

## Full Length Research

# Isolation of Dermatophytes and Screening of selected Medicinal Plants used in the treatment of Dermatophytoses

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**Antidermatophytic activities of five selected medicinal plants (leaves) used traditionally in the treatment of dermatophytoses. The screened plants include; *Euphorbia balsamifera* Ait, *Mitracarpus scaber* Zucc, *Pergularia tomentosa* L, *Streospermum kunthianum* Cham and *Holarrhena floribunda* (g, Don) Dur. And Schiz. A total of two hundred and fifty samples were obtained from infected skin, hair and nails of individuals within Sokoto metropolis. Four dermatophytes were identified to specie level and one to genus level; they include *Trichophyton rubrum*, *Trichophyton mentagrophyte*, *Microsporum audouinii*, *Microsporum gypseum* and *Microsporum* specie. The percentage prevalence of isolated dermatophytes indicated *T. rubrum* had the highest percentage of 27.6 %, while other samples that could not be identified as dermatophytes had the least percentage of 8.4 %. The results of aqueous, hexane and chloroform extracts of *P. tomentosa* and *M. scaber* have exhibited promising antidermatophytic activities against *T. rubrum*, *T. mentagrophyte* and *M. gypseum* at 10 mg/ml respectively. Minimum Inhibitory Concentrations and Minimum Fungicidal Concentrations of the extracts produced inhibitory action on *T. rubrum*, *T. mentagrophyte* and *M. gypseum* at 10 mg/ml each. Findings from this research proved that *P. tomentosa* and *M. scaber* are more active than the conventional antifungal drug Grisiofulvin. Therefore, this reseach reinforce the use of *P. tomentosa* and *M. scaber* in Nigerian traditional medicine for treating skin infections caused by some dermatophytes.**

**Keywords:** Isolation Dermatophytes, medicinal plants, dermatophytoses and screening

## INTRODUCTION

Dermatophytes are the most common agents of fungal infections worldwide (Robert *et al.*, 2004; Yuanwu *et al.*, 2009). Dermatophytic infections have been considered to be a major public health problem in many parts of the world. The infections are common in the developing countries, and are of particular concern in the tropics, especially in infants (Guest and Sam, 1998). The infections are caused by 40 species of fungi which are grouped into three genera; *Trichophyton*, *Microsporum* and *Epidermophyton* (David *et al.*, 1997). The mode of spread is either by direct or indirect contact with an infected particle which is usually a fragment of keratin containing viable fungus. Indirect transfer may occur via the floor of swimming pools, bath rooms or on brushes,

combs, towels and animal grooming implements (Nweze, 2001; Ngwogu and Otokunefor, 2007). Dermatophytes infections are hardly fatal but mostly debilitating and disfiguring diseases that can give rise to permanent deformations if untreated (Elewski, 1996 and Yuanwu *et al.*, 2009).

Dermatophytes are susceptible to common disinfectants, particularly those containing aerosol, iodine or chlorine. In some cases combine topical and systemic treatment are often used. However, in our society (Sokoto metropolis) today lack of education particularly in knowledge related to clinical mycology, such treatments are not administered at the beginning of such infections until when they have progressed to chronic level. More to this are the cost device and availability of such antifungal agents which are sometimes beyond the reach of the common man, as a result people revert as usual to the traditional means of treatment.

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The use of medicinal plants is very wide spread in many parts of the world, Nigeria inclusive (Zavala *et al.*, 1997). Traditional medicine is the oldest method of curing diseases and infections and various plants are used in different part of the world to treat human diseases and infections (Caceres *et al.*, 1991; Venugopal and Venugopal 1995; Nweze *et al.*, 2004; Vineela and Elizabeth, 2005).

In Nigeria, many plants are used against infectious diseases, which today are frequent due to very poor hygienic conditions, cost and microbial resistance to the time- honoured antibiotics (Ngono Ngane *et al.*, 2000). The continuing increase in the incidence of fungal infections together with the gradual rise in resistance of bacterial and fungal pathogens for antibiotics and antifungals highlights the need to find alternative sources from medicinal plants (Berkowitz, 1995, Jennifer and Paul, 2000). This work was designed to isolate and identify dermatophytes from infected individuals with physical skin lesions and to screen five medicinal plants for potential antidermatophytic activity. The selection of these plants for evaluation was based on ethanomedical information obtained from traditional healers in who used the plants for treatment of dermatophytic infection.

