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PHYTOCHEMICAL ANALYSIS AND ANTIFUNGAL ACTIVITIES OF Gynandropsis gynandra (Spider flower) and Buchholzia coriacea (Musk tree) (FAM: CAPPARIDACEAE) ON SOME COMMON FUNGAL ISOLATES

[1]Ogunmefun O.T. and [2]Ajaiyeoba E.O.
[1]Department of Biological Sciences, Afe Babalola University, Ado-Ekiti
[2]Department of Pharmacognosy, University of Ibadan, Ibadan, Nigeria
E-mail: yinkatayo_08@yahoo.com

ABSTRACT

This paper describes the phytochemical and antifungal studies of two members of the Capparidaceae family which are *Gynandropsis gynandra* and *Buchholzia coriacea*. These two plants were screened for the presence of their active constituents. The leaves and the stem were screened separately. The antifungal activities of the leaves and stem were tested using *Aspergillus niger*, *Penicillum sp.*, *Candida albicans*, *Fusarium oxysporium* and *Aspergillus flavus*. The activity of the extract at 200mg/100ml (0.02g/10ml) of methanol was compared with methanol as the control and Tioconazole as reference standard. The result of the antifungal assay of the plant extracts justifies their use in traditional medicine.

Keywords: Gynandropsis gynandra, Buchholzia coriacea Phytochemical analysis, Antifungal activities, Isolates

INTRODUCTION

The family Capparaceae (Capparidaceae) is described from Cape Verde Islands. It comprises of 45 genera and approximately 1000 species, distributed in the tropical and sub-tropical regions, especially East Africa and South America. Plants in this family are annual, biennial and perennial herbs or shrubs, sometimes climbers or trees. Among the members of this family are *Cleome aculeate*, *C. scaposa*, *C. gynandra*, *C. rutidosperma*, *C. brachycarpa* and *C.* viscosa. *C. viscosa* is used as pasture for goats. The first two spices are ruderals (Duarte MC 1995). The plant *C. viscosa* is used to treat various ailments in the Unani system of medicine and its seeds are reported to have nutritive value. The seeds contain 18.3% oil, a mixture of 5 fatty acids, 7 amino acids and sucrose. Acute toxicity (LD50) was determine in mice by the oral route, and sub- acute toxicity was evaluated in a 14 day, repeated – dose study. The results indicated that the seeds are enough to be considered for edible purposes {Naheed – Parveen et al., (1994)}.

The family Capparidaceae are mainly herbs, shrubs or trees, sometimes scandent; leaves alternate, stipulate when present minute or spiny. Flowers mostly hermaphrodite, actinomorphic or rarely zygomorphic, hypogynous, axillary or terminal, variously arranged; perianth of calyx and corolla, or the latter absent. Sepals free or partially united; imbricate or valvate, usually 4. Petals 4 to many or absent. Torus elongated or short rarely with an appendix, Stamens few too many, sometimes, some of them without anthers, filaments sometimes partially adnate to the torus; anthers 2-celled, longitudinally dehiscent. Ovary sessile or more usually supported on a long or short gynophore, 1–celled with parietal placentas or divided into 2 or more cells by spurious dissepiments, ovules few to many. Fruit a capsule or berry sometimes the latter elongate or torulose. Seeds usually reniform or angular, endosperm none or scanty; embryo arcuate or incurved. Tropics and subtropics of both hemispheres, very numerous in East Africa and South America Recognized by the usually showy flowers, dawed petals, mostly stipulate ovary, parietal placentas and curved embryo with scanty or no endosperm.

