

Antimicrobial screening of *Bridelia micrantha*, *Alchornea cordifolia* and *Boerhavia diffusa*

K.A. Abo and J.S.Ashidi*

Department of Pharmacognosy, Faculty of Pharmacy, College of Medicine, University of Ibadan, Ibadan, Nigeria

*Department of Biological Sciences, Ogun State University, Ago-Iwoye, Nigeria.

Summary

This report is on the antimicrobial potential of *Bridelia micrantha*, *Alchornea cordifolia* and *Boerhavia diffusa* sourced from traditional healers through an ethnobotanical survey of anti-infective plants in Egbado South in Ogun State, Nigeria. Extracts of *B. micrantha* and *A. cordifolia* exhibited significant inhibitory activity against the pathogenic organisms. In some cases, the antibacterial activity was comparable to those of ampicillin and gentamycin. However, only the leaf of *A. cordifolia* showed reasonable antifungal activity when compared with Trosyd. The study shows that there is justification for the use of these medicinal plants in traditional medicine.

Keywords: *Bridelia micrantha*, *Alchornea cordifolia*, *Boerhavia diffusa*, antimicrobial activity

Résumé

Ce rapport est sur les potentiels antimicrobiennes du *Bridelia micrantha*, *Alchornea cordifolia* et *Boerhavia diffusa*. Provenant des medecins traditionels (herebeists) a travers une enquete ethnobotanique des plants medicinales a Egbado au sud de l'etat d'Ogun au Nigeria. Les extraits de *B. micrantha* et *A. cordifolia* ont montre une activite inhibitrice significative contre les agents pathogenes. Dans certains l'activite antibacterienne etait comparable a celles de l'ampicillin et gentamycine. Seule les feuilles de *A. cordifolia* a montre une activite contre les champignons compare au Trosyd. L'etude montre qu'il ya une justification sur l'utilisation de ces plantes medicinales dans la medecine traditionnelle.

Introduction

Bridelia micrantha (Hochst) Ball. (Euphorbiaceae), *Alchornea cordifolia* (Schum Thonn.) (Euphorbiaceae) and *Boerhavia diffusa* Linn. (Nyctaginaceae) are known in Yoruba language as "Eran", "Ipa" and "Eti-ponla" respectively [1]. These species are reported to be used in traditional medicine for the treatment of venereal diseases, ulcers and for mouth wash [2-4]. They were chosen from a collection of medicinal plants obtained from traditional healers through a previous ethnobotanical survey of anti-infective plants in Egbado-South in Ogun State, Nigeria.

From the survey, it was observed that these species were among the most popular components of recipes used for the treatment of dysentery, diarrhoea and typhoid fever. They are also components of traditional anti-tuberculosis recipes [5]. This communication describes the antimicrobial potential of the leaf and stem bark of *Bridelia micrantha*, *Alchornea cordifolia* and *Boerhavia diffusa* in view of the limited information on these locally abundant medicinal plants.

Experimental

Plant material

The plants were collected in Ibadan and authenticated at the Forestry Research Institute of Nigeria, Ibadan where herbarium specimens had been deposited. Forty grams of each oven-dried morphological part of each species was macerated in 200 ml of MeOH for 5 days. Each extract was filtered and the solvent evaporated under reduced pressure in a rotary evaporator and weighed. Dilutions of each dried extract was prepared in 70% MeOH to give final test concentrations of 100 mg/ml and 25 mg/ml (dried extract/ml).

Phytochemical screening for various natural products were performed as described by Harborne [6] and the result is shown in Table 1.

Antimicrobial screening

Microorganisms

Microorganisms used were: *Ps. aeruginosa* (NCTC 6750), *Staph. aureus* (NCTC 6571), *E. coli* (NCTC 9750), *Salmonella typhosa*, *Shigella dysenteriae*, *Klebsiella pneumoniae*, *B. subtilis*, *Proteus mirabilis*, *B. megaterium*, *Aspergillus niger*, *Microsporium species*, *Penicillium species* and *Candida albicans*. The microorganisms were obtained from the Department of Medical Microbiology and Parasitology, University College Hospital, Ibadan and the Department of Veterinary Microbiology, University of Ibadan.

Antimicrobial tests

The agar diffusion method [7] was used. About 0.1 ml of a 1 in 100 dilution of the overnight broth culture of each bacterium (about 10^6 - 10^7 cells/ml) was used to seed sterile molten nutrient agar maintained at 45 °C. The plates were allowed to solidify. Solutions containing 25 mg/ml, 50mg/ml of extract in 70% MeOH of each morphological part of each plant were added to appropriate wells' (7 mm diameter) made in seeded plates which were incubated at 37 °C for 24 hours. Gentamycin (10 ug/ml) ampicillin (10 ug/ml) were used as positive control for Gram + and Gram-bacteria, respectively MeOH. (70%) was used as a negative control.

Results and discussion

Alkaloids were detected in *A. cordifolia* and *B. diffusa* while anthraquinone glycosides were present in the leaf and stem bark of *A. cordifolia* (Table 1). Even though the flavonoid content [8] of *A. cordifolia* and triterpenoid constituents of *B. micrantha* [9] had been reported, there is no previous report on the anthraquinone and alkaloid content of *A. cordifolia* and *B. diffusa*.

