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# Fungal Growth and Aerosolization from Various Conditions and Materials

*Jacob Mensah-Attipoe and Oluyemi Toyinbo*

## Abstract

Microorganisms, especially fungi, from damp indoor environments are known to be one of the main causes of degradation of indoor air quality and can pose serious health hazard to occupants because of the production of airborne particles. Particles produced during microbial growth include both living and non-living particles, which can be submicrometer in size. Individuals are exposed to fungi from various sources and in various conditions. The exposure may occur when the fungi grow in hidden areas and on materials that are in common areas and released under various conditions. The proliferation of fungi detected in a particular area depends on the species of fungi, the growth material and the conditions under which they are grown and released. Fungi aerosolized from any growth material include intact spores, which grow when deposited on favorable material surfaces and other fragments of the growth ranging from a few millimeters to micrometers in size. The types and amounts of intact spores and fragments aerosolized depend on factors such as air velocity blowing over the growth surface, the type of substrate, type of fungi, and relative humidity of the growth and the age of the fungal growth.

**Keywords:** fungi, growth, aerosolization, infections, exposure

## 1. Introduction

Fungal spores and fragments usually in the sub-micrometer size range can be released from contaminated materials into air, and if inhaled, may cause adverse health effects for people and animals [1–3]. There is increased interest in the role of aerosolized fungal spores and their submicrometer fragments in adverse effects considering the strong association between the numbers of fine particles and adverse health effects [4–7]. Furthermore, fungal exposures are receiving increasing attention as an occupational and public health problem; this is due to the high prevalence of fungal contamination in buildings. Dampness and moisture-related problems are the main sources of fungal contaminations [8, 9] in homes and other domestic dwellings [10] as well as schools [11].

Fungal spores and fragments are one of the most common classes of airborne biological aerosols in many indoor environments and they form part of the complex community of indoor biological agents [12–17]. Most of these particles are encountered in indoor environments where we spend about 90% of our time [18]. Because of this, it is important to determine the sources of these fungal spores and their

fragments in such environments. Fungi from damp indoor environments are known to be one of the main causes of degradation of indoor air quality and can pose a serious health hazard to occupants [19, 20]. The submicrometer fragments are of utmost importance, because they tend to stay longer in air, and are easily inhaled. The smallest fragments ( $>0.1 \mu\text{m}$ ) can deposit deep in the respiratory tract having the potential for causing adverse health effects [21–23]. Furthermore, the large surface area of the fragments relative to their mass may evoke high biological activity [22].

The high number of released fungal fragments in combination with their potential to deliver harmful antigens and mycotoxins to the alveolar region of the lung suggests the need for their characterization. Furthermore, the properties of spores and fragments released from fungal growth are dependent on the type of materials, the species of fungi, the cultivation time as well as the air volume passing over the growth. The characterization of fungal particles is important to help us understand the potential health effects associated with the exposure [21, 24]. Fungal spores are considered the most abundant fraction of these particles; they have an aerodynamic diameter ( $d_a$ ) in the size range of 1–10  $\mu\text{m}$  [25].

Indoor air, like outdoor air, has many sources of contaminants that affect health adversely. However, it is not clear which source is associated with the adverse health effects. As earlier explained, because we spend most of our time indoors, it is important to characterize fungal fragments based on their origin since this knowledge can improve our understanding of the potential adverse health effects associated with exposure to these particles.

It has been estimated that dampness and mold growth can be detected in most home as reviewed by Mudarri and Fisk [26] and have been associated with increases of 30–50% in several respiratory and asthma-related health outcomes [27]. Furthermore, approximately 8–18% of cases of acute bronchitis and 9–20% of respiratory infections are estimated to occur in environments contaminated with fungi [28].

