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PHYTOCHEMICAL AND ANTIMICROBIAL ACTIVITY EVALUATION OF THE ETHANOLIC LEAF EXTRACT OF *MITRACARPUS SCABRUM*

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ABSTRACT
The present study was designed to evaluate the phytochemical components and antimicrobial activity of Mitracarpus scabrum extracts on selected bacteria and fungi species. Phytochemical screening was carried out using standard protocols while the disc diffusion method was used to test for the activity of the extract against the test organisms. Phytochemical screening of the ethanolic extract of the herb revealed the presence of alkaloids, flavonoids, saponins, terpenoids, and steroids. The antimicrobial activity of the extract was assayed on three bacterial species (Escherichia coli, Staphylococcus aureus and Salmonella paratyphi) and two fungi species (Candida albicans and Aspergillus flavus) at varying concentrations of the extract between 20mg/ml to 80mgl/ml at 37^0C. The results showed that the ethanolic extract of Mitracarpus scabrum has both antibacterial and antimycotic effect on Salmonella paratyphi (zone of inhibition of 9.0mm/ml) and Aspergillus flavus (zone of inhibition of 8.0mm/ml) respectively. Candida albicans, Escherichia coli and Staphylococcus aureus were found to be resistant to the effect of this extract. The plant extract can thus be used in the development of alternative therapeutic agents against Salmonella paratyphi and Aspergillus flavus with less side effects compared with synthetic drugs.

Keywords: Mitracarpus scabrum, phytochemical, ethanolic extract, antimicrobial

INTRODUCTION

Plant compounds are of interest as a source of safer or more effective substitutes than synthetically produced antimicrobial agents. Phytochemical progress has been aided enormously by the development of rapid accurate methods for screening plants for particular chemicals (Abere *et al*., 2007). These procedures have shown that many substances originally thought to be rather rare in occurrence are of almost universal distribution in the plant kingdom. The drugs contained in medicinal plants are divided chemically into a number of groups among which are alkaloids, volatile essential oil, plants and phenolic glycosides, steroids, tannins and terpenes (Reuben *et al.*, 2008).

The use of herbs in the treatment of animal and human diseases has long been established (Adebolu and Oladimeji 2005; Sofowora, 1982). Most plant extracts have been shown to possess anti-microbial agents active against microorganism *invitro*. These plants contain medieval properties that make them potent to cure or prevent diseases (Sofowora, 1982). *Mitracarpus scabrum* belongs to the family Rubiaceae. It is known as "Harwachi" among the "Hausa" speaking people of northern Nigeria. The leaves are highly used for the treatment of skin diseases such as mycosis, scabies and eczema. The young leaves are squeezed and rubbed on the affected corporal parts two or three times per day (Cimanga *et al*., 2004). It is also commonly used for the treatment of headaches, toothache, amenorrhoea, dyspepsia, hepatic and venereal diseases in some parts of West Africa (Obeidat *et al*., 2012).

Therefore, this study was aimed at evaluating the antimicrobial activity of *Mitracarpus scabrum* in line with its ethnomedicinal use.

MATERIAL AND METHODS

Collection and identification of plant materials

The leaves of *Matracarpus scabrum* were obtained from Federal college of Education, Bichi, Kano State and identified at the Botany unit of the Kebbi State University of Science and Technology, Aliero by Dr. Dhramendra Singh. The leaves were dried at room temperature for two weeks and ground using mortar and pestle to obtain the powdered sample which was stored in a paper bag before the extraction process was carried out.

Solvent Extraction

Fifty grammes (50g) of the powdered leave sample was transferred into the soxhlet extractor inside a thimble and 100ml of ethanol was used as the extracting solvent. This was refluxed for about 3 days and the extracting solvent was finally

evaporated using rotatory evaporator to get the crude extract at 70^0C.

Phytochemical Analysis

The extract was evaluated for the presence of saponins, tannins, flavonoids, steroids, glycosides and alkaloids using methods described by Sofowora (1993) and Trease and Evans (2002).

Antimicrobial Activity

The test organisms including three bacterial species and two fungi species were obtained from Federal Medical Centre Birni Kebbi and cultured in bacteriological Mueller-Hinton Broth for bacteria at 37^0C for 24 hours and Sabouraud Dextrose broth for fungi at 30^0C for 48hours. The disc diffusion method was used to test for the activity of the extract against the test organisms. An inoculum suspension was swabbed uniformly to solidified 20 ml Mueller-Hinton Agar (MHA) for bacteria and Sabouraud Dextrose Agar (SDA) for fungi and the inoculum was allowed to dry for 5 minutes. Sterile discs of six millimeter were impregnated in triplicates with 20, 40 and 80 mg/ml of test extract and introduced into the upper layer of the seeded agar plate. The plates were incubated overnight at 37^0C. Amoxicillin and distilled water were used as positive and negative control respectively. The antimicrobial activity was recorded by measuring the width of the clear inhibition zone around the discs using transparent ruler (mm).

