED-XRF showed Ca (91.13 %) and K (4.851 %) as major elements in bark, followed by S (0.758 %), Si (1.361 %), Fe (1.141 %), Cu (0.129 %), and Sr (0.630 %). The bark, especially the inner bark, of mahogany may therefore be useful as a potential source of antifungal active principles for the future antifungal drug.

Keywords: Mahogany bark, antifungal activity, agar well diffusion method, dandruff, athlete's foot disease

Introduction

The earliest recorded use of *Swietenia mahagoni* was in 1514. Other records refered to the use of mahogani between 1521 and 1540, when Spanish explorers employed the wood for making canoes and for repairing ship in the West Indies. Firstly, the use of Mahogani in Spain and England was for ship building, and during the 18th century it was the chief wood employed in Europe for that purpose. Mahogani is a deciduous, erect tree growing to a height of 10 meters, with a heavy, dark-green, and dense crown. Bark is dark gray and ridged. The bark in younger specimens is smooth and grayish, becoming darker and furrowed with age (Figure 1). In the U.S. mahoganies are semi-deciduous, losing all or most of their leaves over winter or shedding at the flush of new growth in spring (Philippine Medicinal Plants, 2016). The bark contains tannin; limonoids and alkaloids, terpenoids, antraquinones, cardiac glycosides, saponins, phenols, flavonoids and long chain unsaturated acid (Mayur *et al*., 2011). The study of traditional medicinal plants and their therapeutics play a very important role in health care system of Myanmar because most of its population is in the rural area and they have been using traditional medicine for centuries. The plant kingdom constitutes an invaluable source of chemical products which may be important due to their biological properties and their potential use in medicine.

Bark of *Swietenia mahagoni* possesses astringent, antipyretic, abortifacient, depurative, anticoagulant, antioxidant, antimicrobial, antidiabetic, antiprotozoal, anthelmintic, cytotoxic, gastroprotective, and hepatoprotective properties (Philippine Medicinal Plants, 2016).

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(a) The whole plant (b) bark

Figure (1) Photographs of *Swietenia mahagoni* (Mahogani)

Materials and Methods

Sample Collection

The medicinal plant namely *Swietenia mahagoni* L. (Mahogani) was chosen in this study. The bark of Mahogani was collected from East Dagon Township, Yangon Region, Myanmar. The collected bark samples were cleaned by washing with water and then were dried in the shade. The dried sample was ground into coarse powder. The dried powder sample was stored in airtight container to prevent moisture changes and other contaminations

Preliminary phytochemical tests

A few grams of powder sample were subjected to the tests of alkaloids, α-amino acids, anthraquinones, carbohydrates, flavonoids, glycosides, phenolic compounds, reducing sugars, saponins, starch and tannins as the preliminary phytochemical test according to test tube methods. (Tin Wa, 1972 and Trease and Evans, 1980).

Determination of Nutritional Values

In the present study, some nutritional values such as moisture, ash, protein, fiber, fat, carbohydrate and energy values of *Swietenia mahagoni* (Mahogani) bark were determined by AOAC methods (A.O.A.C, 2000).

Semi-Quantitative Elemental Analysis of Plant Samples by Energy Dispersive X-Ray Fluorescence (EDXRF)

Elemental analysis of *Swietenia mahagoni* (Mahogani) bark was done by EDX-8000.

Determination of Extractive Values

Dried powdered sample of the bark of *S.mahagoni* bark (50 g each) was extracted separately with five different solvents (250 mL each), namely petroleum ether (PE) (60-80°C), 70%, ethanol, ethyl acetate, n-butanol and water. In each extraction the collected samples were macerated for 1 week in the corresponding solvent, shaking from time to time, filtered and evaporated to dryness.

Preparation of Various Crude Extract

The dried powder sample (300 g) was macerated with 70% ethanol (3 liter) for one week. The ethanolic extract was filtered and rotatory to dryness. The aqueous portion was divided into two portions. The first portion was evaporated carefully until to obtain EtOH extract from the powder. A few amount of the second portion was added to separating funnel

to extract with EtOAc. The upper layer was evaporated to dryness and the EtOAc extract was obtained. The lower layer transfers to separating funnel.

Screening of Antifungal Activities of *Swietenia mahagoni* (Mahogani)

For the examination of *in vitro* antifungal activity, 70%EtOH, EtOAc, n-BuOH and H_2O extracts of the inner and outer bark of *S. mahagoni* (Mahogani) were investigated by agar well diffusion method.

