Iranian J Publ Health, Vol. 42, No.3, Mar 2013, pp.320-325

Original Article

The Incidence of Onychomycosis Infection among Patients Referred to Hospitals in Kermanshah Province, Western Iran

Ali MIKAEILI [1]***,* Isaac KARIMI*** [2]

1. *Dept. of Medical Mycology, School of Medicine, Kermanshah University of Medical Sciences, Kermanshah, Iran*
2. *Laboratory of Molecular and Cellular Biology, Department of Basic Veterinary Sciences, School of Veterinary Medicine, Razi University, Kermanshah, Iran*

***Corresponding Author:** Email: isaac_karimi2000@yahoo.com

(Received 12 Oct 2012; accepted 11 Jan 2013)

Abstract

Background: Onychomycosis is a nail disorder associated with aesthetic problems, discomfort, physical injury and loss of dexterity. The purpose of the present study was to isolate and identify the causative fungi of onychomycosis in 2402 patients in Kermanshah Province, western Iran in 1994 to 2010.

Methods: Mycologic assessment was carried out by standard methods including either microscopic or cultural procedures.

Results: Direct microscopy of the nail clips was positive in 1086 (45.2%) and fingernail and toenail onychomycoses were recognized in 773 (71.1%) and 313 (28.8%), respectively. Yeasts were detected in 853 (78.5%), dermatophytes in 201 (18.5%) and non-dermatophyte fungi in 32 (2.9%) patients. The results of fungal culture showed *Candida albicans* isolated in 384 (45.0%) and other *Candida* spp. isolated in 361 (54.0%) of the cases as the most common agents of onychomycosis while among dermatophytes, *Trichophyton rubrum* was found in 63 (37.0%) of the cases as the main dermatophytic agent followed by *T. mentagrophytes* 32 (15.9%) and *Epidermophyton flocosum* 30 (17.6%). Among the non-dermatophyte moulds, *Aspergillus flavus* was the most prevalent species 12 (37.5%) followed by *A. niger* 8 (25.0%) and *A. fumigatus* 4 (12.5%). Moreover, 139 (12.8%) samples with positive direct microscopy yielded no growth. The highest rate of onychomycosis was found in patients between 30-40 years of age.

Conclusion: In sum, the current results identified the aetiological agents and primary epidemiological aspects of onychomycosis in west Iran.

Keywords: *Onychomycosis*, Fungal diseases, *Candida* spp., *Trichophyton* spp. Iran

Introduction

Onychomycosis is a common fungal infection affecting both fingernails and toenails and represents up to 20% of all nail disorders and 30% of all superficial fungal infections of the skin (1). The worldwide incidence of onychomycosis is increasing and a number of factors such as diabetes mellitus, poor peripheral circulation, immunodeficiency, adverse drug reactions, nail trauma and genetic defect contribute to this rise (1). Onychomycosis, though not a serious disease in terms of morbidity and mortality causes significant psychological aesthetic distress and therapeutic difficulties. The prevalence rate of onychomycosis is determined by age, predisposing factor, social stratum, occupation, climate, living environment and frequency of travel (2). The causal agents of onychomycosis include three groups of fungi: yeasts, dermatophytes and non-dermatophyte moulds. However, there are distinctive geographic differences in the epidemiology and aetiology of onychomycosis; especially in the frequency with which each group of fungi is responsible for the

infection (3). Thus mycological studies are very important in order to define the aetiology of onychomycosis for applying a proper treatment.

The epidemiology of onychomycosis in different parts of globe has been subject of several studies that shown various aetiologies and predisposing factors are involved in the prevalence of onychomycosis (e.g., 4-6). The major defect of these studies is traditional comparison of epidemiology and aetiology of onychomycosis of unrelated regions. Therefore, the region- or even country-based comparisons of prevalence of onychomycosis may lead to find aetiologies more correctly and to design better campaign strategies for prevention and treatment of this disorder. Like other superficial dermatomycosis, onychomycosis is also a common clinical presentation in Iran (e.g., 7-10). The prevalence rates of onychomycosis in some regions of Iran have been reported from 7.0 % to 64.1% (7, 9). Nevertheless, little information exists on the prevalence of the pathogens and their clinical presentation in Kermanshah.

The aim of the present study was to determine the prevalence of onychomycosis among individuals that presented to hospitals in the Kermanshah district of Iran, as well as the mycological features of the disease.

