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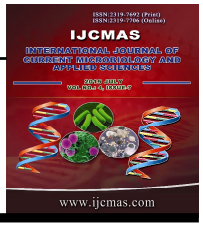

Original Research Article

Clinico-Mycological Study of Dermatophytosis – Our Experience

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K e y w o r d s

Fungal infections, Dermatophytic infections, Lactophenol Cotton Blue, *Tinea corporis*, *Trichophyton rubrum*

A B S T R A C T

Superficial mycosis refers to the diseases of the skin and its appendages by the fungi. Infection by Dermatophytes is restricted to non living cornified layers of the skin. The Present study was conducted to study the pattern of dermatophytic infections in patients suffering with fungal infections. The study group includes all the confirmed cases of Dermatophytosis. The scrapings were collected from the edges of the lesions by using sterile blunt scalpel blade after swabbing the lesion with 70% alcohol. All the specimens were observed under the microscope after performing KOH wet mount examination.10% KOH was used for Skin scrapings and Hair plucking. Slants were observed for a period of 6 weeks. Any growth on the slant was performed wet mount by using Lactophenol Cotton Blue and observed under the Microscope for the presence of hyphal structures and modifications. Out of 298 clinical specimens from the confirmed cases 220(73.83%) were skin scrapings, 34(11.41%) were nail clippings and 44(14.77%) were hair plucking with hair stub. The commonest clinical type of dermatophytosis was *Tinea corporis* with 96/298(32.21%) followed by *Tinea cruris* 83/298(27.85%), *Tinea pedis* 41/298(13.76%), *Tinea unguinum* 34/298(11.41%), *Tinea capitis* 18/298(6.04%), *Tinea faciae* 14/298(4.70%) and *Tinea barbae* 12/298(4.03%). Out of 298 cases of Dermatophytosis 140 were positive for culture. *Trichophyton rubrum* was the commonest isolate 97/140(69.29%). Dermatophytic infections are of concern because of their character of chronicity of the disease. The study highlighted the various types of Dermatophytic infections in and around the places of Nellore. The associated factors like climatic conditions, Socio economic status, Occupation and low literacy rate, lack of knowledge about the disease makes once again a concern. The KOH wet mount examination can be used a screening method for identification of Dermatophytic infections.

Introduction

Superficial mycosis refers to the diseases of the skin and its appendages by the fungi. Over the past decades, they are on the rise and accounts for 20-25% of world's population.[1] They are more common in tropical countries due to humidity, elevated temperatures and sweating[2, 3]. This include Dermatophytosis, Pityriasis versicolor and Candidiasis.[4] Dermatophytes which form the common in the group has the ability to infect the keratinized tissue and cause dermatophytosis commonly called as Ring worm.

Infection by Dermatophytes is restricted to non living cornified layers of the skin. The severity of infections caused by Dermatophytes are influenced by multiple factors which include the virulence of the infected strain or species, anatomic location of the infection, environmental factors and the hosts reaction to fungal metabolic products .[5] However the Dermatophytes and the type of Dermatophytosis are geographically variable because of life style of the population, climatic conditions and migration of population. Therefore some are widely distributed and some are geographically restricted.

In the past decades major changes in the epidemiological pattern of dermatophytic infections have occurred because of intercontinental transport and migration of population. The Present study was conducted to study the incidence and type of dermatophytic infections in patients suffering with fungal infections.

Materials and Methods

The present study was conducted by the Department of Microbiology along with Department of Dermatology, Narayana medical college and Hospital for a period of one year from March 2013 to February 2014. The study was approved by the Institutional ethical committee. Detailed demographic data of the cases which include the duration of the disease, socio economic status, Occupation, H/O of antifungal therapy, source of infection was collected.

The study group included all the confirmed cases of Dermatophytosis. Patients who were on treatment with Topical and systemic antifungal drugs were not included in the study.

