

Study on Prevalence , Identification and Antifungal Susceptibility pattern of Dermatophytosis

INTRODUCTION

Dermatophytosis is a superficial fungal infection of keratinized tissue caused by a group of filamentous fungi such as *Trichophyton*, *Epidermophyton*, *Microsporum*. They are a group of taxonomically related fungi that utilize keratin as a source of nutrients and colonize keratinized tissues including stratum corneum of epidermis, nail, and hair¹. Even though it's a superficial infection, dermatophytes do evoke inflammatory responses like scaling, vesiculation, pustulation, and abscess formation sometimes, because of their metabolic activities. Thus, the clinical infection occurs if fungi penetrate the host's protective barrier. Dermatophytosis is by far the most common fungal disease in humans and sometimes it becomes invasive.

Multiple factors may affect the incidence of this fungal infection within a population. These include geographical area, climate, immunocompetence of the host, pathogenicity of the agent, and availability of the treatment. The growing number of immunocompromised patients due to chemotherapy, transplant, HIV has led to an increased incidence of dermatophytosis. These infections are both endemic and epidemic spreading easily and rapidly, especially in lower economic classes, and therefore require early and safe therapy. Epidemics of dermatophytosis have also been reported in the area of overcrowding and poor hygienic conditions.

The typical infections of dermatophytes are generally referred to as ringworm infections due to their ring-like appearance². These infections are also known as 'tinea infections' and are named according to the location of the lesion of the body example- tinea capitis refers to the ringworm infection of the head region. Since these infections are often confused with other skin disorders, it is, therefore necessary to make early laboratory diagnosis for the better management of the condition. Dermatophytosis are prevalent globally but they are common in tropics and may reach epidemic proportions in geographical areas with higher humidity¹. This can be also due to overpopulation and poor hygienic living conditions, urbanization, shared accommodation such as living in hostels, the use of occlusive footwear, tight-fitting clothes, community showers, and sports activities. The hot and humid climate of India makes dermatophytosis a very common superficial fungal infection of the skin. The distribution, frequency, and causative agents involved vary from place to place depending upon the climatic, socioeconomic conditions, and population density, age, climatic variations⁵. Remissions and exacerbations, and chronicity mark in the course of the disease if left untreated, and remains as a challenge⁶.

With an increasing armamentarium of drugs potentially available for the treatment of dermatophytosis, there is the need for a reference *in vitro* susceptibility test method and the evaluation of more clinical isolates using different antifungal evaluation techniques. They are most often treated with topical antifungal drugs such as ketoconazole, terbinafine which exhibit *in-vitro* antifungal activity. However the most severe dermatophytosis tinea capitis, and tinea unguium, often require the administration of systemic antifungal drugs such as griseofulvin, terbinafine, and

itraconazole³. Some new systemic antifungal drugs such as Posaconazole also appear to be effective *in-vitro* against dermatophytes and are currently being evaluated in the treatment of dermatophytosis. Treatment of dermatophyte infections is usually based on clinical presentation, fungal element detection, and identification of the causative agents. Only a few reports have addressed drug resistance mechanisms in dermatophytes, and most of these were described for *Trichophyton rubrum* such as modifications of target enzymes in squalene epoxidase leading to resistance to terbinafine.

This study was carried out to find out prevalence of dermatophytes infection as well as prevalence of drug resistance to fungal isolates. For Antifungal susceptibility testing, the agar-based disk diffusion susceptibility method for dermatophytes is being the focus of interest of many researchers, since its simple, and inexpensive and does not require specialized training⁷.

MATERIALS & METHODS

This is a Prospective study conducted over 24 months in the Department of Microbiology in association with Department of Dermatology, venerology & Leprosy (DVL), in a tertiary care hospital. The sample size was 100 patients who are clinically suspected cases of Dermatophytosis attending Dermatology, venerology & Leprosy (DVL) OPD. The subjects fulfilling the inclusion criteria of the study were included in the study. Patients of all age groups and both sexes having suspected Dermatophytosis were included in the study. HIV patients, and those who are already diagnosed with Dermatophytosis were excluded. Demographic data and history were obtained from the patient. Patients were explained about the study procedure. Ethical clearance for the study was obtained from the Institutional Ethics Committee prior to commencement. Informed written consent was taken from all study participants or their legal guardians, and all procedures were conducted in accordance with the principles of the Declaration of Helsinki. In the form, the patient has given consent for his/her images and other clinical information to be reported.

The clinical conditions were diagnosed by the clinician himself while examining the patients and the same were processed. Relevant samples like skin scrapings, nail clippings, hair stubs from the scalp were collected from those who are clinically diagnosed with Dermatophytosis.

The infected area is cleaned with 70% alcohol, and allowed to dry. Skin was scraped from active margins of lesions with the help of a sterile blunt scalpel, nails were collected by clipping and hairs were plucked with sterile forceps, collected in sterile brown paper packets and labelled. One part of the sample was directly observed under microscope by potassium hydroxide (KOH) mount. Another part of the sample was inoculated on Sabouraud's Dextrose Agar (SDA) with chloramphenicol (0.05mg/ml) and cycloheximide (0.5mg/ml) and also on Dermatophyte Test medium (DTM).

