



## CLINICO- MYCOLOGICAL STUDY OF DERMATOPHYTOSIS IN A TERTIARY CARE CENTRE

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**Abstract:**

**Background & Objectives:** Dermatophytosis is an emerging problem with increased morbidity in many patients. These infections are seen more in tropical countries. Overcrowding, low socio-economic factors, humidity and poor hygiene are the pre-disposing factors for the fungal infection. Dermatophytosis is a superficial fungal infection which is caused by 40 species of 3 different genera: Trichophyton, Epidermophyton and Microsporum of family Arthrodermataceae. These organisms are keratinophilic and has affinity towards keratin rich structures such as skin, hair and nails and causes keratin degradation. The aim of the study is to identify most common clinical type of dermatophytosis and most common organism causing the disease.

**Methods:** This was a cross-sectional study conducted between March 2021 to September 2022 in the department of Dermatology, venereology and Leprosy of a tertiary care centre. Patients who came under the inclusive criteria were taken into the study after taking informed consent. Detailed history is taken followed by which sample is collected for KOH, Calcofluor and culture from the affected area.

**Results:** In this study, most common age group affected are between 31-40 years with male predominance. Lower socio-economic status people are more affected.

Mixed site involvement is seen in 68.18% individuals with most common site to be affected is neck (55.45%). Co-morbidities such as Diabetes, hypertension and Thyroid is seen in few.

In this study, KOH mount positive was seen in 67.27%, calcofluor white in 84.54% and culture in 51.81% individuals. Most common diagnosis was found to be Tinea corporis with Tinea cruris (42.73%). Most common organism isolated from culture positives were Trichophyton mentagrophytes (24.55%), followed by Trichophyton rubrum (22.73%), Trichophyton tonsurans (2.73%) and Trichophyton violaceum (1.81%).

**Interpretation & Conclusion:** In this study, most common type was tinea corporis with tinea cruris. Most common dermatophyte isolated from the specimens was Trichophyton mentagrophytes.

**Keywords:** Dermatophytosis, tinea corporis, tinea cruris, Trichophyton mentagrophytes, followed by Trichophyton rubrum, Trichophyton tonsurans.

**INTRODUCTION**

Dermatophytosis are a group of superficial fungal infection of keratinized tissues, such as the epidermis, hair, and nails.<sup>1</sup> Dermatophytosis causes 16–75% of all the mycological infections worldwide and severe diseases in immunocompromised patients. Frequency, distribution, and their etiologic agents of dermatophytosis vary based on the age, topography, socioeconomic status, climate, and domestication of animals.<sup>2</sup> It is more prevalent in tropical countries such as India, due to climate & living condition.<sup>3</sup>

Dermatophytosis are included in the category of difficult to treat dermatoses such as psoriasis, vitiligo, pemphigus, and erythroderma. It was previously considered as most trivial condition to which has now become most stubborn disease to treat.<sup>4,5</sup> Quality of life of patients is affected significantly due to its frequent relapses. The complex interplay of host, environment, and agent factors are attributed for the challenging scenario in treatment of the disease which varies as per the topography. The following study aimed to identify the most common causative organism; clinical patterns associated risk factors in this geographic area. Also to evaluate the sensitive method of investigation to find the fungal elements.

**METHODOLOGY:**

It was a cross sectional study of 110 patients attending the Dermatology outpatient of the Tertiary care center for 18 months from March 2021 to September 2022. Clinically suspected cases of dermatophytosis with informed consent were included in the study. A pre structured proforma was used to collect data on history, clinical examination, KOH Mount, SDA Culture & Calcoflour stain. Patients on antifungals for >4 weeks & whose KOH or Cultures showed organisms other than dermatophytes were excluded.

Skin scrapings, Hair strands and Nail clippings were collected and transported to Microbiology lab wrapped in a dark colored paper. **KOH Mount:** sample was seen under microscope for fungal elements by using 10% KOH for Skin, 20% for Hair and 40% for Nails. **Calcofluor white** (AMD labs Bangalore India): sample was stained with Calcofluor white and 10% KOH for one minute and looked for fungal elements in fluorescent microscope.

**Culture (MicroExpress):** The material is inoculated onto two SDA (Sabouraud's dextrose agar) slope with Chloramphenicol and Cycloheximide. One tube incubated in Biological Oxygen Demand (BOD) incubator at 22°C, one tube at Room temperature (37°C) and observed for growth for 4-6 weeks.