The specimens included Skin scrapings, nail clippings and hair plucking with the hair stub. However multiple specimens were collected from patients having lesions at multiple sites and were processed as individual specimens.

Collection of Skin scrapings

The scrapings were collected from the edges of the lesions by using sterile blunt scalpel blade after swabbing the lesion with 70% alcohol. Care was taken not to cause any bleeding from the lesions while collecting the specimens. Scrapings were collected on Black photographic paper for better visualization.[6,7]

Collection of nail clippings

The affected nails were swabbed with 70% Alcohol and Scrapped up to the affected region and also nail clippings were also collected.

Collection of hair plucking

The affected hairs were epilated using sterile forceps and also scales from the surrounding area were collected after swabbing with 70% alcohol. Care was taken to include the basal portion of the hair [Hair stub].

Processing of the specimens

Microscopy: All the specimens were observed under the microscope after performing KOH wet mount examination.10% KOH was used for Skin scrapings and Hair plucking. Nail clippings were kept in 40% KOH over night and examined under 10x objective of the microscope for the presence of septate hyphae and arthrospores. Confirmation was made after examining under 40x objective of the microscope.

Culture: The specimens were inoculated on Sabourad's Dextrose agar with cycloheximide [0.5mg/ml] and Chloramphenicol [0.05mg/ml], Dermatophyte test medium with added supplements for isolation of the dermatophytes [Himedia manufacterrers Ltd.Mumbai].DTM is a modification of commercial formulation made by Taplin et al in 1969 which is used for the isolation and cultivation of pathogenic Dermatophytes[8]. The specimens were inoculated at different sites on the slants of the medium and incubated in BOD at 28^0c and also at room temperature. The slants were inspected at regular intervals to assess the growth rate and to identify the colony characters like morphology, pigmentation on the reverse and obverse.

Slants were observed for a period of 6 weeks and absence of growth after the time period was discarded. Any growth on the slant was performed wet mount by using Lactophenol Cotton Blue and observed under the Microscope for the presence of hyphal structures and modifications, reproductive structures like microconidia and macroconidia. Tease mount and slide culture was also performed for study of the microscopic morphology.[9-11]

Results and Discussion

The study group included 298 diagnosed cases of dermatophytosis. There were 168 males (56.38%) which outnumbered the females 130 (43.62%). The predominant age group in both males and females was 21-30yrs with males around 14.77% and females 26.85% followed by 31-40 yrs [Table-1]. Out of 298 clinical specimens from the confirmed cases 220(73.83%) were skin scrapings, 34(11.41%) were nail clippings and 44(14.77%) were hair plucking with hair stub. The commonest clinical type of Dermatophytosis was *Tinea corporis* with 96/298(32.21%) followed by Tinea cruris 83/298(27.85%), Tinea pedis 41/298(13.76%), Tinea unguinum 34/298(11.41%), Tinea capitis 18/298(6.04%), Tinea faciae 14/298(4.70%) and Tinea barbae 12/298(4.03%) [Table-2].

Out of the total 298 dermatophytosis cases 165(55.37%) were direct KOH wet mount examination positive and 140(46.97%) were culture positive. 118(39.6%) were positive for both KOH and culture. In 22 (7.37%) cases culture was positive but KOH wet mount negative. 111 (37.26%) were KOH wet mount as well as culture negative. Sensitivity was 71.52%, Specificity was 83.46%%, Positive predictive value was 84.29%, Negative predictive value was 70.25%. [Table-3]

Out of the total 298 cases of Dermatophytosis 140 were positive for culture. *Trichophyton rubrum* was the commonest isolate 97/140 (69.29%).Other isolates in order of frequency are *Trichophyton mentagrophytes* 23/140 (16.43%), Trichophyton violaceum 9/140 (6.43%), Microsporum audonii 6/140 (4.29%) and last include *Epidermophyton flocculosum* 5/140 (3.57%).

