

Monitoring of anamorphic fungal spores in Madeira region (Portugal), 2003–2008

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Abstract Seven years of aeromycological study was performed in the city of Funchal with the purpose to determine the anamorphic spore content of this region and its relationship to meteorological factors. The sampling was carried out with a Hirst-type volumetric spore trap following well-established guidelines. A total of 17,586 anamorphic fungal spores were recorded during the studied period, attaining an annual average concentration of 2931 spores m⁻³. Anamorphic fungal spores were observed throughout the year, although the major peaks were registered during spring (April–June) and autumn period (September–November). The lowest spore levels were recorded between December and February months. Over 14 taxa of anamorphic fungal spores were observed with *Cladosporium* being the most prevalent fungal type

accounting for 78 % of the total conidiospores. The next in importance was *Alternaria* (5.4 %), *Fusarium* (4.7 %), *Torula* (3.9 %) and *Botrytis* (1.9 %). Temperature was the meteorological parameter that favoured the most release and dispersal of the conidiospores, whereas rainfall revealed a negative effect. Despite the low concentration levels found in our region, the majority of the fungal types identified are described as potential aeroallergens. This study provides the seasonal variation of the conidiospores and the periods when the highest counts may be expected, representing a preventive tool in the allergic sensitization of the population.

Keywords Aerobiology · Anamorphic fungi · Meteorological conditions · Seasonal variation · Spore calendar

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1 Introduction

Fungal spores are microscopic organisms that possess a great capacity to colonize several kinds of substrates, being ubiquitous in nature. Fungi live as saprophytes on organic material or as parasites (mainly plant pathogens), so the majority of fungal spores in the air outdoors come from farms, forest stands and decomposing plant matter and represent an important fraction of bioaerosol (Sabariego et al. 2007; Huffman et al. 2012; Bowers et al. 2013). Measurements of fungal aerosols from Tulsa, Oklahoma, report average ground-level

concentrations of around 10,000–50,000 spores m^{-3} , sometimes exceeding even 200,000 spores m^{-3} (Levetin 1995). Simulations conducted by Heald and Spracklen (2009) came to the conclusion that 23 % of all primary emissions of organic aerosol are of fungal origin. The release of fungal spores and consequently their concentrations in the atmosphere depends on biological, local and environmental factors such as: life cycle, geographic location, air pollution, weather conditions, human activity and local source of vegetation. Their airborne occurrence is markedly seasonal and is highly dependent on temperature and relative humidity and can be dramatically changed by weather events (Grinn-Gofroń and Strzelczak 2012). In general, moulds are able to live in warm temperatures and are resistant to cold conditions (Krouse et al. 2002).

Some fungi and their spores have a negative effect on human health and are a frequent cause of allergies and immunotoxic diseases (Ataygul et al. 2007). The presence of these aeroallergens in a particular locality can be correlated with clinical symptoms of respiratory illness (Tripathi et al. 2004). The number of inhaled particles is directly related to their concentration in the air and also with the daily activity of the exposed individuals. The type and severity of the disease depend on the time and dose of exposure, individual susceptibility, the enzyme content and antigenic capacity of each fungus (Platt-Mills and Solomon 1998).

Previous aerobiological studies in various locations in Portugal, including Porto, Coimbra, Lisboa and Évora, have shown the predominance of anamorphic fungi in the air, followed by spores of basidiomycetes and ascomycetes (Nunes et al. 2008; Oliveira et al. 2009, 2010). The results published in this work are the first such detailed analysis of most types of fungal spores in the air of Madeira. Considering the utility of these studies in the diagnosis and treatment process of allergic illness, the aeromycological surveys have been carried out in several countries and regions: Spain (Herrero et al. 1996; Elvira-Rendueles et al. 2013), USA (Burch and Levetin 2002), Greece (Gioulekas et al. 2004), Turkey (Ataygul et al. 2007), NW Iberian Peninsula (Aira et al. 2008), Poland (Grinn-Gofroń and Mika 2008; Kasprzyk et al. 2013), Italy (Rizzi-Longo et al. 2009), Jordan (Abu-Dieyeh et al. 2010), England (Sadyś et al. 2015).

The aim of the study is to determine the anamorphic spore content of this region and its relationship to

