### **CLINICO-MYCOLOGICAL STUDY OF ONYCHOMYCOSIS**

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**ABSTRACT:** Onychomycosis is defined as the fungal infection of the nail by any fungus including non-dermatophytes and yeasts. Tinea unguium is clinically defined as a dermatophytic infection of the nail plate. **OBJECTIVES:** To assess the prevalence of fungal infections in nail disorders and to find out incidence of dermatophytes, candida and other non-dermatophytic moulds. **METHODOLOGY:** History of patient was noted. The material was collected from lesions which were inoculated to the media. Slants were incubated and examined. If there was growth of dermatophytes, they were subjected to lactophenol cotton blue staining. **RESULTS:** Incidence was 100 (out of 260 nail cases). Male: female ratio was 1.3:1.69% presented with subungual hyperkeratosis. Distal subungual onychomycosis was most common (51%), proximal (46%) and superficial white (3%). Predisposing factor- occupation associated with wet work (48%). Commonly isolated species were dermatophytes (31%) in which more common was trichophyton rubrum (27%), Non dermatophytes (48%), Aspergillus species (41%), and candida albicans (4%). **INTERPRETATION AND CONCLUSION:** Finger nails were commonly affected. Associated diseases were Tinea pedis, Tinea manum, diabetes mellitus and anemia. It is a significant and important disease which can generate physical, psychological and occupational problems.

**INTRODUCTION:** Onychomycosis, a chronic fungal infection of the toe nails or finger nails, is a relatively common disease, accounting for upto 50% of all nail disorders. The prevalence of onychomycosis rises with age, with a 2-8% incidence in the general population rising to 14-28% in adults over age 60. Several risk factors for onychomycosis have been identified, including older age, Tinea pedis, cancer, psoriasis, cohabitation with family members with onychomycosis, immunodeficiency, swimming, diabetes and smoking.<sup>1, 2</sup>. Anxiety, depression, loss of self-esteem and confidence, avoidance of intimacy, and impaired relationships are among the negative impacts reported <sup>2, 3, 4</sup>. In today's world when people are becoming more aesthetically oriented, flawless skin, well groomed hair and healthy nails are of prime importance. Nails being one of the most visible part of the body, are of real aesthetic and psychological importance. Healthy nails are a thing of beauty and people take pride in flaunting them. While unhealthy and deformed nails give a psychological setback. The nail acts as a protective covering of the finger tip<sup>5</sup>.Nail disorders can be associated with both physical and psychological sequelae which may have significant adverse effects on patient's daily lives, social functioning, and mental health, which collectively involve what is known as quality of life. Hence nails form a vital part of an individual's personality.

The etymology of the name for onychomycosis comes from the Greek "Onyx" means nail, "mykes" means fungus and "osis" means condition i.e. fungal infection of the nail <sup>6, 7</sup>. Most cutaneous infections are the work of the homogenous group of keratinophilic fungi known as dermatophytes. The dermatophyte, Trichophyton rubrum is the major cause of Onychomycosis. In 20<sup>th</sup> century's developments including wars, the modern health movement, the associated use of occlusive

footwear, locker rooms and migration of people in the jet age increased the incidence of tinea pedis and onychomycosis. It is the most frequent alteration of the nails as it is in excess of 50% of the nail complaints in any dermatology consultation. This study is undertaken to know the incidence of both age and sex, morphological patterns of the disease, predisposing factors, associated conditions, distribution in finger/toenail or both, identification of fungus by direct microscopy and cultural methods to rule out the individual fungal species involved.

#### **OBJECTIVES:**

- 1. To assess the prevalence of fungal infection among cases of all nail disorders.
- 2. To find out the incidence of dermatophytes, candida and other non-dermatophytes moulds in the causation of onychomycosis.
- 3. To assess the occurrence of different fungal species, Precipitating factors, Numbers of nails involved finger/ toe nails.
- 4. To find out the age group, sex, and occupational distribution of onychomycosis.
- 5. To assess cosmetic and social problems associated with this fungal nail infection.

