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Screening of some plants for antimycotic effects

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ABSTRACT: The efficacy of some plant extracts in controlling some pathogenic fungi namely: *Aspergillus niger*, *Aspergillus flavus*, *Fusarium solani*, *Curvularia pallescences* and *Rhizopus stolonifer* was carried out. Crude extracts from five locally occurring plants: *Ocinum gratissimum* (OG: Efinrin), *Vernonia amygdaklina* (VA: Ewuro, Bitter leaf), *Azadirachta indica* (AI: Dongoyaro), *Allium sativum* (AS: Aayu, garlic) and *Anacardium occidentale* (AO: Cashew leaf) were prepared in the laboratory using cold water, hot water and ethanol extraction methods. These extracts were assayed for their antinycotic activities. Observations showed that not all the plants have antifungal effects to these tested organisms.

Key Words: Medicinal plants; Drug resistance; Antimycotic effects; Antifungal effects.

Introduction

Owing to the increase in resistance of microorganisms to available control measures, efforts have been geared at searching for alternative methods of taking care of them. This led to the screening of trees and scrubs especially their barks, stems, roots and leaves for antimicrobial properties based on the fact that many plant materials had been exploited by our forefathers through instinct, intuition, trial and error to combat various ailments. As ideas of different tribes, communities and cultures permeated each other, the use of these plant materials became wide spread. A lot of research is now being conducted to screen as many plants as possible for antimicrobial activities. For instance, Bhowmick and Vardhan (1) studied the effect of leaf extracts from *Cinamomum camphors* and *Cathararithus rosecus* on radial growth, spore germination and sporulation in *Curvularia lunata* and reported that these parameters were completely checked by the extracts. The essential oil from *Corcuma longa* and *Lepidium raderale* shoots were also shown by Singh et al (2) to be inhibitory to mycelial growth of *Aspergillus flavus*. Some spices and vegetables commonly used in human diet have also been shown to possess antimicrobial properties against plant pathogens, for example, Ahmed and Sultana (3) obtained extracts from bulb that has biological activity against *Botryodiplochia theobromae* and *Collectotrichum corclori*. Although, a lot of research work has been done and is still being done to screen plants for antimicrobial activities, more still have to be done for nature has blessed mankind with a lot of plants, moreover, there are a lot of diseases that need to be treated.

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This work therefore was designed to investigate the effectiveness of extracts from the leaves of some plants of local interest against different fungi isolates that play significant role in disease production.

Materials and Methods

Fungi isolates: The following fungi isolates were used *Aspergillus flavus, Aspergillus niger, Rhizopus stolonifer, Fusarium solani, Curcularia pallescences*. Pure cultures of each of the fungi isolates were collected from the Food and Industrial Microbiology Unit, Federal University of Technology, Akure and maintained throughout the period of the research on potato dextrose agar slant.

Preparation of plant extracts: Three methods of extraction were used for the preparation of extracts from the leaves of the following plants: *Anacardium occidentale* (Cashew), *Azadirachta indica* (neem), *Ocinum gratissimum* (efinrin), *Veronia amygdalina* (ewuro) and bulb of *Allum sativum* (garlic). The cold water and hot water extractions were done according to the method of Pandey et al (4). The ethanol extraction however was done according to the method of Benjamin and Lamikanra (5).

In-vitro antimycotic assay: This was done according to the method of Ilesanmi et al (6). Each fungi isolates were first seeded separately into different MacCartney bottle containing 10mls of potato dextrose broth and incubated for 24hrs at 28 – 30°C before use. A sterile pipette was used to pipette 0.1ml of the broth on to already prepared potato dextrose agar plates arranged in duplicates for each organism. A sterile spreader was used to spread the inoculum and the petri-dishes were then incubated for 2hrs before wells were made with the aid of a sterile cork borer. Each well was marked and 0.1ml of the different extracts were added separately to each well. The plates were then labelled and incubated at 28-30°C for 72 hrs. Proper controls were set up for each experiment. The sensitivity of the test isolate to each of the extract is indicated by clear zone of inhibition around the wells containing the plant extract and the diameter of the clear zone is directly proportional to the degree of sensitivity.

