

Investigation of fungal flora in hammams, Turkish baths: A field study

Türk hamamlarında fungal floranın araştırılması: Bir saha çalışması

Ünsal Savcı¹, Mustafa Şahin²

¹ Department of Medical Microbiology, Hitit University, Erol Olcok Education and Research Hospital, Corum, Turkey

² Department of Medical Biochemistry, Hitit University, Erol Olcok Education and Research Hospital, Corum, Turkey

ORCID ID of the author(s)

ÜS: 0000-0003-2319-8171

MŞ: 0000-0001-6073-563X

Abstract

Aim: Hammams, also known as Turkish Hammams or Turkish Baths, have preserved their traditional importance and popularity in various countries, namely, Turkey, Morocco, Yemen, and Algeria. In this study, we aimed to evaluate the fungal flora in hammams and its effects on public health.

Methods: This cross-sectional study was performed by collecting two hundred forty samples from different areas of the baths and the tools used. Collected samples were inoculated on Sabouraud dextrose agar and Potato dextrose agar for mycological evaluation. Agar plates were incubated at 25 °C and 37 °C for four weeks and fungal growth was observed every day. For identification of isolated fungi, micro and macro morphology was evaluated; germ tube test, biochemical tests and VITEK®2 Compact (Biomérieux, France) equipment were used.

Results: We determined that molds are the most common fungi in Turkish hammams. *Aspergillus spp.* (n=20), *Scedosporium apiospermum/boydii* (n=5), *Alternaria ulocladium* (n=1), *Rhizomucor spp.* (n=1) and *Penicillium spp.* (n=1) were isolated in collected samples. Isolated yeasts were *Trichosporon spp.* (n=6), *Candida albicans* (n=1) and *Candida tropicalis* (n=1). *Trichophyton tonsurans*, the dermatophyte, was isolated in two samples. Fungus was most commonly isolated from slippers, and not at all isolated from towels or peshtemals.

Conclusion: In our study, the most isolated molds were fungi, found in the nature, and the isolation rate was exceptionally low. Dermatophytes are the most common culprit of fungal transmission in public places such as baths. Compared to previous studies conducted in swimming pools, wrestling cushions, mosque carpets and slippers, our study showed that Turkish baths with high humidity and temperatures are not rich in fungal flora and that the risk of fungal contamination is low.

Keywords: Hammam, Turkish bath, Fungi, Dermatophytes

Öz

Amaç: Türk hamamı veya Türk banyosu olarak da bilinen hamamlar, günümüzde Türkiye, Fas, Yemen ve Cezayir gibi bazı ülkelerde geleneksel önemini ve popülerliğini korumaktadır. Bu çalışmada hamamlardaki fungal floranın ve halk sağlığına etkilerinin değerlendirilmesi amaçlanmıştır.

Yöntemler: Bu kesitsel çalışma, hamamın farklı alanlarından ve kullanılan aletlerden iki yüz kırk örnek toplanarak gerçekleştirildi. Toplanan örnekler mikolojik değerlendirme için Sabouraud dekstroz agar ve Patates dekstroz agara inoküle edildi. Agar plakları dört hafta boyunca 25°C ve 37°C'de inkübe edildi ve her gün fungal büyüme gözlemlendi. İzole edilen fungusların identifikasyonu için; mikro ve makro morfoloji, germ tüp testi, biyokimyasal testler ve VITEK®2 Compact (Biomérieux, France) cihazı kullanıldı.

Bulgular: Çalışmamızın sonunda, Türk hamamlarında küflerin en yaygın funguslar olduğu belirlendi. Toplanan örneklerden izole edilen küfler, *Aspergillus spp.* (n=20), *Scedosporium apiospermum/boydii* (n=5), *Alternaria ulocladium* (n=1), *Rhizomucor spp.* (n=1) ve *Penicillium spp.* (n=1) idi. İzole edilen mayalar *Trichosporon spp.* (n=6), *Candida albicans* (n=1) ve *Candida tropicalis* (n=1) idi. Dermatofit olarak iki örnekte *Trichophyton tonsurans* izole edildi. En sık fungal izolasyon terliklerde görülürken, havlu ve peştemallerde fungal izolasyon görülmedi.

