JOURNAL OF PURE AND APPLIED MICROBIOLOGY, April 2011. Vol. **5**(1), p. 257-261

Isolation and Identification of Dermatophytes

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(Received: 16 November 2010; accepted: 28 December 2010)

To identify the etiological agents of Dermatophytosis in Mysore, the presence study was carried out by the department of Microbiology, Mysore Medical College & Research Institute, Mysore. Skin, hair and nail specimens were collected from a total of two hundred clinically diagnosed randomly selected patients of dermatophytosis attending the out patient department of Dermatology and Venereology, and were processed and identified by standard laboratory techniques. Fungi was demonstrated in 148 cases (74%) either by KoH preparation and / or culture. Most common clinical type was tinea corporis (45%) followed by tinea cruris (19%) and tinea unguium (17%). Most common etiological agent was T. rubrum (62.7%) followed by T.mentagrophyte (25.42%), M.gypseum (5.08%), T. tonsurans (5.08%) and E. floccosum (1.69%). Dermatophytosis was common in the age group of 21 – 30 yrs (28%) and in males (64%). Most of the patients belonged to low socio economic status (85%) and were manual workers (39%).

Key words: Dermatophytosis, *Trichophyton rubrum*,*Trichophyton mentagrophytes*, *Microsporum gypseum, Trichophyton tonsurans*.

Superficial fungal infections are most common skin diseases, affecting millions of people throughout the world.[1] The dermatophytes are by far the most significant fungi because of their wide spread involvement of population at large and their prevalence all over the world.[2] The estimated life time risk of acquiring a dermatophyte infection is between 10 and 20 percent.[1]

Dermatophytes are keratinophilic fungi that cause infections of skin, hair and nails. These infections generally remain limited to non living keratinized layers, but the infection may proceed more deeply than for superficial mycoses, and a variety of pathologic changes can occur depending on the fungus, the site of infection and the immune status of the host. The classical presentation of tinea infection is a lesion with central clearing surrounded by an advancing red, scaly elevated border.[1]

Dermatophytes are assuming greater significance both in developed and developing countries particularly due to the advent of immunosuppressive drugs and disease. Hot and humid climate in tropical and subtropical countries like India makes dermatophytosis a very common superficial fungal skin infection. Although dermatophytosis does not cause mortality, it does cause morbidity and poses a major public health problem and also is of cosmetic importance.[3-6]

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In India, cases of superficial mycosis were first reported from upper Assam by Powell in 1900 AD. Since then various studies have been conducted from different parts of the country.[4] The etiological pattern varies with patient population, geographical region and also tends to vary over a period of time. The clinical presentation, though very typical of ringworm infection is very often confused with other skin disorders, making laboratory diagnosis and confirmation necessary.[3] The present study was undertaken to isolate and identify the etiological agents of dermatophytosis.

MATERIAL AND METHODS

The present study of dermatophytosis was carried out in the department of microbiology, Mysore medical College and Research Institute, Mysore. A total of two hundred clinically diagnosed randomly selected cases of skin, hair and nail infection, of all age groups and of both sexes, attending out patient department of Dermatology and Venereology of K.R.Hospital, Mysore were taken for study. A detailed history of selected cases was taken in relation to name, age, sex, address, occupation, duration of illness and involvement of more than one site. Patients under antifungal treatment were excluded from the study group.

Clinical examination of patient was made in good light and affected area was cleaned with 70% ethyl alcohol. Skin specimen was collected by scraping across the inflamed margin of the lesion; nail specimen was collected by taking clippings of the infected part and scrapings beneath the nail and hair specimen was collected by plucking with epilating forceps along with the base of the hair shaft around follicle. [2, 7-9.]