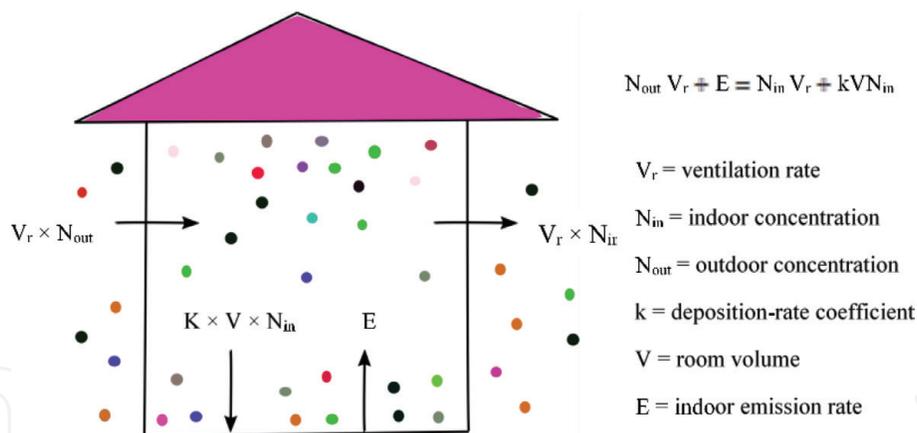
The review of Samson et al. [29] claimed that floods, wet seasons, thermal modernization of residential buildings, air-conditioning systems, construction or material faults, and poor and improper ventilation are the major reasons for increase in the relative humidity and dampness of materials in the indoor environment. When moist conditions are prolonged in indoor environments, for example, when building materials stay damp for a long time, then the growth of microbes is promoted and there is an increased risk of microbial contamination [29–31]. In addition, certain characteristics of the home [32] as well as personal activities of its occupants [33] influence the microbial profile in indoor environments.

Generally, a wide range of fungal species may be encountered in the indoor air. For example, Zyska [34] surveyed the available literature and compiled a list of more than 200 fungal species present in air or growing on structural materials in indoor environments and therefore likely to contribute to the airborne fungal burden. Fungi in indoor environments can be inhaled and exposure via the airways is especially problematic. Furthermore, the presence of fungal particles has been linked to many diseases and symptoms among the occupants of moisture damage buildings [9, 19].

## **2. Indoor sources of fungi**

There are several sources of fungal particles in the indoor environment. This includes fungal particles exclusively generated from indoor sources and those that infiltrate from the outdoor environment as shown in **Figure 1**.

Fungi found indoors may be from different sources. However, the majority (70–80%) of indoor fungal aerosol and fungal allergens (80%) are generated in the indoor environment [3]. In a study by Adams et al. [35], they observed that fungal



**Figure 1.** Schematic diagram showing the sources of fungal particles in the indoor environment [3]. Reproduced with permission from Yamamoto et al.

composition indoor was related to dispersal from the outdoor environment and are passively collected by indoor surfaces, although they rarely grow on the surfaces.

In addition to the above, the basic characteristics and parts of a building can also affect the emergence of fungi. Different researches including Despot and Klarić [36] and Toyinbo et al. [37] have associated buildings with basements with the emergence of indoor mold. This may be due to the high humidity and cold temperature in the building basement. The high humidity and/moisture content may occur from leaky pipes or cracks in the basement walls that allow ground water to penetrate the basement. Another source of moisture in the basement is flooding which makes water to move down to the basement and usually dry at a slow rate due to lack of adequate ventilation. This creates a favorable condition for fungal growth. The kitchen and bathroom sections of a building may also encourage the growth of fungi since these places have a high moisture content and substrates [38].

Outdoor generated indoor fungi enter a building through the ventilation system. This can be a mechanical ventilation system without adequate air filter for pollutants or through a naturally ventilated building with open windows and doors where outdoor to indoor ratio of pollutants can be close to unity. A ventilation system can also be a reservoir for indoor fungi especially when the ducts and filters are dirty with dust that serves as a substrate for fungi growth [39]. A DNA-based analysis of air handling unit filters by Luhung et al. [40] shows diverse genera of fungi, which includes *Cladosporium*, *Aspergillus* and *Lentinus*. Oil residues in ventilation ducts can also trap dusts and serves as a source of nutrients for fungal growth that can be transferred indoor through the ventilation system [39].