**METHODOLOGY:** The patients attending the outpatient department of Skin & STD, Kempegowda Institute of Medical Sciences Hospital with nail changes clinically diagnostic of onychomycosis formed the subjects for the study. The study intended for the period January 2008 to July 2009.A comprehensive history was recorded in every patient with reference to the age, sex, occupation, onset of disease, duration, season and any associated skin or systemic disease and history of various treatment etc, were noted. Clinically the nail changes were carefully defined according to their morphological features. Examination of other systems was also done in detail and the positive findings were noted. Routine investigations like, random blood sugar, blood urea (wherever necessary) and urine examination were done. The material for mycological examination was collected. The nail scraping, clippings were placed on a clean slide and mounted with a 22 mm cover slip. About two drops of KOH were added and slide was left for 6-8 hours and under 40X magnification, the hyphae and arthrospores were looked for. All samples irrespective of KOH examination results were cultured on Sabouraud's dextrose agar with cycloheximide and chloramphenicol. Also on plain Sabouraud's dextrose agar without cycloheximide but with chloramphenicol. Media was prepared as per guidelines by Conant et al.

Glucose	20 gms
Agar	35 gms
Neopeptone	10 gms
Distilled water	1000 ml

The material was inoculated to the media. The slants were incubated at room temperature and they were examined every week for growth of colonies and were discarded after 4 weeks if there was no growth the colonies were identified on the basis of macro and microscopic characteristics, the colour and morphology were studied to identify the species of fungi. In the specimens if there was obvious or suspected growth of dermatophytes, they were subjected to Lactophenol cotton blue staining and examined under microscope. **RESULT-**In the present study totally 100 clinically suspected cases of onychomycosis were studied during the period of January 2008 to July 2009.

**Age Group Incidence-**The incidence is related to number of factors of which include age, sex, social class, occupation, hobbies, underlying diseases and concomitant skin fungal infection.



In the present study the culture isolates were dominated by mould species (58%) followed by dermatophytes (31%) and candida species (8%).

**DISCUSSION:** Onychomycosis is a chronic mycotic infection of nails that affects the quality of life in a significant proportion. A number of factors have contributed to the growing incidence of fungal nail infections, including aging population, an expanding number of immune compromised patients and increasing participation in fitness related activities. The total number of outpatient attendance during the period was 36, 198, of which 260 were the cases of nail abnormalities which were clinically diagnosed as onychomycosis. Among these, 38 cases were presented with nail abnormalities. This finding correlates with Powel et al. (1900) study who observed 14.9% of all ringworm infections in which nails were involved abnormally.

This study shows that out of 280 patients, 100 were clinically diagnosed as onychomycosis and included in the study. This study shows the overall incidence of onychomycosis was 2.58%, of

