

IJBHS 2010212/7202

Assessment of fungi aeroallergens in homes of asthmatic patients in Benin City, Nigeria

E. A. Ophori

Department of Microbiology, Faculty of Life Sciences, University of Benin, P.M.B. 1154, Ugbowo, Benin City, Edo State, Nigeria

(Received December 3, 2010; Accepted February 12, 2011)

ABSTRACT: Atopic individuals have a tendency to suffer from one or more allergic disorders, such as asthma, which development appears to involve the interplay between host factors and environmental exposures. The results of a study of the fungi flora in indoor air of bedrooms of asthmatic patients are discussed. Samples were taken from 104 bedrooms of houses (homes) of asthmatic patients who have been diagnosed clinically and who were attending the Ear, Nose and Throat (E.N.T) Clinic of the University of Benin, Benin City. For each home, a set of 5 sterile Petri-dishes containing Sabouraud dextrose agar, malt extract agar amended with antibiotic mixture of streptomycin and penicillin were exposed for a period of 5-10 min. between 25°C - 28°C for 10 days. Fungi isolates were identified by standard methods. A total of 68 species, comprising of 7 fungal genera were isolated and identified in bedroom indoor air (cfu/m³). Fungi included *Cladosporium herbarium*, *Aspergillus* spp., *Penicillium oxalicum*, *Mucor* sp. and *Alternaria alternata*. In the houses of the asthmatic patients, *Cladosporium herbarium* had the highest percentage frequency of occurrence (29.69% ± 2.01%), followed by *Aspergillus niger* (20.31 ± 1.2%). The least fungus was *Alternaria alternata* (1.56 ± 1.12). Also, in the control group *Cladosporium herbarium* had the highest frequency of occurrence (34.8 ± 4.60%) while the least was *Helminthosporium* sp. (1.4 ± 1.1%). *A.niger* had the highest density of 10⁵ cfu/m³, followed by *Penicillium oxalicum* (10⁴ cfu/m³) in the asthmatic group. The least was also *Alternaria alternata*. The prevalence of these fungi in bedrooms is an indicator of the probable high level of allergens produced and released by these fungi and may be related to allergic symptoms observed in some inhabitants of these houses. Therefore procedures to reduce possible fungal allergens to as low as possible by way of regular cleaning of the home may be a feasible way to preventing continuous and regular fungal allergy.

Keywords: Atopy, allergens, asthma, fungi, Nigeria.

Introduction

Dust is a general term denoting solid particles of different dimensions and origin, which may generally remain in suspension for some time. It occurs in the atmosphere both naturally and as a result of man's activities. House dust is a poorly defined mixture of allergens of varying complexity and antigenicity. Its composition in relation to individual allergens vary both quantitatively and qualitatively from one location to another, thus making precise characterization of its component antigens difficult(1). Rather than a single substance, house dust contains fibres from different types of fabrics, cotton lint, feathers, and danders from dogs, cats and other animals(2), bacteria(3,4,5,6,7,8), fungi (9,10,11,12), plants(13), cockroaches (14) and house dust mites (15):

*To whom correspondence should be addressed.

email eaophori@yahoo.com.

Tel: +2348023393102

There is a growing alarm about the dangerous influence of atmospheric contamination with microbial propagules. Different airborne microorganisms occur in dust and could be frequent cause of allergic respiratory disease, which are common in workers exposed professionally to dust. Varieties of allergens are associated with airborne particles in homes and work environment. If sufficient levels of allergens are airborne at least sporadically during an extended period of time, individuals prone to allergy may become sensitized (10,16)

Airborne fungi may cause infectious diseases, allergic and hypersensitivity reactions, and contribute to the adverse health effects called 'sick building syndrome' caused by *Trichoderma harzianum* Rifai which has also been reported to cause asthma, sinusitis and paralysis of sperm cell motility of boar (17). Common indoor air fungi genera with species of high allergenicity that are associated with hypersensitivity are *Alternaria*, *Aspergillus*, *Aureobasidium*, *Cladosporium* and *Penicillium* (18,19,20). In Nigeria, species of *Aspergillus*, *Penicillium*, *Fusarium*, *Mucor* and *Candida* have been reported (21,22)

The aim of this study was to evaluate the prevalence of fungi aeroallergens in houses of asthmatic patients who have been clinically diagnosed.

Materials and Methods

Patients: Asthma was diagnosed in patients attending the Ear, Nose and Throat (E.N.T) Clinic of the University of Benin Teaching Hospital, Benin City, Nigeria according to predictive index and asthma based on medical history manifestation and pulmonary function test by a trained physician. A total of 104 asthmatic patients aged 6-50 years (mean 26.24± 6.01) who gave informed consent were enrolled in the study. Fifty individuals with no symptoms of any allergic disease as part of their medical history were used as control group.

