

CLINICAL REPORT

Prevalence of Toe Nail Onychomycosis in Diabetic Patients

Ditte Marie L. SAUNTE^{1,2,3}, Jane B. HOLGERSEN², Merete HÆDERSDAL¹, Gitte STRAUSS¹, Mikael BITSCH², Ole L. SVENDSEN⁴, Maiken C. ARENDRUP³ and Else L. SVEJGAARD¹

¹Department of Dermatology and Venereology, ²Copenhagen Wound Healing Centre, ³Department of Internal Medicine, Bispebjerg Hospital, University of Copenhagen, and ⁴Unit of Mycology and Parasitology, Statens Serum Institut, Copenhagen, Denmark

Onychomycosis among diabetic patients has been reported in some studies to be of high prevalence. This study aimed to investigate the prevalence of onychomycosis among diabetic patients at a Danish University Hospital. Clinical and mycological examinations were performed on type 1 and 2 diabetic patients from in- and out-patient clinics. A total of 271 patients were enrolled, 72% males, mean age 61.3 years, 26% of the patients had diabetes type 1. The prevalence of toe nail onychomycosis (positive culture and/or microscopy) was 22% ($n = 59$) of which 55 cases were caused by dermatophytes (93%) and 4 cases by yeasts (7%). A correlation was found between onychomycosis and age ($p = 0.02$) and severity of nail changes ($p < 0.001$), respectively. However, no significant correlation was found to gender, type of diabetes, lower extremity arterial disease, neuropathy, toe amputation or oedema. Onychomycosis occurred with a high prevalence in diabetic patients, especially among older patients and those with severe nail changes. *Key words: diabetes mellitus; dermatophytosis; candidosis; epidemiology; tinea unguium; onychomycosis.*

(Accepted May 8, 2006.)

Acta Derm Venereol 2006; 86: 425–428.

Ditte Marie Saunte MD, Unit of Mycology and Parasitology Building 45/112, Department for Bacteriology, Mycology and Parasitology, State Serum Institute, Artillerivej 5, DK-2300 Copenhagen S, Denmark. E-mail: dms@ssi.dk

Onychomycosis in toe nails is one of the most common nail diseases (1). Prevalences in European countries vary from 2.8% to 11.1% (2, 3). Onychomycosis is caused mainly by dermatophytes, most often by *Trichophyton rubrum* and *T. mentagrophytes* var. *interdigitale*, which are both also associated with tinea pedis (1). Non-dermatophyte moulds and yeasts are the pathogens in 7% of fungal nail infections (4). Male gender and old age are predisposing factors for fungal nail infection, as well as diabetes, psoriasis, peripheral arterial disease and immunosuppression (HIV, chemotherapeutic treatment) (1–12). Moreover, environmental factors, such as being active in sport or attending public bathing facilities, enhance the risk of contracting onychomycosis and

some patients are thought to be genetically predisposed to *T. rubrum* infections due to an autosomal dominant inheritance (3, 5, 13).

Diabetes mellitus (DM) is a worldwide problem of increasing importance. It is estimated that about 200 million people will suffer from DM in 2010 and approximately 70 million of these may develop onychomycosis (8, 10). It is well known that diabetic patients often have problems with their feet, due mainly to neuropathy and arterial insufficiency. The risk of toe or lower leg amputation may be increased if fissures or traumatic ulcerations are followed by a secondary bacterial infection (14). A mycotic nail may be the first step in such a process, as an abnormal nail plate may cause lesions in the surroundings due to hyperkeratosis and sharp nail edges. It is therefore relevant to study the frequency of nail mycosis in diabetic patients.

MATERIALS AND METHODS

Patients

During the period from April 2003 to March 2004, patients with DM type 1 or 2 and older than 18 years were recruited from in- and outpatient clinics at Copenhagen Wound Healing Centre, Department of Dermatology, and Department of Internal Medicine, Bispebjerg Hospital, University of Copenhagen. Patients were referred with one of the following conditions: arterial, venous or diabetic ulcers, for a foot-ulcer preventive consultation, dermatological disease or for diabetic controls.