The information about the applications of antifungal therapy was obtained through an inquiry from the patients and outpatient chart if any treatment was taken during the past 2-3 months. Also, the other information regarding immunosuppressive/immunocompromised state including diabetes and other hormonal diseases was recorded. In addition, the age and sex of infected patients were noted down. The samples were transported to and processed at the Microbiology laboratory. A small quantity of sample (skin, hair) was placed in a drop of 10% KOH solution on a clean glass slide, preparation was kept aside for 30 minutes, then observed microscopically (40X) for presence of fungal

elements. Nail samples were placed in 20% KOH solution with 40% Dimethyl sulfoxide (DMSO) and observed under high power of microscope. In case of very thick nail specimens, preparation was kept in a moist chamber and observed next day.

The samples (skin, hair, nails) were cut into small pieces approximately 1- 2mm in size and incubated on SDA slants with chloramphenicol and cycloheximide irrespective of their KOH positivity and incubated at 25°C for 4 weeks. Tubes were observed for growth at least twice during first week, and once a week thereafter, for a total of 4 weeks. Colony morphology and rate of growth, on obverse and pigment production on reverse of the test tube were observed. If there was no growth in the test tubes after four weeks, it was taken as negative for fungal growth. The growth obtained were examined microscopically by Lactophenol cotton blue (LPCB) mount. Size, shape, arrangement of microconidia and macroconidia, type of hyphae, any special structures like favic chandelier, spiral hyphae etc were noted, in order to identify the dermatophytes. Other tests like Slide culture technique, Dermatophyte test media (DTM), Hair perforation test, Biochemical tests like urease test were performed when necessary.

Dermatophyte Test Medium (DTM) is useful to isolate and distinguish dermatophytes from fungal and bacterial contaminants. DTM was equilibrated to room temperature prior to sample inoculation and made sure that the agar surface is dry. Placed the sample centrally on the surface of the medium and pressed it gently to ensure firm contact. Allowed the cap on the tube to remain loose to ensure gaseous exchange during incubation. Incubated at 25°C for up to 2 weeks in ambient air. The colour change from yellow to red indicates the growth of dermatophytes. The release of alkaline metabolites raises the pH and changes the colour of phenol red in the medium. But there are more chances of false-positive and false-negative results.

Physiological tests for species identification

- a) Urease test – This test is done on Christensen's urea agar medium for the distinction between *Trichophyton rubrum* and *Trichophyton mentagrophytes*. Urease produced by *Trichophyton mentagrophytes* splits urea into ammonia which raises the P^H . This changes the colour of media from amber to pinkish red due to the phenol red indicator.
- b) In vitro hair perforation test –
 - Hair cut in to short pieces of approximate size 1cm are kept in sterile distilled water in a suitable vial inoculated with small fragment of test fungus. Incubated at room temperature. Individual hairs are removed at intervals up to 4 weeks and examined microscopically in Lactophenol cotton blue. The test is taken as positive when the patient shows wedge-shaped perforations in the hair. It is positive in *Trichophyton mentagrophytes* with localised area of pitting and marked erosion and negative in *Trichophyton rubrum*.

ANTIFUNGAL SUSCEPTIBILITY TESTING

Recently CLSI (Clinical and Laboratory standards institute) has approved a microbroth dilution methods for antifungal susceptibility testing of moulds but these tests are difficult to be performed

routinelaboratorysetup. The agar-based disk diffusion is an easy method to determine the antifungal susceptibility of dermatophytes. The application of *invitro* antifungal susceptibility testing for guidance of antifungal therapy has been limited dueto uncertain correlation between *invitro* and *in vivo* action of the drugs.

Antifungal susceptibility testing of the isolated dermatophytes was performed by agar-based disc diffusion method for 5 antifungal drugs. The antifungal disc was procured in readymade form. They are as follows: Ketoconazole (KT), Fluconazole (FLC), Itraconazole (IT), Nystatin (NS). Griseofulvin commercially available in powdered form not readymade as disk and stock solution was prepared in dimethyl sulfoxide as 1.25 mg/ml. Blank discs of 6mm were loaded with 20 microliters of prepared stock solution to obtain the desireddrugconcentrationperdisc.TheisolatesweresubculturedonPotato Dextrose agar (PDA) at 28°C for 7 days to enhance sporulation. The growth was harvested in sterile saline and the suspension was adjusted to 1×10^6 / ml. Plates of Muller Hinton Agar (MHA) plate of 4mm depth and Sabouraud’s Dextrose agar (SDA) were inoculated. The surface of the plates was streaked in 4 different directions(90degree) to cover the entire surface, then kept aside for few minutes for drying and discs were applied using sterile forceps. The plates were incubated for 5-10 days. After sufficient growth occurred, the diameters of zones of inhibition surrounding the antifungal discs were measured and results interpreted. The zone of inhibition varied from 10-32 mmfor Fluconazole,17-36mmforItraconazole,21-22.3mmforketoconazole, nystatin 1.04 ± 20 mm, Griseofulvin with mean \pm SD of 22.6 ± 4.2 , 27.3 ± 6.2 , 32 ± 6.1 and 35.9 ± 4.9 respectively.Statistical analysiswasdoneby Regression analysis.