Materials and Methods

Study population

The study included 2402 clinically suspected cases of onychomycosis that presented to the Kermanshah University of Medical Education Mycology Laboratory in Kermanshah Province, west Iran (34°18'N, 47°3'E and 1420 m above sea level) over a period of 16 years (from Jan 1994 to March 2010). The province of Kermanshah is located in west of Iran, with a moderate climate and near two million population of whom mostly are resided in farms and rural areas. Patients on systemic antifungal therapy or topical therapy during the last month were excluded from the study. The Medical University of Kermanshah Ethics Committee reviewed and approved the study protocol.

Mycological methods

The clinical appearance and location of onychomycosis (toenail and/or fingernail) were recorded. After cleaning the affected area with 80% ethanol, the specimens were obtained from clinically abnormal nails by a vigorous scrapping of the nail bed, underside of the nail plate and the hyponychium. Nail scrapings and/or clippings were collected in sterile Petri dishes and a portion of specimens was dissolved in 20.0% potassium hydroxide (KOH) overnight and subjected to direct microscopy for identification according to previously reported procedures (11, 12). For culture, clinical specimens were inoculated into three isolation media: (i) Sabouraud's dextrose agar (SDA, Merck), (ii) SDA with 5% chloramphenicol and cycloheximide for dermatophyte and (iii) SDA with 5% chloramphenicol for mould isolation. The culture tubes were incubated at 25°C and examined daily for 4 weeks. Culture medium was checked for fungal growth of dermatophytes and their identification was confirmed by micromorphological aspects on slide culture and positive urease test, hair penetration and growth on *Trichophyton* agar media (Hi Media). *Candida* species were detected using germ tube test and morphology on cornmeal agar and carbohydrate assimilation using API-20 test (Bio-Merieux; 11, 12**).** The criteria for the diagnosis of onychomycosis caused by non-dermatophytic moulds were based on: (i) microscopical observation of fungal elements in KOH preparations of nail scrapings, (ii) growth of the mould in the culture media and (iii) no growth of a dermatophyte or yeast in all the culture media. When the light microscopy of a nail specimen showed the presence of filaments concurrently with a non-dermatophytic growth in culture media, a second nail specimen was examined by light microscopy and again culture to confirm the presence of non-dermatophytic mould infection (11, 12). The identification of non-dermatophytic fungi species was accredited by direct microscopical examination followed by microscopical and macroscopical evaluations of the primary and slide cultures (1, 13, 14).

Results

Among 2402 (1680 females and 722 males) clinically suspected cases of onychomycosis, 1086 patients (45.2%) were confirmed to be affected with onychomycosis. Onychomycosis was the most prevalent in the 30- to 40-years age group (mean age 45) and the ratio of male (25.6%) to female (74.5%) onychomycosis patients was approximately 1:3. Fingernails were the most frequent anatomic site of onychomycosis in 773 patients (71.4%) while toenail was found in 313 patients (28.8%). However, simultaneous infections of both fingernails and toenails were seen in 77 patients (7.1%). Furthermore, direct microscopy may not necessary yield positive culture. In this regard, 139 (12.8%) nail clippings with positive direct microscopy had no isolated pathogens in culture. The data revealed a higher rate of fingernail than toenail onychomycosis whereas, frequency of toenail infection in both the genders were nearly the same (Table 1).

Table 1: Onychomycosis distribution based on gender and fungi (number (%))

	Sex				Total
	Male		Female		
	Fingernail	Toenail	Fingernail	Toenail	
Yeasts	101 (9.3)	41 (3.7)	635 (58.4)	76 (6.9)	853 (78.54)
Dermatophytes	19 (1.7)	103 (9.4)	14 (1.2)	65 (5.9)	201 (18.50)
Moulds	1 (0.09)	14 (1.2)	3 (0.2)	14 (1.2)	32 (2.94)
Total	121 (11.14)	158 (14.54)	652 (60.3)	155 (14.27)	1086 (100)

It was found that yeast candidal onychomycosis (78.5%) was the most prevalent form followed by that of dermatophytes (18.5%) and non-dermatophyte moulds (2.9%; Tables 2 and 3).

Table 2: Non-dermatophytic moulds isolated as agents of onychomycosis

Moulds	n (%)
Aspergillus flavus	12 (37.5)
Aspergillus niger	8 (25.0)
Aspergillus fumigatus	4 (12.5)
Aspergillus spp.	4 (12.5)
Unknown	4 (12.5)
Total	32 (100)

Table 3: Dermatophytic species isolated as agents of onychomycosis

Dermatophytes	n (%)
Trichophyton rubrum	63 (35.2)
Trichophyton mentagrophytes	32 (15.0)
Epidermophyton floccosum	30 (15.0)
Microsporum canis	3 (1.7)
Microsporum gypseum	2 (1.1)
Trichophyton spp.	40 (20.0)
Total	170 (100)

Among *Candida* species, *C. albicans* was isolated in 45.0% of the cases, mainly from fingernails of women (Fig. 1). Other *Candida* species were isolated in 54.0% of candidal infections.

