Environmental Contributions to Allergic Disease

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Exposure to indoor allergens and contaminants has been linked to the severity of allergic disease. The contribution of house dust mites, cockroaches, animal dander, and indoor molds was studied intensively and reviewed in several recent papers [6–9]. Perhaps some of the strongest evidence suggests a compelling link between exposure to passive smoking and the development and manifestation of childhood asthma [1••,2]. While not denying the importance of indoor exposure, we feel that the emphasis on indoor allergens has overshadowed the contribution of outdoor exposure. As a result, this review primarily examines some of the recent literature relating to the contribution of outdoor factors to allergic disease.

Influence of Exposure to Pollutants on the Manifestation of Allergic Disease

Ozone

Single ozone (O₃) exposure was shown to evoke cough, breathlessness, and chest pain on inspiration. These symptoms signal restriction of ventilation resulting in decreased forced vital capacity (FVC) and FEV₁. In addition, O₃ was shown to induce neutrophilic inflammation of the airway submucosa and to increase airway reactivity [1••]. However, the response to ozone showed large intersubject variability [10], with only 10% to 20% of the population sensitive to lung function reductions after O₃ exposure [11]. Inflammatory changes in the airways resulted in greater numbers of neutrophils. However, eosinophils, several cytokines, and leukotrienes did not vary when measured in bronchoalveolar lavage fluid and in large airway mucosal biopsy specimens [12]. Airway inflammation seen after O₃ exposure differed substantially from inflammatory changes that occurred in the airways of asthmatic subjects. However, asthma was determined to be a risk factor for bronchial response to inhaled ozone. After 2 hours of exposure to 0.4 ppm ozone, those subjects with asthma had a greater decline in pulmonary function [13]. In a more recent study, Basha et al. [14] did not find any reduction in pulmonary function after 6 hours of exposure to 0.2 ppm O₃, but bronchoalveolar lavage showed significant increases in bronchial hyperresponsiveness (BHR). Because of the marked inflammatory responses in the airways and increased BHR in healthy subjects, there is concern that ozone may enhance the development of asthma in populations exposed to significant concentrations [1••].
Romieu et al. [15] found significant relationships between daily emergency visits for asthma at a pediatric hospital in Mexico City and air pollutant levels. A multivariate regression model predicted that each 50-ppb rise in the one-hour maximum O₃ level would lead to a 43% increase in asthma visits during the following day. This level rose to 68% if O₃ levels were greater than 110 ppb for 2 consecutive days. In another study, White et al. [16] found that the average number of visits to an emergency department for asthma or reactive airway disease in children in Atlanta was 37% higher on days after elevated O₃ values. A similar lag in response was found by Bates and Sitzo [17] in southern Ontario, Canada. These lags suggested an augmentation of airway sensitization or inflammation rather than a direct O₃ effect on the lungs. Kehrl et al. [18] reported that the exposure of subjects with mild asthma to O₃ levels of 0.16 ppb resulted in modest lung impairment but that the reactivity to dust mite allergen was bolstered. Similar results were found with grass and ragweed allergens [19]. These studies suggested that ambient allergen exposure combined with high O₃ levels may contribute to the aggravation of asthma. The effects of longer term O₃ exposure remain controversial; however, recent work by Kinney and Lippmann [20] showed a seasonal decline in respiratory function as well as increased reports of cough, chest tightness, and sore throat in subjects after a summer of outdoor exposure.

**Sulfur dioxide and nitrogen dioxide**

The association of sulfur dioxide (SO₂) with respiratory effects was demonstrated, and it led to the development of common pharmaceutical agents used to treat asthma [21•]. Studies on SO₂ inhalation showed that this gas not only leads to bronchoconstriction in both healthy and asthmatic subjects but that the response may vary, with asthmatics being much more sensitive. For example, inhalation of 1.0 ppm of SO₂ for 10 minutes during moderate exercise can decrease FEV₁ by 23% and increase total lung resistance by an average of 67%. In addition, exposure for as little as 2.5 minutes is sufficient to cause SO₂-induced bronchoconstriction in asthmatics. Decreased FEV₁ values and FVC have occurred at concentrations as low as 35, 70, and 100 µg/m³ for durations of 30 to 40 minutes during exercise [21•].

