Effect of indoor lights on development of Aspergillus versicolor in buildings

Borui Chen¹, Edward Barrett², Spyros Efthymiopoulos² and Hector Altamirano²

- ¹ MSc Environmental Design and Engineering, University College London, UK;
- ² Bartlett School Environment, Energy and Resources, Faculty of the Built Environment, University College London, UK, correspondence email: h.altamirano-medina@ucl.ac.uk.

Abstract: The presence of moulds inside buildings has been a growing concern as it may affect the integrity of buildings and the health of occupants. Most studies have focused on growth conditions involving moisture, temperature and construction materials. Though moulds are known to have light responses, ranging from developmental regulating to metabolites producing, it is unclear whether they respond to light levels found indoors. This study analysed the influence of indoor light of varying colour temperature and illuminance on germination, growth and sporulation of *Aspergillus versicolor*. Mould germination was not significantly affected by colour temperature or light intensity. The development of mycelia was inhibited when completely exposed to light (1050 lx), both warm and cool lighting, and the cool colour temperature had a stronger effect. If the illuminance increases from darkness to 1050 lx, changes in the morphology and biological process were observed. Colonies exposed to high illuminance were high and denser; however, incubation under dark or weak light intensity triggered mould conidiation earlier. This difference in light responses of *A. versicolor* may result from protective mechanisms against environmental or light-relevant stress. With the results, this study demonstrates the role of indoor lights in regulating fungal development in buildings.

Keywords: Aspergillus versicolor; Mould lifecycle; Artificial lights; Illuminance; Colour temperature

1. Introduction

The prevalence of mould growth in buildings has been highlighted around the world. According to the World Health Organization (2009), between 10 % and 50 % of buildings were estimated to have mould and dampness issues spanning Europe, Asia and America, which may be due to poor performance and maintenance of buildings. The presence of mould indoors can affect the integrity of buildings and the health of occupants. The building structure may be at risk of damage due to biodeterioration caused by moulds, such as the wood structure in an old house (Viitanen, 1994). Also, the health of occupants may be adversely affected by mould growth within buildings. Microbial volatile organic compounds (MVOCs) can be released by moulds, as well as unpleasant smells. Clinical research demonstrated a strong link between mould-infested houses and respiratory disease (asthma, coughing, respiratory infections) (Quansah et al., 2012; Kanchongkittiphon et al., 2015). Mould pollution would potentially develop the progression and exacerbation of respiratory disorders (Mendell et al., 2011). In particular, moulds forming toxins cause intoxication reactions in humans, expanding body immune responses and thus inducing adverse effects on the respiration of dwellers (Jarvis et al., 2005). In addition, mould growth threatens heritage collections as the degradative abilities of the mould are devastating in archives (Bastholm et al., 2022).

The growth of mould is a consequence of interactive effects between environmental factors. Humidity and temperature are the main environmental factors affecting the growth of mould in buildings and have been studied extensively. Growth conditions vary between

mould species. In general, moulds have a wide range of temperature tolerances and thrive between 5 °C and 35 °C (Panasenko, 1967). Some moulds can grow at relatively low relative humidity levels of 75% (Johansson et al., 2012), while most mould species require high relative humidity to achieve growth conditions (Heseltine et al., 2009). Even for the same species, their growth condition differs between life phases (Grant et al., 1989). Building materials can be used as nutrients. Different materials have distinct susceptibilities to mould growth and exhibit different growth propensities (Vacher et al., 2010). In addition to these, light may also influence mould growth.

Light can affect fungal germination and proliferation, given the spectrum and species (Bühler et al., 2015). Exposure to the ultraviolet spectrum may be detrimental and even lethal. The effects within this wavelength band are typically associated with the germination and sporulation stages of fungi (Cheong et al., 2016). Some mould species can produce spores in the darkness (Fuller et al., 2015), and the presence of light may encourage sporulation, while inhibition of growth happens accompanied by direct light or in a particular part of the light spectrum (Faneil et al., 2012; Cheng et al., 2012). These effects are significant in determining the ability of mould to grow in various lighting conditions. However, the effects of mould development and viability by light exposure are no more thoroughly understood than those of other factors. There is a great need to study the effects of lighting on mould, particularly of species developing in the built environment.

In this study, the influence of light levels of varying colour temperature and intensity on a common mould species, *Aspergillus versicolor*, has been assessed. Emphasis was laid on germination, growth and sporulation and the differences that could be found in the reaction between light conditions.

2. Materials and methods

2.1 Strains and growth conditions

Aspergillus versicolor strain was obtained from the Biology Lab in this experiment. It was inoculated onto malt extract agar (MEA). Each inoculated point in a Petri dish was marked and performed in triplicate. The mould cultures were incubated at 30 °C.

