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Short Communication

Antimicrobial screening of Terminalia avicennoides and Acalypha wilkesiana

Akpomie, O. O*. and Olorungbon, S.

Department of Microbiology, Delta State University, Abraka, Delta State, Nigeria.

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The antimicrobial activities of extracts of Terminalia avicennoides (Mexican tea) and Acalypha wilkesiana (copper leaf) were screened against Pseudomonas aeruginosa, Bacillus subtilis, Escherichia coli, Klebsiella, Salmonella typhi, Staphyloccus aureus and Candida albicans. The extracts showed a better activity against all organisms but T. avicennoides showed high activity against C. albicans. The antimicrobial activities for both plant extracts were more pronounced at a high concentration than at low concentration of the extracts. The extracts of the two plants can be used in the treatment of diseases where the test organisms are implicated.

Key words: Antimicrobial activities, Terminalia avicennoides, Acalypha wilkesiana.

INTRODUCTION

The problem of the emergence of strains of micro-organisms that are resistant to most of the present day antibiotics requires the use of new antimicrobial agents. Plants have been found to possess the broadest spectrum of synthetic activity and are the source of many useful antimicrobial compounds (Ayodele and Banuso, 2004). Many plants, for example, Thymus vulgaris have both antibacterial and antiviral properties, Cassia fistula and Mesua ferrea have antibacterial and antifungal activities (Ali et al., 2004). Many naturally occurring substances of plant origin cover a wide range of antimicrobial activity than synthetic chemical. The use of plants offers a better opportunity of providing antimicrobial activity against such organisms that are resistant to known antimicrobial agents (Amadi et al., 2007). There are still so many plants whose medicinal uses have not been ascertained. Plants which constitute an active part of the ecosystem have been found to be useful to man both as sources of foods and medicine. Only about 1 to 10% is used as food by both man and animals (Smith et al., 1996), so the remaining part can be used for the treatment of ailments.

In view of the resistance of so many organisms to the available antibiotics and the fact that many organisms are still resistant to the plants that have been researched on, there is the need to find out more on the potentiality of diverse plants as antimicrobial agents. This study therefore aims at assessing the antimicrobiall efficacy of Terminalia avicennoides and Acalypha wilkesiana on selected microorganisms.

*Corresponding author. Email: bunmakp@yahoo.com

Abbreviations: TAEE, *Terminalia avicennoides* ethanol extract; **TAAE,** *Terminalia avicennoides* acetone extract; **AWAE,** *Acalypha wilkesiana* acetone extract; **TAWE,** *Terminalia avicennoides* water extract; **AWWE,** *Acalypha wilkesiana* water extract; **TAS,** *Terminalia avicennoides*.

MATERIALS AND METHODS

Sample collection

The leaves and stem barks of T. avicennoides and A. wilkesiana were collected from Gyellesu, Zaria and were identified in the Herbarium of the Department of Biological Sciences, Ahmadu Bello University, Zaria. Organisms used in the study were gotten from pure culture isolates obtained from Microbiology Laboratory, Ahmadu Bello University, Zaria. They were sub cultured on nutrient agar (oxoid) slants and stored at 4°C.

Extraction of plant materials

The plant materials were washed, air dried and separately ground to powder in a moulinex blender. The powder of each plant was

Table 1. Phytochemical analysis of the plant extracts.

Component	TAEE	TAAE	TAWE	AWEE	AWAE	AWWE
Alkaloids	+	+	+	-	-	-
Tannins	+	+	+	+	+	+
Saponins	+	+	+	+	+	+
Flavoniods	+	+	+	+	+	+

+ Present; - absent.

Table 2. Minimal inhibitory concentration of ethanol, acetone and water extracts of the plant samples.

Test organism	TAEE	TAAE	TAWE	AWEE	AWAE	AWWE
P. aeruginosa	15.625	15.625	125	62.5	62.5	125
S. typhi	15.625	31.25	250	125	125	250
E. coli	31.25	125	250	31.25	31.25	62.5
B. subtilia	62.5	125	250	62.5	62.5	125
Klebsiella phenmoniae	-	250	62.5	125	125	-
S. aureus	-	125	250	31.25	31.25	-
C. albicans	-	125	250	125	125	250

stored in an airtight container and kept in the refrigerator at 4°C. 200 g of each powdered sample was extracted with 150 ml of 40% ethanol using soxhlet extractor. The resulting extract was evaporated to a constant volume and labeled T. avicennoides ethanol extract (TAEE). Soxhlet extraction was also done with 150 ml redistilled acetone. It was evaporated to dryness at 70°C and labeled T. avicennoides acetone extract (TAAE) and A. wilkesiana acetone extract (AWAE). 200 g of powder was dissolved in 100 m distilled water and maintained in a waterbath for about 4 h at 45°C and labelled T. avicennoides water extract (TAWE) and A. wilkesiana water extract (AWWE).

