Clinico-etiologic correlates of onychomycosis in Sikkim

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ABSTRACT

Aims and objectives: The etiological spectrum of any superficial mycosis is largely dependent on the flora in the immediate environment of the individual. It is influenced by the geographic, climatic and occupational factors. The study is basically to formulate baseline data for the species prevalence of various dermatophytes in patients with onychomycosis attending the Central Referral Hospital, Gangtok, Sikkim. Materials and Methods: Thirty-four clinically suspected cases of onychomycosis were subjected to mycological studies. Results: Thirty-two (94.12%) cases were positive for fungal elements by direct microscopy and 28 (82.35%) by culture. Young adults in the age group of 21-30 years were mainly affected. The male:female ratio was 1.125:1. Dermatophytes were isolated in 18 cases (64.29%). Trichophyton tonsurans (44.44%) was the most common isolate followed by T. mentagrophytes (22.22%), T. rubrum (11.11%), T. verrucosum (11.11%) and Microsporum audouinii (11.11%). Apart from dermatophytes, Aspergillus niger (21.43%) and Penicillium marneffei (14.28%) were also isolated. Conclusion: Dermatophytes, mainly T. tonsurans, as well as moulds other than dermatophytes were isolated from onychomycosis patients in Gangtok, Sikkim.

KEY WORDS: Dermatophytes, Gangtok, onychomycosis

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INTRODUCTION

Onychomycosis refers to fungal infection of the nails with various etiological agents, involving dermatophytes, yeasts and non-dermatophytic moulds (NDM). It represents 18.4% of the onychopathies and about 30% of the mycotic cutaneous infections.[1] Dermatophytes, especially Trichophyton rubrum, are the most frequently implicated causative agents in onychomycosis. Previously regarded as contaminants, yeasts are now increasingly recognized as pathogens in finger nail infections, as are moulds.[2]

Onychomycosis is a common nail disorder that has a substantial impact on the patients quality of life. Diagnosis is made through clinical presentation, potassium hydroxide preparations and culture of tissue/nail samples.[3]

The spectrum of dermatophytes isolated from skin lesions had changed in the last 70 years. Before the Second World War in Germany, Microsporum audouinii and Epidermophyton floccosum ranked the first, whereas T. rubrum is the most common dermatophyte since the fifties of the last century, accounting for 80-90% of the strains, followed by T. mentagrophytes. This evolution is typical for Central and North Europe and it needs to be connected with the increase in the incidence of Tinea pedis. In contrast, in Southern Europe and in Arabic countries, zoophilic dermatophytes, such as M. canis or T. verrucosum, are the most frequently isolated. In Europe, especially in the Mediterranean countries, the incidence of M. canis infection has strongly increased during the recent years and this dermatophyte is now the most prevalent in Tinea capitis in children. An analysis of the frequency and distribution of Tinea pedis in different occupations and leisure-time activities as well as the routes of infection are reported. The spreading of this disease in most developed countries of the world represents a considerable economic problem because it was accompanied by a parallel increase in the frequency of onychomycosis, which implies, as Tinea pedis, large financial charges. In poor, developing countries, mycoses appear endemically, primarily with children, and their treatment often fails because of the lack of efficient antifungal agents.[4]

A high rate of false-negative dermatophyte detection is observed when common laboratory methods are used. These methods include microscopic observation of potassium hydroxide-digested nail clippings and culture methods using agar-based media supplemented with cycloheximide, chloramphenicol and gentamicin to isolate dermatophytes. Microscopic detection methods that use calcofluor white staining or periodic acid-Schiff staining may also be substituted for and have previously been reported to be more sensitive than potassium hydroxide-digested nail clippings. DNA protocol is an alternative method for detecting Trichophyton infections. When this protocol is used, the presence of T. rubrum DNA is directly detected. However, the viability of the dermatophyte is not addressed and further methods need to be developed for the detection of viable T. rubrum directly from nail samples.[5]

Onychomycosis is an important public health problem because of the increase...
in immunosuppressive states. Large-scale studies in India are scarce and so the baseline incidence of onychomycosis is not firmly established.\textsuperscript{[6]}

The etiological spectrum of any superficial mycosis is largely dependent on the flora in the immediate environment of the individual. It is influenced by the geographic, climatic and occupational factors. The present study was conducted with the object to formulate baseline data on etiological agents of onychomycosis in Gangtok.

