# Effect of Outdoor Fungus Concentrations on Symptom Severity of Children with Asthma and/or Rhinitis Monosensitized to Molds

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**SUMMARY** Although the relationship between asthma severity and exposure to airborne fungi has been well studied, little is known about the contribution of outdoor molds to the symptoms of children monosensitized to molds. In this study, we aimed to investigate the effect of outdoor mold spore concentrations on daily asthma and/or rhinitis scores in children monosensitized to molds. Nineteen children with asthma and/or rhinitis sensitized only to molds recorded their daily symptoms and PEF values to the diaries, from February 2005 to January 2006. Additionally, mold spores were measured daily using a Burkard 7-day recording volumetric spore trap in city atmosphere and compared with meteorological data. Total number of mold spores in atmosphere was found to be 352,867 spore/$m^3$ during the study period. *Cladosporium* (53%) was the most common encountered outdoor fungi, followed by *Alternaria* (29%) and 1-septate *Ascospore* (3%). Outdoor fungi concentrations were significantly correlated with mean monthly rhinitis score ($r$ = 0.877, $p$ < 0.001) and mean monthly asthma score ($r$ = 0.831, $p$ = 0.001), and mean monthly morning PEF ($r$ = -0.741, $p$ = 0.006) and evening PEF ($r$ = -0.720, $p$ = 0.008), and climatic conditions. The effect of outdoor fungi was highly evident on the symptoms of our patients with asthma and/or rhinitis monosensitized to molds.

The possibility that fungal spores may cause allergic asthma has been recognized for over 30 years.[1] Epidemiological studies have generally concentrated on the association of respiratory symptoms to indoor or outdoor exposure to molds.[2-4] Previous studies reported inconsistent findings on the relationship between symptom severity and outdoor fungi concentrations.[5-9] Although some studies have found an association between total counts of mold spores and asthma symptoms in children and adults,[1,5,7,9,10] others have reported a correlation between severity and specific groups or species.[6,7,9] Previously we have shown that the rate of sensitization to molds in skin prick tests was 11.6% among children with respiratory allergy.[11] Therefore, molds have been accepted as significant allergens of our city following mites (61.7%) and grasses (19.8%). The objectives of this study were (1) to investigate the effect of outdoor mold spore concentrations on

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daily disease severity of children with asthma and/or rhinitis monosensitized to molds throughout one year, (2) to describe the types and concentrations of outdoor fungi in the city atmosphere for the same year and (3) to analyze the relation between the concentration of outdoor fungi and meteorological conditions.

## SUBJECTS AND METHODS

### Subjects

Twenty-nine children aged 4-13 years who fullfilled the GINA[12] criteria for asthma and/or ARIA[13]/EAACI[14] criteria for rhinitis were recruited from our outpatient clinic. Skin prick tests were performed with a standard panel of respiratory allergens including mites (*Dermatophagoides pteronyssinus* and *D. farinae*), grass mix, tree mix, mold mix, *Alternaria*, *Cladosporium*, *Penicillium*, *Aspergillus*, eucalyptus pollen, olive pollen, cat and dog dander and some food allergens (milk, egg, wheat and peanut) (Allergopharma, Reinbeck, Germany). A mean wheal diameter greater than 3 mm was considered positive if no dermographism and/or positivity of negative control were recorded. All patients had at least one positive reaction to mold allergens (*Cladosporium*, *Alternaria*, *Penicillium*, *Aspergillus*) in skin prick tests. Presence of sensitization to other aeroallergens was considered as exclusion criteria.

The study was conducted from February 1, 2005 to January 31, 2006. Each day during the study, the participants recorded a morning PEF (peak expiratory flow) reading, an evening PEF reading, medications taken and symptoms related to asthma and/or rhinitis. Patients were instructed to check "yes" for each symptom occurred during the day. Symptoms related to rhinitis (running nose, sneezing, nasal obstruction) and asthma (cough, wheezing, chest tightness, shortness of breath) were calculated and expressed as rhinitis or asthma score which the daily maximum score was 3 for rhinitis and 4 for asthma. Upper respiratory tract infections were excluded from scoring of rhinitis.