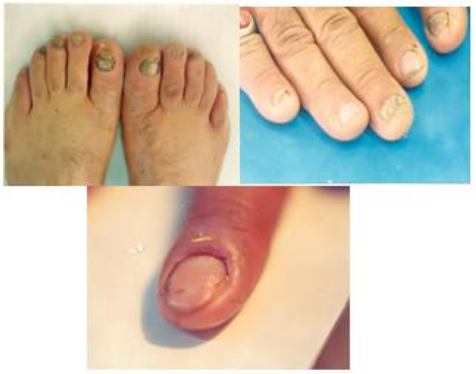

Fig.1: The selected photos of various onychomycoses in Kermanshah province of Iran. From left to right: Toenail, fingernail and candidal infection, respectively

Discussion

As previously concluded by Kaur and colleagues (15), the onychomycosis rate in the present study is probably underestimated because of the difficulty in diagnosis, inappropriate collection of specimens

and fruitless therapies that make it hard to determine the true feature of such onychopathies (15). The present study described, and evaluated the frequency of onychomycosis in individuals representing different strata of population in Kermanshah, Iran, in an attempt to define the epidemiology of this disorder in general population. In this study, the incidence of onychomycosis was confirmed in about 45.2% of the patients analysed, and these data exceeding those published in other regions of country like Qazvin (8), Isfahan (9), Tabriz (10), Khoozestan (16), Kashan (17) and Sari (18), however, lower than the results reported from Tehran (7, 19) and same to the other results of Tehran (20, 21). It is difficult to know to what extent these differences among regions reflect true differences in the aetiological agents of onychomycosis. Although variations among the studies may represent real differences in the geographic groups studied and climate may be one reason, the differences also depend on sampling variations, sex, age, location, clinical type of the infection and other factors (5).

Onychomycosis was more common in the population of 30-40 years of age in this study. This age-related increase of onychomycosis may be resulted from higher probability of nail microtrauma, exposure to pathogenic fungi, and venous insufficiency in older patients as described previously (22). Finally, our data indicate that onychomycosis is uncommon in children in our country thus corroborating the epidemiological reports of other countries (e.g., 23). In the present study, onychomycosis affected more females (74.5%) than males (25.6%). Fingernails were affected more often than toenails in females, which can be explained by sexual hormonal difference and the work habits such as those of housewives, who generally perform farm works or involve in carpet-producing companies. In this sense, Hashemi et al. (24) have proposed that lower concentration of testosterone may increase the susceptibility to some kinds of dermatophytosis. On the other hand, this observation was based on the group of patients examined by the dermatologists and we must consider that some male patients with toenail onychomycosis were not examined, as they did not ask for medical advice or mycological examination. Thus, hands remained wet for most of the day. This fact is mainly due to onycholysis and paronychia of the fingernails caused by *Candida* spp. (67.7%; 25). However, in our study, onychomycosis of fingernails only due to dermatophytosis was prevalent in men (1.7%), whereas the infection of fingernail was lesser prevalent (1.2%) in women. The increased prevalence of onychomycosis of toenails in men compared with women could be the result of more traumas in the nails and the more common use of occlusive footwear. In Iran, household wet works such as laundry and house cleaning are mostly done by housewives, showing a preponderance of female to male patients, at a ratio of 3:1. A similar preponderance of females in onychomycosis cases were reported in other studies from Iran (16, 17).

The previous reports on the prevalence of onychomycosis in different locations of Iran are compared in Table 4. The epidemiology and aetiology of agents of onychomycosis vary in different geographic areas, especially in the frequency with which each group of fungi is responsible for the infection. Dermatophytes were reported as the major causative pathogens in regions such as Ghazvin (8) and Tehran (7), while a high prevalence of yeast infections of the nail has been reported in Isfahan (13), Kashan (17), Khoozestan (16) and Tehran (19-21). In our study, the aetiological fungal agents were 18.50% dermatophytes, 78.54% yeasts and 2.94% moulds. From an aetiologic point of view, dermatophytic moulds were reported as the least prevalent agent of onychomycosis from different regions of Iran (7, 8, 13) and Kermanshah province. On the other hand, in tropical parts o f the world, most infections are attributed to non-dermatophyte mould fungi (5, 26). The two conventional methods for the identification of fungi are direct microscopy under potassium hydroxide and fungal culture. The microscopical method is more sensitive for showing the presence of fungi; however the identification of the fungal pathogen at genus and species levels requires fungal culture. The positivity from direct microscopical examination in our case was 87.2%. This may be considered high when compared with

the work of other researchers (27, 28). Culture was positive in 586 cases including 50 (4.6%) with negative direct examination and 536 (95.4%) with positive direct examination.