The study had the approval of the Ethics Committee of University of Benin Teaching Hospital, Benin City. Patients had resided in sampled houses for at least 1 yr before the study.

Sampling of homes and fungi culture

Samples were taken from 104 non-air conditioned houses of asthmatic patients and the control group in areas in Benin City. Samples were taken after personal communication with each resident, from their bedrooms (as this is where most people spend approximately 6-9 hr/day). Householders were requested not to do extra cleaning but to follow their normal routine before each visit as described (21).

The airborne fungal spores were collected by sedimentation method (23,24). For each home, a set of 5 sterile Petri dishes containing Sabouraud dextrose agar, malt extract agar amended with antibiotic mixture of streptomycin and penicillin were exposed for a period of 5-10 min(25). The Samples were caught 1.5 m above the floors of the bedroom(12). Plates were incubated at 28 °C for a period of 10 days. Plates were examined periodically. The fungal growths were identified using standard procedure based upon gross cultural and microscopic morphology(26)

Statistics: SPSS 10.0 for windows was used for the statistical analyses. P –values < 0.05 were accepted as significant

Results

The fungal species and their frequency of occurrence in the homes of the asthmatic patients and the control group are shown in Table 1. A total of 68 species, comprising of 7 fungal genera were isolated and identified in all houses. It was observed that the frequency of occurrence (32.5%) of fungal spores in the bedrooms of male inhabitants was less than that of female inhabitants (67.5 %), but this was not statistically significant (p>0.05).

The densities of fungi in indoor air are shown in Table 2. *Aspergillus niger* had the highest density (10^5 cfu / m³) in all the homes of asthmatic patients sampled. This was followed by *Penicillium oxalicum* (10^4 cfu / m³). The least density was recorded with *Alternaria alternata* (10cfu / m³). It was observed that patients that lived in houses with more fungi density/species had regular asthmatic allergy than those with less density/species from their medical records. All the fungal species identified in the homes of the asthmatic patients were also identified in the houses of

the control group, apart from *Helminthosporium* sp. which was not identified in the former group (table 2). The densities of fungal species in the houses of the control group were higher. *A. niger* and *P. oxalicum* had the highest density (10^8 cfu/m³ each) compared to the 10^5 and 10^4 cfu/m³ respectively recorded in the asthmatic group. This was statistically significant ($p < 0.05$)

Table 1: Fungal genera and their percentage frequency of occurrence in houses of asthmatics and control group

Organisms	% Frequency of occurrence	
	Asthmatic Patients (n = 104)	Control Group (n = 50)
<i>Cladosporium herbarium</i>	29.69 ± 4.10	34.8 ± 4.60
<i>Aspergillus niger</i>	20.31 ± 3.82	26.0 ± 3.21
<i>Penicillium oxalicum</i>	17.19 ± 3.46	10.0 ± 2.60
<i>Aspergillus fumigatus</i>	12.5 ± 2.60	13.5 ± 2.10
<i>Rhizopus</i> spp.	9.37 ± 1.22	7.4 ± 1.60
<i>Mucor</i> spp.	6.25 ± 2.24	4.8 ± 2.42
<i>Candida albicans</i>	6.25 ± 2.24	-
<i>Alternaria alternate</i>	4.41 ± 2.48	2.1 ± 0.11
<i>Helminthosporium</i> sp.	0.0	1.4 ± 1.1

Table 2: Fungal genera isolated from bedroom indoor air and their densities in houses of asthmatic patients and in control group

Organisms	Approximate density in indoor air (cfu/m ³)	
	Asthmatic Patients (n = 104)	Control Group (n = 50)
<i>Aspergillus niger</i>	10^5	10^8
<i>Penicillium oxalicum</i>	10^4	10^8
<i>Aspergillus fumigatus</i>	10^4	10^7
<i>Cladosporium herbarium</i>	10^3	10^3
<i>Rhizopus</i> spp.	10^3	10^5
<i>Candida albicans</i>	10^2	10^5
<i>Mucor</i> spp.	10^2	10^4
<i>Trichoderma viridae</i>	10^2	10^2
<i>Alternaria alternate</i>	10	10^3
<i>Helminthosporium</i> sp.	-	10^2

Discussion

Allergy to environmental factors has become an important health concern in Nigeria. In our study, all the houses of asthmatic patients and the control group sampled showed the presence of similar fungi species with *A.niger* and *P.oxalicum* having the highest densities in homes of asthmatic patients (10^5 and 10^4 cfu/m³ respectively) and in control group (10^8 cfu/m³ each). All the houses had fungi growth, people are therefore likely to come in contact with these fungi and aggravate health situations of allergic individuals. A high incidence of *A.niger* (65.9%) and *P.oxalicum* (19.0%) from indoor air of living rooms of individuals living in Benin City, Nigeria had been reported²¹. *A.niger* has been reported to be an imported human health risk for those who are susceptible to infection or allergy (12,27,28). The high densities of fungi in indoor air can be attributed to the mixture of those which have entered from outdoors and those which readily grow and multiply.