## MATERIALS AND METHODS

### Sample collection

Fresh leaves of *Euphorbia balsamifera* Ait, *Mitracarpus scaber* Zucc, *Pergularia tomentosa* L, *Streospermum kunthianum* Cham and *Holarrhena floribunda* (g, Don) Dur. And Schiz, were collected around Usmanu Danfodiyo University (permanent site) Sokoto. The plants were identified and confirmed at Usmanu Danfodiyo University, Sokoto Herbarium (Botany unit, Department of Biological Science). Voucher specimens were deposited in the Herbarium. The plant materials (fresh leaves) were air dried, pulverized into a fine powder.

A total of two hundred and fifty (250) samples of infected skin, hair and nail were collected from infected patients with clinical manifestations of dermatophytosis within Sokoto metropolis (schools, barbing salons and hospitals). The sites of infections were first cleaned with surgical spirit, scales from the skin lesions were collected by scraping outwards with a blunt scalpel from the edge of the lesion. Specimens from the scalp were collected using forceps to pluck from the scalp. Samples of nail were collected by scraping materials from underneath the nail and from edge of the nail. All the samples were collected on a clean piece of paper (5cm square) the papers were folded to enclose the specimen, they were labelled and transferred to mycology laboratory of the Biological Sciences Department of Usmanu Danfodiyo University, Sokoto for the culture of associated dermatophytes.

### Extraction and fractionation procedure

Extraction and fractionation of the plants extract was carried out by activity guided fractionation according to Moris and Aziz, 1976. The procedure was carried out using ethanol-water (1:1 v/v) and different organic solvents, (Hexane, Petroleum ether and Chloroform).

Forty grams of the powdered plant materials were extracted using

percolation process in 250ml distilled water and 250ml ethanol at 35°C overnight. The extract was filtered and the filtrate was partitioned with 250ml hexane. The extract was separated by filtration. The hexane filtrate was evaporated to dryness at 40°C to obtain residue. The remaining water ethanol extract was further partitioned with petroleum ether and chloroform using the same procedure above. The last remaining water ethanol extract was also evaporated to dryness to yield residue. The dried extracts were reconstituted in water at different concentrations of 10, 20, 40, 80 and 160 mg/ml respectively. Another extraction was carried out using 40g of procured plant material with 500 ml distilled water at 35°C overnight. The extract was filtered and evaporated to dryness, and residues were obtained in gram.

## Isolation and identification of dermatophytes

### Microscopic examination

Microscopic examination was carried out in accordance with Mackie and McCartney (1999). The hair, nail and skin scrapings were examined microscopically for the characteristic of macroconidia and microconidia, presence of hyphae and arthroconidia. Samples were treated with 20% potassium hydroxide (KOH) solution by flooding on slides. Cover slips were used with application of gentle heat. Microscopy was carried out under low power and subduced light. Infected hair and skin were seen encased in regular sheath of arthrospores that doubled their normal thickness; lactophenol cotton blue was used to improve visualization.

### Inoculation and isolation of dermatophytes from samples

Scrapings, of skin and nail were reduced in size to pieces approximately 1 mm across and the hair roots were cut into similarly sized fragments. Both samples were planted on the surface of selected medium that is Sabourad Dextrose Agar containing chloramphenicol at 500 mg/L. The culture media were incubated at 30°C for up to 21 days. After isolation the cultures were transferred to freshly prepared SDA media to obtain pure cultures. Pure cultures were also maintained in SDA slants at 5±1°C. The test dermatophytes were identified by their cultural morphology and microscopic characteristics (Hartman and Rohde, 1980 and Cheesbrough, 2003).

### Identification of dermatophytes

Identification of dermatophytes was done in accordance with Hartman and Rohde (1980). The identification was based on colonial appearance, pigment production and the micro morphology of the spore produced. Cultures were examined at 4 or 5 days intervals from the onset. Some characteristics were also noted the texture, colour and shape of the upper thallus and the production of pigment on the underside. Identified isolates were sent for confirmation at IITA Ibadan (Germ plasm unit). The confirmed isolates were: - *T. rubrum*, *T. mentagrophyte*, *M. gypseum*, *M. audouinii* and *Microsporum specie*.