2.2 The light incubation equipment

Three light boxes were constructed to enable the incubation of the cultures under different colour temperatures of light. Each light box was subdivided into six rooms with the following illuminances, reduced through the use of neutral density (ND) filters: room 1, ND3 filter (427 lx); room 2, ND6 filter (240 lx); and room 6, ND 9 filter (157 lx). Room 3 was fully exposed to the lighting (1050 lx), and room 5 was covered with black cardboard as a control group in darkness. An internal humidity, temperature and illuminance data logger (Onset HOBO U12-012, USA) was mounted inside room 4. Three different indoor LED lights (Lumenpulse, Manchester, United Kingdom) were used with respective colour temperatures: warm (3000 K for light box 1), cool (5600 K for light box 2) and neutral (4000 K for light box 3). The light intensity was kept at the same level on top (2135 lx) and inside (1050 lx) of the box. All measurements of light intensities were measured with an illuminance meter (Konica Minolta T-10A, Japan). Light boxes were placed in a test chamber (JTS Ltd, United Kingdom) isolated from the external environment during incubation. Inoculated Petri dishes were incubated in the boxes at 30 °C and sealed with laboratory film maintaining a stable relative humidity of 80 to 90 %.

2.3 Growth assessment

For analysing germination, growth and morphology the colonies were single point inoculated on MEA agar, grown under the respective light conditions and imaged. The diameter of a colony was measured in two orientations at right angles to one another. Three replicates were measured and made up of a measurement sample of one mould colony.

3. Results

3.1 Influence of light levels on mould germination

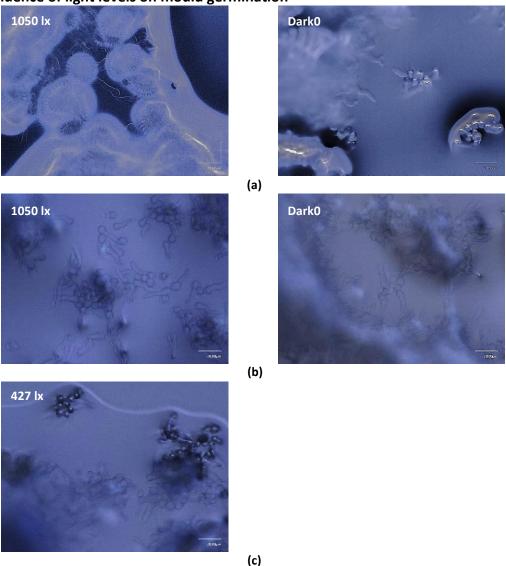


Figure 1. Spores and germ tubes of Aspergillus versicolor under different colour temperatures: warm light 3000 K (a); cool light 5600 K (b); and neutral light 4000 K (c). The diameter of spores was approximately 5 μ m. All scale bars represent 20 μ m in length.

Mould germination was checked after 24 hours, including samples grown at different illuminances (0 lx, 427 lx and 1050 lx) under warm and cool colour temperatures, and the sample cultivated at 427 lx under neutral illumination as a substitute for the contamination of whole light and darkness. **Figure 1** shows images of spores and germ tubes under the digital microscope. Most (over 50 %) spores germinated for the samples observed since germ tubes that had grown were approximately triple the diameter of spores. For different light conditions, the length of the germ tubes did not vary. In addition, minor changes in the

morphology of germinated spores occurred, and slight branching hyphae were noticed in the sample incubated without light. Though some spores were inactive, the mould incubated under different light environments did not show a significant difference in germination compared to controls grown in the dark. This indicates that the germination was not significantly affected by colour temperature or light intensity.

3.2 Influence of light levels on mould growth

Hyphae grew and interlaced in mass on inoculated points to form mycelium, which saturated the area of points five days after inoculation. Colonies grown for 30 days reached 6 to 8 mm in diameter, with high and dense hyphal aggregates under total light exposure; those in the dark featured flat appearances but were slightly larger. However, no colour pigmentation of hyphae was seen in these samples.

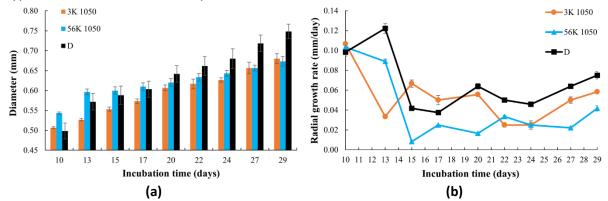


Figure 2. Colony diameter (a) and radial growth rate (b) of *Aspergillus versicolor* under different light levels: 3K 1050, warm light 3000 K 1050 lx; 56 K 1050, cool light 5600 K 1050 lx; and D, dark. Error bars are standard errors of three replicates for each light treatment.

