

Relation of indoor and outdoor airborne fungal spore levels in the Kansas City metropolitan area

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ABSTRACT

Background: Environmental control is an important component of asthma management for persons with asthma. A damp indoor environment and elevated airborne spore levels are factors in housing environmental control.

Objectives: We investigated if indoor airborne fungal spore levels correlated with outdoor ground-level airborne fungal spores or outdoor centrally collected spore levels as to types and abundance.

Methods: Air collections were taken from home interiors, outdoor areas adjacent to the homes, and at a central location in the metropolitan area at the approximate same time. All air collections were examined and enumerated microscopically, and airborne spore estimates per cubic meter of air were reported for total fungal spores and for 11 identifiable spore groups.

Results: The 244 homes in the study were typical of the North American Midwest. The overall mean total spore counts in spores per cubic meter of air was indoors (4076 spores/m³), outdoors at ground level (8899 spores/m³), and outdoor metropolitan area (8342 spores/m³). All of the major indoor taxa were strongly correlated with the mean total spores present in the home. Total outdoor ground spore levels were highly correlated with levels of major outdoor taxa, such as ascospores and *Cladosporium*. Correlations of indoor spore levels with outdoor spore levels are strong for most major outdoor taxa. Indoor *Aspergillus-Penicillium* and *Chaetomium* are significantly correlated between indoor and local ground-level outdoor air.

Conclusion: Although conditions may exist where indoor or outdoor spore levels were not well aligned, in most circumstances, the outdoor airborne spore community was reflected in the indoor airborne spore community.

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The National Heart, Lung, and Blood Institute's 2007 Expert Panel Report 3 "Guidelines for the diagnosis and management of asthma" recommends that multifaceted allergen-control education interventions be provided in the home setting. It is stressed that environmental control is an important component of asthma management for persons with asthma who are at risk for recurrent exacerbations or that are difficult to manage.¹ Although the prevalence of fungal sensitivity is not well defined, in one U.S. study, 21% of subjects with atopy were positive *via* skin testing to one or more fungal allergens.² Results of a recent study indicated that, in children with asthma and who were sensitized to fungus, the outdoor mold correlated more with asthma exacerbations than did indoor mold.³ A meta-analysis of 33 epidemiologic studies showed an increase of 30 to 50% in adverse health outcomes in occupants because of dampness and mold exposure in the home.⁴ A damp indoor environment is linked to a factor in asthma development.⁵ Work in asthmatic populations in inner-city environments indicated an

increased number of homes with issues, including mold and moisture.⁶

Fungi represent a diverse group of eukaryotic organisms within their own kingdom. Fungi are identified primarily by their method of spore production and spore morphology. Spore production is controlled by internal factors but is impacted by the environment. The outdoors' environmental effect on fungal spore production has been well studied.⁷ Weather conditions, such as rain, humidity, wind speed and direction, temperature, and sunshine, all have varying effects on fungal aeroallergens.⁸ Fungi can have seasonal patterns that coincide with the host's ability to colonize or respond to temperature and moisture.⁷ But, although temperature, dew point, and air pressure have all been shown to positively correlate with spore concentrations, no single meteorologic parameter can predict the appearance of airborne spores.^{9,10}

Fungi have adapted to grow on a wide variety of materials and are often seen indoors. A study by Adams *et al.*¹¹ showed that fungal spores collected from passively settled dust did not differ among rooms despite differences in room use, water sources, and potential substrates for fungal growth. A separate study by Adams *et al.*,¹² on the fungal profile in a healthy building, indicated that aerosolization of particular fungi from growth on indoor surfaces is only minor in comparison with the fungal contribution from outdoor air. Indoor environments are often climate controlled,

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and a correlation between indoor and outdoor airborne fungi has not been as thoroughly studied. Our study was designed to examine common fungal taxa and to compare data from indoor and outdoor airborne spore evaluations in relation to various seasonal and environmental factors in the Kansas City, Missouri, metropolitan area. The aim of the study was to determine the relationship of indoor fungal spore levels, outdoor ground-level fungal spore levels, and outdoor spore levels at a central collection point in the Kansas City metropolitan area. Our hypothesis was that indoor mold spore levels correlate with outdoor mold spore levels for commonly evaluated taxa.