**MATERIALS AND METHODS**

A total of 34 clinically suspected cases of dermatophytosis attending the dermatology outpatient department of the Central Referral Hospital, Tadong, Gangtok, during a 2-year period were studied.

A detailed clinical history with particular emphasis on history of trauma, infections, occupation, diabetes mellitus and personal habits were taken. Nail clippings were collected and subjected to mycological study both by direct microscopy in 20\% KOH solution and culture on Sabouraud’s dextrose agar medium and Dermatophyte test medium with chloramphenicol (0.05 mg/mL) and cycloheximide (0.5 mg/mL). The media were incubated at 25 and 37\°C for a minimum period of 3 weeks. Fungal species were identified on the basis of cultural characteristics, pigment production, microscopic examination in lactophenol cotton blue preparation and slide culture (wherever necessary).

**RESULTS**

The maximum number of patients was found in the age group of 21-30 years (20/30; 58.82\%). There were 18 males (52.94\%) and 16 females (47.06\%), the male to female ratio being 1.125:1.

Finger nails were involved in 14 cases (41.18\%) and toe nails were involved in 20 cases (58.82\%)\textsuperscript{[1]} [Table 1]. Both toe and finger nail involvements were not noticed in the same patient. Big toe nail was the most common toe nail involved and it was seen in 11 cases (55\%). Of 34 patients, 32 (94.12\%) cases were positive for fungal elements by direct microscopy and 28 (82.35\%) by culture [Table 2]. Of 28 isolated fungi, 18 (64.29\%) isolates were identified as dermatophytes. Trichophyton species and Microsporum species were isolated. T. tonsurans was the most common species isolated (44.44\%), followed by T. mentagrophytes (22.22\%), T. rubrum (11.11\%), T. verrucosum (11.11\%) and M. audouinii (11.11\%), although T. rubrum is the most common dermatophyte in other studies followed by T. tonsurans.\textsuperscript{[4,6]}

Recently, there had been a noticeable worldwide increase in the incidence of onychomycosis. This has been related to a variety of etiologic factors, including the rise in immunocompromised patients, an aging worldwide population and a rise in environmental risk factors secondary to life style changes.\textsuperscript{[7]}

The prevalence of onychomycosis has been reported to increase with advancing age. Fifteen to 20\% of the occurrence is found in patients aged 40-60 years and the incidence rises in those over 60 years of age.\textsuperscript{[8,9]} In the present study, 20 (58.82\%) patients were within the 21-30 years age group. Similarly, a high prevalence of onychomycosis in the younger age group was found in another study conducted in Bangalore.\textsuperscript{[10]} This increased incidence in the younger population could be because they are more often exposed to occupation-related trauma, predisposing them to onychomycosis. They may also be cosmetic conscious than the older age group.

Various studies have shown no sex differences.\textsuperscript{[10]} In our study also there was no sex difference.

Like in other studies,\textsuperscript{[11]} we also found a high incidence of onychomycosis of the toe nail. In the present study, the big toe nail was the most common toe nail involved. This is in agreement with other studies,\textsuperscript{[8,9]} possibly because of its greater size predisposing to increased trauma.