Symptom diaries and all forms were approved by the medical committee and all patients and their caregivers gave informed consent to the study.

### Outdoor air sampling

The outdoor samples of airborne fungi were collected by Burkard 7-day recording volumetric trap. The device was placed on the roof of the town hall at a height of 15 meters (m) above the ground and samples were collected from February 2005 to January 2006. The air was sucked at a flow rate of 10 liters per minute, and spore grains impacted on to tapes that were coated with a thin film of Vaselin-paraffin wax in toluene. The tape was then mounted with glycerin jelly,[15] and 24 transverse travels were observed for a daily slide. Spores were identified visually with light microscope at a magnification of 400x by an experienced aerobiologist (M.P.). Fungal spore counts were expressed as fungal spores per $m^3$ of air.

### Meteorological data

Daily and monthly meteorological data, *i.e.* temperature, relative humidity, precipitation, velocity of the wind, were obtained from the bulletin of meteorological station located at the city center.

### Statistics

All the analyses were performed using computer software (SPSS version 11.0; SPSS; Chicago, Illinois, USA). Descriptive statistics were expressed as mean + SD, geometric mean, median and range. Spearman correlation test was used to test the relation between outdoor spore counts and meteorological data, symptom scores and PEF variables.

## RESULTS

### Study area

The city located on east Mediterranean coast, at the sea level, in a temperate climate and characteristically has mild winter and very humid summer months. The average level of humidity varies between 50% and 70% all year long and makes its peak during the summer, sometimes up to 80%.

**Summary of patient group**

Among the 29 patients enrolled to the study, only 19 patients completed a great portion of the diaries and thus included in the analysis. Two patients had only rhinitis, while 17 patients had both asthma and rhinitis. Daily diaries were obtained for a total of 6,777 person-days of observations. During one year study, noncompliance with diary completion accounted for 158 person-days.

The PEF data of the five asthmatic patients were not included in the analysis because of their compliance problem. There were a total of 4,142 person-days of observations for morning PEF and 4,161 person-days of observation for evening PEF, with most of the remaining missing observations for times when subjects did not perform. Demographic data of the patients and the mean scores of asthma and rhinitis are shown in Table 1.

**Outdoor fungal spore counts**

The total number of mold spores in the city atmosphere was 352,867 spores/m$^3$ from February 1, 2005, to January 31, 2006. Thirty-five species were recorded and identified during the one-year period outdoor sampling. The fungi most commonly recovered from outdoor air were *Cladosporium* (53%), *Alternaria* (29%), 1-septate *Ascospore* (3%) and *Exosporium* (2.7%). The most common encountered 10 outdoor fungi spore concentrations are shown in Table 2.

Outdoor fungal concentrations were highest in June (89,089 spores/m$^3$), July (58,813 spores/m$^3$) and August (64,266 spores/m$^3$), while the lowest value was detected in January (876 spores/m$^3$) (Fig. 1). From April to July, *Cladosporium* spp. was predominant, from November to March, *Cladosporium* and *Alternaria* were equally predominant, and in August *Alternaria* spp. was predominant.

**Meteorological data**

During the sampling period, the air temperature was in the range of -2.90°C-37°C, and the relative humidity ranged from 62.9% to 79.3%. Summary of meteorological data is given in Table 3.

The outdoor fungi concentrations were significantly correlated with the monthly average temperature ($r$ = 0.883, $p$ < 0.001), average relative humidity ($r$ = 0.622, $p$ = 0.03), monthly average precipitation ($r$ = -0.685, $p$ = 0.01), the number of

