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Short Communication

The phytochemical analysis and antimicrobial screening of extracts of *Olax subscorpioidea*

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The antimicrobial activities of the aqueous and ethanoic extracts of the stem of *Olax subscorpioidea* were evaluated. The ethanoic extract showed considerable activity on both the fungi and bacteria with zones of inhibition ranging from 7.2 mm to 21.5 mm, with minimum inhibitory concentrations ranging from 5 to 45 mg/ml, while the aqueous extract was only active against three of the test organisms used. Phytochemical test reveals the presence of alkaloids, steroids, and flavonoids together with other active ingredients in the ethanoic extract with the exception of saponins which is present in the aqueous extract alone.

Key words: Antimicrobial activity, bacteria, fungi, alkaloids, tannins, glycosides, steroids, flavonoids, saponins.

INTRODUCTION

Medicinal plants are used locally in the treatment of infections caused by fungi, bacteria, viruses and parasites. And over 60% of people in Nigeria rural areas depend on the traditional medicine for the treatment of their ailments (Ghani et al., 1989). Different plants have been used as a source of inspiration in the development of novel drugs (Robbers et al., 1996). Plant derived medicines are widely used because they are relatively safer than the synthetic alternatives, they are easily available and cheaper (Iwu et al., 1999). Many plant species have been evaluated for their antimicrobial activity in the past 20 years (Castello et al., 2000). And since then efficacy of many medicinal plants in the treatment of many diseases have been put to test in many laboratories (Shajahan and Ramesh, 2004).

Olax subscorpioidea can either be a shrub or tree; it is up to 10 m or more in height. This plant is widely distributed in Nigeria, Zaire and Senegal part of Africa. The stem part of this plant is believed to possess medicinal properties. In this study, we report the antimicrobial activity of the aqueous and ethanoic extract of the stem of *O. subscorpioidea* and the preliminary screening for phytochemicals in the aqueous and ethanoic extracts.

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MATERIALS AND METHODS

The stem of *O. subscorpioidea* was bought in a local market in Ogbomoso, Nigeria. The identity of the plant was confirmed at the Department of Pure and Applied Biology, LAUTECH, Ogbomoso. Stems were air dried for 2 months, powdered and then extracted with 95% ethanol and distilled water for 48 h. The extract was filtered and the filtrate concentrated over a water bath to get the crude extract.

Antimicrobial activity screening

The antimicrobial activity of the crude extract was screened against four gram negative bacteria and gram positive bacteria together with four fungi isolated from the Baptist Medical Center, Ogbomoso and the effluents of a pharmaceutical industry in Lagos, Nigeria. The organisms are *Staphylococcus aureus, E. coli, Salmonella sp., Pseudomonas aeruginosa, Proteus vulgaris, Aspergillus niger, Aspergillus tamarii, Saccharomyces cerevisiae* and *Fusarium oxysporum.*

The antimicrobial activity was determined by the paper disc diffusion method (Garg and Jain, 1988) using nutrient agar and potato dextrose agar plates previously inoculated with 18 h old broth culture or spores suspension in sterile distilled water of the test organisms respectively. Sterilized paper discs (6 mm), soaked in the crude ethanoic and aqueous extracts of *O. subscorpioidea* (0.05 g/ml and 0.072 g/ml respectively) were then lay on the agar surface. Antibiotic discs of gentamycin (10 µg/m) and ofloxacin (30 µg/ml) were used as control for bacteria and clotrimazole (30 µg) for fungi. The experiment was performed in triplicates and were then incubated at 37°C for 24 – 48 h for bacteria and room temperature for 72 h for the fungi after which the zones of inhibition formed were

Table 1. Preliminary phytochemical screening of aqueous and ethanoic extracts of *O. subscorpioidea*.

Tested group	Aqueous extract	Ethanoic extract
Colour	Light brown	Wine
Tannins	+	+
Alkaloids	-	+
Glycosides	+	+
Saponins	+	-
Flavonoids	-	+
Steroids	-	+

Key: + = present, - = absent.

