Field validation of a dusting cloth for mycological surveillance of surfaces

Nicoletta Vescia, MD, ScD,[a] Marie-Pierre Brenier-Pinchart, MD, PhD,[b] John Frederik Osborn, BSc, PhD,[a] Franco Cerquetani, MD,[c] Roberta Cavarischia, ScD,[d] Renée Grillot, PhD, PharmD,[b] and Daniela D'Alessandro, MD, MPH, PhD[e]
Rome, Italy, and Grenoble, France

Efficient monitoring of surfaces for spores of filamentous fungi is essential for detecting minor contamination even when air samples test negative for fungi. This study evaluates and compares a pad prepared using a dusting cloth with Rodac contact plates and humidified swabs for detecting mycological contamination, and concludes that the new method is superior and cheaper.
Key Words: Environmental monitoring; *Aspergillus* species; sampling methods.

Invasive aspergillosis (IA) is a major cause of death in severely immunocompromised patients, especially those undergoing cytotoxic therapy for hematologic malignancies with associated prolonged neutropenia, and recipients of a solid organ transplantation.[1] The concentration of spores of *Aspergillus* and other fungal species in the air in hematologic units has been correlated with epidemic and nonepidemic situations of hospital-acquired IA.[1,2] Because the diagnosis of IA is difficult, treatment success is poor, with mortality approaching or exceeding 90% in high-risk patient groups. Controlling air contamination by mechanical and physical means is imperative,[3,4] and periodic environmental surveillance via air and surface sampling is recommended.[5] Surface sampling is a simple and efficient monitoring method that can detect minor contamination even when air samples test negative for fungi;[2,5] however, current methods for sampling surfaces can capture <30% of real microbial contamination.[6]

With the aim of simplifying and improving surface sampling, we developed a simple flat tampon (Ø 4.5 cm) prepared covering a cotton disk (2 mm thick) with a common electrostatic dusting cloth (DC pad) selected among those commonly available in the market. Results of a previous experimental study comparing the efficiency of these DC pads with contact plates and cotton swabs in sampling aspergillar spores from surfaces suggested that the DC pads were significantly more effective.[7]

The aim of the present study was to compare DC pads with Rodac contact plates and humidified swabs in terms of their ability to identify a contamination and to capture filamentous fungi (FF) on surfaces in high-risk hospital environments.

From the Department of Public Health Sciences, Sapienza University of Rome, Italy[a]; Parasitology-Mycology, Teaching Hospital A. Michallon, Grenoble, France[b]; Medical Direction Unit, "San Filippo Neri" Hospital, Rome, Italy[c]; Clinical Pathology Unit, "Villa Betania" Hospital, Rome, Italy[d]; and Department of Civil Building and Environmental Engineering, Sapienza University of Rome, Rome, Italy.[e]

Address correspondence to Daniela D'Alessandro, MD, MPH, PhD, Department of Architecture and Planning for Engineering, Sapienza University, Via Eudossiana, 18, 00186 Rome, Italy. E-mail: daniela.dalessandro@uniroma1.it.

Conflicts of interest: None to report.

0196-6553/$36.00

doi:10.1016/j.ajic.2010.05.018

METHODS

Three hospitals (two in Rome and one in Grenoble) were included in this study. The Italian group collected samples in 18 operating theaters before routine cleaning, whereas the French group collected the samples in 12 rooms in two hematology units. Sampling points in each operating theater included an air outlet grid, the top of a scialitic lamp, and an electrical socket. In the hematology units, samples were taken from an air inlet grid, a television screen, an adaptable table, a bar under the bed, an electric appliance, the technical rail, and electrical sockets. In the operating theaters, 126 surfaces were sampled for a total of 292 samples, and in the hematology units, 96 surfaces were sampled, yielding 192 samples. Each surface was divided into two equal parts, with the DC pad used in one part and the Rodac plate or swab used in the other part. A total of 484 samples were collected, as follows:

- Top of scialitic lamp: 42 samples for the DC pad and 84 samples for two Rodac plates

Table 1. Comparison of DC pads and other methods of sampling for FF

Surfaces	Number of pairs	DC$^+$/other$^+$	DC$^+$/other$^-$	DC$^-$/other$^+$	DC$^-$/other$^-$	% positive		P*	Geometric means ratio†	95% CI	P‡
						DC	Other				
Air grid	53	21	9	1	22	56.6	41.5	<.001	3.72	2.32-5.95	<.001
Scialitic lamp	42	25	12	0	5	88.1	50.0	<.001	3.54	2.60-4.81	<.001
Electric socket	66	24	26	2	14	75.8	39.4	<.001	2.72	2.04-3.64	<.001
Other surfaces	60	14	22	2	22	60.0	26.7	<.001	3.45	2.28-5.23	<.001

*McNemar's test.
†Comparison between DC pad and other methods.
‡*t* test for dependent sample.