Phytochemical analysis

The method of Trease and Elaise (1996) was used to detect the presence of phytochemicals. The filtrate from each extraction was tested for the presence of tannins, alkanoids, saponins and flavonoids.

Antimicrobial screening

The antibacterial and antifungal screening was done with all extracts against Pseudomonas aeruginosa, Staphyloccus typhi, Escherichia coli, Klebsiella sp., Staphyloccus aureus and Candida albicans. Plates of nutrient agar (for antibacterial screening) and potato dextrose agar (for antifungal screening) were seeded heavily with test organisms. Holes (diameter 0.5 cm) were made in them and filled with 250 mg/ml of the sample solution. The plates were incubated at 37°C for 24 h for bacterial isolates and 48 h for fungal isolates. The diameter of zones on inhibition was measured and recorded (Amadi et al., 2004).

Determination of minimal inhibitory concentration

One gram of each extract was dissolved in 4 ml nutrient broth to give 250 mg/ml. Serial dilutions were made to give 125, 65.5, 31.25 and 15.625 mg/ml. Three drops of overnight broth culture of the test organisms were inoculated into the dilutions and inoculated at 37°C for 24 and 48 h for fungi. The lowest concentrations of the extracts that inhibited the growth of the test organisms were recorded as the minimal inhibitory concentration.

RESULTS AND DISCUSSION

The phytochemical analysis of the plant extracts as shown in Table 1 indicates that all the extracts of the two plants contain flavonoids, tannins, saponins and tannins. The extracts of A. wilkesiana did not contain alkaloids. Brantner and Grein (2004) were able to isolate the compounds in plant extracts and found them to be antimicrobial.

Table 2 shows that the extracts had varying degrees of antimicrobial activity against the test organisms. The ethanol extract of T. avicennoides (TAS) was more effective against the bacterial and fungal isolates. It inhibited the growth of 1, 2, 3 and 4 at a lower concentration than the other extracts. This can be attributed to the fact that activity of the biological active components of plants is better enhanced in the presence of ethanol and the higher extraction capacity of ethanol (Ali et al., 2004). Acetone extract of TAS also inhibited the growth of all the organisms but was more effective against P. aeruginosa and S. typhi at a low concentration. It inhibited the growth of the other organisms but at a high concentration. The other extracts were able to inhibit the growth of organisms at very high concentration and with some, there was no inhibition. An increase in the concentration of the extract brought about an increased antimicrobial activity, which agrees with the work of Yongabi et al. (2002) that higher concentrations of antimicrobial substances showed appreciable growth inhibition of bacteria. All the extracts

Table 3. Diameter of zones of inhibition of the extracts (cm).

Test organism	TAEE	TAAE	TAWE	AWEE	AWAE	AWWE
P. aeruginosa	20	16	10	10	12	10
S. typhi	16	14	8	5	10	5
E. coli	14	11	8	12	14	12
B. subtilia	8	9	6	5	10	5
K. phenmoniae	10	8	12	0	14	0
S. aureus	12	10	8	0	14	0
C. albicans	12	10	8	0	10	0

apart from TAEE were able to inhibit the growth of the fungus, C. albicans, but at high concentrations. The poor antimicrobial activity of some of the extracts might be attributed to the solvent extraction methods and the concentration of the active ingredients in the extracts. Uzel et al. (2005) showed that tannins and flavonoids were extracted with ethanol, water and ether, whereas saponins and alkaloids were not.

From the results of the diameters of zones of inhibition (Table 3), the AWEE, AWWE and AWWE showed moderate activity against P. aeruginosa, S. typhi, E. coli and B. subtilis. There was no activity against Klebsiella, S. aurerus and C. albicans. The TAEE, TAEE, TAWE and AWAE showed appreciable activity against all the organisms. TAEE was more effective against the fungus (C. albicans) than the other extracts.

Conclusion

The extracts of the two plants (T. avicennoides and A. wilkesiana) have appreciable antimicrobial activity against the test organisms. The solvent used in the extraction methods determines the chemicals (active ingredient) extracted which affects the concentration and invariably the antimicrobial activity of the plants. TAEE, TAAE, TAWE and AWAE were able to inhibit C. albicans. The extracts of the two plants are thus recommended in the treatment of diseases in which the test organisms are implicated. Further work is encouraged on: 1. the choice of solvents for the extraction of the active ingredients in the plant samples; 2. production of pure active compound, which can be used to formulate drugs against these organisms or used in combination with resistant drugs that are being used presently; 3. toxicological and clinical trials on experimental animals.

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