RESULTS

TABLENo:1.PREVALENCEOFDERMATOPHYTOSIS

TOTALSAMPLESCOLLECTED	TOTALCULTUREPOSITIVE
100	52

TabledepictsthePrevalenceofdermatophytosisinthepresentstudy.Thestudygroupconsistsof100patient samples,collectedfrompatientswho were clinically diagnosed with dermatophytosis.Outofthetotal100 samples,52wereculturepositiveshowingthePrevalence of dermatophytosis as 52% in the present study.

AGE	MALES (n=37)	PREVALENCE (%)	FEMALES (n=15)	PREVALENCE (%)	TOTAL (n =52)	PREVALENCE (%)
1-15 years	00	00	00	00	00	00
16-31 years	21	56.7%	07	46.6%	28	53.8

32-47 Years	03	8.1%	04	26.6%	07	13.4
48-63 years	11	29.7%	03	20%	14	26.9
64- 79 years	02	5.5%	01	6.8%	03	5.9
TOTAL	37	100	15	100	52	100

TABLE NO 2: AGEWISE DISTRIBUTION

Table no. 2 depicts the Age-wise distribution of 52 culture Positive cases. A high prevalence of dermatophytosis was observed in the age group 16-31yrs (53.8%) followed by 26.9% in 48-63yrs and 13.4% in 32-47years, 5.9% in 64-79years. Mean age group is 35.79 years.

TABLE No.3. GENDER-WISE DISTRIBUTION OF CASES

TOTAL	MALES	PREVALENCE IN MALES	FEMALES	PREVALENCE IN FEMALES	162 163 164 165 166 167 168 169 170 171 172 173 174 175
n = 52	37	71.1%	15	28.9%	

Table no.3 depicts the Gender-wise distribution of 52 culture positive cases. Out of the total 52 culture positive samples, 37 were males (71.1%) & 15 were females (28.9%). Males outnumbered females in this study. MALE: FEMALE ratio = 2.4:1

TABLE NO 4: DISTRIBUTION OF CASES ACCORDING TO SOCIO-ECONOMIC STATUS

ECONOMIC STATUS	TOTAL NO OF CULTURE POSITIVES (52)	PERCENTAGE (%)

LOW SOCIO-ECONOMICSTATUS	36	69.2
MIDDLEECONOMIC STATUS	16	30.8
HIGHECONOMIC STATUS	00	00
TOTAL	52	100

UNDER PEER REVIEW IN IJAR

Table No 4 depicts the distribution of cases according to the socio-economic status. Out of the total 52 culture positive samples, 36 cases belong to Low- socioeconomic status, 16 belongs to Middle socio-economic status .

TABLE NO:5.ASSOCIATED SYSTEMIC DISORDERS

SL.NO	SYSTEMIC DISORDERS	No of Patients in the total Culture Positive Samples	Prevalence (%)
1	Diabetes Mellitus	18	34
2	Hypothyroidism/Thyroid disorders	10	18
3	Hypertension	08	16
4	Bronchial Asthma	07	13
5	Chronic Obstructive Pulmonary Disease	06	12.5
6	Chronic Kidney Disease	03	6.5

OBSERVED IN DERMATOPHYTOSIS

Table No 5 depicts the co-morbidities associated with Dermatophytosis. The most common co-morbidity associated with dermatophytosis is Diabetes Mellitus, followed by Thyroid disorders. There is also an association of Hypertension, Bronchial asthma, Chronic Obstructive Pulmonary disease and chronic kidney disease. A few cases are associated with more than one co-

morbidity.

TABLENO:6.RISKFACTORASSESSSED

RISKFACTOR	TOTALNUMBER
Changeundergarmentsdaily	46
Sharingof towel	45
Bath daily	44
Tightclothes	31
Sharingof clothes	24
Sharingof footwear	21
Seasonalexacerbation	21
Familyhistoryofdermatophytosis	16

TableNo6depictstheriskfactorsassociatedwith Dermatophytosis.Theunhygienicconditionsareclosely associatedwithdermatophytosis.Maintenanceofpersonalhygieneisessentialtopreventthecondition.

TABLENO:7.MORPHOLOGYOFLESIONS

TYPEOFLESION	PERCENTAGE
Classicallesion	91.5%
Pustular	3
Eczematous	2
Bullous	2
Pityriasisrosealike	0.9
Papulosquamous	0.6

TableNo7depictsthevariousMorphologyoflesions.ThemostcommonlesionobservedisClassical typelesion91.5%,followedby pustular 3%, Eczematous 2%, Bullous 2%.

❖ **TABLENO:8.TOTALBODYSURFACEAREAINVOLVED**

TOTALBODYSURFACEAREAINVOLVED(%)	PERCENTAGE
1-5	78.8
5-10	12.4

>10	8.8
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Table No 8 depicts the total body surface area involved in dermatophytosis. 1-5 percentage body surface area involved in 78.8% and in 12.4%, 5-10 percentage body surface area involved, whereas in 8.8% more than 10 percentage body surface area involved.