### Invitro Anti-Fungal Assays

#### Determination of the antidermatophytic activity using Agar incorporation method

The preliminary anti-dermatophytic activities of the aqueous and organic solvents extracts were evaluated using dermatophytes

isolated from infected individuals. The dermatophytes were maintained on Sabouraud Dextrose Agar (SDA) medium. The preliminary screening for antidermatophytic activity was carried out using agar incorporation method, as described by Zacchino *et al.*, (1999) and Hassan *et al.*, (2007). The solvents used were water, hexane, petroleum ether and chloroform.

The fungal isolates (dermatophytes) were cultivated on Sabouraud Dextrose Agar (SDA) medium in 90mm petridishes. Five milliliter of water solution of each extract of the plants at concentrations, 10, 20, 40, 80 and 160 mg/ml, were aseptically mixed with 15ml of SDA (Liquified and maintained at melting point in water bath at 45°C. Griseofulvin, positive control (Clarion medicals Ltd. Lagos, Nigeria), was measured from the pulverized 500 mg tablet. Five milliliter of water solution of griseofulvin at concentrations of 10 and 160 mg/ml were aseptically mixed with 15ml of SDA. After cooling and solidification of the medium, the seeding was carried out by inoculation of all the dermatophytes isolates in the middle of the petridishes. The treated and control petridishes were incubated at ambient laboratory condition for 21 days. Three replicates for each concentration were made. Growth was observed after 7 days. Water was used as negative control. Presence of growth (+) is a negative test (indicating the non-potency of the drug) and absence of growth (-) is a positive test (showing the potency of the drug).

#### Determination of Minimum Inhibitory Concentration (MIC)

The Minimum Inhibitory Concentration of the plant extracts that showed antidermatophytic activity was assayed using the standardized procedure described by Gbodi and Irobi (1992); Ngonu Ngane *et al.*, (2000) and Wokoma *et al.*, (2007) with slight modifications. A total number of twelve test tubes (Khan Tubes) were used for the determination of MIC. The MIC was determined by the tube dilution technique. 1 ml of Sabouraud dextrose broth was dispensed into test tubes 2 to 12 each. From the stock solution of the plant extracts (160 mg/ml), 1 ml was dispensed into tube 1 and another 1 ml into tube 2. From the content in tube 2 serial dilutions were carried out up to the test tube number 10. From tube 10, 1 ml was pipetted out and discarded. The concentrations in the tubes were 160, 80, 40, 20, 10, 5, 2.5, 1.25, 0.625 and 0.3125 mg/ml. 1 ml of dermatophytes spore suspension of each of the test organisms previously diluted to give  $10^5$  spores per ml was dispensed into tubes 1 to 12 with the exception of tube 11. To tube 11 1 ml of sterilized S.D.B. was added. Tube 1 which contained 1 ml of the spore solution of the test organism and 1 ml of the plants extracts served as control for the extracts. Tube two which contained 1 ml of sterilized S.D.B and another 1 ml of S.D.B served as a control for the sterility of the medium. Tube twelve with 1 ml of spore solution of the test organisms and 1 ml of sterilized S.D.B, served as a control for the viability of the test organisms. All the tubes were incubated at ambient laboratory conditions and growth was observed from 7 - 21 days. MIC was regarded as the concentration in the tube that fails to show evidence of growth (turbidity) just immediately after the last one that show growth.

#### Determination of Minimum Fungicidal Concentration (MFC) of the extracts

The MFC was determined by culturing the content of the tube cultures that showed no visible growth in the MIC determination test described above. A loopful of the mixture contained in the tubes was subcultured on freshly prepared Sabouraud dextrose agar plates and incubated at ambient laboratory conditions for 21 days. A control comprising the test organism grown on fresh agar medium was also set up. The minimum fungicidal concentration was regarded as the lowest concentration of the extracts that did not

permit any visible colony growth on the agar medium after the period of re-incubation. The MFC test was set up in three replicates.

