STUDY REPORT

A PILOT STUDY TO CHARACTERISE THE BIOCONTAMINANT CLOUD WHILE DECLADDING "LEAKY BUILDINGS"

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Malcolm Cunningham, Caroline Shorter, Nick Waipara and Julian Crane

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Preface

This report examines the exposure of remediators of leaky building to the biocontaminant they are subjected to as they remove the orignal cladding of the building they are remediating.

Acknowledgments

Note

This report is intended for researchers and all those involved in the process of remediating leaky buildings

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Reference

Abstract

There are a large number of "leaky buildings" in New Zealand with a repair bill estimated to be at least $11 billion. As claims for these damaged buildings are settled, remediation has begun. In remediating these buildings, as workers remove the old cladding they are exposed to a cloud of biocontaminants sourced from the very wet deteriorated building materials inside the wall.

A pilot study was undertaken to develop methods to characterise this biocontaminant cloud, assess its content and toxicity, assess the exposure of remediators to this cloud, and understand the cloud's dispersion.

Three units in apartment complexes were studied before, during, and for the first two units, after remediation. Biocontaminant spore trap, culturable air sampling and mycotoxin sampling was undertaken at each visit, including following the remediators around as they declad the building and sampling in real time the air close to the remediators. For the culturable fungi, each recognisable taxonomic unit (RTU) was identified to genus or species level using standard identification methods. Where possible, some representative RTU identifications of frequently obtained taxa were carried out to species level using DNA molecular techniques.

The clouds were found to be a complex, allergenic, pathogenic and toxigenic mix of biocontaminants, including very high levels of *Stachybotrys*, *Cladosporium*, *Pencilium* sp., *Aspergillus* sp., *Alternaria/Ulocladium*, *Epicoccum*, gram positive and gram-negative bacteria including *Bacillus*, *Pseudomonas* and others; spore clusters, hyphal fragments, *Basidiomycete*; and particulates consisting of bacterial clusters, siliceous particles and amorphous particles. Potentially pathogenic species *Aspergillus fumigatus* was frequently detected. Other toxigenic species present included*; Stachybotrys chartarum, Aspergillus flavus, Paecilomyces, Pencilium chrysogenum* and *Pithomyces chartarum*.

The outdoor pre-remediation background levels of fungal spores was around 500 to 800 CFU/m³. During remediation there was up to a 7-fold increase in indoor viable spores with a maximum level found of 3610 CFU/m³. The maximum level found close to the remediators was 6866 CFU/m^3. These counts showed large spikes assumed to coincide with the manual tearing off of the cladding, which entrains biocontaminants from the wall behind, and into the air in the vicinity of the remediator. The maximum level found was found was 6.36 mg/m^3 for $PM_{2.5}$ and 3.1 mg/m^3 for PM_{10}. Particulate counts fell to low levels when the workers ceased remediation activities.

Air samples were obtained with the purpose of identifying and quantifying the mycotoxin trichothecene, using 2 L sampling pump and a SKC button sampler and quantified with an ELISA analysis. Before remediation the mycotoxin trichothecene was measured indoors at very low levels but during remediation indoor levels reached very high values, of up to 0.69 ppb. High levels of trichothecene were also found on the gloves of the remediators and in the pieces of the old cladding handled by remediators.

CFD modelling and the temporal behaviour of the particulate data, showed that the presence of a temporary canopy and heavy plastic curtaining surrounding the building for containment purposes, had the effect of retaining each cloud episode in the vicinity of the remediator at high concentrations for a few seconds, and then dispersing it under the eaves of the canopy.

Nasal samplers and measurements of mycotoxins within the protective equipment showed that dust masks and respirators were only moderately effective in protecting the remediator from this toxic cloud. There were many opportunities for mycotoxins on equipment and clothing to be spread around the worksite, and even beyond.

New techniques trialled in this pilot study that worked well included nasal sampling, swabbing the interiors of respirators and disposable masks and testing for the presence of spores and bacterial levels, and rapid serial air sampling around the remediators using agar plates to pick up changing levels of spores in the air over short time periods.

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Introduction

During the 1990's and early 2000's many New Zealand dwellings were built using new construction methods and materials. These included use of monolithic claddings, untreated timber framing, new Mediterranean design styles frequently lacking eaves, having flat roofs and balconies incorporated within the structure of the houses, all of which were often combined with a failure in the workmanship in using the new materials. As a result many of the new constructions have not withstood the weather, allowing water to seep into the building envelope, without adequate means of the water re-escaping. These buildings are known in New Zealand as "leaky buildings".

One estimate is that there are 40,000 to 90,000 (Howden-Chapman et al. 2009 affected with a repair cost of at least $11 billion (stuff.co.nz)

In 2002 the Building Industry Authority (now part of the Ministry of Business, Innovation and Employment) commissioned the Weathertightness Overview Group to report on the situation. They made 20 key recommendations which helped to reform the building code, and largely halted the creation of these substandard buildings (Hunn et al. 2002).

These failures have resulted in gradual moisture damage, over longer time frames (up to 10 years or more) and as a result, have lead to severe damage to the structural integrity of the building and extensive accumulation of microbial damage to inner walls, ceilings and floors. The levels of microbial damage observed in leaky buildings are unprecedented when compared to other types of moisture damaged New Zealand houses.

While much emphasis has been placed on litigation for home owners and preventing future leaking in buildings, very little research has been conducted into the health implications to the occupants or remediators of a leaking home, even though indoor fungi can significantly affect respiratory health (IOM 2004, Fisk et al. 2007).

The most obvious threat to occupiers and remediators is the widespread and inevitable presence of *Stachybotrys chartarum,* particularly in the walls of these leaky buildings where it thrives under the very wet conditions. More than just allergenic, it releases particularly dangerous toxins that have been implicated in pulmonary bleeding including fatalities (Montana et al. 1997)

Aside from *Stachybotrys* there are always many other fungi species in leaky buildings most of which are allergenic and their spores and fungal fragments pose a health threat to susceptible individuals. There is a large international literature (Mendell et al. 2011, Bornehag et al. 2001, Singh 2010) on the ill-health effects associated with mould including eye, nose and throat irritation, cough, wheeze, respiratory tract infections and asthma. Other reported symptoms include depression, fatigue, arthritis, headache, fever, vomiting, sore throat, and rhinitis.

In particular for leaky buildings with *Stachybotrys* there is strong anecdotal evidence within New Zealand of immediate negative health impacts on remediators, even including anaphylactic shock (Howden-Chapman et al 2009) if a remediator is not adequately protected from these biocontaminants; but the size of the problem is

unknown. It has been estimated that the health costs of leaky buildings to the occupants are $25.9 million per year in New Zealand (Howden-Chapman et al. 2009)

The lack of scientific studies on remediators health is of some concern and needs to be addressed, especially in the New Zealand context, considering the large number of leaky buildings that will need to be remediated in coming years - estimates place the number of homes affected at somewhere between 40,000 to 90,000 (Howden-Chapman et al. 2009). To remediate these buildings workers will be coming into contact with the contaminated materials with increasing frequency. Some contractors concentrate solely on the de-cladding process exposing themselves to frequent high levels of microbial contaminants, which are potentially allergenic or toxic substances. This particular exposure represents a new and emerging hazard for many New Zealand builders many not trained in this particular type of work.

The health of building occupants is also of concern. Recent evidence also suggests that there is high risk of respiratory and other health effects from indoor fungal exposure (Fisk et al. 2007, Fisk et al. 2010, Mendell et al. 2011), which may also apply for occupational exposure. Other high exposure workplace environments such as farms have been found to have an occupational risk of increased health issues, thought to be contributable to microbial contaminants (Eduard 2009). Additionally the ecology and biodiversity recorded from New Zealand environments represents a discreet microbial population assemblage not recorded elsewhere (Waipara et al. 2002). For example, the mycotoxin profiles of fungal isolates in the New Zealand environment are unique with often deleterious impacts (Di Menna et al. 2009, Walter et al. 2008, Lauren et al. 1988). The unknown potential of these communities to also produce deleterious impacts on human health remains a significant knowledge gap in New Zealand.

Leaky buildings are remediated by taking off the external wall cavity and rebuilding the wall with a different design which includes a cavity to drain and ventilate away unwanted water.

During remediation of a water damaged home there is the potential for a very high exposure of the remediator to fungal contaminants, as most of the work carried out during remediation uses hands-on forcible removal of contaminated materials. Spores are designed to become airborne, which they do during a removal event. Fungi produce masses of spores, and even a relatively small volume of fungal contamination can contain many millions of spores. Consequently remediators are exposed to a large shower of biological based contaminants during the removal phase resulting in clouds of spores and fungal fragments, some quite dangerous, entering the workers breathing zone.

Less dramatically and more insidiously house occupants are exposed to these biocontaminants finding their way from the envelope to indoors in unremediated houses, and if present, will also be exposed to the cloud of biocontaminants at remediation. They may also be exposed post-remediation to the remnants of the cloud that have spread throughout the house, posing a health risk during and after remediation for an unknown period of time. These size of these occupant pre- and post- exposures are unknown but of real concern.

This potential for exposure to microbes for building remediators has not previously been examined in the New Zealand setting, and has only rarely been looked at in overseas research (Rautiala et al. 1996, 1998, 2004 - from Finland). Most overseas remediation is instead concerned with occupant health, and fixing leaks to roofs, pipes or to fix sudden moisture damage associated with flooding events (e.g.

Hurricane Katrina 2005, Brisbane floods 2011), which represent very different types of microbial events to the 'leaky building' situation. Rautiala (et al. 1996) conducted a study which was similar in nature to ours, investigating the elevated levels of spores in three homes during mould remediation in Finland. Although the study only looked at surface contamination of fungi (rather than building cavity contamination as in our study) they found fungal concentrations increased from $10^3 - 10^5$ spores/m³ to 10^5 to 10^6 spores/m³ during building remediation activities. However in the Finnish study sampling was carried out on filters, giving no real-time measurement of spores, and few replicates per house, so no indication could be given about how spore levels changed over the course of remediation, and whether certain remediation activities caused greater spore release than others. Further studies by Rautiala et al. (1998, 2004) included whether using dust control techniques could reduce spore exposure, and whether mycobacteria were released during the remediation of the buildings. In contrast to the work by Riautiala, et al., our study has used real time evaluation of fungal spores in the immediate vicinity of the remediators, taking measures every 30 seconds, to gain a greater understanding of the spore cloud that workers may be exposed to, and what activities lead to greater elevation of spores.

Current measures to prevent remediator exposure include use of face masks, gloves and suits, but it is not known how widely and effectively these are used, and whether they are adequate in preventing inhalation of such large quantities of spores.

The purpose of this pilot study was to establish measurement techniques and investigate potential exposures of building remediators to fungal components during the remediation process in three case study homes.

Personnel

The Study was lead by Malcolm Cunningham, a Principal Scientist at BRANZ. The mycology study team consisted of: Caroline Shorter an indoor environment researcher based at Otago University, Wellington School of Medicine; Nick Waipara, a mycologist based at Auckland Regional Council; and Nigel Copas and Adrienne Burnie mycologists based at Biodet Services, a mycology research laboratory who carry out fungal assessments of leaky homes commercially.

Aims

Primary Aim

Characterise the biocontaminant cloud emitted as the cladding is removed, assess its content and toxicity, assess the exposure of remediators to this cloud, and understand its dispersion.

Secondary Aims

- assay indoor airborne mould biocontaminant levels before, during and after remediation to understand exposures of occupants of unremediated leaky houses.
- investigate what protective equipment was currently used by the remediators and obtain a qualitative understanding of its effectiveness
- observe the remediators work practices and make recommendations on how these might improve to reduce exposure to biocontaminants and toxins

- where needed develop and pilot new techniques to obtain the above data

Methodology and research design

Identification of houses
Several remediation companies working in the area of leaky building work were contacted. Cove Kinloch, based in Newmarket in Auckland, were able to provide properties that were being remediated in the timeframe needed. They were able to provide us with apartment units in two apartment complexes. Two units were sampled within the first complex, and one in the second.

Experimental Design
In the first complex each unit under study was visited three times, while the unit 3 in complex two was visited twice, see Table 1. The first visits were undertaken before removing the original cladding, the second during the cladding removal operation, and the third a few weeks after the new cladding had been fixed.

Biocontaminant spore trap, culturable air sampling and mycotoxin sampling was undertaken at each visit as detailed below. Additionally to understand how remediators are exposed to the shower of biocontaminants a computational fluid dynamics (CFD) was constructed allowing the air currents around the remediator to be modelled and then the trajectory of the particulates followed as they are transported by these currents.

	Pre-remediation	During remediation	Post-remediation
Unit 1	8/3/2011	4/4/2011	5/7/2011
Unit 2	13/5/2011	30/5/2011	3/9/2011
Unit 3	31/5/2012	4/7/2012	NA

Table 1: Timetable of visits to the Units

As the main aim of the study was to examine the nature, exposure to, and dispersal of, the biocontaminant cloud created when showers of biocontaminants are released from the wall structure as it is declad, intervals between sampling of the air around the remediators were made as short as practical, around 30 seconds.

Biocontaminant sampling regime – Biodet Ltd

Biodet Services Ltd carried out routine fungal sampling at all visits, to assess exposure levels of contamination in a number of areas within the units. Spore trap samples and culturable air samples were taken from the lounge, bedroom and outdoor air. Mycotoxin sample testing was taken from the bedroom area before and after remediation (but not during). Resultant samples were cultured where appropriate and analysed by Biodet technicians.

Sampling regime – Wellington School of Medicine

Two MAS-100 eco (MERCK Ltd) units were used to collect spores onto agar plates by drawing a known volume of air through a series of fine holes (400 total), see Figure 1, similar to an Andersen sampler. A range of volumes of air were selected, 50, 100 and 500 L samples were taken and two types of agar used viz DG18 (Fort Richard Laboratories) and low nutrient agar (Biodet Ltd). Ten replicates of each agar were taken at each sampling location, alternating between the two agar types,

starting with DG18. Each 100 L sample took approximately 1 minute to collect. Indoor and outdoor samples were collected with the units simultaneously, before remediation had started and during remediation.

Figure 1: MERCK MAS-100 Eco sampler.

The second type of air sampler used in the study was a TSI SidePak AM510 laser photometer, see Figure 2. This measures aerosol mass concentrations of $PM_{2.5}$ and PM_{10} in real time by drawing a sample of air across a laser and closely estimating particulate mass concentrations. These samplers are widely used in industrial workplaces to assess human exposure to particulates, specifically those of a respiratory and thoracic size. $PM_{2.5}$ or PM_{10} levels were taken every 2 seconds and averaged over 10 seconds over the recording time period. Zero calibrations were conducted prior to each sampling day, and a background level of particulates were collected for at least 15 minutes prior to remediation or building activity began on site.

Figure 2: SidePak AM510 sampler

During the initial sampling phase the MAS-100 eco and AM510 samplers were set up on tripods so the sampling inlets were at a height of 1.5 m outside the dwelling. However, in the second phase of sampling the units were carried by hand by the researchers, so that they could be held closer to the remediators work zone and could more easily follow the remediators around the building, see Figure 3.

Figure 3: SidePak sampler held near the working space of the building remediators

All resultant microbial agar plates were grown under controlled laboratory conditions for 7 days (20°C and 65% RH) before colony forming unit counts were taken. Samples were then refrigerated (4°C) until recognisable taxonomic unit (RTU) counts were undertaken.

Due to the very large number of individual colony forming units isolated during the survey, for example ~400< CFUs could be counted on one plate alone;   identification of every plate was logistically impractical.   Therefore taxonomic diversity was enumerated by counting total number of recognisable taxonomic units (RTU) per plate using previously described methods for New Zealand fungi (Walter et al. 2007, Waipara et al. 2002). Post refrigeration, isolation plates were incubated back at room temperature (18-22°C) for up to 7 days to allow optimum development of diagnostic morphological characters such as sporulation, mycelial pigmentation and vegetative hyphal growth.

A sub-sample of each representative RTU colony type obtained on the plates were sub-cultured onto potato dextrose agar (PDA) or potato carrot agar (PCA) for further diagnostic examination. Each RTU were identified to genus or species level using standard identification methods for common indoor fungi (Samson et al. 2004, 2010). Where possible, some representative RTU identifications of frequently obtained taxa were carried out to species level using molecular techniques. Fungi were identified by comparing genetic sequences of their ITS region (using primers ITS1-F and ITS4) and comparing these to sequences deposited at the GenBank database (White et al. 1990; Gardes & Bruns 1993).

In addition to quantifying levels of microbial contamination in leaky buildings (CFU levels), it is equally important to assess what the diversity and type of microorganisms were present at each sampling time. It is critical to detect and differentiate ubiquitous benign species from deleterious species during remediation. RTU assessment informs potential worker exposure to previously known pathogenic, allergenic and toxigenic species and therefore potential occupational health and safety risks that exist during remediation and building projects.

Building Material Samples

Samples of contaminated building materials which had been extracted by the remediators from the three Units were collected, including parts of the gypsum board, building paper, and insulation. These samples were then examined under the microscope by Biodet and Dr Nick Waipara for the fungal community assemblages.

Mycotoxin detection – trichothecene

Mycotoxins are naturally occurring substances produced by fungi as secondary metabolites (Ingle et al. 2010). The trichothecene mycotoxin is highly cytotoxic and can be produced by fungal species such as *Fusarium* and *Stachybotrys chartarum* (Ingle et al. 2010, Waipara 2010). It can be ingested both dermally and orally.