METHODS

Outdoor regional spore data for selected days were retrieved from the Kansas City mold and spore data base. This data base contains 19 years of data collected according to the specifications of the National Allergy Bureau of the American Academy for Allergy Asthma and Immunology.¹³ Indoor and outdoor ground-level data were retrieved from air samples of families who participated in one of two Kansas City Safe and Healthy Housing research projects.^{14,15} Households in these studies contained at least three persons, with at least one person between 2 and 17 years of age. All the households were in the Kansas City area, and all the families had lived in the same home, whether rented or owned, for at least 2 months before sampling. The demographics of these homes was previously reported.¹⁶ All human subjects research was approved by the Children's Mercy Hospital Institution Review Board.

The homes for this study were chosen as a convenience sample of the >400 homes that participated in these projects. Air sampling was conducted in at least four locations in the homes included in this study. Areas sampled included the main living space, the bedroom of the enrolled child, the basement, and outdoors separately by using an Allergenco MK3 (EMS Sales, North Charleston, SC) or a Buck Bioaire sampler (A. P. Buck Inc., Orlando, FL). All the air samples were taken for 10 minutes with an inlet velocity of 15 L/min. The ground-level outdoor air samples were collected at 3 feet above the ground and at a distance of at least 10 feet from the house or apartment, with the preferred location being on the sidewalk in front of the structure.¹⁶ All the air collections were examined and enumerated microscopically, and airborne spore estimates per cubic meter of air were reported for total fungal spores and for 11 identifiable spore groups.¹⁶ *Alternaria*, undifferentiated ascospores, *Aspergillus-Penicillium*, undifferentiated basidiospores, *Chaetomium*, *Cladosporium*, *Curvularia*, *Epicoccum*, *Pithomyces*, smuts and/or myxomycetes, and *Stachybotrys*. Simple statis-

Table 1

Description of Indoor Collections	No.
Total homes	244
Subject bedrooms	244
Bathrooms	211
Family or living rooms	239
Kitchens	221
Basements	232
Other rooms	157
Total rooms	1304

tics were compiled on Excel (Microsoft Corp., Redmond, WA), and Spearman rank correlation coefficients (used because the spore data were not normally distributed) were calculated by using SPSS 18 (IBM Corporation, Chicago, IL).

RESULTS

The basic profile of the data is displayed in Table 1. The homes were typical of the North American Midwest. The characteristics of these homes were reported previously.¹⁵ Briefly, these homes were all one- or two-story wood frame construction, typically over a basement. The homes had natural gas forced air heating, and all had some form of cooling, either by a window unit or a central unit integrated into the heating, ventilation, and air conditioning system. The location and the socioeconomic levels of the occupants of these homes varied, with some homes located in upscale suburban developments and some homes located in blighted urban settings. The total number of homes included in the study was 244. They were evaluated over a time interval of 8 years, from December 2003 until January of 2012. The homes were visited throughout the year, with the fewest homes visited in June (14) and the most homes visited in September (35). Overall, 132 inspections were performed in the heating season (November-April) and 112 during the cooling season (May-October). The overall mean total spore count per home taken indoors was 4076 spores/m³ and taken outdoors at ground level was 8899 spores/m³, which is marginally higher than the mean outdoor central spore count of 8342 spores/m³. Data ranges were from 0 to 64,010 total spores/m³ indoors, with a median of 2347 total spores/m³. Outdoor ground-level counts (Table 2) ranged from 0 to 92,456 spores/m³, with a median of 6584 total spores/m³, and regional outdoor counts ranged from 0 to 39,833 spores/m³, with a median of 7358 spores/m³.