### 3. Health effects of fungi in indoor environment

The health effects associated with fungal exposures may be caused by the fungi themselves, fungal mycotoxins, and fungal cell wall components or metabolically produced volatile compounds. The health effects can be categorized into three groups: (1) infections, which are caused mostly by the viable cells; (2) allergic reactions, which are usually caused by both viable and non-viable cells and components of the cell wall of the fungi if they carry antigens and (3) toxic responses, usually in response to the mycotoxins produced by the fungi.

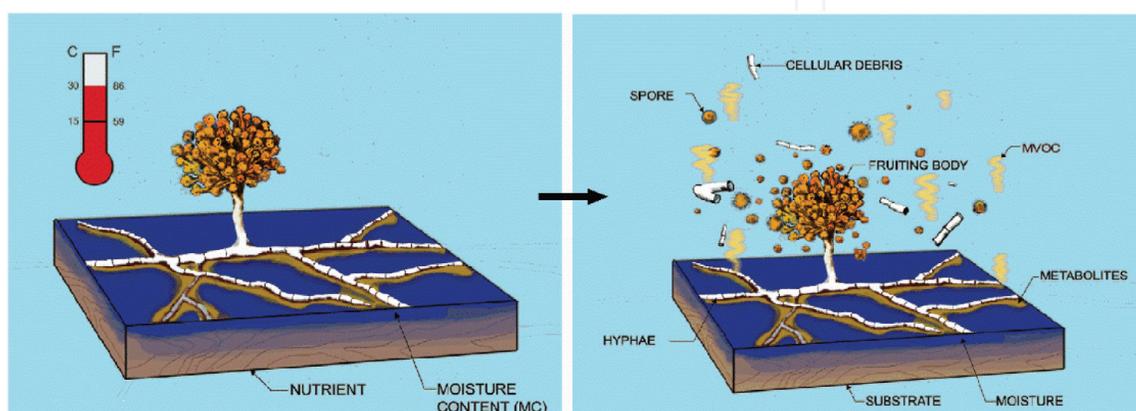
Exposure to fungal particles has been linked to a range of adverse health effects [41]. For example, exposure to fungi has been associated with the onset of asthma in both infants and adults [42–47].

There is convincing data in the literature suggesting an association between moisture damage in a building and the incidence of diseases such as new asthma cases, current asthma, respiratory infections, cough, allergic rhinitis, eczema and bronchitis [2, 42, 43, 46–49]. In contrast, quantitative assessments have not detected any consistent associations between fungal measurements and adverse health effects. Nevertheless, limited or sufficient associations have been documented between the fungal concentration in dust by qPCR, cultured airborne fungi sampled from indoor air as well as several microbial compounds such as ergosterol, endotoxins and beta-glucans in dust and adverse health effects [50–53]. There is credible scientific evidence to support the association between moisture damage, visible fungal growth measured indoors and adverse health effects. The World Health Organization (WHO) has stated that approximately 25% of residents in social housing stocks are prone to experience elevated health risks associated with their exposure to indoor molds.

#### 4. Fungi and fungal growth

Fungi are eukaryotic organisms that lack chlorophyll and obtain their nutrients from the growth media by the use of enzymes that they secrete. On the other hand, molds are filamentous fungi that grow with branched multi-cellular filamentous structures called mycelium [54]. In general, fungi are characterized by a visible vegetative body or a colony composed of a network of threadlike filaments which infiltrate the materials on which they feed. Fungi are usually saprophytic in nature; thus, they obtain nutrients from dead organic matter provided there is sufficient moisture. They can live off many of the materials present in the indoor environment such as wood, cellulose, insulations, wallpapers, glue and everyday dust and dirt [55–57]. Thus, fungi have the remarkable capability to degrade almost all natural and man-made materials [15, 58, 59] especially if they are hygroscopic [10, 60]. Fungi obtain nutrients by releasing extracellular enzymes and acids that break down the materials prior to their absorption. In the process, particles, including microbial degraded materials as well as gases, especially microbial volatile organic compounds (MVOCs), are released into the environment [61].