The Regional Scientific-Ethical Committee approved the study and all participants signed a written informed consent. Pregnant women and patients formerly recruited were excluded from the study.

Clinical examinations

Experienced podiatrists or physicians performed the clinical observations and the mycological sampling was performed by especially trained healthcare workers.

The following parameters were registered: age, gender, thickened nail plate, nail ingrowth, onycholysis, leg oedema and the number of toe amputations. The most severely affected toe nail was scored by a visual analogue scale (VAS) from 0 to 15: normal nail 0; mild nail changes 1–5; moderate nail changes 6–10; and severe nail changes 11–15. Patients with no pulsation of *arteria dorsalis* pedis and *a. tibialis posterior* or with an ankle-brachial index less than 0.80 were recorded as having lower extremity arterial disease (LEAD). Similarly, the presence/absence of lower extremity neuropathy (sensibility) measured by a 10 g Semmes-Weinstein monofilament test was recorded.

Mycological examinations

Nail material was taken from clinically abnormal nails or from the first right toe nail if all nails appeared normal. Direct microscopy was performed using blanchophor and fluorescence microscopy. Cultures were performed using Sabouraud-glucose-agar+chloramphenicol +/- cycloheximid (Bie & Berntsen, Rødovre, Denmark) and incubation at 25°C for up to 6 weeks.

Presence of dermatophytes in patient material was demonstrated by direct microscopy (regularly septated hyphae equally sized with or without arthroconidia) and/or culture and species identifications performed based on micro- and macro-morphology of colonies. Yeasts were detected by direct microscopy and cultures. Numerous budding blastoconidia with or without pseudohyphae defined a positive microscopy, and cultures with more than 10 colonies were considered positive. Identification to species level was done by using CHROM-agar Candida plates or by the API 32-C System Biomerieux yeast identification programme (Biomerieux, Lyon, France).

Malassezia was diagnosed if spherical to oval unipolar budding cells were observed by direct microscopy. Cultures were not performed. Non-dermatophyte moulds were registered, when positive in both microscopy and culture, however, *Malassezia* and non-dermatophyte moulds were not regarded as true pathogens in this study, as repeated sampling for confirmation of infection was not performed in these cases. They were thus excluded for risk factor analyses.

Statistical analyses

A logistic regression model was used to evaluate correlations between onychomycosis and age, gender, LEAD, neuropathy, toe amputation, oedema and type of diabetes (PROC LOGISTIC, SAS version 9.1.3). Odds ratios (OR) with 95% two-sided confidence intervals (CI) are presented with corresponding *p*-values (Wald-tests).

RESULTS

Patients

A total of 271 patients were enrolled, 194 men (72%), 77 women (28%), 70 had diabetes type 1 (26%) and 201 had diabetes type 2 (74%). Ages ranged from 29 to 91 years, mean age 61.3 years (95% CI: 59.8–62.7 years).

Prevalence of onychomycosis

Fungal infection of the toe nails was found in 59 individuals (46/194 men (23%) and 13/76 women (17%)) with an overall prevalence of 22%. Four patients (3 men and 1 woman) with *malassezia* and non-dermatophyte mould were excluded. Dermatophytes were involved in 55 cases (93%) and yeasts in 4 cases (7%) (Table I).

Table I. Mycological culture and microscopy results from 271 diabetic patient samples

Fungal organism	Number of specimens (microscopy positive)
<i>T. rubrum</i>	11 (8)
<i>T. mentagrophytes</i>	5 (4)
<i>T. tonsurans</i>	2 (2)
<i>C. albicans</i>	2 (1)
<i>C. krusei</i>	2 (2)
<i>C. glabrata</i>	1 (0)
<i>C. parapsilosis</i>	1 (1)
Mould ^a	2 (2)
<i>Malassezia</i> sp ^a	2 (2)
Microscopy positive (dermatophyte), culture negative	37 (37)
Microscopy positive (yeast), culture negative	13 (13)
Negative culture and microscopy	193 (–)
Total	271 (72)

^a Not confirmed by resampling. Excluded for further risk factor analysis.

