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Comparison of background levels of culturable fungal spore concentrations in indoor and outdoor air in southeastern Austria

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HIGHLIGHTS

- Fungal spore concentrations in uninfected rooms are comparable to those of outdoors.
- *Cladosporium* sp. had a strong outdoor presence.
- *Aspergillus* sp. and *Penicillium* sp. were typical indoor fungi.
- The 3 genera of fungi are important for the assessment of indoor and outdoor air.

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ABSTRACT

Background concentrations of airborne fungi are indispensable criteria for an assessment of fungal concentrations indoors and in the ambient air. The goal of this study was to define the natural background values of culturable fungal spore concentrations as reference values for the assessment of moldy buildings. The concentrations of culturable fungi were determined outdoors as well as indoors in 185 dwellings without visible mold, obvious moisture problems or musty odor. Samples were collected using the MAS-100® microbiological air sampler.

The study shows a characteristic seasonal influence on the background levels of *Cladosporium*, *Penicillium* and *Aspergillus*. *Cladosporium* sp. had a strong outdoor presence, whereas *Aspergillus* sp. and *Penicillium* sp. were typical indoor fungi. For the region of Styria, the median outdoor concentrations are between 100 and 940 cfu/m^3 for culturable xerophilic fungi in the course of the year. Indoors, median background levels are between 180 and 420 cfu/m^3 for xerophilic fungi. The I/O ratios of the airborne fungal spore concentrations were between 0.2 and 2.0. For the assessment of indoor and outdoor air samples the dominant genera *Cladosporium*, *Penicillium* and *Aspergillus* should receive special consideration.

1. Introduction

Background concentrations of airborne fungi are indispensable criteria for the assessment of fungal concentrations indoors and in the ambient air. The ambient air contains a large variety of fungal spores. Their concentration in the air mainly depends on vegetation, weather, season, time of day, traffic, waste treatment facilities, farms as well as the meteorological and climatic conditions (Reinthaler et al., 1999; Haas et al., 2005). Temperature and relative humidity (RH) influence the concentration of airborne fungal spores (Troutt and Levetin, 2001; Jones and Harrison, 2004; Kaarakainen et al., 2008; Nasir and Colbeck, 2010; Grinn-Gofroń, 2011).

Fungal spores can be transported directly by air movement and/or attached to aerosol particles. As early as 1975, Stix demonstrated a dissemination of spores over a distance of more than 1000 km (Stix, 1975). The differentiation of fungal spores in “dry air spora” and “wet weather spora” is generally accepted (Troutt and Levetin, 2001; Grinn-Gofroń, 2007). Various genera and species of fungi such as *Cladosporium*, *Alternaria*, *Penicillium*, *Aspergillus* and others,

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the so-called “dry air spora”, can be disseminated by air movements at low relative humidity and wind speed (Hirst and Stedman, 1963). At low RH and high wind speeds, these species of fungi increase in the ambient air on warm afternoons (Rich and Waggoner, 1962; Levetin, 1991). *Cladosporium*, which is part of the Dematiaceae family, is the most frequent species in the ambient air. They are also referred to as “summer air spores” because they occur in particularly high concentrations during the summer months (Reiss, 1998; Grinn-Gofroń and Rapiejko, 2009).

Hydrophilic spores, often referred to as “wet weather spora”, are disseminated passively via rain (http://pollen.utulsa.edu/Spores/AIRSPORA.html). During and after rainfalls, the ambient air, therefore, contains particularly high numbers of ascospores and basidiospores (Li and Kendrick, 1995b; Troutt and Levetin, 2001).

The fungal spore concentration varies seasonally. The background concentration of fungal spores in the air increases in spring, reaches high concentrations in the summer, and, in the case of many fungal species, attains a maximum in September (Levetin, 1995). In climate zones with snowy winters and cold temperatures, outdoor fungal spore concentrations are very low in winter (Ebner et al., 1989). In spite of low outdoor background concentrations during winter months, *Aspergillus* and *Penicillium* species are dominant indoors (Millington and Corden, 2005). According to Gregory and Hirst, 1957, there are high outdoor spore concentrations of *Cladosporium* sp. from June through October, additionally influenced by specific weather conditions, the phenology of the local vegetation and its associated fungal flora.

High quality outdoor air is a prerequisite for high quality unpolluted indoor air. An increase of background concentrations outdoors may lead to an increase in indoor spore concentration. Fungal spores enter the air of homes and buildings from the ambient air when windows or doors are opened, then attach to dust particles and/or settle.

