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Health hazards related to conidia of *Cladosporium*—biological air pollutants in Poland, central Europe

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ABSTRACT

The spores of Cladosporium Link. are often present in the air in high quantities and produce many allergenic proteins, which may lead to asthma. An aerobiological spore monitoring program can inform patients about the current spore concentration in air and help their physicians determine the spore dose that is harmful for a given individual. This makes it possible to develop optimized responses and propose personalized therapy for a particular sensitive patient. The aim of this study was to assess the extent of the human health hazard posed by the fungal genus Cladosporium. For the first time, we have determined the number of days on which air samples in Poland exceeded the concentrations linked to allergic responses of sensitive patients, according to thresholds established by three different groups $(2800/3000/4000 \text{ spores per 1 } \text{m}^3 \text{ of the air})$. The survey was conducted over three consecutive growing seasons (April-September, 2010-2012) in three cities located in different climate zones of Poland (Poznan, Lublin and Rzeszow). The average number of days exceeding 2800 spores per cubic meter (the lowest threshold) ranged from 61 (2010) through 76 (2011) to 93 (2012), though there was significant variation between cities. In each year the highest concentration of spores in the air was detected in either Poznan or Lublin, both located on large plains with intensive agriculture. We have proposed that an effective, science-based software platform to support policy-making on air quality should incorporate biological air pollutant data, such as allergenic fungal spores and pollen grains.

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Introduction

Fungi of the genus Cladosporium Link. occur in different climatic zones as cosmopolitan, ubiquitous organisms. Genus Cladosporium, composed of numerous species, belongs to the family Davidiellaceae, order Capnodiales, class Dothideomycetes, and phylum Ascomycota (Index Fungorum). Dugan et al. (2004) reported 772 species names, and the current list of Index Fungorum (November 2016) contains 779 names, including 651 species and 128 varieties. They are secondary parasites and saprotrophs, widespread through the growing season. The fungi can inhabit living plant tissue such as stems, leaves, flowers and fruit (Ellis and Ellis, 1985; Marcinkowska, 2003; Horst, 2013; Lee and Liao, 2014), as well as dead stubble, hay and different composts (Lacey and Dutkiewicz, 1994). The fungi are commonly found on a variety of other substrates, such as rubber, insulation materials, stones, concrete, bricks and mortar, as detailed in the review by Gutarowska (2014). Moreover, Cladosporium readily grows on paper, and greatly contributes to the biodegradation of museum and library collections (Zielinska-Jankiewicz et al., 2008; Skóra et al., 2015). The substrates for fungal development include wood, textiles and wallpaper, so it is also responsible for the deterioration of the interiors of residential buildings (Andersen et al., 2011; Mandal and Brand, 2011). The mycelium of Cladosporium grows in refrigerators and on moist window frames (Barnes et al., 2001).

The spores of Cladosporium contribute to and often dominate in atmospheric bioaerosols (Elbert et al., 2007). According to Fröhlich-Nowoisky et al. (2009), concentrations of conidia of Cladosporium in the atmosphere over cultivated regions usually range between 1 and 10,000 spores per cubic meter of air. The spores of Cladosporium are biological air pollutants that pose a threat to human health. They irritate the respiratory tracts and contain numerous proteins that cause inhalant allergies in people (Resano et al., 1998; Kurup et al., 2002; Thompson et al., 2000; Rapiejko et al., 2004). Due to their small size (3–35 μ m) the spores of Cladosporium can easily reach the lower respiratory tract, including the lungs (D'Amato and Spieksma, 1995; Reponen et al., 2001; Lee and Liao, 2014). The symptoms of inhalant allergies include allergic rhinitis, sinusitis and conjunctivitis, mold asthma and allergic alveolitis (Knutsen et al., 2012). It seems likely that the history of human exposure to these allergens will date back tens of thousands of years, since the spores of Cladosporium are abundant in the air inside of caves (Docampo et al., 2011). In Poland, the highest intensities of Cladosporium-associated allergic symptoms were recorded in summer and autumn (Lipiec et al., 2007). In museums and art conservatories, personnel are exposed to these fungi all year round; allergy to fungal spores, mainly consisting of Cladosporium, was observed in up to 85% of staff (Wiszniewska et al., 2010). The two most common species present in biological aerosol particles are Cladosporium cladosporioides and Cladosporium herbarum (Després et al., 2012).

