

Phytochemical and Antimicrobial Screening of the Stem Bark Extracts of *Pterocarpus erinaceus* (Poir)

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ABSTRACT: The stem bark of *Pterocarpus erinaceus* was successively extracted with n-hexane, ethyl acetate and methanol using a soxhlet extractor. Antimicrobial screening was carried out on some pathogens and the result showed a broad spectrum of activity against *Bacillus subtilis*, *Pseudomonas aeruginosa*, *Staphylococcus aureus*, *Escherichia coli* and *Candida albican*. Phytochemical screening revealed the presence of tannins, flavonoids, phenols and saponins.

Keywords: *Pterocarpus erinaceus*, antimicrobial activity, phytochemical screening

INTRODUCTION

Plants have formed the basis of traditional medicine system which has been used for thousand of years. Traditional medicine refers to health practices, approaches, knowledge and beliefs incorporating plant, animal and mineral-based medicines, spiritual therapies, manual techniques and exercises, applied singularly or in combination to treat or to diagnose and prevent illnesses or maintain well being (World Health Organization, 2003). In developing countries where orthodox medicines are quite expensive, traditional medicine is widely practiced thus, screening for antimicrobially active compounds from ethnomedicinal plants is vital so as to ascertain genuine active plants and active compounds. Ultimately, these phytochemicals will find their way into the arsenal of antimicrobial drugs prescribed by physicians (Cowan, 1999).

Pterocarpus erinaceus is a deciduous legume tree of African savannahs and dry forests famous for producing one of the finest woods in its native region. It also produces leafy fodder high in protein, which makes an excellent animal feed crucial for the survival of livestock during dry season (Hutchinson et. al. 1958). It has several common names including vene in French. It is called palissandre in Senegal, Kino in Gambia, Bani or Banuhi in Fulfude (Burkina Faso), Gwani, Ngueni in Bambara, Tolo in Djerma, Bu Natombu in Gourmantche, Noega, Noeka,

Pempelaga in More, Ban in Serer, Ven or Yirik In Wolof (Sandrine P., 2006) Madubiya in Northern Nigeria and Osun dudu in Southwest Nigeria.

The foliage of *Pterocarpus erinaceus* is a nutritious fodder for farm animals and Mali has an active market for this which is in high demand by sheep farmers for fodder (Hutchinson et.al., 1958).

Medicinal uses of *Pterocarpus* plant include the use of the leaves as a febrifuge, the bark for tooth and mouth troubles, and bark resin as astringent for severe diarrhoea and dysentery. The grated root is mixed with tobacco and smoked in a pipe as a cough remedy. It has also been found useful in the treatment of fever (Hutchinson et. al. 1958; Sandrine, 2006).

The leaves appear to have some insect repellent property. Tenda tribe of Senegal/Guinea use them to protect their graineries against termites. The flowers attract bees. The tree may have value for producing honey. The value of the fruit as food is not reported but the fruit of the closely related *P. santalinoids* is said to be edible if cooked and intoxicating if not. Similarly the seeds are reported to induce intoxication (Hutchinson et. al., 1958; Sandrine, 2006).

This paper reports on the preliminary screening of the stem bark of *Pterocarpus erinaceus* against some American Type Culture Collection (ATCC) and clinically isolated pathogens.

MATERIALS AND METHODS

Plant Collection, Identification and Pre-Treatment:

The stem bark of *Pterocarpus erinaceus* was collected with the aid of a sterilized axe at Bissallam in Dange Shuni local government area in Sokoto, Northern Nigeria. Identification was done at the National Institute of Pharmaceutical Research and Development Idu Abuja.

Extraction: Crude extraction was carried out using a soxhlet extractor. About 500 g of the powdered plant material were taken in a thimble and placed into the extraction chamber of the soxhlet apparatus and three solvents were used successively in order of increasing polarity for exhaustive extraction of the powdered stem bark namely: hexane (2.5L), ethyl acetate (2.5L) and 95 % methanol (2.5L).

