

Fungal Airborne Contamination as a Serious Threat for Respiratory Infection in the Hematology Ward

Ali Ghajari ¹, Ensieh Lotfali ¹, Mansour Azari ², Roohollah Fateh ³, Saba Kalantary ⁴

¹Department of Medical Parasitology and Mycology, School of Medicine, Shahid Beheshti University of Medical Sciences, Tehran, Iran, ² Safety Promotion and Injury Prevention, Shahid Beheshti University of Medical Sciences, Tehran, Iran, ³ Department of Microbiology and Immunology, Faculty of Medicine, Qom University of Medical Sciences, Qom, Iran, ⁴ College of Public Health Tehran University of Medical Sciences, Tehran, Iran.

Received: 27 July 2015

Accepted: 29 October 2015

Correspondence to: Ghajari A

Address: School of Medicine, Shahid Beheshti University of Medical Sciences, Tehran, Iran

Email address: alighadjar@sbmu.ac.ir

Background: Fungi existing in hospital departments may grow and produce micro-colonies. The spores arising from these micro-colonies circulate easily and could be inhaled by patients and cause infections in immune-compromised subjects. Due to the lack of an acceptable method of sampling and evaluation of microbiological quality of air in the isolation units, the purpose of this study was to determine the concentrations of airborne fungi through active and passive sampling and also identify fungi genera in the air of the isolation unit.

Materials and Methods: The air of the isolation unit was monitored through active and passive sampling. In passive sampling, the plates were placed in the room. The active sampling was performed in the hematology unit by using a slit-to-agar biological air sampler with a flow rate of 10 L/minute. Plates were incubated at 30°C for 10 days and were examined daily for fungal growth. Fungal species were identified on the basis of their macroscopic and microscopic morphological features.

Results: In active samples, *Penicillium spp.* was the predominant genus (66.8%), followed by *Aspergillus spp.* (23.9%) and *Cladosporium spp.* (2.5%). *Yeast spp.* accounted for only 2.2% of the isolated fungi.

In passive samples, *Penicillium spp.* (94.4%) was the most frequently found fungi, followed by *Aspergillus spp.* (2.2%), *Cladosporium spp.* (1.1%) and *Yeast spp.* (0.5%). The identified genera included *Penicillium*, *Aspergillus*, *Alternaria*, *Mucorales*, *Cladosporium*, *Yeasts* and other filamentous fungi.

Conclusion: Active and passive sampling can be used for monitoring the fungal content of air. Assessment of fungal contamination profiles in hospitals may provide important information about the level of fungal concentration in the hospitals and for the control of nosocomial infections. In addition, installation of special ventilation systems equipped with HEPA filters in hematology wards could enhance the quality of air. Also, observing sanitary protocols for disinfection of the surfaces is imperative for infection control.

Key words: Fungal contamination, Active and passive sampling, Hematology unit

INTRODUCTION

Some hospital infections are caused by fungi. Fungal contamination of air has been the subject of several researches (1). Fungi existing in hospital units may grow

and produce micro-colonies. The spores arising from these micro-colonies circulate easily and could be inhaled by patients and cause respiratory infections in immune-compromised subjects (2). Level of fungal contamination of

indoor air of hospital units depends on several factors such as air circulation, moisture and temperature (3). Infectious diseases caused by inhalation of different fungal bio-aerosols depend on the biological and chemical properties, the number and the size of fungi inhaled and the site of their deposition in the respiratory system (4).

Fungi species with diameters smaller than 5 μm such as *Aspergillus fumigatus* spores (2 to 3 μm) are able to penetrate into the alveoli and are capable of causing pulmonary infections in humans. Aspergillosis and other systemic mycoses have caused an increasing morbidity among immunologically compromised hospitalized patients (5).

The climatic conditions of the hospitals were reported to play an important role in the biological quality of their indoor air (6-8) and the quality of filtered air conditioning in hospitals was reported to be an effective means of decreasing nosocomial infections (6, 9, 10). The use of the High Efficiency Particulate Air (HEPA) filtration system was reported to prevent invasive pulmonary aspergillosis in immune-suppressed patients in isolation units (6, 11).

Active or passive sampling of indoor air for possible airborne microbial contamination has been reported by several authors (6, 12, 13). There are currently no specific protocols for airborne monitoring of fungal contamination in the form of passive or active sampling (12, 14).

The content of microorganisms in the air through active sampling is expressed in Colony Forming Units (CFUs)/ m^3 and sampling is performed when the concentration of microorganisms is not very high. However, passive sampling provides an actual assessment as it measures the airborne particles, which fall onto a critical surface, and results of passive monitoring are expressed in CFUs/ m^2 /hour (6, 11, 15).