In the present study, species of *Aspergillus*, *Penicillium* and *Cladosporium* were the most common fungi in indoor air of bedrooms of both asthmatic and control group. Exposure to *Aspergillus* spp. has been reported to cause several types of human health problems such as allergies, infection and toxic effects (12). The results of this study showed that *Cladosporium herbarium* was most prevalent (29.69±4.10%). Several studies have reported this fungus as the most abundant genera identified from both indoor and outdoor samples^{29,30,31}. This is in line with the result of this study. However, it was the least fungus reported in indoor of living rooms during the rainy and dry seasons in Nigeria (0.43 and 0.0% respectively) (21). *Aspergillus* spp. has been reported as the most abundant fungi in indoor air of living rooms (22,23). The result of the present study is in contrast to these earlier findings.

Alternaria alternata was the least fungus in houses of asthmatic patients (4.41± 2.48 %). This is contrary to some studies where it has been reported to be high in indoor (12,32). This fungus is associated with asthma and other indoor allergic diseases (11,12,33). One of the major findings of this study is the presence of *Helminthosporium* sp. which was isolated in houses of the control group individuals. It has been implicated in allergies and several infections³⁴. Within the study area, this fungus has not been reported in indoor air in houses.

Interestingly, the houses of asthmatic patients had lower fungal densities than those of the control group. This is perhaps due to the intense house cleaning by the patients as part of the advice given to them by their physicians to do regular cleaning so as to reduce environmental triggers of asthma. From the results obtained here, it can be concluded that there is a high incidence of airborne fungal spores in bedroom of both asthmatic and control individuals and there is a similarity in the species composition of indoor fungi in Benin City, Nigeria. The potential human health effects of these fungi have been well documented. We suggest that public enlightenment programmes and the use of electronic / print media to educate the general public should be intensified so as to adequately clean our environment to reduce fungal spores and fungi allergy.

ACKNOWLEDGEMENT: We sincerely thank all those who gave informed consent to participate in this study. We are also grateful to the Staff and Management of University of Benin Teaching Hospital, Benin City.

References

- Twarog FJ, Picone FJ, Strunk RS, So J, Collen HR. Immediate hypersensitivity to cockroach. Isolation and purification of the major antigens. *J Allergy Clin Immunol* 1976;**59**(2):154-160.
- Schou C. Defining allergens of mammalian origin. *Clin Exp Allergy*. 1993; **23**: 7 – 14
- Olenchock S, May J, Pratt D, Piacelli L. Presence of endotoxin in different agricultural environments. *Am J Ind Med*. 1990;**18**:279- 284.
- Dutkiewicz J, Tucker J, Burrell R. Ultrastructure of the endotoxin produced by Gram negative bacteria associated with organic dusts. *System Appl Microbiol*. 1992; **15**:474-485.
- Donham KJ (1994). Swine confinement buildings. In: *Organic Dusts, Exposure, Effects and Prevention* Eds R Raylander and R Jacobs. Lewis Publishers, London pp 438-356.
- Andersson M, Laukkanen M, Nurmiäho-Lassila EL, Rainey F, Neimela S, Salkinoja-Salonen M. *Bacillus thermosphaericus* sp.nov. a new thermophilic ureolytic *Bacillus* isolated from air. *System Appl Biochem* 1995, **23**:13-18.
- Rylander R. Microbial cell Wall constituents in indoor air and their relation to diseases. *Indoor Air*. 1998, **4**:59-65
- Andersson AM, Weiss N, Rainey F, Salkinoja-Salonen MS. Dust borne bacteria in animal sheds, schools and children's day care centers. *J Appl Microbiol*. 1999; **86**:622-634.
- Pauli G, Lemuo J, Hoyet C. Tools for the study of household allergens. *Presse Med*. 1989; **18** (41): 2015 - 2017
- Tovey ER. (1997). Environmental control In: Eds PJ Barnes, MM Grunstein, AR Leff, AJ Woolcock. *Asthma* Lippincott - Raven Publishers, Philadelphia. pp 1883 - 1904

