

Clinico-Mycolological Study of Onychomycosis in HIV Positive Patients

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Abstract

Materials from clinically diagnosed onychomycosis in HIV positive individuals were subjected to potassium hydroxide (KOH) observation and cultured on Sabourad Dextrose Agar (SDA) with chloramphenicol and cyclohexamide and SDA without cyclohexamide. Growths were examined macroscopically for colony morphology, color, topography and rate of growth. Lacto phenol cotton blue(LPCB) mounts were examined microscopically; slide cultures were done wherever necessary. CD4 counts were done for all patients. The study revealed 18.18% onychomycosis in HIV positive patients. Males in age group of 20-40 and patients with CD4 cell count less than 450 cells/mm³ were much affected. Non-dermatophyte fungi are being isolated frequently from onychomycosis in HIV positive patients. Hence it is extremely important for proper treatment selection, since not all antifungal agents have a spectrum that covers all the types of fungal infections.

KEYWORDS: Onychomycosis, HIV positive patients, CD4 count.

Introduction:

Onychomycosis constitute all fungal infections affecting the nail apparatus, i.e., nail matrix, nail plate, cuticle, mesenchymal tissue and nail folds [Haley L et al 1990]. This may occur as a primary event or a secondary infection of a previously diseased or traumatized nail. The primary pathogens are dermatophytes, nondermatophytes moulds(NDM) and yeast. Clinically, onychomycosis is classified usually into five types; 1) Disto-Lateral Subungual Onychomycosis (DLSO), when the fungus invade the distal nail bed and hyponychium, developing onycholysis and later hyperkeratosis; 2) Superficial White Onychomycosis (SWO), when the fungus directly invade the nail plate surface, which results in a crumbly nail plate surface; 3) Proximal Subungual Onychomycosis (PSO), when the fungus invade under the cuticle and infect the proximal nail bed; 4) Total Dystrophic Onychomycosis (TDO), when the fungus invade the entire nail plate surface, as a result may resemble any of the clinical forms of onychomycosis 5) Candidal paronychia and onycholysis [Eleswki and Zias et al.1998&2007]. The causative pathogen and incidence of onychomycosis depends on age, gender, geographic and climatic conditions, living habits and immune status of the host[Xess and Kaur et al. 2007&2008]. Onychomycosis in immunocompromised patients, such as those infected with HIV, can pose a more serious health problem. Only a few studies have given attention to the nail infections that can occur in association with HIV infection[Valenzano and Daniel et al. 1998&1997]. The prevalence of onychomycosis in HIV infected patients has been reported to be 15-40%, all varieties of onychomycosis are more frequent in HIV positive individuals which may be related to degree of immunosuppression. Onychomycosis is most likely to develop when the CD4 cell count drops to approximately 450cells/ μ L[Goodman, Prose and Conant et al. 1987,1992&1994. The present study

was carried out to know the clinical pattern, aetiological agent and diagnosis of onychomycosis in HIV infected patients, attending the ICTC centre, Mamata General Hospital, Mamata Medical College, Khammam, which hitherto is not done.

Materials and Methods:

This study was conducted from September 2009 to July 2011. A total of 550 HIV positive patients attended to the ICTC centre, Mamata General Hospital, Mamata Medical College, Khammam, were examined for onychomycosis. Among them 100 cases were diagnosed as onychomycosis on clinical examination. The most severely affected nail was thoroughly cleaned with 70% alcohol(ethanol) and nail clippings/scrapings were collected in sterilized brown paper. Then the paper was folded to form a flat packet and it was kept in sterile container. The container was labelled with the patients name, age, sex, outpatient number and date. The sample was processed on the same day. A part of this sample was dissolved in 20% potassium hydroxide (KOH) and examined directly under light microscope for fungal mycelia and yeast. The remaining part was inoculated on Sabouraud Dextrose Agar (SDA) with chloramphenicol and cycloheximide and SDA without cycloheximide supplement. Dermatophyte Test Medium (DTM), a specific medium for isolation of dermatophytes were also inoculated. These culture slants were incubated in BOD at 25^oC and 37^oC. The cultures were observed twice a week for a period of four weeks. The positive culture slants were examined for colour of the colony (obverse and reverse), topography, texture, rate of growth and microscopic examination by Lactophenol Cotton Blue (LCB) mounts. Urease and germ tube tests were specifically done to confirm *T.mentagrophytes* and *C.albicans* respectively. For all the 100 cases CD4 cell count was done by flowcytometry.