Compared with dark conditions, regarding the radial growth, the light was found to have a slightly positive influence on growth in a few days following inoculation (**Figure 2 a**), and treatment with warm lighting was able to attain the growth rate to the extent of darkness sometimes, but as the incubation time increased, the development of mycelia was also inhibited (**2 b**). Significant inhibition of colony growth was found in both light groups. In the presence of cool lighting, the reduction in the radial growth rate of *A. versicolor* was more significant. The complete stalling effect on the growth was not observed under a warm or cool light. The mould colony could grow and expand better under dark than full light and showed a slightly increased growth rate later.

These results suggest that *A. versicolor* is responsive to light levels found indoors with respect to its difference in growth and morphology. Moreover, a cool colour temperature leads to more growth suppression than a warm one.

3.3 Influence of light levels on sporulation

As described above, light treatments did affect the growth rate of mould colonies. In order to test the influence of light levels on the sporulation, the dense mycelia were microscopically observed in the central area of mould colonies. The differences in the shape of stalks (or spore-bearing stalks) were noted under different light intensities. Many conical-shaped and swollen apices (vesicles) were observed on stalks in control groups, suggesting initiation of spore formation occurred, and spores would be further produced. The stronger the light intensity to which the mould was exposed, the less the number of swollen portions was found. A further increase in the illuminance to 1050 lx led to the complete abolition of detectable vesicles, and only thin mycelia were found (Figure 3). Comparisons at different colour

temperatures showed the initiation of sporulation occurred more noticeably when the mould was exposed under cool lighting. This result shows that incubation in a dark or dim environment resulted in mould conidiation earlier.

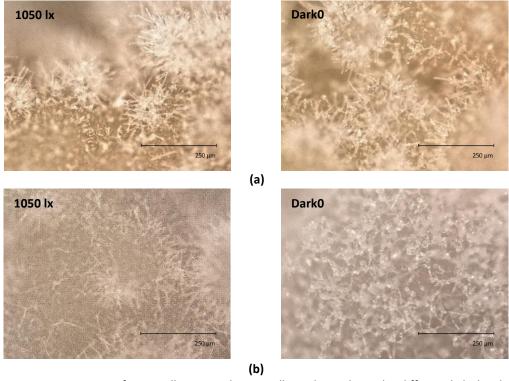


Figure 3. Image comparison of *Aspergillus versicolor* on stalks and mycelia under different light levels: warm light 3000 K (a); and cool light 5600 K (b). All scale bars represent 250 µm in length.

4. Discussion

The previous analysis of Schmidt-Heydt et al., (2011) had reported that Aspergillus incubated under white light generally grew better than in darkness, indicating that the light could promote the growth of Aspergillus, which may also apply to *A. versicolor* in the same genus. However, this conclusion is contrary to the results of the present study. When the mould was exposed to indoor lights, colonial growth was inhibited. One reason for this disparity could be that tested species varied, as these influences may be specie-dependent as white light slowed down the radial growth for *A. fumigatus*, instead of promoting it, (Fuller et al., 2013). A likely explanation is that the mixed spectrum lights used in their experiment led to different responses in the species tested. These influences were then integrated by themselves as a cooperative effect (Cheng et al., 2012). The red and blue light was shown to influence Aspergillus in previous studies, depending on the specific species (Schmidt-Heydt et al., 2011; Cheng et al., 2012). Treatment with multiple wavelengths of light may have a more significant regulatory effect on growth, using monochromatic blue could not reach the extent of white light on growth inhibition in *A. fumigatus* (Fuller et al., 2013).

When *A. versicolor* was incubated under intense light, there were changes in morphology and biological process. While variations in stalk shape and structure had been noted, it is unclear how the mould itself determines the initiation of sporulation. A study of *A. nidulans* suggested that the initiation of sporulation is controlled in two pathways; sporulation could occur as a programmed event or in response to environmental stress (Adams et al., 1998). The difference in sporulation in this study may be a protective mechanism when *A. versicolor* was exposed to light-relevant stress or other environmental stress.

Overall, these findings demonstrated that the presence of light with different colour temperatures or intensities did not influence spore germination of *A. versicolor*. Light exposure could alter the morphology, more importantly, reduce the radial growth rate; moreover, incubation under cool lighting had a strong inhibitory. A significant biological feature to note was that the initiation of sporulation occurred earlier under darkness or weak light, which did not occur at strong light intensity of 1050 lx. These results suggest the role of lights found indoors in regulating the growth and sporulation of *A. versicolor*.

