Antifungal Activity of *Xylopia aethiopica* on Some Clinical Organisms in Nigeria

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The antifungal properties of water, ethanol and chloroform extracts of the fruits of *Xylopia aethiopica* were investigated using agar well diffusion method. Aqueous extract was not potent against all the organisms tested. Chloroform and ethanol extracts were active against *Candida albicans* and *Aspergillus niger* at different concentrations (50 mg/ml, 75 mg/ml and 100 mg/ml) with varying zones of inhibition. The chloroform extracts of *Xylopia aethiopica* had the highest zone of inhibition of 5.4 mm at 75 mg/ml which is comparable to that of Nystatin. Ethanol extract also had strongest inhibition at 75 mg/ml with 5.0 mm mean zone of inhibition against *Aspergillus niger*. The crude extracts of *Xylopia aethiopica* contained Saponins, Flavonoids, Phenol, Alkaloids and Terpenoids.

Keywords: Antifungal activity, Crude extract, *Xylopia aethiopica*.

Introduction

*Xylopia aethiopica* is an angiosperm belonging to the family Annonaceae used mainly as spices and in traditional medicine. A tree of 20 m height or more with a clear straight bole in 75 cm girth, often with short prop roots, smooth grey bark and scented when fresh (Boakye-Yiadom *et al.*, 1971). The fruit is the most important part of the tree, it is narrow, dark brown or black about 2 inches long with separate carpel on a stout peduncle. The plant is distributed in all forest regions of Senegal to Zaire and has been located in Gambia, Nigeria, Gabon, Ivory Coast, Togo and Tanzania (Day *et al.*, 1990).

Recently, traditional medicine has been accepted as an alternative form of health care largely because of the increase in microbial resistance to available antibiotics given to patients with a range of disease (Dalziel, 1992). The fruit of *Xylopia aethiopica* is a common herb used in the treatment of different ailments like cough, bronchitis, dysentery condition and asthma and other infections in Nigeria and this has necessitated our interest in the antifungal study of the plant.

Materials and Method

**Source of Plant Material**

The fruit of *Xylopia aethiopica* was obtained from Oyingbo Market, a native medicinal plant market in Lagos, Nigeria. The plant part was dried under laboratory condition. Samples of the plant were authenticated by Prof. Olowokudejo of Department of Botany and Microbiology, University of Lagos, Nigeria as well as the text in vernacular names of medicinal plants by Gbile, (1984) and Sofowora, (1996). The antifungal assay was carried out against *Candida albicans*, *Aspergillus niger*, and *Aspergillus termini*. 
Source of Micro-organisms

The fungi used in this work were *Aspergillus niger*, *A. tamarii* and *Candida albicans*. They were collected from Nigerian Institute of Medical Research (NIMR), Yaba, Lagos. The organisms were stored in Sabouraud Dextrose Agar (oxide) slants in the refrigerator at 4°C prior to use.

Preparation of Plant Materials

The ground sample (40 g) was extracted separately overnight with distilled water (250 ml), 99.7% ethanol and 98.5% chloroform (200 ml) respectively. The aqueous extract was prepared at 70°C for 10 mins while the ethanol and chloroform extracts were obtained at room temperature. The three extracts were filtered into vial bottles using Whatman No. 1 filter paper and allowed to air dry. The weight of each extract was determined and the extracts were stored in the refrigerator. The aqueous, ethanol and chloroform extracts were reconstituted into the following concentrations: 50 mg/ml, 75 mg/ml and 100 mg/ml. The concentrations of positive and negative controls were 125 mg/5 ml and 100 ml (Nystatin and sterile water).

Preliminary Phytochemical Analysis

The Preliminary phytochemical studies were carried out using the methods of Harborne (1998), Dawson et al., (1986), and Sofowora (1993), Farnsworth and Euler (1962). The ground sample of the fruit of the plant was screened for the presence of flavonoids, saponins, alkaloids and tannins.

Screening for Antifungal Activity

The antifungal activity testing was carried out using the agar plate well-diffusion method of Irobi and Daramola (1993). About 10 ml of previously prepared Sabouraud Dextrose Agar were poured into Petri dishes (9 mm diameter) and allowed to solidify. Seven days old fungal cultures were adjusted with sterile water to a concentration of $10^6$ cells/ml using haemocytometer. A micropipette was used to introduce 0.1 ml of the spore suspension into the agar plate and spread with a glass spreading rod under aseptic conditions. Later three 5 mm diameter wells were dug on the surface of the agar plate with a sterile cork borer. The plant extract (0.1 ml) was introduced into the wells. Three plates were prepared for each fungus per extract. There were two controls: one containing fungal inocula filled with sterile water (negative control) while the second type had the wells filled with Nystatin (125 mg/5 ml). All plates were incubated at 28°C-31°C. Zones of inhibitions were measured after 72 hours of incubation.