❖ **TABLE NO:9. VARIOUS TYPES OF SAMPLES**

COLLECTED

TYPE OF SAMPLE	TOTAL NUMBER OF SAMPLES COLLECTED	TOTAL CULTURE POSITIVE
SKIN LESION	86	49
HAIR STUBS	03	02
NAIL	11	01

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TableNo9depictsthevarioustypeofsamples collected.Outoftotal100samples,86wereskinlesions,03werehairstubs,11were nail. Out of the total 86 skin lesions, culture was positive in 49, culture was positive in 02 out of 03 hair stubs, and 01 out of total 11 nail samples were positive.

❖ **TABLENO:10.DISTRIBUTIONOFCLINICALTYPESOF
DERMATOPHYTOSISINTHETOTALCULTURE
POSITIVE SAMPLES**

SL.NO	Clinicaltype	Males	Females	Prevalence (%)
1	Tinea corporis	28	12	76
2	Tineacruris	03	02	10
3	Tineafaciei	02	01	06
4	Tineacruris+ Tinea corporis	02	00	04
5	Tineacapitis	02	00	04

The tableNo10depicts,thedistributionofclinicaltypeof dermatophytes in the 52 culture positive samples.ThemostcommonclinicaltypeisTineacorporis(76%),andTinea cruris (10%).TineacrurisandTineacorporisaretogetherfoundinfew patients.

TABLENO:10a.DISTRIBUTIONOFTINEACORPORISIN VARIOUS PARTS OF BODY

SL.NO	Tineacorporis=40	Number	PERCENTAGE
1	Trunk	17	42.5
2	Neck	11	27.0
3	Arms	10	25.5
4	Legs	02	5.0

TableNo10depictsthedistributionofTineacorporisinvariousbody parts.

- Themostcommondistributionisaroundtrunk-17,followedbyneck,arms, legs.

❖ **TABLENO:11.KOHMOUNT&CULTUREGROWTHCOMPARIS ON**

TOTAL KOH MOUNT POSITIVE: 49

TOTALKOHMOUNTNEGATIVE:51

KOHMount	CULTURE	TOTAL
NEGATIVE	NEGATIVE	48
NEGATIVE	POSITIVE	03
POSITIVE	NEGATIVE	00
POSITIVE	POSITIVE	49

TableNo11depictscomparisonofKOHmountwithculture growth.

- 49sampleswereKOHmountandculture positive.
- 48sampleswereKOHmountandculturenegative,whereas03sampleswere KOH mount negative and Culture Positive.

TABLENO:11a.PERCENTAGEOFDERMATOPHYTES

ISOLATEDINTHESTUDYPOPULATION

TOTAL SAMPLES PROCESSED	TOTALKOH POSITIVE	PERCENTAGE OF KOH POSITIVITY	TOTAL CULTURE POSITIVE	PERCENTAGE OF CULTURE POSITIVITY
100	49	49%	52	52%

TableNo11adepictspercentageofdermatophytesinKOHmountand culture.InKOHmountthepercentageis49%,andinculturethepercentageis 52%.

❖ **TABLENO:12.DERMATOPHYTESISOLATEDINTHE CULTURESAMPLE**

SL.NO	Agent	NoofSamples positive	Prevalence(%)
1	<i>Trichophytonrubrum</i>	28	53.8
2	<i>Trichophytonmentagrophytes</i>	14	26.9
3	<i>Trichophyton tonsurans</i>	5	9.6
4	<i>Microsporumgypseum</i>	3	5.9
5	<i>Epidermophytonfloccosum</i>	2	3.8
TOTAL		52	100

TableNo12depictsthevariousdermatophytesisolatedinclinicalsamples.

- *Trichophytonrubrum* wasthemostcommonagent-28, followedby *Trichophytonmentagrophytes*-14.

**TABLENO:13a.PERCENTAGEOFTRICHOPHYTONRUBRUM
ISOLATEDWITHCLINICALSAMPLES**

CLINICAL TYPES	Prevalencein %	<i>Trichophyton rubrum</i>	Total Prevalence (%)
Tineacorporis	76	22	28.9%
Tineacruris	10	04	40%
Tineafaciei	6	01	16.6%
Tineacruris+Tinea corporis	4	01	25%
Tineacapitis	4	00	0%

Table no 13a depicts the percentage of trichophyton rubrum isolated among the clinical samples.

- The table shows the prevalence of trichophyton rubrum in Tinea corporis as 28.9%, Tinea cruris as 40%, Tinea faciei as 16.6%, Tinea cruris + Tinea corporis as 25%, Tinea capitis as 0%.

**TABLENO:13b.PERCENTAGEOFTRICHOPHYTONMENTAGROPHYTESISOLATEDWITH
CLINICAL SAMPLES**

CLINICAL TYPES	Prevalencein %	<i>Trichophyton mentagrophytes</i>	Total Prevalence (%)
Tineacorporis	76	11	14.4%
Tineacruris	10	03	30%
Tineafaciei	6	00	0%
Tineacruris+Tinea corporis	4	01	25%
Tineacapitis	4	00	0%

- Tableno 13b depicts the percentage of trichophyton mentagrophytes isolated among the clinical samples
- The table shows the prevalence of trichophyton mentagrophytes in Tinea corporis as 14.4%, Tinea cruris as 30%, Tinea faciei as 0%, Tinea cruris + Tinea corporis as 25%, Tinea capitis as 0%.