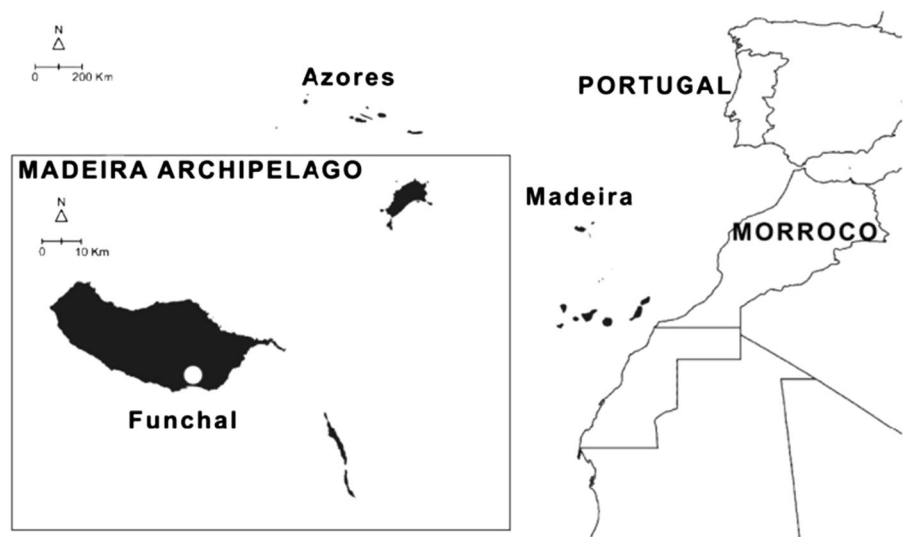
meteorological factors in order to identify the potential aeroallergens that might affect the local population and visitors as well. According to Gams and Seifert (2008), anamorphic fungi are defined as an artificial assemblage of asexual stages of ascomycetes and basidiomycetes. They are classified together with asexually reproducing fungi that lack sexual structures.

2 Materials and methods

The survey of fungal spore concentration took place in Funchal, the capital of Madeira Island, one of the most important touristic regions of Portugal, between January 2003 and December 2008. Madeira is a volcanic island with a surface area of 739 km^2 located 900 km south-west of Portugal and 700 km west of Morocco (Fig. 1) in the Atlantic Ocean.

Air sampling was performed on the roof of the Dr. João de Almada Hospital, about 10 m above ground level. The hospital is located in an urban area about 3 km from the centre of Funchal city. Funchal is located at coordinates 32°39'N, 16°55'W, on the south coast of Madeira at the base of an amphitheatre up to 1800 m tall. Funchal is the most populous insular city in Portugal, in an island with a population of approximately 240,000 (Oliveira and Pereira 2008). Bioclimatically, Madeira Island presents a temperate hyperoceanic submediterranean bioclimate, with a mediterranean pluviseasonal oceanic bioclimate on the south coast, with Funchal in particular lying within the thermosubmediterranean and thermomediterranean thermoclimatic belts (Rivas-Martínez 2001). In Madeira Island, the temperatures are mild all year round (average temperature of 18.7°), varying between 15.9° in February and 22.3° in August, the relative humidity ranges between 55 and 75 %, and rainfall ranges between 500 and 1000 mm yearly (Quintal 2007). The prevailing wind direction is from the south-west during the winter and from north in the summer (Santos et al. 2004). About two-thirds of Madeira Archipelago is a nature reserve in which the laurel forest is found. It contains a wide array of plant species and the most common trees belong to the Lauraceae family, like the Canary Laurel (*Apollonias barbuiana*), Laurel (*Laurus novocanariensis*), Fetid Laurel (*Ocotea foetens*) and Madeira Mahogany (*Persea indica*), besides trees from other families like

Fig. 1 Location of Madeira Archipelago and the city of Funchal



the Lily of the valley tree (*Clethra arborea*) and Wax Myrtle (*Myrica faia*). It also includes abundant endemic bushes, besides many bryophytes, lichens, mosses and liverwort species (Borges et al. 2008).

Funchal is often considered a Garden City due to the diversity of green spaces and floristic richness contained therein. The city offers 17 public green spaces in which exotic plants from around the world thrive, and the most common trees in the city are *Tipuana tipu*, *Jacaranda mimosifolia*, *Agathis robusta*, *Spathodea campanulata*, *Casuarina equisetifolia*, *Melia azedarach*, *Erythrina* spp., *Brachychiton* spp., besides several other species from families like Arecaceae, Cupressaceae and Cyatheaceae. In urban areas, there are many patches of land used for agricultural purposes, mostly cultivated with *Musa acuminata*, *Vitis vinifera* and with horticultural production for domestic consumption. Uncultivated land presents a variety of grasses dominated by *Phleum* spp., *Arundo donax* and *Dactylis* spp. The outskirts of the city are dominated by exotic forest from the genera *Acacia*, *Eucalyptus* and *Pinus*. The fertile soils and mild subtropical climate enable the coexistence of a noticeable variety of tropical and subtropical plants, representing a suitable substrate for fungal growth and reproduction (Quintal 2007; Borges et al. 2008).