all dermatophytosis cases. Puri DKK et al. (1978) states that onychomycosis constitutes 10% of cases of dermatophytosis. Whereas BP Lalithamma et al. states 5.45% as the incidence of Tinea unguium of all (550) clinically suspected cases of dermatomycoses <sup>8</sup>. The overall prevalence of the disease in this study is about 0.72% and nail abnormalities were present in 0.1% on hospital based manner. Madhuri et al. (2002)<sup>9</sup> study concurs with this study, who showed 0.94% as the prevalence rate of the disease, whereas Eleweski (USA, 1996) and Evans and Robers (UK, 1996 and 1992) reported a prevalence rates of 8-9%, 5% and 2.7% respectively in their general population. This study shows the incidence of disease is more from rural area presenting 60% (36 patients) of study included cases. Madhuri et al. (2002) also reported very high prevalence of the disease from rural areas (94%). In this study, the highest incidence was seen between age groups of 21-40 years i.e. about 47% with maximum between 21-30 years (25%). Similarly observed in studies of Madhuri et al. (2002) and Reddy et al. (1982) showing maximum incidence of the disease between age group of 21-30 years. This study shows the incidence of disease more in male patients (57%) than females (43%). Male preponderance observed in this study is similar to that reported by authors Reddy et al. (1982) – 68.9%, and Sujatha et al. (2000) showed Male: female ratio of 1.33:1. In contrast Madhuri et al. (2002), Mercantini et al. (1996), Lopes (1999) and Bokhari (1999) reported a higher prevalence of 51.96%, 72.1%, 62.7% and 72% respectively among women. This study shows finger nails, mainly right hand especially thumb, index and ring fingers were frequently involved than toenails in both sexes. This observation correlates with the studies of Madhuri et al. (2002), Reddy et al. (1982), Velez et al. (1997), Rigopououlos et al. (1998), Sujatha et al. (2000) who also reported an increased involvement of finger nails. In our country finger nails especially the thumb nails is used as a multipurpose tool for jobs such as peeling vegetables, feeding children, etc. making them more prone to injuries and infections. Open footwear could explain the decreased incidence of toenail involvement. In this study, patients with toenail onychomycosis are often associated with Tinea pedis, this observation also correlates with Cheng and Chong study. This study shows the duration of disease ranging from 1 month to 48 months with maximum cases complaining between 1 year and <1 year duration (65%) with a mean duration of 13.9 months. Similarly observed in Madhuri et al. (2002) study, showing the disease duration in their cases ranging between 3-15 years, out of which 38.23% of patients were of less than 1 year duration. Seasonal variations of disease among patients were not able to observe in this study, and most of the patients presenting with the disease of longer duration were not able to correlate the disease with climate and season. Among 6 clinical types, common clinical type presented in the study was DSO, reporting in 51%. This clinical type was also reported as the commonest one in studies of Reddy et al. (1982) and Sujatha et al. (2000) <sup>10</sup>, Banerjee et al. (1990). In contrast Madhuri et al. (2002)<sup>9</sup> reported candidal onychomycosis as the commonest type in their study. In this study the second commonest clinical type reported is followed by proximal subungual onychomycosis in 46% and superficial white onychomycosis in 3%. In contrast Madhuri et al. (2002)<sup>9</sup> study reported distal subungual onychomycosis as the second common, followed by white superficial onychomycosis and proximal subungual onychomycosis. In this study common clinical presentation observed in all types of onychomycosis was nail discoloration (18%) and subungual hyperkeratosis (16%). This concurs with the study done by Reddy (1982) et al. In this study, the common predisposing factor preceding to the development of onychomycosis observed were occupation associated with wet work (27%) including housewives – 25%, employee – 22%,

labourer 15%, student – 5%, agriculturist – 10%, transport – 5%, businessmen – 5%. Patients with increased physical activity were, labourers – 15%, agriculturists – 10%, housewife – 25% and others (businessmen – 5%, students – 17%, sportsman – 1%). The above stated predisposing factors in this study almost correlate with the study reported by Madhuri et al. (2002).Domestic activity associated with wet work associated with constant trauma could explain the probable reason for the high prevalence of onychomycosis in wet work occupational workers (43.3%). Increased physical activity (26.6%) with trauma facilitating easy entry of the fungal pathogens could probably explain the second contributing factor. The above all factors almost correlates with the study of Madhuri et al. (2002) regarding the contributing factors for the predisposition of the disease.

Superficial white onychomycosis, observed in 3 patients in this study had the habit of wearing shoes regularly, which supports the observation of English MP (1976) <sup>11</sup> that superficial white onychomycosis (SWO) occurs in toe nails of predominantly shoe clad population. According to observation done by Ploysangan T et al. (1997)<sup>12</sup>, PSO due to trichophyton rubrum may extend to superficial nail plate producing a clinical picture that resemble SWO which is exclusively seen in finger nails. This observation is similarly noted in these patients. According to Timorthy Berger et al. (1995)<sup>13</sup> in areas of high HIV endemicity onychomycosis is often recognized as a marker of HIV infection.