The Spearman correlation chart for the relationship between taxa in the indoor collection is presented in Table 3. Basically, all of the major indoor taxa listed on the chart were strongly correlated with the mean total

Table 2 Mean and median spore values for each collection type

	Total Spores for Indoor Collections		Mean Total Spores for Ground-Level Collections		Mean Total Spores for Outdoor Central Collections	
	Mean	Median	Mean	Median	Mean	Median
Overall (Jan-Dec)	4076	1309	8899	6584	8322	7358
Heating season (Nov-Mar)	3901	657	2740	590	2601	1847
Cooling season (May-Oct)	4093	1353	11903	8443	11056	9278

spores present in the home. The correlations were slightly lower between genera expected to be found mostly indoors, such as *Chaetomium* and *Stachybotrys*, and major outdoor genera, such as *Alternaria* and *Cladosporium*. But, generally, correlations were strong even between *Alternaria* and *Cladosporium* and indoor *Aspergillus-Penicillium* levels. *Aspergillus-Penicillium*, *Cladosporium*, and undifferentiated *Ascospore* are all very strongly correlated with the mean total spore level for rooms in the home. The equivalent correlation chart for outdoor ground level taxa (Table 4) indicated that *Aspergillus-Penicillium* spores were poorly correlated with major outdoor taxa, such as undifferentiated ascospores and *Cladosporium*, and only moderately correlated with the overall spore count. The total outdoor ground level spore levels were highly correlated with levels of major outdoor taxa such as *Ascospore* and *Cladosporium*, as expected.

The Spearman rank correlations of indoor spore levels with outdoor spore levels are presented in Table 5. The correlations were strong for most major outdoor taxa. *Cladosporium*, basidiospores, ascospores, *Alternaria*, *Pithomyces*, *Epicoccum*, and *Curvularia* levels in indoor and outdoor air were all strongly correlated. Dominant outdoor taxa, such as *Cladosporium* and undifferentiated ascospores, showed the greatest correlation of the taxa represented. Two major taxa associated with the indoor environment (*Aspergillus-Penicillium* and *Chaetomium*), were significantly correlated between indoor and local ground-level outdoor air. Only *Stachybotrys* and smuts and/or myxomycetes were not well correlated between outdoor and indoor collections. For the comparison of indoor spore levels with the regional outdoor spore levels taken at a central location, the correlations were much the same as for the outdoor collections taken at the home, the exceptions being that *Aspergillus-Penicillium* and *Curvularia* spores were not as well correlated, and that *Stachybotrys* and *Chaetomium* were so rarely seen in these central outdoor collections that they were not evaluated. Nearly all of the fungal taxa analyzed in the comparison of outdoor regional spore levels and local ground-level spores were strongly correlated (Table 5). The exception being the correlation for *Aspergillus-Penicillium* and *Curvu-*

laria; the correlations for *Chaetomium* and *Stachybotrys* are not presented in the table.

DISCUSSION

Historically, indoor spore levels have been evaluated with consideration of outdoor spore levels at the same location.¹⁷ We observed (Table 3) that all major indoor fungi correlated with each other and with the total spore count, which indicated that these fungi grew under similar conditions. Significant taxa that dominated the total spore level when elevated, such as *Aspergillus-Penicillium*, *Cladosporium*, and undifferentiated ascospores, were all correlated with mean total spore counts when seen indoors. The correlation chart for indoor taxa (Table 3) was contrasted with the correlation chart for outdoor taxa (Table 4), in which positive correlations were typically slightly higher. Also, major genera associated with the indoors, such as *Chaetomium* and *Stachybotrys*, were not correlated with either total spore levels or major outdoor taxa because they were infrequently observed in the outdoor collections.

In general, indoor spore counts showed a strong correlation with both outdoor ground-level counts and outdoor regional counts (Table 5). The major spore group associated with the indoor environment, *Aspergillus-Penicillium*, was not as strongly correlated with outdoor air as the dominant outdoor taxa such as *Cladosporium* and ascospores. *Aspergillus-Penicillium* as a spore type was even negatively correlated between indoor levels and regional outdoor levels collected at a remote site, whereas it was positively correlated with outdoor ground-level collections at the home site. *Stachybotrys* and *Chaetomium* were not as highly correlated between outdoor and indoor collections, likely due to competition for growth resources of other, more dominant, fungi. *Chaetomium* and *Stachybotrys* were not represented in regional counts because they were rarely seen in the central outdoor collections and were not specifically tabulated.