Airborne fungal spores were collected by Hirst-type volumetric trap (Burkard Manufacturing Co. Ltd., UK) (Hirst 1952) designed to perform analyses according to the European Aerobiology Network recommendations (Galán et al. 2014). The equipment

draws air at a rate of 10 L/min, impacting particles to a drum containing an adhesive tape covered with silicon solution. The drum was changed regularly every week. After sampling, tape was cut into segments corresponding to 24-h sampling (48 mm each) and mounted in a mixture of glycerine with fuchsine. The Hirst trap has stopped during January and February of 2006 due to a mechanical breakdown. The identification and counting of fungal spore were conducted on 4 longitudinal transects along the slides under a light microscope (Olympus BX50) at magnification 400 \times . The spore categories include the following types of: *Cladosporium*, *Alternaria*, *Curvularia*, *Nigrospora*, *Tetraploa*, *Drechslera* type, *Torula*, *Botrytis*, *Fusarium*, *Gliomastix*, *Arthrinium*, *Epicoccum*, *Polithrinium* and *Spegazzinia*. Fungal spores that did not fit into the above categories were denominated as “other spores”, which include partial or unidentifiable fungal spores. The fungal spore counts were converted into atmospheric concentrations and expressed as fungal spores per cubic metre of air. In order to verify the accuracy of calculations performed under the microscope, most of the samples have been reviewed using a microscopic camera connected to a computer screen.

Due to nonlinearity and non-normality of the analysed variables, Spearman’s rank correlation was applied in order to examine the studied relationships.

The calculations were made based on the daily average concentration of the airborne particles and daily weather data. Statistical analysis was carried out using a SPSS 17.0 programme.

Meteorological data for air temperature (mean, max and min), relative humidity and rainfall were provided by the Institute of Ocean and Atmosphere (IPMA)—Regional Station in Funchal—located approximately 5 km south-east of the sampling site.

3 Results

A total of 17,586 anamorphic fungal spores were recorded during the studied period, attaining an annual average concentration of 2931 spores m^{-3} . This class made up 58 % of the total fungal spores detected in the atmosphere of Funchal during 2003–2008. The year with the highest fungal spore concentration was 2003, with 4427 spores m^{-3} , while in 2004, the lowest concentration was recorded, 1935 spores m^{-3} (Table 1).

Anamorphic fungal spores were observed throughout the year, with the highest levels occurring during the spring (April–June) and autumn (September–November). The lowest fungal spore levels were recorded between December and February months (Fig. 2).

The highest daily values were 218 fungal spores m^{-3} , recorded on the 20 September 2008, on the 26 April 2003, with 204 fungal spores m^{-3} and on the 23 April 2008 with 185 fungal spores m^{-3} . The general pattern with maximum values detected during spring season was shown by *Alternaria*, *Botrytis*, *Cladosporium*, *Drechslera* and *Torula* types, whereas

Epicoccum, *Fusarium* and *Nigrospora* showed maximum concentrations in the autumn season (Fig. 3).

The total number of taxa belonging to anamorphic spores observed during the study period was 14, with *Cladosporium* being the most prevalent fungal type accounting for 78 % of the total conidiospores. The next in importance was *Alternaria* (5.4 %), *Fusarium* (4.7 %), *Torula* (3.9 %) and *Botrytis* (1.9 %) (Table 2).

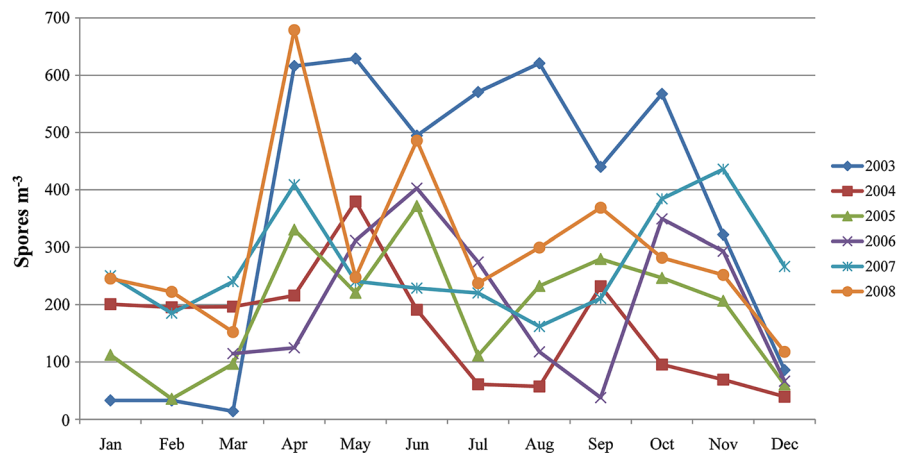
The prevailing weather conditions recorded during the study period revealed an average temperature of 19.6°, where the minimum and maximum temperatures ranged between 17.3° and 23.3°, respectively. The relative humidity recorded an average value of 62 %, and precipitation reached 1.6 mm on average (Fig. 4). The predominant wind quadrant observed in Funchal city was south-west.

The results from correlation analysis between anamorphic spore concentrations and meteorological parameters are shown in Table 3. *Alternaria*, *Curvularia*, *Nigrospora* and *Tetraploa* presented a significant positive correlation with air temperature. *Cladosporium*, *Drechslera* type and *Torula* presented also positive correlation with air temperature, but negative correlations with rainfall. On the other hand, a negative correlation with air temperature appeared in *Botrytis*, *Fusarium* and *Gliomastix* types. Finally, *Arthrinium*, *Epicoccum*, *Polithrinium* and *Spegazzinia* spores did not present significant correlation coefficients with any of the meteorological parameters analysed.

