Airborne mould spores of allergenic importance

Powietrznopochodne zarodniki pleśni o znaczeniu alergenowym

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Abstract

The author described highly allergenic mould spores which may be classified as indoor and outdoor. 10 most common mould spores being detected in Europe were presented. Some distinct differences between outdoor and indoor species of various regions in Europe were noted and discussed in European research literature.

Key words: mould spores, air, allergy.

Streszczenie

W pracy przedstawiono ważne alergenowo, powietrznopochodne zarodniki pleśni. Szczegółowo omówiono zarodniki pleśni znajdujące się w warunkach domowych (tzn. "indoor") oraz w atmosferze (tzn. "outdoor"), zestawiając 10 najczęściej występujących zarodników w każdej z tych grup. Przedstawiono także wyniki badań europejskich, które wykazały istnienie różnic gatunkowych zarodników w warunkach "domowych" i "zewnętrznych" w różnych regionach Europy.

Słowa kluczowe: zarodniki pleśni, powietrze, alergia.

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Introduction

Moulds (or Fungi) are heterotrophic organisms which do not use light for photosynthetic assimilation processes. They need organic matter as nutrients, playing an important role in the natural biologic recycling processes by digesting and degrading organic substrates to chemically more simple components.

Many species of moulds live a very concealed life, and stay almost invisible during most of their biologic cycle. Some fungi form very diffuse systems of hyphae (mycelium) in the soil in the rhizosphere of "higher" plants, or in me organic substrate they *feed* on. Only their spore forming tissue systems become visible as mouldy of mildewed surfaces or as the better known fructification bodies like toad stools or mushrooms.

More biologic information about fungi, particularly in relation to aerobiology and allergy, can be found in handbooks and atlases (Gregory, 1973; Nilsson, 1983; Al-Doory and Domson, 1984; Wilken-Jensen and Gravesen, 1984; Ingold and Hudson, 1993; Gravesen et al, 1994; Samson et al, 1994; Cox and Wathes, 1995). Moulds reproduce and spread mainly by means of spores. They produce different kinds of spores originating from sexual or asexual reproduction. Spores can be distributed mostly by water and air, sometimes by specific vectors.

Airborne spores: outdoors, indoors

Of all the various kinds of biologic particulate matter suspended in the air in the atmosphere all over the world, mould spores constitute the largest portion, both in indoor and in outdoor environments. Under "natural" conditions, concentrations of over 100 000 spores per cubic metre of air are not exceptional. They cover a wide range of (aerodynamic) sizes from more than 100 μ m to less than 2 μ m. With breathing, large number of mould spores are inhaled, and a substantial portion, particularly of the smaller spores, penetrate into the deeper airways.

Although the atmospheric presence of mould spores has been known *for* long, knowledge of the qualitative and quantitative aspects of this natural bio-aerosol is still

 Table 1. Comparison of methods for sampling airborne moulds

 spores

ANDERSEN sampler REUTER centrifugal sampler	HIRST-type sampler (Burkard, Lanzoni)	
BIAP slit sampler (open dish)	(ROTOslide, ROTOrod)	
accurate identification	+ continuous, volumetric	
isolation; cultivation	+ preservable preparation	
dis-continuous; semi volumetric	- small spores under-represented	
selection by medium	- no isolation; no cultivation	
time and room consuming	no isolation; no cultivation	

Table 2. Provisional list of 10 most common outdoor and indoor airborne mould spores, based on data from European studies

For the outdoor mould spores estimations for ranges of relative amounts (in percentages), and for annual sums of daily average concentrations are given.

For the indoor mould spores only a tentative rank order of frequencies is given.

Outdoor			Indoor
Cladosporium	4-80	600 000	Cladosporium
Basidiospores	5-30	25 000	Penicillium
Ascospores	5-20	15 000	Alternaria
Yeasts, Sporobolomyces	2-20	15 000	Aspergillus
Botrytis	2-20	12 000	Botrytis
Aspergillus, Penicillium	2-20	10 000	Wallemia
Alternaria	1–10	7 500	Yeasts
Didymella	1-10	7 500	Mucor
Fusarium	1–10	7 500	Epicoccum
Ustilago	1–10	7 500	Basidiospores

rather fragmentary. There are two factors in the study of me presence of atmospheric mould spores which are responsible for this relatively poor level of knowledge. First, there are the differences and interactions between the more general outdoor mould spore spectrum and the more specific indoor mould spectrum, which can be very different between individual locations. And second, there are methodological difficulties in reliably collecting and identifying the spores from the air. This will be discussed briefly in the next paragraph.

The number of spore types, as well as their concentration in the outdoor air at any given time is depending on several interacting variables such as **seasonal** climatic factors, including temperature, precipitation, wind, air humidity, and diurnal patterns of



Fig. 1a. Provisional European outdoor mould spore calendar for a selection of ten microscopically identifiable spore types and *Aspergillus/Penicillium*. Presentation is on exponential scale to allow visualisation of small and big numbers in one graph

light and darkness. Quite pragmatically, one may speak of moulds producing "dry spores" and those producing "wet spores". Dry spores are released from the spore producing parts of the mould under conditions of decreasing humidity and increasing airflow. Examples of dry spores are those' from *Alternaria*, *Cladosporium*. Wet spores, such as those from many Ascomycetes and Basidiomycetes, are released into the atmosphere by processes related to high humidity conditions or rain.

