

# Invitro Comparative Antifungal studies of *Alphonsea sclerocarpa* with Local herbal shampoo against clinically isolated *Aspergillus flavus*

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**Abstract:** The present study aimed to evaluate the antifungal activity of *Alphonsea sclerocarpa* by comparing with local herbal shampoo (Prakruthi vanam shampoo) available in Madanapalle, Chittoor district, Andhra Pradesh. Clinical isolate of pathogenic fungi *Aspergillus flavus* was selected and plated on Potato Dextrose Agar. The plant samples were extracted with solvent methanol and the test Shampoo was diluted in water to get 1:5, 1:10 dilutions. The antifungal activity of both the plant extract and local herbal shampoo were tested against pathogenic fungus *Aspergillus flavus* by agar well – diffusion method. Both plant extract and shampoo shows antifungal activity, in which shampoo was effective against the tested fungi, whereas plant extract were showed moderate antifungal activity in comparison with shampoo.

Keywords: *Alphonsea sclerocarpa*, local Herbal shampoo, *Aspergillus flavus*, Antifungal activity, Agar well diffusion method.

## 1. Introduction

Medicinal Plants play an important role in human life to combat diseases since the times immemorial. Therapeutic properties of medicinal plants are well recognized at global level (Anwar, 2009). It is estimated over 50% of modern clinical drugs have natural products origin (Sturesson, 1982). Most plants contain several compounds with antimicrobial properties. Active compounds are usually responsible for the biological properties of some plant species used throughout the globe for various purposes, including treatment of infectious diseases (Silva, 2010). Medicinal plants have been traditionally used to treat different kinds of infectious diseases (Vijayan, 2004). Infectious diseases represent an important cause of morbidity and mortality among the general population, particularly in developing countries (Silva, 2010). In recent years, antimicrobial properties of plant extracts have been reported with increasing frequency from different parts of the world. *Alphonsea sclerocarpa* is a small tree with simple leaves and aggregated berries, belongs to family *Annonaceae*, distributed in South India and Sri Lanka; in the Western Ghats, South Sahyadri (common in Thirunelveli hills of Agasthyamalai range). It is commonly known as pulusu mamidi (Madhava chetty, 2013). Whole plant used in the treatment of cancer. There were no scientific articles on this plant, thereby indicating lack of exploration into its pharmaceutical benefits. Several synthetic shampoos are available in the market. They may be anionic, cationic, and non ionic surfactants but may be toxic and cause damage to eye on continuous use also may cause drying of hair (Mohamed, 2009). Thus now a days herbal shampoos are widely used than synthetic shampoos. Many shampoos are available in Indian market under the label of natural, herbal. However these formulations are based on synthetic detergents, other chemical additives and contain herbal extracts added more for their marketing value than for their efficacy (Mainkar, 2001). Risk of opportunistic fungal infection has greatly increased in patients who are severely immune compromised. Fungal organisms

have been widely associated with dandruff, majorly the lipase producing fungi *Malassezia furfur*. Dandruff is a common scalp condition that affects most people at something during and after puberty. Flaking of the scalp is a symptom of seborrhoeic dermatitis also called dandruff (Elakkiya, 2014). The main causes of scalp infections are various pathogens or microorganisms like bacteria, fungus, virus or parasites. Investigation strongly supports the use of all the herbal preparations and their combinations as they inhibited in vitro growth of dermatophytes and *Aspergillus* which may cause opportunistic mycoses (Sumangala Rao, 2013). Among these microorganisms, the most common pathogens involved in skin and hair infections are bacteria and fungus (Clayton, 1967). Fungal organisms have been widely associated with dandruff. To control superficial mycoses, cleanliness of hair and scalp are considered as most important. The use of plant derived products and some traditionally used products as antifungal agents have low mammalian toxicity, less environmental effect and wide public acceptance (Selvakumar, 2012). The study was to investigate invitro antifungal activity of *Alphonsea sclerocarpa* in comparison with herbal shampoo by agar well diffusion method against clinically isolated fungi *Aspergillus flavus*.

## 2. MATERIALS AND METHODS

### 2.1 Preparation of plant extract

Fresh Plant leaves of *Alphonsea sclerocarpa* was collected from Tirumala hills, Chittoor district, Andhra Pradesh, India.

### 2.2 Preparation of leaf powder

The leaves of plant were collected and dried under shade. These dried material was mechanically powdered, sieved and stored in an airtight container. These powdered material was used to check Antifungal activity.

### 2.3 Selection of Fungi

*Aspergillus flavus* was collected from clinic and subcultured on Potato dextrose agar to check antifungal activity of herbal shampoo and *Alphonsea sclerocarpa* plant.

### 2.4 Preparation of test samples

Coarsely powdered leaf (100 g) was placed in soxhlet apparatus. Extraction was performed with methanol for 48 hours at room temperature. Then the Extract was filtered. The resulting filtrate was concentrated in vacuum to dryness to give methanolic extract. The extract was allowed to make different concentrations (50 mg/ml and 100 mg/ml). Local herbal shampoo was diluted with distilled water to get 1:5, 1:10 dilutions(Sumangala Rao,2013).