Sonuç: Çalışmamızda en fazla izole edilen küfler doğada ve çevrede oldukça yaygın bulunan funguslardır ve çalışmamızda izolasyon oranı oldukça düşük bulunmuştur. Hamam gibi ortak kullanılan yerlerde fungal bulaş için en önemli grup dermatofitlerdir. Yüzme havuzları, güreş minderleri, cami halı ve terliklerinde önceki çalışmalar ile karşılaştırıldığında, bizim çalışmamız, yüksek nem ve sıcaklığa sahip Türk hamamlarının fungal flora açısından zengin olmadığını ve fungal kontaminasyon riskinin düşük olduğunu göstermiştir.

Anahtar kelimeler: Hamam, Türk Banyosu, Fungi, Dermatofitler

Corresponding author / Sorumlu yazar:

Ünsal Savcı

Address / Adres: Hitit Üniversitesi, Tıbbi Mikrobiyoloji Anabilim Dalı, Erol Olcok Eğitim ve Araştırma Hastanesi, Çorum, Türkiye
e-Mail: unsalsavci@gmail.com

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Introduction

For thousands of years, people have retreated to hammams to sweat, clean, get scrubbed and socialize with other people. In daily life, hammams are not only traditional cleaning areas, but also centers for health, social and cultural activities. Studies report more than four hundred species of pathogenic fungi commonly isolated from humid locations such as pools, saunas, and public baths, some of which are responsible for serious infections [1]. It is well known that fungal infections are directly transmitted through infected persons, animals, and soil, and indirectly through infected skin and hair follicles [2]. In addition, it is thought that humid environments such as swimming pools and public baths are the main sites for fungal contamination and spreading [3]. Fungi are responsible for allergic conditions and respiratory, dermatological, and central nervous system diseases. Considering their frequent use, hammams are particularly important and effective in terms of public health. However, studies on fungal flora and fungal contamination on Turkish hammams are limited. This study on hammams in cities with medium-sized populations (40,000-260,000 persons) may provide general information about the fungal flora of Turkish hammams.

Materials and methods

This study was performed in accordance with the principles of Declaration of Helsinki. No human or animal material was used in the study and permission was obtained from all bath-hammam establishments. Nine traditional hammams from five different cities in Turkey were included in this study: Tokat, Kırıkkale, Çorum, Yozgat, and Havza. A total of two hundreds forty samples were collected from different areas of the hammams and the tools used: The marble floor, central massage platform, sauna, dressing room, *kurna* edge, body shaving area, slippers, bowl, bath loofahs, towels and *peshtemals*. *Kurna* is a round, mostly marble basin, located under a tap in Turkish hammam, for storing water. *Peshtemal* is a traditional pareo-like cloth, made from special cotton or silk, used to cover and dry the lower part of body. Central massage platform or the navel stone, which is the hottest area of the hammam with underfloor heating, is made from marble, and used for lying down on and having a massage. Body shaving area is the place where body hairs are shaven before entering the hammam. Bath loofah, which is produced from plant or animals, is a coarse bath scrub used for exfoliating the skin. The traditional Turkish hammams usually work in split-schedules specified for male and female customers. Some hammams have separate rooms for men and women, while others serve men and women at different times and days. In our study, four hammams were used by men and women at different times, and five hammams were used by men only. The hammams are usually open from 08.00 to 22.00 on weekdays, and from 08.00 to 24.00 on weekends and vacation days. The hottest places of the hammams were the central massage platforms and saunas. Temperature ranged between 20-60 °C in different areas. After closure, all surfaces were cleaned daily with 5% sodium hypochlorite. For this reason, the samples were collected in the afternoon and before the evening cleaning.