**Table 1** Demographic data and some characteristics of the patients

| **Gender** | **Males(13)/females(6)** |
|---|---|
| **Age** (range in years) | 4-13.3 |
| mean ± SD | 8 ± 3.1 |
| **Diagnosis** | Asthma and rhinitis (17), Rhinitis only (2) |
| **Skin prick test positivity** (≥ 3 mm) | *Cladosporium* and *Alternaria* and *Penicillium* and *Aspergillus* (10) |
| | *Penicillium* and/or *Aspergillus* (5) |
| | *Cladosporium* only (3) |
| | *Alternaria* only (1) |
| **Treatment protocol** | |
| **For asthma** | Budesonide 200-400 μg/day or Fluticasone 100-250 μg/day |
| **For rhinitis** | Nasal steroid and/or oral antihistamines |
| **Duration of the disease** (years), mean ± SD | 4.89 ± 2.47 |
| **Mean ± SD of asthma symptom scores** | 0.11 ± 0.06 |
| **Mean ± SD of rhinitis symptom scores** | 0.17 ± 0.07 |

rainy days per month ($r$ = -0.613, $p$ = 0.03), and average velocity of the wind of the month ($r$ = -0.821, $p$ = 0.001) (Fig. 1).

**Symptom scores and outdoor fungal spore counts**

Mean monthly rhinitis scores were highest in June, July and August, while the peak of mean asthma scores were in July, August and September. The mean monthly rhinitis scores were significantly correlated with both mean monthly outdoor *Cladosporium* spore concentrations ($r$ = 0.818, $p$ < 0.001) and *Alternaria* spore concentrations ($r$ = 0.898, $p$ < 0.001) (Fig. 2). Outdoor fungi concentrations were significantly correlated with mean monthly asthma scores ($r$ = 0.831, $p$ = 0.001), with mean monthly morning PEF ($r$ = -0.741, $p$ = 0.006) and mean monthly evening PEF ($r$ = -0.720, $p$ = 0.008).

## DISCUSSION

In the present study, we investigated the effect of outdoor fungi on symptom severity and PEF

**Table 2** Types and frequencies (spores/m$^3$) of the most common encountered 10 outdoor fungi during the study period

| Fungi species | Sum | Range | Median | % |
|---|---|---|---|---|
| *Cladosporium* | 187,325 | 165-46,101 | 11,274 | 53 |
| *Alternaria* | 102,474 | 111-31,683 | 3,976 | 29 |
| *1-septate Ascospore* | 10,513 | 87-2,783 | 464 | 3 |
| *Exosporium* | 9,658 | 12-4,095 | 306 | 2.7 |
| *Ustilago* | 6,065 | 37-1,372 | 364 | 1.7 |
| *Drechslera* | 5,786 | 48-2,385 | 473 | 1.6 |
| *Epicoccum* | 4,846 | 3-2,077 | 107 | 1.4 |
| *Pleospora* | 4,291 | 17-2,130 | 48 | 1.2 |
| *Periconia* | 3,728 | 14-1,477 | 102 | 1.0 |
| *Leptosphaeria* | 3,571 | 114-762 | 258 | 1.0 |

**Table 3** Meteorological data of the city during the study period

| | Temperature (°C) Range (mean) | Mean relative humidity (%) | Mean monthly precipitation (mm) | Maximum wind speed (m/s) |
|---|---|---|---|---|
| **February** | -2.90-22.30 (10.30) | 63.80 | 75.60 | 13.40 |
| **March** | 4.20-23.30 (13.90) | 71.80 | 61.10 | 17.40 |
| **April** | 5.70-34 (18.10) | 68.70 | 53.00 | 10.30 |
| **May** | 9.20-35.20 (22.00) | 67.80 | 41.20 | 13.20 |
| **June** | 15.80-35.20 (25.70) | 72.40 | 16.10 | 8.60 |
| **July** | 22.20-35.20 (28.70) | 79.30 | 7.60 | 8.10 |
| **August** | 21.60-37 (29.20) | 76.40 | 24.40 | 9.10 |
| **September** | 16.10-35.10 (26.00) | 69.20 | 28.10 | 13.60 |
| **October** | 7.70-33.20 (19.80) | 60.60 | 37.90 | 12.40 |
| **November** | 3.60-27.80 (13.90) | 66.60 | 64.60 | 13.50 |
| **December** | -2-27.40 (12.10) | 69.70 | 64.10 | 13.70 |
| **January** | -1.20-19.10 (8.80) | 62.90 | 36.30 | 14.70 |

values of 19 children with asthma and/or rhinitis monosensitized to molds throughout one-year. Outdoor fungi concentrations were highest in the summer and significantly correlated with both asthma and rhinitis scores, and PEF values. Outdoor fungi were also significantly correlated with the average temperature, relative humidity, precipitation and velocity of the wind of the month.