Nitrogen dioxide (NO₂) was shown to lead to epithelial dysfunction and/or damage resulting in acute inflammatory response in the upper and lower airways [22•]. Exposure to NO₂ or the combination of NO₂ and SO₂ was shown to cause a significant reduction in the dose of inhaled allergen needed to provoke an allergic response, and the reaction may persist for 24 to 48 hours [22•]. Using nasal lavage, it was found that prior exposure to NO₂ primes eosinophils for subsequent activation by allergen [23], as shown by a significant increase in eosinophil cationic protein (ECP) when allergen is introduced. Peak hourly NO₂ concentrations were associated with asthma visits to emergency departments during the winter months in northern California and Athens, Greece, as well as both winter and summer in Barcelona, Spain. In addition, a recent study of daily emergency department visits for asthma at 12 hospitals in London reported significant increases in visits associated with rises in SO₂, NO₂, and particulate (PM₁₀) concentrations [24]. These studies not only showed the association between high levels of these gases and disease manifestation but also pointed to the difficulty in determining single causative environmental factors.

**Diesel particles**

Diesel exhaust particles (DEPs) are the single greatest contributor to particulate pollution in most urban areas and account for up to 90% of the total particle mass in some major cities. The median mass diameter of a DEP is approximately 0.2 µm with more than 90% being less than 1 µm [10••], thereby allowing the penetration of particulate matter deep into the lungs. Diesel exhaust particles consist of an inert carbon core containing unburned fuel petrochemicals that are mostly polyaromatic hydrocarbons (PAHs), with the majority being three- to five-benzene–ring compounds [25]. The PAHs are essentially nonvolatile and remain with the diesel particles until they reach the mucosa, where their hydrophobic nature allows them to diffuse easily through cell membranes and bind to a common cytosolic receptor of the aromatic hydrocarbon receptor complex. In addition, DEPs and their associated PAHs have been shown to absorb allergens and may act as adjuvants by prolonging the retention of the allergen, leading to an enhanced immune-allergen response.

There is growing evidence that DEP exposure may result in enhanced IgE production. For example, Takenaka et al. [26] showed an increased production of human B cells and IgE after exposure to PAHs from DEPs by modifying ongoing transcriptional programs. IgE production in cells currently synthesizing the antibody rises after exposure to PAH [27]. The effects of DEP on the allergic response likely involves T cells, mast cells, macrophages, and epithelial cells. Nel et al. [28] provided a thorough discussion of the biochemical and molecular components of these systems. Diaz-Sanchez et al. [29] showed that the concentration of ragweed-specific IgE was 16 times higher following ragweed plus DEP challenge compared with ragweed alone. In addition, DEPs were shown to absorb allergens from grass pollen onto their surface [30], whereas Ormstad et al. [31] reported that DEPs bind with cat, dog, grass, and birch allergens in vitro under aqueous conditions. While studies indicate that exposure to DEPs might exacerbate pre-existing allergic conditions, there is no indication that they induce new cases of atopy or asthma [1••].

High particulate levels were found to increase emergency room visits for asthma in several studies. For example, in Seattle, Washington significant rates of hospitalization occurred during the day following high levels of PM₁₀, whereas mean PM₁₀ levels for the previous four days provided an even better predictor of admittance [32]. The study further showed that a change of 11 µg/m³ in PM₂.₅ was associated with a 15% increase in hospital visits. Similar results were found in
Anchorage, Alaska, where an increase of 10 µg/m³ in PM₁₀ was associated with a 3% to 6% rise in asthma visits. Concern about the health effects of these respired particles has led to the tightening of ambient air quality standards in the United States, Europe, and the United Kingdom. Recent proposals to increase regulation of PM₂.₅ by the US Environmental Protection Agency remain controversial, but have led to a re-analysis of air quality data and studies that have shown increased morbidity during high PM₂.₅ conditions.

