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Swimming pool deck as environmental reservoir of *Fusarium*

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While investigations on fungal contamination of swimming pools usually focus on dermatophytes, data on other potentially pathogenic molds are scarce. Here, we report the investigation of fungal colonization of the deck surrounding a hospital physical therapy swimming pool. Five series of samples from 8 sites were collected over one year from the pool surroundings. Concomitantly, 58 patients using the swimming pool were examined and samples obtained from those with suspected onychomycosis. All surface samples were positive for fungi, with *Fusarium* the most frequently recovered from 22 of 27 samples of sites surrounding the pool. Among the outpatients evaluated, two presented with a mixed onychomycosis from which *Fusarium* and *Trichophyton rubrum* were isolated. The questions of possible acquisition from the swimming pool area must be considered in both cases as the ungual lesions had developed within the previous three months. This warrants further studies to better understand the epidemiology of potentially pathogenic molds in areas surrounding pools in order to adopt appropriate measures to avoid contamination. This is of particular importance within medical institutions, considering the potential role of *Fusarium* onychomycosis as a starting point for disseminated infections in immunocompromised patients.

Keywords *Fusarium*, swimming pool, environment, onychomycosis

Introduction

Fusarium species are hyaline filamentous fungi distributed widely in the environment. They are primarily plant pathogens which can have major economic impact on food [1]. In human medicine, *Fusarium* spp. are etiologic agents of superficial infections such as keratitis and onychomycosis [2–4]. More recently, they have emerged as life-threatening pathogens in immunocompromised patients, particularly those with hematological malignancy and/or having undergone bone marrow transplantation [5,6]. While trauma resulting from a thorn or from walking barefoot are usually considered the mode of acquisition of superficial infections, recent studies have suggested that Fusarium contaminated water may act as the source of infections for hospitalized immunocompromised patients [7]. Herein, we report the results of an epidemiological survey of the fungal colonization of a hospital physical therapy swimming pool that demonstrates the importance of *Fusarium* contamination in surrounding areas. The occurrence of two cases of *Fusarium*-associated onychomycosis in patients attending the pool raises the question of local acquisition.

Materials and methods

The physical therapy swimming pool, 15 × 7 meters long with a depth of 1.5 meters, was located in the basement of our institution and consequently, there was no surrounding vegetation. There were five entry steps into the water, the temperature of which is controlled at 34°C, with the air temperature maintained at 30°C. Patients who utilize the pool mainly suffer from rheumatologic diseases such as rheumatoid arthritis and some of them were being treated with corticosteroids. During the period of the study, a total

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of about 200 patients attended the pool, on average twice a week, but about 10 utilized it every day. The pool deck was cleaned daily using Solsynthene® (IFA-Credo, Lyon, France), the formulation of which is propanol-25g, laurylpropylene diaminoacetic acid 10g, N-alkyldimethylbenzylamonium chloride 10g, NaOH 0.05g, EDTA 0.02g qsp water 100 ml The product claims to be fungicidal using the French standardized norm AFNOR NF T72-201. As recommended by the manufacturer, the solution was first diluted 0.25% in cold water then wiped on the floor without further rinsing. Cleaning was performed at the end of every working day when all attending patients had left the pool.

Five surveys were performed over one year at the following eight sites; three showers used by patients before entering the swimming pool, three synthetic carpets (one in the locker room) laid on the floor to prevent the patients from falling, and two tiled locations (one close to the basin, the other in the locker room). Sampling was performed on dry surfaces after all patients had left the pool, prior to the cleaning of the floor with a single sample from each site. On two instances, control samples were collected after floor cleaning. For each site, Rodac contact plates (6 cm diameter; Oxoid, Dardilly, France) containing Sabouraud glucose agar plus chloramphenicol (200 mg/l) and gentamycin (40 mg/l) were used for surface sampling. The plates were incubated at 27°C for one month.

During the same period, 58 patients using the swimming pool were examined by a dermatologist for cutaneous lesions. In those cases in which a fungal infection was suspected, samples were collected from nails, toe webs, palm and/or plant. Portions of the samples were prepared in 10% sodium hydroxide for direct microscopic examination to determine the presence of fungal elements [8]. In addition, scales were inoculated on Sabouraud glucose agar with antibiotics with and without cycloheximide (0.5 g/l) and incubated for one month at 27°C.