Possible risk factors and onychomycosis

Old age was found to be associated with a higher prevalence of onychomycosis ($p < 0.02$, OR = 1.04, 95% CI 1.01–1.07) whereas there were no significant correlations to gender, type of diabetes, LEAD, neuropathy, toe amputations and oedema. One fourth of the patients had LEAD, 72% had neuropathy, 19% had one or more toes amputated and 25% had oedema. The prevalence of onychomycosis according to the possible risk factors is illustrated in Table II.

Correlations between mycology and clinical findings

The severity of the clinical nail changes was correlated with the presence of onychomycosis ($p = 0.0007$, OD 1.15, 95% CI 1.06–1.25). This is illustrated in Fig. 1 and the type of nail disorder in relation to mycological findings is shown in Table III.

DISCUSSION

The prevalence of onychomycosis in diabetic patients was 22%. Similar findings have been reported previously (8, 9). In Western Europe onychomycosis occurs in 2.8–11% of the general population and in a Danish population (mean age 55 years old) of 5755 patients visiting general practice 4.9% of the patients

Table II. Prevalence of onychomycosis according to possible risk factors: lower extremity arterial disease (LEAD), neuropathy, toe amputation, oedema and diabetes type. Figures in parentheses represent isolates that were microscopy positive

	LEAD ^a		Neuropathy ^b		Toe amputation		Oedema ^b		Diabetes type	
	+	-	+	-	+	-	+	-	1	2
No. of patients	70	193	197	67	53	214	68	196	69	198
Onychomycosis (%)	20 (29)	37 (19)	44 (22)	14 (21)	15 (28)	44 (21)	19 (28)	39 (20)	15 (22)	44 (22)

^a263 of 267 patients examined for LEAD.

^b264 of 267 patients examined for neuropathy and oedema.

Table III. Mycological findings in nails with and without clinical signs

Observations (n = 274) ^a	Patients (n = 267)	Dermatophytes (n = 55)	Yeasts ^b (n = 4)	Negative ^c (n = 208)
	n (%; 95% CI)	n (%; 95% CI)	n (%; 95% CI)	n (%; 95% CI)
Normal appearing toenails (n = 109)	14 (13; 7–21)	2 (2; 0–6)	93 (85; 77–91)	
Thickened nail plate (n = 133)	40 (30; 22–39)	2 (2; 0–5)	91 (68; 60–76)	
Nail ingrowth (n = 20)	1 (5; 0–25)	0 (0; 0–17)	19 (95; 75–100)	
Onycholysis (n = 12)	2 (17; 2–48)	0 (0; 0–26)	10 (83; 52–98)	

^aFourteen of the patients had combinations of two different clinical signs. Seven patients had no records.

^bDetected by positive microscopy and culture.

^cNo fungal elements were detected (n = 193), or yeast was detected either by microscopy (n = 13) or culture (n = 2), but not considered as pathogen.

had onychomycosis, including a prevalence of 7% in a subgroup of patients with DM (2, 3, 15). The much higher prevalence in diabetic patients in the present study might be explained by difference in patient groups, i.e. patients hospitalized vs. patients in general practice, the former with more severe disease than the latter. Also differences in age may influence the prevalence as old age was found to be associated with a higher prevalence of onychomycosis in our study as well as in other epidemiological studies (15, 16). It is noteworthy that the presence of onychomycosis in this study was independent of gender and diabetes type. In addition, clinical conditions such as LEAD, neuropathy, oedema and toe amputations were not significantly related to fungal infection. However a correlation between toenail onychomycosis and LEAD has been observed in other studies (9, 12), and we can not rule out that a statistically significant association between LEAD and onychomycosis could have been found if more patients had been enrolled in the present study.

The pathogenic role of *Candida* infections in nails is controversial. Some authors state that the demonstration of yeasts generally represents contamination/colonisation of a dermatophyte infected nail (17). Others consider yeasts as aetiological agents if demonstrated by microscopy as well as by culture of the nail (18, 19). The later is confirmed by the fact that detection of yeast/mycelium in the nail plate by direct microscopy or histopathological examination in combination with the isolation of *C. albicans* in a study by Hay et al. was associated with good response to antifungal treatment (20). In agreement with this, only specimens which were fulfilling the combined criterion were considered as *Candida* onychomycosis in the present study.

