

Full Length Research Paper

Does host plant affect the antibacterial activity of *Tapinanthus bangwensis* (Engl. and K. Krause) Danser (Loranthaceae)?

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The extracts of fresh leaves and twigs of *Tapinanthus bangwensis* (Engl. and K. Krause) obtained from different host plants were screened for their antibacterial activities against six human pathogenic bacteria. The methanolic extracts showed activity against *Shigella dysenteriae* and *Salmonella typhimurium*. The antibacterial activity of chloroform extracts, where activity was shown, revealed more activity than those of methanolic extracts. Antibacterial activities vary according to host plants on which *T. bangwensis* was obtained.

Key words: *Tapinanthus bangwensis*, antibacterial activity, Loranthaceae, Pathogens, Nigeria.

INTRODUCTION

Herbal medicine is an alternative form of therapy and has become the mainstream throughout the world due to the growing resistance of pathogens to conventional antibiotics (De Smet, 2002). The screening of plant extracts and plant products for their antimicrobial activity has in most cases involved higher plants, many of which have shown clinical relevance as sources of potential chemotherapeutic agents (Essawi and Srour, 2000; Srinivasan et al., 2001; Hamil et al., 2003; Arias et al., 2004; Shilpi et al., 2006; Van wyk, 2008; Kosalge and Fursule, 2009). The development of herbal products is dependent on local botanical flora. Medicinal plants are distributed worldwide and many abound in tropical countries. Nigeria has a rich variety of medicinal plants distributed in the different geocological regions of the country. The Loranthaceae constitutes the largest group of parasitic plants with about 950 distributed in 77 genera (Engone et al., 2006). Some Loranthaceae are known to be real pestilences in the natural forests, plantations,

cultivated fruit trees and ornamental plants and they cause important damages on the harvests (Sonke et al., 2000; Boussim et al., 2004). Didier et al. (2008) reported eight species belonging to 4 genera in Douala, Cameroon.

Their report also indicated that the plants are found parasitizing a large range of host plants. *Tapinanthus* spp. was found to be ubiquitous and more abundant in their studied sites. Loranthacean mistletoe (including *Tapinanthus bangwensis* (Engl. and K. Krause) Danser and other species are widely distributed in Nigeria and the plants are found on many host trees. The leaves and young twigs of the plants have been used in folklore medicine to treat different diseases such as circulatory and respiratory disease problems, malaria, diabetes, hypertension and sterility in cow. There is paucity of reports on the antimicrobial properties of these plants and no report exists on whether the hosts on which they are found could influence their antimicrobial property. The present study was undertaken to ascertain the antimicrobial potency of *T. bangwensis* against some diarrhea-causing bacteria and to detect if its antibacterial potency is influenced by the host on which it is found.

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MATERIALS AND METHODS

Source of plants

Fresh leaves and twigs of *T. bangwensis* were collected from Iseyin, southern part of Oyo state and Chaga village in Abuja, Nigeria. The plants were authenticated by the taxonomist at the Department of Botany and Microbiology, University of Ibadan, Nigeria and were designated T1 - T4 on the basis of the host plants from which they were collected. T1 was from *Triclisia gillettii* (De Wild.) Staner, T2 from *Parkia biglobosa* (Jacq.) Benth, T3 from *Citrus aurantifolia* (Christm.) Swingle and T4 from *Phyllanthus muellerianus* (Oktze) Exell.

Preparation of plant materials and extracts

Leaves were air-dried on the herbarium table (25 - 28°C) after which they were shredded and preserved in airtight cellophane bags. The shredded leaves were milled into powder form using a warring commercial blender. Fifty grams of each of the powdered plant materials was soaked in 200 ml of methanol and chloroform separately and stoppered. The flasks were manually agitated at intervals for 5 days. The resulting extracts from each flask was filtered rapidly through layers of gauge and then through Whatman No. 1 filter paper. The resulting filtrates were then concentrated by evaporation in a rotary evaporator. The yield of extracts obtained from methanol was 2.3 and 1.4 g from chloroform.