## RESULTS AND DISCUSSION

A total number of two hundred and fifty (250) samples were isolated for dermatophytes; out of these samples four (4) fungal organisms (dermatophytes) were identified to specie level and one (1) was identified to genus level. The species identified include; *Trichophyton rubrum*, *Trichophyton mentagrophytes*, *Microsporium audouinii*, *Microsporium gypseum* and *Microsporium specie*. The percentage prevalence of dermatophytes isolated is shown in figure 1.0. *Trichophyton rubrum* had the highest number of occurrence with 27.6% followed by *Trichophyton mentagrophyte* 20.8%, *Microsporium audouinii* 16.0%, *Microsporium gypseum* 12.8%, *Microsporium species* 14.4% and others that cannot be identified as dermatophytes had 8.4%. The percentage prevalence of *Trichophyton rubrum* agrees with the findings of Hartman and Rohde, (1980) and Anonymous, (2008), who reported that *T. rubrum* is one of the most common fungi pathogenic for man; about 58% of the dermatophytes species isolation is *T. rubrum*. *T. rubrum* has been reported as one of the anthropophilic species of dermatophytes, and anthropophilic species are primarily adapted for parasitism of man (Mukoma, 2000). The frequency of occurrence of *T. rubrum* may be associated with the fact that anthropophilic dermatophytes are mainly associated with community life (Mukoma, 2000). The frequency of occurrence of this organism may also be attributed to collection of many samples from infected hair and nails. The next organism in terms of percentage prevalence is *T. mentagrophyte* 20.8%. According to Anonymous (2008) about 27% of the dermatophytes species isolation is *T. mentagrophyte*. The high frequency of occurrence of this specie may be attributed to the source of collection of samples, This also agree with findings of Mukoma (2000), in which the high frequency of occurrence of *T. mentagrophyte* was attributed to the sources of the samples used. Other samples that could not be identified as dermatophytes had the least percentage prevalence of 8.4%.

The results of antidermatophytic activity of aqueous extract of *P. tomentosa* tested at different concentrations of 10, 20, 40, 80 and 160 mg/ml is presented in Table 2. Aqueous extract of the plants was active on *Trichophyton mentagrophyte* and *Trichophyton rubrum* at all the concentrations employed 10 to 160 mg/ml. Griseofulvin at concentrations of 10 and 160 mg/ml showed total effect on the growth of only *Trichophyton rubrum*.

The results of antidermatophytic activity of aqueous extract of *M. scaber* at various concentrations of 10, 20, 40, 80 and 160 mg/ml is shown in Table 3. The aqueous extract exhibited high activity on *Trichophyton mentagrophyte* and *Trichophyton rubrum* at 10 mg/ml. The extract was active on the growth of *Microsporium*

**Table 1:** Percentage prevalence of dermatophytes isolated

| Dermatophytes                     | Frequency of isolation (n = 250) | Percentage prevalence (%) |
|-----------------------------------|----------------------------------|---------------------------|
| <i>Trichophyton rubrum</i>        | 69                               | 27.6                      |
| <i>Trichophyton mentagrophyte</i> | 52                               | 20.8                      |
| <i>Microsporum audouinii</i>      | 40                               | 16.0                      |
| <i>Micsporum gypseum</i>          | 32                               | 14.4                      |
| <i>Microsporum specie</i>         | 36                               | 12.8                      |
| Others                            | 21                               | 8.4                       |
| <b>Total</b>                      | <b>250</b>                       | <b>100%</b>               |

**Table 2:** Antidermatophytic activity of aqueous extract of *Pergularia tomentosa*

| Plant /control<br>Conc.mg/ml | Growth of pathogens |                         |                    |                   |              |
|------------------------------|---------------------|-------------------------|--------------------|-------------------|--------------|
|                              | <i>T. rubrum</i>    | <i>T. mentagrophyte</i> | <i>M. audouini</i> | <i>M. gypseum</i> | <i>M. sp</i> |
| 10                           | -                   | -                       | +                  | +                 | +            |
| 20                           | -                   | -                       | +                  | +                 | +            |
| 40                           | -                   | -                       | +                  | +                 | +            |
| 80                           | -                   | -                       | +                  | -                 | +            |
| 160                          | -                   | -                       | +                  | -                 | +            |
| Griseofulvin 10              | -                   | +                       | -                  | +                 | +            |
| 160                          | -                   | -                       | +                  | -                 | -            |
| Water                        | +                   | +                       | +                  | +                 | +            |