**RESULTS:**

Among 110 patients of the study population, mean age group affected was 34.2 years. Males are more affected than females (54.55%) who were daily wage workers from lower socioeconomic strata. [Table 1]. Most common site of infection was Groin (54.54%) followed by buttocks (44.54%). The risk factors associated are poor hygiene & fomites among subjects [Table 2]. Most common clinical variant of dermatophytosis was Tinea corporis with cruris (42.73%). (Figure 1) Commonest organism isolated from the culture was Trichophyton mentagrophytes (24.55%) [Figure 3], followed by Trichophyton rubrum (22.73%) [Figure 4]; Trichophyton tonsurans (2.73%) and Trichophyton violaceum (1.81%) [Figure 5] with average time taken for the culture to grow was around 14 to 21 days. KOH & Calcofluor positivity was seen in 84 (98.82%) whereas KOH & culture positive was seen in 43 (75.43%). We found calcofluor with KOH could pick up faint fungal elements which was missed in culture. (Table 3 & 4) (Figure 2)

Demographic factor	Most involved	Percentage
Age	31-40 years	29.09%
Socioeconomic status	Lower socioeconomic status	50%
Occupation	Daily Wages	32.73%
Duration	Months	62.73%
Symptoms	Itching	97.27%
	Hyperpigmentation	90.90%
	Scaling	89.09%
	Pain	10.09%
	Discharge	9.09%
	Thickening of skin	9.09%
	Hair loss	7.27%
	Burning sensation	6.36%

<b>Risk factors</b>	<b>Frequency (N=110)</b>	<b>Percentage</b>
Past history	Positive: 39	35.45%
Family history	Positive: 41	37.27%
Fomites	Towel: 22 Razor: 22 Soap: 17 Comb: 8 Foot wear: 3	30.55% 30.55% 23.6% 11.11% 4.16%
Personal Hygiene (Frequency of bathing)	Alternate days: 71 Daily: 25 Once in 3 days: 14	64.54% 22.73% 12.72%
Comorbidities	Diabetes: 5 Hypertension: 2 Thyroid: 1 Diabetes with hypertension: 1	4.35% 1.82% 0.91% 0.91%
Pets	Present: 7	6.36%
More than 2 sites involved	75	68.18%

<b>KOH</b>	<b>Calcofluor</b>		<b>Culture</b>	
	<b>Positive</b>	<b>Negative</b>	<b>Positive</b>	<b>Negative</b>
<b>Positive</b>	84 (98.82%)	1 (1.17%)	43 (75.43%)	31 (58.49%)
<b>Negative</b>	12 (48%)	13 (52%)	14 (24.56%)	22 (41.51%)

<b>Culture Report</b>	<b><i>T. mentagrophyte</i></b>	<b><i>T. rubrum</i></b>	<b><i>T. tonsurans</i></b>	<b><i>T. violaceum</i></b>
<b>T. Corporis</b>	7(25.9%)	12(48%)		1(50%)
<b>T. Cruris</b>	1(3.7%)	1(4%)		
<b>Onychomycosis</b>	1(3.7%)	1(4%)		
<b>T. Corporis, T. Cruris</b>	13(48.14%)	10(40%)	3(100%)	
<b>T. Corporis, T. Cruris, T. faciei</b>	2(7.4%)			
<b>T. Corporis, T. Cruris, T. pedis</b>	1(3.7%)			
<b>T. Corporis, T. faciei, T. Barbae</b>		1(4%)		1(50%)
<b>T. Corporis, T. pedis</b>	1(3.7%)			
<b>T. Corporis, T. capitis</b>	1(3.7%)			