Air samples of trichothecene were taken using 2 L sampling pump and a SKC button sampler, see Figure 4, loaded with cellulose filters. Two sampling pumps and SKC button samplers were used in two locations prior to remediation, one was placed in the living area and one was placed to measure the air inside a wall cavity in the living room. The samplers were left to run for 24 hours, collecting 48 litres of air. The resultant samples were then frozen until ELISA analysis could be carried out.

Additionally in Unit 1 and 2 samples were taken during remediation, one inside the Unit, and one outside. These samplers were left running while remediation activities were carried out, taking 4 L and 3 L samples of air respectively. Due to the short sampling time available for remediation in Unit 3, airborne mycotoxin samples were not taken during remediation. Samples of contaminated building paper and gypsum board were also taken for analysis of mycotoxin content. Samples were also taken from the gloves of a researcher who had been handling the building material, by touching the gloves to a cellulose filter. All samples were stored frozen until analysis could be conducted (-20°C). ELISA analysis of trichothenes were conducted at the Wellington School of Medicine by Rob Siebers using methods outlined previously (Park et al. 1996). Several dilution levels were chosen to measure both air, building and glove samples.

Figure 4: SKC button aerosol sampler (SKCinc.com)

Workers' exposure

Nasal samplers

Maggie Nasal Samplers (supplied and designed by Dr Euan Tovey, University of NSW) were worn to assess levels of inhaled spores. For the remediation of Unit 1 the sampling, shown in Figure 5, were filled with 15 µl of melted Vaseline. Two nasal

samples were worn by each researcher, one prior to remediation, and one during remediation, with no other safety equipment masks worn, as was observed to be the case with the builders on Unit 1. The resultant samples were examined by Biodet Services for spore identification under a microscope. For Units 2 and 3 the cups were encased in a double sided adhesive tape outer layer, which allowed easier identification of spores than the Vaseline, when viewed at higher magnification. In Unit 2 one set of nasal samplers were worn during remediation with no masks, and then a further set were worn during remediation with either respirators or a dust mask, to see whether these reduced the types of spores inhaled during remediation. In Unit 3, because of a much reduced remediation sampling time, nasal samples were worn during remediation using the two NZ safety recommended respirators. Resultant nasal plugs were again examined for spore presence by Biodet Services Ltd.
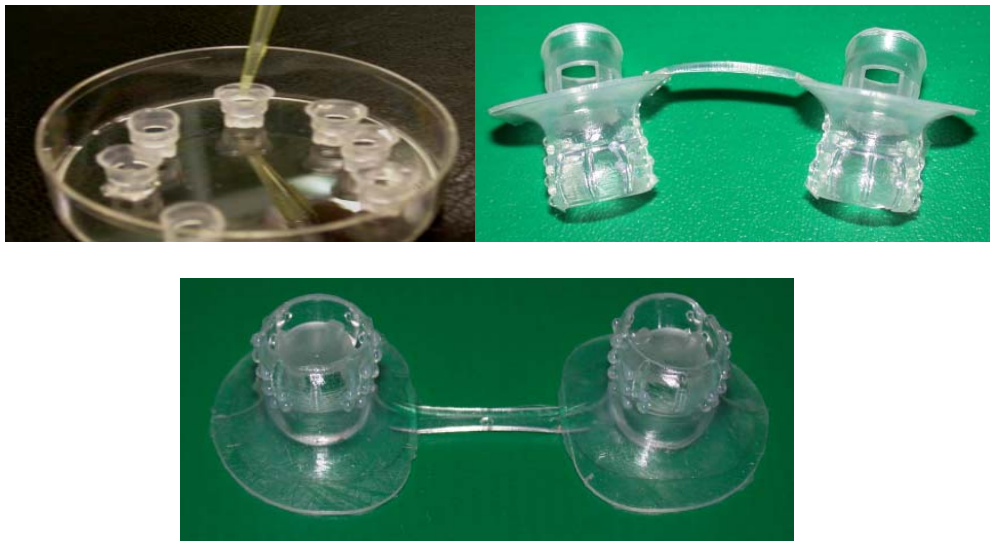
Figure 5: The nasal sampler

Figure 6: Nasal Samplers being worn onsite

Mask Filters

Both the respirator filters (Unit 2) and the disposable mask (Unit 2) were swabbed and plated onto agar, then examined by Biodet under a microscope for the presence of spores and bacterial level assessments.

Provenance of sampling techniques

The techniques chosen in this study are a mix of internationally established techniques (colony forming unit counts, recognisable taxonomic unit counts, microscopy spore counts, particulate matter laser photometer counts) and new techniques (nasal sampling spore inhalation counts, mycotoxin inner wall cavity sampling). While new techniques to measure fungal spores are beginning to emerge (such as using DNA fungal methods), the use of agar plates and microscopy slides are still the most appropriate for giving real time exposures, particularly when levels are expected to rapidly change (Flannigan et al. 2001, Prezant et al. 2008). Filters and dust collection provide only a cumulative exposure – one reading only, which is inappropriate when we need to understand how breathing exposures change within minutes – workers are not inhaling just once every 4 hours.

Computational fluid dynamical modelling

The airflow in and around the apartment complexes was modelled approximately using the multiphysics software COMSOL4.3a.

It must be understood that the CFD model is not for quantitative predictive purposes but for qualitative explanatory purposes. For quantitative prediction the geometry, initial and boundary conditions must be accurately known, and experimental data should be available with which validation can be carried out. In our case none of these pre-conditions are available. Rather currently we have no idea what the biocontaminant cloud might look like, how it evolves and what the consequent exposure of the remediator might be. The CFD model has provided valuable insights towards answering these questions.

Details of the CFD model

The exposure of the remediators is strongly conditioned by the surrounding physical structures.

Figure 7: View of the temporary canopy of long run metal and translucent plastic sheets and plastic curtain surrounding the building being remediated

Figure 8: View of underside of the canopy

Figure 9: Interior view from the upper level scaffolding of the canopy and curtaining

In order to contain the biocontaminants and direct them away from the immediate surroundings the building is contained within a temporary roof of long run steel and translucent long run plastic sheeting and a surrounding heavy plastic curtain, see Figure 7, Figure 8, Figure 9 and Figure 11. This gives rise to very complex air flow paths as the cladding is demolished making it non-obvious how the contaminant cloud is dispersed.

The situation was modelled using the turbulent k-ω turbulent air option of the "Non-Isothermal Flow", and the "Particle Tracing for Fluid Flow" modules from within the multi-physics software, COMSOL, version 4.3a. The non-isothermal module provides CFD modelling coupled with heat transfer in solids, while as its name suggests, the particle tracing module makes it possible to track the flow of particles within the moving air streams.
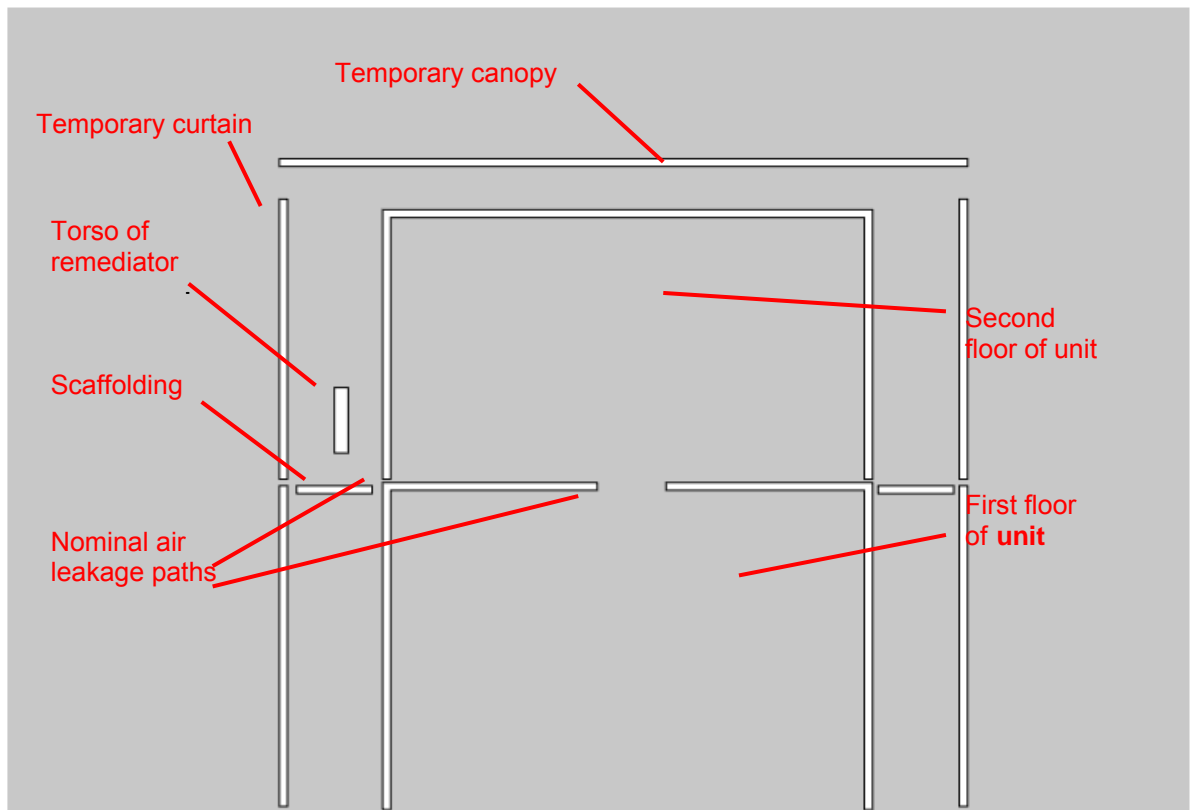
Figure 10: Two dimensional model of the building, canopy and curtaining, and torso of the remediator

The building, canopy and curtain, and remediator are modelled two dimensionally with gaps left between these elements to provide an approximation to air flow paths between them, see Figure 10.
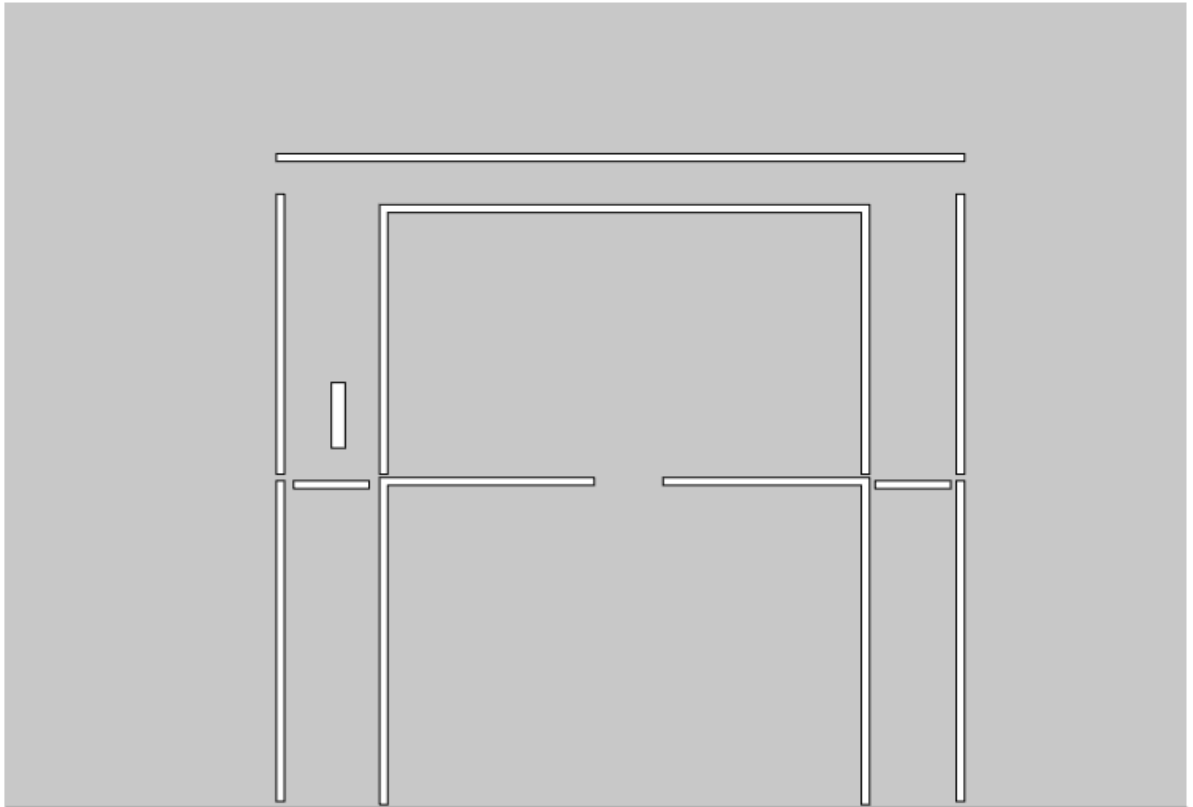
 The torso of the person is represented by a rectangle, 1 metre high and 20cm wide. To provide external wind driving forces the building is placed in a nominal open space. Temperature driving forces are provided by setting the roof canopy to 298 K (24.85 $^{\circ}$C) and releasing heat from the remediator at the rate of 100W, spread around the entire surface of the rectangles representing the torso of the person.

The incoming air is set at 290 K (16.85 $^{\circ}$C) with three different velocities modelled, viz 2 m/s, 0.1 m/s and -1 m/s. Particulates of 10 microns diameter and 1g/cm^3 density (neutrally buoyant) were released and their dispersion modelled. Particles of this size and of densities of the order of 1g/cm^3, have a terminal velocity in the order of 3 × 10^{-3} m/s, which is negligible compared to the modelled air velocities and thus will have their trajectories determined by the air currents only with density differences and settling effects unimportant.

Results

Observational photographs

Figure 11: (Photo 1) Units in Complex 1 prior to remediation and scaffolding – dark areas of water damage are clearly visible as dark areas on the monolithic cladding

Figure 12: (Photo 2) Units during remediation phase in Complex 1 with scaffolding, temporary roof and screens attached

Figure 13: (Photo 3 & 4) The decladding process in Complex 1 – boards of cladding are pulled from the buildings by the remediators. This is thought to be a time of possible high spore exposure. Sampling machines can be seen in the foreground.

Figure 14: (Photo 5) Once cladding is removed water damage is often seen with the disintegration of building paper

Figure 15: (Photo 6 & 7) Building paper and insulation layers are also removed.

Figure 16: (Photo 8 – 10) Cladding removal also shows biodeterioration of the wooden structure of the Unit

Figure 17: (Photo 11) Sometimes this deterioration is sometimes quite severe

Figure 18: (Photo 12 & 13) Inside units fungal, biocontamination from water damage is apparent, particularly around windows

Figure 19: (Photo 14) Fungal contamination from water damage is also apparent on skirting boards and carpets.

Figure 20: (Photo 15) Plastic wrapped carpets. Some owners opt to retain original carpets after remediation to cut down on repair costs.

Figure 21: (Photo 16) Complex 2 apartment block, Unit 3 (located on second floor).

Figure 22: (Photo 17 – 22) All of the building materials damaged by water can support fungal and microbial growth. These pictures demonstrate fungal colonies seen on some of these materials.

Biocontamination of building materials

All three house units showed high levels of biocontamination on all types of building materials being removed during remediative work. Both external cladding materials and internal materials were observed to be colonised by a range visible rot fungi. In all houses fungal colonisation had led to extensive biodeterioration of most types of

materials. Destructive removal of contaminated materials enabled a range of representative samples to be collected and examined.

Stachybotrys chartarum was frequently observed across all buildings. Its distinctive 'sooty' sporulating colonies were often recorded producing a wet rot on obviously water damaged materials such as wallpaper, carpet backing, building paper, particle board, gypsum board/fibre cement, plasterboard and insulation materials (Photos 23 & 24). As *Stachybotrys* was found to be well established and very abundant on most biodeteriorating materials being removed, all workers involved with destruction and disposal of these materials would have been constantly exposed to the fungus during this activity. Presence of large amounts of *Stachybotrys* wet rot is a common bioindicator of water damage and leaks as it is usually only recovered from materials or environments with a very high water activity (a w > 0.90-0.95).