Sample collection and identification method in hammams

Dry sterile cotton swabs were used for wet surfaces. Cotton swabs, moistened with sterile saline (0.09% NaCl), were used for dry surfaces. All samples were collected by sweeping an area of 10 cm in diameter with circular motions. Collected samples were inoculated on unsupplemented and chloramphenicol-and-cycloheximide-supplemented Sabouraud dextrose agar (Oxoid Limited, Basingstoke, Hampshire, UK) for mycological evaluation. Plates were incubated at 25 °C and 37 °C for four weeks and fungal growth was observed every day. The isolated fungi and suspicious dermatophyte samples were subcultured on plates of Potato Dextrose Agar (Oxoid Limited, Basingstoke, Hampshire, UK) for fungal growth. Growing dermatophytes in the plates were observed and sufficiently grown dermatophytes were examined under the microscope using lactophenol cotton blue. Yeasts were identified by using several conventional methods including evaluation of macro-morphology, germ tube test and biochemical tests if necessary. VITEK[®]2 Compact (Biomerieux, France) equipment was used for advanced mycological identifications.

Results

In our study, thirty nine fungi were isolated from two hundred and forty samples of nine Turkish hammams. The provinces where the Turkish Baths where fungal specimens were collected are shown in Figure 1. Microscopic views of fungal specimens collected from Turkish baths are shown in Figure 2. Among the isolated fungi species, molds were the most common. The isolated molds were as follows: *Aspergillus spp.* (n=20), *Scedosporium apiospermum/boydii* (n=5), *Alternaria ulocladium* (n=2), *Rhizomucor spp.* (n=1) and *Penicillium spp.* (n=1). As yeasts, *Trichosporon spp.* were detected in six samples, *Candida albicans* in one sample and *Candida tropicalis* in one sample. As a dermatophyte, *Trichophyton tonsurans* was isolated in two samples. Among personal tools, fungi were most commonly isolated from slippers, and not at all detected in towels and *peshtemals* used in hammams.



Figure 1: The provinces with Turkish Baths where fungal samples were collected

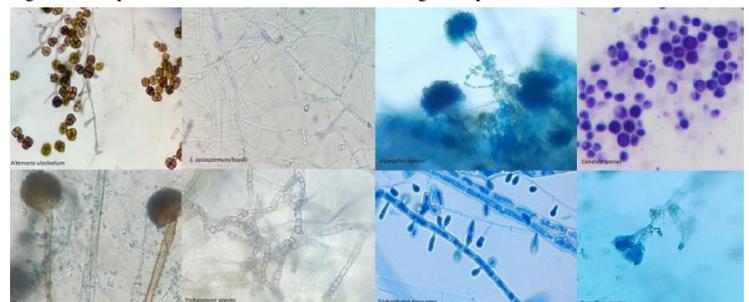


Figure 2: Microscopic views of fungal specimens collected from Turkish baths

Table 1: Fungi distribution in different parts of the hammams and using tools

Fungi	n(%)	Dressing Room Carpet	Floor	Slippers	Bowl	"Kurma"	Body shaving area	Sauna	Central massage platform	Bath loofah	Towel	"Peshtemal"
Mold												
<i>Aspergillus fumigatus</i>	14(35.9)	2	3	5	1	0	2	0	1	0	0	0
<i>Aspergillus terreus</i>	2(5.1)	0	1	0	0	0	0	0	1	0	0	0
<i>Aspergillus niger</i>	2(5.1)	1	0	0	0	0	1	0	0	0	0	0
<i>Aspergillus flavus</i>	1(2.6)	0	0	0	0	0	0	0	1	0	0	0
<i>Aspergillus species</i>	1(2.6)	0	0	0	0	0	1	0	0	0	0	0
<i>S. apiospermum/boydii</i>	5(12.8)	0	1	0	0	2	1	1	0	0	0	0
<i>Alternaria ulocladium</i>	2(5.1)	0	0	2	0	0	0	0	0	0	0	0
<i>Rhizomucor species</i>	1(2.6)	1	0	0	0	0	0	0	0	0	0	0
<i>Penicillium species</i>	1(2.6)	0	0	1	0	0	0	0	0	0	0	0
Yeasts												
<i>Trichosporon species</i>	6(15.4)	1	2	1	1	0	0	0	1	0	0	0
<i>Candida albicans</i>	1(2.6)	1	0	0	0	0	0	0	0	0	0	0
<i>Candida tropicalis</i>	1(2.6)	0	0	0	0	0	0	0	0	1	0	0
Dermatophytes												
<i>Trichophyton tonsurans</i>	2(5.1)	0	0	1	0	0	0	0	1	0	0	0
Total	39(100.0)	6	7	10	2	2	5	1	5	1	0	0