Results and Discussion

The result obtained from the *in-vitro* assay revealed that the cold water extract of *O. gratissimum* (efinrin) has no inhibitory effect on all the tested fungi isolates. Also, the ethanol extract of the same plant has no inhibitory effect on all of the tested fungi isolates only the hot water extract gave 10.5mm zone of inhibition on *Rhizopus* species (Table 1). Also, only the hot water extract of *A. occidentale* (cashew leaf) had inhibitory effect of *A. flavus*, both the cold and ethanol extracts have no effect on all of the other tested fungi isolates. The inhibitory effect of *A. occidentale* on *A. flavus* can be seen in Table 2. Cold water extract of the leaves of *A. indica* (Neem) had a degree of inhibition on *A. niger* and not on the other tested fungi isolates (Table 3). There was no zone of inhibition from the assay involving the use of hot water and ethanol extract of the leaves. *V. amygdalina* (ewuro) had no inhibitory effect on all the tested fungi isolates using the three different methods of extraction.

The inhibitory effect of OG on *Rhizopus spp* is similar to the findings of Awuah (7) who reported that hot water extract from fresh leaves of OG and *Chromolaena odorata* and dry fruits of *Xylopia aethilopica* reduce growth of the fungi by 10 – 60%. The result of the inhibitory effect of AI on *A. niger* also agrees with the report of Vir and Sharma (8) who demonstrated that extracts from AI possess antifungal activity against *A. niger, Drechsleria rostrum* and *Macrophomina phaseoli.*

The inhibition of all the tested fungi isolates by AS showed that AS has a high antimycotic property therefore its usage should be encouraged. Garlic is majority used as a spice in foods but when it is cooked, allicin which has been reported to be responsible for the antimicrobial properties (9) is converted to diallyl disulphide which has a much weaker fungicidal and bactericidal properties. Therefore, it is advisable to eat the garlic raw or cut into pieces and take as a medicine with water.

Table 1: The effect of extracts from the leaves of *Ocinum gratisium* on some selected fungi.

Extraction method	**Zone of inhibition of extracts on tested fungi (mm)**				
	A. flavus	*A. niger*	*R. stolonifer*	*R. solani*	*C. pallescences*
Cold water	0.0	0.0	0.0	0.0	0.0
Hot water	0.0	0.0	10.5	0.0	0.0
Ethanol	0.0	0.0	0.0	0.0	0.0

Table 2: The effect of crude extracts of the leaves of *Anacardium occidentale* on some selected fungi

Extraction method	**Zone of inhibition of extracts on tested fungi (mm)**				
	A. flavus	*A. niger*	*R. stolonifer*	*R. solani*	*C. pallescences*
Cold water	0.0	0.0	0.0	0.0	0.0
Hot water	5.0	0.0	0.0	0.0	0.0
Ethanol	0.0	0.0	0.0	0.0	0.0

Table 3: The effect of crude extracts of the leaves of *Azadirachta indica* on some selected fungi.

Extraction method	**Zone of inhibition of extracts on tested fungi (mm)**				
	A. flavus	*A. niger*	*R. stolonifer*	*R. solani*	*C. pallescences*
Cold water	0.0	8.5	0.0	0.0	0.0
Hot water	0.0	0.0	0.0	0.0	0.0
Ethano	0.0	0.0	0.0	0.0	0.0

Table 4: The effect of crude extracts of the bulb of *Allium sativum* on some selected fungi.

Extraction method	**Zone of inhibition of extracts on tested fungi (mm)**				
	A. flavus	*A. niger*	*R. stolonifer*	*R. solani*	*C. pallescences*
Cold water	15.0	16.0	13.5	16.5	11.5

In conclusion, this work has been able to show that not all the plant extracts studied have antifungal activity but the ones that have should be exploited.

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