

Full Length Research Paper

Phytochemical and antimicrobial activity of *Terminalia avicennioides* extracts against some bacteria pathogens associated with patients suffering from complicated respiratory tract diseases

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Ethanollic extracts of the stem bark, root bark and leaves of *Terminalia avicennioides* were assayed against *Staphylococcus aureus*, *Escherichia coli* and *Pseudomonas aeruginosa* obtained from patients suffering from complicated respiratory tract infections for their antimicrobial activity. Analysis of variance showed that the ethanollic extracts of the plant material exhibit a significant ($P > 0.05$) antimicrobial activity against *S. aureus*. The various zone of inhibitions on *Escherichia coli* was moderate ($P < 0.05$) at different concentrations, very low antimicrobial activity was observed against *P. aeruginosa*. Important bioactive agents like phenols, steroids, glycosides, flavonoids, tannins, ellagic acids were detected in the bark, leaves and root of *T. avicennioides*. Anthraquinone was not absent in all the plant parts tested. The bioactive constituents detected in *T. avicennioides* may be responsible for the observed antimicrobial activity of the plant and hence, its potential use as antimicrobial agent.

Key words: Antimicrobial activity, bacterial pathogens, Phytochemical, respiratory tract diseases, *Terminalia avicennioides*.

INTRODUCTION

Medicinal plants are major sources of obtaining antimicrobial agents (Sofowora, 1986). Plants are used medicinally worldwide as sources of many potent drugs (Iwu et al., 1999). Traditional medical practitioners in Nigeria use a variety of herbal preparations to treat different kinds of diseases including microbial infections (Sofowora, 1993). The search for new bioactive agents led to the screening for bioactive compounds in *Terminalia* species (Combretaceae). The selection of this genus is based on its uses in traditional medicine both in Africa and in Asia for the treatment of microbial infections. Many of the species from Combretaceae family are reputed to contain antimicrobial constituents (Baba-Moussa et al., 1999). *Terminalia* species occurring in southern Africa have exhibited substantial antifungal activity (Baba-Moussa et al., 1999). Most of the crude extracts had minimum inhibitory concentration (MIC) values of 0.08 mg/ml, and some had MIC's as low as 0.02 mg/ml (Masoko et al., 2005). Therefore, research into the anti-

microbial activity of these local medicinal plants is expected to enhance the use of these plants against diseases caused by bacteria. The active principles of many drugs found in plants are secondary metabolites (Cragg and Newman, 2005). Hence, basic phytochemical investigation of the extracts for their main phytochemicals is very vital. *T. avicennioides* Hoshst, family: Combretaceae, found in Nigeria with these Vernacular names: Nupe - Kpace, Gwari - Kpayi, Hausa - Baushe, Yoruba - Igiodan, Igbo - Edo have been reported to possess antimicrobial activities (Abdullahi et al., 2001; Akinside et al., 1995; Akinyemi et al., 2000; Akinyemi et al., 2005. Mann et al., 2008)

There is still little evidence on the antimicrobial properties of the plant under investigation against majority of the microbes particularly clinical isolates of patients suffering from complicated respiratory tract diseases. The fact that this medicinal plant plays paramount role in the management of ailments in rural communities, there is need for further verification of its activities against endemic microbial pathogens in order to establish scientific rationale for its use as antimicrobial agents. In the present study, the ethanollic extracts from different parts of *T. avicennioides* were screened for phytochemical constituents and anti-

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Table 1. Phytochemical screening of *T. avicennioides* extracts

Plant part	Alkaloid	Glycoside	Steroid	Phenol	Tannin	Saponin	Flavonoid	Ellagic acid	Anthraquinone
Leaves	ND	+	ND	+	+	ND	ND	+	ND
Stem barks	ND	+	ND	+	+	+	+	+	ND
Root barks	+	+	+	+	+	+	ND	+	ND

(+) = detected; ND = Not detected

Table 2. Effect of graded concentrations of *T. avicennioides* on *Ps. aeruginosa*.

Concentration (mg/ml)	Mean diameter of zone of inhibition (mm) ± S.D				
	Control	Stem	Root	Leaves	Root/stem/leaves
10	NZI	NZI	NZI	NZI	NZI
20	NZI	NZI	NZI	NZI	NZI
30	NZI	NZI	NZI	NZI	3.50±1.40
40	NZI	7.00±0.00	8.00±0.00	5.50±1.70	9.50±0.00
50	NZI	10.50±1.40	9.00±0.00	12.50±0.00	11.50±1.10

S.D: Standard deviation; NZI: No zone of inhibition; Values represent means of triplicate determinations

icrobial activity against *S. aureus*, *P. aeruginosa* and *E. coli* clinically isolated from patients suffering from complicated respiratory tract diseases.

MATERIALS AND METHODS

Plant materials

The stem bark, root bark and leaves of *T. avicennioides* were collected from Emitete village, near Bida, Niger State. The plant parts were dried at room temperature for one month. The dried plant parts were then pulverized and stored in polythene bag until required. The plant was identified at National Institute for Pharmaceutical Research and Development (NIPRD) where the voucher specimen was deposited with the Herbarium number NIPRDH 5735.

Test organisms

Fresh clinical isolates of *E. coli*, *S. aureus* and *P. aeruginosa* isolated from patients suffering from complicated respiratory tract diseases were used in this study. The organisms were obtained from the medical microbiology laboratory of the Federal Medical Centre, Bida, Niger State and maintained on nutrient agar slant until needed.