5. References

- Adams, T.H., Wieser, J.K., and Yu, J.H. (1998). Asexual sporulation in Aspergillus nidulans. *Microbiology and molecular biology reviews*, 62(1), pp.35-54.
- Bastholm, C. J., Madsen, A. M., Andersen, B., Frisvad, J. C., and Richter, J. (2022). The mysterious mould outbreak-A comprehensive fungal colonisation in a climate-controlled museum repository challenges the environmental guidelines for heritage collections. *Journal of Cultural Heritage*, 55, pp.78-87.
- Bühler, R.M.M., Müller, B.L., Moritz, D.E., Vendruscolo, F., de Oliveira, D. and Ninow, J.L., (2015). Influence of light intensity on growth and pigment production by Monascus ruber in submerged fermentation. *Applied biochemistry and biotechnology*, 176(5), pp.1277-1289
- Cheng, C.W., Chen, C.K., Chang, C.J. and Chen, L.Y. (2012). Effect of colour LEDs on mycelia growth of Aspergillus ficuum and phytase production in photo-fermentations. *Journal of Photochemistry and Photobiology B: Biology*, 106, pp.81-86.
- Cheong, K.K., Strub, C., Montet, D., Durand, N., Alter, P., Meile, J.C., Galindo, S.S. and Fontana, A., (2016). Effect of different light wavelengths on the growth and ochratoxin A production in Aspergillus carbonarius and Aspergillus westerdijkiae. *Fungal Biology*, 120(5), pp.745-751.
- Fuller, K.K., Loros, J.J. and Dunlap, J.C. (2015). Fungal photobiology: visible light as a signal for stress, space and time. *Current genetics*, 61(3), pp.275-288.
- Fanelli, F., Schmidt-Heydt, M., Haidukowski, M., Susca, A., Geisen, R., Logrieco, A. and Mulè, G. (2012). Influence of light on growth, conidiation and fumonisin production by Fusarium verticillioides. *Fungal biology*, 116(2), pp.241-248.
- Grant, C., Hunter, C. A., Flannigan, B., and Bravery, A. F. (1989). The moisture requirements of moulds isolated from domestic dwellings. *International Biodeterioration*, 25(4), pp.259-284.
- Heseltine, E., and Rosen, J. (2009). 'Moisture control and ventilation Mould and mites as indicators of building performance' in Heseltine, E., and Rosen, J. (ed.) WHO guidelines for indoor air quality: dampness and mould. Denmark: WHO, pp.37-41.
- Jarvis, B.B. and Miller, J.D. (2005). Mycotoxins as harmful indoor air contaminants. *Applied microbiology and biotechnology*, 66(4), pp.367-372.
- Johansson, P., Ekstrand-Tobin, A., Svensson, T., and Bok, G. (2012). Laboratory study to determine the critical moisture level for mould growth on building materials. *International Biodeterioration and Biodegradation*, 73, pp.23-32.
- Kanchongkittiphon, W., Mendell, M.J., Gaffin, J.M., Wang, G. and Phipatanakul, W. (2015). Indoor environmental exposures and exacerbation of asthma: an update to the 2000 review by the Institute of Medicine. *Environmental health perspectives*, 123(1), pp.6-20.
- Mendell, M.J., Mirer, A.G., Cheung, K., Tong, M. and Douwes, J., (2011). Respiratory and allergic health effects of dampness, mold, and dampness-related agents: a review of the epidemiologic evidence. *Environmental health perspectives*, 119(6), pp.748-756.
- Panasenko, V.T. (1967). Ecology of microfungi. *The botanical review*, 33(3), pp.189-215.
- Quansah, R., Jaakkola, M.S., Hugg, T.T., Heikkinen, S.A.M. and Jaakkola, J.J. (2012). Residential dampness and molds and the risk of developing asthma: a systematic review and meta-analysis. *PloS one*, 7(11), e47526.
- Schmidt-Heydt, M., Rüfer, C., Raupp, F., Bruchmann, A., Perrone, G. and Geisen, R. (2011). Influence of light on food relevant fungi with emphasis on ochratoxin producing species. *International journal of food microbiology*, 145(1), pp.229-237.
- Vacher, S., Hernandez, C., Bärtschi, C., and Poussereau, N. (2010). Impact of paint and wallpaper on mould growth on plasterboards and aluminum. *Building and Environment*, 45(4), pp.916-921.
- Viitanen, H. (1994). Factors affecting the development of biodeterioration in wooden constructions. *Materials* and *Structures*, 27(8), pp.483-493.