**TABLENO:13c.PERCENTAGEOFTRICHOPHYTON TONSURANS
ISOLATEDWITHCLINICALSAMPLES**

CLINICAL TYPES	Prevalence in %	<i>Trichophyton tonsurans</i>	Total Prevalence (%)
Tinea corporis	76	04	5.2%
Tinea cruris	10	00	0%
Tinea faciei	6	00	0%
Tinea cruris+Tinea corporis	4	00	0%
Tinea capitis	4	01	25%

Table 13c depicts the percentage of *Trichophyton tonsurans* isolated among the clinical samples

- The table shows the prevalence of *Trichophyton tonsurans* in Tinea corporis as 5.2%, Tinea cruris as 0%, Tinea faciei as 0%, Tinea cruris + Tinea corporis as 0%, Tinea capitis as 25%.

TABLE NO: 13d. PERCENTAGE OF MICROSPORUM GYPSEUM ISOLATED WITH CLINICAL SAMPLES

CLINICAL TYPES	Prevalence in %	<i>Microsporum gypseum</i>	Total Prevalence (%)
Tinea corporis	76	02	2.6%
Tinea cruris	10	00	0%
Tinea faciei	6	00	0%
Tinea cruris+Tinea corporis	4	00	0%
Tinea capitis	4	01	25%

- Table 13d depicts the percentage of *Microsporum gypseum* isolated among the clinical samples
- The table shows the prevalence of *Microsporum gypseum* in Tinea corporis as 2.6%, Tinea cruris as 0%, Tinea faciei as 0%, Tinea cruris + Tinea corporis as 0%, Tinea capitis as 25%.

**TABLENO:13e.PERCENTAGEOFEPIDERMOPHYTON
FLOCCOSUM ISOLATED WITH CLINICAL
SAMPLES**

CLINICAL TYPES	PREVALENCE in (%)	<i>Epidermophyton floccosum</i>	Total Prevalence (%)
Tineacorporis	76	02	2.6%
Tineacruris	10	00	0%
Tineafaciei	6	00	0%
Tineacruris+ Tineacorporis	3	00	0%
Tineacapitis	3	00	0%

Tableno13edepictsthepercentageofEpidermophytonfloccosum isolatedamong the clinical samples

- ThetableshowstheprevalenceofEpidermophytonfloccosuminTinea corporisas2.6%,Tineacrurisas0%,Tineafacieias0%,Tineacruris +Tineacorporisas%,Tineacapitisas0%.

❖ **TABLENO.14.ANTIFUNGALSUSCEPTIBILITYPATTERN
OFTHE DRUGS IN DISK DIFFUSION TEST**

NAME OF THE DRUG	PERCENTAGE OF SENSITIVITY
Itraconazole	97.04%
Fluconazole	70.3%
Ketoconazole	55.5%
Nystatin	32.98%
Griseofulvin	30.84%

- TableNo14depictstheAntifungalsusceptibilitypatternofvariousdrugsin disk diffusion test.
- MostoftheisolatesweresensitivetoItraconazole-97.04%,followedby Fluconazole - 70.3%.
 - TheleastssensitivedrugwasGriseofulvin– 30.84%

TABLENO.14a.ANTIFUNGALSUSCEPTIBILITYPATTERNOF TRICHOPHYTON RUBRUM

NAMEOFTHEDRUG	<i>Trichophytonrubrum</i> (28)
Fluconazole	89.2%
Ketoconazole	85.7%
Itraconazole	92.4%
Nystatin	67.8%
Griseofulvin	64.2%

- TheabovetableshowsAntifungalsusceptibilitypatternoftrichophyton rubrum.
- Thesensitivityismaximumtoitraconazole(92.4%)andleasttogriseofulvin 64.2%.

TABLENO.14b.ANTIFUNGALSUSCEPTIBILITYPATTERNOF TRICHOPHYTON MENTAGROPHYTES

NAMEOFTHEDRUG	<i>Trichophytonmentagrophytes</i> (14)
Fluconazole	85.7%
Ketoconazole	78.5%
Itraconazole	92.8%
Nystatin	57.1%
Griseofulvin	50%

- TheabovetableshowsAntifungalsusceptibilitypatternoftrichophyton mentagrophytes.
- Thesensitivityismaximumtoitraconazole(92.8%)andleasttogriseofulvin 50%

TABLENO.14c.ANTIFUNGALSUSCEPTIBILITYPATTERNOF TRICHOPHYTON TONSURANS

NAMEOFTHEDRUG	<i>Trichophytontonsurans</i> (5)
Fluconazole	60%
Ketoconazole	80%
Itraconazole	100%

Nystatin	40%
Griseofulvin	40%

- The above table shows Antifungal susceptibility pattern of trichophyton tonsurans
- The sensitivity is maximum to itraconazole (100%) and least to nystatin, griseofulvin 40%.

TABLE NO. 14d. ANTIFUNGAL SUSCEPTIBILITY PATTERN OF MICROSPORUM GYPSEUM

NAME OF THE DRUG	<i>Microsporum gypseum</i> (3)
Fluconazole	66.6%
Ketoconazole	33.3%
Itraconazole	100%
Nystatin	0%
Griseofulvin	0%

- The above table shows Antifungal susceptibility pattern of microsporum gypseum.
 - The sensitivity is maximum to itraconazole (100%) and least sensitivity to nystatin, griseofulvin 0%.

