Epidemiology of Dermatophytes and Non Dermatophyte Molds among Patients Attending Rank Higher Specialized Dermatology Clinic, Addis Ababa, Ethiopia

Shambel Araya (✉ shambel.araya@aau.edu.et)

Addis Ababa University, College of Health Sciences <https://orcid.org/0000-0002-0109-6062>

Betelhem Tesfaye

Rank Higher Specialized dermatology clinic

Desalegn Fente

Addis Ababa University, College of health science, Department of Medical Laboratory

Research article

Keywords: Dermatophytes, Non dermatophytes, Dermatophytosis, fungi

Posted Date: July 9th, 2019

DOI: <https://doi.org/10.21203/rs.2.11152/v1>

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Abstract

Background: Dermatophytes are keratinophilic fungi that infect keratinized tissues causing diseases known as dermatophytosis. Dermatophytosis is common worldwide and continues to increase. **Objective:** This study was undertaken to determine the prevalence of dermatophytes and the spectrum of fungal agents in patients attending Rank higher clinic. **Methods:** a cross sectional study has been conducted, in which 318 Samples were collected from patient's hair, nail and skin. A portion of each sample was examined microscopically and the remaining portion of each sample was cultured onto plates of Sabouraud's dextrose agar containing chloramphenicol with and without cycloheximide. Dermatophyte isolates were identified by studying macroscopic and microscopic characteristics of their colonies. **Result:** Of 318 samples, fungi were detected in 133 (54.4%) by direct wet mount while 148/315(46.5%) of them were culture positive. From these 72/148(46.8 %) were dermatophytes. Among dermatophyte isolates *T. tonsurans* 29/72(40.2%) was the most common cause of infection. Tinea capitis was the predominant clinical manifestation accounting for 170/315(53.4%) of the cases. Patients with age group 1-14 years were more affected. *T. tonsurans* was the most common pathogen in tinea capitis, whereas *T. mentagrophytes* was the most common pathogen in tinea corporis. **Conclusion:** In this study the prevalence of dermatophytes were higher. Further intensive epidemiological studies of dermatophytes induced dermatophytosis which have public health significance are needed. **Key words:** Dermatophytes, Non dermatophytes, Dermatophytosis, fungi

Background

Dermatophytosis represents one of the common infectious diseases worldwide and causes serious chronic morbidity, especially in developing countries [1]. The condition is caused by dermatophytes, which are fungi basically molds that require keratin for growth. An increase in the incidence of dermatophytosis has been noted worldwide, especially in developing countries [2, 3]. In particular, tinea capitis one of the most common dermatophytosis represents a major public health issue among children in developing countries; mainly in children's of African or Caribbean origin [3].

Geographic location, health care, immigration, climate (temperature, humidity, wind, etc.), overcrowding, environmental hygiene culture, awareness to dermatophytes , age of individuals, hygiene and socioeconomic conditions have been described as major factors for these variations of dermatophyte epidemiology [4, 5,6,7]. As human contact among children is more frequently between the ages of 4 and 16 years than in very early childhood; this age group is similarly at greater risk of contracting infectious diseases from different sources [8, 9, 10].

Dermatophyte fungi have a worldwide distribution, and now days, there are about 40 known species in the genera of dermatophytes. Of these species, about 25 species belong to the three most recognized and prevalent worldwide genera's; *Epidermophyton*, *Microsporum* and

Trichophyton and these genera's are presently known to infect humans [7, 11, 12].

Ethiopia is a developing nation located in the tropical region with wet humid climate which makes it to fall into regions with high prevalence of dermatophytosis. So, conducting further studies to know the actual magnitude of dermatophytosis as well as the spectrum of its etiological agents among the general population is of the highest priority [9, 10].

Methods

Study areas and population

A prospective cross-sectional institution based study was carried out from January 2018 to June 2018 among dermatophytosis suspected patients who visit Rank higher specialized dermatology clinic, Addis Ababa, Ethiopia.

Sample size determination and sampling technique

The required sample size of the study was determined using the formula for single population proportion. A total of 318 clinical samples were collected from patient visiting the Dermatology clinic during the study period were included in the study. Single specimen was taken from each patients based on their clinical manifestation.

