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The effect of *Leptadanea hastata* stem on some cultured microorganisms

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ABSTRACT

An ethanol extract of the stem of *Leptadanea hastata* (Pers) Decne was partitioned between chloroform and water. Saponins were detected in the aqueous fraction, while the chloroform fraction, which showed significantly inhibitory activity against *E. coli, Klebsiella pneumonae, Staphylococcus aureus, Pseudomonas sp. And Salmonella sp. b*acteria, was shown to contain alkaloids. The chloroform fraction was further partitioned into aqueous methanol and petroleum ether. The aqueous methanol portion showed activity against only *K. pneumonae* while the pet. ether portion showed actitivy against *E. coli, Pseudomonas sp. and salmonella sp*. These results justify the use of the plant in ethnomedicine.

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INTRODUCTION

Leptadanea hastata (Asclepiadaceae) is a common shrub in the northern part of Nigeria, it is called 'Yadiya' in Hausa. It is a climber with broad, well-developed, half-succulent leaves and thick greenish sap. The leaves and flowers are used as a food source [1], and have been shown to possess high levels of beta-carotene, amino acids, and low levels of hemagluttinin and saponins [2]. Elemental analysis indicate that the leaves are rich in iron, with calcium, potassium and magnesium also present in significant amounts [3].

A number of polyoxypregnane ester derivatives have previously been reported from *L. hastata*, [4,5]. While lupeol and its acetate and palmitate esters have been shown to display significant anti-inflammatory activity [6].

The stem latex of *L. hastata* is used traditionally in the treatment of dandruff, ringworm diseases (*Tinea capitis*), bacterial tract infections and bacterial conjunctivitis [7].

EXPERIMENTAL

Stems of *L. hastata* were collected from the University farm of Ahmadu Bello University, Zaria, Nigeria. A voucher specimen number 578 of the plant material was deposited in the herbarium of the Department of Biological Sciences.

Air dried, ground stems (200g) were extracted by percolation with 95% ethanol at room temperature. The percolate was concentrated at 40°C under reduced pressure to give residue (F_1). This residue was partitioned between $CHCl_3$-H_2O (400mL, 1:1) affording a $CHCl_3$ soluble fraction (F_2) and an aqueous fraction (F_3), which were concentrated separately.

Residue F_2 was further partitioned between 80% MeOH and petroleum ether (bp. 60^0-80^0C) (400mL, 1:1) to give petroleum ether (F_4) and aqueous methanol (F_5) soluble fractions, which were also concentrated separately.

Antimicrobial sensitivity tests were carried out using the five standard microorganisms *E. coli*, *Klebsiella pneumonae*, *Staphylococcus aureus*, *Pseudomonas* sp. and *Salmonella* sp.

Activity of the extracts were tested using the penicylinder cup technique, which relates the growth of the organism to the activity of the extracts. The prepared nutrient agar (Muller Hinton Agar (oxoid) was melted and cooled in water bath to between 45°C and 50°C. The medium was poured into a sterile petri dish and was allowed to solidify. The medium was inoculated with the test organism. A sterile penicylinder (usually made of porcelain or glass) was placed in the top layer of agar with an open end of the cylinder pointed up. After the medium has hardened, two drops of 100mg of the extract dissolved in 1ml dimethylsulphoxide (DMSO) were introduced with the help of dropping pipette into the embedded cylinder and the plate was incubated in an upright position at 37°C for 24 hours. Clear zones of inhibition adjacent to the cup were seen and measured by measuring the diameter in millimeter (mm), as in Table 1.

Phytochemical tests were carried out to establish the presence of alkaloids, tannins, saponins,

Table 1: Zone of inhibition (in mm) of four partitioned fractions of ethanol extract of *L. hastata* on Bacteria

	E.coli	*Klebsiella pneumonae*	*Staphylococcus aureus*	*Pseudomonas* sp.	*Salmonella* sp.
$CHCl_3$ Fraction (F2)	20	17	20	19	20
Aq. Fraction (F_3)	-	-	-	-	-
Petroleum ether Fraction (F_4)	11	-	-	12	14
MeOH Fraction (F_5)	-	14	-	-	-

- = no inhibition.

anthraquinones and carbohydrates using standard procedure [8].

RESULTS AND DISCUSSION

Four partitioned fractions (F_2-F_5) were obtained from the crude fraction (F_1) of the stem of *L. hastata.* Alkaloids were found to be present in the chloroform fraction (F_2), when the fraction was treated with Mayer's, Wagner's and Dragendorff's reagents and was confirmed when the tests were repeated after removing the non-alkaloidal compounds capable of given false-test results with either reagents, the procedure involves using 1ml of the acidic aqueous extract and treating it with 25% ammonia solution until the solution was alkaline to litmus paper and then extracted several times with chloroform. The chloroform extracts were combined and concentrated, the test for alkaloid was then conducted. A creamy white precipitate with Mayer's and a reddish- brown precipitate with Wagner's and Dragendorff's reagents were evidence for the presence of alkaloids. Saponin was detected in the aqueous fraction (F_3), this was observed when 0.1g of the extract was shaken with 5ml distilled water in a test tube for 1min, and kept aside. Persistent frothing for more than 15min was observed and this indicated the presence of saponins.

These preliminary phytochemical studies suggest that the alkaloids present in the $CHCl_3$ fraction (F_2) might be the one responsible for the considerable antimicrobial activity against all the five microbes displayed, and hence support the traditional ethnomedicinal usage of this plant. Effort is under way to elucidate the structure of the active component.

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