Figure 1: Clinical variants of Dermatophytosis

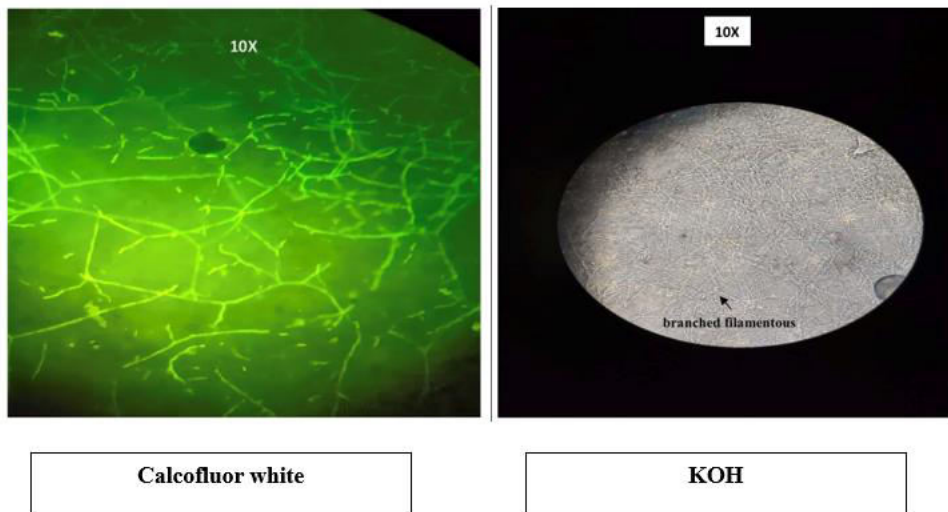


Figure 2: Calcofluor White & KOH in 10X

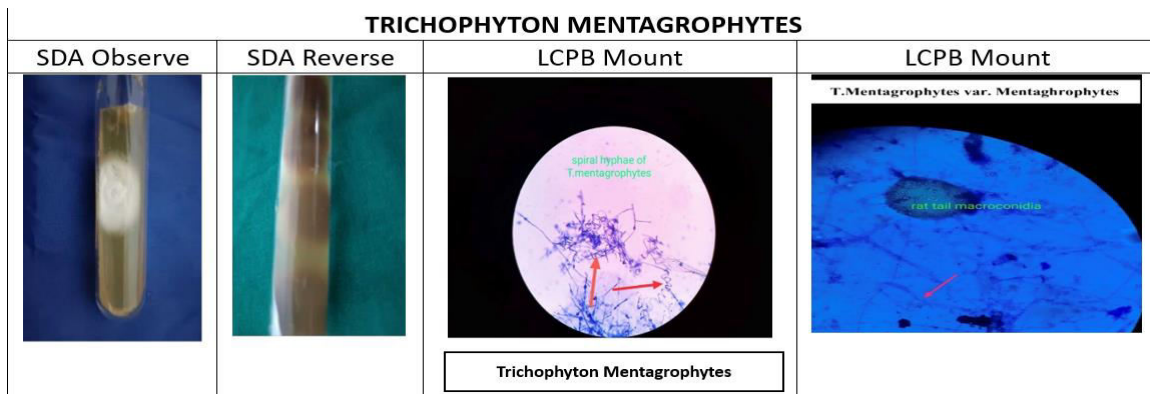


Figure 3: Macro and Microscopic appearance of Trichophyton Mentagrophytes

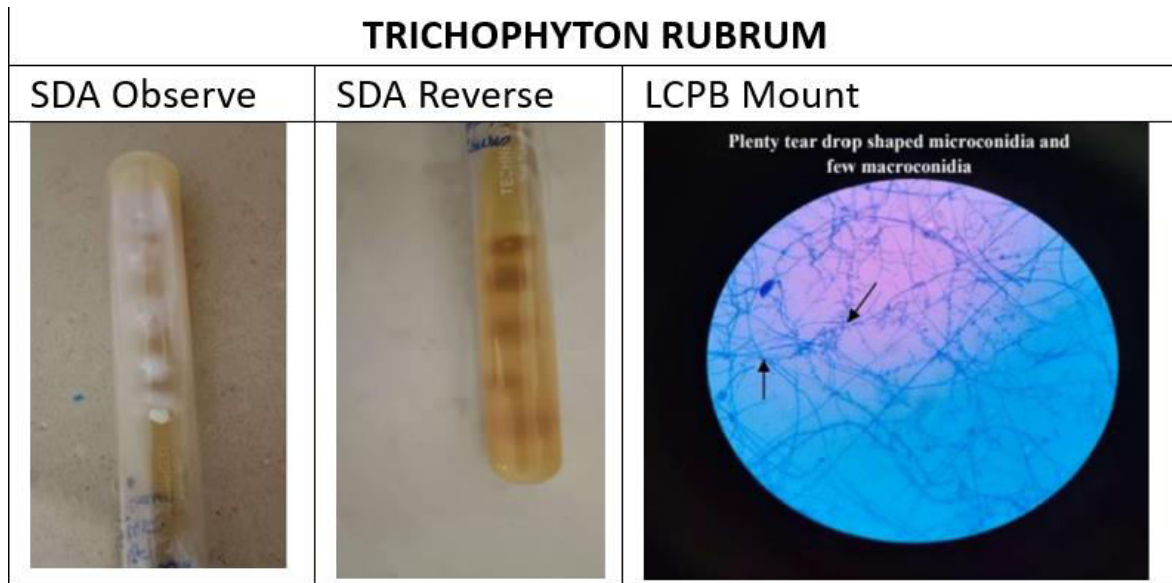


Figure 4 Macro and Microscopic appearance of Trichophyton Rubrum

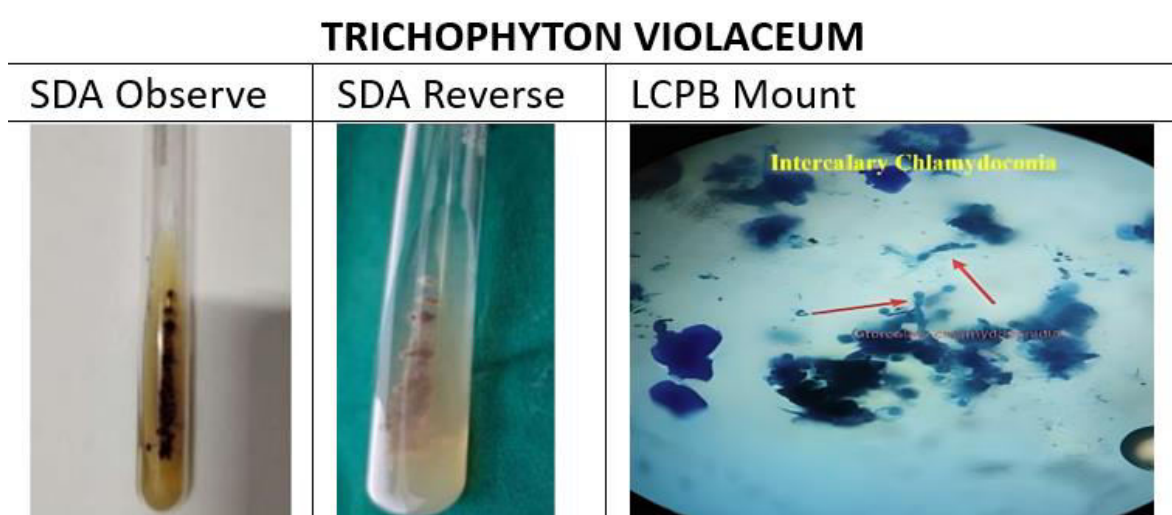


Figure 5: Macro and Microscopic appearance of Trichophyton Violaceum

**DISCUSSION:**

Cutaneous fungal infections constitute a substantial amount of health problems all the world due to antifungal abuse, steroids misuse, and immunocompromised status.<sup>4</sup> Dermatophytosis is one of the most common superficial fungal infections.<sup>5</sup>

In this study, results of patient's age, gender, socioeconomic factors, occupation duration and symptoms were in co-relation with the studies conducted by Shenoy MM et al,<sup>6</sup> Gu D et al<sup>7</sup>. This could be due to overcrowding, poor hygiene, sharing of clothes, nutritional deficiency and lack of awareness on sanitation among low socioeconomic groups.

Risk factors associated with this study are past & family history of similar infection, fomites, poor personal hygiene, diabetes as co-morbidity, pets and sites involved were in co-relation with studies conducted Costa JEF et al<sup>8</sup> and de Macedo GMC et al.<sup>9</sup> This could be linked to their poor living & working condition with lack of awareness on personal hygiene. Changing inner garments regularly, use of tight inner wears, working in hot & humid climate for long durations and use of synthetic

clothing/uniforms.

In this study, Among KOH positivity, Calcofluor white positive and negative were seen in 98.82% and 1.17% respectively. Similarly, Culture positive and negative cases were 75.43% and 24.56% respectively. This study correlates with studies conducted by Sakshi R et al<sup>11</sup> with KOH mount positivity of 69%, Calcofluor white positivity of 80% and Culture positivity of 54% respectively and studies conducted by Das Set al<sup>10</sup> with KOH positivity of 56%, Calcofluor white of 63.33% and culture positivity of 39.33% respectively.