Due to importance of quality of air within an isolation unit with high-risk patients, the purpose of the present study was to determine the quality and quantity of airborne fungi in the isolation unit of a hospital via active and passive sampling.

MATERIALS AND METHODS

This study was performed to determine the microbiological quality of air in the isolation unit of the Hematology Ward of Taleghani Hospital located in Tehran in March 2014. This ward has 6 separate rooms with 27 beds. Air sample was collected once a week for passive sampling and once a week for active sampling from 30 locations at 10:00 a.m., with a total of 30 passive and 30 active samples. In both samplings, the plates were placed in room approximately 1m above the floor and about 1m away from the walls. The number of plates per square meter was 6/18. The distance between the six measuring plates was 3m.

The number of colonies counted on each petri dish with a diameter of 9cm sampled for one hour was expressed as CFUs/ m^2 /hour (16).

Active samplings were performed in all rooms of the Hematology Unit using a slit-to-agar biological air sampler (Casella Air Bacteria Sampler MK II with Casella pump T 13692), with a flow rate of 10 L/minute. This sampler drew air at high speed through a narrow slit and blew it over a solid Sabouraud chloramphenicol dextrose agar (SabC, Becton-Dickinson, Heidelberg, Germany) plate. The plate rotated at a uniform speed under the slit (17).

The number of CFUs was adjusted using the conversion table provided by the manufacturer and the value was expressed in CFUs/ m^3 . Samples were processed in the clinical mycology laboratory of the Shahid Beheshti University of Medical Sciences. Plates were incubated at 30°C for 10 days and were examined daily for fungal growth. Fungal species were identified on the basis of their macroscopic and microscopic morphological features (18).

RESULTS

In this study, 60 samples (passive and active) were taken during the same period. The mean load of isolated fungi was 10 CFUs/ m^3 and

1 CFU/m²/hour in active samples and in passive samples, respectively (Table 1).

The frequencies and the loads of the fungi isolated from the Hematology Unit are demonstrated in Table 2. The identified genera included *Penicillium*, *Aspergillus*, *Alternaria*, *Mucorales*, *Cladosporium*, *Yeasts* and other Filamentous fungi. The highest mean fungal load was noticed for the *Penicillium spp.* (12CFU/m²/h) in passive samples. The mean fungal load of other fungi was low, ranging from 1 to 2CFUs/m²/hour. But in active samples, the dominant mean fungal load was *Penicillium spp.* (6 CFUs/m³), followed by *Aspergillus spp.* (3 CFUs/m³).

Concerning percentages of fungi isolated from the air of the Hematology Unit, *Penicillium spp.* was the predominant genus (66.8%), followed by *Aspergillus spp.* (23.9%) and *Cladosporium spp.* (2.5%), and *Yeast spp.* accounted for only 2.2% of the isolated fungi in active sampling.

In passive samples, *Penicillium spp.* (94.4%) was the most frequently found fungi, followed by

Aspergillus spp. (2.2%), *Cladosporium spp.* (1.1%) and *Yeast spp.* (0.5%).

DISCUSSION

In our study, indoor air contamination of the Hematology Ward of Taleghani Hospital was assessed by both active and passive sampling. Both sampling methods detected fungal air contamination and preference of either active or passive sampling was not observed. According to our data, the passive sampling and subsequent analysis predicted the possible contamination rate at the surgical site and it permitted direct measurement of the quantity of microorganisms on selected surfaces, and these observations have also been confirmed by other authors (6, 12, 19, 20). In contrast, the active sampling and analytical method were performed to obtain information on the concentration of all inhalable viable particles, which has also been confirmed by other authors (6, 12).

Table 1. Concentrations of fungi collected in the Hematology Unit with the active and passive sampling methods

Methods	Total number of plates	Total fungal count	Mean load of fungi
Active method	30	43.8 CFU/m ³	10 CFU/m ³
Passive method	30	180 CFU/m ² /h	1 CFU/m ² /h

Table 2. Fungi identified in the air of the Hematology Unit during the study period.