Specimen collected was subjected to potassium hydroxide (KoH) wet preparation of various concentrations (10%, 20% and 40%) depending on the type of clinical specimen for the presence of fungal elements. Another portion of the specimen was inoculated on to three sets of test tubes, one containing sabouraud's dextrose agar with 0.05% chloramphenicol, sabouraud's dextrose agar with 0.05% chloramphenicol and 0.5% Cycloheximide and the other to dermatophyte test medium. [2, 7-9]

Sabouraud's dextrose agar with 0.05% chloramphenicol, sabouraud's dextrose agar with 0.05% chloramphenicol and 0.5% Cycloheximide was incubated at 28ºC for up to four weeks, and

Table 1. Various clinical types in relation to occupication

Clinical Types	Occupation					Total
	Manual	HouseWife	Students	Professional	Miscellaneous	
Tinea corporis	36 (40%)	16 (17.78%)	22 (24.44%)	10 (11.11%)	6 (6.67%)	90 (45%0)
Tinea Cruris	14 (36.84%)	16 (42.11%)	8 (21.05%)	-	-	38 (19%)
Tinea Unguium	16 (47.06%)	14 (41.18%)	2 (5.88%)	2 (5.88%)	-	34 (17%)
Tinea Capitis	-	-	8 (66.67%)	-	4 (33.33%)	12 (6%)
Tinea Pedis	-	2 (50%)	2 (50%)	-	-	4 (2%)
Tinea Faciei	-	-	4 (100%)	-	-	4 (2%)
Tinea Manuum	2 (100%)	-	-	-	-	2 (1%)
Tinea Corporis With Tinea Cruris	10 (6.25%)	6 (37.5%)	-	-	-	16 (8%)
Total	78 (39%)	54(27%)	46(23%)	12(6%)	10(5%)	-

was observed periodically for growth. If no growth was found after four weeks it was taken as negative for growth and discarded. Dermatophyte test medium was incubated at 28ºC for up to ten days and was observed for colour change. Fungal isolate was identified based on colony morphology, pigmentation, growth rate, microscopy (LPCB), slide culture, urease test, hair perforation test and rice grain test. [2, 7-9.]

RESULTS

Most common age group affected was 21 -30 yers with 56 cases (28%) and the least common was 61 - 70 years with 10 cases (5%). Mean age was 33.15 years. Males were more commonly affected with 128 cases (64%) than females with 72 cases (36%). Male to female ratio was 1.78:1.

Majority of the cases were from low income group with 170 cases (85%) followed by middle income group with 20 cases (10%) and high income group with 10 cases (5%).

Dermatophytosis was predominantly seen in manual workers (39%) followed by house wives (27%), students (23%), and professionals (6%) and miscellaneous (5%). *Tinea corporis*, *Tinea cruris* and *Tinea unguium* were more commonly seen in manual workers followed by house wives. *Tinea capitis* was more commonly seen in school going children (66.67%). Various clinical types in relation to occupication is shown in Table 1.

Out of the two hundred patients, 148 (74%) were found to be positive for fungal etiology either by direct microscopy and/or culture. 114 cases (57%) were positive by both microscopy and culture. 30 cases (15%) were positive by microscopy and negative by culture. four cases (2%) were negative by microscopy but culture positive. 52 cases (26%) were negative both by microscopy and culture.

Tinea corporis was the commonest clinical type with 90 cases (45%) followed by *Tinea cruris* (19%), *Tinea unguium* (17%) and *T. capitis* (6%). Dermatophytes isolated in relation to clinical types is shown in Table 2.

Table 2. Dermatophytes isolated in relation to clinical types

Clinical type	No	*T. rubrum*	*T. mentagrophyte*	*M.gypseum*	*T.tonsurans*	*E.floccosum*	Total isolated
Tinea corporis	90	36 (66.67%)	12 (22.22%)	6 (11.11%)	-	-	54 (60%)
Tinea cruris	38	16 (66.67%)	6 (25%)	-	-	2 (8.33%)	24 (63.16%)
Tinea unguium	34	6 (60%)	4 (40%)	-	-	-	10 (29.41%)
Tinea capitis	12	-	-	-	6 (100%)	-	6 (50%)
Tinea pedis	4	2(100%)	-	-	-	-	2(50%)
Tinea faciei	4	4 (100%)	-	-	-	-	4 (100%)
Tinea manuum	2	2 (100%)	-	-	-	-	2 (100%)
Tinea corporis with *Tinea cruris*	16	8 (50%)	8 (50%)	-	-	-	16 (100%)
Total	200	74 (62.71%)	30 (25.42%)	6 (5.08%)	6 (5.08%)	2 (1.69%)	118 (59%)

DISCUSSION

The present study shows that dermatophytosis was more common in the age

group of 21-30 years (28%) followed by 31-40 years (18%), which is comparable with other studies done by Sen SS[10], Mishra M[6], whereas Veer P[11] has reported that the most common age group affected was 31-40 years followed by 41-50 years. The highest incidence in young adults aged 21-30 years may be due to increased physical activity and increased opportunity for exposure.

