

Effect of the seed extracts of some medicinal plants on the control of fungi associated with dandruff.

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Abstract

Fungi cause diseases upon the skin and scalp. In the present study, fungi were isolated from scalp and grew on Sabouraud Dextrose Agar medium. The identified fungi include *Aspergillus niger* and *Aspergillus flavus*. They are controlled by using the seed extracts of four medicinal plants. The efficiency of the seeds of *Brassica nigra*, *Nigella sativa*, *Piper nigrum*, and *Terminalia chebula* extracts in different concentrations such as 0.5ml, 1ml, and 2ml were analyzed by using minimum inhibitory concentration method. The fungi isolated from the scalp infection were effectively controlled by *Brassica nigra*, *Nigella sativa*, *Piper nigrum* and *Terminalia chebula* against *Aspergillus niger* and *Aspergillus flavus* respectively.

Keywords: *Aspergillus flavus*, *Aspergillus niger*, *Brassica nigra*, *Nigella sativa*, *Piper nigrum*, scalp infection, *Terminalia chebula*.

INTRODUCTION

Fungi had been recognized as causative agent of human diseases earlier than bacteria. Fungal infections had been described as early as in 1839. Fungal infections however are extremely common and some of them are serious and even fatal. Fortunately though there are over 1,00,000 species of fungi, only about 50 of them are known to be pathogenic, and most of them are opportunistic pathogens (Ahamad *et al.*, 1995).

In humans, the scalp is a specialized area of skin on top of the head, usually covered in both sexes. The scalp contains as many as 1,50,000 hair follicles. It is usually described as having five layers (Anantanarayan, 1997).

Scalp itchiness is a problem affecting 43% of the general population at least once in 12 month period. Many factors contribute to this including blow dryers, cosmetic products, hats allergies, dandruff, mites and even stress. The scalp can reflect the overall conditions of the body and is affected by stress and hormonal changes.

The main causes of scalp infections are various pathogens or microorganisms like bacteria, fungi, viruses or parasites (Ajello *et al.*, 1996). The present paper deals with the control of fungi associated with dandruff using some medicinal plants including *B. nigra*, *N. sativa*, *P. nigrum* and *T. chebula*.

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MATERIALS AND METHODS

Collection of sample

The fungal samples were collected from infected patient. Sterile dry forceps were used for scrubbing the scalp and obtaining dandruff flakes. The flakes were obtained from different areas of the head of different individuals separately using separate sterile forceps and collected in a clean dry Petri dish. Healthy seeds of *B. nigra*, *N. sativa*, *P. nigrum* and *T. chebula* were collected from market in Mannargudi, Thiruvavur (Dt) and transferred to the laboratory in sterile plastic container for anti-fungal assay.

Isolation of fungi

Sabouraud Dextrose Agar medium was prepared. 0.5g of streptomycin was added to the SDA medium and mixed well. The medium was poured into the sterile Petri plates. The plates were incubated and pure culture was obtained and maintained by inoculating the colonies on new SDA medium. Then the fungi were observed by wet mount technique for identification.

Antifungal assay by well diffusion method (Fourneir *et al.*, 2002)

The Sabouraud dextrose agar medium was prepared and sterilized at 120°C and 15lbs for 15 minutes. Then the medium was poured into Petri plates and allowed to solidify. The identified fungal species such as *A. niger* and *A. flavus* were spread on the plates containing SDA medium separately. The wells were made in the agar plates by using cork borer.

Then the seed extracts of *B. nigra*, *N. sativa*, *P. nigrum* and *T. chebula* were prepared by washing the seeds with

clean water and sterilized with 1% ethyl alcohol for removal of contaminants and crushed with sterile pestle and mortar. From that different concentrations of seed extracts such as 25%, 50%, 75% and 100% were made and aseptically poured into Petri plates containing well separately. The plates without plant extracts were maintained as control. Then the plates were incubated at 25±2°C for 3 days. The zone of inhibition was observed and measured.

Minimum inhibitory concentration of seed extracts against fungal pathogens

Sabouraud dextrose broth was prepared and small pinch of streptomycin was added to the medium. The different concentrations of seed extracts (0.5ml, 1ml and 2ml) were added to each tube separately. Then the fungal spores were inoculated into the broth containing different concentration of plant extracts. Then the tubes were incubated at 25±2°C and continuously examined for spectroscopic assay for 3-6 days on daily basis.