Fungal species	Positive samples (active)		Positive samples (passive)	
	Absolute number	Mean fungal load (CFUs /m ³)	Absolute number	Mean fungal load (CFUs /m ² /hour)
<i>Acremonium spp.</i>	2	1	1	1
<i>Aspergillus spp.</i>	105	3	4	1
<i>Alternaria spp.</i>	2	2	1	1
<i>Cladosporium spp.</i>	11	1	2	1
<i>Epicoccum spp.</i>	1	1	1	1
<i>Fusarium spp.</i>	1	1	1	1
<i>Mucor spp.</i>	3	1	-	-
<i>Paecilomyces spp.</i>	5	1	3	1
<i>Penicillium spp.</i>	293	6	170	12
<i>Trichoderma spp.</i>	8	1	2	2
<i>Yeast spp.</i>	10	1	1	1

In our study, concerning the mean load of fungi isolated in both sampling, *Aspergillus spp.* (10.5 CFUs/m³) and *Penicillium spp.* (6 CFUs/m³) in active sampling and *Penicillium spp.* (12 CFUs/m³) in passive sampling were the most frequent. The incidence of these two genera has been formerly observed in other hospitals and authors suggested that airborne contamination mainly relates to micromycetes that have a specific ability to adapt to the environment within buildings (21-23).

In another study, Sautour et al. described the tree leaves containing nutrients, which promote the growth of fungi especially *Aspergillus* spore in autumn and winter (1). However, results of this study showed the abundance of *Penicillium spp.*, which might be related to plants and soil and the construction activities close to the hospital. This phenomenon was also reported in cases of earth removal, wood-cutting operations and digging, and air currents through windows could increase the number of spores in the rooms (1, 24).

Comparing the results of this study with previous ones about the quality of indoor air of hospitals, lower airborne concentrations of *Cladosporium spp.* were observed in our study than in the hospitals in France and Asia (21, 23, 25). This phenomenon might be due to less humidity and open air circulation of hospitals in Tehran.

The greatest difference between active and passive samples in this study was related to the detection of *Aspergillus spp.*. This finding might be due to the adherence quality and existence of reservoirs for this species in the environment (26). It is possible that a more thorough daily cleaning of surfaces in hospital units was responsible for control of this species observed in passive samples. The sources of infection in many outbreaks of chronic pulmonary diseases were nosocomial aspergillosis, which might be due to the spores of *Aspergillus spp.* (27-29).

Spore concentration in this study was 10 CFUs/m³. Although the threshold spore concentration has not been defined by the health authorities, few studies demonstrated significant risk of fungal infection

occurrence with *Aspergillus* spores >2 CFUs/m³ in indoor air (29-31).

Since transmission of the spores of *Aspergillus spp.* through breathing of contaminant air is considered as the main route of pulmonary infection (5), installation of special ventilation system equipped with HEPA filters in hematology wards may enhance the quality of air. Observing sanitary protocols for disinfection of the surfaces is imperative for infection control.

REFERENCES

1. Sautour M, Sixt N, Dalle F, L'Ollivier C, Fourquenot V, Calinon C, Paul K, Valvin S, Maurel A, Aho S, Couillault G, Cachia C, Vagner O, Cuisenier B, Caillot D, Bonnin A. Profiles and seasonal distribution of airborne fungi in indoor and outdoor environments at a French hospital. *Sci Total Environ* 2009; 407 (12): 3766-71.
2. Singh N, Paterson DL. *Aspergillus* infections in transplant recipients. *Clin Microbiol Rev* 2005; 18 (1): 44-69.
3. Medrela-Kuder E. Seasonal variations in the occurrence of culturable airborne fungi in outdoor and indoor air in Cracow. *International Biodeterioration & Biodegradation* 2003; 52(4): 203-5.
4. Pastuszka JS, Paw UK, Lis DO, Wlazlo A, Ulfik K. Bacterial and fungal aerosol in indoor environment in Upper Silesia, Poland. *Atmospheric Environment* 2000;34(22):3833-42.
5. Streifel AJ, Lauer JL, Vesley D, Juni B, Rhame FS. *Aspergillus fumigatus* and other thermotolerant fungi generated by hospital building demolition. *Appl Environ Microbiol* 1983;46(2):375-8.
6. Napoli C, Marcotrigiano V, Montagna MT. Air sampling procedures to evaluate microbial contamination: a comparison between active and passive methods in operating theatres. *BMC Public Health* 2012;12:594.
7. Sehulster L, Chinn RY; CDC; HICPAC. Guidelines for environmental infection control in health-care facilities. Recommendations of CDC and the Healthcare Infection Control Practices Advisory Committee (HICPAC). *MMWR Recomm Rep* 2003;52(RR-10):1-42.