Fungal allergens are proteins and glycoproteins with molecular weights usually between 6 and 90 kDa (Burge and Rogers, 2000). Thompson et al. (2000) documented two allergens of *C. cladosporioides* and nine allergens of *C. herbarum*. Additionally, the Allergome web site (www.allergome.org) lists six other *Cladosporium* allergens. The Allergome database contains data on the allergenicity of allergens in their natural conformations and also for allergens obtained by means of molecular biology techniques, if available. There is also one in silico predicted protein from *Cladosporium malorum* Alt mr 1 (137aa) classified as an Alt a 1-related allergen. The total number of proteins identified as allergens in *Cladosporium* fully or partially characterized and listed in different international scientific journals and web databases is close to twenty, of which thirteen have been cloned.

While species of Cladosporium are capable of producing multiple allergens, it should be noted that not all may be expressed in conidia. According to Aukrust (1979), out of ten allergens found in the genus Cladosporium, eight were detected in *C. herbarum*, but only one of these ten allergens was detected in fungal conidia (Cla h HCh-1); the remaining ones were found in mycelium. Dixit and Kwilinski (2000) reported allergenic cross-reactivity between *C. cladosporioides*, *C. herbarum* and *Cladosporium sphaerospermum*.

Volumetric traps frequently record *C. herbarum*-type spores, which occur mainly in temperate and cold climate zones (Hjelmroos, 1993). Concentrations and annual total sums of these spores depend on geographic area, the location of measuring devices and timing of the growing season (Grinn-Gofron et al., 2011; Aira et al., 2012). Thus, aerobiological monitoring may be a valuable tool to patients and physicians that helps to identify a harmful dose of allergenic spores.

Various authors have established different threshold values for the Cladosporium spore concentrations causing allergy symptoms in average sensitive patients. The threshold value to evoke the first allergy symptoms in humans is 80 to 100 spores per 1 m³ of air, as reported by Rapiejko et al. (2004) and Gravesen (1979), respectively. The spores of Cladosporium are small (4–25 \times 3–7 μ m), so high concentrations were postulated to induce allergenic symptoms. For severe asthma, Rantio-Lehtimaki et al. (1991) proposed the threshold of 4000 spores per 1 m³ of air, D'Amato and Spieksma (1995) reported 3000 spores and Rapiejko et al. (2004) cited a figure of 2800 spores per 1 m³. Rantio-Lehtimaki et al. (1991) based the threshold on the relation of the size of Cladosporium spores to the size of birch pollen grains (0.03). D'Amato and Spieksma (1995) established the threshold using a combination of the results of aerobiological studies, positive diagnostic tests and careful recording of symptoms elicited by the allergens produced by Cladosporium. The development of the threshold by Rapiejko et al. (2004) was based on studies conducted in the years 2002-2003, concerning the intensity of allergic rhinitis and bronchial c asthma symptoms in 600 patients living and working in two districts of Warsaw, Poland. Patients were sensitive to the allergens of some pollen grains, as well as to the spores of Cladosporium. The studies took into account aerobiological measurements, patient self-observation sheets and medical examination, including endoscopy of nasal cavities assessing color, edema of the nasal turbinates and the presence and intensity of nasal congestion, as well as conjunctival symptoms.

The aim of this study was to assess the extent of the human health hazard posed by the fungal genus *Cladosporium* by determining the number of days on which the spore concentration threshold for allergic responses was exceeded.

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