Identification of filamentous fungi was based on macroscopic and microscopic characteristics [9]. Yeast isolates were numbered and one colony of each was identified by its phenotypic appearance using Auxacolor system (Biorad, Marnes-la Coquette, France).

Results

Surface samples

All samples ($N=27$) were positive for fungi (Table 1). Yeasts were detected in 14 samples with *Candida famata* proving to be the most common in shower areas. *Trichophyton interdigitale* was the only dermatophyte isolated but was recovered only once from a single site.

Among the molds, *Fusarium* spp. were isolated from 22 (81.5%) samples and were found in all sites studied. These same fungi were quantitatively the most abundant filamentous fungi, with more than 50 colonies per site in five instances. Macroscopic and microscopic examination of the colonies revealed that they were consistent with the features of *Fusarium oxysporum* in the majority of the cases except two, where *Fusarium solani* was identified. Results from samples performed after routine cleaning did not show any significant change in the fungal flora (data not shown).

Patients examination

Among the 58 outpatients evaluated, 13 were suspected of having onychomycosis. Samples were not collected from four of these patients as they were receiving antifungal therapy. No fungi were recovered from another three of the patients. Of the remaining 6 patients, *Trichophyton rubrum* was isolated in one case, *T. interdigitale* was recovered in samples from three patients, and both *T. rubrum* and *F. oxysporum* were isolated in the remaining two cases of suspected onychomycosis. Similar results were obtained in the latter two cases when samples were collected 1 week and 10 days after the initial collection. In both cases, patients stated that they were free of ungual lesion when they started to frequent the swimming pool and that the lesions (subungual and superficial onychomycosis of the left first toenail and subungual onychomycosis of the 4th and 5th left toenails, respectively) appeared within the first trimester of attending the pool. One of these patients was ongoing corticosteroid therapy at the time of diagnosis.

Discussion

Our work clearly demonstrates the frequency, predominance and abundance of *Fusarium* in the environs of a hospital physical therapy swimming pool. *Fusarium* spp., along with members of *Cladosporium* and *Acremonium*, were the most frequently isolated molds from these surfaces. While a number of reports describe the contamination of swimming pools with dermatophytes [10–12], surveys to evaluate the presence of other molds are scarce [13,14] and the recovery of *Fusarium* unusual. The use of culture media containing cycloheximide, an antifungal agent known to inhibit the growth of most molds, excluding dermatophytes, may explain the results of these earlier studies. However, our data demonstrate that the recovery of other molds is greater than that of dermatophytes from the swimming pool environment. This is supported by the results of a previous investigation which reported that *Fusarium* spp. were isolated in 18.6% of surfaces of a public swimming pool [13]. Recently, *Fusarium* was shown to be the most frequently isolated genus from the surfaces of a panel of ten swimming facilities, representing 10.5% of

Table 1 Results (genera, species and CFU/sample) of microbiologic analysis for detection of fungi in environmental samples from a hospital physical therapy swimming pool in Paris, France.

	May		October		March		April		May	
Site of isolation	Species	Colonies	Species	Colonies	Species	Colonies	Species	Colonies	Species	Colonies
Shower 1	*F. oxysporum*	>50	*F. oxysporum*	10	Yeasts†	>100	not done		*Cladosporium* sp	10
	Penicillium sp	30	*Cladosporium* sp	2	*F. oxysporum*	13			*Acremonium* sp	1
					F. solani	1				
Shower 2	*F. oxysporum*	6	*F. oxysporum*	27	Yeasts†	>100	Yeasts†	30	not done	
	Acremonium sp	2			*F. oxysporum*	9	*F. oxysporum*	10		
	Chaetonium sp	2			*Cladosporium* sp	2	*Cladosporium* sp	20		
	Rhodotorula rubra	1			*Acremonium* sp	3	Yeasts††	50		
					Penicillium sp	2	*Trichosporon* sp	50		
Shower 3	*Cladosporium* sp	1	*F. oxysporum*	>50	Yeasts†	>100	Yeasts†	50	*F. oxysporum*	1
			Acremonium sp	3	*Acremonium* sp	>50	*Acremonium* sp	2	Yeasts†	4
					Cladosporium sp	2	*Cladosporium* sp	3	*Cladosporium* sp	10
							Rhodotorula rubra	5	*Penicillium* sp	11
							Trichosporon sp	15	*Aspergillus niger*	1
Carpet 1	not done		not done		Yeasts†	>100	*F. oxysporum*	15	*F. oxysporum*	2
					F. oxysporum	30	*F. solani*	1	*Cladosporium* sp	10
					Cladosporium sp	1	*Cladosporium* sp	>50	*Penicillium* sp	16
					Penicillium sp	2	Yeast††	5		
Carpet 2	*F. oxysporum*	3	*F. oxysporum*	15	*F. oxysporum*	45	not done		not done	
	Acremonium sp	1	*Penicillium* sp	3	Yeasts†	20				
			S. brevicaulis	2	*Cladosporium* sp	5				
					Geotrichum sp	50				
					Trichophyton interdigitale	3				
Tiled floor next to the basin	not done		*F. oxysporum*	5	*F. oxysporum*	>50	*F. oxysporum*	>50	*F. oxysporum*	>50
			Penicillium sp	1	*Cladosporium* sp	4	*Acremonium* sp	>50	*Trichosporon* sp	1
Locker room tiled floor	not done		not done		*Acremonium* sp	5	*F. oxysporum*	>50	not done	
					Cladosporium sp	1	*Cladosporium* sp	6		
					Geotrichum sp	1	Yeasts†, ††	1,8		
							Aspergillus niger	2		
Locker room carpet	not done		not done		*F. oxysporum*	5	not done		*F. oxysporum*	1
					Yeasts†	25			*Cladosporium* sp	5
					Penicillium sp	20			*Penicillium* sp	20

