



CLINICOMYCOLOGICAL STUDY OF VARIOUS DERMATOPHYTIC INFECTIONS ATTENDING DERMATOLOGY OP IN A TERTIARY CARE HOSPITAL, KANCHIPURAM, WITH SPECIAL MENTION ON SIGNIFICANCE OF PROPER SAMPLE COLLECTION FOR THEIR DEFINITIVE LABORATORY DIAGNOSIS

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ABSTRACT

Dermatophytic infections are worldwide in distribution. They often present with superficial skin infections. Severe infections suggest underlying immunodeficiency state of particular clinical case. They are usually caused by anthropophilic dermatophyte species such as Trichophyton, Microsporum, Epidermophyton. Common age group being 30-40 and males are usually infected. Certain infections are commonly seen in paediatric age group. (T. capitis). The prevalence and distribution of species vary in various geographical areas. Following guidelines for appropriate collection of sample, clinical specimen is collected and 10-20% KOH mount preparation of skin scrapings, hair follicles, nail clippings are examined under microscope for fungal elements, followed by inoculation on to SDA and special dermatophyte media for culture confirmation. Results are interpreted based on colony morphology, microscopic identification of microconidia and macroconidia. The most commonest dermatophyte isolated being T. rubrum, T. mentagrophytes followed by T. tonsurans, M. audouinii. Most of the dermatophytic infections are diagnosed and treated depending on clinical manifestations and 10-20% KOH mount. Dermatophytic infections that need prolonged antifungal therapy and those with atypical clinical presentation need confirmation by culture as it takes long time for their isolation. Proper identification needs appropriate sample collection. Among superficial skin infections dermatophytic infections are the most common. As their infection results in disfigurement and discomfort and significant morbidity, avoiding contact with infected animals and persons as well as maintaining good environmental, personal hygiene can decrease dermatophytic infections.

KEYWORDS: *Dermatophyte, Trichophyton, Epidermophyton, Microsporum, sample collection, SDA, 10-20% KOH, disfigurement*



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INTRODUCTION

Dermatophytes are ubiquitously distributed¹ eukaryotic fungi, confined to superficial keratin layers²⁾ of living hosts that rarely give rise to serious consequences, however for considerable disfigurement and discomfort as well as their contagious nature makes them of much human concern.³ Further severe infections or those refractory to treatment represent underlying immunodeficiency.⁴ Dermatophytes eventhough are categorized into anthropophilic (humans), zoophilic (animals) and geophilic (soil) that are usually caused by the three species Trichophyton, Microsporum, Epidermophyton,⁵ human infections (skin, hair, nail) are caused by all the three varieties. The dermatophytic infections are commonest superficial skin infections.⁵ They exist in Anamorphic forms (asexual) with microconidia and macroconidia and sexual forms (telomorphic forms)⁶ Usually anamorphic forms are seen in laboratory cultivation. Eventhough there is rapid evolution in molecular diagnostic techniques, microscopy and cultivation for species identification are still conventional confirmatory diagnostic methods. The clinical presentation usually called ring worm infections consists of Tinea corporis (body), Tinea capitis (scalp), Tinea cruris (buttocks), Tinea barbae (beard), Tinea pedis (foot), Tinea incognito (atypical), Tinea unguium (nails) etc. They present with different clinical lesions at different sites. The collection of possible appropriate sample and its transportation for performing laboratory tests and identification is essential prerequisite⁶ The present study includes careful collection of ample sample from the infected site using sterile scalpel, taking universal precautions and their transportation in a square shaped brown coloured dry paper for performing 10-20% KOH mount (the squamous cells are destroyed keeping the fungal elements intact) as well as cultivation using special dermatophyte media and after 2 weeks of incubation microscopically examined for species identification.

MATERIALS & METHODS

*Specimen collection guidelines*⁶

The following guidelines are observed for collecting the sample. 1. Sample is collected by wearing sterile surgical gloves and face mask. 2. Active growing margin of lesion is identified for skin lesions, by holding the sterile surgical scalpel vertically to the surface ample material is collected into brown coloured square shaped paper that is properly labelled with patients details including age, sex, place, travel history, animal exposure and transported to laboratory. 3. For scalp infections infected hair follicle taking care to preserve the root is collected, for few cases the hair is brushed using a sterile tooth brush to collect the sample. 4. For nail infections the material from underside of the proximal portion of the nail is collected.