Influence of Exposure to Aeroallergens on the Manifestation of Allergic Disease

Within the past few years, several studies showed that exposure to outdoor fungal spore concentrations, especially Alternaria, has an adverse effect on asthma severity. In a study of asthma deaths in Chicago from 1985 to 1989, Targonski et al. [33] showed that airborne fungal spore concentrations were significantly higher on days when asthma deaths occurred than on days when no asthma deaths occurred. The odds ratio was 2.16 for asthma-related deaths occurring when the outdoor spore concentration was 1000 spores/m³ or greater compared with other days. It should be noted that limited information is available from this statistic. Since the number of asthma deaths was obtained from death certificates, the investigators did not gather any information about other exposures or other risk factors. Also, fungal spores were counted as a single group, with no attempt to identify individual genera. Although this study provided limited information, it agreed with other investigations that showed correlations between fungi and asthma severity.

In 1991, O'Hollaren et al. [34] reported on a group of 11 asthma patients who had sudden respiratory arrest. This group was compared with 99 control patients from the same clinic who had no history of respiratory arrest. Skin-test records showed that 10 of the 11 with respiratory arrest (91%) were sensitive to Alternaria, compared with 31% of the control patients. This difference was significant (P < 0.001). In three of the arrest patients, Alternaria sensitivity was the only positive skin test. Overall, the risk of respiratory arrest is approximately 200 times greater in the skin-test-positive group. Serum IgE antibodies to Alternaria were elevated in nine of the 11 patients tested. In addition, all cases of respiratory arrest occurred during the Alternaria aeroallergen season.

Recently, Neukirch et al. [35] examined the association of Alternaria sensitivity to severe asthma. Approximately 1000 people were skin tested with 11 common allergens; from this group, 108 subjects were asthmatic. Investigators used several diagnostic criteria to classify the asthmatics as having mild, moderate, or severe disease. Analysis of the asthmatic group showed that the percentage of subjects with skin tests positive for Alternaria, Timothy, olive, dust mite, and cat increased as asthma severity increased. However, in a multiple regression model used to test the associations between severe asthma and sensitization to allergens, only Alternaria was independently associated with severe asthma.

Delfino et al. [36] examined the interaction as well as the independent effects of ozone and aeroallergens on self-reported asthma symptoms in a group of asthmatic children in the San Diego area. The subjects recorded daily asthma symptoms and inhaler use during September and October. The investigators found that outdoor fungal spore levels and personal ozone exposure were significantly associated with asthma severity. The effect of fungal spores varied with the type of spore; the greatest effect on symptom scores seemed to be from those fungi not included in skin-test panels, such as basidiospores and ascospores. There was no association between asthma severity and increased pollen levels.

In a follow-up study with a different panel of asthmatic subjects, Delfino et al. [37] found similar results showing a relation between exposure to fungal spores and asthma symptoms, even though a second study was conducted in a different climatological region (inland vs coastal) and different season (spring vs fall). In the second study, those fungal spores included in skin-test panels had a greater effect than non-test fungi. The investigators found that for an increase of 1000 spores/m³, asthma symptom scores (on a scale of 0 to 5) increased by 0.16 for all test fungi and 0.14 for all non-test fungi. Both increases were significant. Asthma symptoms again were associated strongly with basidiospore levels. Symptom scores increased by 1.5 for every increase in 1000 spores/m³. In addition, asthma symptom scores were correlated strongly with the basidiospore levels on the previous day, suggesting a possible lag effect. The effect of basidiospore levels on symptom scores was even more pronounced when the association with Coprinus (one type of basidiospore) was examined. Symptom scores increased 2.3 for every increase in 1000 spores/m³. This second study included both adults and children in the panel, and when individual regression analysis was performed, it showed that basidiospores were significantly associated with asthma symptom scores in seven of the 13 pediatric subjects but in none of the adults. Symptom scores among the pediatric subjects changed from 2.5 to 6.0 for every 1000 spores/m³. In contrast to symptom scores, neither peak flow values nor inhaler use was associated with basidiospore levels. Also, in this second study, none of the outcome variables was associated with ascospore or pollen levels.