Table 1 Annual and monthly totals of the daily concentrations of anamorphic spore types in Funchal (highest monthly concentrations in bold)

	2003	2004	2005	2006	2007	2008	Total
Jan	32.9	200.9	112.3	–	250.6	245.6	842.3
Feb	32.9	195.5	35.6	–	185.2	222.5	671.8
Mar	14.0	196.6	96.7	114.7	240.3	152.3	814.5
Apr	616.1	216.0	330.5	124.7	408.8	678.8	2374.9
May	629.1	379.6	220.9	311.6	240.8	247.9	2029.9
Jun	494.6	191.2	371.9	402.8	229.0	486.0	2175.5
Jul	570.8	61.0	111.2	274.3	220.5	237.6	1475.4
Aug	621.0	57.2	232.7	117.7	162.0	299.7	1490.3
Sep	440.1	232.2	279.7	37.8	211.1	368.8	1569.8
Oct	567.5	95.6	246.8	349.4	384.5	281.9	1925.7
Nov	321.8	69.1	206.8	293.3	435.9	252.0	1579.0
Dec	85.9	39.4	60.5	66.4	266.7	117.7	636.6
Average	4426.9	1934.3	2305.6	2092.8	3235.3	3590.7	17,585.6

Fig. 2 Annual and monthly variation of anamorphic fungal spores concentrations in the atmosphere of Funchal city (2003–2008)

