Vol. 3 no. 2, pp 103-106 (September 2006)
http://www.ajol.info/journals/jpb

Journal of
PHARMACY AND
BIORESOURCES

Phytochemical screening and antibacterial activity of *Cissampelos mucronata* a. Rich (Menispermaceae) extracts

Terrumun A. Tor-Anyiin[1*], Rufus Sha'ato[1] and Hyacinth O.A. Oluma[2]

[1]Department of Chemistry; [2]Department of Biological Sciences, University of Agriculture, P.M.B. 2373, Makurdi. Benue State. Nigeria.

Received 19th April 2006; Accepted 11th August 2006

Abstract

Phytochemical screening of various extracts of *Cissampelos mucronata*, obtained using soxhlet apparatus showed presence of alkaloids, tannins and saponins. These crude extracts demonstrated moderate inhibition activity using Nutrient agar Swab sticks method against four species of bacteria: *Streptococcus pyogenes* and *Staphylococcus aureus* (gram positive), and *Salmonella typhi* and *Escherichia coli* (gram negative). Water and methanol – dichloromethane (1:1) extracts exhibited highest activity. These results lend credence to the use of this plant in traditional medicine in treating bacteria related diseases.

Keywords: Garlic; *Cissampelos mucronata;* Antibacterial activity; Phytochemical screening; Alkaloids.

Introduction

The dependence of man on plants as source of medicine, food, shelter, clothing, etcetera, has been since creation (Gamaniel, 2000; Cordell, 2000). Plants have also been known to produce an amazing array of novel phytochemicals, many of which are biologically active. Thus, many communities especially in poor countries of the south still depend on plants for cure of many external and internal infections and disease of both man and livestock. Indeed interest in plant-derived medicines as a re-emerging health care aid has assumed global proportions owing to rising costs of prescription drugs, as well as a renewed interest in folk traditions of the ancients. Advances made in bioprospecting for cheaper plant-derived (or traditional herbal) remedies have continued to encourage research in this area, worldwide (Hoareau and Dasilva, 1999).

Cissampelos mucronata A. Rich (Menispermaceae) is a perennial herbaceous climbing shrub common in dry zones of tropical Africa. Leaves alternate, serrate at mid and mucronate; petiole 2 – 4 cm long. Female flowers occur in clusters with a small foliaceus bract at the base. Male flowers occur in racemes. Fruit are small rounded berries, yellow at maturity (Adjanahoun *et al*, 1991; Hutchinson and Dalziel, 1954). The Yorubas call it *"jenjokoo"*, *"jokoo-jee"* Tiv: *"igbe-alom"*.

This plant is important trado-medically. For instance, it is used in treatment of ulcer and stomach-ache (Akah *et al.*, 1998),

* Corresponding author. *E-mail address*: toranyiint@yahoo.com Tel: +234 (0) 805 2821118
ISSN 0189-8442

abdominal pain and irregular menstrual cycle (Adjanahoun *et al.*, 1991), and malaria fever (Tor-Anyiin *et al.*, 2003, Gessler *et al.*, 1994, 1995).

Despite the seeming importance of this plant, there is paucity of literature on it. There is the thorny issue of credibility of herbal medicines arising from discrepant therapeutic claims, inconsistent and undocumented product quality, insufficient efficacy validation and safety verification. This study was therefore undertaken to provide answers to some of these questions and also as a first step in the isolation of biologically active component(s) of this plant.

Experimental

Plant collection, identification and preparation. Aerial parts of *C. mucronata* were collected at April, near Makurdi (Benue State) in January. It was identified by Mr. P.O. Ekwuno of Forestry and Wildlife Department, University of Agriculture, Makurdi where voucher specimen has been deposited. The fresh sample was room air-dried, ground with mortar and pestle into fine powder.

Extraction. The pulverized plant material, 45g was gradiently extracted using soxhlet apparatus with 750 ml, 650 ml. 500 ml, 500 ml and 700 ml each of petroleum ether (60^0 – 80^0C), $CHCl_3$, dichloromethane – methanol (1:1), methanol and water, respectively. The organic extracts were distilled to dryness on a thermostated ($50\ ^0$C water bath; water extract was concentrated on a sand bath maintained at $60\ ^0$ C . These crude extracts were labeled as PE, CE, DM, ME and WE, for petrol, $CHCL_3$, DCM –MeOH, MeOH and water, respectively.