Biological sample collection

The samples were collected from January 2018 to June 2018 using convenient sampling technique. Before collecting the sample the infected area was cleaned with 70% (v/v) ethanol. Skin, nail, and finger scrapings were collected aseptically using sterile blades and transferred into sterile plastic petri-dishes. In tinea capitis suspects, dull broken hairs from the margin of the scalp lesion with forceps was sampled and transferred to sterile folded papers and transferred in to sterile petri dishes.

Culture and microscopic examination: A portion of each sample from a nail, finger skin and scalp scraping was mounted in a drop of 10% (w/v) potassium hydroxide on a clean microscopic slide. After 5 minutes; the mount preparation was examined under low ($\times 10$) and high ($\times 40$) power magnification for the presence of any fungal elements. The remaining portion of each clinical sample from the different sites was cultured in to Sabouraud's dextrose agar plates containing chloramphenicol with and without cycloheximide (Oxoid, Basingstoke, England) prepared according to the instruction of the manufacture. All inoculated plates were then incubated at inverted position at 25–30 °C for 4–6 weeks. Culture plates containing the Sabouraud's dextrose agar were examined twice a week for any fungal growth. In the absence of growth during 6th week, the results were considered negative. Those suspected colonies for dermatophytes were sub cultured into potato dextrose agar for the production of spores. Cultures of dermatophytes were identified by examining macroscopic and microscopic characteristics of their colony on Sabouraud's dextrose agar. For the macroscopic identification; rate of growth, texture, topography, and pigmentation of the reverse and front side of the culture were employed. Mold isolates was identified microscopically by placing pieces of the colony from Sabouraud's dextrose agar plates in to clean

microscopic slide and stained using lacto phenol cotton blue stain. Each preparation was observed microscopically after placing a cover slip. In addition, urease test was also used in differentiating between some members of *Trichophyton* species. For the identification of yeasts, candida CHROME agar was also used.

Statistical analysis

Data was collected, double entered, cleaned and analyzed using SPSS version 20 software according to the study objectives. Frequency and percentage were used for investigation of the outcome.

Result

A total of 318 dermatophytosis suspected patients were included in this study. Among these; fungi were detected in 133(41.8%) samples using potassium hydroxide wet mount while, 148 (46.5%) were culture positive (Table 1).

In the present study a total of 318 clinical samples were collected from suspected cases of dermatophytosis of which 125 (39.3%) were males and 193(60.7 %) females (Table 2).

The age of the study subject was ranged from 1 to 88 year with a mean age of 16 year. Dermatophytic infection rate was high in the age group of 1-14 years 144/318 (45.2%), followed by the age group of 25-44 years with 96/318(30.1%) patients, and old age group (>81 years) with only 7 (2.2%) (Table 3).

A total of 148 fungi were recovered. Of these, 72/148(48.6%) were dermatophytes, 63/148(42.56 %) non-dermatophyte mold and (13/148) 8.7% were yeasts. *T. tonsurans* was the predominant dermatophyte followed by *T. Mentagrophytes* and *M. audouinii*. Among non dermatophyte molds *Cladosporium spp* was the predominate isolate followed by *Neoscytalidim dimidatum* and *Alternaria spp* respectively.

Yeasts were the least common isolates accounting 13(8.7 %) of the total suspects of dermatophytosis (Table 4.)

Further identification of dermatophytic fungi showed the presence of *Trichophyton mentagrophytes*, *Trichophyton rubrum*, *Trichophyton tonsurans*, *Trichophyton soudanense*, *Trichophyton violaceum*, *Trichophyton verrucosum*, *Trichophyton schoenleinii*, *T. tonsurans* and *Microsporum audouinii*. Among all the dermatophyte isolates *T. tonsurans* was the most common (19.59%) cause of infection, followed by *T. mentagrophytes* (8.78%) and *M. audionii* (8.78%), whereas *Trichophyton schoenleinii* (0.67%) was the least common (Table 4). Clinical manifestation in relation to age group depicted that patients with age group 1-14 were highly affected accounting for 45.2 % of the cases followed by age group 25-44 years accounting for 30.1 %. Tinea capitis was found to be more in patients of age group 1-14 years and tinea corporis in patients of age group 45–64 years. Tinea unguium was common in patients of age group of 25–44 years (Table 2).