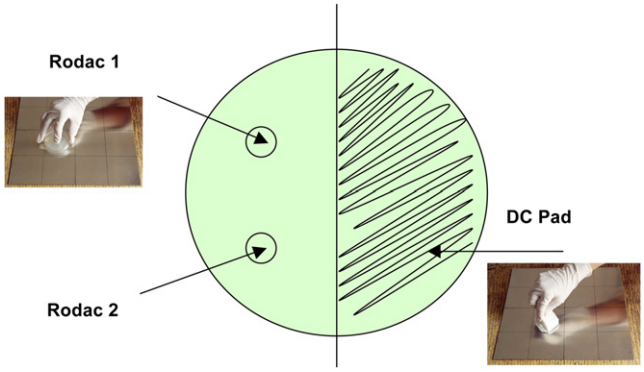

Fig 1. Sampling points and methods used on the scialitic lamp.

- Air grids and irregular surfaces: 53 samples for the DC pad and 53 samples for the humidified swab
- Electrical sockets: 66 samples for the DC pad and 66 samples for one Rodac plate
- Smooth, regular surfaces: 60 samples for the DC pad and 60 samples for one Rodac plate.

The sampling method used for the swabs and Rodac plates was as recommended in European Standard - International Organization for Standardisation (EN ISO) 14698-1:2004.[8] For sampling with the DC pad, the dry sterilized tampon was rubbed on the surface, and the dust captured was inoculated on a Petri plate (Ø 9 cm). The Rodac and Petri plates contained Sabouraud dextrose agar + chloramphenicol + neutral (Merck in Grenoble; BD in Rome). The plates were incubated for 7 days at 27° ± 1°C, and molds were identified to the genus level.[5,9]

The sampled area was identical for most of the sampling points, with the exception of the scialitic lamp, on which the half sampled with the DC pad was rubbed with a single pad, whereas two Rodac plates were used to sample the other half (Fig 1). We were interested not only in quantifying the number of colony-forming units (CFU) captured per cm^2, but also in increasing the sensitivity for identifying a contaminated surface.

The χ^2 test was used to compare the overall proportions of samples found to be positive for FF. The proportions of positive samples were also compared for venue (Grenoble and Rome), surface investigated, and sampling method. The McNemar test was used to test the difference between paired proportions. Surfaces on which neither method detected colonies were excluded. To quantify the differences in capturing mold between the DC pads and contact plates or cotton swabs, the *t* test for dependent samples was used after logarithmic transformation. This test was applied to each sampling surface.

RESULTS

Overall, 69.2% (153/221) of the DC pad samples were positive, compared with 40.3% (106/263) of the other samples ($P < .005$). The difference persists when stratifying by venue (Grenoble or Rome), by type of sampled surfaces (air grids, scialitic lamps, electric sockets, other surfaces), or by sampling method (Table 1).

DISCUSSION

The incidence of IA has been correlated with surface and air fungal contamination in both epidemic and nonepidemic situations.[2] This suggests the need for environmental control interventions and periodic mycological surveillance in high-risk units to quickly detect malfunctioning air-conditioning systems or inadequate application of cleaning procedures and measures to prevent fungal contamination of the ward.[5] It has been established that (1) air fungi spores tend to settle on surfaces, and fungal contamination of surfaces is considered to reflect previous air contamination;[2] (2) surface sampling can detect minor contamination even when air samples test negative for fungi;[5] (3) monitoring of surfaces is cheap, easy and efficient; and (4) the methods for sampling surfaces in use are able to capture about 30% of real microbial contamination.[6]

Our evaluation of the DC pads suggests that these pads can be used to sample any type of surface and will detect more than twice the CFU as Rodac contact

plates and humidified swabs. Furthermore, the DC pad can monitor an entire critical surface with just a single sample, thus reducing costs. We conclude that the DC pad can be used for environmental monitoring during the implementation and testing of air-conditioning systems, after the maintenance of these systems, and for periodic mycological surveillance, as has been recommended.[9]

References

1. Warnock DW, Hajjeh RA, Laser BA. Epidemiology and prevention of invasive aspergillosis. Curr Infect Dis Rep 2001;3:507-16.
2. 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 hematology patients. J Hosp Infect 2001;48:198-206.
3. Centers for Disease Control and Prevention. Prevention and control of health care–associated pulmonary aspergillosis. Morb Mortal Recomm Rep 2004;53(RR-03):15-7.
4. Bénet T, Nicolle MC, Thiebaut A, Piens MA, Nicolini FE, Thomas X, et al. Reduction of invasive aspergillosis incidence among immunocompromised patients after control of environmental exposure. Clin Infect Dis 2007;45:682-6.
5. Gangneux JP, Bretagne S, Cordonnier C, Datry A, Derouin F, Grillot R, et al. Prevention of nosocomial fungal infection: the French approach. Clin Infect Dis 2002;35:343-6.
6. Association pour la prévention et l'étude de la contamination. La biocontamination: Salles propres, environnements maîtrisés & zones de confinement. ASPEC Guide, October 2008. p. 194-5.
7. Vescia N, D'Alessandro D, Osborn JF, Grillot R. Development of an innovative method for the evaluation of fungal contamination of surfaces. Ann Ig 2008;20:1-6.
8. EN (European Standard) - ISO (International Organization for Standardisation) 14698–1:2004. Clean rooms and associated controlled environments. Biocontamination control. Part 1: General principles and methods, 2004.
9. Gangneux JP, Poirot JL, Morin O, Derouin F, Bretagne S, Datry A, et al. Surveillance mycologique de l'environnement pour la prévention de l'aspergillose invasive: propositions de standardisation des méthodologies et des modalités d'application. Press Med 2002;31:841-8.