Other species part of these wet rot assemblages included *Chaetomium globosum*, *Aspergillus niger, Ulocladium chartarum* and various unidentified *Penicillium* species. Cellotape samples collected by Biodet also identified that the common airborne contaminant *Cladosporium* was also colonising to contaminated building papers.
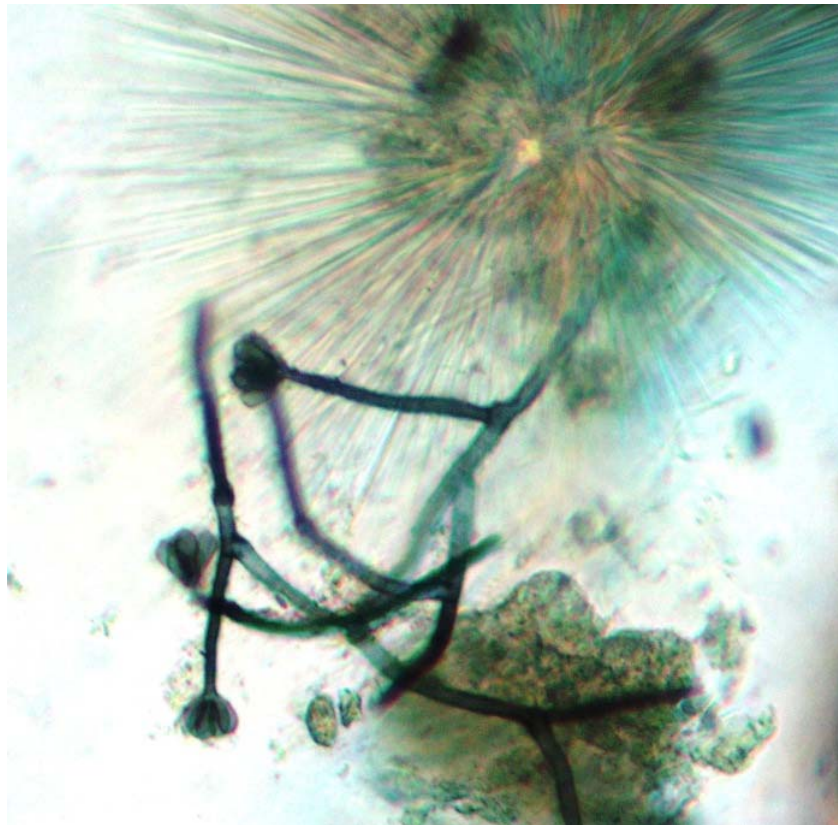
Figure 23: (Photo 23) An example of water-damaged fibre cement abundantly colonised by *Stachybotrys chartarum.*

Figure 24: (Photo 24) Visible 'soot-like' biocontamination of fibre cement (left) and insulation batts (right) by toxigenic fungus Stachybotrys chartarum.

Rhizomorphs and mycelium wood-rotting decay fungi, mostly basidiomycetes, were observed to be associated with severe biodegradation of structural building timbers (Photo 25).   Significant to extreme loss of strength was noted for many of these timber samples with advanced brown rot decay and or occasional fungal soft rot being present.   Such brown rots are very destructive and can occur on both treated and untreated building timbers, as was observed in this project.   A deteriogenic Ascomycetous species *Nodulisosporium* was also recovered from brittle rotting timber.

Figure 25: (Photo 25) Rhizomorphs of a basidiomycete wood decay fungus causing destructive brown rot on timber framing

A species of *Taeoniella,* morphologically most similar to *T. rudus*, was obtained from severely contaminated wet indoor building paper adhering to external timber framing that was abutting garden soil. While the fruiting body of a basidiomycete decay fungus was even observed contaminating synthetic plastic building wraps. Basidiomycetous fungi have been previously reported to have allergenic properties (Bryant & Rogers 1991), particularly those producing fruiting bodies and thus releasing spores (Lehrer et al. 1994). More research is required to determine exposure risks to workers removing wood decay fungi during remediation projects.

Areas of water damaged carpet (Photo 14) also were observed to have been colonised with elevated levels of fungi, including yeasts, and gram negative bacteria. Four swab samples taken from carpets in both house 1 (unit38) and 2 (unit 40) were found to higher than baseline levels of >10,000 bacterial CFUs and >500 fungal CFUs per 10 cm^2. Proliferation of species isolated from water damaged carpet piles included bacteria; *Bacillus, Pseudomonas* and fungi; *Aspergillus, Cladosporium, Chaetomium, Mucor, Penicillium,* and yeasts. Workers could therefore be exposed to these elevated levels of bacteria and fungi whilst undertaking activities within the units including when removing/replacing the damaged carpets and flooring.

Workers removing contaminated building materials across all sampled units were clearly in direct and indirect contact, and thus a potential range of exposure types, with an elevated level of spores and mycelium of a range of building rot fungi. Further research is required to ascertain level of individual exposures and what if any could have deleterious health impacts. Of concern was the well established growth and frequent occurrence of *Stachybotrys chartarum* on a wide range and type of biodeteriorating building materials that were being removed at all three houses. Health and safety precautions should be undertaken at all times when removing items contaminated by this toxigenic fungus.

Viable spore levels during remediation

Levels of viable spores were measured at several time points during the remediation process. In Unit 1 colony forming unit levels taken shortly before remediation activities started indicated that spore levels were approximately equal indoors and outdoors (538 CFU/m³ indoors versus 498 CFU/m ³ outdoors, see Figure 26. Samples taken 30 minutes later, during initial remediation, which started on the lower level of the Unit, show a 7 fold increase in spore levels inside, with 3610 CFU/m³, and a 2.5 fold increase outside with 1264 CFU/m³. These static samples were taken from fixed positions, inside the home on the kitchen bench (1.2 m height), and outside 2 metres away from the walls of the Unit, on a tripod (1.5 m height). Actively following the builders with the samplers outside (approx 1 – 2 m from where they were working), also showed high mean levels of spores, between 1264.5 – 3770 CFU/m³. The error bars indicate that there were higher levels of variability between replicates during remediation activities, suggesting that spore levels were highly variable during this time. Little variation was observed in the levels of spores before remediation began, see Figure 7.
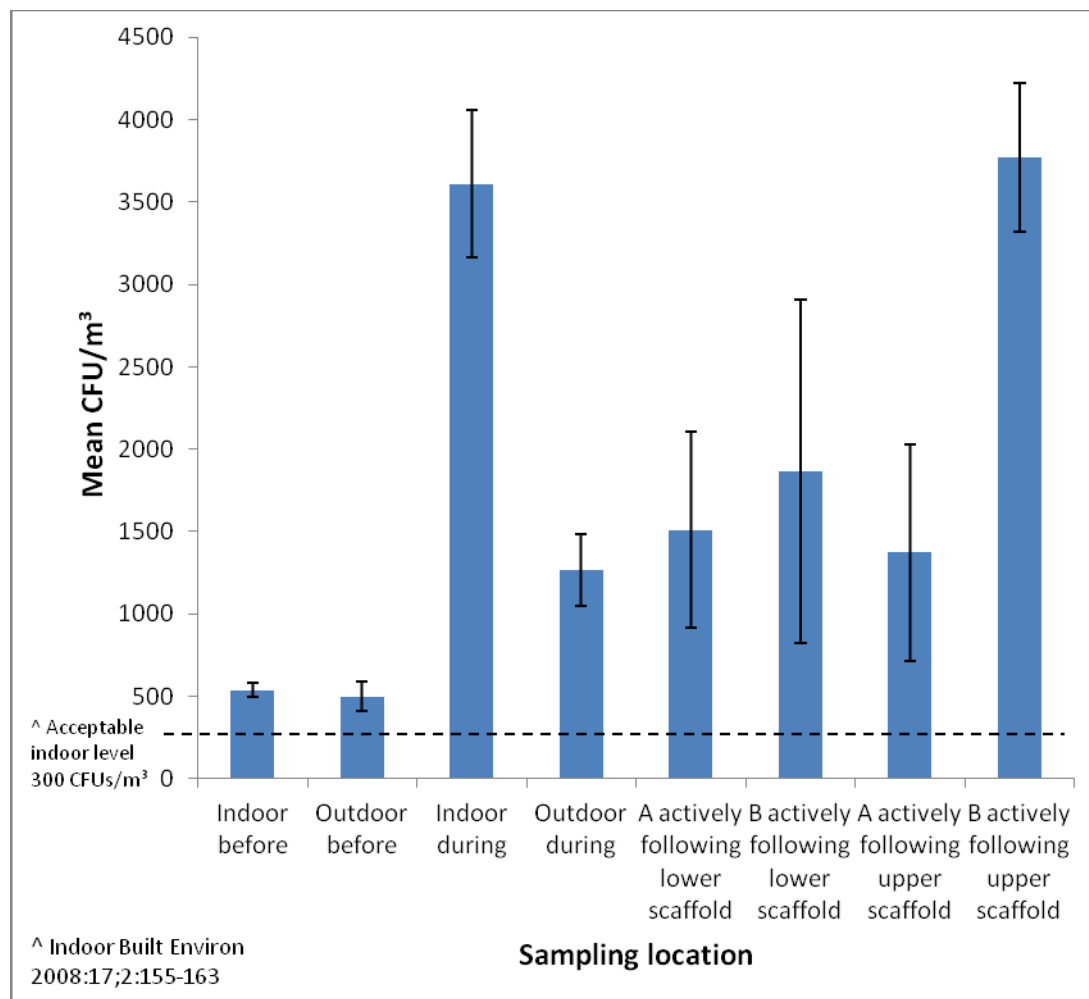
Figure 26: Mean CFU levels taken during remediation of Unit 1 (error bars = ±1.96 × std error around the mean)

The level of spores prior to remediation was higher in Unit 2 than Unit 1, and levels indoors were twice those in outdoor air (1688 CFU/m³ indoors versus 834 CFU/m³ outdoors, Figure 27). Despite levels of spores being higher indoors prior to remediation they only increased by 1.6 times indoors during initial remediation activities (Figure 27: Indoor during). This may have been because unlike Unit 1, remediators started cladding removal at the top of the Unit, furthest away from the indoor sampling in the downstairs lounge area. The levels of spores outside were 5 times higher during initial remediation activities compared to prior to remediation activities beginning. This was higher than in Unit 1, which again may have been due to the starting position of remediation, with remediators directly above the sampling, spores may have been disturbed and fallen down into the samplers from above.

The chart shows CFU/m³ on the y-axis (0 to 8000) and Sampling location on the x-axis with the following categories:

- Inside pre removal
- Outside pre removal
- Inside during removal
- Outside during removal
- Outside Top level scafolding A
- Outside Top level scafolding B
- Outside Bottom level scafolding A
- Outside Bottom level scafolding B
- Inside after break

^ Acceptable indoor level 300 CFUs/m³

^ Indoor Built Environ 2008:17;2:155-163

Figure 27: Mean CFU levels taken during remediation of Unit 2 (error bars = ±1.96 × std error around the mean)

The samples taken when actively following the remediators also indicate high spore exposures occurring outdoors (range 3284 – 6866 CFU/m³) during cladding removal, both on the upper scaffolding and the lower scaffolding, see Figure 27. These exposures were again highly variable. Some of the variability will be due to different positional relationships between the remediator and the sampler held in the hands of the researcher. However, most of the variability will come from spikes of biocontaminant concentration as individual parts of the cladding are torn off the building. These spikes can be quite clearly in the particulate data, discussed below, see also Figure 41 and Figure 42.

A further static indoor sample in the lounge was taken during the smoko break, by which time remediation activity had ceased. This indicated that spore levels had increased indoors compared to those taken during the initial phase of remediation, and also that the levels of spores continued to be high at least 30 minutes after remediation activities had stopped on the home. This is of note because during

lunchtime some workers were observed to sit in the lounge area of this unit to eat their lunches, when no doubt spore levels were still very high indoors.

Background levels of spores taken prior to remediation activities began on Unit 3 were comparable between the indoors and outdoors (487 CFU/m³ versus 453 CFU/m³, see Figure 28, a similar pattern to Unit 1. During initial remediation activities on Unit 3, which being a 1 level home occurred next to the indoor and outdoor samplers, the indoor samples were 6 times higher compared to the indoor pre-remediation levels, indicating a rapid increase in spores had occurred through the vibration associated with cladding removal, see Figure 28.
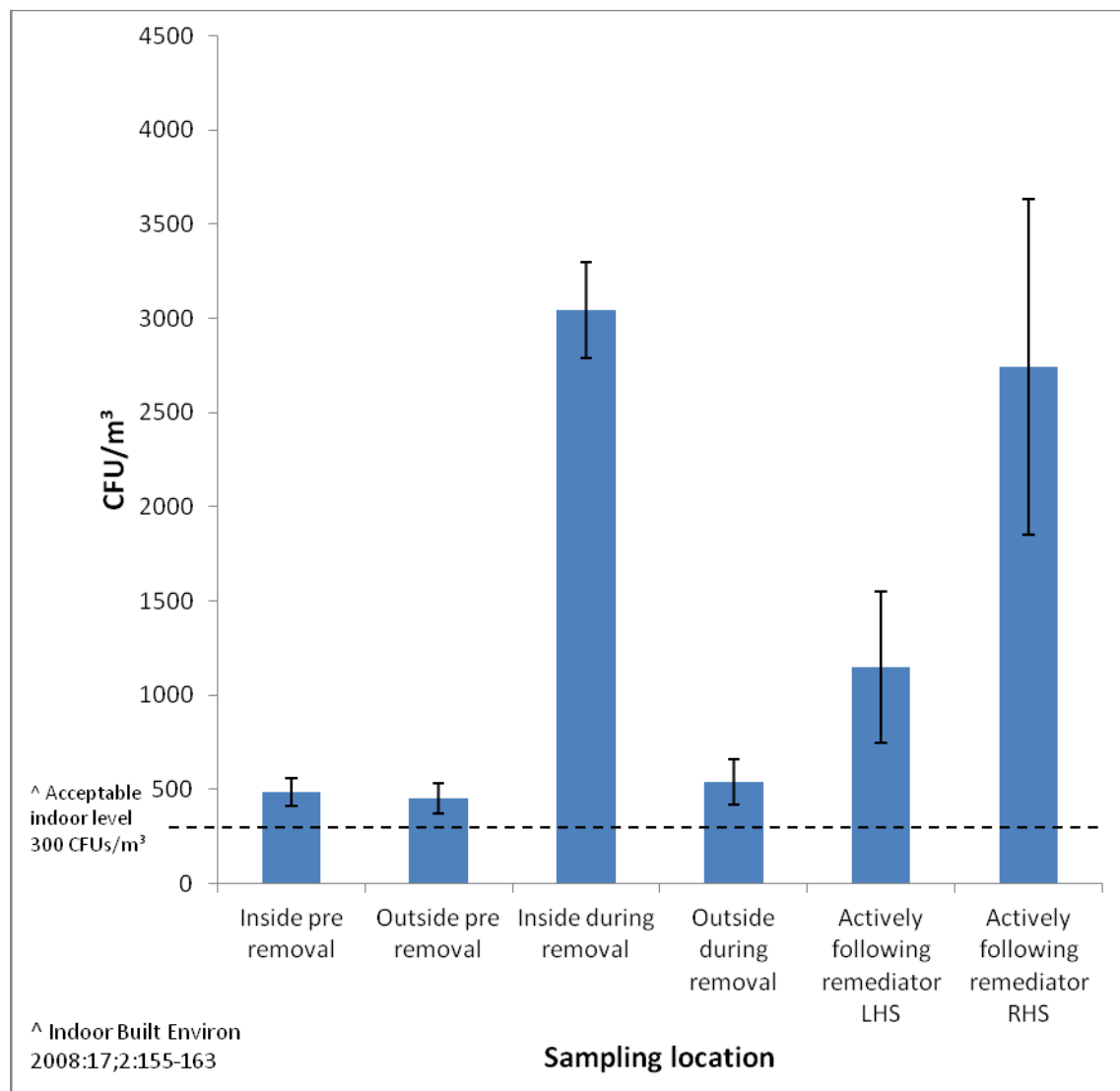
Figure 28: Mean CFU levels taken during remediation of Unit 3 (error bars = ±1.96 × std error around the mean)

However levels in outdoor air during the initial remediation activity only increased slightly compared to the pre-remediation outdoor levels (540.5 CFU/m³ versus 453 CFU/m³, see Figure 28), which may have been due to the positioning of the outdoor sampler unit, which was upwind of the remediation activity. This suggests that spore clouds may be highly influenced by wind direction and may be very different in the breathing space of the remediator compared to 2 metres back. This theory appears to be strengthened by the higher levels of CFU/m³ collected when actively following the remediator, with the samples collected on the right hand side of the remediator

being almost 2.5 times higher than those collected by a second sampler located to the left hand side of the remediator (2743 CFU/m³ versus 1149 CFU/m³, Figure 28). Ideally a sampler held as close to the remediator as possible would yield the best data on potential exposure.

RTU levels during remediation

A relatively small number of taxa (n= 38), to what was actually present were able to be identified to species level from isolation plates (refer to RTU table below). As this total species number is only obtained from a relatively small sub-sample from all plates, the diversity does not represent all culturable isolates observed on plates However, the RTU count does represent the most frequent taxa recovered and therefore most likely to result in exposure to workers undertaking remediation. Most taxa encountered are both widespread and common airborne fungi, and thus have mostly been previously reported from both New Zealand and overseas building surveys.