Table 2. Antimicrobial activity of *Olax subscorpioidea.*

Organisms	Sources of organism	Ethanolic extract(mm)	Water extract(mm)	Gentamycin (mm) 10 µg	Ofloxacin (30µg)
Bacteria	Hospital (HVS isolate)	14.8	NA	11.3	NA
S. aureus	Hospital (wound isolate)	17.5	NA	15.0	17.0
S aureus	Pharmaceutical industry waste water isolate	21.5	NA	14.7	NA
S. aureus	Hospital (urine isolate)	10.5	NA	31.5	22.0
E. coli	Pharmaceutical industry waste water isolate	7.5	NA	21.6	NA
E. coli	Pharmaceutical industry waste water isolate	7.2	NA	14.2	7.0
Pseudomonas sp	Hospital isolate	18.5	14.5	22.0	22.0
Proteus vulgaris sp	Hospital (blood isolate)	15.7	8.7	21.5	11.5
Salmonella Sp.	Pharmaceutical industry waste water isolate	11.7	NA	Clotrimazole	
Fungi	Pharmaceutical Industry waste water isolate	10.5	NA	(30 µg)	
Aspergilus niger	Laboratory isolate	11.0	NA	32.5	
A. tamarii	Laboratory isolate	9.5	NA	37.7	
Saccharomyces cerevisiae				21.5	
Fusarium oxysporum				30.5	

Key: NA – Not active

measured and average diameter of zone of inhibition were obtained.

The minimum inhibitory concentration (MIC) of the crude extracts were also determined using the same method except that the paper discs were soaked in different concentrations of the crude extracts in water

Preliminary phytochemical studies

The extracts were subjected to various phytochemical tests to determine the active constituents present in the crude aqueous and ethanoic extracts. The slightly modified method of Okerulu and Ani (2001) was used.

RESULT AND DISCUSSION

The phytochemical screening of the aqueous and ethanolic extracts of *O. subscorpioidea* was depicted in Table 1. The phytochemical results reveals the presence of tannins, glycosides and saponins in the aqueous extract while the ethanoic extract shows the presence of tannins, alkaloids, glycosides, steroids and flavonoids.

The ethanoic extract of *O. subscorpioidea* showed considerable antibacterial activity against the bacterial isolates with zones of inhibition ranging between 7.2 – 21.5 mm in diameter (Table 2). The highest zone was against the *S. aureus* isolated from the effluents of a pharmaceutical company while the least zone was against *P. aeruginosa* isolated from the effluents of a pharmaceutical company. The crude aqueous extract was only active against species of *Pseudomonas* isolated from the hospital, *Proteus* sp. and *salmonella* sp. also isolated from hospital with zones of inhibition of 15.5, 14.5 and 8.7 mm, respectively. The potency of the crude extracts was comparable to those of antibiotics which are pure substances. Gentamycin have a universal activity

Table 3. The zone of inhibition formed by ethanolic extract of O. *subscorpioidea and MICS.*

Micro organisms	Source of organisms	Zones of inhibition (mm)	MICS (mg/ml)
+*E. coli*	Hospital (urine isolate)	7.3	5.0
Salmonella sp.	Hospital isolate	11.5	15.0
Pseudomonas sp.	Hospital (wound isolate)	12.8	37.5
S. aureus	Pharmaceutical industry waste	15.0	45.0
S. aureus	water isolate	13.7	25.0
S. aureus	Hospital (wound isolate)	10.3	15.0
Proteus vulgaris	Hospital (HVS isolate) Hospital isolate	14.2	40.0

against the entire test organism, with zones of inhibition ranging from 11.3 to 31.5 mm, while four organisms were resistant to ofloxacin (Table 2). The ethanoic extract also showed moderate activity against the fungal isolates with zones of inhibition ranging from 9.5 to 11.7 mm while the aqueous extract showed no antifungal activity (Table 3). The minimum concentration of the ethanoic extract to inhibit the test organism ranged from 5 to 45 mg/ml. The lowest MIC of 5 mg/ml was recorded against the *E. coli* isolated from the urine in hospital while the highest MIC of 45 mg/ml was recorded against *S. aureus* isolated from the pharmaceutical industry waste water.

In conclusion, results from this research shows that the ethanoic extract of *O. subscorpioidea* is a broad spectrum agent which can be used against both gram positive and gram negative bacteria and also fungi. While the aqueous extract has little antibacterial property.

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