Preparation of plant extracts

A sample (50 g) each powered plant parts (stem, root and leaves) were soaked in 95% ethanol (200 ml) for 72 h. At the end of the extraction, each extract was filtered using Whatman No.1 filter paper. The filtrate was concentrated in vacuum at 30°C and stored at 4°C until use. The graded concentrations (10 mg/ml, 20 mg/ml, 30 mg/ml, 40 mg/ml and 50 mg/ml) of the extracts were prepared for the bioassay.

Bioassay

The antimicrobial test was performed using the agar diffusion method of Nair and Chanta (2005). The test organisms were inoculated on nutrient agar plates and spread uniformly using sterile glass spreader. Wells of 5 mm diameter were made on the nutrient agar using a sterile cork borer. The cut agar disks were carefully removed by the use of forceps sterilized by flaming. To each well was introduced to the graded concentrations (10, 20, 30, 40 and 50 mg/ml) of the plant extracts. Control experiments comprising inoculums without plant extracts were set up. The plates were inoculated at 37°C for 24 h. The zones of inhibition were then recorded.

Phytochemical screening

The phytochemical analysis of the plant extracts was performed using the methods described by Harborne (1998); Evans (1989) and Sofowora (1993).

RESULTS

Bioactive ingredient such as tannins, alkaloids, saponins, steroids, phenols and glycosides were detected in most of the plant parts tested (Table 1). Anthraquinone was not detected in any of the plant extracts.

Alkaloids, glycosides saponins, steroids, phenols, tannins and ellagic acid were detected in the root bark of the plant. Alkaloids and steroids were however absent in the leaves and stem bark (Table 1). The results in Table 2 show the antimicrobial activity of the ethanolic extract of *T. avicennioides* against *P. aeruginosa* which was relatively low from those of Table 3. It was observed that *T. avicennioides* has a moderate antimicrobial activity against *E. coli* but high ($P > 0.05$) antimicrobial activity was observed against *S. aureus*.

Table 3. Effect of graded concentrations of *T. avicennioides* on *E. coli*.

Concentration (mg/ml)	Mean diameter of zone of inhibition (mm) ± S.D				
	Control	Stem	Root	Leaves	Root/stem/leaves
10	NZI	NZI	NZI	NZI	NZI
20	NZI	NZI	NZI	NZI	4.00±0.17
30	NZI	NZI	NZI	NZI	7.50±0.00
40	NZI	8.00±0.00	8.50±0.00	4.00±1.70	10.00±1.80
50	NZI	12.50±1.40	12.50±1.18	7.50±0.21	15.50±0.00

S.D: Standard deviation; NZI: No zone of inhibition; Values represent means of triplicate determinations

Table 4. Effect of graded concentrations of *T. avicennioides* on *S. aureus*

Concentration (mg/ml)	Mean diameter of zone of inhibition (mm) ± S.D				
	Control	Stem	Root	Leaves	Root/ stem /leaves
10	NZI	NZI	3.50±0.00	NZI	4.50±0.00 ^a
20	NZI	4.00±0.17 ^a	7.00±0.00 ^c	3.50±0.00 ^a	3.00±0.00 ^a
30	NZI	4.50±0.41 ^a	8.50±0.00 ^c	4.00±0.00 ^a	3.00±0.00 ^a
40	NZI	12.00±0.00 ^a	10.50±0.00 ^c	8.00±0.00 ^b	7.00±0.00 ^b
50	NZI	18.00±1.40 ^b	15.00±0.00 ^d	11.00±0.17 ^b	11.00±0.17 ^b

S.D: Standard deviation; NZI: No zone of inhibition; Means ± SD followed by different letters in a column are significantly different at P < 0.05

DISCUSSION

Phytochemical screening of ethanolic extracts of *T. avicennioides* shows the presence of glycosides, saponins, tannins, alkaloids, steroids and phenol (Table 1). According to Sofowora, 1986, the presence of secondary metabolites in plants, produce some biological activity in man and animals and it is responsible for their use as herbs. Hence, the presence of the secondary metabolites in *T. avicennioides* may be responsible for its potential use as drug by the Nupe indigenes of Bida and its environs. Analysis of variance shows that graded concentrations of *T. avicennioides* exhibits significant (P < 0.05) activity against *S. aureus*. The inhibitory activity exhibited by the secondary metabolites tends to agree with the reports of Leven et al. (1979) and Scherbonvaski (1971) both of which linked the antibacterial properties of plants to the presence of secondary metabolites. Several plants which are rich in alkaloids have been shown to possess antimalarial activity against a number of microorganisms. For example, Adebayo et al. (1983) investigated the antimicrobial activity of leaf extract of *Eugenia uniflora* and reported that alkaloids, tannins and glycosides were detected and that the ethyl acetate and methanolic leaf extracts of the plant were active against *E. coli*, *P. vulgaris*, *K. pneumoniae* and *A. niger*. Low antimicrobial activity was observed against *E. coli* (Tables 3 and 4). The zone of inhibition of the tested organisms indicated their susceptibility to the plant parts used in this study. It was observed that the zone of inhibition varies from one

organism to another and from one plant parts to another at different concentrations. According to Prescott (2002) the activity of antimicrobial agent is concentration dependent. Hugo (1998) also indicated that the position of the zone edge (diameter of inhibition) is determined by the initial population density of the organism, their growth rate and the rate of diffusion of the antimicrobial agent. The results obtained in this study showed that the ethanolic extracts of stem bark, root bark and leaves of *T. avicennioides* has antimicrobial activities on test organisms used in this study. It may therefore be suggested that the constituents of this plant can be used in chemotherapy. This study represents the preliminary report on antimicrobial activity of the crude extracts of *T. avicennioides* against the clinical isolates of complicated respiratory tract infectious diseases in Niger state, Nigeria.

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