It should be noted that the fungal spore levels during these California studies [36,37] generally ranged from daily averages of 650 to 7500 spores/m³. Asthma severity was found to increase when concentrations increased within this range. Similar studies need to be conducted in other regions where spore concentrations are higher. In Oklahoma, fungal spore levels can be an order of magnitude greater; there are days when the average daily concentration of spores exceeds 50,000 spores/m³. During a two-week period in September 1998, three such days occurred in Tulsa (Fig. 1): September 18, 21, and 30. These days saw increases of 20,000 spores/m³ from the previous days and even greater hourly changes. Examination of the hourly data on these days showed a peak hourly concentration of 85,059 spores/m³ on September 18.
While bioaerosols typically show a diurnal rhythm with a mid-day peak for most asexual fungi and an early morning peak for basidiospores, the levels seen during these spore plumes are four to five times higher than the normal diurnal peak. Although basidiospores were not the dominant spore type in the atmosphere on these days, their atmospheric concentrations showed remarkable increases as well (Fig. 2). It seems likely that asthma severity could increase dramatically in regions such as Tulsa, where spectacular increases in spore levels may occur.

The results from the San Diego studies [36,37] concurred with previous studies that showed the importance of basidiospores. Although basidiospore extracts generally are not included in skin-test panels, a series of studies with non-commercial extracts showed levels of sensitivity comparable with those fungi traditionally used in testing [38]. Horner et al. [39] examined the risk of asthma associated with the sensitization to basidiospores and compared it with risk associated with the sensitization to the more commonly recognized asexual fungal (mold) spores. The study examined skin-test reactivity in 701 clinic attendees from seven US and five European cities. They found that asthma was more closely associated to sensitivity to basidiospores than to mold spores. The relative risk (RR) of developing asthma was significantly related to basidiospore sensitivity (RR = 1.37) but not significantly related to sensitivity of the mold spores used in the test panel (RR = 1.15). The RR of asthma associated with basidiospore sensitivity was even greater (1.87) in subjects less than 20 years old and also greater (1.45) in those not sensitive to dust mites. The authors concluded that routine skin testing with basidiospore extracts is useful for the diagnosis of atopic asthma.

Epton et al. [40] examined the influence of climate and aeroallergens in a prospective study of 139 mild to moderate asthmatics in New Zealand. They found that from all the pollen and spore categories, only basidiospores were significantly associated with asthma exacerbation. High basidiospore levels were associated with high symptom scores, increased risk of waking at night, and high medication use. Recently, Rosas et al. [41] studied the relationship between asthma emergency admissions to a Mexico City hospital and aeroallergen levels, air pollution, and weather. Asthma admissions data were stratified by age into children, adults, and senior citizens; and weather was categorized into wet and dry seasons. Separate analyses were carried out for each age group and each season, resulting in six separate data sets. Investigators developed one or more models of environmental variables that best explained the asthma admissions for each data set. They found that grass pollen was included in the predictive models for both children and adults during both seasons. For children, aspergillus concentrations were also associated with asthma admissions during the wet season. The association with ascospores (coefficient = 0.27) was almost as important as the association with grass pollen (coefficient = 0.39). During the dry season, either asexual (mold) spores or basidiospores were included in grass pollen in separate models for children. None of the fungal spore categories was included in models for either adults or senior citizens in either season. Surprisingly, there were no strong statistical associations between any of the air pollutants and asthma admissions. Also, this study differed from those of Delfino et al. [36,37] in finding a significant association between asthma and pollen levels.

Because pollen grains deposit in the nasopharynx, they are associated with rhinitis, sinusitis, or conjunctivitis. As a result, it is generally believed that exposure to pollen is not a risk factor for asthma [7,40]. Even though many asthmatics are sensitive to various pollen types, the sensitivity has not been associated with increased risk for asthma [6]. However, several studies, such as the one cited above [41], have implicated pollen exposure with asthma exacerbation.
Grass pollen was implicated in outbreaks of asthma associated with thunderstorms in Australia and England. In each of those epidemics, the incidence of asthma increased dramatically following thunderstorms during the grass pollen seasons. A number of researchers investigated the episodes and described the involvement of grass pollen [42,43,44,45•]. Much of the work focused on *Lolium perenne* (rye grass). Rye grass pollen grains rupture in rainwater releasing about 700 starch grains. These starch grains, which are from 0.6 to 2.5 μm in size, are associated with the major Lol p allergens and can trigger allergic reactions in sensitive individuals. Unlike intact grass pollen, these particles are small enough to enter the lower airways. Four patients showed severe bronchial constriction after inhalation challenge with starch grains [42]. Air sampling also showed that the concentration of starch grains increases in the atmosphere following rain. Grote et al. [44] used immunogold electron microscopy to show that both group 1 and group 5 allergens were found among the cytoplasmic materials expelled from the grains. They could not detect the allergens within the starch grains but found allergen-containing cytoplasmic debris attached to the surface of the starch grains.