Our results were mostly consistent with Adams *et al.*,¹² who showed, by using microbiome methods, that indoor fungal communities correlated with outdoor

Table 3 Spearman correlation between indoor spore taxa levels

	Ascomycetes	Aspergillus- Penicillium	Basidiospores	Chaetomium	Cladosporium	Curcularia	Epicoccum	Pithomyces	Smuts and/or Myxomycetes	Stachybotrys	Mean Total Spores
<i>Alternaria</i>	0.32**	0.31**	0.43**	0.02	0.53**	0.28**	0.33**	0.39**	0.32**	-0.1	0.42**
Ascomycetes	1	0.47**	0.53**	0.1	0.53**	0.2**	0.08	0.25**	0.23**	0.13*	0.65**
<i>Aspergillus- Penicillium</i>	1	1	0.35**	0.32**	0.44**	0.13*	0.15*	0.22**	0.32**	0.25**	0.79**
Basidiospores			1	0.09	0.63**	0.18**	0.26**	0.36**	0.28**	0.13*	0.52**
<i>Chaetomium</i>			1	1	0.12	0.04	0.15*	0.08	.30**	0.36**	0.28**
<i>Cladosporium</i>					1	0.15*	0.26**	0.26**	.039**	0.08	0.69**
<i>Curcularia</i>						1	0.18**	0.39**	0.11	0.12	0.20*
<i>Epicoccum</i>							1	0.32**	0.33**	0.1	0.23**
<i>Pithomyces</i>								1	0.25**	0.09	0.27**
Smuts and/or myxomycetes									1	0.24**	0.40**
<i>Stachybotrys</i>										1	0.26**
Total spores											1

*Significant at <0.05.

**Significant at <0.01.

Table 4 Spearman correlation between outdoor ground-level spore taxa

	Ascomycetes	Aspergillus- Penicillium	Basidiospores	Chaetomium	Cladosporium	Curcularia	Epicoccum	Pithomyces	Smuts and/or Myxomycetes	Stachybotrys	Total Spores
<i>Alternaria</i>	0.57**	-0.01	0.58**	-0.13	0.75**	0.37**	0.45**	0.41**	0.47**	0.07	0.66**
Ascomycetes	1	0.17*	0.65**	-0.08	0.73**	0.29**	0.24**	0.29**	0.43**	0.06	0.86**
<i>Aspergillus- Penicillium</i>	1	1	0.12	0.01	0	0.06	0.06	0.15*	0.12	0.19**	0.15*
Basidiospores			1	-0.09	0.66**	0.22**	0.36**	0.38**	0.41**	0.1	0.67**
<i>Chaetomium</i>				1	-0.6	0.11	-0.01	-0.08	0.06	-0.02	-0.04
<i>Cladosporium</i>					1	0.38**	0.42**	0.42**	0.51**	-0.01	0.86**
<i>Curcularia</i>						1	0.29**	0.49**	0.30**	0.09	0.35**
<i>Epicoccum</i>							1	0.27**	0.37**	-0.06	0.35**
<i>Pithomyces</i>								1	0.31**	-0.04	0.39**
Smuts and/or myxomycetes									1	0.06	0.50**
<i>Stachybotrys</i>										1	0.05
Total spores											1

*Significant at <0.05.

**Significant at <0.01.

Table 5 Spearman correlation tables for indoor, ground-level outdoor and regional outdoor collections

	Spearman Correlation between Indoor and Ground-Level Outdoor Spores for Each Taxon	Spearman Correlation between Indoor and Regional Outdoor Spores for Each Taxon	Spearman Correlation between Regional Outdoor and Ground-Level Outdoor Spores for Each Taxon
<i>Alternaria</i>	0.47**	0.43**	0.55**
Ascospores	0.48**	0.32**	0.52**
<i>Aspergillus-Penicillium</i>	0.21**	-0.17	0.07
Basidiospores	0.54**	0.49**	0.65**
<i>Chaetomium</i>	0.22**	Not observed	Not observed
<i>Cladosporium</i>	0.56**	0.38**	0.70**
<i>Curvularia</i>	0.38**	0.07	0.12
<i>Epicoccum</i>	0.40**	0.42**	0.35**
<i>Pithomyces</i>	0.46**	0.27**	0.28**
Smuts and/or myxomycetes	0.44**	0.24**	0.35**
<i>Stachybotrys</i>	0.06	Not observed	Not observed
Total spores	0.42**	0.25**	0.66**