Results:

Onychomycosis in HIV positive patients with reference to age and sex are shown in [Table.1]. Out of 100 patients 33(33%) were between 20-30 years of age, 35(35%) were between 31-40 years of age, 28(28%) were between 41-50 years of age and 4(4%) were between 51-60 years of age. Male patients in the age group of 20-40 were affected more. Toe nail involvement was seen in 62 patients(62%), finger nail involvement in 14 patients(14%), while 24(24%) patients had involvement of both finger and toe nails. Twenty six patients(26%) had TDO, 18 patients(18%) had WSO, 4 patients(4%) had PSO, while the majority i.e. 52 patients(52%) had DLSO [Table 2].

Among 100 clinically suspected onychomycosis cases, 74 were culture positive, out of these seventy four, dermatophytes isolated were 25(33.7%), non-dermatophytes in 35(47.2%) and yeasts in 14(18.9%) cases. Out of 25 dermatophytes; *T.mentagrophytes* were grown on 10 cultures(40%), *Epidermophyton floccosum* 7(28), *T.verrucosum* 4(16%) and *T.rubrum* 4(16%). Out of the 35 non-dermatophytic isolates; *Aspergillus* spp. 8(22%), *Epicoccum* spp. 8(22%), *Curvularia* 4(11%), *Fusarium* 6(17%), *Scopuloriopsis* 3(8%), *Trichosporon beigeli* 4(11%) and *Scytadium dimidiatum* 2(5%) were isolated. Out of 14 yeast isolates; *Candida albicans* 8(57%), 6(43%) were non-*Candida albicans*(NAC). These 100 HIV positive patients were subjected to CD4 cell count. Among them 79(79%) patients had counts less than 450cells/mm³, of them 63 were culture positive and 21(21%) patients had counts more than 450cells/mm³, of them 11 were culture positive. Fungal isolates

from patients with CD4 count <450cells/mm³, non-dermatophytes were 30, dermatophytes were 21 and yeast were 12. Whereas patients with CD4 count >450cells/mm³, non-dermatophytes 5, dermatophytes 4 and yeasts 2 were isolated [Table 3].

Discussion:

The incidence of onychomycosis due to dermatophytes and other molds varies from place to place.^[Godey et al.2004] Moulds are common contaminants on the skin as well as in the laboratories. Studies on onychomycosis in HIV infected patients are scanty in India[Prose and Surjushe et al. 1992&2007].

The prevalence of onychomycosis in HIV positive patients revealed in our study was 18.18%. It has been reported by other workers that the prevalence of onychomycosis in HIV infections varies between 15%-40%[Surjushe, Gupta et al. 2007,2008].

Our findings on distribution of age group and sex in HIV positive patients with onychomycosis are shown in Table 1. This corresponds to the findings of Criber 1998, Grover S 2003, Veer P 2007 and Garg A 2004 et al. The increased prevalence of onychomycosis in HIV positive patients in men compared to women could be due to more traumas to the nails and common use of occlusive foot wear. The increase in cases with age could be due to minor nail trauma, more prolonged exposure to pathogenic fungi, greater work activity and venous insufficiency[Kaur et al. 2007].^[18]

Our study revealed that toe nail infection is common [Table 2]. Our findings are consistent with other studies[Surjushe 2007, Criber 1998, Bonifaz 2007, et al. In present study, DLSO was seen in 52(52%), followed by TDO 26(26%), WSO 18(18%) and PSO 04(4%). Various studies also reported DLSO was commonest clinical pattern[Gupta 2000, Groover 2003, Veer 2007, Gupta 2007 et al.