Schappi et al. [45•] compared the atmospheric concentration of grass pollen with the atmospheric concentration of group 5 grass pollen allergens and correlated these levels with patient symptoms. They used a Burkard spore trap (Burkard Manufacturing Co., Hertfordshire, UK) to monitor the airborne pollen levels and a high-volume cascade impactor to sample for allergens. The impactor separated particles into two fractions: nonrespirable particles (>7.2 μm) and respirable particles (<7.2 μm). The authors found that group 5 allergen levels in nonrespirable particles were significantly related to the grass pollen counts (r = 0.842, P < 0.001). On dry days, 37% of the total allergen load was associated with respirable particles; however, on days with rain, 57% of the total allergen level was detected in the respirable fraction. The investigators also were able to identify starch grains in the respirable fraction. On these days, the atmospheric pollen levels decreased during periods of rainfall. There was no significant correlation for asthma patient data and grass pollen levels or with allergen level associated with nonrespirable particles. However, the 4-day running mean of grass allergens detected in smaller respirable particles was significantly correlated with diagnoses of asthma in the emergency room of a Melbourne hospital (r = 0.334, P < 0.001). By contrast, hay fever symptoms were correlated with the grass pollen count. These data suggest that there are different environmental risk factors for hay fever and asthma in grass pollen–sensitive patients. A similar mechanism was found in birch pollen; however, the starch grains were released from pollen tubes produced by germinating grains.

Recently, D’Amato et al. [46] described a case of bronchial asthma caused by exposure to *Parietaria* pollen. The subject was a scuba diver who suffered a severe asthma attack while diving at 27 meters. He was skin-test positive to Parietaria and had a high concentration of serum IgE to *Parietaria* allergens. The only factor that distinguished this dive from previous ones was that the scuba tanks were filled in a new area. Upon investigating, it was found that no air filter was used when the tanks were filled, and testing showed that the air in the tanks contained *Parietaria* pollen. *Parietaria* pollen grains are very small, averaging 1.2 to 16 μm. In addition to the small pollen size, the mouth breathing of scuba divers facilitates access to the lower respiratory tract.

O’Rourke et al. [47] studied the relationship between pollen levels and symptoms in Tucson, Arizona. The study population of 121 subjects included control subjects, atopic subjects, and asthmatics. Subjects kept daily diaries of symptoms, which included peak flow measurements. The investigators found a direct relationship between rhinitis and ragweed, mulberry, and total pollen. However, decrease in peak flow values among the asthmatic group was significantly associated only with mulberry pollen. The investiga-
tors suggested that the small size of mulberry pollen (10 to 15 μm) allowed for tracheobronchial deposition.

Although atmospheric pollen concentrations are generally much lower than fungal spore levels, average daily concentrations reported from a central location at rooftop level may underestimate the true nose-level exposure. In addition, hourly concentrations show great variability throughout the day. Hourly concentrations may sometimes reveal remarkably high concentrations; these pollen plumes are comparable to the spore plumes described above. We saw such levels during our studies of *Juniperus ashei* (mountain cedar) pollen. *Juniperus ashei* is responsible for a severe form of allergic rhinitis, known as cedar fever, in Texas, Oklahoma, and other states of the central plains. Although Tulsa is approximately 200 km from the closest populations of *J. ashei*, long-distance transport carries this pollen to the Tulsa atmosphere each winter [48]. The highest daily concentration observed in Tulsa during the past 20 years was greater than 2400 pollen grains/m³. The hourly peak on that day was 5964 pollen grains/m³. Although this is a remarkable level when considering the distance the pollen was carried, it is far lower than levels recorded in Texas. Hourly *J. ashei* pollen concentrations in excess of 40,000 pollen grains/m³ were recorded from our samplers in Texas (Fig 3A). In fact, hourly concentrations above 10,000 pollen grains/m³ were recorded 25 times during three winters in Austin and 12 times during two winters in Junction, Texas. Although pollen release typically occurs during the daytime, with peaks in the late morning, several high concentrations of *J. ashei* were recorded in the middle of the night (Fig 3B). The late-night peaks are thought to be due to the size of the *J. ashei* population on the Edwards Plateau, the peculiar geography of this area, and the meteorological conditions conducive for pollen settling [49]. These pollen peaks may represent significant exposure risks to sensitive individuals.
Avoidance