Tinea capitis was the predominant clinical manifestation accounting for 53.4% of the cases; of which 98 (57.6%) were females and 72 (42.4 %) males. This was followed by tinea corporis and tinea capitis accounting for 30.5% and 16% of the cases, respectively (Table 5). According to species frequency in different areas of involvement, *T. tonsurans* was the most common pathogen in tinea capitis, whereas *T. mentagrophytes* was the most common pathogen in *tinea corporis* (Table 5).

Discussion

Accurate diagnosis based on the clinical symptoms alone is often difficult. Currently, the diagnosis of dermatophytosis is confirmed by clinical examination and screening of the collected clinical specimen by direct microscopy and fungal culture [13]. An accurate diagnosis of dermatophytosis is important for its successful treatment. The risk of developing adverse drug reactions, the cost and long duration of the therapy, and possible interactions with concomitant medications all affects the importance of accurate diagnosis of the condition before commencing therapy [13, 14]. In the present study, direct microscopy positivity rate is 41.8% and culture positivity rate is 46.5 % (Table 1). This was in line with study by Tekleberhan *et al*, in Ethiopia, found that 31.1% and 42.6% using potassium hydroxide direct wet mount and culture results respectively [9]. This high prevalence could be due to that; Ethiopia is a tropical country with wet humid climate, large population size, and low socioeconomic status and this is supported by other studies conducted on dermatophytosis etiologies and risk factors [15, 16].

The most susceptible persons to tinea capitis were among patients 1-14 years (45.2%) this could be because of the lack of protective fatty acids in their scalp. This infection was rarely reported in persons above fifty years of age. Earlier, several authors have supported this finding [17, 18]. Many cases occurring in adults is involved with hormonal disorders resulting in carryover of tinea capitis from childhood or in patients with severe immunodepression due to leukemia, lymphoma, or treatment with immunosuppressant drugs [19, 20].

From 46 species of Fungi causing tinea capitis; *T. tonsurans* 26/46(56.5%) were the most common dermatophytes followed by *M. audouinii* (8/46(17.3%) and *T. verrucosum* 4/46(8.6%). A similar study conducted in Kenya supports our present finding with *Trichophyton tonsurans* (45.3%) being the most prevalent followed by *T. verrucosum* (4.3%) and *Trichophyton rubrum* (4.3%) being the least [25]. Even though it is not in line with the current finding; a similar study in Ethiopia showed that the prevalence of *T. tonsurans* in the rate 18.4% [9].

However, in a recent study conducted by Bitew A, 2018 in Addis Ababa, Ethiopia *T. violaceum* was the dominant causative agent of tinea capitis [21] and this study shows dissimilarity results with the current study. But other studies showed that species of *T. tonsurans* are circulating in the population. In a study conducted by Raccurt *et al*, 2009; three anthropophilic species were identified (*T. tonsurans* 63.6%; *M. audouinii* 12.7%; *T. rubrum* 7.3%), one zoophilic species (*T. mentagrophytes* 14.5%), and one geophilic species (*M. gypseum* 1.8%).. Social phenomena of today on the continent could explain the emergence of

T. tonsurans in different areas. Recent studies underline the fact that *T. tonsurans* is highly contagiousness and the role of children scalp for tinea capitis [22, 23].

In developing countries such as Kenya and other sub-Saharan countries, the most common agent is *M. canis* followed by *T. tonsurans*. *Trichophyton* species had the highest prevalence of 61.3%, with *T. tonsurans* being the most predominant dermatophyte due its ubiquitous in nature and abundance among human carriers [20, 26, 27]. This could show that *T. tonsurans* is spreading in Ethiopia.

