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Antifungal activity of a Nigerian herbal plant *Chrysanthellum americanum*

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ABSTRACT

The water and methanol extracts of the leaves of Chysanthellum americanum were screened for antifungal activity using paper disc assay. The aqueous extract was active against Microsporum audouinii Trichophyton mentagrophyte, T. rubrum, Candida. tropicalis, Penicillum chrysogenum and Candida albicans. Methanol extract was not active against all the species of fungi. The susceptibility of the microorganisms to the aqueous extract of the leaves increased with increasing concentration. Microsporum audouinii, showed the highest susceptibility to the aqueous extract at 200mg/0.8ml. The least activity of the aqueous extract of C. americanum was against C. albican and at 50ml/0.8ml. The activity of aqueous extract on M, andouinii, C: albicans, T. rubrum, C. tropicalis, Penicillum chrysogenum and T. mentagraphyte was comparable to that of the standard (mycotin). Preliminary phytochemical screening of the leaves of the plant showed that Chrysanthelum americanum contained flavonoids, saponins and tannins.

Keywords: *water and methanol extracts, paper disc assay, flavonoid, tannin,*

INTRODUCTION

Chysanthellum americanum is found in the Asteraceae family with 10 species (Ernst, 1995). The plant is an annual herb, erect or prostrate, glabrous with feathery bipinnate, compound leaves, which have distinctive aromatic odor. The flowers are sessile, several and crowded on the flat axis. The fruit is dry, indehiscent, winged and one-seeded (Adjanohoun *et al.,* 1989). The plant originated from Peru and was imported by botanists to Africa where it slowly colonized fallow land and roadsides especially in Sudan-Guinea regions (Farnsworth & Soegarta, 1991).

The leaves are used in the treatment of upper respiratory tract infection (Echinacea), lithiasis, inflammations, migraine, cirrhosis of liver, and dermatoses (Farnsworth & Soegarta, 1991). Honore-Thorez (1985) also cited the plant, always in decoction, employed as a mouthwash, to calm toothaches and to wash the head in cases of migraine.

In recent times traditional medicine has been accepted in Africa as an alternative form of health care (Bisignano *et al.,* 1996) largely because of the increase in microbial resistance to available antibiotics given to patients with a range of disease. The leaf of *Chrysanthellum americanum* is a common herb used in the treatment of different skin diseases and other infections in Nigeria and this has necessitated special interest in the suspected antifungal properties of the plant, since there has not been any documented report on this .

Materials and Method

The leaves of *Chrysanthellum americanum* were purchased from local herb dealers in July 2005 in Mushin market and authenticated by Dr O. Kadiri of herbarium section of Botany Department, University of Lagos, Akoka, Lagos, Nigeria. The antifungal assay was carried out against six fungi, *Microsporum audouini, Candida albican, Trichophyton mentagrophyte, T. rubrum, C. tropicalis* and *Penicillum* sp collected as stated earlier from Lagos University Teaching Hospital Mycology Laboratory. The organisms were preserved by storage at 4^0C.

The ground leave sample (10g) was extracted by soaking in 100ml of distilled water and 95% methanol respectively. The water extract was filtered using Whatman No. 1 filter paper and evapourated to

dryness by heating for 2hrs while the methanol extract was dried overnight under the fume cupboard. The weight of the extracts were recorded and the final residue discarded. The aqueous extract of the leaves was reconstituted in distilled water into varying concentrations (50mg/0.8m, 100mg/0.8ml and 200mg/0.8ml) and the methanol extract was in 95% methanol into the same concentration as above. Mycotin (50mg/0.8ml) was the positive fungal standard. The standard was mixed in water and methanol to prepare the two solvent extracts.

The ground sample of the leaves of the plant was also screened for the presence of flavonoids, saponins and tannins.

Sample of ground leaves (2.0g) was boiled with 10ml of ethyl acetate in a water bath, cooled and filtered. Four ml of the filtrate was mixed with 6ml of bromine water and the colour of the bromine water noted.

Sample of ground leaves (2.0g) was boiled in 10ml of distilled water in a water bath for 5mins and the solution was filtered while hot and then cooled. Dilute H_2SO_4 (3ml) was mixed with the filtrate and boiled for 15mins.The mixture was boiled with 2.5ml benedicts solutions and the colour changes noted

Each of the fungi was grown in 50ml of sterile Sabouraud agar in 150ml conical flask for 48hr at room temperature after which 0.1m1 of the culture was aseptically transferred into another flask containing sterile fresh medium. The transfer was repeated twice to obtain actively growing cells.