A fresh portion of the powdered stem bark (about 200 g) was used to carry out bulk extraction using 95 % methanol.

Phytochemical and antimicrobial screening of *Pterocarpus erinaceus*: The powdered stem bark of *Pterocarpus erinaceus* was screened for secondary metabolites such as tannins, anthraquinones, saponins, flavonoids and phenols using standard methods (Trease and Evans 1983; Olurinola, 1996).

Antimicrobial assay was carried out on the

hexane, ethyl acetate and methanol fractions and also the crude methanol using the cup-plate method on gram positive and gram negative pathogens viz: *Candida albicans*, *Staphylococcus aureus*, *Escherichia coli*, *Bacillus subtilis* and *Pseudomonas aeruginosa* along with controls- Organism Viability Control (OVC), Extracts Sterility Control (ESC) and Medium Sterility Control (MSC) using standard methods (Trease and Evans 1983; Bauer *et. al.*, 1996; Olurinola, 1996).

RESULTS

500g of dried pulverized stem bark of *Pterocarpus erinaceus* was successively extracted with n-hexane, ethyl acetate and methanol to yield three fractions with the following masses:

a yellowish green solid 1.45 g (0.29 %), a brown solid 1.56 g (0.31 %) and a reddish brown solid 39.65 g (7.93 %) respectively.

200 g of dried pulverized stem bark of *Pterocarpus erinaceus* extracted with 95 % methanol yielded 38.40 g (19.20 %) methanol crude extracts.

The phytochemical screening results are as shown in Table 1. Tables 2 and 3 show the antimicrobial assay results on the fractions and crude extracts.

Table 1: Phytochemical screening results of *Pterocarpus erinaceus*

COMPOUNDS	PRESENCE / ABSENCE
Tannins (Condensed and Hydrolysable)	+
Anthraquinones	-
Saponins	+
Flavonoids	+
Phenols	+

key: (+) = Presence; (-) = Absence

Table 2: Sensitivity test results of the extracts

Extracts	Organisms / Zones of Inhibition (mm)							
	Ca	Sa	Ec	Bs	Ps	OVC	ESC	MSC
Crude Methanol	-	-	20	20	20	N	N	N
Hexane Fraction	30	30	-	20	-	N	N	N
Ethyl acetate Fraction	-	-	-	20	10	N	N	N
Methanol Fraction	30	30	30	20	10	N	N	N

Key: { (+) = Active; (-) = Inactive; N = Normal}; OVC = Organism Viability Control; ESC = Extract Solubility Control; MSC = Medium Sterility Control; Ca = *Candida albicans* (ATCC 10231); Sa = *Staphylococcus aureus* (ATCC 13709); Ec = *Escherichia coli* (ATCC 9637); Bs = *Bacillus subtilis* (Clinical Isolate); Ps = *Pseudomonas aeruginosa* (Clinical 7853)

Table 3: Minimum Inhibition Concentration (MIC) values of the extracts

Extracts	Organisms / MIC (mg/ml)				
	Ca	Sa	Ec	Bs	Ps
Crude Methanol	-	-	1	1	1
Hexane Fraction	1	0.5	-	1	-
Ethyl acetate Fraction	-	-	-	1	1
Methanol Fraction	0.5	1	0.5	1	0.5

Ca = *Candida albicans* (ATCC 10231); Sa = *Staphylococcus aureus* (ATCC 13709); Ec = *Escherichia coli* (ATCC 9637); Bs = *Bacillus subtilis* (Clinical Isolate); Ps = *Pseudomonas aeruginosa* (Clinical 7853)

DISCUSSION

Exhaustive, successive extraction using n-hexane, ethyl acetate and methanol yielded three crude fractions, 1.4 g (0.29 %), 1.56 g (0.31 %) and 39.65 g (7.93 %) respectively. Phytochemical screening of the powdered stem bark showed that *Pterocarpus erinaceus* contains both condensed and hydrolysable tannins, saponins, flavonoids and phenols (Table 1).