Phytochemical studies

Phytochemical tests were carried out to determine the presence of flavonoids, tannins, alkaloids, saponins and anthraquinones using the methods described by Odebiyi and Sofowora (1978).

Test organisms

Staphylococcus aureus, *Salmonella typhimurium*, *Salmonella typhi*, *Pseudomonas aeruginosa* and *Escherichia coli* strains were obtained from the Nigeria Institute of Medical Research (NIMR), Yaba, Lagos, Nigeria while *Shigella dysenteriae* was obtained from the Department of Medical Microbiology, University College Hospital, Ibadan, Nigeria. Isolates were from clinical sample of diarrhoeic patients. The isolates identities were further confirmed in our laboratory using standard biochemical procedures (Barrow and Feltham, 1993). The isolates were maintained on Tryptone Soy agar (TSA) (Oxoid) at 4°C before use for this work.

Determination of antibacterial activity

The medium used was Mueller Hinton agar (Oxoid, U.K). The bacterial inoculum were adjusted to 0.5 McFarland turbidimetric standard and inoculated onto the medium using sterile swabs. For each extract, three replicate plates were prepared against the test organisms. Antibacterial activity of the methanolic and chloroform extracts of the plant samples was evaluated by the agar well diffusion method. Using sterile cork-borer of 6 mm diameter, equidistant wells were cut in each of the agar plates. Different concentrations of the extracts, 100, 150, 200, and 250 mg/ml were introduced into the wells. The plates were left for 2 h at room temperature to allow the extract to diffuse. The solvents used for extraction served as controls and were introduced into a separate well as appropriate. Gentamicin disc (30 µg/ml) was used as standard antimicrobial for comparison. The plates were then incubated at 37°C for 24 h. Antimicrobial activity was determined by

measurement of zone of inhibition around each well using a pair of calipers (in mm.) and read on a meter rule.

RESULTS AND DISCUSSION

Results of the phytochemical analysis as summarized in Table 1 showed that both the methanolic and chloroform extracts revealed the presence of tannins, flavonoids, alkaloids and saponins while anthraquinones were not detected. Majority of the phytochemical constituents present have been known to possess antibacterial activities (Sofowora, 1992; Lullerodt et al., 1999; Pretorius and Watt, 2002). Results of the antibacterial activities of both the methanolic and chloroform extracts show that the plant demonstrated activities against the test organisms to varying degrees. For methanolic extracts, only T3 showed no activity against *P. aeruginosa*. T2 showed no activity against *E. coli* while T1 and T2 showed no activity against *S. typhi*. For the chloroform extract T1 and T3 showed no activity against *S. typhimurium*. T1 and T2 showed no activity against *P. aeruginosa*. T3 and T4 showed no activity against *S. typhi*, *S. aureus* and *S. dysenteriae*. It would appear that the methanolic extract showed activity against many of the test organisms at the different concentration tested. However where activity was shown against the test organisms in the chloroform extract the zones of inhibition exceeded, in many cases, those obtained for the methanolic extract. For example, the chloroform extract had a higher inhibition than the methanolic extract for *S. aureus* with T2 and T3. Whereas the methanolic extract of T4 showed activity against *S. aureus*, the chloroform extract of T4 showed no activity at all against the same organism (Table 2).

The results obtained indeed suggest that the host plants on which *T. bangwensis* are found can influence the antibacterial activity of the plant. All Loranthaceans are parasites of the xylem tissue and depend on their hosts for water, nutrients and some carbon compounds (Didier et al., 2008). This dependence may in fact have added to their antibacterial activity since many of the plants on which they parasitize are known to have medicinal properties, and hence there may have been nutritional exchange with the host tissues. For example, *Phyllanthus* sp, one of the host plants in this investigation, has been reported to have many traditional uses for a wide variety of diseases such as diarrhea, kidney disorders, gonorrhoea and cough in Malaysia (Burkill, 1996). However, it has also been reported that some host plants, for example *Mangifera indica*, contain polyphenolic compounds rich in tannins and flavonoids which are deleterious to the survival of some cultivars of mistletoe (Hariri et al., 1991). The report of Ermel et al. (1986) showed that the concentration of insecticidal principle in seed kernels of *Azadirachta indica* A. Juss. Varied with humidity, geographical location, individuality of trees among other factors. Olafimihan (2004) also