S. apiospermum/boydii: *Scedosporium apiospermum/boydii*

Scedosporium apiospermum/boydii was isolated from one sample collected from the sauna, one of the hottest area of hammams. *Aspergillus spp.* (n=3), *Trichosporon spp.* (n=1) and *Trichophyton tonsurans* (n=1) were isolated from another hot area, the central massage platform. *Trichophyton tonsurans*, a dermatophyte, was isolated once from a slipper sample and once from a central massage platform sample. Among common tools, fungal colonization was most frequently seen in slippers, no colonization was detected on towels and peshtemals. *Candida albicans* was isolated from the dressing room carpet in one sample. *Candida tropicalis* was isolated from a bath loofah in one sample. *Aspergillus fumigatus*, and *Trichosporon spp.* were each isolated once from the bowls used in hammams. The distributions of the fungi isolated from different parts of the bath and the tools used are shown in Table 1.

Discussion

Turkish hammams are Finnish sauna-variants, only humid. The average temperatures in Finnish saunas and Turkish hammams are 80°C and 60°C, respectively [4]. In previous studies, the presence of dermatophytes was shown on the wrestling mats, floors of swimming pools and carpets [5]. Positive results were obtained for fungal contamination in all studies conducted in hammams. Physical environmental factors, including humidity and temperature, not only initiate fungal growth, but also stimulate fungal colonization [6]. Dermatophytes can survive for several years on the epithelium sheds and be transmitted indirectly to healthy people through the remnants of the epithelium sheds in the environment [7]. Hannuksela et al. [8] investigated the benefits and risks of Finnish saunas on human health and reported that fungal infections increased the risk. Although there are many scientific reports on fungal infections, investigations about fungal diversity and contamination in hammams in Turkey still do not suffice. Up until now, only one study regarding fungal flora conducted in two hammams has been published by Goksugur et al. [4] in 2006 in Turkey. In that study, Goksugur et al. [4] isolated 16 fungi species from 209 samples collected from traditional Turkish hammams. The isolated fungi species in this study were *Aspergillus spp.* (n=4), *Penicillium spp.* (n=4), *Trichophyton rubrum* (n=3), *Trichophyton mentragrophytes* (n=1), *Epidermophyton floccosum* (n=1), *C. albicans* (n=2), and *C. tropicalis* (n=1). The results of this study were similar to ours.

Being the most isolated mold, *Aspergillus* is known to cause many diseases in humans, from allergic reactions to respiratory tract colonization, aspergilloma and tissue invasions, mostly through pre-existing colonization [9]. In our study, *S. apiospermum/boydii* was isolated in five samples. *Scedosporium* is a rare pathogen which most frequently causes sinopulmonary and central nervous system diseases in both immunocompromised and immunocompetent humans [10]. These fungi are airborne saprobes that survive and grow in decomposing organic matter, which is why they reside in sewage areas, places with polluted water and urban soil, affecting nearby populations and causing serious infections [11]. Molds are quite common in the environment we live in. Reports show that *Fusarium* induces onychomycosis and lower-extremity skin lesions in neutropenic patients, and *Rhizopus* induces sinus infections in patients with diabetic ketoacidosis. *Alternaria* is an important culprit of paranasal sinusitis in both healthy humans and immunocompromised patients [9]. In our study, common tools were evaluated for fungal reproduction. Although there was no fungal growth or activation on towels and peshtemals, slippers were most frequently found to house fungal colonies. Washing towels and peshtemals in the washing machine at high temperatures after each use and providing disposable articles for each customer may be effective for preventing fungal contamination. Insufficient disinfection and washing of the slippers only once at the end of the day may explain the rich fungal colonization. The central massage platforms and saunas are the hottest parts of the Turkish Hammams with an average temperature around 60°C. In our study, *S. apiospermum/boydii* was isolated in only one sample collected from saunas. The high temperature limit for growth of any thermophilic eukaryotic organism is about 62-65 °C, so dermatophytes and other infective microorganisms are thought to be easily killed by the high temperatures in the Hammams [12]. In our study, five fungal colonies were grown from samples collected from the central massage platform, which were *Aspergillus spp.* (n=3), *T. tonsurans* (n=1) and *Trichosporon spp.* (n=1). Central massage platforms and saunas had similar ambient temperatures, so what was the reason for the difference in fungal colonization? This question can be answered by comparing the use of saunas and central massage platforms. In traditional hammam culture, customers use slippers and sit in the saunas with peshtemals. The saunas in hammams are used for sweating purposes only. There is little skin contact with the floor and sitting areas, which is quite different from the central massage platforms. The customers lie on the marble central massage platforms with their