Table 2 shows the antimicrobial potential of the plant species against the pathogenic organisms. The figures in the tables are the calculated zones of inhibition (determined as the difference between the diameter of observed zone of inhibition [in mm] and the diameter of the cork borer). The leaves and stem bark of *B. micrantha* and *A. cordifolia* exhibited significant inhibitory activity against the test organisms. In some cases, the antibacterial activity was comparable to those of ampicillin and gentamycin at the doses examined.

Table 1: Result of phytochemical screening of *Bridelia micrantha*, *Alchornea cordifolia* and *Boerhavia diffusa*

Natural	<i>Bridelia micrantha</i>		<i>Alchornea cordifolia</i>		<i>Boerhavia diffusa</i>
	leaf	stem bark	leaf	stem bark	leaf
Tannins	+	++	+	+	+
Saponins	+	++	+	+	+
Antraquinones	-	-	+	+	-
Cardenolides	-	-	-	-	-
Alkaloids	-	-	++	+	+

[+] = Positive; [++] = Highly positive; [-] = Negative

The extract of the leaf of *B. diffusa* exhibited minimal activity. Table 3 shows that only the leaf extract of *A. cordifolia* exhibited reasonable antifungal activity. Extract of *B. micrantha* and *B. diffusa* were practically inactive against the test fungi. The inhibitory effect of extracts of *A. cordifolia* on *Candida albicans* and *Microsporium* species is significant since these fungi are implicated in vaginal candidiasis and for aflatoxin production respectively.

Table 3: Antifungal activity of extracts of *bridelia micrantha*, *Alchornea cordifolia* and *Boerhavia diffusa*

Fungi	Dose mg/ml	<i>Bridelia Micrantha</i>		<i>Alchornea cordifolia</i>		<i>Boerhavia diffusa</i>
		leaf	stem bark	leaf	stem bark	leaf
<i>Candida albicans</i>	100	-	12	12	-	12
	50	-	-	9	-	9
	25	-	-	-	-	7
<i>Aspergillus niger</i>	100	-	-	10	14	-
	50	-	-	8	10	-
	25	-	-	-	-	-
<i>Penicillium spp.</i>	100	-	-	11	-	-
	50	-	-	-	-	-
	25	-	-	-	-	-
<i>Microsporium spp.</i>	100	10	-	11	10	-
	50	8	-	9	-	-
	25	-	-	7	-	-

Tioconazole (0.5 mg/ml); *C. albicans* (32mm); *A. niger* (15mm); *Penicillium spp.* (15mm); *Microsporium spp.* (19mm); = no activity. Zone of inhibition = Difference between the diameter of observed zone (mm) and the diameter of cork borer.

Table 2: Antibacterial activity of extracts of *Bridelia micrantha*, *Alchornea cordifolia* and *Boerhavia diffusa*

Micro-Organisms*	Dose mg/ml	<i>Bridelia Micrantha</i>		<i>Alchornea cordifolia</i>		<i>Boerhavia diffusa</i>
		leaf	stem bark	leaf	stem bark	leaf
PA	50	-	12	30	18	12
	25	-	12	25	15	-
SD	50	18	8	12	9	7
	25	15	-	9	-	-
ST	50	10	11	8	9	7
	25	8	9	-	7	-
EC	50	18	10	20	22	-
	25	16	9	18	18	-
KP	50	10	8	-	10	8
	25	9	-	-	8	-
PM	50	20	11	12	15	-
	25	18	10	10	13	-
SA	50	8	12	15	16	11
	25	7	8	11	14	9
BM	50	-	10	18	16	7
	25	-	8	16	14	-
BS	50	10	11	15	15	10
	25	9	9	12	12	-

* Gentamycin (10 ug/ml); Ampicillin (10 ug/ml); PA = *Ps. Aeruginosa* (0 mm); SD = *Shigella dysenteriae* (15 mm); ST = *Salmonella typhosa* (18 mm); EC = *E. Coli* (18 mm); KP = *Klebsiella penumoniae* (18 mm); PM = *Proteus megaterium* (10 mm); SA = *S. aureus* (12 mm); Bm = *Bacillus megaterium* (10 mm); BS = *B. subtilis* (9 mm). Zone of inhibition = difference between the diameter of observed zone (mm) and the diameter of cork borer.

In Egbado-South local government area of Ogun State, traditional healers use extracts of the stem bark of *B. micrantha* in combination with either extracts of *Parkia biglobosa* or *Croton zambesicus* for the treatment of typhoid fever and dysentery. This action is explained by the inhibition shown by the leaves and stem bark of *B. micrantha* against *Salmonella typhosa*, *Shigella dysenteriae* and *Escherichia coli*.

This study justifies the use of these medicinal plants in traditional medicine.

Acknowledgements

We are grateful to Mrs. V.O. Ogunleye of the Department of Medical Microbiology and Parasitology, University College Hospital, Ibadan and Mrs. O.O. Orioke, Department of Veterinary Microbiology and Parasitology, University of Ibadan for the supply of micro-organisms.

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