Nutrient agar was prepared according to the method described by Cruickshank, 1975. Nutrient agar was boiled and 20□25 mL of the medium were poured into the test tube and plugged with cotton wool and autoclaved at 121 ºC for 15 minutes in an autoclave. After this, the tubes were cooled down to 30°-35 ºC and poured into sterilized petri dished and 0.02 mL of spore suspension was also added into the dishes. The agar was allowed to set for 2 hours after which 10 mm agar well was made with the help of sterilized cork burner. After that, about 0.5 mL of sample was introduced into the agar–well and incubated at 37 ºC for 24 hours. The inhibition zone (clear zone) which appeared around the agar-well indicated the presence of antifungal activity.

Results and Discussion

Phytoconstituents present in bark of *Swietenia mahagoni* (L.) Jacq. (Mahogani)

According to the phytochemical result, the bark of *S. mahagoni* (Mahogani) was found to contain alkaloids, α-amino acids, anthraquinone, carbohydrates, flavonoids, glycosides, phenolic compounds, reducing sugars, saponins, starch and tannins. (Table 1)

Table (1) Results of Preliminary Phytochemical Tests on Bark of *S. mahagoni* (Mahogani)

No.	Test	Extracts	Test reagents	Observation	Result
1.	Alkaloids	1% HCl	Wagner's reagent	Reddish brown ppt	+
			Mayer's reagent	Yellow color	+
			Dragendroff's reagent	Brown ppt	+
			Sodium picrate	Orange ppt	+
2.	Anthraquinone	$CHCl_3$	10% NH_3	Pink color	+
3.	α-Amino acids	H_2O	Ninhydrin reagent	Purple spot	+
4.	Carbohydrates	H_2O	10% α-Naphthol & Conc:H_2SO_4	Red ring	+
5.	Flavonoids	EtOH	HCl, Magnesium turning	Reddish brown colour	+
6.	Glycosides	H_2O	10% Lead acetate solution	White ppt	+
7.	Phenolic compounds	H_2O	1% $FeCl_3$ 1% $K_3Fe(CN)_6$	Deep blue colouration	+
8.	Reducing sugars	H_2SO_4 (dil)	Benedict's solution	Yellow ppt	+
9.	Saponins	H_2O	Distilled water	Frothing	+
10.	Starch	H_2O	1% Iodine	Brown colourtion	+
11.	Tannins	EtOH	5% $FeCl_3$	Dark green colour	+

Nutritional Values of the Bark of *S. mahagoni* (Mahogani)

Some nutritional values such as moisture, ash, fibre, fat, protein, carbohydrates and energy values of the bark of *S. mahagoni* (Mahogani) were summarized in table (2).

Table (2) Nutritional Values of *S. mahagoni* (Mahogani) Bark

No.	Nutrients	Content (%)
1	Moisture	11.98
2	Ash	7.01
3	Protein	2.50
4	Fiber	29.34
5	Fat	0.50
6	Carbohydrate	48.67
	Energy Value (kcal/100g)	217

Semi-Quantitative Elemental Analysis of the Bark of Mahogani by Energy Dispersive X- Ray Fluorescence

The EDXRF spectrum of the sample was shown in Figure 2. It can be seen that mineral element such as Ca (91. 13%) is predominant in the sample followed by smaller amounts of K (4.851 %), Si (1.361 %), Fe (1.414 %), S (0.758 %), Sr (0.630 %) and Cu (0.129 %) as shown in (Table 3).

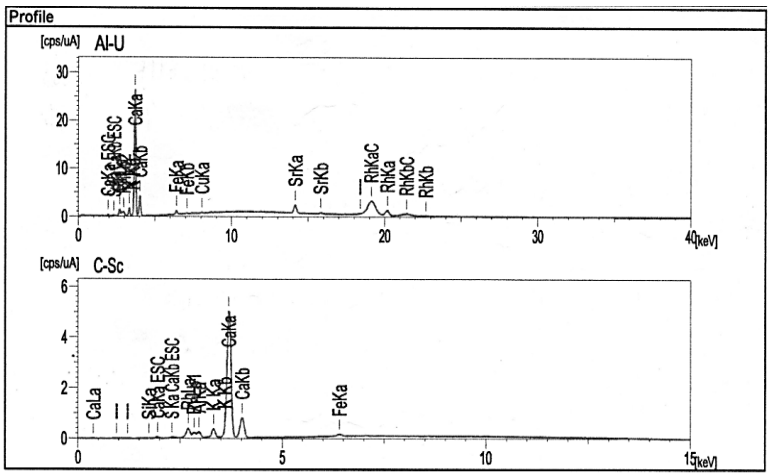

Figure (2) EDXRF spectrum of the bark of *S. mahagoni* (Mahogani)