whole bodies after removing their slippers while they receive the exfoliation and massage services. At this time, the feet and almost all skin surfaces are in contact with the marble floor. We think that the epithelium, poured on the marble floor during massage and exfoliation, may increase the risk of fungal contamination. As mentioned in some studies, it has been reported that central massage platforms are quite rich for keratin derived from skin and other skin waste products, which are nutritional source of fungi [13,14]. Yenisehirli et al. [5] detected dermatophytes in 113 out of 144 samples in a study on mosque carpets. Interestingly, in our study, no dermatophyte could be isolated on the hammam dressing room carpets. However, six fungal colonies were isolated from samples collected from the dressing room carpets in hammams: *Aspergillus spp.* (n=3), *Rhizomucor spp.* (n=1), *Trichosporon spp.* (n=1), and *C. albicans* (n=1). More frequent disinfection, cleaning and washing of hammam dressing room carpets than mosque carpets may explain this significant difference. However, it is advisable to use an easier-to-clean flooring material in the dressing rooms instead of the carpets. Benammar et al. [1] isolated eight yeast species, *C. albicans*, *C. glabrata*, *C. tropicalis*, *C. lipolytica*, *Geotrichum spp.*, *Trichosporon spp.*, *Rhodotorula spp.* and *Cryptococcus spp.* in a traditional hammam study in Algeria. In our study, *C. albicans*, *C. tropicalis* and *Trichosporon spp.* were isolated. Being opportunistic fungi, *Candida* species can cause clinical infections in all organs, which include superficial mucosal infections, cutaneous candidiasis and extensive hematogenous spread involving organs such as the liver, spleen, heart and brain. Mucosal infections (known as oral candidiasis) linked with *Candida* species may be limited to the oropharynx or may spread to the esophagus and the whole gastrointestinal tract [9]. *Trichosporon* species are normal flora members of the gastrointestinal and genitourinary tract in humans but may temporarily colonize the skin and respiratory system [15]. In such studies, the reasons for reduced isolation of yeast groups may be related to the sample collecting method, or some fungi such as the *A. flavus*, which may exert antifungal effects against *C. albicans* and *C. Tropicalis* [16]. On the other hand, filamentous fungal hyphae can cause this result by rapidly growing and limiting the growth of yeasts in the environment [17]. The isolation rates of dermatophytes in hammam studies were reported as 0.98% by Benammar et al. and 2.39% by Goksugur et al [1,4]. In our study, the rate of dermatophyte isolation was 2.3%, which was similar to other studies. In our study, *T. tonsurans* were isolated from two samples only. *Tinea corporis* and *Tinea capitis* are the two most common clinical forms of *T. tonsurans* infection [18].

The limitations of this study are the use of conventional methods and the absence of molecular methods in fungal isolation.

Conclusion

In our study, the most isolated molds were fungi, as found in the nature. The isolation rate was very low. Dermatophytes are the most important culprit for fungal transmission in public places, such as baths. Compared to previous studies in swimming pools, wrestling cushions, mosque carpets and slippers, the outcomes of our study showed that

Turkish Hammams had a very low risk for fungal contamination. Especially in hammams, regular disinfection and cleaning of the common areas and the tools, use of personal or disposable slippers, and informing customers about fungal infection transmission routes and prevention could reduce risk of fungal contamination.

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