Our study revealed a mycological positivity of 94.12\% on direct microscopic examination. Other studies had reported a mycologic positivity of 63.3\%-82.82\%\textsuperscript{[7]} in clinically suspected cases of onychomycosis. A 70\% incidence of dermatophytes, especially T. rubrum in the culture-positive cases, has been reported by some authors.\textsuperscript{[11,12]} The percentage of dermatophytes isolated in

References:

1. Adhikari, et al.: Clinico-etiologic correlates of onychomycosis.

2. Indian Journal of Pathology and Microbiology - 52(2), April-June 2009
our study was close to these studies (82.35%), but we differ from these studies in that we isolated mainly T. tonsurans. We also isolated A. niger in 21.43% of the culture-positive cases and P. marneffei in 14.29% of the culture-positive cases. A. niger has been isolated in other studies. A. niger may not be significant as these organisms are ubiquitous in nature and are common contaminants in cultures. Candida albicans is reported as the most common cause of paronychial onychomycosis, but, in our study, we did not isolate C. albicans.

Another prospective study in New Delhi revealed the incidence of onychomycosis mostly in men aged 21-30 years, like in our study. But, species prevalence was quite different. They found T. rubrum and T. mentagrophytes as the main causative dermatophytes, whereas T. tonsurans was the most common species isolated, followed by T. mentagrophytes in our study.

In our neighboring country, Nepal, onychomycosis commonly caused by two cases of onychomycosis in green tea leaf pluckers. In our neighboring state, Assam, in North-Eastern India, our study was close to these studies (82.35%), but we differ from our study.

In our neighboring state, Assam, in North-Eastern India, onychomycosis in green tea leaf pluckers was caused by Scytalidium dimidiatum, a dematiaceous NDM, which was clinically indistinguishable from that caused by dermatophytes and responded poorly to antifungals.

In our neighboring country, Nepal, onychomycosis commonly affected the age group of 21-40 years, similar to our study. T. mentagrophytes (28.8%) was the most common pathogen isolated followed by T. rubrum (21.2%), T. tonsurans (11.5%), C. albicans (11.5%), Trichosporon beigelli (9.6%), Epidermophyton floccosum (7.7%), T. violaceum (5.8%), and A. flavus (3.9%).

In a study in Central India, T. rubrum and C. albicans were the major pathogens.

In another cross-sectional study in New Delhi, T. mentagrophytes was the most common isolate.

Candida was the most common pathogen, followed by dermatophytes (T. rubrum (31%), T. violaceum (5%), T. mentagrophytes (4%), T. tonsurans (2%) and E. floccosum (1%)).

In the study on human immunodeficiency virus patients in Mumbai, NDM were the predominant causative organisms. Of the 13 positive dermatophyte cultures, T. rubrum was isolated on 11 and T. mentagrophytes on two cultures.

Dermatophytes and yeast (C. albicans) were isolated in 40.8% each of the cultured nail specimens while NDM were cultured in 18.6% of the samples. Various dermatophytes cultured were T. rubrum (32.6%), T. mentagrophytes (6.1%) and T. verrucosum (2.1%), respectively. Aspergillus spp. (6.1%) was the most commonly isolated NDM while other detected molds were Acremonium spp, Fusarium spp, Scopulariopsis spp, Curvularia spp and P. marneffei.

CONCLUSIONS

The findings of the research indicated that onychomycosis is an important health problem in Sikkim with a difference in the etiological agent from the rest of India, as T. tonsurans was the most common species isolated.

LIMITATIONS OF THE STUDY

This study was carried out as part of the M.Sc. program and could not be developed and followed-up later to understand the reason behind this disparity as the resource was limited with no funding authority.

RECOMMENDATIONS

Given the findings in the present study, the following recommendations are made:

Health education
The health education system needs to improve knowledge about onychomycosis among the people by means of improving educational tools, preferably based on audiovisual techniques.

Removal of myths and misconceptions
Making people aware of recent findings to motivate the general public on clearing the myths and misconceptions about onychomycosis.

Provision of better facilities
Provision of better facilities in health care facilities and spreading awareness about the advantages of better diagnosis could be a motivating factor.