**Key:**

+ = presence of growth

- = Absence of grow

*audouinii* and *Microsporum gypseum* at higher concentrations of 80 and 160 mg/ml. Griseofulvin (positive control) showed significant effect on the growth of only *Trichophyton rubrum* at all the concentrations employed, while the drug had activity on *M. audouinii*, *M. gypseum* and *Microsporum species* at 160 mg/ml. The negative control did not show effect on the growth of the test isolates. Antidermatophytic activity of aqueous extract of *S. kunthianum* is presented in Table 4. The plant was not active on any the tested dermatophytes at concentrations used. Griseofulvin the positive control showed activity only on *T. rubrum* at all the concentrations tested (10 and 160 mg/ml). The result of antidermatophytic activity of *E. balsamifera* against the tested dermatophytes is shown in Table 5. The extract of the plant did not show effect on all the tested dermatophytes. Griseofulvin exhibited total inhibition of *T. rubrum* at 10 mg/ml. The extract of *H. floribunda* did not show antidermatophytic effect on the tested organisms (Table 6). The antidermatophytic activities of *P. tomentosa*, *M. scaber*, *E. balsamifera*, *S. kunthanum* and *H. floribunda* Tables 1-5, indicated that the part used (leaves) for *P. tomentosa* and *M. scaber* were active against most of the tested dermatophytes. The extracts of the rest of the plants were less active. It is not surprising

that there were differences in the antidermatophytic effects of these plants due to the phytochemical properties and differences among species. It is quite possible that some of the plants that were ineffective in this research do not possess antifungal properties, or the plant extracts may have contain antifungal constituents not in sufficient concentrations as to be effective. It is also possible that the active chemical constituents were not soluble in the organic solvents used in the extraction. The drying process may have caused conformational changes to occur in some of the chemical concituents found in these plants. Both hexane and chloroform extracts of *P. tomentosa* showed promising antidermatophytic activity against *T. rubrum*, *T. mentagrophyte* and *M. gypseum*, from the least concentration of 10 mg/ml used. This agrees with the findings of Hassan *et al.*, (2007), in which both above organic solvents extract of *P. tomentosa* were active against some fungal isolates including dermatophytes (*T. rubrum* and *M. gypseum*). Action of the aqueous, hexane and chloroform extracts of *P. tomentosa* against the dermatophytes tested may be due to inhibition of fungal cell wall due to pore formation in the cell and leakage of cytoplasmic constituents by the active components such as alkaloids, saponins, protein, amino acid and sphingolipid biosynthesis and electron transport

**Table 3:** Antidermatophytic activity of aqueous extract of *Mitracarpus scaber*.

| Plant /control<br>Conc.mg/ml | Growth of pathogens |                         |                    |                   |              |
|------------------------------|---------------------|-------------------------|--------------------|-------------------|--------------|
|                              | <i>T. rubrum</i>    | <i>T. mentagrophyte</i> | <i>M. audouini</i> | <i>M. gypseum</i> | <i>M. sp</i> |
| 10                           | -                   | -                       | +                  | +                 | +            |
| 20                           | -                   | -                       | +                  | +                 | +            |
| 40                           | -                   | -                       | +                  | +                 | +            |
| 80                           | -                   | -                       | -                  | -                 | +            |
| 160                          | -                   | -                       | -                  | -                 | +            |
| Griseofulvin 10              | -                   | +                       | +                  | +                 | +            |
| 160                          | -                   | -                       | -                  | -                 | -            |
| Water                        | +                   | +                       | +                  | +                 | +            |

**Key:**

+ = presence of growth

- = Absence of growth

**Table 4:** Antidermatophytic activity of aqueous extract of *Stereospermum kunthianum*.

| Plant /control<br>Conc.mg/ml | Growth of pathogens |                         |                    |                   |              |
|------------------------------|---------------------|-------------------------|--------------------|-------------------|--------------|
|                              | <i>T. rubrum</i>    | <i>T. mentagrophyte</i> | <i>M. audouini</i> | <i>M. gypseum</i> | <i>M. sp</i> |
| 10                           | +                   | +                       | +                  | +                 | +            |
| 20                           | +                   | +                       | +                  | +                 | +            |
| 40                           | +                   | +                       | +                  | +                 | +            |
| 80                           | +                   | +                       | +                  | +                 | +            |
| 160                          | +                   | +                       | +                  | +                 | +            |
| Griseofulvin 10              | -                   | +                       | +                  | +                 | +            |
| 160                          | -                   | -                       | -                  | -                 | -            |
| Water                        | +                   | +                       | +                  | +                 | +            |