Our study has shown that incidence of Non-dermatophyte infection was more than the dermatophyte infection, which is consistent with Criber's and Surjushe's findings. This could be because of increased susceptibility to non-dermatophyte infection in HIV-infected patients due to immunosuppression, environmental factors that favour the growth of non-dermatophytes and the ubiquity of a large and varied species of in our environment, as well as the active nature of lifestyles, which increases the vulnerability to trauma, may be probable causes. The role of *Aspergillus* spp. as pathogens has been a topic of controversy as they are commonly considered contaminants. However, various recent studies and case reports have confirmed its pathogenic role [Surjushe et al, 2007, Groover et al. 2003, Veer et al, 2007, Jaharomi et al, 2010]. In our study 14 *Candida* spp. were isolated (14%), these findings correlate with other studies[Goodman et al, 1987, Surjushe et al, 2007, Criber et al, 1998]. We found increased number of onychomycosis in HIV positive patients, where CD4 cell count was below 450cells/mm³. Thus our findings are consistent with other studies[Zias et al, 2007, Gupta et al.,2000].

Conclusion:

The clinical characteristics of onychomycosis, may be mimics to other nail disorders. The clinical diagnosis of onychomycosis should always be confirmed by laboratory tests. HIV(AIDS) is a well known predisposing cause for fungal infections including onychomycosis. Non-dermatophyte fungi are being isolated from nail

infections, more frequently from HIV positive patients. Hence it is extremely important for proper treatment selection, since not all antifungal agents have a spectrum that covers all the types of fungal infections.

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TABLE 1: Distribution of onychomycosis in HIV positive patients according to age group and gender

| Age group(years) | Male (%) | Female (%) | Total (%) |
|------------------|----------|------------|-----------|
| 20-30 | 18(18%) | 16(%) | 34(34%) |
| 31-40 | 26(26%) | 8(%) | 34(%) |
| 41-50 | 16(16%) | 12(12%) | 28(28%) |
| 51-60 | 04(4%) | 00 | 04(4%) |

TABLE 2: Clinical patterns and Nail involvement of Onychomycosis in HIV positive patients

| Clinical pattern | Finger nails only | Toe nails only | Both finger nails and toe nails | Total |
|------------------|-------------------|----------------|---------------------------------|-------|
| DLSO | 4 | 44 | 4 | 52 |
| PSO | 2 | 2 | - | 4 |
| WSO | 2 | 12 | 4 | 18 |
| TDSO | 6 | 4 | 16 | 26 |
| TOTAL | 14 | 62 | 24 | 100 |

TABLE 3: Fungal isolates and CD4 count from the cases of onychomycosis in HIV positive patients

| Isolated fungus | Total (%) | CD4 count | |
|-----------------------------|-----------|---------------------------|---------------------------|
| | | <450cells/mm ³ | >450cells/mm ³ |
| Dermatophytes (n=25) | | 21 | 04 |
| Trichophyton mentagrophytes | 10 (40%) | | |

| | | | |
|---------------------------------|--------|-----------|-----------|
| T. verrucosum | 4(16%) | | |
| T. rubrum | 4(16%) | | |
| Epidermophyton floccosum | 7(28%) | | |
| | | | |
| Non-dermatophytes (n=35) | | 30 | 05 |
| Aspergillus spp. | 8(22%) | | |
| Epicoccum spp. | 8(22%) | | |
| Curvularia | 4(11%) | | |
| Fusarium | 6(17%) | | |
| Scopuloriopsis | 3(8%) | | |
| Trichosporon beigeli | 4(11%) | | |
| Scytadium dimidiatum | 2(5%) | | |
| | | | |
| Yeasts (n=14) | | 12 | 2 |
| Candida albicans | 8(57%) | | |
| Other Candida spp. | 6(43%) | | |
| | | | |
| No growth (n=26) | | 10 | 16 |