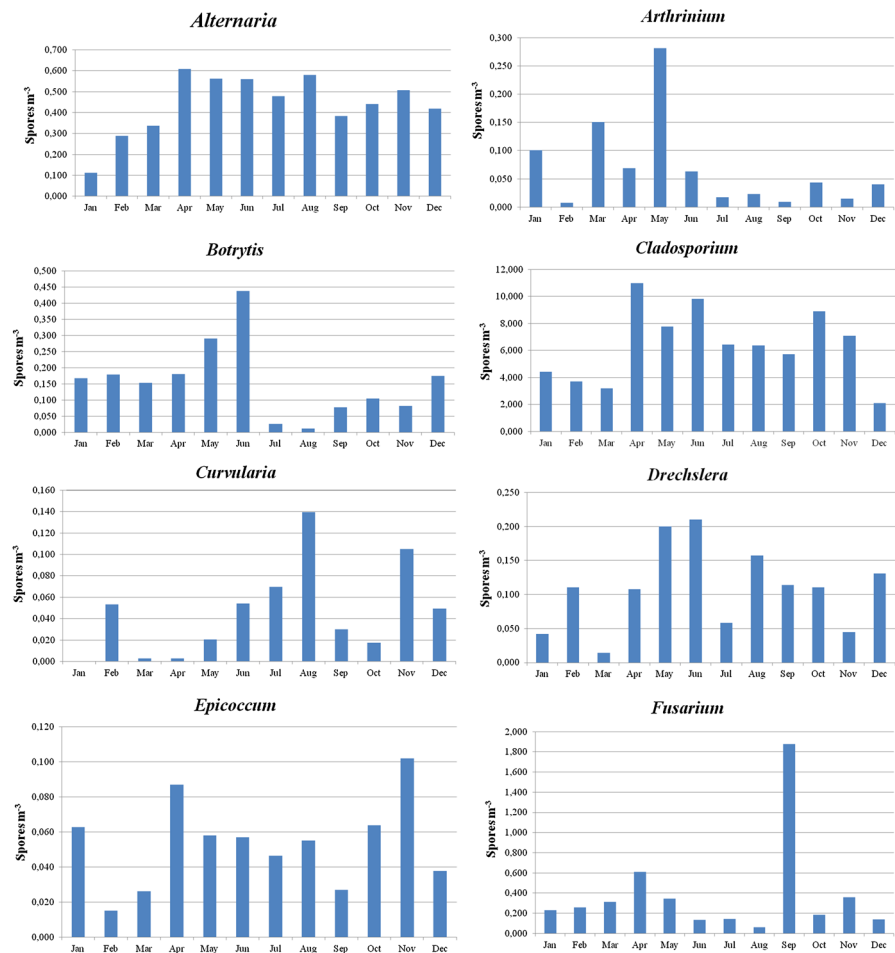


4 Discussion

This study reveals the anamorphic fungi spectrum of the city of Funchal and determines the meteorological factors that influence their occurrence in the atmosphere. Examined fungal spore types were observed throughout the year with an annual average concentration of 2931 fungal spores m^{-3} , while in other European cities, these fungal spores were also detected along the year but in higher levels. From the total of 17,586 fungal spores m^{-3} observed during the study period, most of the counts corresponded to *Cladosporium* (13,715 spores m^{-3}) and *Alternaria* (954 spores m^{-3}) (Tables 1, 2). In the Iberian Peninsula, the annual representation of *Cladosporium* and *Alternaria* alone varies between 70,000 and more than 200,000 fungal spores/year (Aira et al. 2008; Oliveira et al. 2010), whereas in Poland, the annual totals of *Alternaria* conidia varied between 14,000 and 28,000 (Grinn-Gofroń and Rapiejko 2009). In Thessaloniki (Greece), the conidiospore annual average reached 173,784 fungal spores (Gioulekas et al. 2004) and, in Madrid, the monthly concentrations were always above 3000 fungal spores during every month of the year (Sabariego et al. 2007). The lower fungal spore levels detected in Funchal may be explained by the coastal proximity of the city and its insular condition. It has been reported that airborne particle concentrations tend to be lower in coastal stations than in the mainland due to the proximity of the sea (Belmonte et al. 2008), which may be explained by the fact that

there is less land mass for growth and development of fungi and the sea also prevents resuspension of particles. In a survey carried out by Aira et al. (2008) in the north-western Iberian Peninsula, it was shown that aerobiological particles are more abundant inland than in coastal areas due to the presence of larger extensions of substrate available for fungal development and higher temperatures during the main sporulation period. Oliveira et al. (2010) also observed lower fungal spore counts in Porto, an urban and littoral station, comparatively to inland and rural stations, and the phenomenon was explained by the proximity of the River Douro and the Atlantic Ocean, water mass which are known to negatively affect airborne biological particles. The main island has a climate influenced by the subtropical anticyclone of the Azores, with a temperate hyperoceanic sub-mediterranean bioclimate influence (Rivas-Martínez 2001). The low level of fungal spores found in Funchal's stations is partially explained by the prevailing winds from the sea (south-west in Funchal during most of the year). The predominant wind direction varies from south-west quadrant during the winter to north quadrant during the summer (Santos et al. 2004). Data analysis revealed that fungal spore counts in Funchal are mostly influenced by air temperature and rainfall rather than wind direction and humidity (Table 3). Despite this fact, low fungal spore counts occurred in the winter, when the prevailing south-west wind carries clean air from the sea onto Funchal, whereas higher fungal spore counts

Fig. 3 Daily average concentrations of anamorphic spores by month for the 7-year period



were registered in the summer, when the prevailing north wind can carry fungal spores from the forest onto the city.

The anamorphic fungi represent the most important fraction of the air spore content in the atmosphere (Herrero et al. 1996; Sabariego et al. 2007; Ianovici 2011; Elvira-Rendueles et al. 2013), and this fact also occurs in Funchal. This group of fungi contains many known aeroallergens. Fungal spores of *Alternaria* may also adversely affect human health. It has been estimated that 12–42 % of atopic people are mould sensitive and about 70 % of patients respond to the

presence of *Alternaria* spores in air samples (Sanchez and Bush 2001; Knutsen et al. 2012). Approximately 3 % of the Portuguese population suffered from allergy to either *Alternaria* or *Cladosporium*, while in Spain, positive skin tests to both fungi were reported for 20 % of the population (Licorish et al. 1985; D'Amato et al. 1997). Allergic reaction severity to *Alternaria* depends on airborne spore concentrations. In the UK, Frankland and Davies (1965) reported that 50 spores of *Alternaria* m⁻³ of air were enough to trigger symptoms in sensitized individuals, while symptoms are precipitated by *Cladosporium* when

Fig. 3 continued

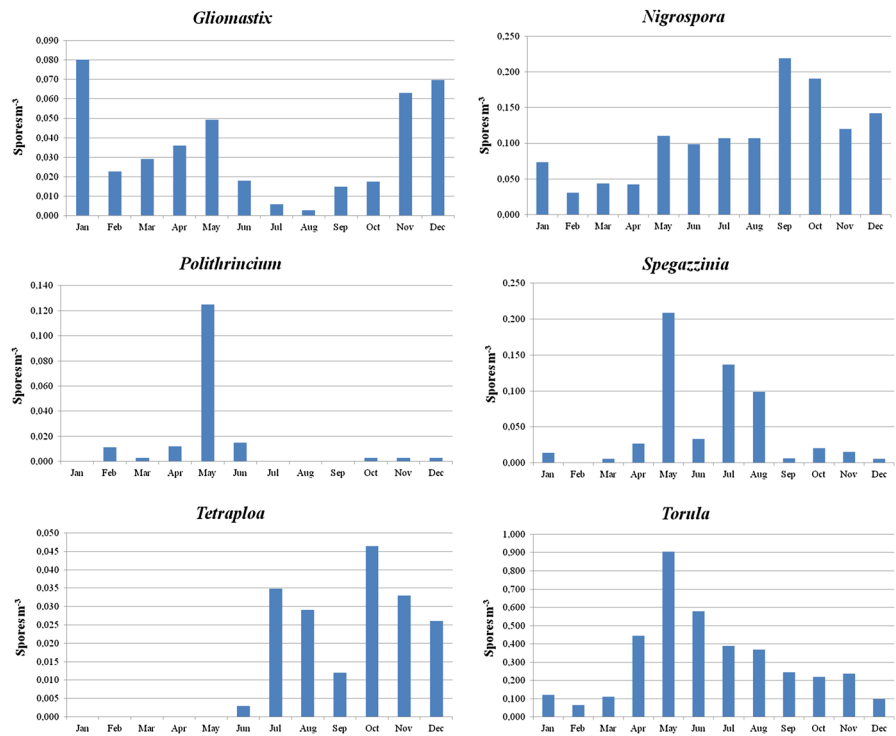


Table 2 Prevalent fungal spores recorded in the atmosphere of Funchal (2003–2008)