Gynandropsis gynandra: Botanical Name

Spider flower, bastard, cat's whiskers – Common (English) name

Description

A herb of about 60cm high, a common plant of waste places and weed of cultivated land, and often itself cultivated, occurring throughout all territories of the Region, and pan-tropics generally. The plant is eaten in all parts of the region and in other country of its worldwide distribution, often as a pot-herb, taken cooked, fresh or dried (Irvine, 1952 ; 1956) or as spinach and in soups (Ainslie,1937; Adjanohun and Ake Assi, 1972). In Tanganyika it is recorded that the leaves require soaking in several changes of water to remove the bitterness (Michelmore 1402), an action which cooking also effects (Burkill, 1935; Watt & Breyer- BrandWijk, 1962). The leaves are said to provide a piquant taste or a sourness, perhaps due to conditions under which the plants has grown. They probably contain sinapin, the substance which gives mustard its biting taste (Irvine, 1956). They are considered very good for the stomach and to have antiscorbic properties (Anslie, 1937). Their vitamin C content has been recorded at 6.0mg per 100gm dry weight (Watt & Breyer- Brandwijk, 1962). They are rich in minerals and content of aluminium (1,390ppm) and of iron (470ppm) is unusually high (Busson, 1965). Though the plant is everywhere considered edible it is curious that it has been used as a fish poison (Watt & Breyer – Brandwikj, 1966).

The leaves have medical uses everywhere the plant occurs, most commonly as counter – irritant for local pain, the leaves being merely rubbed on the part affected or applied as a poultice (Dalziel, 1937; Kerharo & Adam, 1974). The leaves are considered disinfectant and a good remedy for rheumatism in Nigeria (Oliver, 1960), and pounded are applied externally for rheumatism, lumbago etc, but if left too long blisters are produced – indeed, this preparation is used in Nigeria as a vesicant (Ainslie, 1937). A leaf- mash is warmed and laid on swollen armpit bubos on the Ivory Coast (Adjanohun and Ake Assi, 1972) and over the kidneys and poulticing is known and practiced in SE Asia (Burkill, 1935). The leaves are rubbed on hands and inhaled like smelling salts for headache in West Africa (Dalziel, 1937). Leaf-sap is given in nasal instillation in the Ivory Coast (Bouquet and Debray, 1974). Widespread use is made of the leaves for ear trouble. The leaves are rubbed on hands and inhaled like smelling salts for headache in West Africa (Dalziel, 1937).

Leaf – Sap is used in minute quantities in Nigeria as an Eye wash (Oliver, 1960) and in Tanganyika for inflamed eyes (Haerdi, 1964). The seed, and the plant as a whole, contains a volatile oil rich in senevol with properties resembling sulphur derivative found in garlic and mustard oils (Kerharo & Adam, 1974). An acid fixed oil and a brown resin are also present (Watt & Breyer-Brandwijk, 1962). The concentration of the oil is about 17.6% (Kerharo Adam, 1974) and it is regarded as edible and suitable for soup-making. A thick greenish drying oil has been reported in the seeds (Walt & Breyer-Brandwijk, 1962). The seeds have anthelmintic property (Walker, 1953; Walker & Sillans, 1961) and their oil is furthermore used as a fish poison. In Tanganyika, they are used to facilitate childbirth and for internal disorders (Bally, 1937) in undisclosed ways.

Botanical Name: Buchholzia coriacea

English / Common Name: Musk tree

Description

An evergreen under-storey tree of the lowland rainforest, to 20m high, occurs from Guinea to West Cameroons, and in E. Cameroun and Gabon. The tree is often planted around Gabon villages (Walker, 1953). The wood is yellowish white, soft and somewhat fibrous (Aubreville, 1959; Keay et al., 1960, Irvine, 1961). It is probably suitable for house-building. The closely related *B. macrophylla* Pax is used in construction work in Gabon (Walker & Sillans, 1961).