Table 1. Phytochemical analysis of extracts of *Tapinanthus bangwensis*

Extract	Phytochemical constituents				
	Tannins	Alkaloids	Saponin	Anthraquinones	Flavonoids
Methanol	+	+	+	-	+
Chloroform	+	+	+	-	+

Table 2. Antibacterial activity of *Tapinanthus bangwensis* against selected bacteria.

Plant designate	Test organism	Zone of inhibition (mm)							
		Methanol extract (mg/ml)				Chloroform extract (mg/ml)			
		100	150	200	250	100	150	200	250
T1	S.d.	10.3	10.8	11.4	14.0	10.0	10.2	10.2	11.0
	S.ty	10.0	16.0	17.0	19.8	0	0	0	0
	P.a	0	0	0	0	0	0	0	0
	S.t	0	0	0	0	15.0	20.2	21.0	23.0
	E.c	6.0	7.0	10.0	14.0	8.0	12.0	16.0	18.0
	S.a	0	0	0	0	10.0	10.2	10.2	12.0
T2	S.d.	10.0	10.2	10.8	11.4	15.0	20.1	21.3	27.0
	S.ty	0	10.6	10.8	14.0	0	10.0	14.0	20.0
	P.a	0	0	0	0	0	0	0	0
	S.t	0	0	0	0	20.0	20.0	21.0	30.0
	E.c	0	0	0	0	15.0	20.1	20.2	28.0
	S.a	10.2	14.1	15.0	16.2	17.0	17.0	19.8	24.0
T3	S.d.	10.0	17.0	13.0	12.0	10.0	13.0	10.0	10.0
	S.ty	10.0	21.0	10.0	10.0	0	0	0	0
	P.a	10.0	9.0	9.0	10.0	0	11.0	0	0
	S.t	9.0	16.0	11.0	11.0	24.0	13.0	13.0	16.0
	E.c	0	0	0	0	0	0	0	0
	S.a	12.0	12.0	14.0	21.0	16.0	18.0	20.0	22.0
T4	S.d.	0	0	0	9.0	0	0	0	0
	S.ty	9.0	13.0	15.0	15.0	0	0	16.0	16.0
	P.a	0	0	0	0	16.0	14.0	16.0	16.0
	S.t	0	12.0	11.0	11.0	0	0	0	0
	E.c	14.0	17.0	17.0	13.0	0	0	0	0
	S.a	16.0	24.0	18.0	18.0	0	0	0	0

T1 = *Tapinanthus bangwensis* on *Triclisia gillettii*, T2 = *Tapinanthus bangwensis* on *Parkia biglobosa*, T3 = *Tapinanthus bangwensis* on *Citrus aurantifolia*, T4 = *Tapinanthus bangwensis* on *Phyllanthus muellerianus*, S.d = *Shigella dysenteriae*, S.ty = *Salmonella typhimurium*, P.a = *Pseudomonas aeruginosa*, S.t = *Salmonella typhi*, E.c = *Escherichia coli*, S.a = *Staphylococcus aureus*,

reported the influence of season on the antibacterial activity of stem bark of *A. indica*. The fact that there are varying degree of inhibition by extracts obtained using different solvents also reveal that solvents used in extraction may also influence the antibacterial activity of the plant. It may well be that chloroform is a better extracting solvent for the active ingredient as observed in this study. Loranthaceae have higher rate of therapeutic properties (Didier et al., 2008). Hence, their parasitic

nature notwithstanding, the current study could be extended to include other genera and species of Loranthaceae found in Nigeria. Further work could also be done using other extracting solvents.

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