Taxon Unit	DG18	NA	1	2	3
Absidia sp*	+				+
Acremonium sp	+	+	+	+	
Alternaria alternata	+	+	+	+	+
Aspergillus spp.	+++	+++	+++	+++	+++
Aspergillus flavus*	+	+	+	+	+
Aspergillus fumigatus*	+		+	+	+
Aspergillus niger	+			+	
Aspergillus terreus*	+		+	+	+
Aureobasidium pullulans	+	++	+	++	+
Botrytis cinerea		+	+	+	
Candida spp.		++	+	+	+
Candida albicans*		+			+
Candida peltata*		+	+		
Chrysonillia sitophila*	+	+	+	+	+++
Cladosporium cladosporioides*	++	++	++	++	++
Cladosporium herbarum*	++	++	++	++	++
Cladosporium spp.	+++	+++	+++	+++	+++
Clonostachys rosea (Gliocladium roseum)	+		+	+	+
Cryptococcus spp		++	++	++	++
Cryptococcus laurentii*		++	+	+	+
Cylindrocarpon sp	+			+	
Dreschlera sp	+		+		
Epicoccum nigrum	+	+	+	+	+
Fusarium sp	+		+		
Geotrichum candidum*		++	+	+	+
Mucor sp1		+	+	+	+
Mucor sp2		+			+
Mycelia sterilia spp.	++	++	++	++	++
Oidiodendron sp cf griseum	+			+	
Paecilomyces lilacinus	+		+		+
Penicillium spp.	+++	+++	+++	+++	+++
Penicillium brevicompactum*	+		+		
Penicillium chrysogenum*		+		+	

Penicillium expansum*		+			+
Penicillium glabrum*	+		+		
Penicillium janthinellum*	+	+			+
Phoma spp	+	+	+	+	+
Phoma exigua*		+			+
Phomopsis sp	+	+		+	
Pithomyces chartarum	+		+	+	
Rhizopus oryzae		+	+	+	
Rhodotorula spp.		++	+	+	+
Rhodotorula glutinis*		+	+		
Sporobolomyces roseus	+	+	+	+	+
Trichoderma spp.	+	+	+	+	+
Trichoderma viride*	+	+	+	+	+
Ulocladium chartarum	+		+	+	
Yeasts	+	+++	+++	+++	+++

* Identified by DNA analysis of ITS genetic sequences
+ Relatively low levels of isolation frequencies across all plates: occurred on >20% plates
++ Medium levels of isolation frequencies across all plates: occurred on 20-60% plates
+++ High levels of isolation frequencies across all plates: occurred on >60% plates

Table 2: RTU table. List of fungal taxa identified from isolation plates

In addition to fungal taxa listed above, a large number of bacterial colonies were counted on Nutrient Agar plates, bacterial groups including actinomycete species were present in high levels on all nutrient agar plates, but mostly absent from DG18 which is fungi specific. Future surveys should undertake purification into subculture to allow DNA identification of representative bacteria species associated with leaky buildings. This would give added valuable information on the potential exposure regimes to workers.

Using two types of isolation media was valuable in increasing the range of taxa identified. For example yeasts such as *Candida, Cryptococcus* and *Rhodotorula* were only able to be detected using Nutrient agar (NA), whilst DG18 had useful capacity of enhancing direct morphological identification of fungal species such as *Gliocladium roseus Pithomyces chartarum* and *Ulocladium chartarum*. Future surveys should continue to incorporate broad spectrum isolation media to ensure representative airborne taxa are accurately detected.

Although accurate quantitative RTU analysis was compromised by agar media bias, fast growing saprophytes and overcrowded plates due to isolation clouds, the method was very useful in qualitatively identifying a suite of allergenic, pathogenic and toxigenic fungi were present during remediation activities. For example potentially pathogenic species *Aspergillus fumigatus* was frequently detected. Other toxigenic species present included*; Aspergillus flavus, Paecilomyces, Pencilium chrysogenum* and *Pithomyces chartarum*. Quantified CFU counts supported with RTU differentiation of those counts show the potential of worker exposure to very high levels or airborne clouds of fungal and bacterial propagules.

Comparison of RTU of overall levels between pre remediation isolation plates and during remediation showed a very slight trend of a decrease in the total RTU levels. This was probably due to the plates collected during remediation being largely being overwhelmed by the large number of CFU's, and that the five predominant taxon

groups; *Aspergillus*, Bacteria, *Cladosporium, Penicillium* and yeasts were precluding the identification and isolation of other taxa which were more visible pre disturbance and without clouds of dislodged airborne propagules to mask their presence. Therefore the lower RTU numbers was likely an artefact of plating rather than remediation activity itself. Further monitoring and method development across a large sample size of houses is still required to fully determine RTU levels and subsequent worker exposures.
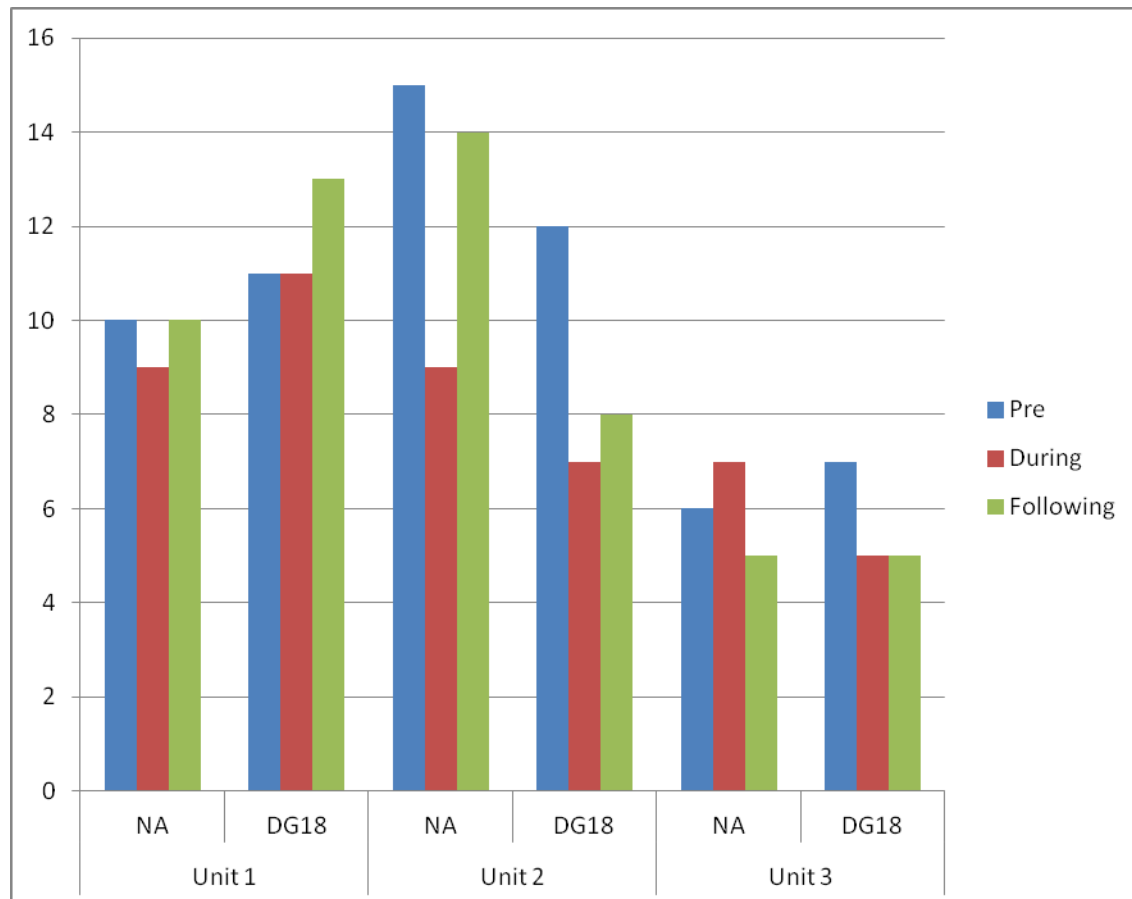
Figure 29: RTU Figure. Levels of common airborne fungi isolated onto Nutrient agar (NA) and DG18 agar pre remediation, during remediation and following workers, across the all three houses.

Indoor exposures

For Unit 1 and 2 we collected data before, during and after remediation. Figure 30 to Figure 39 compare between Unit 1 and 2 the way in which various biocontaminant levels changed before during and after remediation.
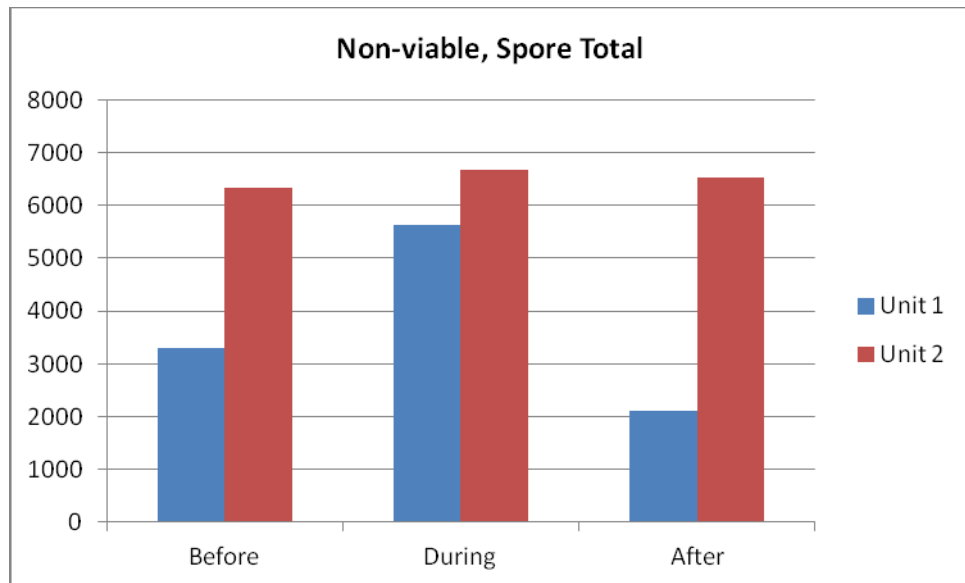
Figure 30: Indoor non-viable spore total counts before, during and after remediation
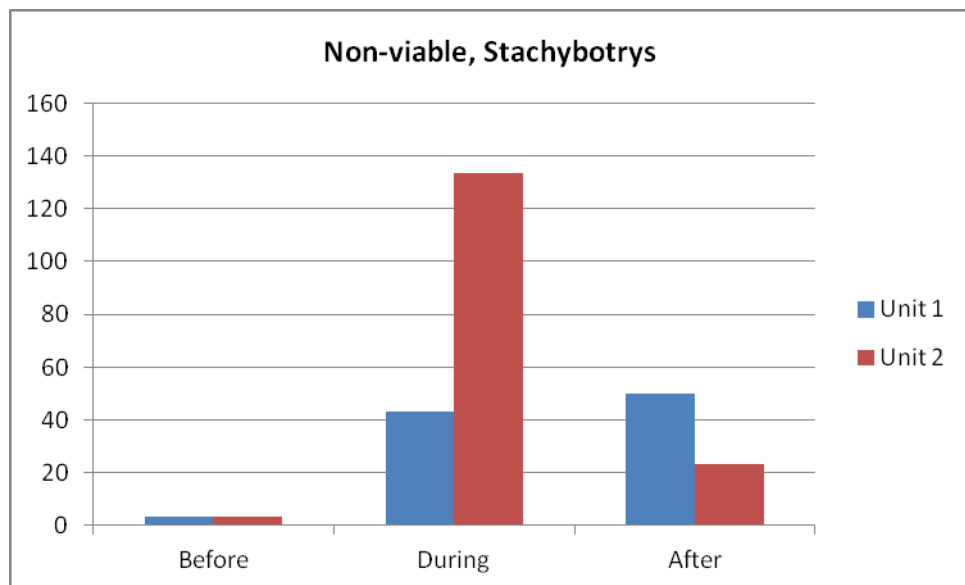
Figure 31: Indoor non-viable *Stachybotrys* counts before, during and after remediation
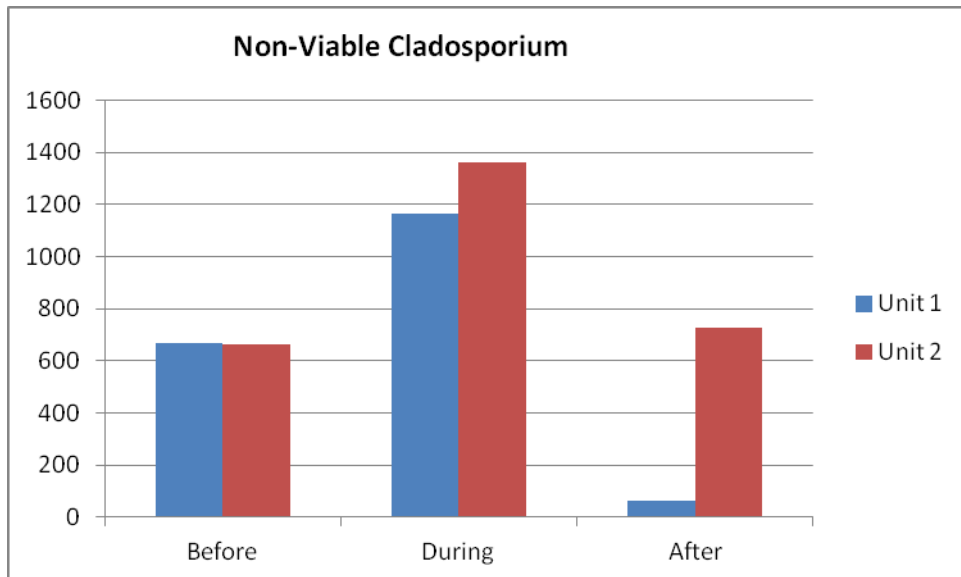
Figure 32: Indoor non-viable *Cladosporium* counts before, during and after remediation
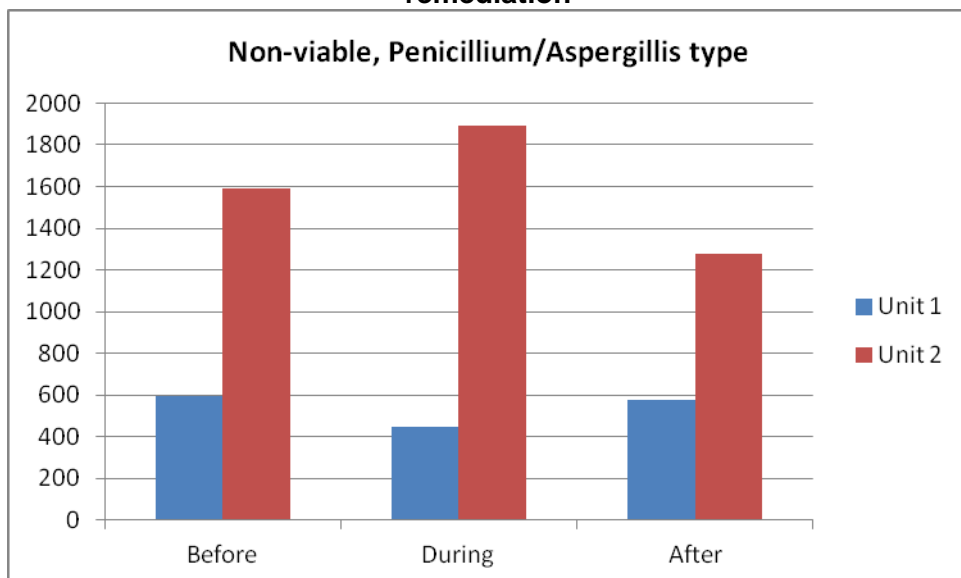
Figure 33: Indoor non-viable *Penicillium/Aspergillus* type counts before, during and after remediation
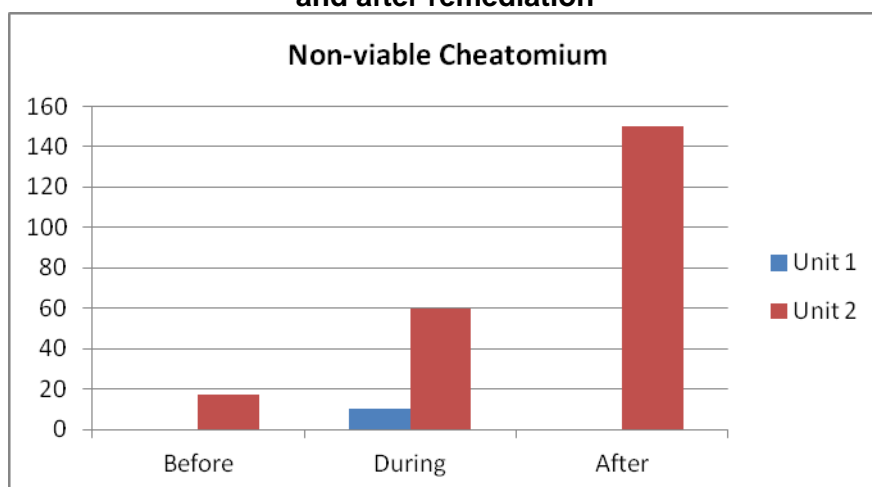
Figure 34: Indoor non-viable *Chaetomium* counts, before, during and after remediation
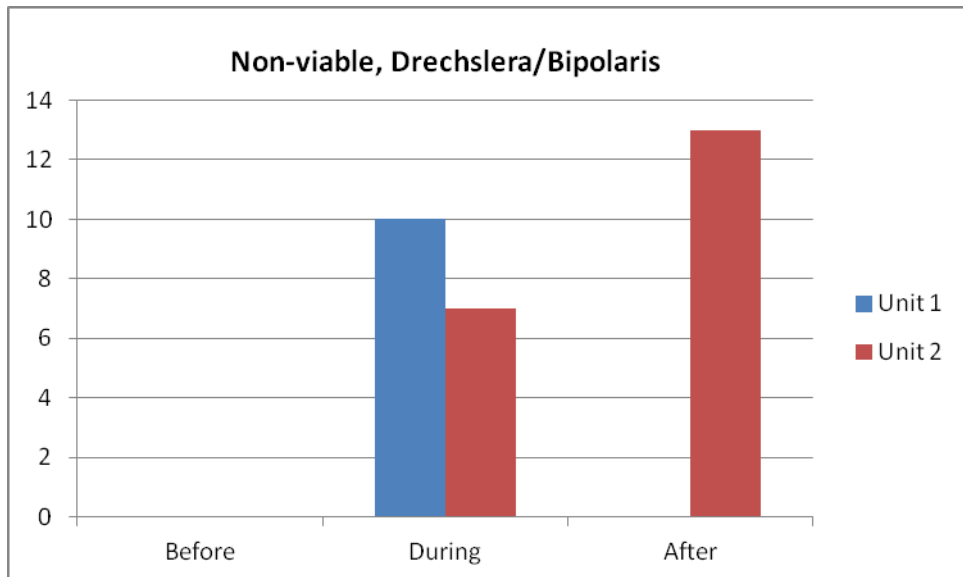
Figure 35: Indoor non-viable *Drechslera/Bipolaris* type counts, before, during and after remediation
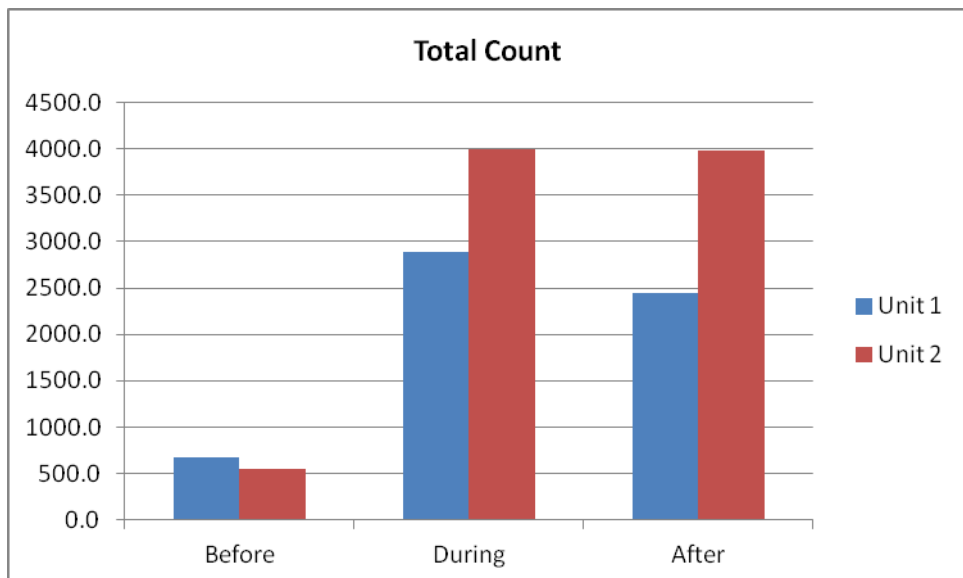
Figure 36: Indoor viable total count, before, during and after remediation
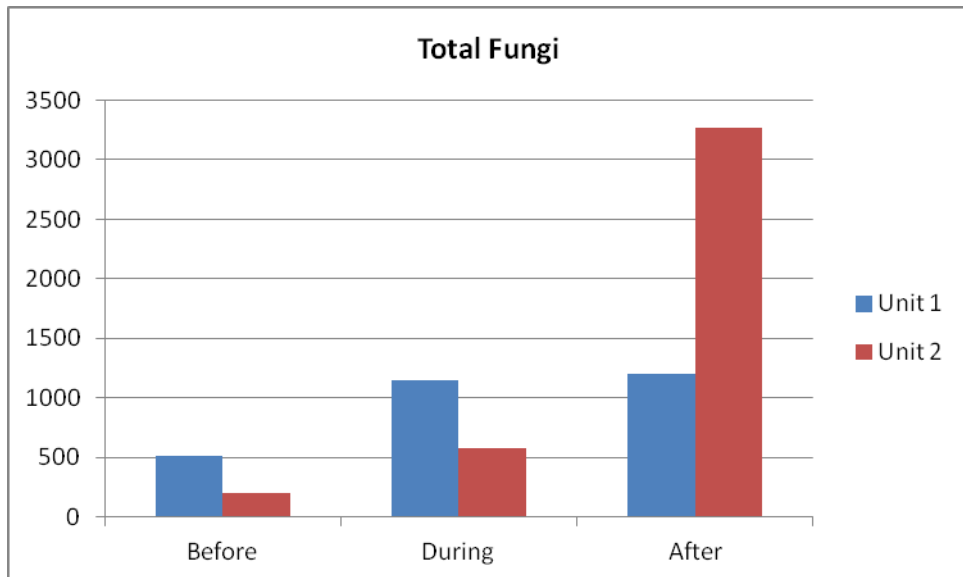
Figure 37: Indoor viable total fungi count, before, during and after remediation
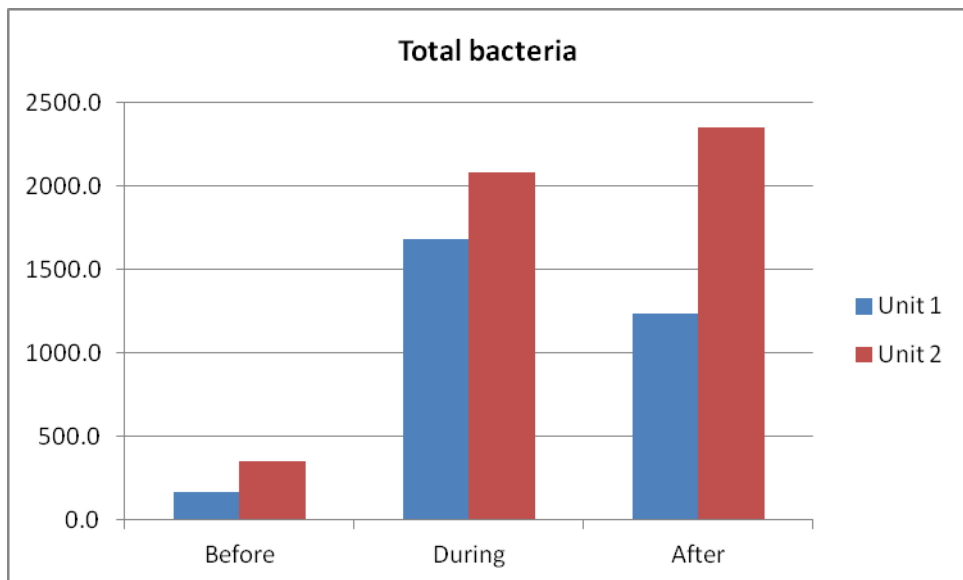
Figure 38: Indoor viable total bacteria count, before, during and after remediation
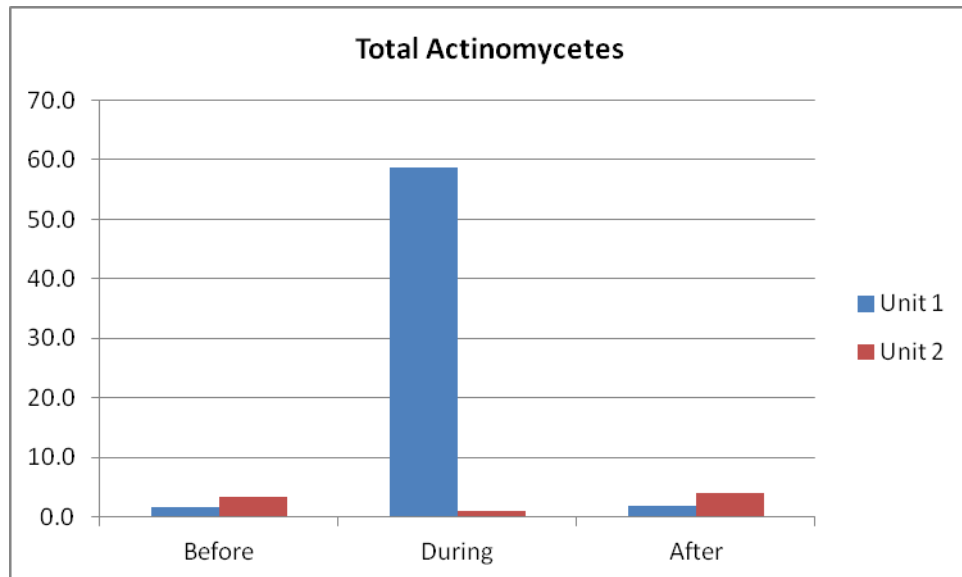
Figure 39: Indoor viable total *Actinomycetes* count, before, during and after remediation