The MVOCs may form sub-micrometer particles through a process of secondary aerosol formation [61, 62]. These sub-micrometer particles have been shown to be aerosolized into the indoor environment following exposure to the effects of airflows and vibration [62, 63] **Figure 2**.



**Figure 2.** Schematic diagram showing the growth of fungi on a material surface with the subsequent release of particles of the fungal growth [64]. Reproduced with permission from Morse and Acker.

## 5. Conditions that promote fungal growth indoors

### 5.1 Material characteristics

Distinct characteristics of the growth material can play an important role in the creation and accumulation of moisture which eventually lead to mold growth on their surfaces [65, 66]. For example, when buildings are constructed with very good insulations in order to reduce heat loss and improve thermal performance, the several layers of insulation prevent easy movement of air in and through the building materials leading to accumulation of moisture within the building materials as well as the building. Consequently, the building becomes a microbiological reservoir and a contributor to the microbial exposure due to their ability to absorb and accumulate moisture [67].

Due to the heterogeneous nature of new buildings, there are varieties of materials that serve as micro-niches, that is, they have a favorable temperature, water activity ( $a_w$ ) and relative humidity (RH). For example, the surfaces of affected building materials (such as concrete and ceramic tiles in moist walls, ceiling tiles, dust laden wooden furniture) create specific niches suitable for the growth of microorganisms including bacteria and fungi. As expected, the climate within the building varies from one part of the indoor environment to the next. Thus, fungal growth would also be predicted to vary with the microclimate created. Moisture damage and dampness in buildings often affect a variety of structural components of building materials, leading to a deterioration of the indoor air quality.

### 5.2 Water, nutrients and temperature requirements

Water-damaged building materials, particularly those rich in organic matter, can support microbial growth if they remain wet for a prolonged period of time [55, 59]. Under certain required conditions such as temperature, nutrient and pH conditions, microbial growth can occur within an hour [24]. Nonetheless, the principal limiting factor is the availability of moisture [55, 68]. It has been established that the lowest RH of a material at which fungi can grow is in a range around 75–80%, which corresponds to a water activity ( $a_w$ ) of 0.75–80 [55, 69, 70]. The moisture of the substrate that is available to the fungi for growth is the so-called free water and this amount is influenced by the relative humidity of the surrounding air. This does not include bound water that is a component of the chemistry of the substrate [24]. Moisture sources for fungal growth on materials indoors may be internal or external with moisture movement into and through building cavities by convection, gravity or capillary action.

Pasanen et al. [71] found that relative humidity values of 70–90% are required if there is to be fungal growth on building materials. Furthermore, the relative humidity required for growth depends on the particular material and the fungal species involved. Since most materials are porous in nature, adsorption of water into the materials first occurs via the pores before the material surface and become available to the microbes. Thus, porous materials support fungal growth when their RH is higher than 80% [68]. These conditions influence the extent of colonization and the types of fungi that will be present, since any changes in moisture availability will change also the composition of the microbial species present in that environment. For example, certain species of *Penicillium*, *Eurotium* and *Aspergillus* grow in relatively dry environments with RH between 75 and 85% (e.g., in settled house dust on material surfaces with a relatively low RH). As RH increases, different species such as *Basidiomycetes* and *Eurotium* begin to grow, requiring continuously wet substrates such as soaked wallboard with RH range of 80–90%, while others like *Fusarium*, *Cladosporium* and *Stachybotrys* only grow at RH exceeding 90% [29, 70–73].