The bark- slash is deep red, and the sap exudes with a violently spicey, poungent smell that causes sneezing (Aubreville, 1959). The bark is made in the Ivory

Coast into a pulp for inhalation (Adjanohun and Ake Assi, 1972) or into a snuff to relieve headache, sinusitis and nasal congestion in headcolds, also otitis and ophthalamus (Kerharo & Bouquet, 1950; Bouquet and Debray, 1974). A bark decoction is applied externally as a general reconstituent (Adjanohun and Ake Assi, 1972) or as a revulsive for pains in the chest, bronchitis, pleurisy and kidney pains (Kerharo & Bouquet, 1950;Bouquet and Debray, 1974. The Gagon of Ivory Coast administer bark-sap as an enema for kidney pains (Kerharo & Bouquet, 1950). The fresh bark is used for earache in Ghana (Vigue fide Irvine, 1961). A bark- decoction is used to wash persons with small-pox (Bouquet and Debray, 1974) and in Gabon the crushed bark is used in frictions on skin-itch (Walker, 1953).

The Guere of Ivory Coast incorporate the bark in arrow poisons and it is highly effective (Kerharo & Bouquet, 1950). In Liberia, the seeds are used on skin-eruption and internally for worms and pains (Dalziel, 1937), while when crushed up they are pasted over the stomach in Ivory Coast for difficult child birth (Adjanohun and Ake Assi, 1972). In Gabon, the fruit is considered anthelmintic (Walker, 1953). The fruit has a disagreeable smell on cutting open. The seeds have a hot spicy flavour. In South Nigeria, the Edo boil and eat the fruit after storage for few days (Irvine, 1961). In Gabon, it is considered poisonous (Bouquet and Debray, 1974). A number of chemical substances have been isolated from the plant and the substances determined (Bouquet and Debray, 1974).

MATERIALS AND METHODS

The plant materials used in this research work were collected from Edo state in Nigeria. *Buchholzia coriacea* was collected from a forest in Edo state through the help of Forestry Research Institute of Nigeria (FRIN) herbarium workers and was authenticated at the Botany Department Herbarium, of the University of Ibadan. The plant belongs to the family Capparidaceae. The local name in Yoruba is Uworo. The fungi isolates used in this study were supplied by the Department of Pharmaceutical Microbiology and Clinical Pharmacy, University of Ibadan, Nigeria. The fungi collected were *Aspergillus niger, Penicillium sp., Candida albicans, Fusarium oxysporium and Aspergillus flavus.*

EXTRACTION AND SOLVENT SEPARATION

Different quantities ranging from 384.5g to 747.5g of the powered samples were extracted with hexane using cold maceration for 72hours which was then filtered and the filtrate concentrated. The residues were air dried and

successively extracted with methanol for 72 hours. The solvent was removed by distillation and the residues properly dried.

The weight of *Gynandropsis gynandra* powdered leaves that were originally extracted with hexane was 404.5g. That of the stem that was extracted with hexane was 499.1g. The weight of *Gynandropsis gynandra* powdered leaves that was extracted successively with methanol was 384.5g showing a loss in weight by 20g from the original weight extracted with hexane. Weight of Buchholzia coriacea powdered stem extracted with hexane was 420.0g. That of stem extracted with methanol was 394.7g showing a loss in weight of 25.3g from the original weight extracted with hexane. Weight of *Gynandropsis gynandra* powdered stem extracted with methanol was 443.9g showing a loss in weight of 55.2g from the original weight that was initially extracted with hexane.

ANTIFUNGAL ASSAY

16.25g of Sabouraud dextrose agar was suspended in minimum volume of water and made up to 250ml. This was boiled to dissolve completely the Sabouraud dextrose agar. It was then dispensed in 15 volumes into universal bottles and autoclaved at121^{0}C for15 minutes. Fungi were introduced into 10ml saline; 0.5ml of the saline suspension was spread over set Sabouraud petri-dishes and incubated at room temperature for 7 days. The same procedure was employed for all the fungi except *Candida albicans* that was subcultured on slopes. For the assay, melted Sabouraud dextrose agar were poured into labeled petri-dishes and allowed to set, dried in the incubator for 20 minutes. A suspension of the organisms were prepared by taking very small amount with inoculating loop from the plates and slopes and inoculated in peptone soya broth. This was incubated at room temperature for 4 days, 0.5ml was introduced into 30ml saline to make 1 in 60 dilution. 0.3ml of each dilution was spread over the surface of the petri-dishes .The petri-dishes were left to stand for 3 hours to allow the suspension of the organisms to diffuse into the medium. By means of sterile cork borer (10mm) wells were made into the plates, after a firm drying.