Results and Discussion

The results of testing the solvent extracts of *Xylopia aethiopica* (fruits) against *Candida albicans, Aspergillus niger, Aspergillus tamarii* are presented in Tables 1, 2 and 3. Although chloroform extract was active against the first two organisms at all concentrations, the extract showed the strongest inhibition zone of 5.4 mm at 75 mg/ml concentration. The water extract was not potent against all the organisms tested. Tables 2 and 3 show that the chloroform extract was more potent than ethanol extract because it was active against *Candida albicans* and *Aspergillus niger* at the lowest concentration of 50 mg/ml. The zones of inhibition of the two active solvent extracts up to 75 mg/ml suggest that the extracts contain substances that are fungicidal.

The mean zones of inhibition of *Aspergillus niger* of the chloroform at 75 mg/ml concentration was comparable to that of the standard (Nystatin). This agrees with the report of Adekunle and Okoli (2002). *Aspergillus niger* was the most susceptible to the solvent extracts of *Xylopia aethiopica* at various concentrations tested. This activity strongly indicates that the chloroform and ethanol extracts of this plant could be useful in treating skin infection, eye irritation, and candidiasis. Observation is in line with the work of Adeleye et al., (2003).
TABLE 1
The Mean Zone of Inhibition of Aqueous Extracts of Xylopia aethiopica on Candida albicans, Aspergillus niger and Aspergillus tamarii Cultures

<table>
<thead>
<tr>
<th>Test organisms</th>
<th>Zone diameter (mm) at concentration (mg/ml)</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>50 mm</td>
<td>75 mm</td>
</tr>
<tr>
<td>Candida albicans</td>
<td>Nil</td>
<td>Nil</td>
</tr>
<tr>
<td>Aspergillus niger</td>
<td>Nil</td>
<td>Nil</td>
</tr>
<tr>
<td>Aspergillus tamarii</td>
<td>Nil</td>
<td>Nil</td>
</tr>
</tbody>
</table>

Nil = No zone of inhibition

TABLE 2
The Zone of Inhibition of Chloroform Extracts of Xylopia aethiopica on Candida albicans, Aspergillus niger and Aspergillus tamarii

<table>
<thead>
<tr>
<th>Test organisms</th>
<th>Zone diameter (mm) at concentration (mg/ml)</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>50 mm</td>
<td>75 mm</td>
</tr>
<tr>
<td>Candida albicans</td>
<td>3.1</td>
<td>3.7</td>
</tr>
<tr>
<td>Aspergillus niger</td>
<td>5.3</td>
<td>5.4</td>
</tr>
<tr>
<td>Aspergillus tamarii</td>
<td>Nil</td>
<td>Nil</td>
</tr>
</tbody>
</table>

Nil = No zone of inhibition

TABLE 3
The Zone of Inhibition of Ethanol Extracts of Xylopia aethiopica on Candida albicans, Aspergillus niger and Aspergillus tamarii

<table>
<thead>
<tr>
<th>Test organisms</th>
<th>Zone diameter (mm) at concentration (mg/ml)</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>50 mm</td>
<td>75 mm</td>
</tr>
<tr>
<td>Candida albicans</td>
<td>Nil</td>
<td>Nil</td>
</tr>
<tr>
<td>Aspergillus niger</td>
<td>4.2</td>
<td>5.0</td>
</tr>
<tr>
<td>Aspergillus tamarii</td>
<td>Nil</td>
<td>Nil</td>
</tr>
</tbody>
</table>

Nil = No zone of inhibition
The phytochemical test of the compound constituents of the extracts revealed that *Xylopia aethiopica* contained tannins, terpenoids, flavonoids, alkaloids and steroids. The presence of these secondary metabolites in *Xylopia aethiopica* has been reported by Barnabas (1998), Asekun and Adeniyi (2004) and implicated the plant to have inhibited cell wall formation in fungi leading to death of the organisms. Ogundipe and Oladipo (2002) also reported that tannin and flavonoids have antimicrobial properties and can coagulate protoplasm of micro-organisms.

Therefore these constituents detected in *Xylopia aethiopica* could be responsible for its antifungal activity. Further investigation is however required to isolate, purify and structurally elucidate the active component of this plant.

The toxicity level of the pure compounds can also be investigated with the view of formulating them into crude antibiotic drugs of the therapeutic threshold that is acceptable.

REFERENCES