TABLENO.14e.ANTIFUNGALSUSCEPTIBILITYPATTERNOF
EPIDERMOPHYTON FLOCCUSUM

NAMEOFTHEDRUG	<i>Epidermophytonfloccusum(2)</i>
Fluconazole	50%
Ketoconazole	0%
Itraconazole	100%
Nystatin	0%
Griseofulvin	0%

- TheabovetableshowsAntifungalsusceptibilitypatternofEpidermophyton floccosum
- Thesensitivityismaximumtolitraconazole(100%)andleasttonystatin, griseofulvin , ketoconazole 0%

DISCUSSION

Dermatophytosis is a common superficial mycosis causing significant cutaneous morbidity. Itching is severe and disabling lesions on the genital and other areas cause social embarrassment and impair quality of life. The incidence of dermatophytosis is increasing in recent times especially in the young age group. The rising prevalence of dermatophytosis has been attributed to many factors including tropical climate, overcrowding, urbanization, shared accommodation such as living in hostels, use of occlusive footwear, tight-fitting clothes, community showers, and sports activity have been proposed but the exact reason has not been elucidated. The present study shows a prevalence of 52% which is comparable with Sudip das et al (2020, 53.4%). This is related to various factors including seasonal variations, personal hygiene, hot & humidity, family history etc.

The youngest patient was 15 years old and the oldest patient was 72 years old. The prevalence of dermatophytosis is more common in an age group of 16-31 years, this can be due to

- a) Increased physical and sports activities.
- b) Sharing of clothes, footwear.
- c) Increased sweating.
- d) Poor hygienic practices.
- e) The increased incidence of dermatophytosis in this age group 15-30 may be because this population group takes part in maximum outdoor activities such as agriculture and manual labour, which predispose to acquire infection from environmental exposure. However, Bindu *et al* observed a higher prevalence in the age group of 11-20 years.
- f) In the present study, there are an observation of sharing towels and footwear, and clothes in our patients. This could contribute to the spread of infection.
- g) Objects such as clothing, bedsheets, and towel harbour the fungal pathogen and are capable of transmitting the disease among the family members.

In addition, fungal spores remain viable for months in household dust leading to recurrent episodes of clinical diseases. Asymptomatic carriers among family members may also be another cause for recurrence. A third of patients wore tight clothes such as jeans, and woollen undergarments in winters, which were often unwashed for weeks creating a damp environment favourable for the proliferation of

dermatophytes. Dermatophytes are the most commonly implicated etiological agents, particularly *Trichophyton rubrum* and *Trichophyton mentagrophytes* var *interdigitale*, followed by candida species and nondermatophytic moulds in immunocompromised patients. Dermatophytes account for approximately 90% of the toenail, and 50% in the fingernail. Dermatophyte invasion of the nail plate is termed tinea unguium. Diabetes and other immunocompromised conditions make the patients more prone to the disease.

The increased prevalence in males may be due to the occupational hazards related to their nature of work and increased risk of exposure to infections, Furthermore, the lower incidence in females may also be due to the nonreporting of female patients to hospitals due to the prevailing social stigma in the rural population of India.

Direct examination using KOH is still a fast and simple technique for dermatophytes diagnosis. Moreover, KOH results corresponded with reference standard cultures. Non visualization of hyphae on direct microscopy could be due to a severe inflammatory reaction that obscures them. Three samples were KOH negative but culture positive. The limitation of KOH mount testing is that it cannot identify the specific genus and species of fungi. Therefore, standard culture and identification are important to confirm the etiological agents and speciation of dermatophytes.

- Out of the total Tinea corporis, the most common distribution was observed in the trunk followed by the neck, legs, and arms. This could be related to the above-specified reasons such as increased sweating, wearing tight clothes, etc.
- Besides the climatic conditions favorable for the growth of dermatophytes, other factors such as migration of laborers, workers, and tourists frequently visiting this region, overcrowding, unhygienic lifestyle of the community with low socioeconomic background might contribute to the development of dermatophytes.

Trichophyton rubrum grew relatively slower (10-15 days), the growth is powdery to velvety with a reddish tinge on the obverse and rusty brown to deep red on the reverse. Well, septate, pencil-shaped hyphae with numerous spherical microconidia along with macroconidia were visible on microscopic examination. As *Trichophyton rubrum* is a slow-growing organism, there is a possibility that other dermatophytes species might overgrow or mask the growth while attempting isolation.

Rarely do these pathogens cause a more aggressive and invasive form of infection.

that may present as a) Majocchi's granuloma (nodular granulomatous perifolliculitis)

is an infection of dermal and subcutaneous tissue related to disruption of hair follicles and spillage of fungi into the dermis, which produces a granulomatous inflammation. Both

immunocompromised and immunocompetent patients may be affected by this type of

infection b) In deep or invasive disease, invasion is limited mainly to the extremities and there is subcutaneous involvement without the involvement of other internal organs. Patients with this syndrome are usually immunocompromised hosts.

c) In exceptional instances, generalized invasive disseminated infection with dermatophytes has been reported. PCR amplification directly from the samples could be a better tool to prove this. In patients who have a superficial infection with dermatophytes, culture from subcutaneous tissue may be contaminated and thus lead to misdiagnosis of invasive infection. Some authors have suggested that specific T-cell immunity plays a role here, but virulence factors of the specific pathogen may also be important.

