Atmospheric Pollution Research 7 (2016) 659–663

Contents lists available at ScienceDirect

Atmospheric Pollution Research

journal homepage: http://www.journals.elsevier.com/locate/apr

Original article

Investigation of fungal volatile organic compounds in hospital air

Lydia Dayanne Maia Pantoja [a, *], Ronaldo Ferreira do Nascimento [b], Ana Barbara de Araújo Nunes [b]

[a] Federal University of Ceará – UFC, Avenida Mister Hulls, S/n–Bloco 713, Campus do Pici, Fortaleza, Ceará, Brazil
[b] Federal University of Ceará – UFC, Fortaleza, Ceará, Brazil

ARTICLE INFO

Article history:
Received 10 November 2015
Received in revised form
21 February 2016
Accepted 23 February 2016
Available online 24 March 2016

Keywords:
Fungi
Microbial volatile organic compounds
Hospital units
2-Heptanone
Aspergillus

ABSTRACT

Fungal growth within the structure of buildings or in ventilation filters generates “hidden contamination”, which cannot be detected only through visual inspection. At the beginning of development, the fungi release fungal volatile organic compounds (FVOCs) into the atmosphere, which can originate from metabolic pathways or from the enzymatic degradation of materials. This study analyzed the air quality of a public referral hospital in Fortaleza, Ceará, Brazil in terms of FVOCs, to establish ways to improve methods of monitoring and control of specific sectors in the hospital. For that, we created and validated a protocol for detection of FVOCs, using GC/MS, while fungal samples were identified by analysis of macro and micromorphology. In total, 48 samples (60.5% positive) were analyzed for FVOCs; 7 were detected in at least one of the sectors analyzed, with 2-heptanone (179.5 $\mu g/m^3$) and 2-methyl-1-propanol (121.5 $\mu g/m^3$) as the most abundant. With respect to fungal findings, 24 samples were analyzed, with a high number of colony-forming units per cubic meter (CFU/m^3) observed in all sectors. The airborne fungal spectrum revealed the existence of 19 genera, composed predominantly by hyaline filamentous deuteromycetes. Analysis with periodic monitoring is still needed to allow improvement in the data quality. Also, further discussion on the subject in the academic and legislative environment is needed to contribute to the systematic study of aerobiology.

1. Introduction

Monitoring the air quality and its biomarkers (fungi) in hospitals is increasingly important because of the growing number of patients with acquired or induced immunosuppression, due to cancer treatment, transplantation of bone marrow or solid organs, acquired immunodeficiency virus infection or prolonged administration of corticosteroids, which make patients vulnerable to opportunistic fungal infections (Moretti, 2007; Munoz et al., 2015).

According to the literature, in developed countries the overall prevalence of nosocomial infections in intensive care units (ICUs) in newborns varies from 8.4 to 26%, while developing countries, such as Brazil, have rates between 18.9 and 57.7%. The reasons for such high percentages are poor working conditions, insufficient numbers of health professionals and improper infrastructure (Pinheiro et al., 2009).

Simple habits, such as hand washing, use of personal protective items and compliance with aseptic procedures, as well as the strict control of the air quality, can avoid some opportunistic fungal infections, as these measures block the transmission of microorganisms.

The presence of hyaline filamentous and dematiaceous fungi in hospital environments must be considered, since they may be responsible for several infections in immunocompromised patients (Munoz et al., 2015). In the group of hyaline filamentous fungi, the genus *Aspergillus*, especially the species *Aspergillus fumigatus*, is one of the opportunistic agents most frequently cited in the literature, acting particularly in bone marrow transplant and neutropenic patients (Lang-Yona et al., 2016). Inhalation of fungal particles is the most common transmission route and aspergillosis outbreaks are commonly associated with remodeling and construction projects within and near hospitals (Diniz-Martins et al., 2005).

In this context, hospitals are environments that require close environmental monitoring, broken down by specific areas, to identify possible sources of contamination/dissemination and

* Corresponding author. Tel.: +55 85 98899 7347, +55 85 3366 9623.
E-mail addresses: pantojalydia@gmail.com (L.D.M. Pantoja), ronaldo@ufc.br (R.F. do Nascimento), abarbara@deha.ufc.br (A.B. de Araújo Nunes).

Peer review under responsibility of Turkish National Committee for Air Pollution Research and Control.

http://dx.doi.org/10.1016/j.apr.2016.02.011

etiologic agents (Diniz-Martins et al., 2005). According to Li and Kuo (1992), indoor bioaerosols are considered a major cause of respiratory problems, causing the absence of students at school and employees at work, or low productivity in hospitals and other occupational environments (Sousa et al., 2011; Pantoja et al., 2012).

The literature indicates that current methods to monitor air quality have a number of drawbacks, such as slow count of colony-forming units and results that often do not relate to the actual environmental situation (Bastos, 2005). Therefore, more accurate methods to characterize the fungal composition in the air are necessary. Recent protocols aiming to analyze microbial volatile organic compounds (MVOCs) in samples with possible fungal contamination are fast and highly sensitive.

VOCs are defined as organic compounds, found in gaseous or vapor state, that can be measured by analytical methods (Tucker, 2004). A portion of VOCs found in the domestic environment comes from outdoor air. However VOCs levels can be higher indoors than outdoor, due to the fact that internal sources may be prevalent, especially in new buildings where materials exhibit higher initial rates of emission, decreasing with time. Factors such as season, temperature and relative humidity are also likely to alter concentrations of VOCs in the air (Wang et al., 2007).