- Cetinkaya Z, Fidan F, Unlu M, Hasenekoglu I, Tetik L, Demirel R. Assessment of indoor air Fungi in Western-Anatolia, Turkey, *Asian Pacific J Allergy Immunol*, 2005; **32(2-3)**, 87-92.
- Hedayati MT, Mayahi S, Denning DW. A study on *Aspergillus* species in houses of asthmatic patients from Sari City, Iran and a brief review of the health effects of exposure to *Aspergillus*. *Environ Monit Assess. DOI*: 2009; **10**:1-5
- Middleton Jr., E.J., Reed CE, Ellis EF, Adkinson F Jr, Yunginger JW, Busse WWC. (1999) *Allergy: Principles and Practice* (5th edn). Academic Press Limited NY.
- Lehrer SB, Elliott Horner W, Menon PK, Oliver J Hauck P. Cockroach allergenic activity: Analysis of commercial cockroach and dust extracts. *J Allergy Clin Immunol* 1991; **88**: 895-901.
- Colloff MJ, Spiekma F. Th M. Pictorial keys for the identification of domestic mites. *Clin Exper Allergy*. 1992; **22**: 823 – 830.
- Ayanru DKG. (1981). Agricultural and clinical significance of some microbial components of the air spora in the Benin- City area. In: *Global Impact of Applied Microbiology* (Eds) SO Emejuaiwe, O Ogunbi, SO Sanni .Academic Press 6th edn London pp107-117
- Peltola J, Andersson M, Haahtela T, Mussalo-Rauhamaa H, Rainery FA, Kroppenstedt RM, Samson RA, Salkinoja-Salonen MJ. Toxic metabolite - producing bacteria and fungus in an indoor environment. *Appl Environ. Microbiol* 2001; **67(7)**: 3269-3274.
- Crow SA, Ahearn DG, Noble JA, Moyenuddin M, Price DL. Microbial ecology of buildings: Effect of fungi on indoor air quality. *Feature Article* 2001; **1**:15-16.
- Lopez M, Salvaggio J, Butcher B. Allergenicity of basidiomycetes *J Allergy Clin Immunol* 1976; **57**:480-485.
- Salvaggio JE, Buge HA, Chapman JA. Emerging concepts in mould allergy. What is the role of immunotherapy? *J Allergy Clin Immunol* 1993; **92(2)**:217-222.
- Ophori EA, Ogbimi AO. Indoor viable dust bound microfungi in residential homes in Benin-City Nigeria. *Tropical J Environ Sc Health*. 1999; **2(1)**:20-23.
- Ophori EA, Ogbimi AO. Indoor viable dust bound microfungi in residential homes in Benin-City Nigeria. *Scientia Africana*. 2004; **3(2)**:67-71.
- Cosentino S, Palmas F. Occurrence of Fungal spores in the respiratory tract and homes of patients with positive skin test to fungi. *Aerobiologia*, 1996; **12**:155-160.
- Yazicioglu M, Asan A, Ones U, Vatansver U, Sen B, Ture M. Indoor airborne fungal spores and home characteristic in asthmatic children from Edirne region of Turkey. *Allergologia et Immunopathologia* (Madrid, Spain) 2004; **32(4)**, 197-203
- Leung R, Lam CKW. The importance of domestic allergens in a tropical environment. *Clin Exp Allergy* 1997, **27**: 856-859.
- Rippon JW. (1974) *Medical Mycology. The Pathogenic Fungi and Pathogenic Actinomycetes*. W. B. Saunders, London.
- McGinnis MR. Pathogenesis of indoor fungal diseases, *Medical Mycology* 2004; **42**:107-117.
- Denning DW. Invasive aspergillosis. *Clin Infect Dis* 1998; **26**:781-803
- Ebner MR, Haselwandter K, Frank A. Indoor and outdoor incidence of airborne fungal allergens at low and high altitude alpine environments. *Mycoses* 2003; **36**:69-73.
- Macher JM, Huang FY, Flores M. A two-year study of microbiological indoor air quality in a new apartment. *Arch. Environ Health* 1991; **46**:25-29.
- Solomon WR Assessing fungus prevalence in domestic interiors. *J. Allergy Clin. Immunol.* 1975; **56**:235-42.
- Su HJ, Wu PC, Chen HL, Lee FC, Lin LL. Exposure assessment of indoor allergens, endotoxin, and airborne fungi for homes in southern Taiwan. *Environ Research Section* 2001; **85**:135-44.
- Sanchez H, Bush RK. A review of *Alternaria alternata* sensitivity. *Review of Iberoam Micol.* 2001; **18**:56-59
- Goldfarb AA (1968). The significance of mold spore allergy in childhood respiratory disease. *Ann. Allergy*. 26:321-325