The anthropophilic *Microsporum* species cause a contagious disease and they are endemic in many countries. In the current study; Tinea capitis is the most common clinical disease followed by tinea pedis and tinea corporis. The zoophilic *Trichophyton* and *Microsporum* species are seldom responsible for more than minor outbreaks of human infections. *T. mentagrophytes*, *T. verrucosum*, *T. tonsurans*, *T. violaceum*, and *M. audouinii* species are causal agents of tinea capitis [24, 25].

Tinea corporis was the second most common infection among the enrolled patients with a significant incidence among age groups of 25-44. The site of infection was mostly restricted to face and neck. *T. mentagrophytes*, *T. rubrum* and *M. audouinii* were the main causative agents. In a study conducted by Teklebirhan et al 2015; tinea unguium was the dominant clinical manifestation involving 51.1% of the total cases of dermatophytosis. [9]. But in our study Tinea corporis was the second common clinical presentation accounting for 33 (10.8%) next to tinea capitis and this is in line with study conducted in Harari regional state, Ethiopia [28].

Tinea unguium was also observed mainly on the age of 15-24 (Table 4). Since onychomycosis infections in children is not common due to many reasons such as; rapid growth of the nail, have less exposure to fungal infection risk factors than adults such as pedicure and manicure repeated aggressiveness, frequent housework and cosmetic reasons [16, 29]. So, the high occurrence of tinea unguium in this study among these age groups could be due to such factors. But in other study more males and elderly patients were highly affected than the adults [30].

Non dermatophytic molds were isolated from 63 cases (42.56%) with *Cladosporium spp.* as a major isolate accounting 30% of the total non dermatophyte mold isolates. Similar recent study done in Ethiopia supports this finding [21]. *Cladosporium* species are dematocious fungi, ubiquitous and they are infrequently associated with human and animal opportunistic infection. It is also the most widely spread fungi in the world. It is true also; most of the time appears as a contaminant. But some studies showed that they are associated as opportunistic infection in subcutaneous and disseminated form, especially among immune depressed individuals [32]. *Neoscytalidium dimidiatum* was the second most common isolated fungi from the non dermatophytes. They were isolated from skin and nail scrapings predominantly of toenails. *Neoscytalidium dimidiatum* and *Scytaalidium hyalinum* are common causative agents of human superficial infections in different parts of the world especially in tropical and subtropical region [21]. Similarly, yeasts were isolated from 13 cases with *C. albicans* as a major isolate accounting 38.4% of the total yeast isolated and this study was similar with study conducted in Saudi Arabia [33]. Non dermatophyte fungi were isolated as a cause dermatophytosis in many studies [33, 34].

Conclusion: This study showed that the prevalence of dermatophytosis was 46.5% which is more or less similar to study conducted in Ethiopia [9] but in developed countries showed that less than 5% [35] which indicates that dermatophytosis is still a common problem in developing countries. Tinea capitis was identified as the most prevalent clinical presentation and children's are the most vulnerable group. This study found that *T. tonsurans* was the most common etiologic agent followed by *M. audouinii* and *T. mentagrophytes*. We recommend further research on the possible risk factors and spectrum for dermatophytes and Non dermatophyte Molds in large clinical setting and the community.

Declarations

Abbreviations

Not applicable

Ethics approval and consent to participate

Ethical clearance for this study was provided from Medical Laboratory science, college of health sciences, Addis Ababa University ethical review committee. Verbal informed consents were also obtained from participants. Assent form was completed and signed by family member and/or adult guardian for participants under the age of 16 years.

Consent for publication

Not applicable as details like; videos or images related to study subjects were not recorded for this study.

Funding

The study was supported by Addis Ababa University, College of health science, department of medical laboratory science. The funder had no role in data collection, study design, data analysis and interpretation.

Availability of data and material

The data sets used or analyzed during the current study are available from the corresponding author on reasonable request.

Competing of interest

The author's listed above declare that there is no competing of interest.

Authors' contributions

SA, BT and DF had participated in culture media preparation, identification of bacterial pathogens, have given final approval of the version to be published; analysis and interpretation of data, and agree to be accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of

any part of the work are appropriately investigated and resolved. All authors participated in this research have read and approved the manuscript.