In addition to humidity and water, fungi need adequate nutrition and temperatures to grow. The availability of nutrients depends on the composition of the building material. Building materials like wood and ceiling tiles are organic in nature; they contain complex polymers like starch, cellulose and lignin. These components are broken down by the extracellular enzymes of the fungi into simple sugars, amino acids and other simple nutrients [74, 75]. As fungi can utilize many complex polymers, a wide range of materials can act as nutrient sources.

Fungi can grow over a wide temperature range (5–39°C), [76]. However, at low temperatures (0–5°C), the fungal metabolic activities necessary for growth are slowed down, rendering the fungi dormant until an optimum temperature is reached [77]. At a higher temperature (34–36°C) the metabolic reaction rates increase and at temperatures above 46°C, the fungi become stressed and die [78]. This is because most of the activities of the fungi are dependent on DNA and enzymes. Due to the above, the concentration of fungi is usually high during the summer season as compared to winter season [79].

### 5.3 Types of building materials

Fungal growth on building materials is dependent on the chemical composition of the materials [58]. The most susceptible materials to microbial growth and biodegradation are those with a natural organic composition, for example, wood and paper. These materials contain starch, cellulose and hemicellulose, pectin and lignin [74, 80, 81]. Based on these components, a wide variety of materials are potentially suitable for supporting fungal growth [15, 58, 59].

Buildings contain a wide variety of materials that affect the germination and growth rate of fungi [82]. Thus, each material serves as a niche for a specific microorganism, depending on the composition of the material, water activity and nutrient content [58, 83]. These properties of the building materials determine the diversity and extent of growth of the microbes [84, 85].

Wood remains the most extensively used material in buildings [81, 86]. Wood is able to absorb and retain water and moisture from both standing water and the environment [81, 87]. This characteristic in addition to the high nitrogen-bound compounds and low molecular carbohydrates that are transferred to the wood surface during processing mean that wood is very susceptible to fungal growth [87]. For example, a study by Meklin et al. [88] found school constructed with wood to have a higher concentration of fungi (5–950 cfu/m<sup>3</sup>) than those constructed with concrete (<2–5 to 500 cfu/m<sup>3</sup>). Although concrete is also hygroscopic, it has a low moisture permeability which reduces its rate of degradation and it contains very little or no nutrient for fungi growth [89]. Fungal species commonly found on moisture-damaged wood include *Aspergillus versicolor*, *Penicillium brevicompactum*, [81, 84, 85].

Gypsum board, on the other hand, is mostly used as the inner wall liners in buildings [90]. The paper liners used to reinforce the gypsum core makes gypsum board susceptible to fungal growth. Since the inner core (gypsum) is able to retain water and make it available to the surface paper lining, there can be a prolonged presence of water and moisture required to sustain fungal growth [10]. While the inner core (gypsum) may not be susceptible to fungal growth, the glue and paper serve as good media due to their organic nature [91]. The fungal species routinely found on gypsum board are the cellulolytic *Stachybotrys chartarum* [70] and *Cladosporium cladosporioides* [91].

Plastic materials are also becoming a common material used in buildings, as either sheets or pipes. As sheets, they are used as material envelopes, which insulate the building. Though plastics are known to be resistant to microbial attack because

microbes do not possess any enzymes capable of degrading synthetic polymers [92], the addition of plasticizers can make the plastics susceptible to microbial growth [93]. These plasticizers are commonly organic acid esters such as dioctylphthalates (DOP) and dioctyladipate (DOA) which are added to the polyvinyl chloride (PVC) to modify the polymer's physical or mechanical properties [93].

Glass fibers used in insulation materials do not support fungal growth. However, the glue used as binders does contain nutrients that may promote fungal growth [90] since these glues can be synthetic or plant-based. For example, the urea-based derivatives, polyurethanes, which are used as binders, are known to support fungal growth [94]. Plant-based binders are also used in binding certain building materials such as plywood, and ceiling tiles and may contain nutrients suitable to allow fungal growth.