Fungal spore type	Spores m ⁻³	%	Maximum daily count (spores m ⁻³)
<i>Cladosporium</i>	13,714.73	78.0	126.9
<i>Alternaria</i>	953.86	5.4	21.14
<i>Fusarium</i>	828.28	4.7	216.54
<i>Torula</i>	687.42	3.9	7.02
<i>Botrytis</i>	332.62	1.9	13.5
<i>Drechslera</i>	232.74	1.3	4.32
<i>Nigrospora</i>	232.2	1.3	5.94
<i>Arthrinium</i>	148.5	0.8	7.02
<i>Epicoccum</i>	114.48	0.7	13.5
<i>Spegazzinia</i>	105.3	0.6	1.62
<i>Curvularia</i>	97.74	0.6	15.66
<i>Gliomastix</i>	71.82	0.4	3.24
<i>Tetraploa</i>	34.02	0.2	4.96
<i>Polithrinium</i>	31.86	0.2	0.54
Total of anamorphic spores	17,585.57	100	216.54

the mean daily concentrations rise to 3000 or more spores m⁻³. In Zagreb (Croatia), Peternel et al. (2004) reported that throughout the day, when *Alternaria* and

Cladosporium spore concentrations rose (between 10 and 12 a.m.), asthma attacks were noted. In north-west Spain, high fungal spore counts were recorded in late

Fig. 4 Prevailing weather conditions recorded during the study period in Funchal city: minimum, average and maximum temperature, relative humidity and rainfall

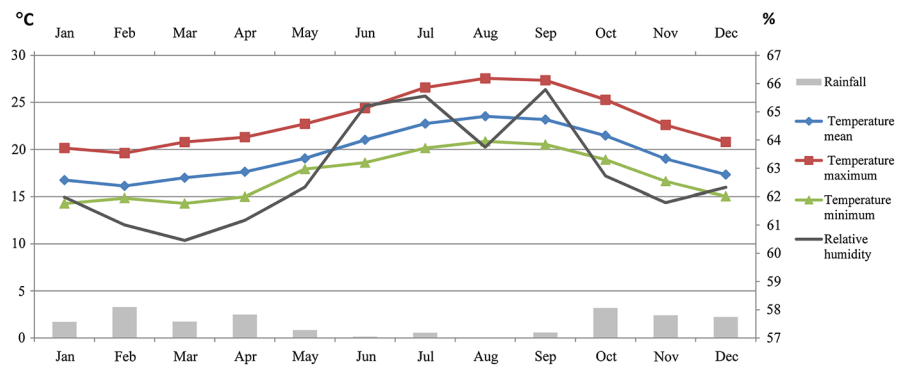


Table 3 Spearman's rank correlation coefficients between anamorphic spore concentration and weather parameters

	Temperature mean	Temperature maximum	Temperature minimum	Relative humidity	Rainfall
<i>Alternaria</i>	0.078**	0.073**	0.074**	0.041	0.005
<i>Arthrinium</i>	-0.054*	-0.032	-0.076**	-0.026	-0.036
<i>Botrytis</i>	-0.118**	-0.108**	-0.133**	-0.042*	-0.013
<i>Cladosporium</i>	0.091**	0.087**	0.076**	0.037	-0.063**
<i>Curvularia</i>	0.085**	0.082**	0.089**	-0.023	-0.026
<i>Drechslera</i>	0.077**	0.082**	0.076**	0.031	-0.058**
<i>Epicoccum</i>	0.009	0.006	-0.006	0.010	-0.051*
<i>Fusarium</i>	-0.087**	-0.082**	-0.096**	0.007	-0.008
<i>Gliomastix</i>	-0.109**	-0.111**	-0.093**	-0.018	0.054*
<i>Nigrospora</i>	0.086**	0.087**	0.090**	-0.029	-0.019
<i>Polthrincium</i>	-0.029	-0.038	-0.002	0.008	-0.035
<i>Spegazzinia</i>	0.016	0.013	0.013	0.020	-0.009
<i>Tetraploa</i>	0.059**	0.058**	0.061**	0.055*	0.019
<i>Torula</i>	0.091**	0.097**	0.077**	0.012	-0.091**
Total of anamorphic spores	0.082**	0.079**	0.063**	0.023	-0.062**