10 -20% KOH mount

The material (skin scrapings, infected nails, infected hair with intact root) is placed in 10 – 20% KOH preparation for few minutes. For nail sample 20% KOH is used and the sample in 20% KOH is warmed under Bunsen flame before being examined. In present study only one case of T. manuum was sent for dermatophyte culture.

Dermatophyte medium

Dermatophyte medium composition was of papaic digest of soybean meal, dextrose, cyclohexamide, phenol red, chloramphenicol, Agar at final Ph 5.6 +/- 0.2 and the media is poured on to a petridish /test tube. The inoculated media is incubated at 25 degree C. Observed for growth after 24 hours every day upto 7 days. Interpretation of results : Red colour around the colony suggests positive for dermatophyte and growth without colour change indicates non-dermatophyte.

RESULTS

In our present study all clinically diagnosed cases of superficial mycotic infections starting from October 2014 to August 2015 attending a dermatology OP in a tertiary care hospital were included. Among 220 clinically diagnosed cases of superficial mycotic infections 132 cases were as dermatophytic infections. Rest of them (88 cases) were due to various other superficial mycotic infections such as P. versicolor, P. rosea, Piedra etc., that are included under exclusion criteria in our study. (table 1 & chart 1)

Table 1

	Number of cases	Percentage
Clinically diagnosed superficial mycotic infections due to dermatophytes	132	60%
Other infections such as P. versicolor, P. rosea, Piedra etc	88	40%
Total number of cases	220	100%

FIGURE - 1

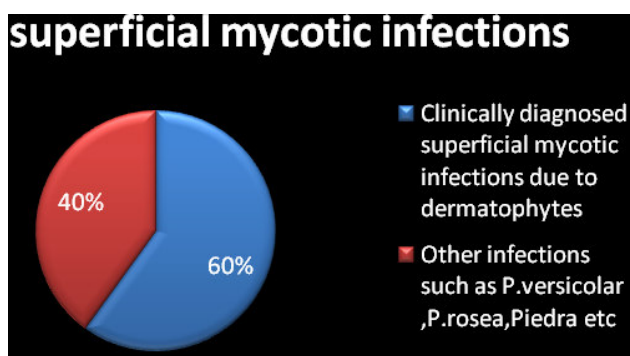


FIGURE 2
Sex distribution of different clinical types of dermatophytes

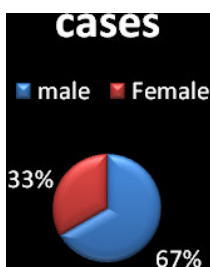
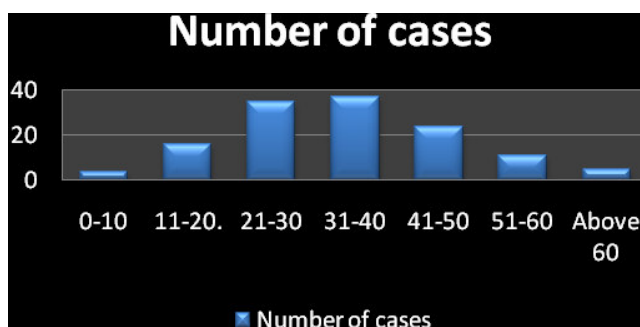


Table 2
Age distribution of various clinically diagnosed dermatophytic infections

s.no	Age	Number of cases
1.	0-10	4(3.03%)
2.	11-20	16(12.12%)
3.	21-30	35(26.51%)
4.	31 – 40	37(28.03%)
5.	41 – 50	24(18.18%)
6.	51 – 60	11(8.3%)
7.	Above 60	5 (3.7%)
Total		132