Indoors, seasonal variations of most fungal spores are not as pronounced as outdoors (Koch et al., 2000), since indoor fungal spore concentrations also depend on other factors. In addition to various outdoor sources such as dust or organic waste, indoor spaces may contain materials which favor fungal growth releasing high spore concentrations into the air. Materials such as furniture, wallpaper, wood, foods or potting soil may create ideal conditions for fungi in combination with an increased substrate and air humidity. Moreover, modes of ventilation, air temperature and air humidity regulate indoor spore concentrations. Indoor human activities disperse spores in the air (Su et al., 1992; Lee et al., 2006a,b; Docampo et al., 2010; Haas et al., 2010). In the winter months, spore concentrations indoors may reach higher levels than outdoors which suggests a contamination by fungi indoors (Li and Kendrick, 1995a). In case of contamination, the affected surfaces are colonized by those fungal species which generally indicate moisture damage (Meklin et al., 2002). Various species of airborne culturable fungi also clearly indicate sources of mold. Total indoor spore concentrations of *Penicillium* and *Aspergillus* are directly related to the extent of mold in living spaces, whereas *Cladosporium* proved to be a characteristic outdoor fungus (Pastuszka et al., 2000; Haas et al., 2007).

In recent years, the health hazard of fungi has received increasing public interest. Measurements of indoor culturable fungi have traditionally led to the detection of hidden mold for an assessment of potential risk to inhabitants. Numerous studies and assessments of indoor and outdoor air have been conducted using different measuring methods, mainly by impaction, to determine airborne culturable fungal spore concentrations. The assessment of the indoor concentrations of airborne, viable spores, however, is difficult because of lacking standards and limits.

1.1. Background

A series of investigations concerning culturable fungal spore concentrations in indoor and outdoor air were conducted in the Austrian State of Styria. Styria is located in the southeastern part of Austria and has a continental climate. Its capital city, Graz, is located southeast of the Alps, lies in a basin, and is influenced by the Mediterranean climate. The town has little wind, around 120,000 cars drive into the city on weekdays, and there is severe fine dust in winter, all of which influence the air quality.

Prior to this study, indoor air quality investigations were performed quantitatively in a large number of homes by using the six-stage Andersen cascade impactor (ACFM©). Additionally, a study was carried out with the goal of comparing the culturable fungal spore concentrations quantitatively and qualitatively in the air of apartments with and without visible mold growth by using MAS-100® impactor (Haas et al., 2007). On the basis of these findings, the question was raised whether the background of fungal spore concentrations in houses without visible mold growth are comparable to the concentrations outdoors which could then be used as reference values for homes with obvious mold problems. Furthermore, a comparison between the most frequent fungal genera indoors and outdoors is needed.

1.2. Goal of the study

The goal of the present study was to define the natural background values of culturable fungal spore concentrations as a reference for measurements in moldy buildings. The concentrations of culturable fungi were determined in outdoor as well as in indoor air in different homes without visible mold, obvious moisture problems or musty odor. The airborne, culturable fungal spores were gathered by impaction and assessed both qualitatively and quantitatively. The focus was on 3 fungal genera, namely: *Cladosporium*, *Aspergillus* and *Penicillium*. The season, air temperature and RH were evaluated.

2. Material and methods

Between 2011 and 2013, a total of 740 samples from 185 rooms of homes without visible mold problems and the ambient air in Styria/Austria, were investigated with regard to the total fungal spore concentrations and their seasonal distribution. On-site inspection ensured that rooms were free from dampness, moldy patches, musty odor and mechanical ventilation systems. Homes of healthy residents, who were willing to participate in this study, were selected randomly. In order to ensure standardized measurements, residents were asked not to ventilate the room for 6 h prior to sampling. Air samples were collected in the room where they spend most of their time and from the corresponding outdoor air. During the measuring time in the morning, the inhabitants did not engage in activities such as showering, cooking or sports. The air sampling outdoors was performed only during dry weather conditions and low wind speeds of 0–2 m/s according to VDI 4251.

Measurements were not conducted in the vicinity of relevant sources of emissions such as organic waste containers or composting locations. Data relating to indoor and outdoor climate such as temperature and RH (testo® 445, GmbH, Wien) were recorded. The calendar months were grouped into four periods, namely spring (March, April, May), summer (June, July, August), fall (September, October, November) and winter (December, January, February).

Samples were collected using the Merck MAS-100® microbiological air sampler with a volume of 100 L/min (Meier and Zingre, 2000) and a cut off size of 1.7 μm (Yao and Mainelis, 2006). The