Table (3) Relative Abundance of Some Elements in the Bark of *S. mahagoni* (Mahogani) by EDXRF Method

No.	Elements	Relative Abundance (%)
1	Calcium	91.130
2	Potassium	4.851
3	Silicon	1.361
4	Iron	1.141
5	Sulphur	0.758
6	Strontium	0.630
7	Copper	0.129

Extractive Values of the Bark of *S. mahagoni* (Mahogani)

Five solvents were used to find the extractive values of the bark. Ethanol, methanol, ethylacetate, PE and water were used as solvents.

The percent crude extracts (w/w) prepared for the selected medicinal plant are described in table (4).

Table (4) Yield Percent of Crude Extracts from the Bark of *S. mahagoni* (Mahogani)

Plant Sample	Solvent	Weight (g)	Yield (%)
Mahogani (Bark)	PE	0.2	0.9
	95% EtOH	2.3	4.6
	EtOAc	1.7	3.4
	MeOH	3.2	6.4
	H_2O	1.4	2.8

Screening of Antifungal Activity of Bark of *S. mahagoni* (Mahogani) by Agar Well Diffusion Method

The different crude extract namely 70% EtOH, EtOAc, n-BuOH and H_2O extract were screening of antifungal activities against fungi causing dandruff and athlete's foot infision and also against *Candida albicans*. Potato-dextrose agar well method was used in the screening. Two species were isolated from dandruff sample and three species were isolated from the socks for athlete's food fungi. Antifungal activities of various crude extracts from the inner bark of *Swietenia mahagoni* (Mahogani) were carried out by the five isolated species and *Candida* albicans. The inhibition zone diameter under the view of microsope is shown in the figures (3, 4 & 5). Among them the EtOAc fraction was the most active one.