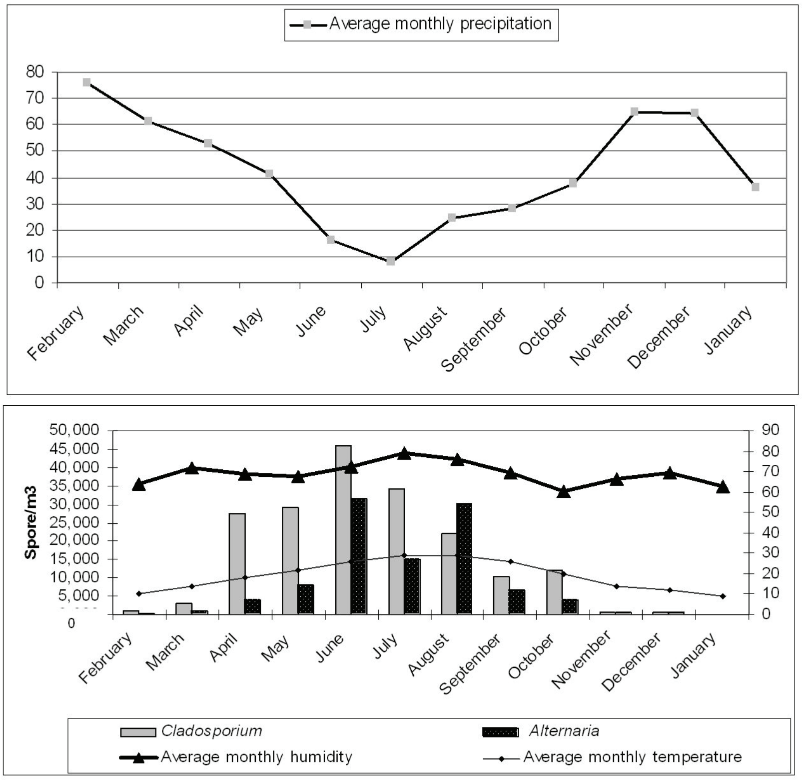


**Fig. 1** The relationship between *Cladosporium* and *Alternaria* concentrations in atmosphere and meteorological data.

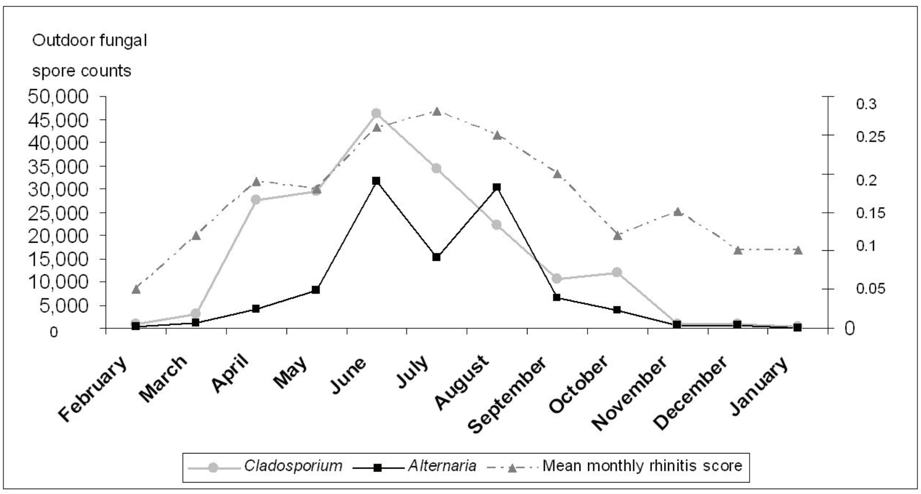


**Fig. 2** Outdoor *Cladosporium* and *Alternaria* counts were found significantly correlated with mean monthly scores of rhinitis ($p < 0.001$).