Unit 2 biocontaminant levels were consistently higher than Unit 1 for all visits. This was particular so for the third visits where the results indicate that after remediation Unit 1 biocontaminant levels fell, while Unit 2 levels did not.

A major exception to this is the case of *Stachybotrys* which is higher for Unit 1.

During remediation - Particulate data

Background concentrations of PM 10 particulates were generally low prior to activities commencing on the building site on Unit 1, with a maximum reading of PM 0.21 mg/m³ in this time. When cladding removal commenced PM10 levels were observed to increase, see Figure 40; however the level to which they did so appeared to depend on where the sampler was located in relation to the building activity, see Table 3, with closer proximity being associated with higher levels. At the highest peak concentrations of particulates were 17 times the background levels (3.1 versus 0.21 mg/m³). Levels of PM10 returned to low levels during the lunch time break, suggesting that disturbed particles outdoors quickly disperse once remediation activity stops, see Figure 40 and Table 3.

	Background 7:15 am – 8:30 am	Cladding removal Detection at ground level 8:30 am – 9:30 am	Cladding removal Detection at upper scaffold level 9:30 am – 11:30 am	Lunch time (no working) 11:30 am – 1:30 pm	Cladding removal In the immediate vicinity of the workers 2:40 pm – 2:55 pm
Number of peaks greater than 0.1 mg/m^3	4	3	15	9	8
Average of the peaks greater than 0.1 mg/m^3	0.16	0.15	0.48	0.28	0.21
Maximum value (mg/m^3)	0.21	0.18	3.1	0.6	0.54

Table 3: Unit 1 PM 10 measurements over 0.1mg/m³ with various activities

Top chart labels:
- Background
- Cladding removal Detection at ground level
- Cladding removal Detection at upper scaffold level

Middle chart labels:
- **Cladding removal** Detection at upper scaffold level
- Lunch time (no working)

Bottom chart label:
- Cladding & building paper removal Detection in the immediate vicinity of the workers
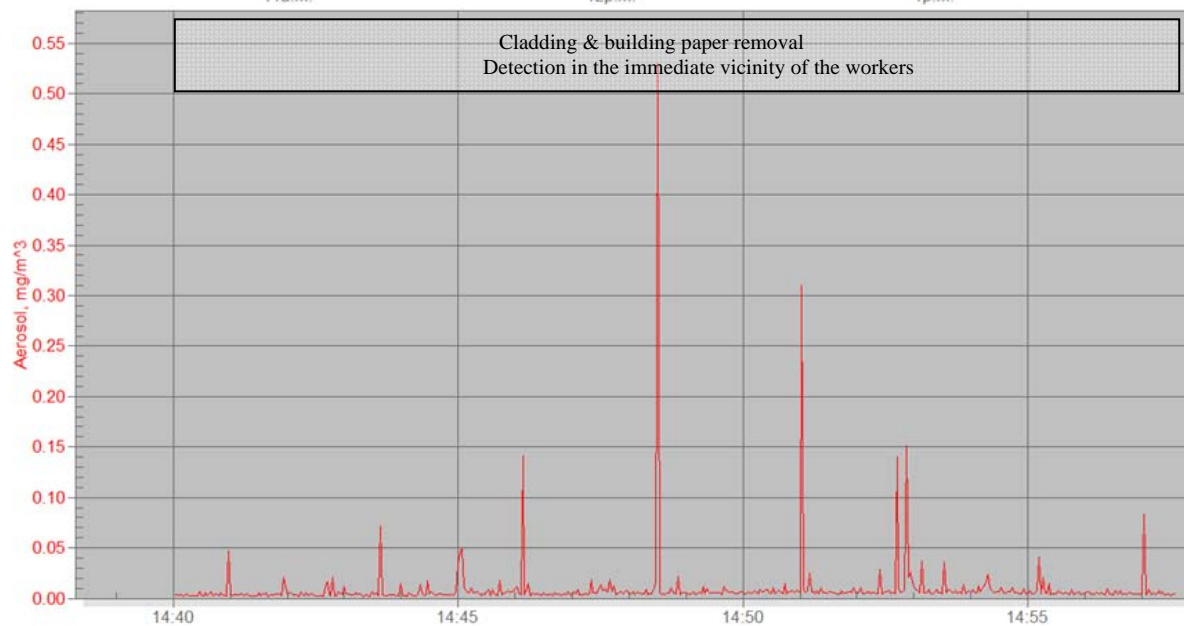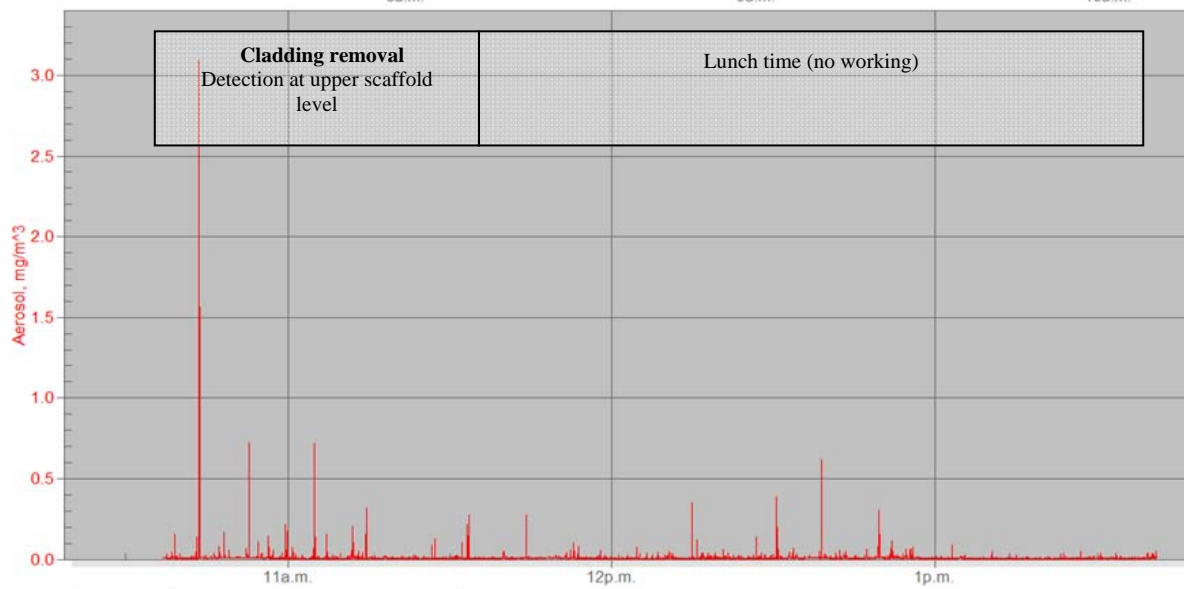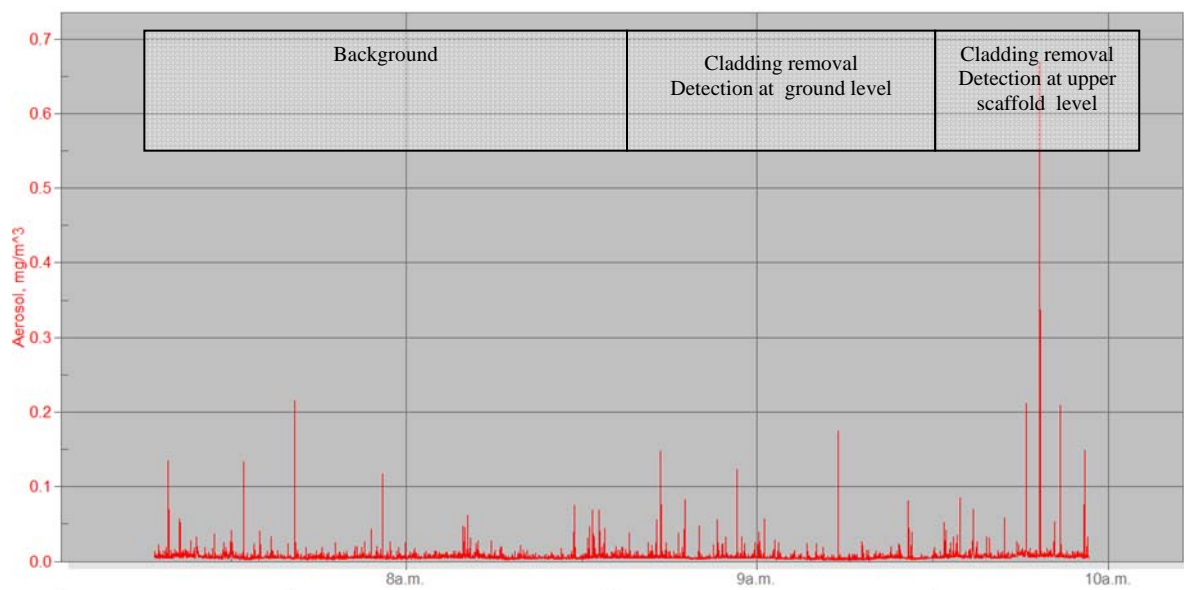
Figure 40: Unit 1 PM10 measured with various activities (NB: scale differs on each graph)

A decision was made to look at smaller particle levels in Unit 2 and Unit 3 by measuring $PM_{2.5}$ concentrations. $PM_{2.5}$ particles are more readily inhaled, and therefore may be more damaging and remain airborne for longer periods, once disturbed.