Indoors, mould spores will become airborne when disturbed by air movements, very often caused by human activities. All kinds of organic materials, but also air conditioners or humidifiers, can be important sources of indoor mould spores. Moreover, spores occurring abundantly in the outdoor air will enter homes, working places, etc., mainly by ventilation. So, the indoor airborne mould-spore spectrum is usually a mixture of spores from both indoor and outdoor sources, depending on many changing and individual conditions. Due to these differences in conditions between individual indoor situations, it is almost impossible to arrive to conclusions which are generally valid for the indoor mould spore





situation (Begum and Nolard, 1994). In a recent monograph (Samson et al, 1994) the many aspects of indoor mould spores and their implications for human health are dealt with. References to European studies on outdoor mould spore concentrations can be found in D'Amato and Spieksma (1995) and Nikkels et al (1996).

Spore sampling and identification

For the collection and identification of airborne mould spores there is no completely adequate method, which combines continuous volumetric sampling with reliable taxonomic identification of all spore types (see Table 1). Some, so-called non-viable sampling techniques, e. g. based on the Hirst-type sampler, can be operated continuously, and give results in numbers of individual spores per cubic metre of air, by microscopic inspection of an impacting strip. But these samplers are size selective and collect particles smaller than 5 μ m with poor efficiency. This leads to under-representation of some abundant spore types such as *Aspergillus*, *Penicillium*, many Basidiospores, and others. Moreover, several spore types cannot be identified my microscopic inspection alone.

With other, so-called viable sampling techniques, like the Andersen sampler, the Renter Centrifugal sampler, or the BIAP slit sampler, it is possible to achieve a rather accurate identification of the moulds grain from the collected spores. But these methods are also selective, as not all spores can be grown to colonies on the usual media. And continuous sampling is impractical because of the very large number of colonies to be grown. Moreover, for many mould taxa the counts cannot be expressed as numbers of individual spores, but are given as numbers of "Colony Forming Units".

So depending on the kind of sampling technique, reports on atmospheric mould spores always give

incomplete information about the quantities of spores in the air. Probably the best solution for the dilemma is the combined operation of both methods: continuous volumetric sampling with the non-viable technique, next to a discontinuously operated viable sampler (Burge et al, 1977; Rubulis, 1984). Descriptions of the methods can be found in publications by Wilken-Jensen and Gravesen (1984), Samson et al (1994), and Cox and Wathes (1995). In practice, most studies on the presence of mould spores in indoor air are performed with discontinuous, viable samplers. Survey on outdoor airborne spores are mostly done with continuous, non-viable techniques. Some useful publications for microscopic identification are by Gregory (1973), Southworth (1973), Nilsson (1983), Wilken-Jensen and Gravesen (1984), Grant Smith (1986), Nikkels et al (1996). Drawn images of some of the commonest airborne mould spore types are shown in Plates A and B.

Summary of European studies

Direct comparisons or conclusions from the several European studies on outdoor mould spores, cited by D'Amato and Spieksma (1995) or Nikkels et al (1996), are difficult to make, because of the differences in sampling techniques and identification methods, as mentioned above. Yet, it is striking to see how much agreement there is between the various observations even when done in distant regions in Europe. In Table 2 the ten most common types of outdoor atmospheric mould spores are provisionally listed, with estimations for the ranges of the relative amounts and for the annual sums of the daily average concentrations. From this Table 2 it becomes dear that Cladosporium spores are by far the most abundant in the outdoor air. This is true not only for Europe, but also for most other places all over the world. Basidiospores and Ascospores, comprising spore types from many taxa, most of which are very difficult to identify, are also very abundant. Less abundant, but still not uncommon, mostly in relative amounts less 5% (not listed in Table 2), are in alphabetical order: Aureobasidium, Chaetomium, Curvularia, Drechslera, Entomophthora, Epicoccum, Erysiphe, Ganoderma, Leptosphaeria, Mucor, Paecilomyces, Phomd, Pithomyces, Pleospora, Polythrincium, Rhizopus, Rhodotorula, Scopulariopsis, Stemphylium, Tilletiopsis, Torula, Trichoderma, Ulocladiun, Urocystis, Uredinales, and many more. Although their average presence is usually less than 5%, under certain meteorological conditions their numbers may peak to high atmospheric concentrations for short periods of time.

Data on the seasonal fluctuations in the airborne presence of mould spores are rather scarce. A selection of some microscopically identifiable spore types and a combination of *Aspergillus* and *Pemcillium* spores, for

which continuous observations from some regions of Europe are available (Wilken-Jensen and Gravesen, 1984; Nildcels et al, 1996), is shown in Figure 1, presenting a provisional European mould-spore calendar. Also for the seasonal fluctuations there is not much difference between the various regions of Europe, except that in the North of Europe the season is somewhat shorter than in the Mediterranean.

The results from studies of indoor mould-spore presence are even less comparable, so that no indication of relative abundances can be given. Tentatively, in Table 2 a rank order of the ten most frequently found indoor spore types is given.

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