The range of outdoor fungi concentrations varies in different parts of the world. It has been reported as total spore concentration < 200 spores/m$^3$ from Palencia, Spain;[16] annual average spore concentration of 173,784 spores/m$^3$ from Thessaloniki, Greece;[17] daily average spore concentration of 1,207 spores/m$^3$ from Melbourne;[18] monthly average spore concentration range of 1,500-4,500 spores/m$^3$ in Santiago.[19] The total number of mold spores was reported as 429,264 spores/m$^3$ in another city of our country.[20] The daily average outdoor spore concentration range of our city was 29-2,969 spores/m$^3$ (monthly geometric mean 13,088 spores/m$^3$) during the study which was a high value for exposure. There have been many reports concerning the monthly (seasonal) variations of fungi in oudoor air in many European and North American countries.[16-19] Most reports showed that peak of concentration of fungi are recorded during the summer and early fall months and the lowest during the winter months.[16,17,21,22] Similarly, we found highest outdoor fungi concentration in June, followed by August and July, and the lowest in January. *Cladosporium* was the predominant fungi in atmosphere throughout the year; but, surprisingly only in August *Alternaria* counts exceeded *Cladosporium*.

Some studies showed that a relationship appeared to exist between *Alternaria* and *Cladosporium* frequency (counts) and climatic conditions.[23,24] In the present study, we also found significant correlation between the concentration of oudoor fungi and meteorological conditions such as average monthly temperature, relative humidity, precipitation and velocity of the wind of the month, and number of rainy days of the month.

Humidity is usually accompanied by precipitation. However, it is surprising that in our city highest humidity was recorded in summer at the time of year as the lowest rainfall. This unusual condition is the characteristic feature of our city and related with its geographic location.

We showed that both asthma and rhinitis symptom scores of patients monosensitized to molds were affected significantly from rises of mold concentrations in the atmosphere. Fourteen of 19 patients in this study had *Cladosporium* and/or *Alternaria* sensitization in skin prick tests, however, we could not correlate symptom scores with counts of causative fungi.

There are some limitations of our study. First, the number of the study patients is few, but the patients with asthma and/or rhinitis monosensitized to only molds are not common in allergy clinics. Secondly, we did not have a control group and therefore, each child act as his/her own control.

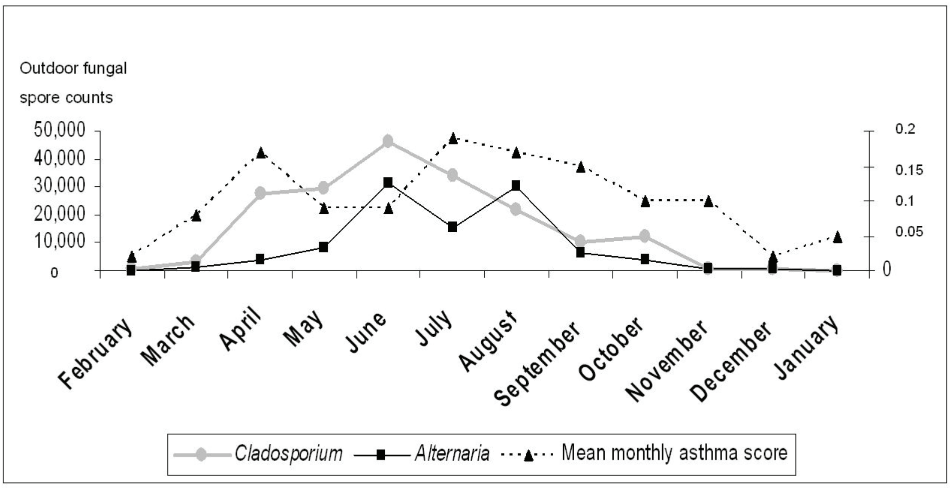


**Fig. 3** Outdoor *Cladosporium* and *Alternaria* counts were found significantly correlated with mean monthly scores of asthma ($p$ = 0.001).

In conclusion, we found that symptoms of children with asthma and/or rhinitis monosensitized to molds have been influenced predominantly by outdoor air fungi. Forecasting of fungi in oudoor air and to take measures against environmental exposure to molds in homes may be helpful in the management of allergic diseases caused by fungi spores.

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