	Background 7:20 am – 7:55 am	Cladding removal upper levels 7:55 am – 9:15 am	Cladding removal directly across 9:15 am – 9:26 am	No working 9:27 am – 10:01 am	Cladding removal In the immediate vicinity of the workers 11:26 pm – 12:03 pm
Number of peaks greater than 0.1 mg/m^3	1	33	98	0	1
Average of the peaks greater than 0.1 mg/m^3	0.10	0.16	0.16	0	0.24
Maximum value (mg/m^3)	0.10	0.18	0.28	0	0.24

Table 4: Unit 2 PM 2.5 measurements over 0.1mg/m³ with various activities

During remediation of Unit 2, the background levels prior to any activity commencing on the building site of $PM_{2.5}$ were found to generally be low, being below 0.05 mg/m³, with just one peak observed over 0.1 mg/m³ see Figure 41 and Table 4. When remediation activity started on the upper levels of the housing, the sampler on the ground did pick up increased $PM_{2.5}$ concentration, with 33 peaks above 0.1 mg/m³, and a maximum of 0.18 see Table 4. When the sampler was moved to be in the immediate vicinity of the remediators on the upper scaffolding (9:15 – 9:26 am, Table), $PM_{2.5}$ concentrations rapidly increased and 98 peaks were observed over a level of 0.1 mg/m³ during this time. Again, as with Unit 1, when activity stopped on the site, the outdoor particulate matter concentration fell rapidly back to low levels. Interestingly when work restarted on the lower level of the Unit (11:26 – 12:03 pm, Table 4), and the sampler was moved to follow the remediators, only one peak over 0.1 mg/m³ was observed, indicating that the location of the workers on the building may be important for exposure, potentially in relation to the temporary roof on the remediation building skin.
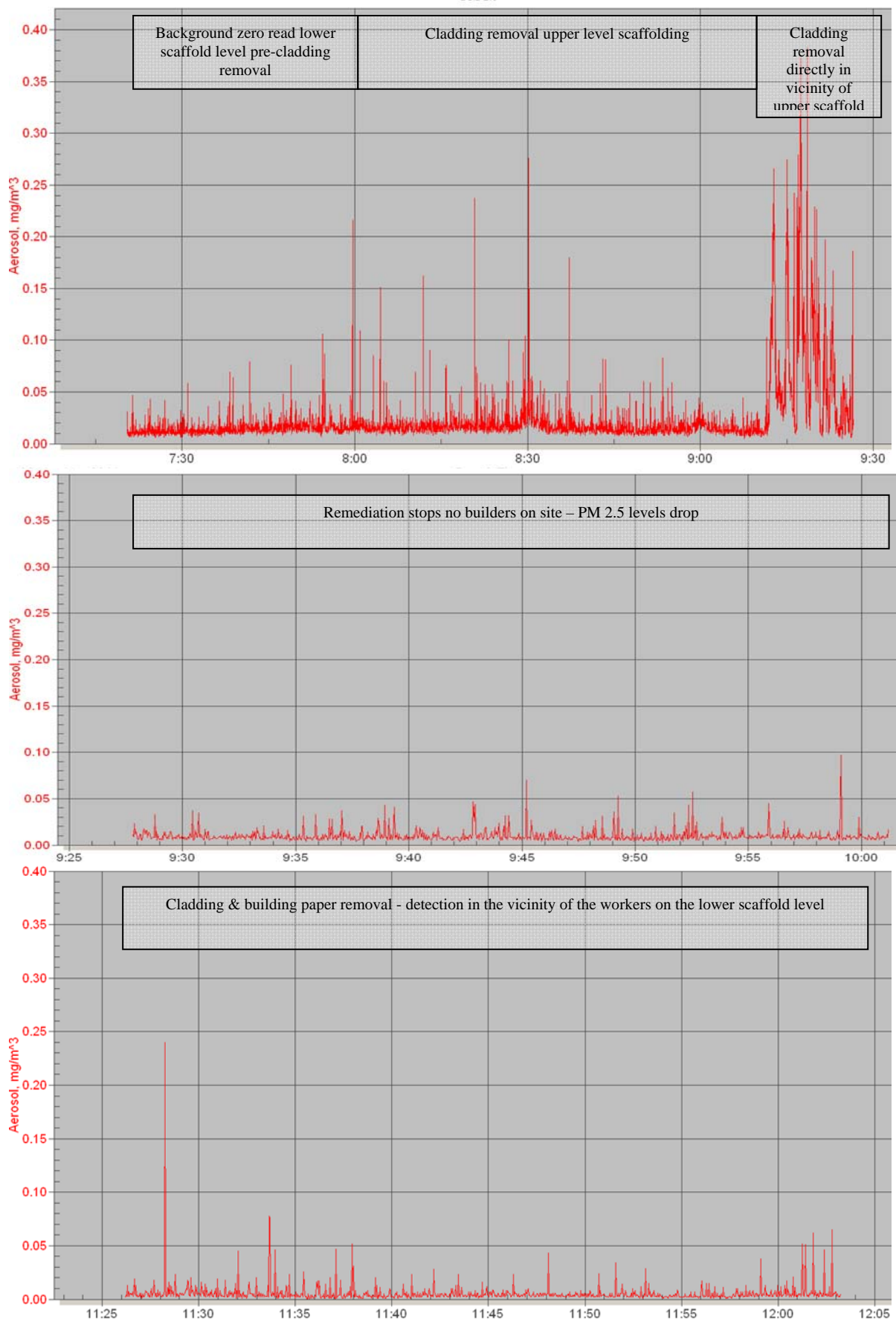
Figure 41: Unit 2 PM2.5 measured with various activities

	Background 7:47 am – 8:20 am	Builders around but no removal 8:33 am – 8:55 am	Cladding removal 10:31 am – 11:00 am	Cladding removal In the immediate vicinity of the workers 11:04 am – 11:16 am
Number of peaks greater than 0.1 mg/m^3	1	1	116	97
Average of the peaks greater than 0.1 mg/m^3	0.126	0.501	0.315	0.52
Maximum value (mg/m^3)	0.126	0.501	6.36	4.43

Table 5: Unit 3 PM 2.5 measurements over 0.1mg/m³ with various activities

Background concentrations of PM $_{2.5}$ prior to workers being on site on Unit 3 were again, as with Units 1 and 2, very low, with a peak of 0.126 mg/m³, the only peak above 0.1. When builders first arrived on site the levels continued to be low, and activities were mainly based inside of the building around the far side of the apartment block, and did not involve any decladding (temporary internal walls were being erected inside the buildings). When the decladding process started concentrations of $PM_{2.5}$ rapidly increased for the static sampler (10:31 – 11:00 am), with 116 peaks recorded over 0.1 mg/m³ and a maximum concentration of 6.36 recorded. Cladding removal in the immediate vicinity of the workers between 11:04 – 11:16 am also showed high levels of particulate matter over 0.1, with 97 peaks observed, and the maximum value of 4.43 mg/m³. It must be noted however, that some of these particulates may have also been generated by the use of a metal grinder around the other side of the building, with the potential for the wind to move it around the building and into the vicinity of our samplers, and so may not be solely contributable to the cladding removal process.

Background zero read lower scaffold level pre-cladding removal

Workers onsite (non-cladding removal)

Cladding removal by workers AM510 on tripod approx 2 m away

Further cladding removal by worker AM510 hand held approx 1 m away from worker
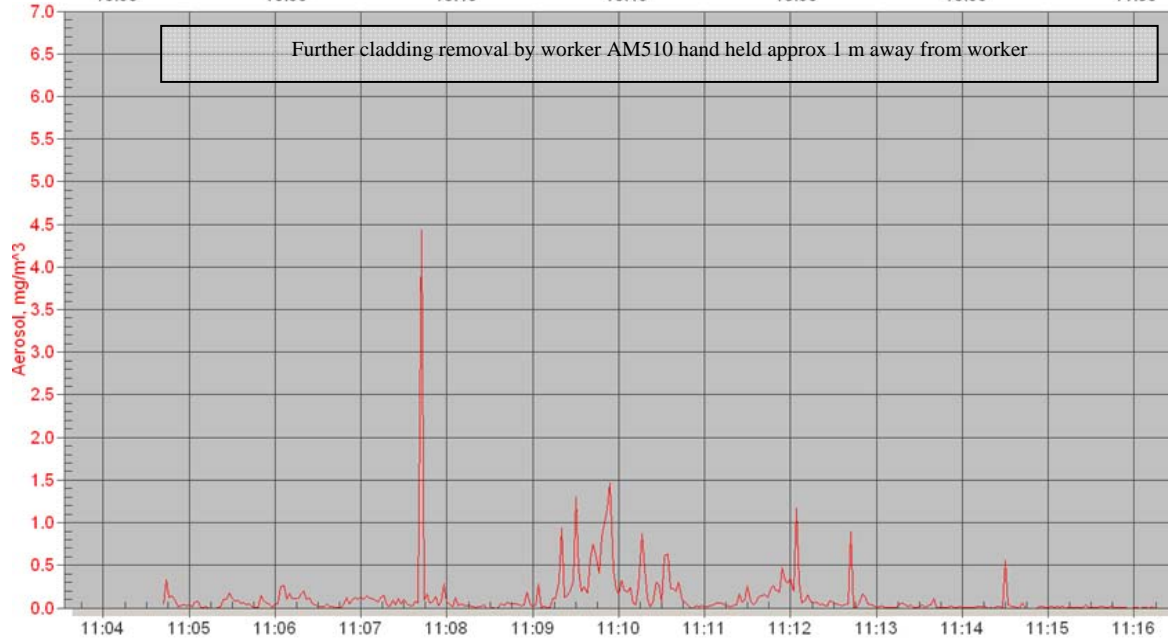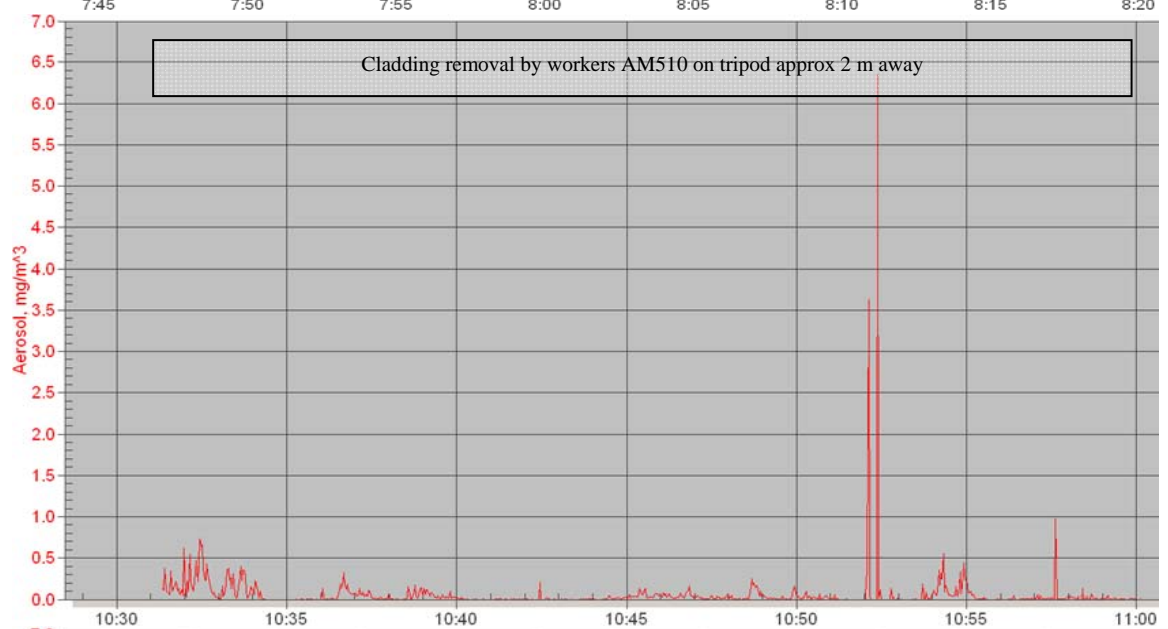
Figure 42: Unit 3 PM2.5 measured with various activities

Nasal sampling

Levels of spores were recorded from the Vaseline coated nasal samplers worn during the remediation of Unit 1 on a 0 – 4 scale, see Figure 43. No masks were worn during this sampling period, after observations of workers on site indicated very few worn respirators. Person A had moderate levels of spores in both nostrils, Person B just in nostril 1, and Person C had sporadic levels in nostril 1. No spores were observed in nostril 2 for Person B or Person C. A large amount of fibres and anomorphous material were also collected in the Vaseline filled nasal sample cups, making spore counting difficult. Additionally the nasal samplers were difficult to wear for Person C and Person D (results not able to be read due to sample preparation problems), as those with larger nostrils found it difficult to keep the samplers in place. Therefore for Unit 2 and Unit 3 samplers were worn by smaller nostrilled researchers, and were also attached to an elasticated band attached to the mask which went around the back of the head, to help keep the samplers in place, particularly as the addition of a respirator was observed to sometimes dislodge the nasal samplers. After advice from Biodet the coating of the samplers in Vaseline was abandoned for Unit 2 and Unit 3, and instead a double sided layer of adhesive tape was applied to the cups, for easier identification of spores under the microscope.
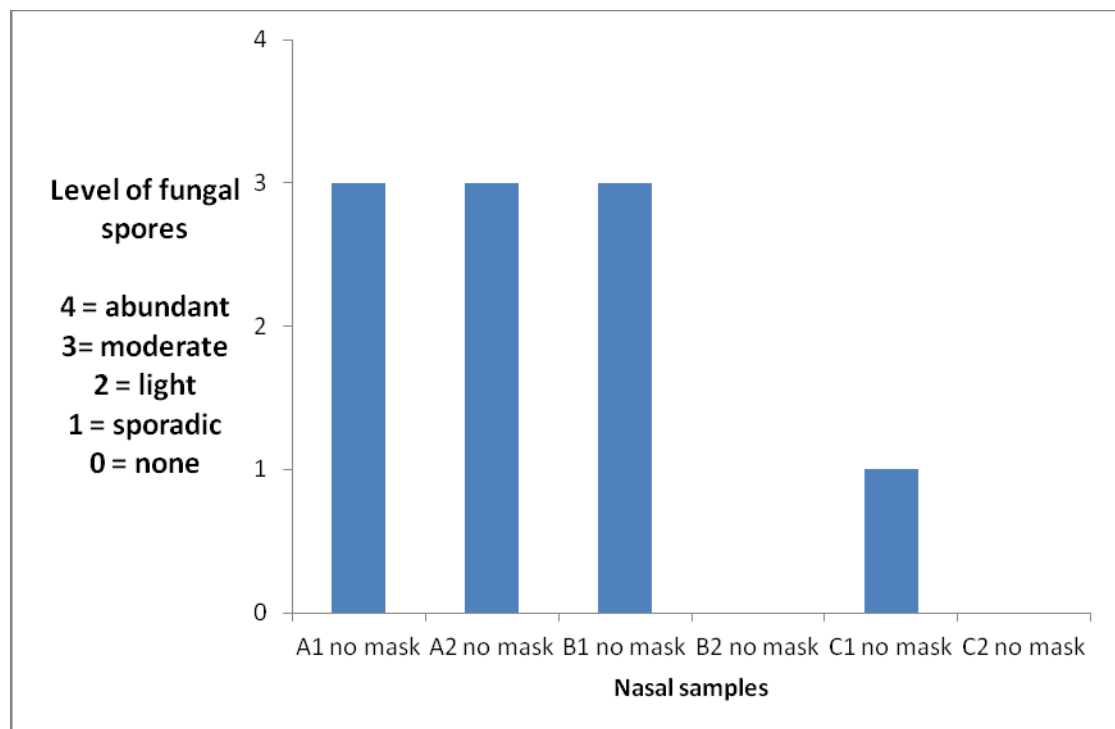
Figure 43: Levels of fungal spores observed in Vaseline coated nasal samplers during remediation of Unit 1, for person A, B and C, wearing no safety masks.
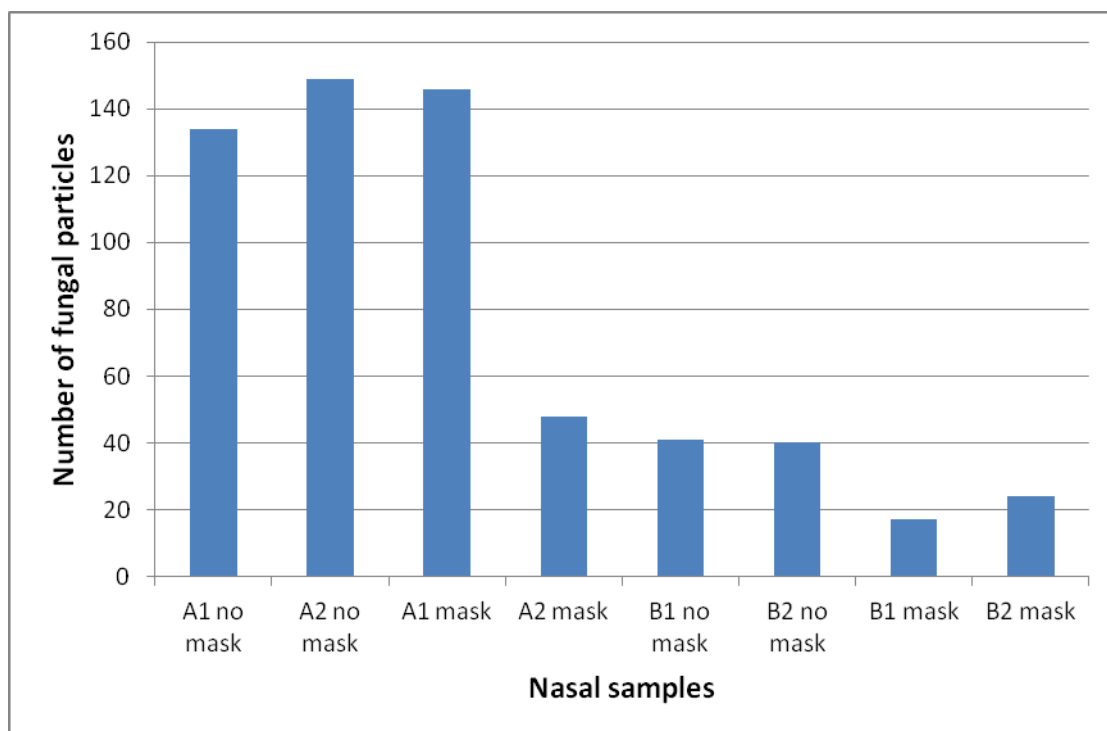
Figure 44: Number of fungal particles found in each sellotape nasal sample during remediation of Unit 2, with and without masks. Person A (respirator) and Person B (disposable mask).