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REFERENCES

1. Achten G, Wanet Rouard J. Onychomycosis in the laboratory: Mykosen 1978;21:125-7.
2. Sujatha V, Grover S, Dash K, Singh G. A clinico-mycological evaluation of onychomycosis. Indian J Dermatol Venereol Leprol 2000;66:238-40.
3. TTrepnier EF, Amsden GW. Current issues in onychomycosis. Ann Pharmacother 1998; 32:204-14.
4. Seebacher C, Bouchara J, Mignon B. Updates on the epidemiology of dermatophyte infections. Mycopathologia 2008;166:335-52.
5. Gupta AK, Zaman M, Singh J. Diagnosis of Trichophyton rubrum from onychomycotic nail samples using polymerase chain reaction and calcofluor white microscopy. J Am Podiatr Med Assoc 2008;98:224-8.
6. Sarma S, Capoor MR, Deb M, Ramesh V, Aggarwal P. Epidemiologic and clinicomycologic profile of onychomycosis from north India. Int J Dermatol 2008;47:584-7.
7. Odom RB. Common superficial fungal infections in immunocompromised patients. J Am Dermatol 1994;31:556-9.
8. Proceeding and Transactions - 2nd international symposium on...
Adhikari, et al.: Clinico-etiologic correlates of onychomycosis

Onychomycosis. Florence, Italy. Int J Dermatol 1997;36:266-33.
9. Roberts DT. Prevalence of dermatophyte onychomycosis in UK: Results of an omnibus survey. Br J Dermatol 1992;126:23-7.
10. Gupta AK, Jain HC, Lynde CW, Watteel GN, Summerbell RC. Epidemiology of unsuspected onychomycosis in patients visiting dermatologists offices in Ontario, Canada: A multi centre survey of 2001 patients. Int J Dermatol 1997;36:783-7.
11. Williams HC. Epidemiology of onychomycosis in Britain. Br J Dermatol 1993;129:101-9.
12. Banerjee U, Sethi M, Pasricha IS. Study of onychomycosis in India. Mycoses 1990;33:411-5.
13. Jean Shadmy H, Pertz J. Deep fungal infections. In: Fitzpatrick, Eisen AI, editors. Dermatology in general medicine. vol. 2. 4th ed. McGraw Hill Inc; 1993. p. 2486.
14. Zaug M, Bergstraesser M. Amrolfine in the treatment of onychomycosis and dermatomycosis: An overview. Clin Exp Dermatol 1992;17: 61-70.
15. Sehgal VN, Aggarwal AK, Srivastava G, Gupta M, Chaudhary A. Onychomycosis: A 3-year clinicomycologic hospital-based study. Skinmed 2007;6:11-7.
16. Barua P, Barua S, Borkakoty B, Mahanta J. Onychomycosis by Scytalidium dimidiatum in green tea leaf pluckers: Report of two cases. Mycopathologia 2007;164:193-5.
17. Agarwalla A, Agrawal S, Khanal B. Onychomycosis in eastern Nepal. Nepal Med Coll J 2006;8:215-9.
18. Garg A, Venkatesh V, Singh M, Pathak KP, Kaushal GP, Agrawal SK. Onychomycosis in central India: A clinicoeotologic correlation. Int J Dermatol 2004;43:498-502.
19. Grover C, Reddy BS, Chaturvedi KL. Onychomycosis and the diagnostic significance of nail biopsy. J Dermatol 2003;30:116-22.
20. Bokhari MA, Hussain I, Jahangir M, Haroon TS, Aman S, Khurshid K. Onychomycosis in Lahore, Pakistan. Int J Dermatol 1999;38:391-5.
21. Surjushe A, Kamath R, Oberai C, Saple D, Thakre M. A clinical and mycological study of onychomycosis in HIV infection. Indian J Dermatol Venereol Leprol 2007;73:397-401.
22. Gupta M, Sharma NL, Kanga AK, Mahajan VK, Tegta GR. Onychomycosis: Clinico-mycologic study of 130 patients from Himachal Pradesh, India. Indian J Dermatol Venereol Leprol 2007;73:389-92.

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