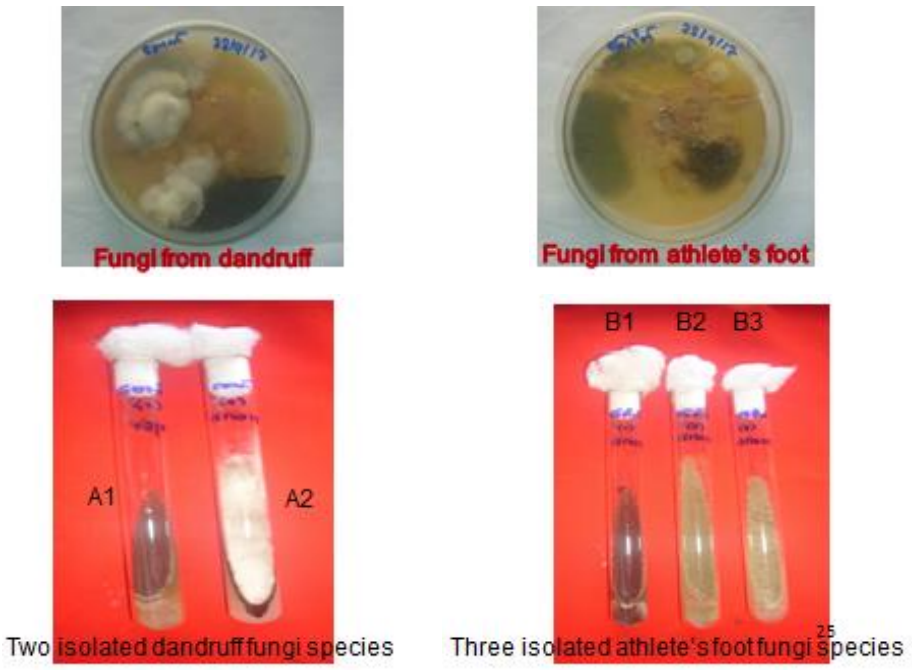

Figure (3) Culture of isolated fungus species

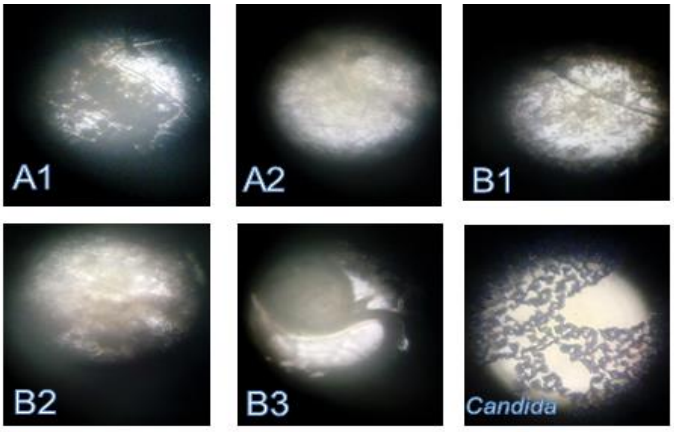

Figure (4) Test organisms

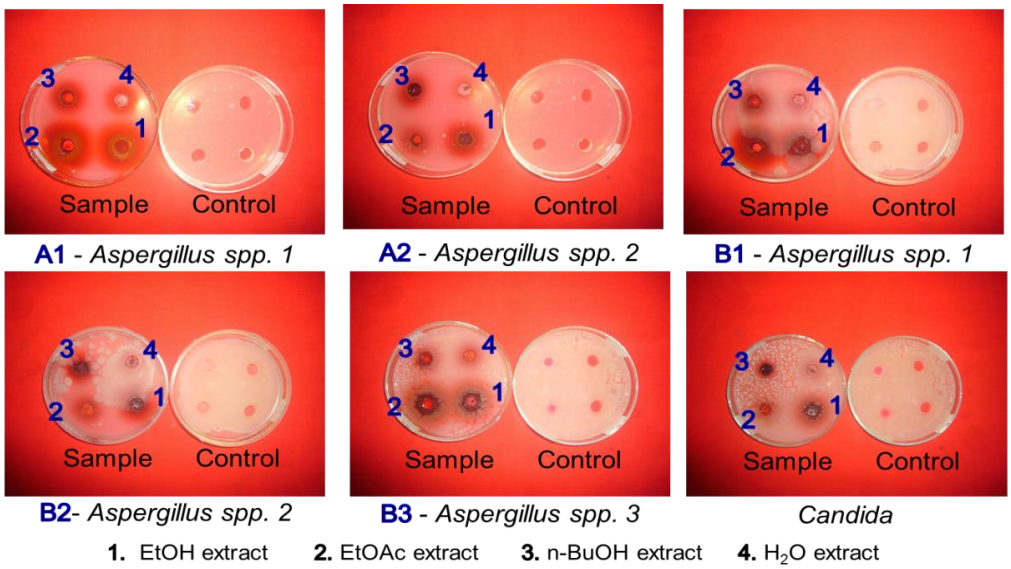

Figure (5) Antifungal activity of various extracts of *Swietenia mahagoni* (Mahogani) inner bark on six microorganisms

Table (5) Antifungal Activity of Various Crude Extracts from Inner Bark of *Swietenia mahagoni* (Mahogani) by Potato-dextrose Agar Well Diffusion Method

Test Organisms used	**Diameter of Inhibition Zone (mm)**			
	EtOH	**EtOAc**	**n-BuOH**	**H_2O**
Aspergillus:spp(A1)	20	25	20	15
Aspergillus:spp (A2)	-	20	15	20
Aspergillus:spp (B1)	15	15	12	13
Aspergillus:spp (B2)	16	18	17	14
Aspergillus:spp (B3)	15	18	18	15
Candida albicans	18	16	15	15

Agar Well – 10 mm
10 mm ~ 14 mm - lower activity
15 mm ~ 19 mm - higher activity
20 mm ~ above - highest activity

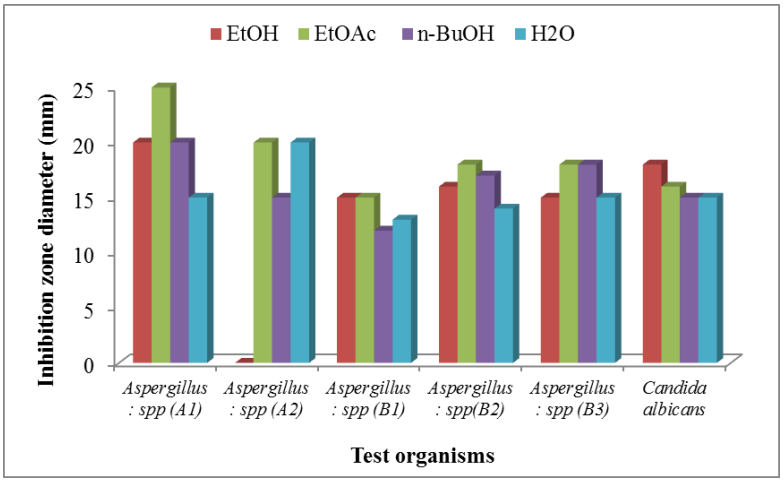

Figure (6) Histogram of inhibition zone diameters of different extracts of *Swietenia mahagoni* (Mahogani) inner bark against six microorganisms