†Identification of 1–3 isolates gave an identification of *Candida famata*. ††Identification of 1–3 isolates gave an identification of *Rhodotorula rubra*. Considering the 6 cm-diameter of the plate a cut-off value of 50 and 100 for the count of colonies was used for filamentous fungi and yeasts, respectively.

the isolates [14]. In addition, *Fusarium* spp. were also repeatedly isolated from water collected from pools and rivers [15]. These results support data suggesting that water may be an environmental reservoir of *Fusarium* spp., including the water supplies of a hospital water system [7]. It was found in an *in vitro* study that 30% of Fusarium spores would germinate in water after 6 h incubation, and up to 60 and 100% after 12 and 24 h of inoculation, respectively. These percentages were significantly higher than those observed with 12 other mold species, including *Aspergillus fumigatus* [16].

These results also raise the question of appropriate measures to control *Fusarium* contamination around swimming pools. To our knowledge, there is currently very little data on the activity of disinfectants against *Fusarium* species using standardized protocols. Using the French standard T72-201, Bobichon *et al*. reported that Decalcite was ineffective (Bayrol manufacturer; containing chlorohydric acid, polyvinyl alcohol, nonionic detergent and isopropylic acid) and that Adilon was moderately effective (Bayrol manufacturer; containing phosphoric acid, isopropanol and alkylphenolethoxylate). The latter in our case, seemed to have no efficacy. Indeed, in two instances, sampling performed at the end of the day, after cleaning revealed no effect on the fungal flora. This is another argument for considering the formulation used as inactive against *Fusarium* and some yeast species such as *Candida famata*. A switch to another disinfectant product could not be implemented due to the termination of the physical therapy activity which included the definitive closure of the swimming pool. Overall, this suggests that when investigated, disinfectants must be tested with standardized protocols against additional fungal species other than *Candida albicans* and *Aspergillus niger*.

Finally, the isolation of *Fusarium* from the nails of two patients suffering onychomycosis, among 58 frequenting the pool during the study, raises the question of the acquisition from the pool deck environs. Only molecular typing of environmental and clinical isolates would provide definitive answers to this question, but having no facility for such investigations, the samples were discarded. These patients had no other risk factor for *Fusarium* onychomycosis such as walking bare-foot or wearing occlusive footwear, and did not attend other pools or sports centers. Even considering the concomitant isolation of a dermatophyte, the role of *Fusarium* in the nails cannot be overlooked and these cases can be considered as mixed onychomycosis. While onychomycosis is often considered a benign infection, *Fusarium* onychomycosis is generally difficult to treat. To date, apart from some successful treatments from case reports, no therapeutic strategy has provided reliable results [17–19]. Also, it has been emphasized that *Fusarium* onychomycosis can be the portal of entry of further invasive infection in immunocompromised patients, most often with hematological malignancies [20,21].

In conclusion, our data warrant further studies of different types of swimming pools to better appreciate the epidemiology of potentially pathogenic molds, such as *Fusarium*. Appropriate measures to control fungal contamination of the pool environs are still to be defined. Effective and safe compounds should be evaluated and applied for appropriate cleaning to prevent the risk of infection.