- Most clinical types of dermatophytosis respond well to topical antifungal therapy, while Tinea unguium, Tinea capitis and extensive type of dermatophytosis require systemic therapy (Priyam Basak et al, 2019).
- Different dermatophyte strains may have different antifungal susceptibility patterns which may vary geographically also.
- Some strains of dermatophytes resistant to particular antifungal agents have been reported.
- Recently there has been a rise in antifungal resistant strains of fungi. Therefore, early initiation of correct antifungal therapy is essential for proper treatment and prevention of spread of disease.
- This makes testing of the susceptibility of dermatophytes more important which may help in surveillance and epidemiological study of resistant strain. (Prabhat Kiran khatri *et al*, 2017).

CONCLUSION

Superficial fungal infections are an important public health problem, especially dermatophytosis infections. Dermatophytosis is a superficial fungal infection of keratinized tissue of the skin, hair, and nails which are rich in keratin (a predisposing factor and nutritional requirements of filamentous fungi) caused mainly by *Trichophyton*, *Epidermophyton*, *Microsporum*. To cause disease, dermatophytes must adhere to a surface such as epithelial cells, and then obtain nutrients for growth from these cells. Dermatophytes secrete keratinases and other proteases which are thought to play a role during infection, however, the temporal expression of these proteases appears to vary between species. The study aimed to detect the prevalence and identify the fungal cause of Dermatophytosis and evaluate the Antifungal Susceptibility pattern. In the study, the Males were affected more common than females, and the most commonly affected age group was 16-31 years. Unhygienic conditions among the low socio-economic group, frequent migration of laborers, workers may be some of the contributing epidemiological factors for the higher prevalence. Diabetes Mellitus, Thyroid disorders, Hypertension, Bronchial asthma were the most frequent systemic association noted in this study. Other systemic conditions like Chronic obstructive pulmonary disease, chronic kidney disease also observed.

Tinea corporis was the most common clinical pattern observed in the study.

Various tinea conditions in the present study were diagnosed by the clinician itself based on the clinical presentation.

Tinea conditions are a consequence of exhaustive physical work and prolonged exposure to the sun leading to excessive sweating. In addition, the tight fittings and synthetic clothing particularly in males provided damp, sweaty, and warm skin conditions factors that favor the growth of dermatophytes or may exaggerate the disease condition. Tinea pedis and Tinea unguium might result from wearing socks and shoes for a long period providing damp conditions, especially in inter-digital spaces. The most common fungal isolate was *Trichophyton rubrum* with *Trichophyton mentagrophytes* being the second commonest one. Injudicious use of topical steroid application was found in alarming numbers both in OTC and prescribed by practitioners. It has been suggested that treatment for deep invasive infection with *Trichophyton* should include either itraconazole, and in some severe cases, surgical debridement is also required. The most potent antimycotic agent against the dermatophytes was itraconazole in the Antifungal susceptibility testing with disk diffusion and also *Trichophyton rubrum* was

sensitive to itraconazole the most. In comparison to other drugs, Griseofulvin was found to be less active. Topical steroid application can also enhance the penetration of fungus to the dermis which can lead to treatment failure. A high incidence of intrafamilial tinea infections, misuse of corticosteroids containing topical antifungal preparations, poor compliance to treatment, and poor personal hygiene were associated with recurrences and poor responses to treatment. Recurrences may also be due to reinfection from family members or the environment, and most of the patients stop treatment as soon as they get partial relief.

Dermatophyte organisms are likely becoming more virulent with a gradual increase in resistance to commonly used antifungal drugs as a result of various host, environmental, and treatment factors posing a great emerging health hazard to the community.

Although the present study focuses primarily on the prevalence of different dermatophyte species, and its Anti-fungal susceptibility pattern, a more systemic study covering a larger population and over a longer period would give a better insight into the epidemiology

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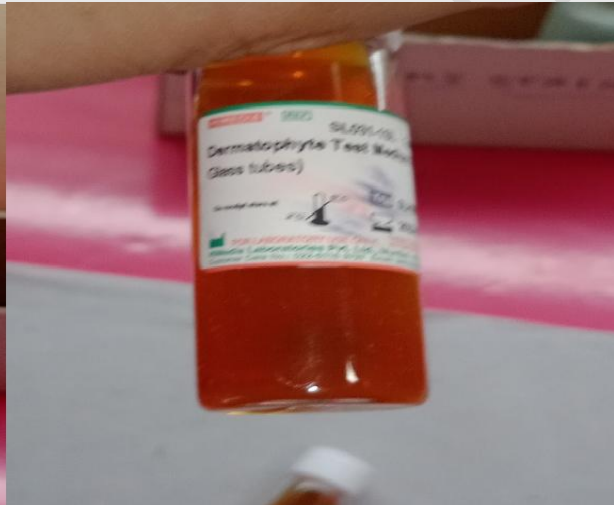
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IMAGES