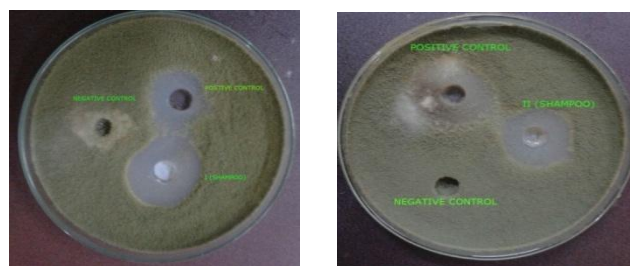
#### 2.4 Determination of antifungal activity

The culture medium potato dextrose agar was inoculated with the fungal strain suspended in. 0.5 cm diameter wells were punched into the agar and filled with plant extract and diluted shampoo solutions. Standard antibiotic (Clotrimazole ) was used as positive control and fungal plates were incubated at 37°C for 72 hours. The diameters of zone of inhibition were measured(Abhishek Mathur,2011).

### 3.RESULTS AND DISCUSSION

The potato dextrose agar plates was cultured with clinically isolated *Aspergillus flavus* were appeared as green colonies. Then the culture was tested with local herbal shampoo and plant extract. Antifungal activity of methanolic extract of *Alphonsea sclerocarpa* against *Aspergillus flavus* as tabulated in table 1.Plant extract shows moderate zone of inhibition with 13mm (50 mg/ ml),8 mm zone (100 mg/ml) shown in fig 1. Antifungal activity (Zone of inhibition) of herbal shampoo against *Aspergillus flavus* shown infig 2 shows 20mm zone (1:5 dilution), 14mm zone (1:10 dilution) with potent antifungal activity as tabulated in table 2.

<i>sclerocarpa</i>	100	8	20
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(a)1: 5 shampoo dilution (b) 1: 10 shampoo dilution  
Fig 2 : Antifungal activity of Prkruthi vanam shampoo against *Aspergillus flavus*

Table 2:Antifungal activity (Zone of inhibition) of Local shampoo against *Aspergillus flavus*

Shampoo dilutions	Conc (ml)	Zone of inhibition (mm)	Positive control(mm)
1:5	0.1	20	20
1:10	0.1	14	20



(a)50 mg/ml plant extract Concentration (b)100mg/ml plant extract Concentration

Fig 1 :Antifungal activity of methanolic extract of *Alphonsea sclerocarpa* against *Aspergillus flavus*

Table1: Antifungal activity (Zone of inhibition) of methanolic extract of *Alphonsea sclerocarpa* against *Aspergillus flavus*

Extract	Conc(mg/ml)	Zone of inhibition (mm)	Positive control
Methanolic extract Of <i>Alphonsea</i>	50	13	20

In recent years, antimicrobial properties of plant extracts have been reported with increasing frequency from different parts of the world(Cowan,1999). *Alphonsea sclerocarpa* belongs to family *Annonaceae*. Previous chemical and pharmacological investigations on some species of *Annonaceae* family have indicated the presence of important bioactive compounds, exhibiting various pharmacological activities including antimicrobial insecticidal and antiparasitic properties(Khan,2002 & Rahman, 2005)*Aspergillus flavus* is a saprotrophic and pathogenicfungusand also an opportunistic human and animal pathogen, causing *Aspergillois* in immune suppressed individuals and it is the most common cause of superficial infection. The increase in antimicrobial resistance and populations of patients at risk, in conjunction with the restricted number of commercially available antifungal drugs that still present many side effects, are the cause for this problem. All commercial synthetic shampoos used exhibited high antifungal activity against *Aspergillus flavus*. six commercially available shampoos (in the Philippines) were assessed for antifungal activity against a human (dandruff) isolate of *M. furfur*: (a) Head & Shoulders (Proctor & Gamble),Gard Violet (Colgate-Palmolive), Nizoral 1% (Janssen), Nizoral 2% (Janssen),Pantene Blue (Proctor & Gamble); and (f) Selsun Blue (Abbott). The results demonstrated that all six of the assayed hair shampoos have some antifungal effect on the test yeast(Bulmer,1999) .The work invitro antifungal activity of different synthetic, local

herbal shampoos and natural products used in traditional medicine was performed on clinical isolates of fungi like *Malassezia* sps, *Trichophyton* sps and *Aspergillus* sps. When compared the effects of test compounds on seven different organisms used, synthetic shampoos showed excellent inhibitory activity on all of them (Sumangala Rao, 2013). Herbal extracts have proved to be good alternatives for the chemical preparations. A number of herbal shampoos and poly herbal hair oils have excellent results due to their synergistic, antifungal, anti-inflammatory and immune stimulatory action (Chandrani, 2012).

#### 4. CONCLUSION

Almost all commercially available hair shampoos publicize that they contain some form of antifungal agents. The present study was concluded that local herbal shampoo and *Alphonsea sclerocarpa* shows antifungal activity against *Aspergillus flavus*. In which, Plant extract shows moderate zone of inhibition and local herbal shampoo shows potent antifungal activity, due to the presence of Shikakai, Aloe vera, Methi, Soap nuts and Amla Prakruthivanam shampoo. This study reveals *Alphonsea sclerocarpa* antifungal activity and might be used for future pharmaceutical formulations.

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