SHORT COMMUNICATION REPORT

ANTIMICROBIAL ACTIVITY OF LEAF EXTRACTS OF Pavetta crassipes (Hutch) AGAINST SOME RESPIRATORY TRACT PATHOGENS.

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The search for antimicrobial agents is an important aspect of human development and will remain relevant as long as there are diseases and the need for new and better drugs. The potentials for finding remedies for various illnesses from plant sources or natural products are only limited by our imagination and capacity to screen. Soforawa (1993) reported that less than 10% of the flora of the world have been screened for their medicinal value. Little wonder that the traditional medicine system which is largely based on plant resources has formed part of primary healthcare (Sofowora 1993).

This study focused on screening the leaves of *Pavetta crassipes* for activity against some respiratory tract pathogens. *P. crassipes* is a local plant that is widely used in Nigerian Hausa ethno-medicine in the treatment of respiratory tract infections,

Collection and identification of plant: Plant samples were collected from Toro local government area of Bauchi State, Nigeria between July and August 2006 and identified by the traditional healers of Yalleman(Kaugama Local Government Area, LGA Jigawa state), Sabuwar Kasuwa, Gama, Brigade quarters(Nassarawa LGA, Kano State, Nigeria. Taxonomic identity of the plants was also confirmed at the herbarium of the Department of Biological Sciences, Bayero University Kano, Nigeria

Extraction and fractionation: The methods of Fatope *et al*, (1993) and Adoum *et al*. (1997) were adopted for extraction and fractionation. 30 g of the powdered, air-dried plant material was percolated in 300 ml of 100 % ethanol for two weeks, after which the extract was filtered using a whatman No 1 filter paper. The crude extract was concentrated using a Rotary evaporator (R110) at 40 °C. The extract was labeled, weighed and kept under refrigeration in the laboratory until required for further analysis.

A fraction of the extract was partitioned between water and chloroform (H_20/CH_3Cl_3) mixture (1:1) (100ml:100 ml), shaken vigorously for about 1 hr and allowed to settle for 24 hr in a separating funnel. The chloroform and water fractions were separated in beakers and labeled separately. The fractions were again weighed and kept in the laboratory for further analysis.

Preparation of sensitivity Discs: A whatman filter paper was punched to obtain 6 mm dics which were placed in sterile bijour

bottles and sterilized in an oven at 160 $^{\circ}\text{C}$ for 15 min and allowed to cool.

Four different concentrations of the different solvent extracts from each of the plants investigated were prepared for the sensitivity testing: 1,000µg/ml, 2,000µg/ml, 5,000µg/ml and 8,000µg/ml respectively and placed in sterile bijour bottles. Subsequently 100 sterile discs each were aseptically placed in labelled bottles and 0.9ml, 0.8ml, 0.5ml and 0.2ml solution of the various plant extracts in DMSO (Dimethyl sulphoxide) were taken out using syringes and transferred into appropriate bottles containing the filter paper discs.

The stock solution of (10,000 µg/ml) concentration was prepared by suspending 1mg of plant extracts in 1ml of solvent (DMSO). The 4 different concentrations of each of the plant extract were prepared in the following manner: for 1,000 µg/ml, 0.1ml of stock solution was added to 0.9ml of DMSO, 0.2ml of stock solution was added to 0.8ml of DMSO to obtain 2,000ug/ml while for 5,000µg/ml, 0.5 ml of stock solution was added to 0.5ml of DMSO. Finally, 0.8ml of stock solution was added to 0.2ml of DMSO to obtain 8,000ug/ml concentration.

Test Culture: 4 pure cultures of respiratory tract bacterial isolates, namely *S. aureus, K. pneumoniae, Proteus specie* and *P. aeruginosa* were collected from Aminu Kano Teaching Hospital (A.K.T.H) and Murtala Muhammad Specialist Hospital (M.M.S.H) and used for the test.

Preparation of culture Media and Bioassay: Nutrient agar was used to culture the test organisms and the media were pepared according to manufacturer's instructions. For the bioassay, the disc-plate method was used as described by Kir-Bauer (1966) and modified by Khan & Saeed (2000). The sterile agar media were carefully transferred into sterile Petri dishes and allowed to solidify after which the plates were placed in the drier to remove excess moisture. The plates were labeled using masking tape to indicate the test organisms and the positions of the four discs of different concentrations of the plant fraction. From a 24 hr culture of each isolate a loopful of bacterial inoculum was taken and streaked on the surface of the dried medium. Thereafter four discs of different concentrations of the same extracts were aseptically placed at the marked positions while standard antibiotic discs comprising ciprofloxacin and streptomycin were placed at the center for comparison.

The plates were later incubated for 18-24 hr at 37°C. After incubation, the plates were observed for presence of clear zones of growth inhibition as evidence of antimicrobial activity. The zones, where present, were measured and recorded in millimetres (mm) using a meter rule.

The sensitivity pattern of the test organisms to chloroform and ethanol fractions of the plant extract is shown in Tables 1 and 2. The ethanol fraction exhibited increased activity with increase in concentration against *S. aureus* as indicated by the diameters of growth inhibition zones. On the other hand the chloroform fraction had activity only at 5,000 and 8,000ug/ml concentrations. According to Vlicktnick *et al.* (1995),

antibacterial activity is recorded when the zone of inhibition is greater or equal to 6mm. It follows that the ethanol fraction is active against the test organism even at the lowest concentration of 1,000ug/ml which suggests that it contains more of the plant constituents that are active against the test organism. The chloroform fraction showed no activity against *K. pneumoniae* while the ethanol fraction was only active at the lowest concentration with a growth inhibition of 8mm. A similar pattern was observed for *P. aeruginosa* even though increased concentration of the ethanol fraction had no effect on the size of the growth inhibition zone which was 8mm throughout as shown in Olowosolu & Ibrahim (2006) reported a similar sensitivity pattern for *P. aeruginosa* when it was tested against extracts of the plant *Raphia nitida* Lodd where they recorded greater sensitivity to ethanol fraction compared with that of chloroform.

Antimicrobial activity was recorded for all concentrations of the chloroform extract when tested against *Proteus* sp. While no activity was observed for the ethanol fractions except for mild activity at 10,000ug/ml. The fact that both ethanol and chloroform fractions of the plant extract showed varying degrees of activity against the tested organisms is a good indication that the plant holds a great potential for the isolation of antimicrobial compounds which could be used in the development of drugs against respiratory tract pathogens.

The study has shown that *P. crassipes* represents a good candidate for the extraction of active antibacterial compounds with potency against respiratory tract pathogens.

TABLE 1. ANTIMICROBIAL ACTIVITY OF ETHANOL FRACTION OF P. CRASSIPES ON THE TEST ORGANISMS

Test Organisms	Concentration of extracts (µg/ml)						
	1000	2000	5000	8000	STD (CPX)		
S. aureus	08	08	10	13	21		
K. pneumoniae	80	00	00	00	48		
Proteus spp.	07	00	00	00	26		
P. aeruginosa	80	80	80	80	22		

TABLE 2. ANTIMICROBIAL ACTIVITY OF CHLOROFROM FRACTION OF *P. CRASSIPES* ON THE TEST ORGANISMS

	Concentration of extracts (µg/ml)						
Test Organisms	1000	2000	5000	8000	STD (S)		
S. aureus	00	00	07	07	29		
K. pneumoniae	00	00	00	00	23		
Proteus spp.	07	10	09	19	38		
P. aeruginosa	00	00	00	00	27		

STD = standard antibiotic

Cpx =Ciprofloxacin; S=Streptomycin.

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