Figure3
Age Distribution of clinically Diagnosed Dermatophytic Infections



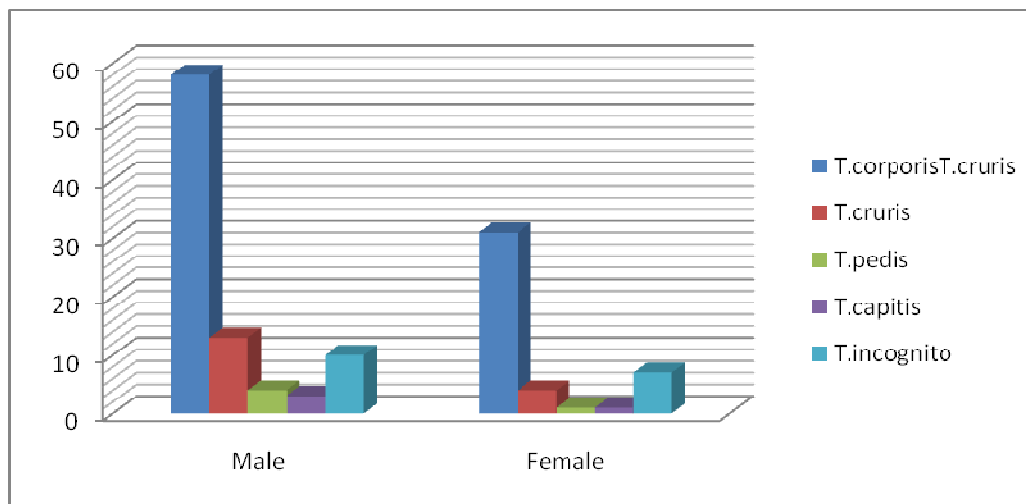
Out of 220 cases 132 cases were due to different dermatophytic infections and 88 cases were due to Pytiriasis versicolor. In present study only dermatophytic infections are included. Among 132 cases 88 (66.6%) cases were male patients and 44(33.3%) cases were female patients. (Figure 2)

Table-3
Sex distribution of different clinical types of dermatophytes

Clinical types	Male	Female	Total
T.corporis	58	31	88
T.cruris	13	4	17
T.pedis	4	1	5
T.capitis	3	1	4
T.incognito	10	7	17
Total			132

Tinea corporis infections were found to be the most common

FIGURE 4
Sex distribution of different clinical types of dermatophytes

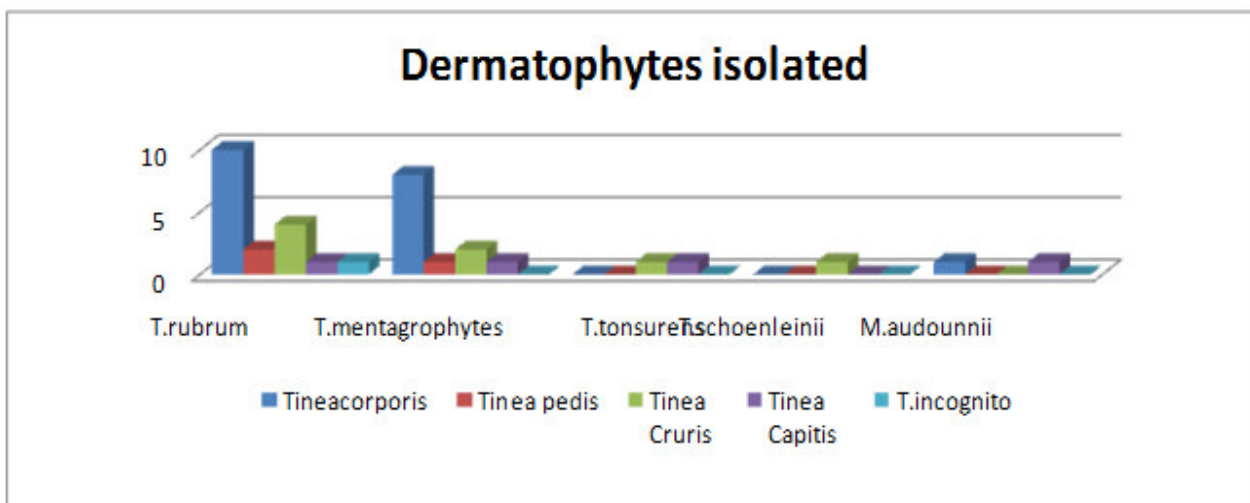


Out of 46 samples collected upon request from dermatologist for 10% KOH mount and fungal culture 35 samples (76%) have given rise to different fungal isolates that included *Trichophyton rubrum*, *T. mentagrophytes*, *T. schoenleinii*, *M. audouinii*, *M. canis*. (Figure 4, Table 5) 11 samples were culture negative. (24%). Among culture negative cases many were from cases of *T. incognito*. out of 46 samples 25 samples (54.34%) showed the presence of fungal elements. 21 samples (45.66%) were negative for 10% KOH mount (Table 6)