RESULTS AND DISCUSSION

Phytochemical screening of the ethanolic leaf extract of *Mictracarpus scabrum*

The result of the phytochemical screening reveals the presence of cardiac glycoside, tannins, saponins, anthraquinone, steroid, flavonoid, terpenoids, glycoside and saponin glycoside while alkaloid was absent. Saponins are special class of glycosides that have been shown to have antifungal property (Ekpendu, 1995). The function of saponins in plants is to protect the plant from parasite. Saponins have similar protective action in humans as they exhibit antibacterial and antiviral properties (Abere *et al*., 2007)

Tannins have been reported to inhibit growth of microorganisms by precipitating microbial pattern and making nutritional proteins unavailable for them (Ekpendu, 1995). Wide range of anti-infective actions has been assigned to tannins (Ceullar *et al.,* 2010). The presence of tannins in the extract may be responsible for the traditional use of *Mitracarpus scabrum* in the treatment of bruises and superficial wounds (Alinnor, 2008)

Table 1: Result of the Phytochemical screening of the ethanolic leaf extract of *Mictracarpus scabrum*

Chemical Constituents	Test Results
Alkaloids	-
Cardiac glycosides	+
Tannins	+
Saponins	+
Anthraquinones	+
Steroids	+
Flavonoids	+
Terpenoids	+
Glycosides	+
Saponin Glycosides	+

Key:

\+ = Present

\- = Absent

Antimicrobial activity of the ethanolic leaf extract of *Mitracarpus scabrum.*

Ethanolic extract of the leaf of *Mitracarpus scabrum* was assayed for its antimicrobial activity on *E. coli*, *Aspergillus flavus*, *C. albicans* and *Salmonella paratyphi* at various concentrations using the disk diffusion method. The result is presented in Table 2. The result revealed that *Salmonella paratyphi* is more sensitive to the ethanolic extract at 80mg with an inhibition of 9.0mm. The extract shows no bactericidal effects on *E. coli* and *Staphylococcus aureus i*n the present study. This result is in agreement with works of Banso, (2009). Other researchers have however found the methanolic extracts of some plant species are active against *E. coli* and *S. aureus* (Bisignano *et al.,* 2000; Ekpendu *et al.,* 2008). This could be attributed to the fact that different solvents have various degrees of solubility for different phytoconstituents (Cowan, 1999). The extract is also found to have strong antimycotic action against *Aspergillus flavus* even at low concentration. This shows that the ethanolic extract of *M. scabrum* has both fungistatic and fungicidal activity against the fungi. The result however shows that the extract is not active against *C. albicans*. The result shows that the plant extract is actually a good source of antimicrobial agents as suggested by the local populace. Due to their bioactive compounds, medicinal plants are known to

harbor prospective endophytic microbes. The genus Mitracarpus, consists of no fewer than 50 species (Arruda and Gomes, 1996). Many authors have reported that the plant has both antibacterial and antifungal activities. Herbal medicines derived from plant extracts are being increasingly utilized to treat a wide variety of clinical diseases including liver diseases (Cowan, 1999; Benjamin et al., 1986; Gyl, 1992). Rather, the extracts ' inhibition of organism development can be ascribed to the potency of active values as well as the strain and potentially weakness in the strength of the used organism. Ahonkhai et al., (1999) reported the activity of the crude extract of *Mitracarpus scarbrum* in a liquid soap formulation and a whole plant was employed, in this study on the ethanol extracts of the leaves were employed.

Table 2: Result of antimicrobial activity o/:"IIIO of the ethanolic leaves extract of *mitracarpus scabrum.*

	Controls		Concentration (mg/ml) Zone of Inhibition (mm)		
Bacteria	**Positive Control (Streptomycin)**	**Negative Control (Distilled Water)**	**20mg**	**40mg**	**80mg**
E.Coli	33.0	-	-	-	-
S.aureus	21.0	-	-	-	-
S. Paratyphi	22.0	-	4.0	8.0	9.0
Fungi					
C. Albicans	22	-	-	-	-
A. flavus	21	-	5.0	6.0	8.0

- = No zone of inhibition

CONCLUSION

The plant studied could be an answer for people seeking for better therapeutic agents from natural sources which are believed to be more efficient at high concentration with little or no side effects when compared to the commonly used synthetic chemotherapeutic agents. The present study verified the traditional use of *Mitracarpus scabrum* for the treatment of human related ailments. Thus this plant could be utilized as an alternative source of useful drugs. Compounds with antimicrobial activity can be isolated further from this plant. With further researches, new chemotherapeutic agents could possibly be developed from these plants.

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