The susceptibility of each organism to the solvent extracts of the leave was tested using paper disc assay according to the method used by Irobi & Daramola (1994). Twenty four old fungal cultures were adjusted with fresh sterile broth to a concentration of 1.2×10^6 conidia/ml at 0.3 O. D. (600nm) using UV-visible Spectrophotometer. An aliquot (0.5ml) of the cell suspension of each fungal culture was spread on Sabouraud Dextrose agar medium with a bent glass rod after about 30min. When the inoculums were completely absorbed, disc prepared from Whatman filter paper were soaked in the solvent extracts, dried and sterilized under ultra violet (UV) light for 1 hour. The disc of each concentration of the solvent extracts was then gently placed on each incubated plate at 37^0C for 24hours. The radius of the zone of inhibition for each and concentration of the solvents extracts were measured in millimeter using transparent ruler.

Results

The results of the phytochemical screening of the leaves of the plant are presented in Table 1. Preliminary analysis of the leaves showed that the plant contained flavoniods., saponins and tannins. The result of the antifungal screening of the crude extracts of the leaves of *Chrysanthellum americanum* are presented in Table 2 and 3.

The aqueous extract of the leaves of the plant at the following concentrations 50mg/l0.8ml,100mg/0.8ml. 150mg/0.8ml and 200mg/0.8ml inhibited the growth of *Microsporum aduoinii, Candida albicans aoduinii, Candida albicans, T. rubrum, C. tropicalis, Penicillum notatum* and *Trichophyton mentagrophttye.* There was an increase in susceptibility of these organisms to the aqueous extract with increasing concentrations. *M. audonii was the most* susceptible to the solvent extract at 200mg/0.8. *C. albicans* was the least susceptible with zones of inhibitions of 15.5,19,0 20.0 and 21.5 at the varying concentrations. The activity of aqueous extract on the three organisms was comparable to that of the standard (Mycotin, UK) but the methanol extract was not active against all the species of fungi.

Discussion

These secondary metabolites, flavonoid, tannin and saponin detected from the leaves could be responsible for the antifungal activity of *Chrysanthellum americanum* (Harborne, 1973). According to Igboanugo, (2006) *Chrysanthellum americanum* contains some antifungal components which explain its potency against the organisms tested. *Trichophyton* species *Candida albicans* and *Candida tropicalis* a*re* associated with skin,nail, mouth and vaginal infections (Uaboi Egbenni, 2000). The activity of the extracts therefore, are strong indications that the aqueous extract could be very useful in treating skin disease incited by these organisms, suggesting the reason for the use of the plant for the treatment of eczema and ringworm in Nigeria (Adjanohoun *et al.*, 1989). Based on this antifungal activity it is recommended that the traditional medicinal use of the leaves of *Chrysanthellum americanum* should be continued.

Table 1: Reaction of leaf of *Chrysanthellum americanum* during the phytochemical analysis

Test	**inference**
Flavoniods	++
Saponin	+
Tannins	+

Table 2: Zones of inhibition of water extract of *Chrysanthellum americanum* on *Microsporum audonii*, *Trichophyton mentagrophyte*, *Trichophyton rubrum* *Candida tropicalis*, *Candida albicans* and *Penicillum notatum*

Test Organism	*Concentration (mg/0.8ml)*				
	50	**100**	**150**	**200**	Mycotin (50mg/0.8ml)
M. audonii	24.5	18.5	30.0	31.0	32.0
T. mentagrophyte	19.5	25.5	28.0	23.0	29.5
C. albicans	15.5	19.0	20.0	21.5	22.0
C. tropicalis	14.0	16.5	21.0	22.45	21.5
T. rubrum	20.0	22.0	24.0	23.5	28.5
P. notatum	17.0	19.7	22.0	22.6	23.0

Table 3: Zones of inhibition of methanol extract of *Chrysanthellum americanum* on *Microsporum audonii*, *Trichophyton mentagrophyte*, *Trichophyton rubrum* *Candida tropicalis*, *Candida albicans* and *Penicillum notatum*

Test Organism	*Concentration (mg/0.8ml)*					
	50	*100*	*150*	*200*		*control*
M. audonii	ni	ni	ni	ni	ni	3.0
T. mentagrophyte	ni	ni	ni	ni	ni	28.5
C. albicans	ni	ni	ni	ni	ni	23.0
C. tropicalis	ni	ni	ni	ni	ni	22.5
T. rubrum	ni	ni	ni	ni	ni	24.5
P. notatum	ni	ni	ni	ni	ni	23.0

ni = no inhibition

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