**Key:**

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**Table 5:** Antidermatophytic activity of aqueous extract of *Euphorbia balsamifera*.

| Plant /control<br>Conc.mg/ml | Growth of pathogens |                         |                    |                   |              |
|------------------------------|---------------------|-------------------------|--------------------|-------------------|--------------|
|                              | <i>T. rubrum</i>    | <i>T. mentagrophyte</i> | <i>M. audouini</i> | <i>M. gypseum</i> | <i>M. sp</i> |
| 10                           | +                   | +                       | +                  | +                 | +            |
| 20                           | +                   | +                       | +                  | +                 | +            |
| 40                           | +                   | +                       | +                  | +                 | +            |
| 80                           | +                   | +                       | +                  | +                 | +            |
| 160                          | +                   | +                       | +                  | +                 | +            |
| Griseofulvin 10              | -                   | +                       | +                  | +                 | +            |
| 160                          | -                   | -                       | -                  | -                 | -            |
| Water                        | +                   | +                       | +                  | +                 | +            |

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chain (Hassan *et al.*, 2007). The present study highlights the use of *P. tomentosa* in folk medicine for the treatment of mycotic infections and underscores the importance of the ethnobotanical approach for the selection of *P. tomentosa* in the discovery of new bioactive compounds which is in agreement with the works of Van-wyk *et al.*, (1997) in which antidermatophytic activity of *P.*

*tomentosa* was tested and results obtained in this study showed that the plant has antifungal effect against *Aspergillus niger*. This finding also supported the results obtained in this work, where different extracts of the plant exhibited significant effect against some fungal (dermatophytes) isolates.

The antidermatophytic activity of hexane, petroleum

**Table 6:** Antidermatophytic activity of aqueous extract of *Hollarhena floribunda*.

| Plant /control<br>Conc.mg/ml | Growth of pathogens |                         |                    |                   |              |
|------------------------------|---------------------|-------------------------|--------------------|-------------------|--------------|
|                              | <i>T. rubrum</i>    | <i>T. mentagrophyte</i> | <i>M. audouini</i> | <i>M. gypseum</i> | <i>M. sp</i> |
| 10                           | +                   | +                       | +                  | +                 | +            |
| 20                           | +                   | +                       | +                  | +                 | +            |
| 40                           | +                   | +                       | +                  | +                 | +            |
| 80                           | +                   | +                       | +                  | +                 | +            |
| 160                          | +                   | +                       | +                  | +                 | +            |
| Griseofulvin 10              | -                   | +                       | +                  | +                 | +            |
| 160                          | -                   | -                       | +                  | -                 | -            |
| Water                        | +                   | +                       | +                  | +                 | +            |

**Key:**

+ = presence of growth

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**Table 7:** Antidermatophytic activity of extract of *P. tomentosa* in organic solvents

| Organic solvent extract<br>Conc mg/ml | Growth of pathogens |                         |                   |
|---------------------------------------|---------------------|-------------------------|-------------------|
|                                       | <i>T. rubrum</i>    | <i>T. mentagrophyte</i> | <i>M. gypseum</i> |
| Hexane                                |                     |                         |                   |
| 10                                    | -                   | -                       | -                 |
| 20                                    | -                   | -                       | -                 |
| 40                                    | -                   | -                       | -                 |
| 80                                    | -                   | -                       | -                 |
| 160                                   | -                   | -                       | -                 |
| Petroleum ether                       |                     |                         |                   |
| 10                                    | +                   | +                       | +                 |
| 20                                    | +                   | +                       | +                 |
| 40                                    | +                   | +                       | +                 |
| 80                                    | +                   | +                       | +                 |
| 160                                   | +                   | +                       | +                 |
| Chloroform                            |                     |                         |                   |
| 10                                    | -                   | -                       | -                 |
| 20                                    | -                   | -                       | -                 |
| 40                                    | -                   | -                       | -                 |
| 80                                    | -                   | -                       | -                 |
| 160                                   | -                   | -                       | -                 |
| Griseofulvin                          |                     |                         |                   |
| 10                                    | -                   | -                       | +                 |
| 160                                   | -                   | +                       | +                 |
| Water                                 | +                   | +                       | +                 |