In the total 69 cases of culture positive *Tinea corporis*, *Trichophyton rubrum* was the predominant isolate 49/69(71.01%) followed in order by *Trichophyton mentagrophytes* 14/69 (20.29%), Trichophyton violaceum 4/69 (5.8%), *Epidermophyton flocculosum* and Microsporum audonii 1/69(1.45%).

In 27 culture positive cases of Tinea cruris, *Trichophyton rubrum* was the predominant isolate 19/27(70.37%) followed by *Trichophyton mentagrophytes* 4/27(14.81%) and *Epidermophyton flocculosum*, Microsporum audonii each 2/27 (7.41%).

In 16 culture positive cases of Tinea unguinum, *Trichophyton rubrum* with13/16(81.25%) and *Trichophyton mentagrophytes*, Trichophyton violaceum and Microsporum audonii each with 1/16(6.25%). In 9 cases of Tinea pedis, *Trichophyton rubrum* was major isolate 6/9(66.67%) followed by Trichophyton violaceum, *Trichophyton mentagrophytes* and *Epidermophyton flocculosum* each 1/9(11.11%).

Trichophyton rubrum, Trichophyton mentagrophytes each 2/5(40%) and Trichophyton violaceum 1/5(20%) accounted for 5 cases of Tinea faciae. *Trichophyton rubrum* was 3/4 (75%) and *Epidermophyton flocculosum* was 1/4(25%) among 4 culture cases of Tinea barbae.

Dermatophytosis accounts for one of the most common fungal infections around the world. 1/5th of the world's population suffers from mycotic infections[12] . The maximum age incidence in the study was 21-30 yrs which is in par with the studies of Kumar et al, Nita Patwar Dhan, Rashmika Dave et al [13] .The maximum incidence is explained by the reason of increased physical activity and exposure, hormonal influences during the period. Studies of V.Rajagopal reports maximum age incidence during 11-20yrs which is in contrary to the present study.[14] Males were predominant in the present study (56.38%) which concur with the studies of Kumar et al, KM Achary, RK Thakur et al [15], SS Sen, ES Rasul et al [16] and many other studies. The reason is males have an increased opportunity for exposure towards fungi and increased outdoor activities. Females being more Cosmetic conscious and go for an early treatment.

In the present study out of 298 cases 46.97% were culture positive and 53.08% were culture negative. However 53.37% were KOH positive and 44.63% were KOH negative. This finding coincides with the studies of Reena ray Ghosh et al [17] and others . Findings in the studies of Bindu .V.et al, S.singh, P.Beena et al and SS Sen, ES rasul reported more number of culture positivity than KOH negative cases. *Trichophyton rubrum* was the predominant isolate [69.29%] throughout the study. This finding coincides with the reports of Singh S et al in 2003 – 73.27% [18]. Mohanthy JC et al in 1998 – 68.34% [19], Bindu V et al in 2002-66.2%, Sumana V et al in 2004 – 60%, Peerapur B V et al in 2004 – 43.7% [20], Gupta BK et al in 1993 -42.42% [21]*Trichophyton mentagrophytes* formed the 2nd common isolate[16.43%] followed by Trichophyton violaceum[6.43%], Microsporum audonii[4.29%] and the last *Epidermophyton flocculosum*[3.57%].

The following findings are in par with the findings of the studies mentioned above.

In Cases of *Tinea corporis* out of 69 total isolates *Trichophyton rubrum* was the predominant isolate 71.01% followed by *Trichophyton mentagrophytes* 20.29%, Trichophyton violaceum5.8%,Microsporum audoni and *Epidermophyton flocculosum* 1.45% .