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References

1 Nelson PE, Dignami MC, Anaissie EJ. Taxonomy, biology and clinical aspects of *Fusarium* species. *Clin Microbiol Rev* 1994; **7**: 479–504.
2 Gupta AK, Baran R, Summerbell RC. *Fusarium* infections of the skin. *Curr Opin Infect Dis* 2000; **13**: 121–128.
3 Guilhermetti E, Takahachi G, Shinobu CS, Svidzinski TI. *Fusarium* spp. as agents of onychomycosis in immunocompetent hosts. *Int J Dermatol* 2007; **46**: 822–826.
4 Alfonso EC, Cantu-Dibildox J, Munir WM, *et al*. Insurgence of *Fusarium* keratitis associated with contact lens wear. *Arch Ophthalmol* 2006; **124**: 941–947.
5 Dignani MC, Anaissie E. Human fusariosis. *Clin Microbiol Infect* 2004; **10** (Suppl. 1): 67–75.
6 Hennequin C, Lavarde V, Poirot JL, *et al*. Invasive *Fusarium* infections: a retrospective survey of 31 cases. The French 'Groupe d'Etudes des Mycoses Opportunistes' GEMO. *J Med Vet Mycol* 1997; **35**: 107–114.
7 Anaissie EJ, Kuchar RT, Rex JH, *et al*. Fusariosis associated with pathogenic *Fusarium* species colonization of a hospital water system: a new paradigm for the epidemiology of opportunistic mold infections. *Clin Infect Dis* 2001; **33**: 1871–1878.
8 Monod M, Baudraz-Rosselet F, Ramelet AA, Frenk E. Direct mycological examination in dermatology: a comparison of different methods. *Dermatologica* 1989; **179**: 183–186.
9 de Hoog GS, Guarro J, Gene J, Figueras MJ. *Atlas of Clinical Fungi*. ASM Press; 2000.
10 Detandt M, Nolard N. Fungal contamination of the floors of swimming pools, particularly subtropical swimming paradises. *Mycoses* 1995; **38**: 509–513.
11 Leoni E, Legnani P, Guberti E, Masotti A. Risk of infection associated with microbiological quality of public swimming pools in Bologna, Italy. *Public Health* 1999; **113**: 227–232.

12 Hilmarsdottir I, Haraldsson H, Sigurdardottir A, Sigurgeirsson B. Dermatophytes in a swimming pool facility: difference in dermatophyte load in men's and women's dressing rooms. *Acta Derm Venereol* 2005; **85**: 267–268.
13 Bobichon H, Amelot O, de Bievre C, *et al*. Contamination fongique des sols d'une piscine. *Bull Soc Fr Mycol Med* 1989; **18**: 289–292.
14 Brandi G, Sisti M, Paparini A, *et al*. Swimming pools and fungi: an environmental epidemiology survey in Italian indoor swimming facilities. *Int J Environ Health Res* 2007; **17**: 197–206.
15 Ali-Shtayeh MS, Khaleel T, Jamous RM. Ecology of dermatophytes and other keratinophilic fungi in swimming pools and polluted and unpolluted streams. *Mycopathologia* 2002; **156**: 193–205.
16 Wadhwani K, Srivastava AK. Fungi from otitis media of agricultural field workers. *Mycopathologia* 1984; **88**: 155–159.
17 Tosti A, Piraccini BM, Lorenzi S, Iorizzo M. Treatment of nondermatophyte mold and *Candida* onychomycosis. *Dermatol Clin* 2003; **21**: 491–497, vii.
18 Brasch J, Koppl G. Persisting onychomycosis caused by *Fusarium solani* in an immunocompetent patient. *Mycoses* 2009; **52**: 285–286.
19 Hattori N, Shirai A, Sugiura Y, *et al*. Onychomycosis caused by *Fusarium proliferatum*. *Br J Dermatol* 2005; **153**: 647–649.
20 Girmenia C, Arcese W, Micozzi A, *et al*. Onychomycosis as a possible origin of disseminated *Fusarium solani* infection in a patient with severe aplastic anemia. *Clin Infect Dis* 1992; **14**: 1167.
21 Nucci M, Anaissie E. Cutaneous infection by *Fusarium* species in healthy and immunocompromised hosts: implications for diagnosis and management. *Clin Infect Dis* 2002; **35**: 909–920.

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