#### **5.4 Contamination or soiling**

All materials, both organic and inorganic, are able to sustain fungal life especially when the materials have dust, dirt or other deposits on their surface which represent sources of carbon and nitrogen [56, 57]. Dust is known to contain microorganisms, debris and other animal or insect parts that serve as nutrients for fungal growth [95]. Thus, more growth is observed on materials with dust on their surfaces compared to those without dust [56, 96]. Furthermore, settled dust or soil alters the water absorbing and retentive characteristics of the material surface, making the material surface continually moist, conditions in which fungi thrive [10]. Dust absorbs water from the atmosphere. It has been shown that dust competes with the material surface for moisture, with the dust holding more water due to its more hygroscopic nature. Therefore, dust may promote fungal growth even on materials that naturally would not support microbial growth [56, 57]. It is therefore important for indoor surfaces to be continually cleaned to avoid fungal growth and any health effect associated with it.

### **6. Aerosolization of fungal spores and fragments**

Forces such as turbulence, temperature, air velocity, vibration and zone of convection are usually associated with the release of fungal spores and hyphae from fungal colonies. In addition, factors such as the maturity of the colony, changes in temperature, relative humidity over the culture surface, light periods, nutritional composition of the substrate and the specific fungal species will determine the frequency and the number of spores that will be liberated and transported into the air at any given time. Furthermore, the dispersal of the fungal particles depends upon their size, shape, roughness, density, electrostatic charge, air movement and activities that influence the circulation of the air [24].

Release of fungal particles usually occurs by two mechanisms; active and passive release [68]. Active release refers to an adaptive type of particle aerosolization, via forces arising inside the fungi attributable to a burst of energy by a mechanism known as osmotic pressure and surface tension discharge [97]. Passive release occurs by energy originating from outside the fungi, such as mechanical disturbances of the fungal colonies by mechanical handling, vibration or air currents. The latter forces can also cause secondary release of settled spores from surfaces. Activities that have been shown to increase fungal spore concentrations in indoor air include daily activities such as vacuuming, sweeping, walking etc. [98–103].

During fungal growth and sporulation, as well as when the culture is in a dormant phase, spores and bioactive agent containing fragments are released into

the indoor environment [21, 61, 104–107]. As mentioned earlier, hyphal fragments are of high importance since they make up about 6–56% of the total fungal particles based on microscopic sample analysis [108, 109]. Aerosolized fungal particles in chamber studies have shown that fungal fragments are released at levels up to 514 times higher than spores [21, 61, 106, 107, 110]. In other studies, Li and Kendrick [111] used microscopic counting and found that hyphal fragments accounted for only 6.3% of the total number of fungal particles in indoor environments. In addition, by applying a biomass determination, Adhikari et al. [112] detected lower amounts of  $\beta$ -N-acetylhexosaminidase (NAHA) enzyme in fungal fragments  $<1 \mu\text{m}$  compared to spores  $>1.8 \mu\text{m}$ .

Though both types of particles (spores and fragments) released from the fungal cultures during aerosolization are potentially harmful, the fragments are of greater importance since they tend to suspend longer in air than the spores [61, 62, 106, 107, 113]. They also have a tendency to penetrate deep into alveolar regions of the respiratory tract when inhaled [21, 114]. Cho et al. [21] have used a computer-based model to assess the deposition of spores and fragments of *A. versicolor* and *S. chartarum* in the respiratory tract. For both fungi, they found that the vast majority, 65–90%, of inhaled fungal spores deposited in the nasal and extra thoracic regions while only 3–15 and 2–5% of the spores deposited in the alveoli-interstitial and bronchial-bronchiolar regions, respectively. They also demonstrated that about 60% of fungal fragments deposited in the alveoli-interstitial region with 14–15% being trapped in the nasal and extrathoracic regions. It can therefore be deduced from the above modeling analysis that the different deposition efficiencies could have consequences on the potential adverse health effects induced by inhaled fungal particles of different sizes.