Antimicrobial activity screening of the three extracts and the methanol crude extract showed a broad spectrum activity. From Table 2, it can be seen that the methanol fraction is the most active of the four extracts tested showing activity against all five pathogens. The n-hexane fraction and the crude methanol extract showed activity against three out of the five pathogens tested. While the n-hexane fraction was inactive against the *E. coli* and *P. aeruginosa*, the crude methanol extract was inactive against *C. albicans* and *S. aureus*. The ethyl acetate fraction was the least active of the four

extracts. It was active against two out of the five pathogens tested, namely: *E. coli* and *B. subtilis*.

Similarly, while the crude methanol extract was inactive against *C. albicans* and *S. aureus* at 1 mg / ml, the methanol and the n-hexane fractions were active against them.

From Table 2, it can be seen that the ethyl acetate exhibited the least sensitivity between 10 mm against *P. aeruginosa* and 20 mm against *B. subtilis*. The crude methanol extract showed sensitivity of 20 mm against all the pathogens it was active against while the n-hexane fraction exhibited sensitivity values of 20 mm against *B. subtilis* and 30 mm against *C. albicans* and *S. aureus*.

The methanol fraction appears to also be the most sensitive of the four extracts with a sensitivity of 30 mm against three out of the five pathogens (Table 2).

It should be noted that the n-hexane fraction was active against *S. aureus* at 0.5 mg / ml with a sensitivity of 20 mm and the methanol fraction was active at 0.5 mg / ml against *B.*

subtilis and *P. aeruginosa* with a sensitivity of 20 mm and 10 mm respectively.

It can therefore be said that of the four extracts tested, the methanol fraction was the only extract that showed activity against all the five pathogens at MIC values of 0.5 mg / ml and 1 mg / ml. Generally, MIC values for the extracts against the various pathogens were between 0.5 mg / ml and 1 mg / ml.

It can be seen from Table 3 that both the n-hexane fraction and the methanol fraction exhibited an MIC value of 0.5 mg / ml.

Secondary metabolites of various chemical types present in the plant species are known to possess antimicrobial activity. Flavonoids are found to be effective antimicrobial substances against a wide range of microorganisms (Tsuchiya *et al.*, 1996). Phenols and phenolic compounds are known to be toxic to microorganisms (Mason T.L. and Wasserman, 1987). The ability of tannins to inactivate microbial adhesins, enzymes and cell envelope transport proteins makes them antimicrobial active (Ya, *et al.*, 1998). Broad spectrum antibacterial activity of leaf extract of *Bolusanthus speciosus* is due to flavonoids (Bojase, 2002). *Landolphia owerrience* is known to possess glycosides, flavonoids, tannins and saponins which either individually or in combination exert antibacterial activity (Ebi and Ofoefule., 1997).

Comparing the antibacterial activity of leaf and stem bark extracts of *Pterocarpus santalinus* (*Fabaceae*) as investigated by (Manjunatha, 2006) and *Pterocarpus erinaceus* in this work, it can be seen that the two plants contains: phenols, saponins, flavonoids and tannins. The two plants also exhibited antibacterial activity against the tested pathogens but *P. erinaceus* exhibited higher sensitivities than *P. santalinus* (Table 2 and 3).

Conclusion: From the results obtained from this work, it can be concluded that the stem bark of *Pterocarpus erinaceus* contains tannins, flavonoids, Phenols and saponins. It has also been confirmed that the stem bark possesses a broad spectrum anti microbial activity against *Bacillus subtilis*, *Pseudomonas aeruginosa*, *Escherichia coli*.

Staphylococcus aureus and *Candida albicans* with varying degrees of MIC values and sensitivities.

The broad spectrum antibacterial activity exhibited by *Pterocarpus erinaceus* in this work may be attributed to the various active components present in it, which either due to their individual or synergistic action exhibit antibacterial activity.

From the foregoing, the findings of this work can serve as a scientific data base for the acclaimed medicinal uses of *Pterocarpus erinaceus*.

Further work will attempt to isolate, characterize and screen phytochemical components which may be responsible for the activity of *Pterocarpus erinaceus*.

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