Phytochemical screening. Small portions of each crude extract were subjected to phytochemical analysis using standard procedures (Farnsworth, 1966; Harborne, 1984).

Antibacterial assay. Freshly prepared Nutrient agar medium plates were smeared using Swab sticks. The Swab sticks were dipped into four bottles of peptone water inoculated with different species of bacteria: *Streptococcus pyogenes*, *Escherichia coli*, *Staphylococcus aureus* and *salmonella typhi*. Paper discs impregnated with each of the crude extracts were placed aseptically and pressed firmly on the surface of the inoculated agar plates. All plates were incubated aerobically at 34^0C for 24 hr. Plates were examined for evidence of growth inhibition which appeared as a clear zone around the disc completely devoid of bacterial growth. The observed diameters of such zones of inhibition were measured in mm using a transparent meter rule. The respective solvents for extraction were used as control (n = 4 for each extract type).

Results and Discussion

The characteristic colour, nature and yield (as percentage of plant material) of each extract is shown in Table 1. Preliminary qualitative phytochemical analysis of these extracts is contained in Table 2. Antibacterial activity (zones of inhibition, mm) of the various extracts of *C. mucronata* are reported in Table 3. Result show mean ± S.E.M (n = 4)

Table 1: Characteristics, nature and yields of extracts.

Solvent	Extract code	Nature/colour	Yield (g)	% yield
Petroleum ether (60° – 80°C)	PE	Greenish oily liquid	1.3	2.8
Chloroform	CE	Dark green paste	0.9	2.0
Dichloromethane-methanol (1.1)	DM	Green solid	0.7	1.5
Methanol	ME	Light green paste	0.7	1.5
Water	WE	Brown paste	1.0	2.2

Table 2: Phytochemical composition of extracts of *C mucronata*

Phytochemical composition	Reagent(s)/test(s) used	Extracts				
		PE	CE	DM	ME	WE
Alkaloid	Dragendorff's	+	+	+	+	+
	Wagner's	+	+	+	+	+
Tannins	$FeCl_3$	-	+	+	-	-
	Bromine water	-	-	+	-	-
Saponins	Frothing	-	+	-	-	-
Flavonoids	Mg chips + H_2SO_4	-	-	-	-	-
Anthraquinones	Borntrager's	-	-	-	-	-
Cardiac glycosides	Kedde's	-	-	-	-	-
	Keller – Kiliani	-	-	-	-	-
Sterols/triterpenes	Salkowski's	-	-	-	-	-
	Liebermann–Burchard	-	-	-	-	-

+ = Presence of component (s) - = absence of component(s)

Table 3: Antibacterial activity (zones of inhibition) of extracts of *C. mucronata*

Bacteria	Inhibition zone diameter (mm) Mean ± SEM (n=4)				
	PE	CE	DM	ME	WE
Streptococcus pyogenes	13 ± 1	13 ± 1	15 ± 1	15 ± 1	17 ± 1
Escherichia coli	9 ± 1	11 ± 1	11 ± 1	12 ± 1	14 ± 1
Staphylococcus aureus	12 ± 1	0 ± 0	13 ± 1	13 ± 1	13 ± 1
Salmonella typhi	10 ± 1	9 ± 0	17 ± 0	9 ± 1	10 ± 2

Discussion

All the crude extracts gave positive test (red precipitate) for alkaloids using Dragendorff's reagent, confirming the sensitivity of this reagent as a general test for alkaloids (Farnsworth, 1966). They also gave brown precipitates with Wagner's reagent. The presence of alkaloid(s) in *C. mucronata* has demonstrated that *Cissampelos* species are generally producers of alkaloid as major secondary metabolites. The presence of saponins (in CE) and tannins (in CE and DM) Table 2 in this plant in particular and species in general, is reported here.

These crude extracts showed moderate antibacterial activity on both gram positive and gram negative bacteria tested (Table 3). These pathogens are responsible for a number of ailments in animals. For instance, gram positive bacteria cause diseases such as boil, sore throat, upper respiration disease, abscesses, conjunctivitis, septicaemia, osteomyelitis pneumonia, food – poisoning, secondary infections of ulcers, burns, wounds and skin disorders. Gram negative bacteria are responsible for typhoid fever, urinary infections, diarrhoeal diseases, meningitis, cystitis, pyelonephritis, pyelitis, etcetera. Chloroform extract, CE, failed to inhibit growth of *S. aureus,* completely.

