

Antimicrobial Activity of Aqueous and Ethanolic Extracts of *Bryophyllum pinnatum*

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Abstract: *Bryophyllum pinnatum* is a crassulescent herb with wide distribution and documented antimicrobial efficacies. Inconsistence in its activities has however, been suggestively linked with some factors. Hence, this study was carried out to establish the effect of some extrinsic factors on the antibacterial activities of *Bryophyllum pinnatum* against some disease causing microorganisms. Antibacterial activity of *Bryophyllum pinnatum* was determined by standard agar-diffusion method. Results from this study showed a significantly higher zone of microbial inhibition with ethanolic extract when compared with aqueous extracts ($P < 0.05$). Conclusion from this study have shown that solvents and concentration of extracts are some of the extrinsic factors that influences the antibacterial activities of *Bryophyllum pinnatum*

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1. Introduction

The increasing occurrence of antibiotic resistant organisms coupled with the non availability and high cost of new generation antibiotics in addition to their limited effective life span have resulted in increased morbidity and mortality (Wright, 2014). This observation have led to the search for alternative therapy with the aim of discovering potentially useful active ingredients that can serve as source and template for the synthesis of new antimicrobial drugs (Pretorius *et al.*, 2003, Moreillion *et al.*, 2005). The plant, *Bryophyllum pinnatum* (Crassulaceae) is commonly known as air plant, love plant, miracle leaf, life plant, Zakham-e-hyat, panfutti, Ghayamari (Jain *et al.*, 2010) and has been accepted as a herbal remedy in almost all parts of the world (Olajide *et al.*, 1998; Gupta *et al.*, 2010).

This plant is a crassulescent herb of about 1 metre in height, with opposite glabrous leaves (with 3–5 deeply crenulated, fleshy leaflets) (John-Ojewole *et al.*, 2002), distributed worldwide but growing primarily in the rain forest. It is grown widely and used as folk medicine in tropical Africa, India, China, Australia, tropical America, Madagascar, Asia and Hawaii (Yadav and Dixit, 2003). It is astringent, sour in taste, sweet in the post digestive effect and has hot potency. It is well known for its haemostatic and wound healing properties. The plant have considerable attention for their medicinal properties and find application in folklore medicine, as well as in the contemporary medicine (Kamboj and Saluja *et al.*, 2009). The antibacterial activity of the leaf juice of

B. pinnatum has been well reported. However, inconsistence in its activities has however, been suggestively linked with some factors. Hence, this study was carried out to establish the effect of some extrinsic factors on the antibacterial activities of *Bryophyllum pinnatum* against some disease causing microorganisms.

2. Materials and Methods

2.1 Source of plants

Bryophyllum pinnatum specimen was obtained from the Agronomy Unit of Dagwon farm (National Veterinary Research Institute, Vom). The plant was authenticated by the taxonomist at the Department of Botany and Biochemistry, University of Jos, Nigeria.

2.2 Preparation of plant materials and extracts

The plucked leaves were washed with running tap water and then thoroughly with distilled water several times. They were then disinfected and weighed using metler's balance. The leaves were further sliced longitudinally. One kilogram of the fresh leaves was air dried on the laboratory table at room temperature (27°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 to give a total weight of 600g. Hundred grams each of the powdered plant was soaked in 500 ml of ethanol and water for 6h using soxhlet apparatus.

2.3 Phytochemical studies

Phytochemical analysis was carried out to determine the presence of flavonoids, tannins,

alkaloids, saponins and anthraquinones using the methods described by Odebiyi and Sofowora (1978).

2.4 Test organisms

Escherichia coli, *Staphylococcus aureus*, *Salmonella typhi*, *Shigella spp*, *Proteus vulgaris*, *Pseudomonas aeruginosa*, *Klebsiella aerogenes*, *Trichophyton mentagraphytes*, *Candida albican* and *Cryptococcus neoformans* were obtained from Bacteriology and Dermatophilosis Unit, Division of NVRI, Vom. The isolates identities were further confirmed in our laboratory using standard procedures (Cheesborough, 2005). The isolates were maintained on Tryptone Soy agar (TSA) (Oxoid) and Sabouraud dextrose agar (oxoid) at 4°C for bacteria and fungi respectively.

2.5 Determination of antimicrobial activity

The medium used was Mueller Hinton agar (Oxoid, U.K). The microbial inoculums were adjusted to 0.5McFarland turbidimetric standard and inoculated onto the medium using sterile swabs. For each extract,

three replicate plates were prepared against each test organisms. Antimicrobial activity of the ethanolic and aqueous 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 while different concentrations of the extracts, 1000, 750, 250, and 100 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 control and was introduced into a separate well as appropriate. Ciprofloxacin /Fluconazole (250/150 µg/ml) was used as standard antimicrobial agent 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.

3.0 Results

Table 1. Effect of solvent of extraction on the antibacterial activity of *Bryophyllum pinnatum*

Solvents of extraction	n	Zones of inhibition
		Mean ± SEM(mm)
Ethanol	10	21.18 ± 1.20
Aqueous	10	14.00 ± 1.50

P < 0.05.

Table 2. Effect of concentrations on the antimicrobial activity of *Bryophyllum pinnatum*

Concentrations	n	Zones of inhibition
		Mean ± SEM(mm)
1000mg/ml	10	24.25 ± 1.06
750mg/ml	10	22.38 ± 1.83
250mg/ml	10	20.10 ± 1.60
100mg/ml	10	19.50 ± 1.35

P > 0.05.

The effect of solvent of extraction on the antimicrobial activities of *Bryophyllum pinnatum* was determined in table1. The zones of inhibition (mm) varied significantly with solvent of extraction (P<0.05). In table 2, when the mean of zones of inhibition of *Bryophyllum pinnatum* at different concentration were compared, an insignificant difference was observed (P>0.05).

4. Discussions

Result of this study have shown that the antimicrobial activities of *Bryophyllum pinnatum* were dependent on the solvent of extraction. This observation supported the alternate hypothesis which says that solvent of extraction influences the antimicrobial activities of *Bryophyllum pinnatum*. The significantly higher inhibitory activities of the ethanolic extract compare to the aqueous extract indicates higher extraction strength of bioactive

ingredients with ethanol (Thomas *et al.*,2012). Also, the fact that *Bryophyllum pinnatum* exhibited inhibitory activities almost at equal level at concentration range of 1000-100mg/ml is indication that there is an equilibrium of activities between the lesser and higher concentrations of *Bryophyllum pinnatum*. This clearly shows that the antibacterial quality of the active constituents of the plant is largely dependent on its molecular weight and diffusion rates through agar rather than its concentration. Since higher zones of inhibition in agar diffusion antibiotic susceptibility tests is an attribute of faster rates of drug diffusion and low molecular weight, it can then be inferred that the active antimicrobial constituent of *Bryophyllum pinnatum* might be among compounds with lower molecular weight. In conclusion, the outcome of this study has demonstrated that antimicrobial activities of concentrates from *Bryophyllum pinnatum* could be best enhanced by

methods that include air drying and ethanol extraction of the plant.

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