Males (64%) were more commonly affected than females (36%). Male to female ratio was 1.78:1, which is comparable with other studies done by Huda[4], Karmakar[12], Bindu[13], Singh[14], whereas and Nada[15] reported that females were more commonly affected than males, with male to female ratio being 0.69:1. Male predominance may be due to increased outdoor physical activities and increased opportunity for exposure to infection than females. *Tinea capitis* was more commonly seen in males (83.33%) and in the age group of <10 years (83.33%), which is comparable with other studies done by Siddappa[16] (77.78%) and Kumar[7] (78%). High occurrence of *Tinea capitis* in less than 10 years of age may be due to lack of fungistatic secretion by scalp in childhood. Adult sebum has fungistatic action. The low occurrence in females could be due to regular application of vegetable oil over the scalp which has fungistatic properties.

Infection of most common in low income group (85%) followed by middle income group (10%) and high income group (5%), which is similar to the observation of Ranganathan[17] who reported that 69.2% of infected people were from the low income group and 23.2% from middle income group. This may be due to poor hygienic condition, over crowding, sharing clothes without washing them properly and also due to poor nutrition.

Dermatophytosis was most commonly seen in manual workers 39% followed by house wives 27%, students 23%, professionals 6% and miscellaneous 5%, which is comparable with the ovservation of Veer[11], whereas Nada[15], reported high incidence in house wives 37.7% followed by professionals 28.3%, manual workers 13.2%, students 13.2% and miscellaneous 7.6%. This may be due to increased physical activity and increased opportunity for exposure in manual workers, and increased wet work in housewives.

In the present study out of 200 clinically diagnosed cases of dermatophytosis 148 cases (74%) wer positive for fungi, either by KOH and / or culture. 140 cases (57%) were positive by both KOH and culture, 30 cases (15%) were positive by KOH and negative by culture, 4 cases (2%) were negative by KOH but culture positive, 52 cases (26%) were negative by both KOH and culture, which is comparable with other studies done by Nada H[15] and Singh S.[3] This variation could be due to non-viability of fungal elements in some cases.

In the present study, *T. rubrum* 37 (62.71%) was the commonest etiological agent in majority of the clinical types followed by *T. mentagrophytes* 30 (25.42%), *M.gypseum* 6 (5.08%) *T. tonsurans* 6 (5.08%) and *E. floccosum* 2 (1.69%). In *Tinea capitis*, *T. tonsurans* was isolated in all the 6 cases (5.08%), which is comparable to other studies done by Bindu[13], Venkatesan[5], whereas Karmakar[12], and Fathi[18] in their study reported *T. violaceum* and *T. verrucosum* as the predominant isolates respectively.

In the present study, *T. tonsurans* was isolated in all the three cases of *Tinea capitis* (100%), which is comparable with the study of Gupta[19] (76%), whereas Kumar[7] reported *T. violaceum* (63.15%) as the commonest etiological agent of *Tinea capitis*.

CONCLUSIONS

Two hundred clinically diagnosed cases of dermatophytosis were studied. *T. corporis* 90 (45%) was the commonest clinical type followed by *T. cruris* 38 (19%), *T. unguium* 34 (17%), *T. corporis* with *T. cruris* 16 (8%), *T. capitis* 12 (6%), *T. pedis* 4 (2%), *T. faciei* 4 (2%) and *T. manuum* 2 (1%). Commonest age group affected was 21-30 years. Male to female ratio was 1.78:1.

- Majority of cases belonged to low socio-economic stutus 85%.
- Infection was more common in manual workers 39%.
- Fungi was demonstrated in 74% cases, either by direct microscopy and / or culture.
- Of the dermatophytes isolated, *T. rubrum* 74 (62.71%) was the commonest etiological agent followed by *T. mentagrophytes* 30 (25.42%), *M.gypseum* 6 (5.08%) *T. tonsurans* 6 (5.08%) and *E. floccosum* 2 (1.69%).