Fungal fragments have been shown to contain antigens [61, 62], allergens [5, 115, 116], mycotoxins [23, 117], and (1  $\rightarrow$  3)- $\beta$ -D-glucans [23, 52]. Their size in relation to their numbers and their biological properties all contribute to their potential to evoke adverse health effects. It is known from atmospheric studies investigating the adverse health effects of ultrafine particles that it is the number concentration rather than mass concentration which is important [118, 119].

Different fungal species have characteristic structures and thus behave differently when they become airborne. In addition, the growth substrate providing the nutrients for the fungi may also affect the properties of the spores and fragments and could contribute to fragments released from the biodegradation of the substrate itself during fungal metabolism. The amount of fungal particles released may also depend on the type of substrate and the conditions under which the fungi were grown. It is very important to evaluate spore properties under a variety of conditions in order to gain insights into the contribution these factors have on the adverse health effects produced by these particles.

## **7. Aerosolization and characterization of fungal spores and fragments**

One of the ways fungal particles are characterized is by their properties when they are released from contaminated materials. The particles released are affected by the growth substrate, fungal species, age of the culture and air velocity to which the cultures had been exposed [120]. The same factors affected the fragment/spore (F/S) ratios [121].

Biological particles are usually distinguished from non-biological particles by their ability to fluoresce when excited with photons at a certain wavelength. The fluorescence property is based on molecules such as tryptophan, tyrosine, or phenylalanine, reduced nicotinamide adenine dinucleotide (NADH), and

nicotinamide adenine dinucleotide phosphate (NADPH) as well as riboflavins, flavin adenine dinucleotide (FADH) and flavin mononucleotide (FMN). Depending on the conditions under which the fungi grow, differences in fluorescence properties are observed. For example, spores obtained from cultures on building materials, such as, gypsum board, have been shown to have lower fluorescent properties than spores from agar. This indicates that cultures growing on nutrient poor substrates contain less compounds capable of fluorescence. Studies by Agranovski et al. [121] and Kanaani et al. [122] measuring fungal amounts from agar using fluorescence measuring devices in laboratory settings resulted in good detection efficiency of the instruments. However, the use of fluorescence properties may underestimate the concentration of fungal particles due to influences of nutrient availability on the growth of the fungi.

The type of species also affects the fluorescence properties. For example, lower fluorescent particle fraction (FPF) values have been observed for *C. cladosporioides* compared to *A. versicolor* and *P. brevicompactum* [120, 123]. The structure of the spore plays a major role in allowing devices to measure fluorescence properties. *C. cladosporioides* has a dark-skinned coating preventing impinging photons from penetrating to reach the exterior pigments to excite fluorescence from internal fluorescence. It can be deduced that *C. cladosporioides* concentrations may be underestimated in field measurements.

In recent study by Mensah-Attipoe et al. [121] and Afanou et al. [104], they observed that *A. versicolor* produced a higher F/S-ratio compared to *C. cladosporioides* and *P. brevicompactum*. The increased sub-micrometer fragments from *A. versicolor* can be attributed to the outer-wall spines, which are easily sheared away during sampling.

Studies have shown that the type of material and nutrient affects how much particles are released [120, 121]. For example, the fragment/spore ratio (F/S) for agar was higher compared to wood and gypsum board. Seo et al. [124] observed a higher F/S ratio for *A. versicolor* cultivated on agar than on gypsum board and ceiling tiles. Generally, higher concentrations of fungal particles are aerosolized from dry surfaces with low moisture contents than wet surfaces with high humidity [62]. Agar may have a different moisture content and moisture dynamics during the fungal growth than wood and gypsum board. During growth, the moisture content becomes reduced [23] and it is possible that agar loses more moisture than wood and gypsum. Therefore, fungal growth on agar undergoes desiccation stress and releases more fragment particles than when it grows on wood and gypsum board.