Tioconazole (Trosyd) 0.01g/ml (1%dilution) was used as the control. The petri-dishes were left at room temperature for 2 hours to allow the extract diffuse into the medium after which it was incubated at room temperature for 48 hours – 4 days to 7 days (Cowan, 1974 and Reeves et al., 1978).

RESULT

Phytochemical Analysis

Table I:

	Gynandropsis gynandra		*Buchholzia coriacea*	
	Leaves	**Stem**	**Leaves**	**Stem**
Alkaloid	+++	+++	+++	+++
Anthraquinone	-	-	±	±
Saponins	-	-	-	-
Tannins	-	-	-	-
Cardiac glycoside	-	-	-	-
Cyanogenetic glycoside	+++	+++	+++	+++
Steroidal nucleus	+++	++	+++	++
Reducing sugars	+++	+++	+++	+++

+ = Slightly present, ++ = Fairly present, +++ = Abundant, - = Absent

Antifungal Studies

Table II:

	Micro-organisms	*Buchholzia coriacea* stem hexane extract	*Gynandropsis gynandra* stem hexane extract	R.S	Control	*Gynandropsis gynandra* leaf hexane extract	*Buchholzia coriacea* leaf hexane extract	R.S	Control
1	*Candida albicans*	21	10	10	10	11.5	13.5	10	10
2	*Aspergillus niger*	19	10	12	10	10	10	10	10
3	*Penicillum sp*	12.5	12	13	10	14.5	17	17	23
4	*Aspergillus flavus*	15.5	13.5	10	10	10	10	10	10
5	*Fusarium oxysporium*	14	13.5	10	13	10	11.5	10	10

Table III

	Micro-organisms	B*uchholzia coriacea* stem methanol extract	*Buchholzia coriacea* leaf methanol extract	R.S	Control	G*ynandropsis gynandra* stem methanol extract	Gynandropsis gynandra leaf methanol extract	R.S	Control
1	*Candida Albicans*	10	14.5	10	10	15.5	17.5	10	10
2	*Aspergillusniger*	12	10.5	17	10	13.5	12.5	14	17
3	Penicillum sp	14	17.5	15	25	17	12	17	16
4	Aspergillus flavus	17	16	10	20	10.5	11	12	10
5	Fusarium oxysporium	11	10	10	15	13.5	10	10	18

Result of Antimicrobial activity (Antifungal studies) in mm
Control = Absolute Methanol
Concentration of extracts = 0.02g/10ml of methanol
Concentration of Reference Standard (Tioconazole) = 10%dilution=0.01g/mlof SDW
8 – 15mm +; 16 – 25mm = + +; 26mm and above = + + +; (-) = No activity

DISCUSSION AND CONCLUSION

From the tests carried out on the plant extracts, it could be concluded that both plants are active on fungi. Therefore, it could be a proof for their being used locally to treat sores and skin diseases. For the antifungal tests, the results were in duplicate and the average taken for the zone of inhibition against fungi. The most active extract against the microbes was found to be *Buchholzia coriacea* stem hexane extract which has the highest zone of inhibition in most microbes. The second most active extract was found to be *Gynandropsis gynandra* stem methanol extract.

From the thin layer chromatography carried out on the most active extract in microbiological studies which is *Buchholzia coriacea* stem hexane extract, about five spots were detected. Also, from the different fractions collected from column chromatography, the TLC done on them revealed that there is a compound present which came out so close to the solvent front in seven different fractions while a group of compound also came out at different retardation factors (RF) in about eleven to twelve fractions.

Therefore, it could be deduced that alkaloids which form the major constituents of this plant have been separated into various components. Further

research would determine which particular alkaloids are responsible for the activities of this plant.

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