* $p < 0.05$; ** $p < 0.01$

summer and early autumn, with a fairly similar hourly spore-count pattern to Zagreb, with high concentrations at late evening (7–10 p.m.) (Rodriguez-Rajo et al. 2005). According to Rapiejko et al. (2004), the threshold concentration in Poland amounts to 80 spores m^{-3} of air, whereas Gravesen (1979) published a threshold value of 100. *Cladosporium* has relatively small spores, and so airborne concentrations must reach high levels in order to induce allergenic symptoms (Brown and Jackson 1978). Gravesen (1979) has estimated an allergenic airborne concentration threshold of 3000 spores m^{-3} for

Cladosporium. More recently, a figure of 4000 spores m^{-3} has been cited (Anon 2002). In Poland, Rapiejko et al. (2004) reported that subjects with hypersensitivity to *Cladosporium* allergens experienced the symptoms during exposure to a concentration of approximately 2800 spores m^{-3} of air.

Regarding seasonal dynamics, the highest fungal spore concentration and diversity in Funchal were detected during late spring and autumn seasons (Fig. 2), similar to the works of Jothish and Nayar (2004) and Halwagy (1994). In Porto city (mainland Portugal), Oliveira et al. (2010) reported the highest

airborne fungal spore concentrations during summer and late autumn, and a typical pattern also observed in the Mediterranean region (Sabariego et al. 2007; Aira et al. 2008). The fact that lowest fungal spore levels occurred during winter months (December and February) is in line with the study of Sabariego et al. (2007) performed in Madrid (Spain) and Ataygul et al. (2007) in Bursa (Turkey). In other European cities, peak concentrations are reached during summer and are characterized by the prevalence of *Cladosporium* and *Alternaria* from April to September–October (Herrero et al. 1996; Gioulekas et al. 2004; Stepalska and Wołek 2005; Sabariego et al. 2007; Ianovici 2011; Kasprzyk et al. 2013). In Funchal, just like in several other aeromycological surveys, *Cladosporium* also represents the most abundant fungal type of airspora (Ataygul et al. 2007; Abu-Dieyeh et al. 2010), which can account for 20–80 % of the airborne spore load, with peaks occurring during the summer (Burge et al. 1997; Ianovici 2011). In Funchal, this fungal type represents 78 % of the total airspora spectrum, followed by a small number of fungal types which contribute in low proportions to the total counts (each with <5 %) (Table 2). In Bursa (Turkey), *Cladosporium* accounted for 88.1 % of the total fungal spore count (Ataygul et al. 2007), followed by *Alternaria* (4.9 %) and *Fusarium* (0.8 %). Likewise, in Cartagena (Spain), *Cladosporium* attained 62.2 % of the total and *Alternaria* (5.3 %) was the second most abundant fungal type (Elvira-Rendueles et al. 2013). A similar trend was reported in Thessaloniki (Greece) by Gioulekas et al. (2004) and Sabariego et al. (2007) in Madrid (Spain).

The type and amount of fungal spores dispersed in the atmosphere of Funchal city might be affected by an array of regional and local factors. In analogous aeromycological surveys performed in other regions, the available substrate, the plant cover and the prevailing weather conditions were pointed out as the most important factors affecting the distribution pattern of the airborne fungal spores. In fact, the peak season for many fungi overlaps with the maturation and harvesting of important crops (Dixit et al. 2000) and is also determined by the geographic features and the land use of each region (Gonianakis et al. 2005). The city of Funchal has been expanding over the years and becoming built up, despite the abundance of public gardens and farmland in the urban area. At first thought, the available organic substrate for common

airborne fungi and the meteorological parameters should have the biggest influence on the airspora content of the city; however, we consider that this influence is surpassed by the dimension and insular condition of the region, where the atmosphere–ocean interaction heavily influences the quantity and seasonality of fungal spores.

The year-round occurrence of the anamorphic fungal types seems to be favoured by the temperate climate of Madeira region, whose mild temperatures and relative humidity levels promote the development and spread of common outdoor fungal spores. Nevertheless, two peak concentrations were observed, one in the spring and summer, reported during April and June, and the other in the autumn that lasted from September until November (Fig. 2). The first peak overlaps with the increase in the air temperature, favouring the onset of dry fungal spores, since for their liberation is necessary a certain degree of dryness in the atmosphere, which occurs when the temperature rises (Morin 2001). The precipitation is scarce but intense, occurring mainly in late spring, which could explain some peak concentration detected. It is the period, when the vegetation is in its maximum growth and diversity due to the rainfall period of winter season that ends immediately after. The second peak of conidiospores (September–November) may be due to an increase in the availability of substrate owing to the seasonal processes of decomposition of plant matter and increased relative humidity (González et al. 2004).