Acknowledgments

We thank Dr. Zemenu Tamir, Daniel Kahase for proof reading and revision of the manuscript. We also thank the help of Dr. Adane Bitew; Addis Ababa University, Medical laboratory department staff for helping the laboratory activities. We are also thanking the study participants.

STROBE

Not applicable

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Tables

Table 1: correlation of direct microscopy with culture (N=318)

Test procedure	Number	Percentage
Direct wet mount (KOH) positive	133	41.8
Culture positive	148	46.5
Direct wet mount((KOH) negative culture positive	72	22.3
Direct wet mount positive (KOH) culture negative	54	16.9
Both Direct wet mount (KOH) and culture positive	76	23.8
Both Direct wet mount (KOH) and culture negative	111	34.9

Table2: Frequency and distributions of clinical manifestations in relation to sex ($n = 318$).

Clinical manifestation	Male	Female	Total n(%)
Tinea capitis	72	98	170(53.4)
Tinea unguium	13	38	51(16.0)
Tinea corporis	40	57	97(30.5)
Total	125(39.3%)	193(60.7%)	318(100%)

Table 3: Frequency of clinical manifestation in different age groups (n=318)

	Age						
Site	1-14	15-24	25-44	45-64	≥65	Total	n(%)
T. Capitis	115	11	34	9	1	170	53.4
T. corporis	21	14	43	14	5	97	30.6
T. unguium	8	16	19	7	1	51	16.0
Total	45.2%	12.8%	30.1%	9.4%	2.2%	100%	

Table 4: Frequency and distribution of dermatophytes and Non dermatophyte Molds

Fungal category	Species	Number	Percentage
Dermatophytes n=72/148(48.6%)	<i>Trichophyton schoenleinii</i>	1	0.67
	<i>M.audouinii</i>	13	8.78
	<i>T.tonsurans</i>	29	19.59
	<i>T.mentagrophytes</i>	13	8.78
	<i>T.rubrum</i>	6	4.05
	<i>T.violaceum</i>	1	1
	<i>T.verrucosum</i>	4	2.70
	<i>T.soudanense</i>	4	2.70
Non- dermatophytes n=63/148(42.56%)	<i>Cladosporium spp</i>	21	14.18
	<i>Colletotricum spp</i>	1	0.67
	<i>Exserohilum spp</i>	1	0.67
	<i>Stemphylium</i>	1	0.67
	<i>Neoscytalidium dimidatum</i>	11	7.43
	<i>Nigrospora spp</i>	1	0.67
	<i>Alternaria spp</i>	9	6.08
	<i>Fusarium spp</i>	6	4.72
	<i>Scopulariopsis brevicualis</i>	6	4.05
	<i>Paecilomyces spp</i>	1	0.67
	<i>Aspergillus fumigatus</i>	1	0.67
	<i>Geotricum condidum</i>	1	0.67
	<i>Aspergillus niger</i>	1	0.67
	<i>Bioplaris</i>	1	0.67
	<i>Helmentho spoium</i>	1	0.67
Yeasts n=13/148(8.78%)	<i>C.cruzie</i>	3	2.02
	<i>C.albicans</i>	5	3.38
	<i>C.glbrata</i>	3	2.02
	<i>R.mucilainosa</i>	2	1.35
Total		148	100

Table 5: Frequency and distribution of dermatophytes in relation to clinical manifestation

	Clinical manifestation			Total n(%)
	Tinea capitis n(%)	Tinea corporis n(%)	Tinea unguium n(%)	
<i>phyton schoeninii</i>	1(2.1)	0	0	1(1.3)
<i>douinii</i>	8(17.3)	5(23.8)	0	13(18)
<i>urans</i>	26(56.5)	2(9.5)	1(20)	29(40)
<i>tagrophytes</i>	3(6.3)	7(33.3)	4(80)	14(19.4)
<i>um</i>	2(4.2)	4(19)	0	7(9.7)
<i>aceum</i>	1(2.1)	0	0	1(1.3)
<i>ucosum</i>	4(8.4)	0	0	4(5.5)
<i>danense</i>	1(2.1)	3(14.2)	0	4(5.5)
	46(100%)	21(100)	5(100)	72(100)