Most common organism isolated from the culture was *Trichophyton mentagrophytes* (24.55%), followed by *Trichophyton rubrum* (22.73%); *Trichophyton tonsurans* (2.73%) and *Trichophyton violaceum* (1.81%) with average time taken for the culture to grow was around 14 to 21 days.

*Trichophyton Mentagrophytes* had macroscopic appearance of cream to yellowish coloured, powdery to floccose colonies on upper side. reverse SDA showed ochre to red brown. Microscopically- few figure shaped 3-8 celled macroconidia with smooth to thin wall spherical shaped sessile microconidia measuring about 2mm diameter, arranged in dense grape like clusters or alongside the hyphae. Frequently spiral hyphae were seen. **[Figure 4]**.

*Trichophyton Rubrum* had macroscopic appearance of white fluffy to cottony colonies sometimes becoming rose when aging; reverse SDA show wine red to olive colour, sometimes yellow. Microscopically few macroconidia seen (Figure shaped). Tear drop shaped microconidia seen alongside of undifferentiated hyphae. **[Figure 5]**.

Macroscopic appearance of *Trichophyton Violaceum* showed slowly growing glabrous, leathery, wrinkled colonies which are purple red in colour. Reverse SDA also showed purple colour. Microscopically, hyphae are highly distorted and have reflexive branching which are strongly septate, disarticulating into arthroconidia. Macroconidia very rare, microconidia ovoida or pyriform. **[Figure 6]**.

Macroscopically *Trichophyton Tonsurans* showed colonies which were mostly suede like which white to yellowish and irregularly furrowed. Reverse SDA shows mahogany-red, yellow or brown colour. Microscopically, abundance of variable size microconidia seen on loosely clustered branches or thickened terminal hyphae. Miroconidia are sessile, clavate to nearly cylindrical, sometimes inflating to balloon shaped. Whereas macroconidia, if present are variable measuring 10-65 x 4-12mm, 2-6 celled with thick walled, cylindrical to cigar shaped. Abundance of terminal an intercalary, swollen chlamyospores are formed.

This study introduces a rapid and valid approach in fungal diagnostics, revealing the efficacy of combining KOH and Calcofluor staining techniques. Our study indicates a significantly higher positivity rate when both KOH & calcofluor methods were used compared to traditional culture & KOH. The observation that Calcofluor with KOH detects faint fungal elements missed by culture emphasis its enhanced sensitivity. This combination of investigations not only expedites diagnosis but also enhances accuracy, potentially reducing diagnostic delays, cost and improving patient outcomes. The study thus offers a refined diagnostic strategy for dermatophytosis.

Strength of this study was that KOH, Calcofluor white and Culture were used for identification of the fungal elements in the sample. Calcofluor white acts as a better stain when compared to KOH in detecting fungal elements. This study is a hospital based cross sectional study which cannot be

generalized to the population. Fluorescent microscope is required for identifying fungal elements through calcofluor white which is available only in tertiary health care centers. In future, Anti-fungal resistance tests to be included in the panel of tests done for dermatophytosis along with KOH & Culture to reduce the development of new antifungal resistance and better management.

Interpretation of this study was that most of the study participants were daily wagers who do not maintain personal hygiene. Most of the patients were having Tinea corporis with cruris among people who work in humid climates and maintain poor hygiene. The most common organism isolated from this topography was *Trichophyton mentagrophytes*.

### Conclusion:

The study highlights Tinea corporis and Tinea cruris as the predominant clinical variant, with *Trichophyton mentagrophytes* emerges as the most frequently isolated organism, emphasizing its significance rampant & resistant dermatophytosis in this topography. Furthermore, the findings suggest Calcofluor staining as a superior method for microscopy in diagnosing dermatophytosis compared to KOH, potentially due to its enhanced sensitivity in detecting fungal elements. These observations provide valuable insights into the epidemiology and diagnostic methods of dermatophytosis but also emphasize the importance of accurate and efficient diagnostic techniques in clinical practice. This contributes to understanding of dermatophytosis & associated risk factors aiding in improved management of these infections.

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