The number of fungal particles that were observed on sellotape traps on the nasal samplers during remediation of Unit 2 indicate that high levels of spores can be breathed in if there is no mask protection, see Figure 44, particularly small spores such as *Nodulisporium* and *Penicillium/Aspergillus* types, see Figure 45. *Stachybotrys* spores were observed in the nasal samples of Person A with no mask present, with 5 spores in sample 1 and 3 spores in sample 2.

With the addition of a respirator there were no *Stachybotrys* spores observed in one nasal sample for Person A. However, one *Stachybotrys* spore was observed in the other nasal sample, suggesting that the respirator was not preventing all spores from entering the breathing cavities.
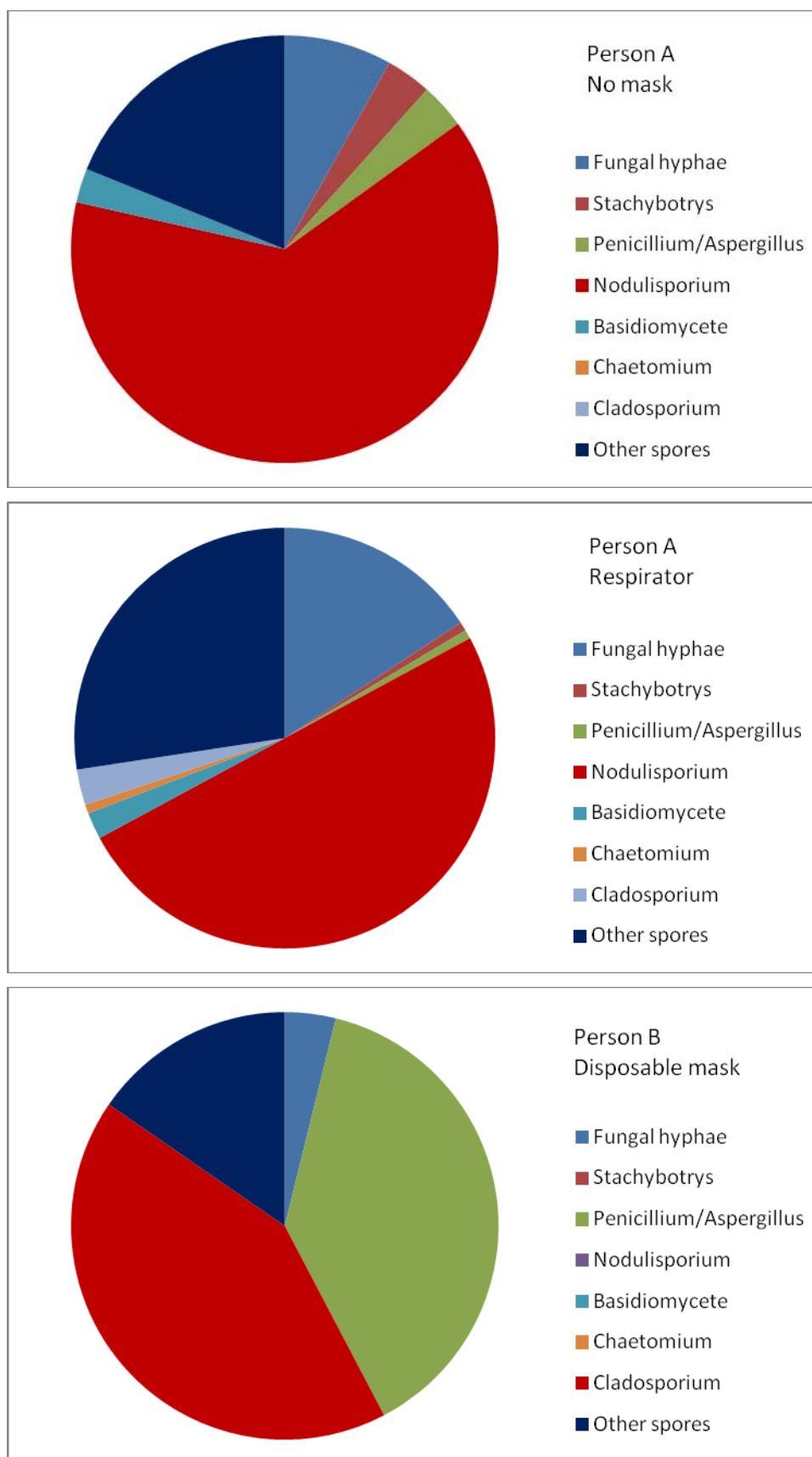
Figure 45: Types of fungal particles found in sellotape nasal samples during remediation of Unit 2, with and without a respirator for Person A and with a disposable mask for Person B

In Unit 3 only a short sampling time became available and only one period of nasal sampling could be conducted. The decision was made to carry out the sampling with respirators, and miss out the no mask samples. Fungal particles were present on the nasal samplers even while using respirators, for both Person A and Person B, see Figure 46. Levels were lower than those observed for Unit 2, and consisted entirely of fungal hyphae and a few *Penicillium phialides* spores. No *Stachybotrys* spores were picked up by any of the nasal samplers on Unit 3, even though they were observed to be present on site on the building materials.
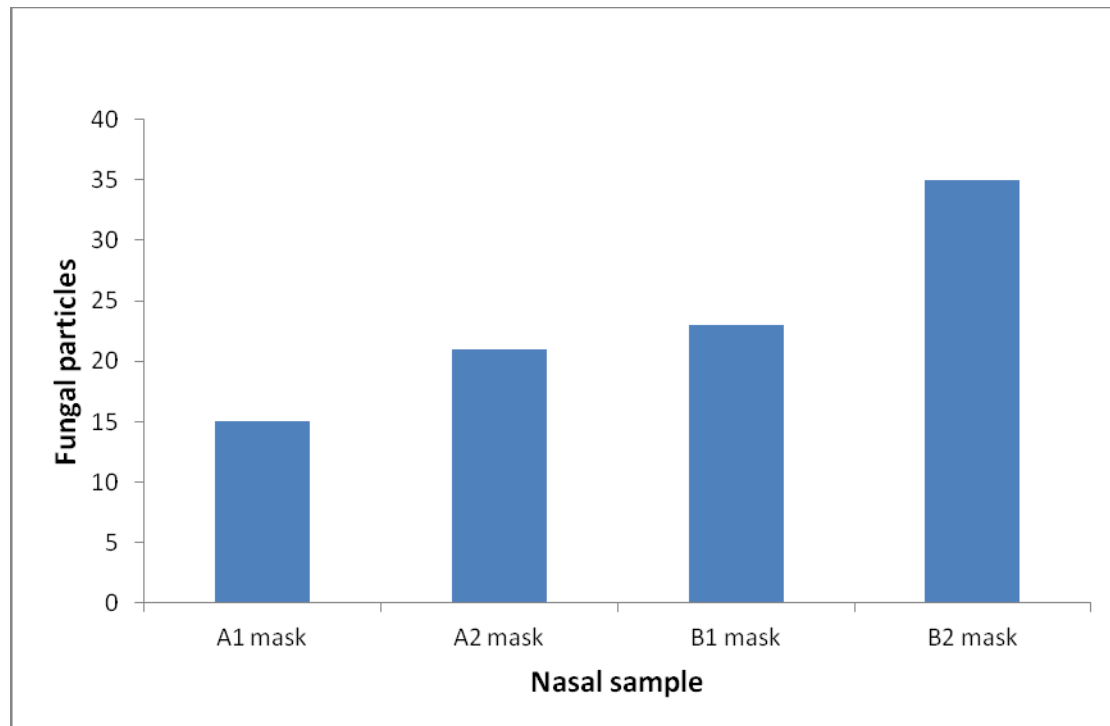
Figure 46: Number of fungal particles found in each sellotape nasal sample during remediation of Unit 3, for person A and person B, both with respirators

Mask swab samples

For Unit 2 a swab sample was taken from the inside of the respirator and the disposable mask to identify whether any contaminants were present on the inside of the mask.

Fungal spores and fragments were observed on the inside of both the respirator (Person A) and the disposable mask (Person B), as were yeasts, Actinomycetes and other bacteria, see Figure 47 and Figure 49. This would suggest that these contaminants were able to breach the both types of masks.
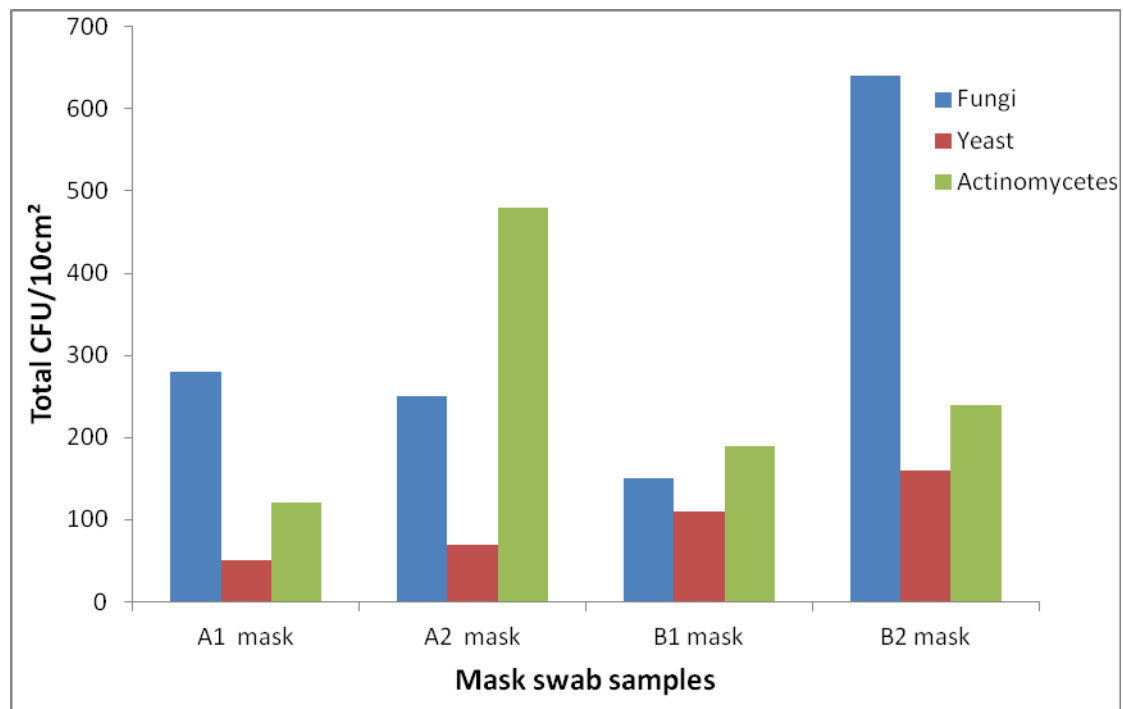
Figure 47: Levels of biocontaminants from swab samples taken inside the masks after being worn during remediation activities in Unit 2

Paecilomyces was the predominant fungi found on the inside of the masks, Figure 48, which is a common decaying fungi of soil and plant material. It has been found to be cause infections such as ocular infections, sinusitis and cellulitis in humans particularly for those who are immune compromised (Jade et al. 1986, Gucalp et al. 1996, Pastor et al. 2006). Non-sporulating fungi were also common on both the respirator and the disposable mask. *Stachybotrys* spores were not found on the inside of the respirator but were found on the disposable mask, which may suggest that the respirator had filtered out these spores. However *Trichoderma*, *Alternaria/Ulocladium* and *Phoma* were all found on the respirator and not the disposable mask, all of which have spores in a similar size range as *Stachybotrys*, suggesting that it was not a size related factor.
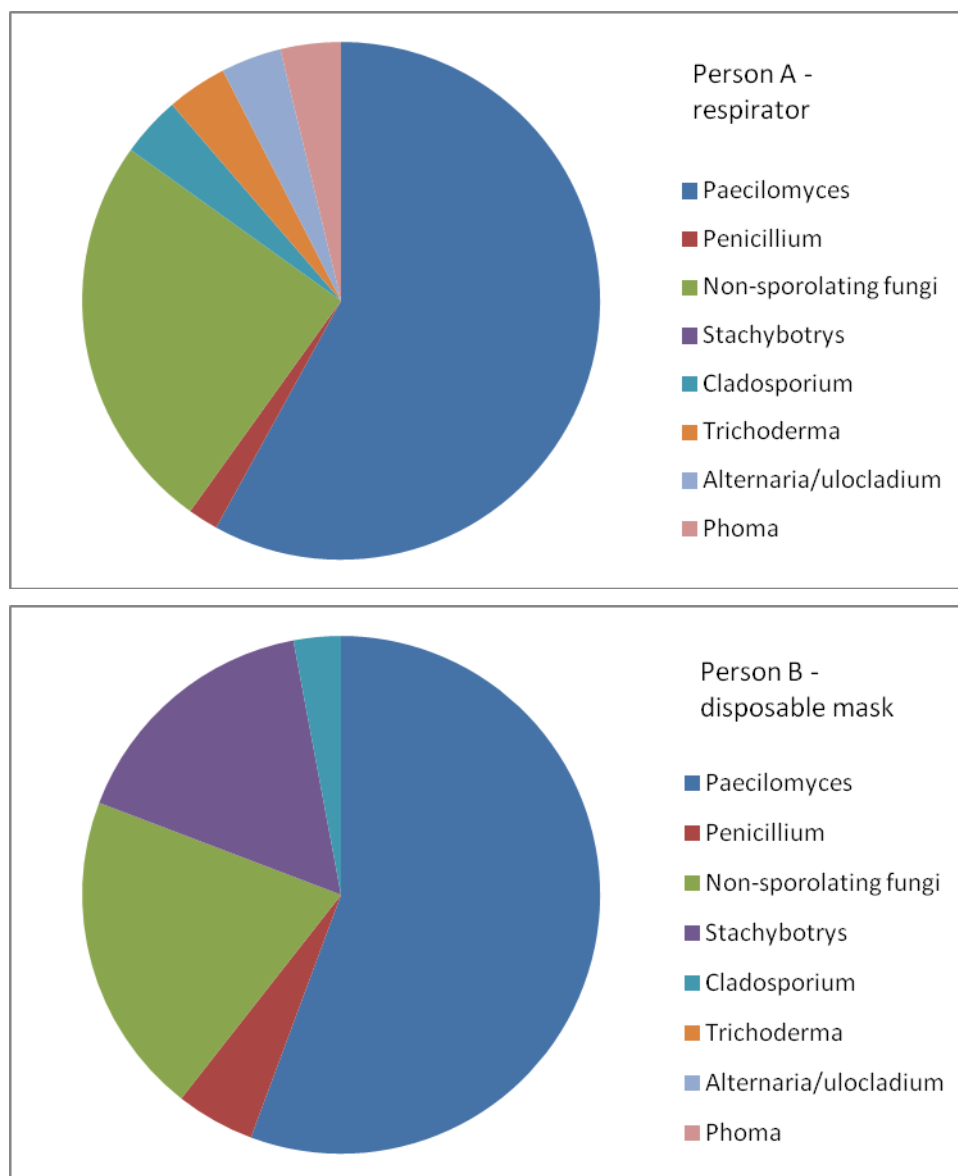
Figure 48: Types of fungi found on swabs of the interior of masks after remediation activities.