Table 5
Number of Dermatophyte Isolated From Different Clinical Types

Dermatophytes isolated	Tinea corporis	Tinea pedis	Tinea Cruris	Tinea Capitis	T.incognito	Total
<i>T. rubrum</i>	10	2	4	1	1	18(51.45%)
<i>T. mentagrophytes</i>	8	1	2	1	-	12(34.28%)
<i>T. tonsurans</i>	-	0	1	1	-	2(5.7%)
<i>T. schoenleinii</i>	-	0	1	-	-	1(2.8%)
<i>M. audouinii</i> , <i>M. canis</i>	1	0	-	1	-	2(5.7%)
Total	19(54.28%)	3(8.5%)	8(22.8%)	4(11.4%)	1(2.8%)	35

FIGURE- 5
Different Dermatophyte Isolated From Different Clinical Types

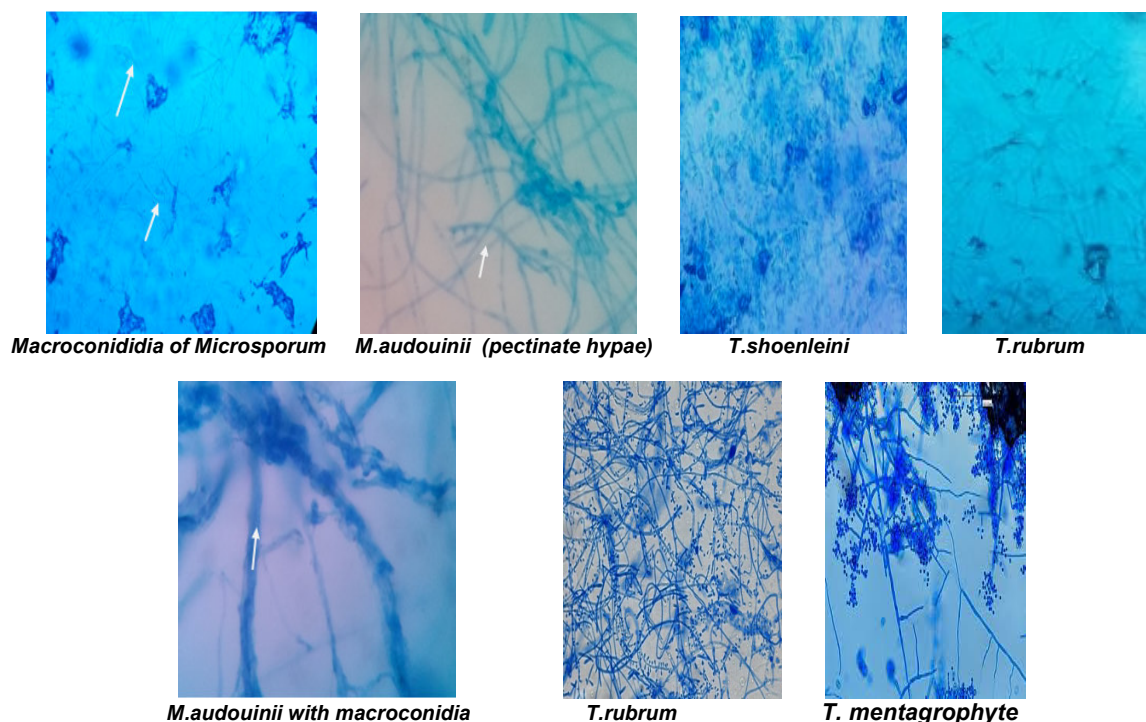


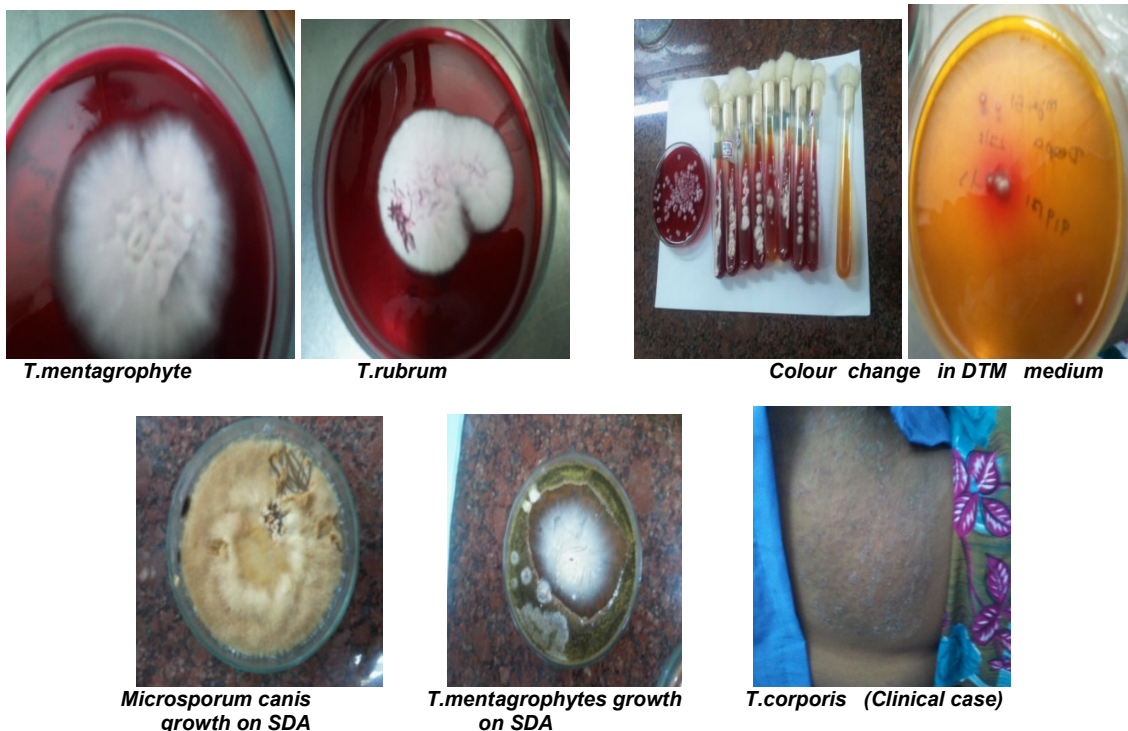
Comparison of culture positivity and KOH positivity

Table 6 is showing 10% KOH result

	KOH positive	KOH negative	Culture positive	Culture negative
Total number of samples processed	25	21	35	11
46				
Culture positivity in KOH positive cases	25	-	25	nil
Culture positivity in KOH negative cases	-	21	10	11
Total dermatophytes isolated - 35				

FIGURE- 6
IMAGES (MICROSCOPIC,MACROSCOPIC,CLINICAL) OF DERMATOPHYTES





DISCUSSION

Most of the dermatophytic infections are usually diagnosed and treated based on clinical presentations alone, as it takes long time to obtain culture report. Microscopic examination of the sample often needs expertise to identify fungal elements with dermatophyte morphology.