Most commonly associated dermatosis in this study was fungal infection over other body areas which were seen in 13 (13%). Similarly observed in Madhuri et al. (2002) study. The other associations according to frequency are diabetes mellitus (11.6%), acne (5%), hyperhidrosis (3.33%), fissure foot (3.33%), Paronychia (3.33%), verruca vulgaris (1.66%), scabies (1.66%), melasma (1.66%), PLE (1.66%), infective eczema (1.66%), LSC (1.66%), HIV with pulmonary tuberculosis (1.66%), leprosy (ENL on high steroids) (1.66%).

Dogra S Kumar et al <sup>14</sup> study shows the prevalence of onychomycosis in diabetic patients were about 17%. In our study, it showed about 1.6%. Out of 60 patients, mycologically positive by direct microscopic results were positive in 50 cases (83%) showing fungal elements as elongated, branching, septate, light retractile structures. The KOH positive result rate in this study similarly correlates with the study of Sujatha et al. (2000)<sup>10</sup> showing who showed 67% as direct microscopic positive result. In this study culture positive result is obtained in 40 cases (67%) who were also gained positive results with KOH examination. Culture positive rates were approximately similar to observations made by Reddy et al. (1982)<sup>15,</sup> who showed 68.8% as culture positive rate. In contrast Madhuri et al. (2002) even high rates of both direct microscopy and culture positive reporting in 93% and 87.2% respectively. Isolated fungi obtained by cultural methods in this study are the dermatophytes 31% (Trichophyton rubrum – 27% and Epidermophyton – 4%). Reddy et al. (1982)<sup>15</sup>, Banerjee et al. (1990) <sup>16</sup>, Bokhari et al. (1999), Gupta AK also reported dermatophytes as the common isolated fungal species in their study. In this study other organism isolated were the yeast and only four species is identified i.e. Candida albicans (20%). In contrast Madhuri et al. (2002), Actheten (1978), Al-Sogair (1991), Rigopuoulos (1998) reported a higher isolation rate of Candida species in their respective studies. In this study, non-dermatophytic moulds were Aspergillus species were identified in study, which are Aspergillus niger (26%) and Aspergillus flavus.

**CONCLUSION:** 18% of patients presented with nail discoloration and 16% of patients presented with subungual hyperkeratosis. Finger nails were commonly affected in both the sexes and most commonly the dominant hand (right) fingers. Commonly associated dermatoses in this study were Tinea pedis, Tinea manum and diseases were diabetes mellitus and anemia. Careful precautions should be taken while inoculating the material into the media and also during the period of growth. Although onychomycosis is not usually a life threatening, it can be a source of significant pain and discomfort; it can also pose a risk for patients, their families and others in contact with them. Onychomycosis can no longer be considered a simple cosmetic nuisance confined to the nails. It is a significant and important disease, which can generate many physical, psychological and occupational problems considerably impairing patient's quality of life.

**SUMMARY:** A clinically diagnostic study of 100 cases of untreated onychomycosis attending Outpatient Department of Skin and STD and other confining Department Wards was undertaken from January 2008 to July 2009.During this period, the incidence of Onychomycosis attending the department 100 (out of 260 abnormal nail cases) out of 36, 198 comprises for 0.1%. In this study of 100 patients, of clinically diagnosed onychomycosis, there were 57 males and 43 females with a ratio of 1.3:1. Their age varied from 15 years to 70 years. Majority of the patients were in 21-40 years age group. 25% in 21-30 years, 22% in 31-40 years. Around 5% of the patient showed positive family history. Most of the patients are from rural areas. 11.7 (7 patients) were associated with pain and 69% of patients presented with subungual hyperkeratosis.

Duration of the disease varied from 1 month to 48 years and with a mean duration 13.9 months. Distal subungual onychomycosis was the most common clinical type (51%) seen, followed, proximal subungual onychomycosis in (46%) and superficial white onychomycosis in (3%). Around 11% were associated with pain/discomfort. Common predisposing factors were the occupation associated with wet work (48%) and increased physical activity (26%).