Dermatophyte infections are very common in our country where hot and humid climate in association with poor hygienic conditions play an important role in the growth of these fungi. There is varying difference in isolation of different species from southern and northen part of India. By and large Trichophyton species forms the commonest etiological agent of dermatophytosis. *T. rubrum* was the commonest isolate in *Tinea corporis*, *Tinea cruris* and *Tinea unguium*. *T. tonsurans* was isolated from all the 6 cases of *Tinea capitis*.

REFERENCES

1. American Family Physician. Diagnosis and management of common Tinea infections. 1998; **58**(1): 163-74,177-8.
2. Chander J., Textbook of Medical Mycology. 3rd ed. New Delhi: Mehta Publisher:pg. 2009; 53-67, 122-42.
3. Singh S, Beena PM. Comparative study of different microscopic techniques and culture media for the isolation of dermatophytes. *Indian J Med Microbiol* 2003; **21**: 21-4.
4. Huda MM, Chakraborthy N, Bordoloi JNS. A clinico-mycological study of superficial mycoses in upper Assam. *Indian J Dermatol Venereol Leprol* 1995; **61**: 329-32.
5. Venkatesan G, singh AJAR, Murugesan AG, Janaki C, Shankar SG. *Trichophyton rubrum* – the predominant aetiological agent in human dermatophytosis in Chennai, India. *Afr J Microbiol Res.*, 2007; 9-12.
6. Mishra M, Mishra S, Singh PC, Mishra BC. Clinico-mycological profile of superficial mycoses. *Indian J Dermatol Venereol Leprol* 1998; **64**: 283-5.
7. Kumar AG, Lakshmi N. *Tinea capitis* in Tirupati. *Indian J Pathol. Microbiol.* 1990; **33**(4): 360-3.
8. Kannan P, Janaki C, Selvi GS. Prevalence of dermatophytes and other fungal agents isolated from clinical samples. *Indian J Med Microbiol* 2006; **24**(3):212-5.
9. Collee JG, Fraser AG, Marmion BP, Simmons A. Mackie and McCartney Practical Medical Microbiology. 14th ed. Edinburgh: Churchill Livingstone; 1996; 695-716.
10. Sen SS, Rasul ES. Dermatophytosis in Assam. *Indian J Med Microbiol* 2006; **24**: 77-8.
11. Veer P, Patwardhan NS, Danle AS. Study of onychomycosis: prevailing fungi and pattern of infection. *Indian J Med Microbiol* 2007; **25**: 53-6.
12. Karmakar S, Kalla G, Joshi KR. Dermatophytosis in a desert district of Western Rajasthan. *Indian J Dermatol Venereol Leprol* 1995; **61**: 280-3.
13. Bindu V, Pavithran K. Clinico-mycological study of dermatophytosis in Calicut. *Indian J Dermatol Venereol Leprol* 2002; **68**(5): 259-61.
14. Singh S, Beena PM. Profile of dermatophyte infections in Baroda. *Indian J Dermatol Venereol Leprol.*, 2003; **69**: 281-3.
15. Allah SS, Nada H, Mokhtar M. Yeast infections as a cause of nail disease in the Western province of Saudi Arabia. *Egypt J Med Lab Sci* 2005; **14**: 2.
16. Siddappa K, Mahipal OA. Dermatophytosis in Davangere. *Indian J Dermatol Venereol Leprol* 1982; **48**(5): 254-9.
17. Ranganathan S, Menon T, Selvi GS, Kamalam A. Effect of socio-economic staus on the prevalence of dermatophytosis in Madras. *Indian J Dermatol Venereol Leprol* 1995; **61**: 16-8.
18. Fathi HI, Alsamarai AM. *Tinea capitis* in Iraq. *East Mediterr Health J* 2000; **6**: 138-48.
19. Gupta AK, Summerbell RC. Increased incidence of T. tonsurans, *Tinea capitis* in Ontario, Canada between 1985 and 1996. *Medical Mycology* 1998; **36**: 55-60.