⁷ However, it offers painless and rapid results. As many of the non-infective conditions often present with similar clinical presentation, culture identification of dermatophytic infections is usually recommended for those cases that require prolonged treatment with antifungal agents.⁴ In our present study, also out of 132 cases clinically diagnosed as dermatophytic infections, only 46 cases were requested for culture confirmation for dermatophytes and for 10% KOH mount. Among 132 clinically diagnosed cases, the male to female ratio was 2:1, i.e., 88:44. This finding is correlating with many studies.^{8,9,10} The age distribution is also correlating to several studies, i.e., more common among 30-40 years age group.^{11,12} Seasonal distribution is more common during October to December months. Out of 46 cases, 35 cases were culture positive, i.e., 76% and 24% were culture negative on special dermatophyte media, whereas 25 cases (54.34%) were positive for 10% KOH mount and 21 cases were negative (45.66%). This is correlated with other studies.^{13,14,15} 59.20%, 53.3%, 49%, however, some of the studies show that 10% KOH preparation is more sensitive.^{16,17,18} The commonest infections were *Tinea corporis* (54.28%) and the commonest causative organism was *Trichophyton rubrum* (51.45%). This is correlating with many other studies in India.⁽¹⁹⁾ In present study, the 10% KOH mount showed 54.34% positive, i.e., 25 positives out of 46 samples, whereas culture positive were 35 out of 46

