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Short Communication

Preliminary phytochemical and antibacterial studies on ethanolic extract of the leaves of *Acacia nilotica*

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ABSTRACT: The ethanolic extract of the leaves *Acacia nilotica* was evaluated for antibacterial activity using disc diffusion method and also to test the presence of the plants. Photochemical screening result revealed the presence of these five plant constituents, namely Resin, Tannins, steroid-Glycoside, Saponin and Alkaloids. Determination of antibacterial activity of the extract was conducted with the following concentrations (1000 µg/ml, 2000 µg/ml, 5000 µg/ml & 10,000 µg/ml) were all tested against *Salmonella typhi* aIid *Escherichia coli.* The result showed activity of the extract on *E - coli* at all concentration of (2000, 5000, & 10,000 µg/ml) respectively, except of 1000 µg/ml, showed resistant. However, for S. *typhi* the extract showed activity with all the concentrations, forming the following zone of inhibition ((7,9,14 & 5)mm) respectively. The inhibitory effect of the ethanolic leaves extract on the growth of test organisms may be attributed to the presence of the four active constituents screened. The results validate the use of the plant part in ethonomedicine.

Keyword: *Salmonella typhi, Escherichia coli, Acacia nilotica,* Phytochemistry, Antibacterial agents.

Introduction

Medicinal plants have become an effective source of both traditional and modern medicine and are genuinely useful for primary health care. Over the years the World Health Organization (WHO) has advocated traditional medicines as safe remedies for ailments of microbial origins (WHO, 1978). The use of plant extracts and phytochemical with non antibacterial properties may be of importance in therapeutic treatments where, in the past few years a number of studies have been conducted in different countries to prove such efficacy (Ikram & Inamu 1984, Almagboul *et al.*, 1995). Medicinal plants would be the best source for obtaining a variety of drugs where about 80% of the population in developing countries use traditional medicines derived from the medicinal plants. Therefore such plants should be investigated thoroughly to determine their structural and functional properties as well as the efficacy of various parts (Ellof,1998).

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Materials and Methods

Collection and Identification of Plant Material

The plant sample *(Acacia nilotica)* was collected from Ungogo Town, Ungogo Local Government, Kano State, Nigeria. The plant materials were located by traditional healers of· Ungogo Town. Taxonomic identification of the plant was confirmed at the herbarium of Biological Sciences Department, Bayero University, Kano.

Extraction Procedure

The World Health Organization (WHO, 1992) procedure of extraction was adopted for this study. Two hundred grams of the powdered *Acacia nilotica leaves* were soaked in 1000 ml of ethanol (solvent). The mixture was kept for two weeks in a tightly sealed conical flask at room temperature. It was then filtered using a Whatman number 1 filter paper.

Preparation of Sensitivity Disc

Four different concentration of (1000µg/ml, 2000µg/ml, 5000µg/ml and 10,000µg/ml) were prepared from the stock solutions of this leaves extract in four different sterile bijou bottles containing a punched Whatman filter paper of 6mm disc, and each bottle was labeled accordingly.

Test Culture

The microorganisms used were the pure culture of gastro intestinal tract bacteria isolate collected from Infectious Diseases Hospital (IDH) Kano, Kano State, Nigeria. These include *Salmonella typhi* & *Escherichia coli.*

Sensitivity disc

The prepared sterile agar media was carefully poured into a sterile petridishes and allow to solidified after which the plates were labeled using masking tape to indicate the test organisms and position of four disc, developed from four prepared concentrations, of 0.1ml disc potency of stock solution. Fresh culture of each isolate of bacteria inoculums was stretched on the surface of the solidified media and four different prepared discs were place at different position, (Amoxicillin 30g) was used as a standard antibiotic and control for the concentrations.

The plates were later incubated for 18-24 hours at 37°, after incubation the plates were observed for the presence of inhibition zones as an evidence of antimicrobial activity. The zones were formed to presence then measure and recorded in millimeters (mm).

Photochemical Screening of Plant Extract

The extract was analyzed for the presence of reducing sugar, Tannin, Resin, f1avonoside, spooning, Alkaloid & steroid glycosides.