Detection of MVOCs or, specifically, of fungal volatile organic compounds (FVOCs) in samples air, is an indication that microbial growth is occurring. These compounds can cause headaches, nasal irritation, dizziness, fatigue and nausea in humans (Wålinder et al., 2008; Nurmatov et al., 2013). For example, Kim et al. (2007) found an association between respiratory symptoms and MVOC concentration inside Swedish public schools.

Polizzi et al. (2012) described a broader influence of fungal volatiles in buildings, for instance in relation to the sick-building-syndrome, where analogous volatile organic compounds play a peculiar role in jeopardizing health. Similar studies have been carried out focusing on different genera of fungi (e.g. *Trichoderma* sp., *Aspergillus* sp., *Fusarium* sp.), as discussed by Lancker et al. (2008), Polizzi et al. (2009, 2011).

Therefore, examination and characterization of typical fungal distribution in a particular environment can be useful to identify associations between domestic fungal sensitization, clinical diagnosis and prevention of seasonal allergic diseases (Pei-Chin et al., 2000). Moreover, such studies contribute to the analysis of ecological relationships in the environment. Some researchers are particularly interested in determining the presence of FVOCs as markers of the contamination of environments (Rosch et al., 2014). In addition, fungal microbiota varies with location and season. The variation of environmental characteristics in regions makes it important to conduct national and international systematic studies to check the dynamics of fungal microbiota.

In this context and highlighting the lack of findings in the national literature, this study aimed to analyze the air quality based on FVOCs, to enhance the monitoring and control of sectors in a tertiary referral hospital in Fortaleza, and also to improve the systematic study of aerobiology.

2. Material and methods

2.1. Research typology

This study is explanatory, of the experimental (Gil, 2007), quantitative and qualitative type, as well as exploratory, applying the hypothetico-deductive method.

2.2. Ethical aspects

The project was approved by the hospital ethics committee.

2.3. Selection of the hospital sectors

Samples were collected from sectors of one of the largest public hospitals in the state of Ceará, located in the capital city, Fortaleza.

Four specific sectors were chosen elected in the hospital, since according to Hess-Kosa (2002), the collection sites should be indicated in advance, and such sites should be framed in one or more categories: (1) place where the worst case of indoor air quality (IAQ) is noticed; (2) areas with greater representation in size and occupation; and (3) special concern sites. In addition to these aspects, two types of air microbiota were considered: in closed and open environments, and also the type of ventilation, whether artificial (air conditioning) or natural (Table 1). Humans were present in all sectors during the collections.

2.4. Sample collection

Samples were collected during the two seasons that predominate in the state of Ceará, the dry season (September, October and November 2014) and rainy season (April, May and June 2015). Rainfall data were provided by the Ceará Foundation for Meteorology and Water Resources (FUNCEME).

Air samples for FVOC analysis were collected in duplicate in each of the selected sectors, by air suction with the aid of an active sampling pump, adapted for a flow range of 80–100 mL/min (USEPA, 1999b), for 1 h, using specific cartridges (Brand 226-01 SKC-ANASORB CSC), as recommended by Harper (2000). The system was mounted at a height equivalent to the area of human breath and away from the walls (Schleibinger et al., 2008; Wålinder et al., 2008; Araki et al., 2009; Sousa et al., 2011).

The samples were analyzed by gas chromatography/mass spectrometry (GC/MS), performed within 6 h after the collection in order to minimize the risk of interference. Ten external standards were used for monitoring: seven alcohols (2-methyl-1-propanol, 2-methyl-1-butanol, 3-methyl-1-butanol, 3-octanol, l-octen-3-ol, 1-pentanol and 2-pentanol) and three ketones (2-hexanone, 2-heptanone and 3-octanone).

The analytic parameters in GC/MS were built from the analysis performance: a DB-5 ms column was used (non-polar, length 30 m, film thickness 0.5 mm, diameter 0.25 mm), with a temperature program from 35 °C (7 min), increase of 20 °C min^{-1}–75 °C, increase of 10 °C min^{-1}–125 °C (2 min). Helium (column flow 0.9 ml/min) was used as carrier gas. The FVOCs were identified by their mass spectra at an interface temperature of 250 °C, scanning range of 45 ± 300 m/z (USEPA, 1999a, 1999b) and according to the studies of Araki et al. (2009), Fiedler et al. (2001), Demyttenaere et al. (2004), Quadros (2008), Schleibinger et al. (2008) and Schuchardt and Kruse (2009).

For the analysis of fungi, the samples were collected using the passive sedimentation method in 150 mm diameter Petri dishes containing potato dextrose agar medium (Himedia®, India). The

Table 1
Selection criteria of the collection sites considering the categories suggested by Hess-Kosa (2002), the two types of air microbiota and the type of ventilation, at a public tertiary referral hospital in the city of Fortaleza, Ceará, Brazil.

Specific sectors	Type of ventilation	Air microbiota
Adult Intensive Care Unit[c] Transplant Ward[b,c] Emergency Reception[b] Elective Reception[a,b]	Artificial (air conditioning)	Closed

[a] Place where the worst case of indoor air quality (IAQ) was noted.
[b] Areas with greater representation in size and occupation.
[c] Special concern sites.