**Key:**

- = Absence of growth

+ = Presence of growth

ether and chloroform extracts of *P. tomentosa* is shown in Table 7. Hexane and chloroform extracts of the plant showed activity on the growth of some tested dermatophytes. Hexane and chloroform extracts of *P. tomentosa* was active on *T. rubrum*, *T. mentagrophyte*

and *M. gypseum* by showing total inhibition of growth of these organisms at 10 mg/ml. Petroleum ether extract did not show effect on the growth of any of the tested organisms. Griseofulvin positive control showed activity only on *T. rubrum*, with less effect on other species of

**Table 8:** Antidermatophytic activity of extract of *M. scaber a* in organic solvents

| Organic solvent/extract<br>Conc mg/ml | Growth of pathogens |                         |                   |
|---------------------------------------|---------------------|-------------------------|-------------------|
|                                       | <i>T. rubrum</i>    | <i>T. mentagrophyte</i> | <i>M. gypseum</i> |
| Hexane                                |                     |                         |                   |
| 10                                    | -                   | -                       | -                 |
| 20                                    | -                   | -                       | -                 |
| 40                                    | -                   | -                       | -                 |
| 80                                    | -                   | -                       | -                 |
| 160                                   | -                   | -                       | -                 |
| Petroleum ether                       |                     |                         |                   |
| 10                                    | +                   | +                       | +                 |
| 20                                    | +                   | +                       | +                 |
| 40                                    | +                   | +                       | +                 |
| 80                                    | +                   | +                       | +                 |
| 160                                   | +                   | +                       | +                 |
| Chloroform                            |                     |                         |                   |
| 10                                    | -                   | -                       | -                 |
| 20                                    | -                   | -                       | -                 |
| 40                                    | -                   | -                       | -                 |
| 80                                    | -                   | -                       | -                 |
| 160                                   | -                   | -                       | -                 |
| Griseofulvin                          |                     |                         |                   |
| 10                                    | -                   | -                       | +                 |
| 160                                   | -                   | +                       | +                 |
| Water                                 | +                   | +                       | +                 |

**Key:**

- = Absence of growth

+ = Presence of growth

dermatophytes.

The antidermatophytic activity of hexane, petroleum ether and chloroform extracts of *M. scaber* is presented in Table 8. Both hexane and chloroform extract exhibited significant effect on the growth of some tested dermatophytes. Hexane and Chloroform extracts of the plant were active against *T. rubrum*, *T. mentagrophyte* and *M. gypseum* at all the concentrations employed. The petroleum ether extract did not show effect on any of the organisms. Griseofulvin (Positive control) was active only on one of the test isolates, *T. rubrum* at all the concentrations used (10 and 160 mg/ml). The antidermatophytic activities of organic solvents extracts of *E. balsamifera*, *S. kunthianum* and *H. floribunda* carried out did not show any effect on the dermatophytes, and therefore the results were not shown. The antidermatophytic activities of the aqueous and organic solvents extracts of *M. scaber* (Tables 3 and 8) showed significant antidermatophytic activities against some dermatophytes, *T. rubrum*, *T. mentagrophyte* and *M. gypseum*. Results obtained on the antidermatophytic activities of extracts of *M. scaber*, indicated that the plant is a potent antidermatophytic agent. Hexane and

chloroform extracts of the plant showed activity against the test isolates (dermatophytes) at low concentration 10 mg/ml. Findings from this study were comparable with the reports of Van-wyk *et al.*, (1997), who reported that *M. scaber* is an effective antifungal agent and also revitalizes areas of hypopigmentation and hyperpigmentation. The antidermatophytic activity exhibited by this plant is strongly in line with the assumption that any plant that proved to be active against bacteria can also be active against dermatophytes. The methanolic extract and isolated constituents of the aerial parts of *M. scaber* were reported to exhibit both antibacterial and antimycotic activities (Aberé *et al.*, 2007), these finding strongly supports the result obtained in this work where the parts used in the work (leaves) exhibited high antidermatophytic activity on some of the dermatophytes tested.