*Significant at <0.05 .**Significant at <0.01 .

communities and that indoor fungal community variation was not significantly different from outdoor community variation.¹² These findings were also consistent with Lee *et al.*,¹⁸ who showed significant positive correlations between indoor and outdoor concentrations of culturable and total fungal spores and with Sterling and Lewis¹⁹ who found similar correlations in trailers in the Southwest. Lee *et al.*,¹⁸ also noted a higher indoor mean spore level than outdoor mean spore level which they believed was due to a more favorable survival environment for culturable fungi indoors; however, in a separate study, they showed total counts that were higher outdoors than indoors.²⁰ We noted a similar trend of higher mean total spore levels when comparing outdoor ground-level counts with indoor counts. We also noted total higher outdoor counts regionally in comparison with indoor counts, which may be due to availability of resources outdoors.

The homes we evaluated were a cross section of homes in our community. Most were homes of children with asthma or other respiratory conditions. Analysis of our data showed ranges from 0 to 64,010 total spores/m³ indoors, with a median of 2347 total spores/m³. Outdoor ground-level counts ranged from 0 to 92,456 spores/m³, with a median of 6584 total spores/m³, and regional outdoor counts ranged from 0 to 39,833 spores/m³, with a median of 7358 spores/m³. In the study by Lee *et al.*,²⁰ they reported indoor ranges from 148 to 6302 spores/m³ and 405 to 7704 spores/m³ outdoors. They looked at the same six homes with no fungal issues over three seasons. Our average was over

an 8-year period. A study by Garrett *et al.*²¹ showed slightly higher indoor and outdoor total counts than in the study by Lee *et al.*²⁰

The most abundant indoor spore taxa found were *Aspergillus-Penicillium*, followed by *Cladosporium*, then ascospores; the most common outdoor ground spores were *Cladosporium*, followed by undifferentiated ascospores, *Aspergillus-Penicillium*, and smuts and/or myxomycetes, and the most-common central outdoor counts were ascospores, *Cladosporium*, and basidiospores. Lee *et al.*²⁰ showed *Aspergillus-Penicillium* as the most common indoor and outdoor spores seen, followed by undifferentiated ascospores, *Cladosporium*, and undifferentiated basidiospores. The comparison of outdoor regional spore counts with outdoor ground-level spore counts showed a strong correlation for most spores. *Aspergillus-Penicillium* and *Curvularia* did not show statistically significant correlations. It was interesting that these taxa showed a statistically significant but weak correlation to ground-level spore counts but not to regional counts. Given the sometimes high spore levels indoors, particularly with regard to *Aspergillus-Penicillium*, this may represent a spilling of indoor spores to the surrounding outdoor areas. Although further studies would need to be done to make this association.

Adams *et al.*¹² also showed that indoor and outdoor variation maintained a correlation despite changes in season. Our study did not look at seasonal species variation. Limitations of our study were that the study did not control for rainfall and that the number of

homes evaluated in the winter was not the same as the number of homes in the summer. The data in Table 2 reflected the strong differences in outdoor spore levels between the heating season and the cooling season. Another limitation was that these were all separate homes and the same home was not examined in the winter and in the summer. The difference in seasons and environments could have changed the growth of particular taxa. Also a limitation of our study was that the majority of these homes contained patients with atopy and at high risk, and this may have biased our data because these patients usually lived in low-middle-to-lower socioeconomic level urban homes. Further studies that control for these factors will help establish environmental roles of fungal growth, which will better allow us to change environments to limit growth for families, particularly those at high risk for respiratory disease.

CONCLUSION

In this group of 244 parallel spore collections from within homes, from the outside of homes, and at a central collection point taken over a period of 8 years, the spore populations were in general accord. Exceptions were noted for spores thought to be associated with the indoor environment. Generally, indoor spore collections reflected those spores found in the outdoor air. Therefore, when evaluating indoor spore levels for amplification, outdoor spore levels should be considered.

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