The occurrence of *Cladosporium*, *Drechslera* and *Torula* (Fig. 3) was favoured by the increase in air temperature and low precipitation levels during May and September (Fig. 4), which was proven by the statistical test (Table 3). These weather conditions seem to be essential for the production and release of these so-called dry fungal spores. The negative association between fungal spore occurrence and precipitation parameter might be a limiting factor for their sporulation and dispersal, as rainfall has the potential to wash out fungal spores from the atmosphere, and absorption of water by spores might make them heavier and less transportable (Minero et al. 1994). *Cladosporium* is considered the main source of inhaled fungal allergens worldwide. The positive correlation between its concentration in the atmosphere of Funchal and air temperature is in accordance with other reports (Stepalska and Wołek 2005; Aira et al. 2008; O’Gorman and Fuller 2008). *Alternaria* is

also considered an important allergen worldwide. It occurs in the atmosphere of Funchal throughout the year, especially during spring and summer (Fig. 3), a pattern similar to that observed by Rizzi-Longo et al. (2009) in Trieste (Italy) and Kasprzyk et al. (2013) in Poland. In addition to *Alternaria*, *Curvularia*, *Nigrospora* and *Tetraploa* spores have also showed a typical behaviour of dry-air fungal spores (Ataygul et al. 2007; Escudero et al. 2011), presenting concentration peaks in the hottest time of the year (July–September) (Fig. 3). *Curvularia* is recognized as an important aeroallergen causing allergic fungal sinusitis, being implicated in hypersensitivity reactions type I (hay fever and asthma) (Chrzanowski et al. 1997; Platt-Mills and Solomon 1998) such as *Nigrospora* (Platt-Mills and Solomon 1998). However, the allergenicity of *Tetraploa* type has not been yet documented.

In turn, *Botrytis*, *Fusarium* and *Gliomastix* have occurred mainly before the hottest period of the year (before June) (Fig. 3), a fact reinforced by the negative and significant value of the correlation between their concentration and air temperature (Table 3). *Botrytis* peaked in May and June, but in other works, a later sporulation season has been reported (Stępalska and Wołek 2005). *Botrytis* is a typical wet spore (Levetin 1995), implicated in allergic episodes (Kurup et al. 2000; Pharmacia Diagnostics 2004/2005) and in the infection of several plants, including species with economical importance such as grapevines (Fernandez-Gonzalez et al. 2012). Like *Botrytis*, the occurrence of *Fusarium* is usually favoured by humid, warm weather and the rain. However, we did not observe a positive correlation with the concentration of those fungal spores and rain. The genus *Fusarium* is a common pathogen of cereal crops worldwide (Keller et al. 2014) and a common allergen implicated in skin reactions, hypersensitivity reactions type I, mycetoma and keratitis (Pharmacia Diagnostics 2004/2005). *Gliomastix* spores have been also detected in patients with chronic rhinosinusitis, but its allergenicity has not been yet determined (Ponikau et al. 1999).

Finally, the spores of *Arthrimum*, *Epicoccum*, *Polythrincium* and *Spegazzinia* have not shown any correlation with the meteorological parameters analysed. *Arthrimum* occurred in springtime, like *Polythrincium* and *Spegazzinia*. *Epicoccum* was found all year round, mainly between April and November

(Fig. 3). In Poland, *Epicoccum* also peaked during summer and autumn (Stępalska and Wołek 2005; Rizzi-Longo et al. 2009), and in Bursa, these spores occurred all of the year, peaking between June and August (Ataygul et al. 2007). *Epicoccum* is considered as a dry-air fungal spore, found in higher concentrations during warm, dry and windy weather conditions. It can trigger high skin reactivity, sharing allergenicity with different fungi responsible for type I hypersensitivity reactions (Chew et al. 2000; Pharmacia Diagnostics 2004/2005). *Polithrincium* presented a seasonal distribution in the atmosphere of Funchal, with higher concentrations in May (springtime), as observed in Porto, mainland Portugal. In the atmosphere of Rzeszów, *Polithrincium* was observed from April to November (Kasprzyk et al. 2004), although in Cracow, these fungal spores were observed from June to October, with peak values in August (Stępalska and Wołek 2005). *Polythrincium* is also considered an important allergen (Platt-Mills and Solomon 1998). *Spegazzinia* peaked in May and July, and its allergenicity has not been documented yet.

5 Conclusions

Anamorphic fungal spores are the most prevalent fungal class detected in the atmosphere of Funchal. Conidiospores were recorded in spring (April–June) and autumn (September–November), with *Cladosporium* being the most prevalent fungal type. Total counts during the period studied were considerably lower comparatively to other mainland stations, probably due to the insular condition of the region. For that reason, the weather-related parameters have a weak influence on airborne conidia counts and their distribution over the year. Nevertheless, temperature reveals a low positive and significant effect on anamorphic fungal spore concentration and rainfall a negative effect. The majority of these fungal types observed are considered potential aeroallergens, and so this study is of interest to allergologists and patients sensitive to these fungal spores, as it provides an indication of the periods, when the highest conidiospore counts may be expected. Further, the co-occurrence of the fungal types observed in our region increases the risk of sensitization to these common aeroallergens.

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