Table 4: Frequency distribution of onychomycosis in various parts of Iran

Year	Area	%	The most infected age group	Reference
1996-1997	Tehran	61.4	Not reported	7
2001-2002	Tehran	38.9	40-50 (F); 60-70 (M)	20
2004-2005	Tehran	47.9	31-50 (both sexes)	19
2005	Tehran	42.8	Not reported	21
1996-2004	Tabriz	7.0	11-40 (both sexes)	10
1987-1997	Khoozestan	43.0	30-35 (F); 0-5 and 50 (M)	16
2001- 2003	Kashan	18.9	Not reported	17
2006-2007	Isfahan	39.8	Not reported	9
2004- 2007	Qazvin	40.2	40-49 (both sexes)	8
2009	Sari	30.0	30-49 (both sexes)	18
1994-2010	Kermanshah	45.2	30-40 (both sexes)	The present study

Among dermatophytes, *T. rubrum*, *T. mentagrophytes* and *E. floccosum* are the three common species isolated from the patients in this study. Irrespective of *T. mentagrophytes* varieties, comparing our results with those from other Iranian studies (8, 19, 20). As our results show, yeast caused most of the cases of onychomycosis (57.7%), similar to findings in other regions of Iran (7, 10). Similar to reports from other parts of the world (6, 15), *C. albicans* was the main species responsible for onychomycosis in Kermanshah and other Iranian regions (8-10, 16, 17). The predominant non-dermatophyte moulds in the present study were species of *Aspergillus* in which *A. flavus*, *A. niger* and *A. fumigatus* were the more frequent isolated species. *Aspergillus* species are the predominant causative moulds in two big Iranian provinces, Isfahan (9) and Tehran (7) in agreement with our results. Onychomycosis was found more frequently in women than in men. Prolonged moisture and cosmetic reasons may account for this, because most of the women in the present study were housewives. The different environment of toenails and fingernails may attribute to the differences in fungal involvement in onychomycosis. It is clear from our study that yeast is predominant isolates from fingernails and dermatophytes from toenails.

Conclusion

The most common fungal isolates from onychomycosis in Kermanshah were *Candida* spp., followed by dermatophytes and moulds. Clinicians, therefore, should inform the general population about onychomycosis prevention. The present findings suggest that clinic-, community-, and school-based onychomycosis prevention programs may benefit Kermanshahi people by addressing the risk factors for onychomycosis infection.

Ethical considerations

Ethical issues (Including plagiarism, Informed Consent, misconduct, data fabrication and/or falsification, double publication and/or submission, redundancy, etc) have been completely observed by the authors.

Acknowledgement

Authors acknowledge support of Department of Medical Mycology, School of Medicine, Kermanshah University of Medical Sciences, Kermanshah, Iran. The authors declare that there is no conflict of interest.

References

1. Pontes ZB, Lima Ede O, Oliveira NM, Das Santos JP, Ramos AL, Carvalho MF (2002). Onychomycosis in Joao Pessoa city, Brazil. *Rev Argent Microbiol*, 34: 95–99.
2. Williams HC (1993). The epidemiology of onychomycosis in Britain. *Br J Dermatol*, 129: 101.