8. Vescia N, Brenier-Pinchart MP, Osborn JF, Cerquetani F, Cavarischia R, Grillot R, D'Alessandro D. Field validation of a dusting cloth for mycological surveillance of surfaces. *American journal of infection control* 2011;39(2):156-8.
9. Beldi G, Bisch-Knaden S, Banz V, Mühlemann K, Candinas D. Impact of intraoperative behavior on surgical site infections. *Am J Surg* 2009;198(2):157-62.
10. Demir F. A survey on prevention of surgical infections in operating theaters. *Worldviews on Evidence- Based Nursing* 2009;6(2):102-13.
11. Oren I, Haddad N, Finkelstein R, Rowe JM. Invasive pulmonary aspergillosis in neutropenic patients during hospital construction: before and after chemoprophylaxis and institution of HEPA filters. *Am J Hematol* 2001;66(4):257-62.
12. Standard B, Iso B. Cleanrooms and associated controlled environments Biocontamination control.
13. Pasquarella C, Albertini R, Dall'aglio P, Sacconi E, Sansebastiano GE, Signorelli C. Air microbial sampling: the state of the art. *Ig Sanita Pubbl* 2008;64(1):79-120.
14. ISPESL: Istituto Superiore per la Prevenzione e la Sicurezza del Lavoro. Linee guida per la definizione degli standard di sicurezza e di igiene ambientale dei reparti operatori. 2009.
15. Gosden PE, MacGowan AP, Bannister GC. Importance of air quality and related factors in the prevention of infection in orthopaedic implant surgery. *J Hosp Infect* 1998;39(3):173-80.
16. Pasquarella C, Pitzurra O, Savino A. The index of microbial air contamination. *J Hosp Infect* 2000;46(4):241-56.
17. Azari MR, Ghajari A, Nejad MR, Nasiree NF. Airborne microbial contamination of dental units. *Tanaffos* 2008;7(2):54-7.
18. de Hoog GS, Guarro J, Gené J, Figueras MJ. Atlas of Clinical Fungi, 3rd. Utrecht: CBS-KNAW Fungal Biodiversity Centre. 2009.
19. Kundsins RB. Architectural design and indoor microbial pollution. Oxford University Press, USA; 1988.
20. Whyte W. In support of settle plates. *PDA J Pharm Sci Technol* 1996;50(4):201-4.
21. Dassonville C, Demattei C, Detaint B, Barral S, Bex-Capelle V, Momas I. Assessment and predictors determination of indoor airborne fungal concentrations in Paris newborn babies' homes. *Environ Res* 2008;108(1):80-5.
22. Sautour M, Sixt N, Dalle F, L'ollivier C, Calinon C, Fourquet V, et al. Prospective survey of indoor fungal contamination in hospital during a period of building construction. *J Hosp Infect* 2007; 67 (4): 367-73.
23. Wu PC, Su HJ, Ho HM. A comparison of sampling media for environmental viable fungi collected in a hospital environment. *Environ Res* 2000; 82(3): 253- 7.
24. Horner WE, Worthan AG, Morey PR. Air- and dustborne mycoflora in houses free of water damage and fungal growth. *Appl Environ Microbiol* 2004; 70(11): 6394- 400.
25. Faure O, Fricker-Hidalgo H, Lebeau B, Mallaret MR, Ambroise-Thomas P, Grillot R. Eight-year surveillance of environmental fungal contamination in hospital operating rooms and haematological units. *Journal of hospital infection* 2002; 50(2):155-60.
26. Cornet M, Levy V, Fleury L, Lortholary J, Barquins S, Coureul MH, et al. Efficacy of prevention by high-efficiency particulate air filtration or laminar airflow against Aspergillus airborne contamination during hospital renovation. *Infect Control Hosp Epidemiol* 1999;20(7):508-13.
27. Cheng SM, Streifel AJ. Infection control considerations during construction activities: land excavation and demolition. *Am J Infect Control* 2001;29(5):321-8.
28. Lutz BD, Jin J, Rinaldi MG, Wickes BL, Huycke MM. Outbreak of invasive Aspergillus infection in surgical patients, associated with a contaminated air-handling system. *Clin Infect Dis* 2003; 37(6): 786-93.
29. Vonberg RP, Gastmeier P. Nosocomial aspergillosis in outbreak settings. *J Hosp Infect* 2006;63(3):246-54.
30. Alberti C, Bouakline A, Ribaud P, Lacroix C, Rousselot P, Leblanc T, et al. Relationship between environmental fungal contamination and the incidence of invasive aspergillosis in haematology patients. *J Hosp Infect* 2001;48(3):198-206.
31. Perdelli F, Cristina ML, Sartini M, Spagnolo AM, Dallera M, Ottria G, et al. Fungal contamination in hospital environments. *Infect Control Hosp Epidemiol* 2006;27(1):44-7.