All four pathogens were more sensitive to DM (containing alkaloids and tannins) and WE (containing only alkaloids). Alkaloids and/or tannins have been shown to exhibit some antimicrobial activities (Nuhu *et al.*, 2000; Ibrahim *et al.*, 2005; Ogukwe *et al.*, 2004). Thus the higher antibacterial activity of dichloromethane-methanol (1:1) – DM-extract could be attributed to the presence of these secondary metabolites. It was noted that these extract were in general more sensitive to gram positive pathogens.

The sensitivity of all extracts of *C. mucronata* to tested organisms has justified the ethnomedicinal usage of this plant in treating bacteria related infections such as ulcer and stomach-ache (Adjanahoun *et al.*, 1991 and abdominal pain (Hutchinson and Dalziel, 1954). Work on isolation and identification of active component(s) is on going. There is however need in conducting toxicity studies on this plant extracts (and

other plants as well) to ascertaining their safety in human systems.

References

Adjanahoun E., Ahyi M.R.A., Akeassi L., Dramane K., Elewude J.A., Fodoju S.O., Gbile Z.O., Goudote E; Johnson C.L.A; Keita A., Morakinyo O., Ojewole J.A.O., Olatunji A.O. and Sofowora, E.A. (1991) Traditional Medicine and Pharmacopoeia: Contribution to Ethnobotanical and Floristic studies in Western Nigeria. Organization of African Unity Scientific, Technical and Research Commission, OAU/STRC Publication, Lagos, Nigeria. Pp. 180-85.

Akah P.A., Orisakwe L.E., Nwafor S.V. and Gamaniel K.S. (1998). Prospects of Natural plant product as antiulcer agents. *J. Pharm. Res. & Devpt.*, 3(2):57-62.

Cordell G.A. (2000). Biodiversity and drug discovery–A symbiotic relationship. *Phytochemistry* 55: 463-480.

Farnsworth N.R. (1966). Biological and phytochemical screening of plants. *J. Pharm. Sci.*, 55(3): 225-276.

Gamaniel K.S. (2000). Toxicity from medicinal plants and their products. *Nig. J. Nat. Prod. & Med.*, 4:4-7.

Gessler M.C., Nkunya M.H.H., Nwasumbi L.B, Heinrich M. and Tanner, M. (1994). Screening Tanzanian Medicinal Plants for Antimalarial Activity. *Acta Tropica* 56(1): 65–77.

Gessler M.C., Tanner M; Chollet J; Nkunya M.H.H. and Heinrich M. (1995). Tanzanian medicinal plants used Traditionally for the treatment of Malaria-*in vivo* antimalarial and *in vitro* cytotoxic activities. *Phytotherapy Res.* 9(7), 504-508.

Harborne J.B. (1984). Phytochemical Methods. A guide to modern Techniques of plant Analysis. 2nd ed Chapman and Hall, New York. 288pp.

Hoareau L. and Dasilva E.J. (1999). Medicinal plants: A re-emerging health aid. *Electronic J. Biotechnology* 2(2): 1–7.

Hutchinson J. and Dalziel J.M. (1954). Flora of West Tropical Africa. Vol. 1 Part 1. 2nd ed. (Reviewed by R.J.W. Keay). Crown Agents for Oversea Government and Administration, London. 544pp.

Ibrahim H., Amupitan J.O. and Coombes P.H. (2005). The effect of *Leptedanea hastata* stem on some cultured microorganisms. ChemClass Journal, 2:45-46.

Nuhu A.M., Mshelia M.S. and Yakubu Y. (2000) Antimicrobial screening of the bark extract of *Pterocarpus erinaceus* tree, *J. Chem. Soc. Nigeria*, 25: 85-87

Ogukwe C.E., Oguzie E.E; Unaegbu C. and Okolue B.N. (2004); Phytochemical screening on the leaves of *Sansevieria trifasciata, J. Chem. Soc. Nig.*, 29(1): 8-10.

Tor-Anyiin T.A., Sha'Ato R. and Oluma H.O.A. (2003). Ethnobotanical survey of antimalarial medicinal plants amongst the Tiv people of Nigeria. *J. Herbs Spices & Medicinal Plants*, 10(3):61-74.