3. Garg A, Venkatesh V, Singh M, Pathak KP, Kaushal GP, Agrawal SK (2004). Onychomycosis in India: a clinicoaetiologic correlation. *Int J Dermatol*, 43: 498–502.
4. Abanmi A, Bakheshwain S, El Khizzi N, Zouman AR, Hantirah S, Al Harthi F, Al Jamal M, Rizvi SS, Ahmad M, Tariq M (2008). Characteristics of superficial fungal infections in the Riyadh region of Saudi Arabia. *Int J Dermatol*, 47: 229–35.
5. Velez A, Linares MJ, Fenandez-Roldan JC, Casal M (1997). Study of onychomycosis in Cordoba, Spain: prevailing fungi and pattern of infection. *Mycopathologia*, 137: 1–8.
6. Romano C, Gianni C, Difonzo EM (2005). Retrospective study of onychomycosis in Italy: 1985–2000. *Mycoses*, 48: 42–44.
7. Khosravi AR, Mansouri P (2000). Onychomycosis in Tehran, Iran: prevailing fungi and treatment with itraconazole. *Mycopathologia*, 150: 9.
8. Aghamirian MR, Ghiasian SA (2008). Dermatophytoses in outpatients attending the dermatology center of Avicenna hospital in Quazvin, Iran. *Mycoses*, 51: 155–60.
9. Chadeganipour M, Shadzi S, Dehghan P, Movahed M (1997). Prevalence and aetiology of dermatophytoses in Isfahan, Iran. *Mycoses*, 40:321-4.
10. Kazemi A (2007). *Tinea unguium* in the Iran (1996-2004). *Rev Iberoam Micol*, 24: 113-117.
11. Khosravi AR, Aghamirian MR, Mahmoudi M (1994). Dermatophytoses in Iran. *Mycopathologia*, 37: 43–48.
12. Larone DH (2002). Medically Important Fungi: A Guide to Identification, 4th edn. Washington, DC: American society for microbiology press.
13. Chadeganipour, M., Nilipour, S. and Ahmadi, G (2010). Study of onychomycosis in Isfahan, Iran. *Mycoses*, 53: 153–157.
14. Koursidou T, Devliotou-Panagiotidou D, Kara-Katsanis G, Minas A, Mourellou O, Samara K (2002). Onychomycosis in Northern Greece during 1994–98. *Mycoses*, 45: 29–37.
15. Kaur R, Kashyap B, Bhalla P (2007). A five-year survey of onychomycosis in New Delhi, India: epidemiological and laboratory aspects. *Indian J Dermatol*, 52: 39–42.
16. Yaghoobi R, Hoghooghi-Rad N (2000). A retrospective study on onychomycosis in Khoozestan, Iran. *Med J Iran Hosp*, 3: 43–47.
17. Asadi MA, Dehghani R, Sharif MR (2009). Epidemiologic study of onychomycosis and *tinea pedis* in Kashan, Iran. *Jundishapur J Microbiol*, 2: 61–64
18. Shokohi T, Hajheidari Z, Haghani I, Khalilian A, Aghili SR, Miahi S (2009). The study of 101 cases of onychomycosis and associate factors in patients referred to Boali Sina Hospital and Toba dermatology outpatient clinics in Sari. *J Mazandaran Uni Med Sci*, 19: 33-43.
19. Zaini F, Mahmoudi M, Mehbod ASA, Kordbacheh P, Safara M (2009). Fungal Nail Infections in Tehran, Iran. *Iranian J Publ Health*, 38: 46–53.
20. Gerami shoar M, Zomorodian K, Emami M, Tarazoei B, Saadat F (2002). Study and identification of the etiological agents of onychomycosis in Tehran, capital of Iran. *Iranian J Publ Health*, 31:100–104.
21. Hashemi SJ, Gerami M, Zibafar E, Daei M, Moazeni M, Nasrollahi A (2010). Onychomycosis in Tehran: mycological study of 504 patients. *Mycoses*, 53: 251-255.
22. Heikkala H, Stubbs S (1995). The prevalence of onychomycosis in Finland. *Br J Dermatol*, 133: 699–703.
23. Gupta AK, Sibbald RG, Lynde CW et al (1997). Onychomycosis in children: prevalence and treatment strategies. *J Am Acad Dermatol*, 36: 395–402.
24. Hashemi SJ, Sarasgani MR, Zomorrodian K(2004). A comparative survey of serum androgenic hormones levels between male patients with dermatophytosis and normal subjects. *Jpn J Infect Dis*, 56: 60–62.
25. Romano C, Giani C, Difonzo E (2005). Retrospective study of onychomycosis in Italy: 1985-2000. *Mycoses*, 48: 42-44.
26. Ng KP, Saw TL, Madasamy M, Soo Hoo TS (1999). Onychomycosis in Malaysia. *Mycopathologia*, 147: 29–32.
27. Kam KM, Au WF, Wong PY, Cheung MM (1997). Onychomycosis in Hong Kong. *Int J Dermatol*, 36: 757–61.
28. El Sayed F, Ammoury A, Haybe R, Dhaybi R (2006). Onychomycosis in Lebanon: a mycological survey of 772 patients. *Mycoses*, 49: 216.