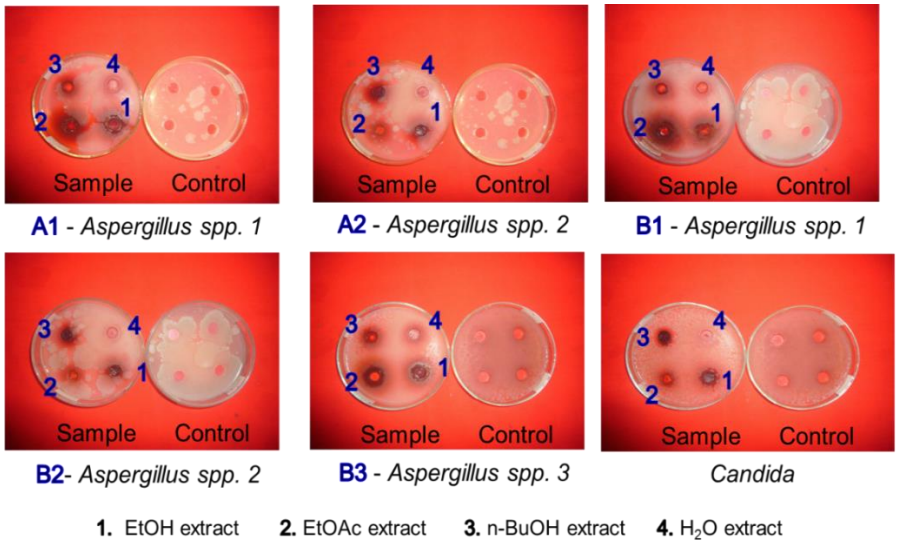

Figure (7) Antifungal activity of various extracts of *Swietenia mahagoni* (Mahogani) inner bark on six microorganisms

Table (6) Antifungal Activity of Various Crude Extracts from Outer Bark of *Swietenia mahagoni* (Mahogani) by Potato-dextrose Agar Well Diffusion Method

Test Organisms used	**Diameter of Inhibition Zone (mm)**			
	EtOH	**EtOAc**	**n-BuOH**	**H_2O**
Aspergillus:spp (A1)	15	13	15	14
Aspergillus:spp (A2)	15	17	18	13
Aspergillus:spp (B1)	20	15	12	13
Aspergillus:spp (B2)	15	16	13	14
Aspergillus:spp (B3)	15	15	15	13
Candida albicans	18	15	17	13

Agar Well – 10 mm
10 mm ~ 14 mm - lower activity
15 mm ~ 19 mm - higher activity
20 mm ~ above - highest activity

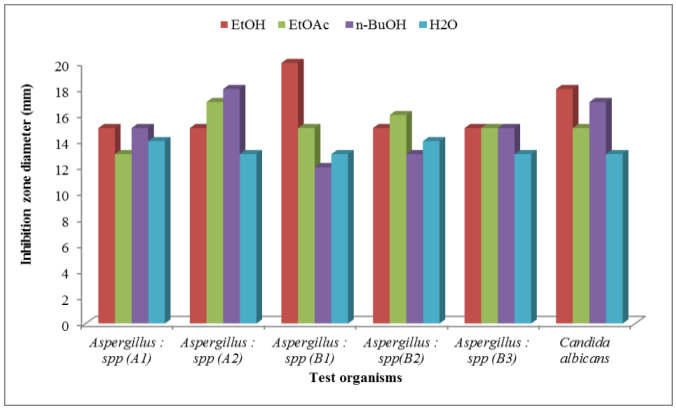

Figure (8) Histogram of inhibition zone diameters of different extracts of *Swietenia mahagoni* (Mahogani) outer bark against six microorganisms

Conclusion

The 70% EtOH extracts of the inner and outer barks of mahogany, as well as the EtOAc, n-BuOH and aqueous fractions of these extracts showed activity against the isolated fungal strains from dandruff and athlete's foot infections. Among them the EtOAc fraction was the most active one. The bark, especially the inner bark, of mahogany may therefore be useful as a potential source of antifungal active principles for the future antifungunal drugs.

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