Test for Reducing Sugar

0.1ml of ethanolic extract was taken into at test-tube, which was diluted with 2.0ml of diluted water followed by addition of Fehling's solution (A+B) and the mixture warmed, Brick red precipitates at the bottom of the test tube would indicate the presence of reducing sugars in accordance with Brain and Turner, (1975).

Test for Tannin

Two millilitres of the ethanolic extract was diluted with distilled water in a test tube, then 2 - 3 drops of 5% ferric chloride solution was added. The appearance of black or blue coloration would indicate the presence of Tannin as reported by Ciulei (1994).

Test for Steroid Glycosides

Two millilitres of ethanolic extract was taken into a test tube and evaporated to dryness. The residue was dissolved in acetic anhydride and chloroform was then added. By means of a pipette, concentrated sulphuric acid was added by the side of the test tube. A brownish ring at the inter face of the two Liquids and the appearance of violet color at the supernatant layer would indicted the presence of steroid as reported by Ciulei (1994).

Test for Resins

This method was reported by Evans (1996). 2.0g of the ethanotic extract was dissolve in 10ml of acetic anhydrate on drop of concentrated sulphuric acid was added. Appearance of purple colour dovish rapidly changes to violet would indicate the presence of Resins.

Test for Flavonsides

Two grams of the ethanolic extract was dissolved in 50% methanol by heating magnesium metal and 5 drops of concentrated hydrochloric acid were added. The appearance of green colour would indicate the presence of Flavonside as reported by Ciulei (1994).

Test of Alkaloid

1.0ml of the ethanolic extract in test tube, 2 drops of Dragendoff's reagent was added. An orange red precipitate/ turbidity would indicate the presence of alkaloid as reported by Ciulei (1994).

Test for Saponins

Half gram (0.5g of the powdered plant material was taken into a test tube, 5.0ml of distilled water was added and shaked vigorously. A persistent forth that last for at least 15 minutes would indicate the presence of saponin in accordance with the method Brain and Turner (1973).

Results

The results are presented in Tables 1 and 2.

Table 1:· Some Photochemicals identified in ethanolic leaves extract of *Acacia niotica*

S/N	Constituents
1	Resin
2	Tannin
3	Steroid Glycoside
4	Saponin
5	Alkaloid

The result of photochemical screening showed that the *Acacia nilotica* leaves extract was positive for five photochemical except f1avonoside and reducing sugar.

Table 2: Antibacterial activity of ethanolic leaves extract of *Acacia nilotica*

S/No.	Test Organism	Disc Potency (g)	Zone of inhibition (mm)			Control
		1000	2000	5000	10,000	Amoxicillin
1.	*E.Coli*	0.0	10.0	11.0	13.0	40.0
2.	S. *Typhi*	7.0	9.0	14.0	15.0	20.0

Key: *E. coli = Escherichia coli, S. typhi = Salmonella typhi*

The result of antibacterial effect showed the activity on *S. typhi* of all concentration but on *E coli* no activity at 1000mglml concentration.

Conclusion

The results confirmed *Acacia nilotica* to have antibacterial activity on *Salmonella typhi* and *Escherichia Coli* which may be due to the presence of plant *active* constituent such as Resin, Tannin, steroid glycoside, saponin and alkaloid.

Discussion

The present result of photochemical screening carried out on *Acacia nilotica* leaves ethanolic extract revealed the presence of *fives* photochemical out of *seven* expected ones. the report of may dell 1990, was not in accordance with present study in his work or the plant *Acacia niotica* only three photochemical were formed to be present that Resin, Saponin and tannin. Based on the antimicrobial effect of *Acacia nilotica* the present study has shown the activity of ethanolic *leaves* extract against *Salmonella typhi* and *Escherichia coli*, while more reported by (Satony *et al* 1995), on the antimicrobial activity of *Acacia niotica revered* The activity only on *Clostridium perfringes*, but not on *Salmonella typhi* and *Escherichia Coli.* This may be due to the fact that preparing an extract with organic solvent (e.g. Ethanol) have been shown to provide a better antibacterial activity than using water solvent (Nair et al. 2005).

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