Two-thirds of the specimens were dermatophyte positive in microscopy (n = 37) but negative in culture. Failure to culture from microscopy positive nails is a well-known phenomenon and has been reported in 12–50% of the nail specimens (18, 21, 22). A study by

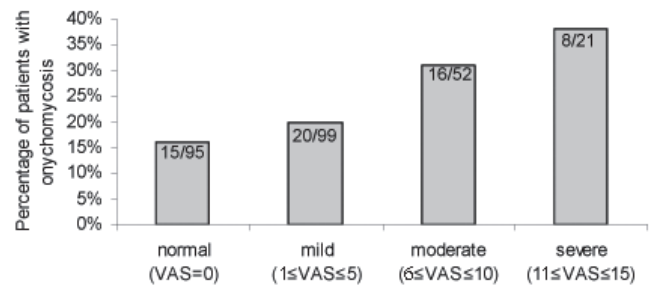


Fig. 1. Severity of nail changes according to the Visual Analogue Scale (VAS) score from zero to 15: normal nail 0; mild nail changes 1–5; moderate nail changes 6–10; and severe nail changes 11–15. The figures in the columns show the number of infected patients in each clinical subgroup.

Gentles (21) demonstrates that it is possible to reduce this inconsistency by resampling from microscopy-positive nails but, even though this is done more than four times, 18% of the patient samples remain culture negative. Another possible explanation for this might be that the specimens were stored for up to 3 months before examinations were performed. This delay may have weakened the viability of the fungi. Inadequate sampling technique also reduces positivity rates; although the risk of this was minimized as only especially trained healthcare workers performed the sampling. The morphology observed in direct microscopy provides a presumptive identification of the fungus involved (i.e. dermatophyte, yeast or mould), but final genus identification is necessary to guide the choice of optimal antifungal treatment why treatment should never be initiated on a positive microscopy alone. Re-sampling from the culture negative toe nails would have been optimal to confirm the diagnosis. This was not possible from a practical point of view, as we did not have ethical approval of contacting the patients more than once. Since dermatophytes are regarded as true pathogens (18, 23) we decided to consider all cases in which dermatophytes were demonstrated by direct microscopy as tinea unguium, realizing that the reported prevalence will be higher than if only culture positive cases were included. In support of our choice, direct microscopy was recently found to be 74% sensitive for dermatophytes in a study by Summerbell et al. (24).

Even though we demonstrate a statistical correlation between the clinical appearance and the mycological result, clinical evaluation is not sufficient to diagnose fungal infection. Thus, 68% of the nail material from thickened toe nails, 95% from ingrown toe nails and 83% from nails with onycholysis were negative in both microscopy and culture. This clearly demonstrates that a proper mycological diagnosis is required before a systemic antimycotic treatment for onychomycosis is initiated.

Notably, 15% of clinically normal toe nails were mycology positive, whereas this was only the case in 1.5% of the patients with normal toenails in the study of Gupta et

al. (9). This colonization may in itself be considered a risk factor for future infection in predisposed individuals.

This study confirms that diabetic patients are at high risk of having or contracting onychomycosis. Further studies are needed to investigate whether treatment of this patient group may prevent severe complications such as secondary bacterial infections.

ACKNOWLEDGEMENTS

The authors thank all the patients who volunteered, Hanne Anderson for her excellent technical assistance, Susanne Krid for her enthusiasm when recruiting patients, and Anders Mørup Jensen for his help with the statistical analysis.