Direct microscopy examination of half of the nail scrapings/clipping samples mounted with 20-30% KOH demonstrated fungal elements in 87 (87%) patients. Out of 100 patients nail samples, positive results were obtained in 94 patients (94%). Common isolated species were the dermatophytes (31%) in which more commonly isolated fungus was Trichophyton rubrum 27 (27%). Followed by non-dermatophytes which were isolated in only 48 cases. Aspergillus species like e.g. Aspergillus niger 26 (26%) and Aspergillus flavus 15 (15%) were identified. Next common isolated fungal species were yeasts and only Candida albicans (4%) is identified. Negative result by direct microscopic examination (6 cases) and culture negativity in 6 cases indicates that more strict precautions are necessary, while collecting the nail samples. Proper collection of nail samples is needed to avoid false negative results as well as to eliminate contaminants and it is important to collect more nail material as nail may contain only a few fungal element. Histopathological examination was undertaken in 8 cases in which 6 cases showed positive results showing long slender hyphae and spores and 2 cases which were negative were subjected for special stain i.e. Grocott's staining. One stained positively by this special stain showing short slender hyphae.

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Sex	No.	%
Male	57	57
Female	43	43
Table 1: Sex incidence		

Occupation	NO.	%
Employee	22	22
Transport	5	5
Labour	15	15
Student	17	17
Housewife	25	25
Sports	1	1
Agriculturist	10	10
Businessmen	5	5
Table 2: Occupation		

Aggravating factors	No.	%
No	52	52
Moisture	27	27
Sweating	7	7
Warmth	5	5
Tight footwear	9	9
Total	100	100
Table 3: Aggravating factors		

Finger nail infection occurs more commonly in women.

In this study the maximum incidence of Onychomycosis was seen in the age group of 21-30 years.

No.	Minimum	Maximum	Mean
100	1.00	48.00	13.9360
Table 4: Duration of infection			

In our study the duration of infection varied from 1 months to 48 months, but majority of the cases were within 14 months.

In this study the predominant clinical signs were, discolouration (18%) and subungual hyperkeratosis (16%) and with discoloration and subungual hyperkeratosis (35%), making totally (69%).

<b>Clinical types</b>	No.	%
DSO	51	51
PSO	46	46
SWO	3	3
Total	100	100
Table 5: Clinical types		

Of the 100 cases examined in the present study distal subungual Onychomycosis commonest (51%) followed by proximal subungual onychomycosis (46%) followed by superficial onychomycosis (3%).

КОН	No.	%
Negative	33	33
Positive	67	67
Total	100	100
Table 6: KOH		

The high incidence of culture positivity in this study could be because of the fact that the scrapings were subjected to culture despite negative KOH reporting and that the scraping were subjected to culture and subculture more than once in some cases.

Fungal species	No.	%
Negative	6	6
Aspergillus flavus	15	15
Trichophyton rubrum	27	27
Aspergillus niger	26	26
Candida	4	4
Rhizopos	4	4
Geotrichum	4	4
Contaminants	1	1
Epidermophyton	4	4
Pencillium	2	2
Fusarium	5	5
Scytalidium	1	1
Alternaria	1	1
Total	100	100
Table 7: Distribution of various fungal species		

In the present study in Distal subungual Onychomycosis moulds were isolated in 14 cases and dermatophytes were isolated in 27 cases. The proximal subungual type was showing a high predominance of Mould species in 55 cases and followed by dermatophytes in 6 cases. In the 4 cases of Candidal Onychomycosis Yeasts species were isolated in 4 cases



DLSO associated with hyperkeratosis and onycholysis



Total dystrophic onychomycosis due to T. rubrum



PWSO due to Fusarium



TDO with Paronychia



T.RUBRUM-Macroscopic appearance of the colony



Scopulariopsis brevicollis – Macroscopic appearance of the colony three weeks after inoculation



FUSARIUM SOLANI; macroscopic appearance of the colony one week after inoculation

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