All the organic solvents used for extraction of antidermatophytic components of the plants in this research are known to enjoy extensive patronage in the fractionation of antifungal and antidermatophytic components of medicinal plants (Subramanian *et al.*,

**Table 9:** Minimum inhibitory concentration (MIC) in mg/ml of hexane and chloroform extracts of *P. tomentosa* and *M. scaber*

| Extract in solvent | MIC values in mg/ml         |                         |                   |                           |                         |                   |
|--------------------|-----------------------------|-------------------------|-------------------|---------------------------|-------------------------|-------------------|
|                    | <i>Pergularia tomentosa</i> |                         |                   | <i>Mitracarpus scaber</i> |                         |                   |
|                    | <i>T. rubrum</i>            | <i>T. mentagrophyte</i> | <i>M. gypseum</i> | <i>T. rubrum</i>          | <i>T. mentagrophyte</i> | <i>M. gypseum</i> |
| Hexane             | 10                          | 10                      | 10                | 10                        | 10                      | 10                |
| Chloroform         | 10                          | 10                      | 10                | 10                        | 10                      | 10                |
| Griseofulvin       | 10                          | 80                      | -                 | 10                        | 80                      | -                 |

Key :

- = No MIC value

**Table 10:** Minimum Fungicidal Concentration (MFC) in mg/ml of hexane and chloroform extracts of *P. tomentosa* and *M. scaber*

| Extract in solvent | MIC values in mg/ml         |                         |                   |                           |                         |                   |
|--------------------|-----------------------------|-------------------------|-------------------|---------------------------|-------------------------|-------------------|
|                    | <i>Pergularia tomentosa</i> |                         |                   | <i>Mitracarpus scaber</i> |                         |                   |
|                    | <i>T. rubrum</i>            | <i>T. mentagrophyte</i> | <i>M. gypseum</i> | <i>T. rubrum</i>          | <i>T. mentagrophyte</i> | <i>M. gypseum</i> |
| Hexane             | 10                          | 10                      | 10                | 10                        | 10                      | 10                |
| Chloroform         | 10                          | 10                      | 10                | 10                        | 10                      | 10                |
| Griseofulvin       | 10                          | 80                      | -                 | 10                        | 80                      | -                 |

Key :

- = No MFC value

2006). This may be the reason for reasonable antidermatophytic activities exhibited by the extracts of *P. tomentosa* and *M. scaber*. The results obtained in this work is comparable to that of Padmaja *et al.*, (1995) who reported that the hexane extract of the leaves and stem bark of *Amona globra* although a different plant showed substantial antidermatophytic activity. Chakraborty *et al.*, (1995) also showed that the alcoholic extract of the stem bark of *M. scaber* was found to be active against fungi, gram positive and negative bacteria.

The Minimum Inhibitory Concentrations (MIC) of hexane and chloroform extracts of *P. tomentosa* and *M. scaber* is presented in Table 9. Both the Hexane and Chloroform extracts of *P. tomentosa* and *M. scaber* had produced inhibitory at 10 mg/ml against *T. rubrum*, *T. mentagrophyte* and *M. gypseum*. Griseofulvin the positive control had an MIC of 10 mg/ml against *T. rubrum*, while at 80 mg/ml the drug had effect on *T. mentagrophyte*. The results obtained from Minimum Fungicidal concentration (FMC) of the extract is reflected on Table 10. Fungicidal effects were observed at the same concentrations as the MIC. Results of Minimum Inhibitory Concentrations (MIC) and Minimum Fungicidal Concentration (FMC) of hexane and chloroform extracts of *P. tomentosa* and *M. scaber* (Tables 9 and 10) showed that when the dermatophytes were cultured in broths

containing extracts, the inhibitory activity was observed at 10 mg/ml each, suggesting that the extracts possess potential antidermatophytic agents. The dermatophytes (*T. rubrum*, *T. mentagrophyte* and *M. gypseum*) were particularly sensitive. This findings correspond with the findings of Gbodi and Irobi 1992 who observed the MIC of *A. quadriclineatus* though a different plant against the same species of dermatophytes (*M. gypseum*, *M. audouinii* and *T. rubrum*) at low doses of 0.090 mg/ml to 0.192 mg/ml.

## CONCLUSION

Antidermatophytic activities of aqueous and organic solvents extracts of five medicinal plants screened revealed two out of the plants as the most active; the plants are *P. tomentosa* and *M. scaber*. They were active against *T. rubrum*, *T. mentagrophyte* and *M. gypseum*. The MIC and MFC results were 10 mg/ml each.

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