REFERENCES

- Gill D, Marks R. A review of the epidemiology of tinea unguium in the community. *Australas J Dermatol* 1999; 40: 6–13.
- Perea S, Ramos MJ, Garau M, Gonzalez A, Noriega AR, del Palacio A. Prevalence and risk factors of tinea unguium and tinea pedis in the general population in Spain. *J Clin Microbiol* 2000; 38: 3226–3230.
- Sigurgeirsson B, Steingrimsson O. Risk factors associated with onychomycosis. *J Eur Acad Dermatol Venereol* 2004; 18: 48–51.
- Gupta AK, Lynde CW, Jain HC, Sibbald RG, Elewski BE, Daniel CR, et al. A higher prevalence of onychomycosis in psoriatics compared with non-psoriatics: a multicentre study. *Br J Dermatol* 1997; 136: 786–789.
- Gupta AK, Konnikov N, Lynde CW, Summerbell RC, Albreski D, Baran R, et al. Onychomycosis: predisposed populations and some predictors of suboptimal response to oral antifungal agents. *Eur J Dermatol* 1999; 9: 633–638.
- Gupta AK, Taborda P, Taborda V, Gilmour J, Rachlis A, Salit I, et al. Epidemiology and prevalence of onychomycosis in HIV-positive individuals. *Int J Dermatol* 2000; 39: 746–753.
- Levy LA. Epidemiology of onychomycosis in special-risk populations. *J Am Podiatr Med Assoc* 1997; 87: 546–550.
- Dogra S, Kumar B, Bhansali A, Chakrabarty A. Epidemiology of onychomycosis in patients with diabetes mellitus in India. *Int J Dermatol* 2002; 41: 647–651.
- Gupta AK, Konnikov N, Macdonald P, Rich P, Rodger NW, Edmonds MW, et al. Prevalence and epidemiology of toenail onychomycosis in diabetic subjects: a multicentre survey. *Br J Dermatol* 1998; 139: 665–671.
- Scher RK, Baran R. Onychomycosis in clinical practice: factors contributing to recurrence. *Br J Dermatol* 2003; 149 (suppl 65): 5–9.
- Larsen GK, Haedersdal M, Svejgaard EL. The prevalence of onychomycosis in patients with psoriasis and other skin diseases. *Acta Derm Venereol* 2003; 83: 206–209.
- Gupta AK, Gupta MA, Summerbell RC, Cooper EA, Konnikov N, Albreski D, et al. The epidemiology of onychomycosis: possible role of smoking and peripheral arterial disease. *J Eur Acad Dermatol Venereol* 2000; 14: 466–469.
- Faergemann J, Baran R. Epidemiology, clinical presentation and diagnosis of onychomycosis. *Br J Dermatol* 2003; 149 (suppl 65): 1–4.
- Rich P. Onychomycosis and tinea pedis in patients with diabetes. *J Am Acad Dermatol* 2000; 43: 130–134.
- Svejgaard EL, Nilsson J. Onychomycosis in Denmark: prevalence of fungal nail infection in general practice. *Mycoses* 2003; 47: 131–135.
- Heikkilä H, Stubb S. The prevalence of onychomycosis in Finland. *Br J Dermatol* 1995; 133: 699–703.
- Ellis DH, Watson AB, Marley JE, Williams TG. Non-dermatophytes in onychomycosis of the toenails. *Br J Dermatol* 1997; 136: 490–493.
- English MP. Nails and fungi. *Br J Dermatol* 1976; 94: 697–701.
- Lateur N, Mortaki A, Andre J. Two hundred ninety-six cases of onychomycosis in children and teenagers: a 10-year laboratory survey. *Pediatr Dermatol* 2003; 20: 385–388.
- Hay RJ, Baran R, Moore MK, Wilkinson JD. Candida onychomycosis – an evaluation of the role of Candida species in nail disease. *Br J Dermatol* 1988; 118: 47–58.
- Gentles JC. Laboratory investigations of dermatophyte infections of nails. *Sabouraudia* 1971; 9: 149–152.
- Petrini B, von Rosen ML. Optimal dermatophyte diagnosis requires both microscopy and culture. *Läkartidningen* 2002; 99: 4084.
- Elewski BE. Onychomycosis: pathogenesis, diagnosis, and management. *Clin Microbiol Rev* 1998; 11: 415–429.
- Summerbell RC, Cooper E, Bunn U, Jamieson F, Gupta AK. Onychomycosis: a critical study of techniques and criteria for confirming the etiologic significance of non-dermatophytes. *Med Mycol* 2005; 43: 39–59.