clinical cases (76%). All 10% KOH positive cases were positive even for culture. This can be attributable to person-to-person collection of sufficient samples from the active growing edge of the lesion and their proper transportation. According to some studies, 10% KOH is more sensitive than fungal culture. However, in our study, fungal culture was more sensitive (76%), this is correlating with other studies where culture positive was 79.1%,³ 62.8%,⁽¹⁶⁾ 51%.²⁰ Usually *Tinea corporis* (^{21,22}) infections are common, that present with inflammation, erythema, scaling, blister formation, especially in the case of *T. rubrum*, presents with nodular granulomatous perifolliculitis-like lesions.⁽²³⁾ The lesions are often seen over trunk, neck, hands, and buttocks. In current study, many of *T. corporis* infections (54.28%) were presented with typical clinical presentations. Few were presented with blister formation and few with localized abscess in the thigh, gluteal region and over limbs and some with typical nodular granulomatous perifolliculitis-like lesions characteristic of *T. rubrum*. Usually *T. corporis* infections were caused by *T. rubrum*, *T. mentagrophytes*, *M. audouinii*, *M. gypseum* etc. In present study, many of the infections were caused by *T. rubrum* and *T. mentagrophytes*. Two cases by *T. tonsurans*. *T. cruris*, second commonest ring worm infection and in our study, also it was second commonest (22.8%). *T. cruris* presents with erythematous, scaly, raised sharply demarcated edges with central clearing, often associated with pruritis and burning.⁶ The causative organisms include *T. rubrum*, *T. mentagrophytes* var. *interdigitale* and *E. floccosum*. The most common being *T. rubrum*. In our present study, also this is correlating, *T. rubrum* being the most common isolate. *T. capitis* infections are common in paediatric age group.²⁴ Usually caused by *M. audouinii*, *T. schoenleinii*, *T. tonsurans* and *T. violaceum*. The

lesions are scaly, annular, elevated, erythematous edge, sharp margin, central clearing, with ectothrix or an endothrix type of lesions. In the present study four cases of *T. capitis* identified one female patient with 7 years and one male patient with 17 years, and two other from males age group 21 and 8. (*T. rubrum*, *T. mentagrophyte*) and Female patient presented with alopecia, scaling with black spots and male patient with endothrix type of lesion. *M. audouinii* and *T. tonsurans* are isolated from these two patients. *T. pedis* also called athlete's foot presents with dryness, fissures white scales or moist macerated lesions another form scaling of soles and lateral surfaces with inflammation and dryness. Third form with erythema, pustules, vesicles, bullae over feet.¹³ In our present study out of 5 cases two cases presented as blister over the foot 3 cases with fissures and scaly lesions. The commonest causative organism *T. rubrum*, *mentagrophytes* var. *interdigitale* and *E. floccosum*. In our present study *T. rubrum* and *T. mentagrophytes* were isolated. *T. incognito*, these lesions are due to inadvertent use of steroids by non-dermatologist to treat skin infections. They present with the atypical clinical presentation less scaly and less raised margins pruritic,

erythematous and mimic other clinical conditions.²⁵ The commonly isolated organism in these cases is *T. rubrum*.²⁶ In our study *T. incognito* cases were 17 out of 132 cases (12.87%) only one case had given rise to culture positive for *T. rubrum*.

CONCLUSION

In general dermatophytic infections are more common in males with age 30-40 years and among superficial mycotic infections, dermatophytic infections (*T. corporis*) are common. They are caused most commonly by *T. rubrum*. By improving living conditions and effective treatment anthropophilic dermatophytic infections can be kept under control. Good hygiene and isolation of individual cases can prevent their transmission. Infected animals also should be isolated and treated. Proper disinfection of fomites, maintaining environmental hygiene and moisture control especially for certain dermatophytic infections like *Tinea pedis* is essential to prevent further transmission, as prevention is always better than cure in all human health related problems.

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