Much has been written about measures to control indoor allergen exposure, and data suggest that these may reduce sensitization and symptoms [6,8,9]. While avoidance measures seem to be effective in reducing indoor exposure, little can be done to totally eliminate exposure to outdoor bioaerosols. Sensitive individuals are cautioned about being outdoors during ragweed and tree pollen seasons, and those with fungal sensitivity usually are warned to avoid walking in the woods on warm and windy days during the fall, when spore counts are high. However, even a brief exposure to outdoor levels, such as those occurring during times of a spore or pollen plume, may trigger symptoms in sensitive individu-

als. In addition, outdoor bioaerosols can penetrate indoors. Any time doors or windows are open, pollen and spores are introduced. Also, bioaerosols can penetrate through tiny cracks around windows, doors, and walls [50]. Clearly, total avoidance is difficult.

Avoidance measures are most successful when the environmental factors that influence aeroallergen levels are known to sensitive individuals. Current pollen and spore counts available in the media typically provide information that is 24 to 48 hours old. But aerobiology is beginning to make the transition from descriptive to predictive. With the greater availability of meteorological data, aerobiologists now are studying the meteorological conditions correlated with the onset of pollen or spore release as well as those associated with peak atmospheric concentrations. These are the necessary first steps to developing valid forecasts. Accurate forecasts can provide guidance on avoidance and on prophylactic medication. Few forecasts of this type are available now. The Aerobiology Laboratory at The University of Tulsa provides forecasts for mountain cedar pollen in the southern plains (Fig. 4). These forecasts are available on the Internet (http://pollen.utulsa.edu) and furnish sensitive individuals with warnings when a “pollen cloud” is likely to be in their area.

Air pollution models have been developed that predict days when ozone or other pollutants are likely to be high. In most major urban areas, pollution forecasts now are provided to the general public through local news media.

Conclusions

Incidence of allergic disease has increased in recent decades, and some of this increase is linked to environmental exposure. Although indoor allergens are clearly involved in this rise, the outdoor contribution to allergic disease still is of major importance.

Rising levels of atmospheric pollution are linked to the exacerbation of allergic disease. Exposure to high levels of ozone, SO$_2$, NO$_2$, and diesel exhaust particles were shown to reduce lung function. Not only do these chemical species illicit allergic response, most are known to act as adjuvants, resulting in the prolonging of symptoms. In addition, diesel particles were shown to attach to common allergens, thus promoting delivery into the lungs. High atmospheric concentrations are correlated with increased emergency room visits, and in the case of diesel exhaust particles, which are contained within the PM$_{2.5}$ fraction, tightened regulation is being proposed.

The link between exposure to fungal spores and exacerbation of allergic disease was shown in several recent studies. Alternaria has been associated with severe asthma and respiratory arrest, and several studies showed basidiospores to be associated with asthma exacerbation. Although pollen concentrations are generally linked to allergic rhinitis, grass pollen also has been associated with asthma epidemics, especially following thunderstorms. The mechanism is the release of allergen-bearing starch
grains following rupture of pollen in water. Both *Parietaria* and mulberry pollen also have been associated with asthma, suggesting these small pollen grains may be able to penetrate the lower airways. Clearly, these studies emphasize that the outdoor environmental contribution to allergic disease should not be ignored.

References and Recommended Reading

Papers of particular interest, published recently, have been highlighted as:

- Of importance
- Of major importance


This is an outstanding review article that relates environmental pollutants, lifestyle factors, and allergen exposure to the prevalence of allergic disease.


This study correlated self-reported asthma symptoms and peak flow readings with outdoor allergen concentrations and also personal ozone levels. Investigators found that fungal spore levels were associated with all outcomes.


This study investigated grass pollen asthma by comparing levels of grass pollen allergens with airborne pollen concentrations. On dry days, the allergen levels were correlated with pollen levels, but following rain, 57% of the allergen was detected in respirable-sized particles not intact pollen grains.


