Sensitivity to Moulds
In Egyptian Asthmatic Children

Thesis

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List of Abbreviations

ABP A	Allergic bronchopulmonary aspergillosis
ABPM	Allergic bronchopulmonary mycosis
AD	Atopic dermatitis
AFS	Allergic Fungal Sinusitis
AIDs	Acquired immunodeficiency syndrome
CDC	Centers for Disease Control and Prevention
CT	Computed tomography
CXR	Chest X-ray
DNA	Deoxyribonucleic acid
EAA	Extrinsic allergic alveoli
ECRHS	European Community Respiratory Health Survey
ELISA	Enzyme-linked immunosorbent assay
ER	Endoplasmic reticulum
GM	Galactomannan
HP	Hypersensitivity pneumonitis
IA	Invasive Asperagillus
ICS	Inhaled corticosteroids
IDT	Intradermal test
IFN-γ	Interferon- γ
IgE	Immunoglobulin E
IgG	Immunoglobulin G
IOM	Institute of Medicine
LABA	Long-acting inhaled β-agonists
LPR	Late-phase reaction
MVOCs	Microbial volatile organic compounds
PATY	Pollution and the Young study
RAST	Radioallergosorbent testing
SAFS	Severe asthma with fungal sensitization
SD	Standard deviation
SPT	Skin prick test
UV	Ultraviolet rays
Th2	T-helper cell type 2

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INTRODUCTION

Most asthma patients have mild symptoms which are well controlled with anti-inflammatory and bronchodilator therapy but a minority of asthma patients has severe airway inflammation and airflow obstruction requiring multiple hospital admissions ***(O'Driscoll et al., 2005).***

Sensitization to airborne allergens might be involved in the underlying mechanisms of severity. Sensitization to moulds has been suggested as a risk factor for life threatening asthma ***(Zureik et al., 2002).*** It was reported that 20 of 37 (54%) patients admitted to an intensive care unit for asthma had a positive result on skin testing for one or more fungal allergens ***(Black et al.,2000).***

Filamentous microfungi (moulds) can threaten human health through release of spores that become airborne and can be inhaled. Some moulds produce metabolites (mycotoxins) that can initiate a toxic response in humans or other vertebrates ***(Robbins et al., 2000).***

Depending on patients' geographic locations, their mold allergies can lead to seasonal or perennial clinical disorders. Perennial mold allergies are particularly prevalent, both in consistently humid and warm climates. Of the thousands of types of fungal spores found in indoor and outdoor environments, adverse health effects in humans have most frequently been associated with *Alternaria*, *Aspergillus*, *Cladosporium*, and *Penicillium* ***(Stark et al., 2003; Hossain et al., 2004; Jarvis & Miller, 2005 and O'Driscoll et al., 2005).***

The health effects of exposure to mold in the indoor environment have been extensively studied ***(Belanger et al., 2003; Portnoy et al., 2005).*** Among children living in the southern United States who are not yet school-aged and develop a confirmed allergy to indoor allergens, 80% have sensitivity to mould spores. An Institute of Medicine (IOM) committee concluded in 2004 that there is sufficient evidence of a causal link between indoor dampness and upper respiratory tract symptoms, cough, wheeze, and asthma symptoms in sensitized people ***(IOM, 2004).***

The panel further concluded that there is suggestive evidence of an association between damp indoor environments and dyspnea, lower respiratory illness in healthy children, and new-onset asthma. A large population-based prospective cohort study found that the presence of mold odor in the home was associated with a 2.4-fold increased incidence rate of asthma among children ***(Jaakkola et al., 2005).***

Although indoor fungal allergen exposure occurs, outdoor exposure is generally more relevant in terms of sensitization and disease expression ***(Bush et al., 2006).***

A large Canadian time-series study reported that daily fluctuations in ambient mold spores are directly associated with childhood asthma attacks requiring a visit to an emergency department (***Dales et al., 2004).*** Researchers in Southern California reported an association between ambient mold spore concentrations and childhood asthma attacks even in areas where the airborne spore concentrations are relatively low ***(Delfino et al., 1997).***

Aim of the work:

There are no published data on the rates of mould sensitization in Egypt. The aim of this pilot study is to screen for mould sensitivity by skin prick testing in a group of Egyptian asthmatic children in relation to clinical severity in a trial to evaluate its importance as an allergen in our community.

Mould

Definition:

Moulds (fungi) are eukaryotic non-chlorophyll ***(Terr, 2009)*** filamentous, and mostly spore-bearing organisms, which exist as saprophytes or as parasites of animals and plants ***(Kurup et al., 2000)***. It has been estimated that moulds have existed for more than 500 million years and currently occupy 25% of the earth's biomass. Moulds can be found in any building, and their airborne spores are easily detected indoors as well as outdoors ***(Terr, 2009)***.

It is estimated that more than a million species of fungi exist, and many of these produce airborne spores, conidia, hyphae or other fragments that can be inhaled by humans. Fungal conidia and their associated components are unlike any other bioaerosol in that they are heterogeneous, biologically dynamic particles and are actively able to secrete molecules that have a variety of pathogenic, inflammatory and allergic properties ***(Green et al., 2006).***

Taxonomic Classification:

There are four major phyla of fungi; each is extremely diverse, and fungi are assigned to a phylum on the basis of their mechanism for producing asexual spores. These phyla are the Chytridiomycota (primitive aquatic fungi), Zygomycota (e.g., black bread mold, *Rhizopus nigricans*), Ascomycota (sac fungi, e.g., *Saccharomyces, Candida, Aspergillus, Neurospora*, and morel mushrooms), and Basidiomycota (e.g., mushrooms, rot fungi, and puffballs) ***(Varki et al., 2009).***

In considering known and potential allergens, 5 major classes of fungi have particular clinical significance: Oomycetes, Zygomycetes, Ascomycetes, Basidiomycetes, and Deuteromycetes (Table 1). The most common and best described mold allergen sources belong to the taxonomic group Fungi Imperfecti (Deuteromycetes; asexual stages of Ascomycetes), which includes *Alternaria, Cladosporium, Aspergillus* ***(Huang, 2006)*** and *Penicillium* ***(Helbling and Reimers, 2003).***

Table 1: Fungi associated with IgE-mediated allergy.