Table.1 Dermatophytosis in relation to age and sex

AGE(Yrs)	MALE =n (%)		FEMALE =n (%)		TOTAL= n (%)	
0-10	6	2.01	8	2.68	14	4.70
11- 20	19	6.38	12	4.03	31	10.40
21-30	44	14.77	36	12.08	80	26.85
31-40	38	12.75	24	8.05	62	20.81
41-50	22	7.38	17	5.70	39	13.09
51-60	20	6.71	17	5.70	37	12.42
>65	19	6.38	16	5.37	35	11.74
TOTAL	**168**	**56.38**	**130**	**43.62**	**298**	

Table.2 Clinical types of Dermatophytosis

Clinical types	Cases(n)	Percentage(%)
Tinea corporis	96	32.21
Tinea unguinum	34	11.41
Tinea pedis	41	13.76
Tinea cruris	83	27.85
Tinea barbae	12	4.03
Tinea capitis	18	6.04
Tinea facie	14	4.70
TOTAL	**298**	

Table.3 Results obtained after direct examination and culture

	KOH positive (n%)	KOH negative (n%)	Total (n%)
Culture positive	118(39.6%)	22(7.37%)	140(46.97%)
Culture negative	47(15.77%)	111(37.26%)	158(53.03%)
TOTAL	165(55.37%)	133(44.63%)	298(100%)

Table.4 Dermatophytes in different clinical types

	Tinea corporis	***Tinea unguinum***	***Tinea pedis***	***Tinea cruris***	***Tinea barbae***	***Tinea capitis***	***Tinea facie***	**Total**
Trichophyton rubrum	49 (71.01%)	13 (81.25%)	6 (66.67%)	19 (70.37%)	3 (75%)	5 (55.56%)	2(40%)	97 (69.29%)
Trichophyton mentagrophytes	14 (20.29%)	1 (6.25%)	1 (11.11%)	4 (14.81%)	0	1 (11.11%)	2(40%)	23 (16.43%)
Trichophyton violaceum	4(5.8%)	1 (6.25%)	1 (11.11%)	0	0	2 (22.22%)	1(20%)	9 (6.43%)
Microsporum audoni	1(1.45%)	1 (6.25%)	0	2 (7.41%)	0	1 (11.11%)	0	6 (4.29%)
Epidermophyton flocculosum	1(1.45%)	0	1 (11.11%)	2 (7.41%)	1(25%)	0	0	5 (3.57%)
Total	69	16	9	27	4	9	5	

The findings of our study coincides with the findings of Nita patwardhan, Rasmika Dave in 1999[13], Seema Bhaduria, Neetu Jain in 2001[12] and G. Venkatesan, AJA Ranjit Singh et al in 2007. [23] In 27 cases of Tinea cruris, *Trichophyton rubrum* accounted for 70.37% forming the major isolate and the rest included *Trichophyton mentagrophytes*(14.81%), *Microsporum audoni* and *Epidermophyton flocculosum* (7.41%) each. The finding of our study is supported by the studies mentioned earlier.

Among the clinical types of Dermatophytosis, *Tinea corporis* accounted for the majority 96/298(32.21%) followed by Tinea cruris (83/298) 27.85%, Tinea pedis 41/298(13.76%),Tinea unguinum34/298(11.41%),Tinea capitis 18/298(6.04%),Tinea faciae 14/298(4.70%) and the least Tinea barabae 12/298(4.03%). Studies by G. Venkatesan, A.J.A Ranjit Singh et al in 2007, Nita Patwardhan et al in 1999, Seema Bhaduria et al in 2001 also reported *Tinea corporis* as the major clinical Dermatophytosis. The reason why *Tinea corporis* and Tinea cruris are common is because of their pruritic nature which seeks the medical attention.

Out of all Dermatophytic fungi, Trichophyton was the most common followed by Microsporum and Epidermophyton in the present study. These findings were substantiated by many studies in India and abroad.

In conclusion, Dermatophytic infections are of concern because of their character of chronicity of the disease. The study highlighted the various types of Dermatophytic infections in and around the places of Nellore. The associated factors like climatic conditions, Socio economic status, Occupation and low literacy rate, lack of knowledge about the disease makes once again a concern. The KOH wet mount examination can be used a screening method

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