RESULTS AND DISCUSSION

The locally available plant extracts such as *B. nigra*, *N. sativa*, *P. nigrum*, and *T. chebula* can be used as the low cost home remedy for scalp infection caused by fungal pathogens. The higher concentration of *B. nigra* and *P. nigrum* extracts inhibited the growth of the organisms such as *A. niger* and *A. flavus*.

Antifungal assay

Among the four plant extracts, *B. nigra* effectively inhibited the growth of *A. niger* at 100% concentration (25.6±0.58mm). At 100% concentration, the aqueous extracts of *B. nigra* also effectively inhibited the growth of *A. flavus*. Guerra *et al.* (1992) also reported that *A. niger* was effectively inhibited by the extract of *B. nigra*. The measured zone of inhibition was 26.6 ± 0.58mm (Table 1-4). This possesses significant threat for being a highly potent opportunistic pathogen. In immuno compromised patients this fungi can cause the release of aflatoxin with significant effects which can even be fatal.

Table 1 : Effect of plant Extracts on the growth of *A. niger*

Name of Medicinal plants	Zone of inhibition[in mm]			
	Concentration			
	100%	75%	50%	25%
<i>Brassica nigra</i>	25.6 ± 0.583	19 ± 1.732	15.3 ± 0.583	-
<i>Nigella sativa</i>	19.3 ± 1.55	15.6 ± 0.583	-	-
<i>Piper nigrum</i>	20.3 ± 1.55	17.6 ± 0.583	13.6 ± 1.732	-
<i>Terminaliachebula</i>	17.6 ± 0.583	14 ± 1.732	-	-

Values are expressed as Mean ± Standard deviation

Table 2 : Effect of plant Extracts on the growth of *A. flavus*

Name of Medicinal plants	Zone of inhibition[in mm]			
	Concentration			
	100%	75%	50%	25%
<i>Brassica nigra</i>	26.6 ± 0.583	18.3 ± 1.55	14.6 ± 1.732	-
<i>Nigella sativa</i>	18.6 ± 2.311	12.6 ± 0.583	-	-
<i>Piper nigrum</i>	20.6 ± 0.583	17.3 ± 1.55	13.3 ± 1.55	-
<i>Terminaliachebula</i>	17.6 ± 2.311	14.3 ± 1.55	-	-

Values are expressed as Mean and Standard deviation

Table 3: Spectrophotometric analysis of antifungal Activity of Plant Extracts by Minimum Inhibitory (Concentration Method Against *A. niger*)

Concentration of Plant Extracts			
Name of Medicinal plants	.5ml	1ml	2ml
<i>Brassica nigra</i>	1.08	2.21	4.08
<i>Nigella sativa</i>	1.08	2.21	4.08
<i>Piper nigrum</i>	1.06	2.16	3.81
<i>Terminaliachebula</i>	1.02	1.81	2.18

Significant number of *A. niger* and *A. flavus* was obtained in the plates for all individuals. This exhibits the higher chances of pathogenicity and toxicosis in immunocompromised individuals. Thus the chance of ill effects by this fungus in such individuals is fuelled up. Invasive sinus aspergillosis due to *A. niger*, *A. flavus* and massive intracranial infections also have been reported (Denning and Hope, 2010). Chances of secondary ill effects such as carcinoma is also at higher probability in such individuals. Hence significant consideration should be given for treating the mycotic infection of this fungus, and presence of these fungi should be prescreened for such individuals to prevent such infections to develop and proceed at later stage. Though the drugs like amphotericin B, itraconazole, voriconazole and posaconazole show significant activity against *Aspergillus species*, but their side effects cannot be ignored. Acute aflatoxicosis (hepatacorcinoma) is also caused by the fungus, which should also be treated effectively. An approach to develop an ideal vaccine for fungal infections is still on prowl and significant work needs to be done on this aspect. Aflatoxin should be primarily concentrated upon due to its serious nature (Woloshuk *et al.*, 2000). The treatment of the diseases caused by the fungi with natural plant products doesnot cause any serious side effects. Hence screening of plants and their parts against various diseases caused by fungi in the need of the hour.

Table 4: Spectrophotometric analysis of antifungal Activity of Plant Extract by Minimum Inhibitory (Concentration Method Against *A. flavus*)

Concentration of Plant Extracts			
Name of Medicinal plants	.5ml	1ml	2ml
<i>Brassica nigra</i>	2.18	3.85	4.89
<i>Nigella sativa</i>	1.06	2.18	2.89
<i>Piper nigrum</i>	2.08	3.18	3.80
<i>Terminaliachebula</i>	1.03	1.19	1.85

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