Zygomycetes	Ascomycetes	Basidiomycetes	Deuteromycetes (fungi imperfecti)		Yeasts
Mucor	*Chaetomium*	*Agaricus*	*Acremonium*	*Helminthosporium*	*Saccharomyces*
Rhizopus	*Claviceps*	*Boletus*	*Alternaria*	*Neurospora*	*Candida*
	Daldinia	*Calvatia*	*Aspergillus*	*Nigrospora*	*Rhodotorula*
	Didymella	*Coprinus*	*Aureobasidium*	*Paecilomyces*	
	Erysiphe	*Ganoderma*	*Botrytis*	*Penicillium*	
	Eurotium	*Lentinus*	*Cephalosporium*	*Phoma*	
	Microsphaera	*Merulius*	*Chrysosporium*	*Scopulariopsis*	
		Pleurotus	*Cladosporium*	*Stachybotrys*	
		Psilocybe	*Coniosporium*	*Stemphylium*	
		Puccinia	*Curvularia*	*Torula*	
		Tilletia	*Cylindrocarpon*	*Trichoderma*	
		Urocystis	*Drechslera*	*Trichophyton*	
		Ustilago	*Epicoccum*	*Trichothecium*	
			Fusarium	*Ulocladium*	
			Gliocladium	*Wallemia*	

(Kurup, 2003).

Prevalence of mould contamination:

Reported prevalence rates of home mould/dampness range widely around the world: from 10–13% in Russia and Taiwan, to 15% in United Kingdom, Finland, and Poland, to 25% in the Netherlands, to 27–28% in Sweden, to 38% in China and Canada, and up to 50% in the USA. The prevalence rate of reported mould/dampness in Italy was low (mean 10%) with a significantly lower prevalence of mould/dampness in southern (7%) than in northern/central Italy (12%) ***(Simoni et al., 2005).***

A study performed in the Netherlands reported that the prevalence of indoor dampness and mould growth in the homes of Dutch (24%) was more or less comparable with the prevalence found in other European studies ***(Hagmolen of ten Have et al., 2007)***.

In the European Community Respiratory Health Survey (ECRHS), adult asthmatics were questioned on water damage, water on the basement floor, and prevalence of mould in the home; the prevalence was 12%, 2%, and 22%, respectively ***(Zock et al., 2002).*** It has been estimated that 20% to 40% of homes in Northern Europe and Canada have mold contamination. A study in the USA showed that *Aspergillus, Penicillium,* and *Cladosporium* spores were the most common types of fungi found ***(Niemeier et al., 2006).***

After Hurricane Katrina and Rita had struck New

Orleans, Louisiana, USA, a survey by the Centers for Disease Control and Prevention (CDC) found that 46% of randomly selected homes had visible mold growth, and 17% had heavy mold coverage ***(CDC, 2006).*** The most common genera of mold detected in indoor samples was *Cladosporium* (20–50%) and outdoor samples were *Aspergillus* and *Penicillium* (20–70%) at any given location ***(Solomon et al., 2006).***

The average values indicated Cladosporium (83%), Aspergillus (10%), Penicillium (5%) and Alternaria (1%) as the main fungal spore types present in the atmosphere of Porto (Portugal) ***(Oliveira et al., 2010)***. Also, 87% of the fungal spores in outdoor air of Copenhagen were accounted for by *Cladosporium*, *Alternaria*, *Penicillium* and *Aspergillus* ***(O'Driscoll et al., 2005)***.

The majority of the fungi isolated from the Belgrad Forest (Turkey) were of the form class Deuteromycetes (Fungi imperfecti). In the air, *Aspergillus* (24.333%), *Penicillium* (19%) and *Cladosporum* (17%) were the most populated genera ***(Çolakoglu, 2003)***.

Mould Morphology:

The phenotype of molds ranges from a unicellular to a dimorphic or filamentous appearance ***(De Hoog et al., 2000)***. They are non-motile and have life cycles that incorporate both sexual and asexual reproduction. They typically have elongated filaments or hyphae, which have cell walls that comprise complex polysaccharides including mannans, galactans, glucans, and chitin ***(Varki et al., 2009)***.

A) Gross Morphology:

The gross morphologies of fungi are incredibly diverse. Closer inspection of these structures reveals that all are composed of aggregated long, branching threads termed hyphae (singular hypha), organized to support spores for reproduction and dissemination ***(Kavanagh, 2005)***.

1) Hyphae:

The hyphae extend and branch within the supporting substratum as a network, termed a mycelium, from which the apically growing hyphae seek out, exploit and translocate available nutrients (Fig.1). The hyphae of individual fungi may extend endlessly via apical growth, provided they are supported with appropriate nutrients and other environmental conditions. Apically growing hyphae usually have a relatively constant diameter ranging from 1 to 30 μm or more, depending on species and growth conditions ***(Kavanagh, 2005)***.

The hyphae of a single species can differ considerably in diameter, depending on environmental conditions, their position in a colony, and the age of the colony ***(Carlile et al., 2001)***.