It has been observed that fragment/spore ratio (F/S) increases with increasing age of the culture. Moisture content of wood and gypsum increases with incubation time. Therefore, before aerosolization can yield enough particles, the material must be dried. With differences in the absorption and retention of moisture by the various materials, fungal biomass is also affected and hence affects the release dynamics of fungal particles from the material surfaces. Seo et al. [124] demonstrated that F/S increased with age. They attributed the increase in particle release from older cultures to changes in fungal biomass and moisture content. Dryness on the surface of the culture increases the aerosolization of fungal particles by reducing the adhesion forces between the fungal structures and making these structures more brittle [124]. Therefore, it has been concluded that with time, fungal growth in buildings may increase the contribution of sub-micrometer-sized fungal fragments to the overall mold exposure [124]. Spores aerosolized from older cultures displayed lower fluorescence than younger cultures. Kanaani et al. [125] reported a decrease in fluorescence emitted by *Penicillium* and *Aspergillus* from 2 days to 21 days. They suggested that fluorescent intensity of biomolecules such as nicotinamide-adenine dinucleotide phosphate NAD(P)H and surrogates of metabolic function such as

riboflavin found in fungal spores may vary according to the environmental conditions under which the fungal colonies are growing and also on their concentration at a particular point in time. The decrease in fluorescence with age could also be due to changes in the fluorescent compounds as the culture ages.

Concentration of fungal spores and fragments has been shown to increase with increasing air velocity, but the F/S ratios decreased with increase in air velocity. A decrease in fluorescence per spore was observed when the air velocity was increased. It is also possible that as larger particles are carried along with the increased air currents in the sampling lines, they impact on the sides of the walls resulting in the breakage; as posited by Afanou et al. [104, 105].

Fragments have been proposed to be secondary organic aerosols formed from MVOCs released from fungal growths (secondary formation of aerosol particles) [61]. If fragment particles are formed by this mechanism in the presence of ozone, the concentration of fragments should decrease with higher flow rates due to their increased dilution. However, the opposite was observed by Mensah-Attipoe et al. [121], meaning that secondary aerosol formation may not be a relevant process for origin of fungal fragments. Instead, fragments are mainly formed through mechanical processes. It has been shown that fungal fragments are aerosolized at low air velocity [61]. Studies by Mensah-Attipoe et al. [121] show that fragments and spore concentrations increased with greater air velocities, however, the spore concentration increased more than the fragment concentration. This explains the decrease in F/S ratio when the air velocity is increased. A decrease in fluorescence in response to the increase in air velocity has been postulated to be due to a decrease in relative humidity of the culture causing desiccation stress to the fungal spores [125]. In addition, due to the increased air velocity, larger fungal hyphae are aerosolized together with spores due to increased stress and desiccation of the colony. The desiccation stress and decrease in fluorescence induced by increased air velocity has been attributed to a loss of spore viability [125].

## **8. Conclusions**

The type of building material and fungal species affect the amount of growth measured on the contaminated surfaces. In addition, these factors together with air velocity and age of the culture affect the properties of the fungal particles aerosolized from fungal contaminated surfaces. The nutritional value, chemical composition and moisture requirements as well as sources of external nutrients potentially affect fungal growth.

Fluorescence property of the particles which is sometimes attributed to their viability decreases when fungi are grown on poor nutrient substrates, released from older cultures and released in the presence of high air velocities. Since a building has many different materials in its structure and varying airflows passing over different ages of the growths at any point in time, it is concluded that fungal viability and their ability to cause infections may vary under different conditions.

F/S ratios decrease with increasing air velocity while spore concentration increase. This suggests that the conditions under which individuals are exposed to fungal particles may be different. A fraction of the fragments could be derived from building materials due to biodegradation of substrates when they are subjected to fungal metabolism. Fragments aerosolized from building materials could represent a potential health hazard depending on the composition of the material.

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