1.DERMATOPHYTETESTMEDIUM



Trichophyton rubrum



Trichophyton mentagrophytes

2.SDASLANTS-FUNGALCULTUREOF
DERMATOPHYTES

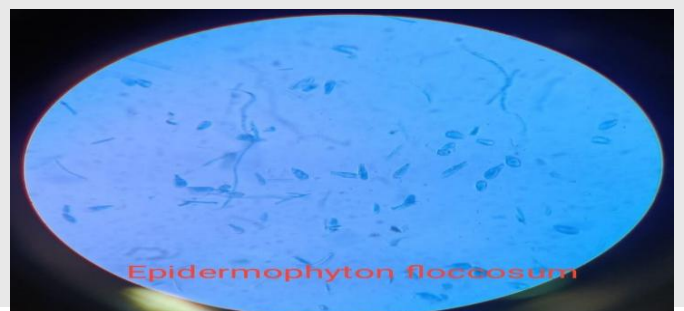
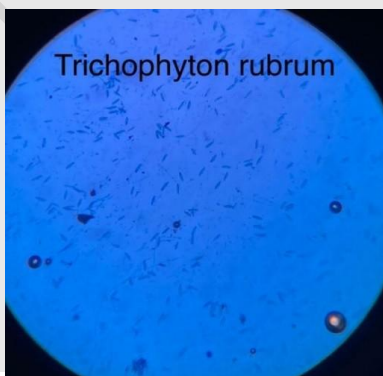


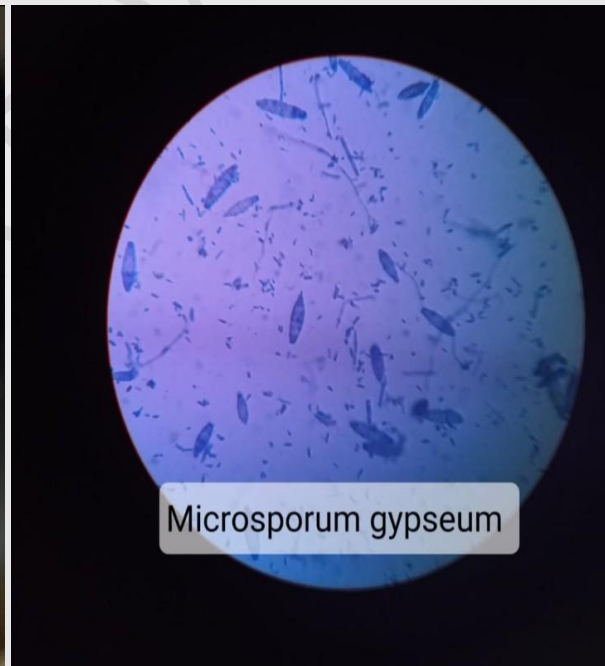
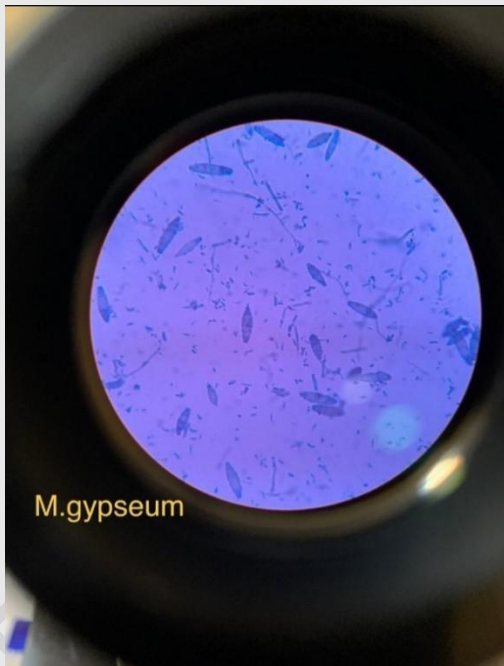
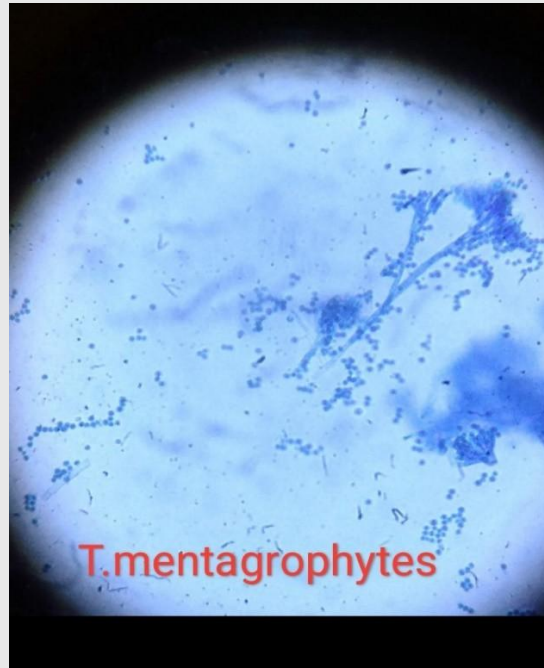
Microsporiumgypseum



Trichophyton tonsurans

3-LPCBMOUNTOFTRICHOPHYTONRUBRUM







4.TINEA CORPORIS



5.TINEA CAPITIS



UNDER PEER REVIEW IN IJAR