Bacterial levels inside the mask were particularly high on one side for Person A, who was wearing the respirator, with 5.2×10^4 bacteria present, which mainly comprised of *Staphylococcus/micrococcus* bacteria (50%) see Figure 50. The bacterial flora was different between person A and person B on the inside of the masks, with the predominant bacteria for Person B being *Bacillus*, see Figure 50. Again it is possible that some of these bacteria were naturally occurring, and not from the building site, particularly as they can be associated with skin and saliva.
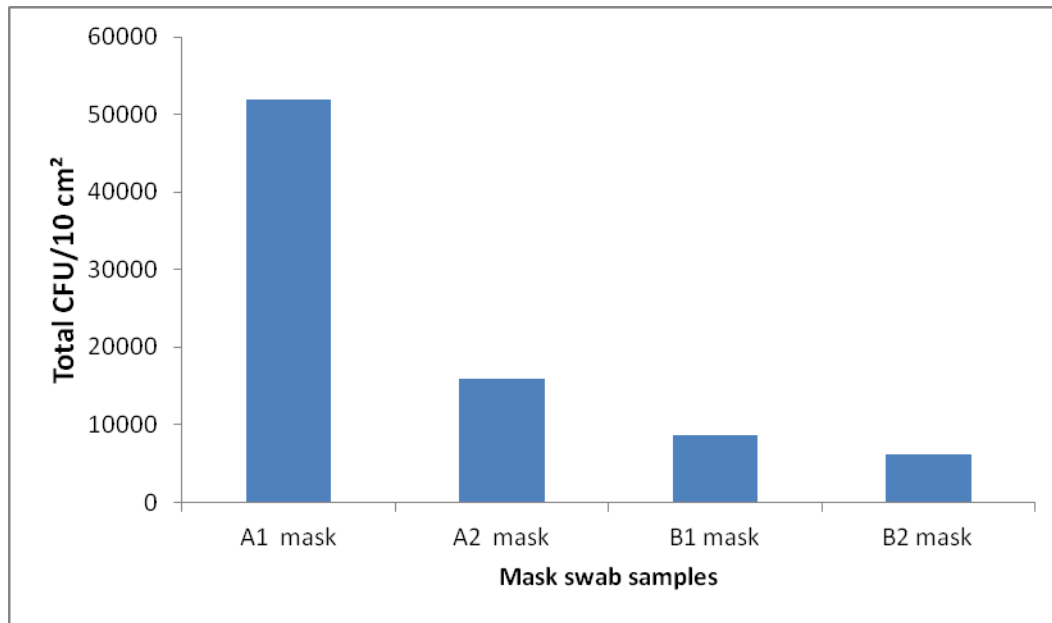
Figure 49: Levels of bacteria from swab samples taken inside face masks after being worn during remediation activities in Unit 2
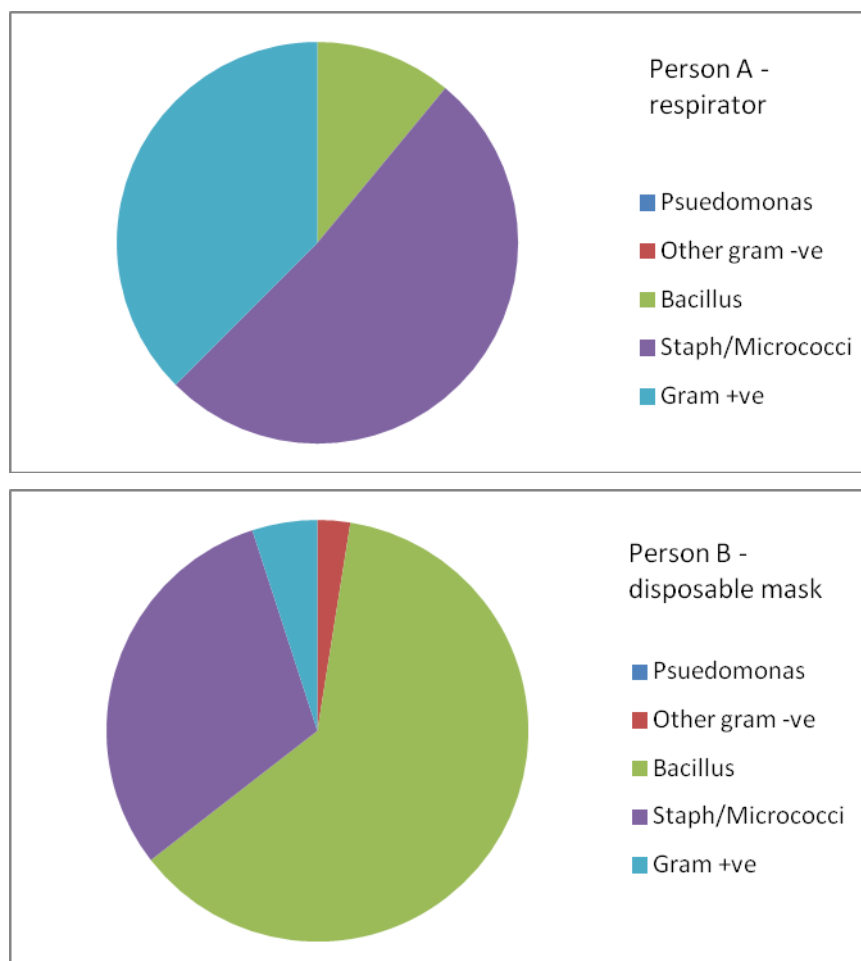
Figure 50: Types of bacteria found on swabs of the interior of masks after remediation activities

While it is not a surprise that biocontamination of fungal spores occurred in the case of disposable masks, which do not provide a good airtight seal around the mouth and nose area, and therefore allowed contaminants through, it is somewhat surprising that the respirator was found to have high levels of contaminants, since it should have a good airtight seal. As respirators are the recommended safety equipment for working in a dust contaminated environment this requires further investigation. It is possible that the respiratory was not fitted correctly which resulted in an insufficient seal around the respirator mask, or that some of the biocontaminants were already present on Person A's skin prior to the mask been fitted. Certainly humans can be a source for yeasts and bacteria, and it is also possible that some contamination from the building site may have adhered to Person A's skin prior to donning the mask.

Mycotoxin samples

Air samples were taken to examine the presence of trichothecene mycotoxins from Units 1 – 3 before remediation had occurred. The samples were taken in the lounge and bedroom of each Unit by collection of air onto a filter for a 24 hour period, using a 2 L sampling pump. Samples of mycotoxins were found to be below the limit of detection (0.14 parts per billion) in two of the Units prior to remediation, including one placed in a contaminated lounge wall cavity. Unit 3 had detectable amounts of mycotoxin present in the before remediation air samples both in the lounge and in the bedroom, which were found to be between 31.7 – 35. ppb/m³ of air. Trichothecene samples taken during remediation activity were measurable, despite the shorter sampling time (1.75 hours versus 24 hours), indicating higher levels of mycotoxin were present during the remedial activity than during no activity. Air samples were taken for Unit 1 and 2 only during remediation, due to the aforementioned short period of remediation activity available for Unit 3 (less than 30 minutes).

Sample Description	Air sample (litres)	Mycotoxin level (ppblitre)	Mycotoxin level (ppb/m³)
Lounge wall cavity Unit 1 before remediation	48	< LOD	< LOD
Lounge Unit 1 before remediation	48	< LOD	< LOD
Bedroom Unit 2 before remediation	48	<LOD	< LOD
Lounge Unit 2 before remediation	48	< LOD	< LOD
Bedroom Unit 3 before remediation	48	0.0317	31.7
Lounge Unit 3 before remediation	48	0.0358	35.8
Indoor during remediation Unit 1	3.5	0.1486	148.6
Outdoor during remediation Unit 1	3.5	0.6857	685.7
Indoor during remediation Unit 2	3.5	0.1600	160.0
Outdoor during remediation Unit 2	3.5	0.1714	171.4

Table 6: Trichothecene levels in air samples taken before and during remediation

For Unit 3 a number of other mycotoxin samples were taken of fungal contaminated building materials and of gloves that had handled the building materials, see Table 7. The top layer of the building material samples with the contaminated materials was removed and cut into 2 cm × 2 cm sub-samples before being analysed for trichothecene levels. The filters used in the air sampling were pressed against the glove finger tips and the transferred material analysed for trichothecene levels. The

results indicated that mycotoxin levels were in excessive of 180 parts per billion per ml of extract, the highest dilution level chosen. This suggests that the handling of the contaminated building materials themselves could be the highest potential risk of exposure, and that the mycotoxins are able to be transferred by direct contact with the materials. Further investigations should be conducted to examine how far this contamination may spread, i.e. is it also present on builders clothing, shoes and skin after remediating buildings, and if so, how it may best be removed from these areas so that contamination does not spread further. Further consideration should also go to how and where the fungal contaminated building materials are disposed of, as they appear to be a potent source of mycotoxins.

Sample Description	Mycotoxin level (ppb/ml)
Unit 3 building material gibboard sample A (2 x 2cm)	180+
Unit 3 building material gibboard sample B (2 x 2cm)	180+
Unit 3 building material building paper sample (2 x 2cm)	180+
Glove filter sample A	180+
Glove filter sample B	180+

Table 7: Trichothecene levels in material samples taken from Unit 3

CFD Modelling

Figure 51 shows a general view of the modelled airflow over the building with an external velocity of 2 m/s. Figure 52 and Figure 53 are close-ups of the buildings and its surroundings showing the consequent airflows in the various spaces.

Figure 54 and Figure 55 and show the resulting trajectory of the biocontaminant cloud for the external velocities of 2 m/s and -1 m/s respectively.
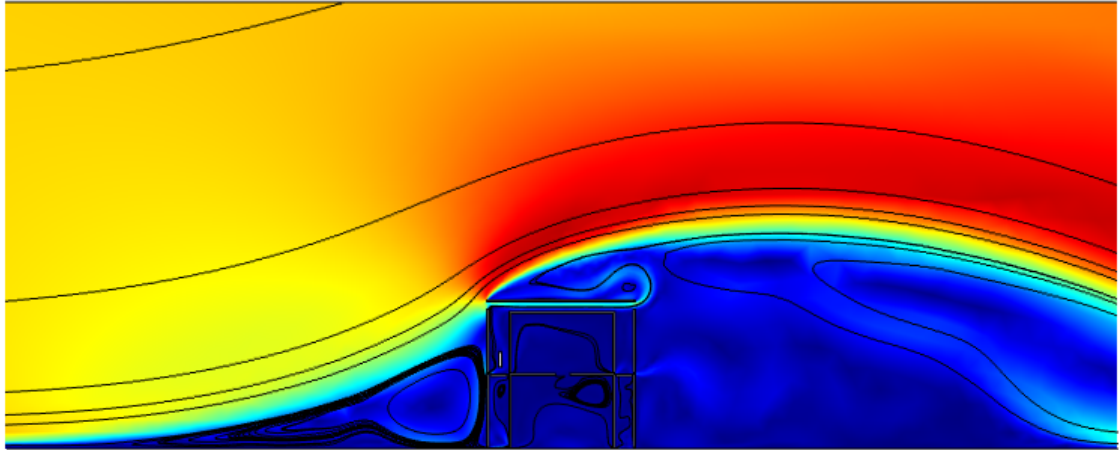
Figure 51: Airflow over the building with a wind of 2 m/s
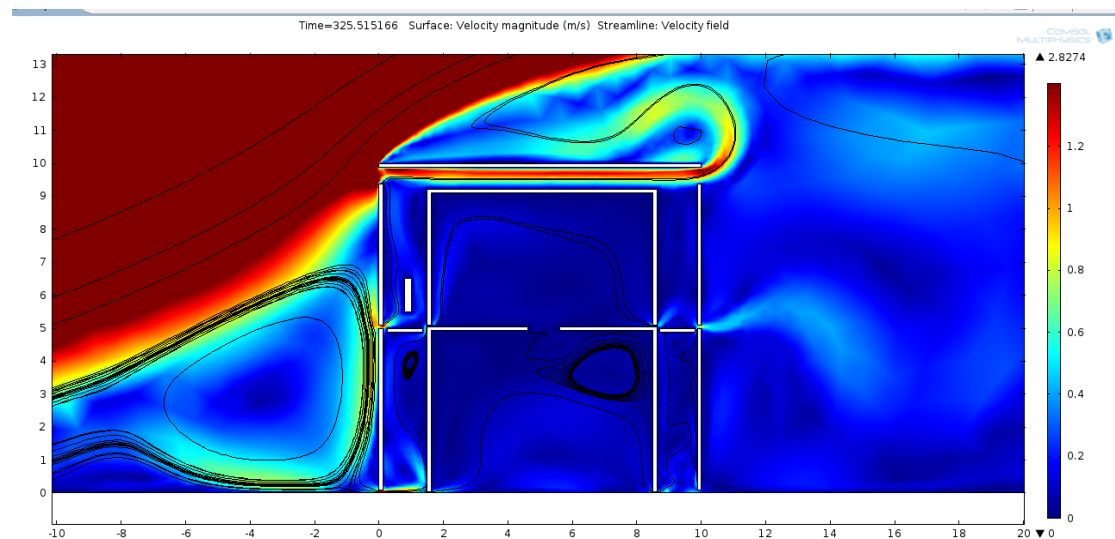
Figure 52: Airflow within building and protective surrounds with a wind velocity of 2 m/s
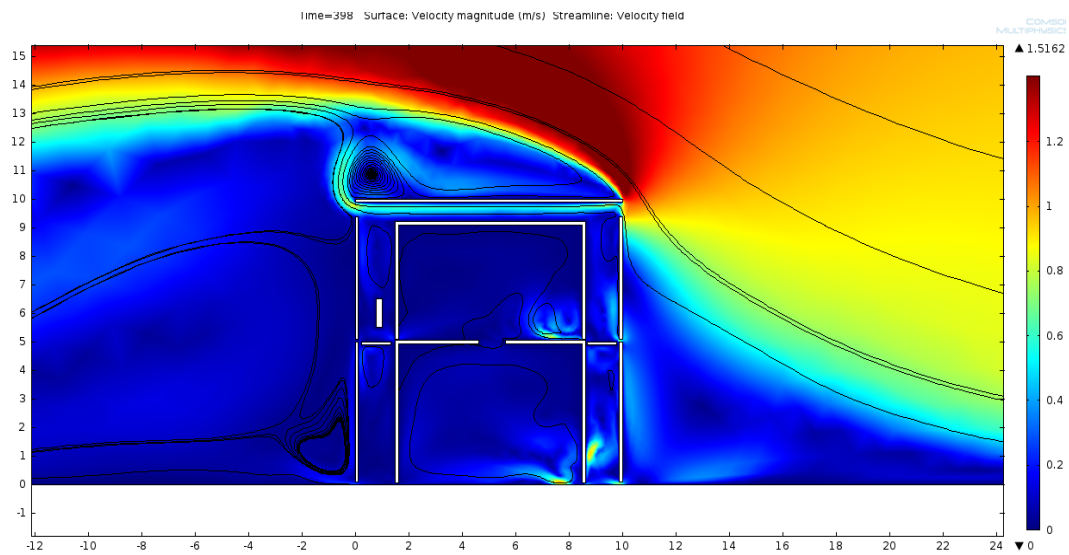
Figure 53: Airflow within building and protective surrounds with a wind velocity of – 1 m/s

5 seconds

10 seconds

15 seconds
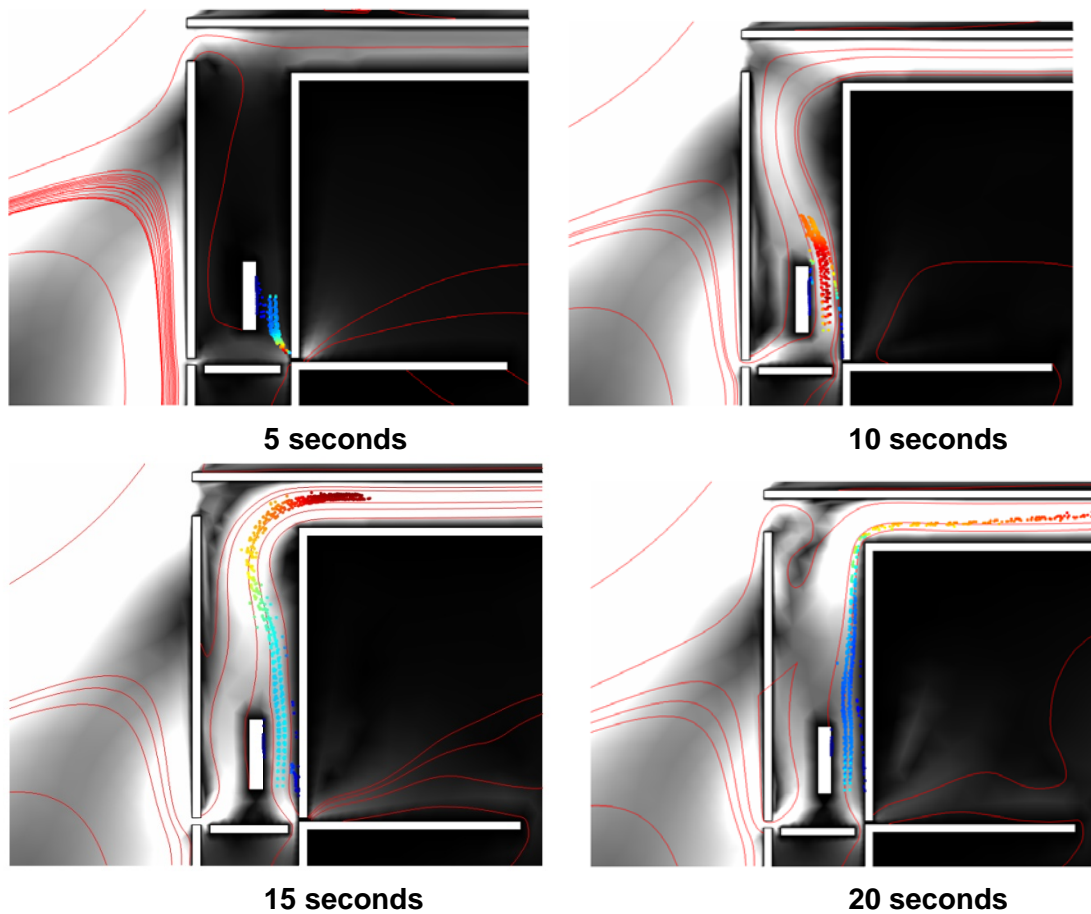
20 seconds

Figure 54: Trajectory of biocontaminant cloud with wind velocity of 2 m/s

5 seconds   **10 seconds**
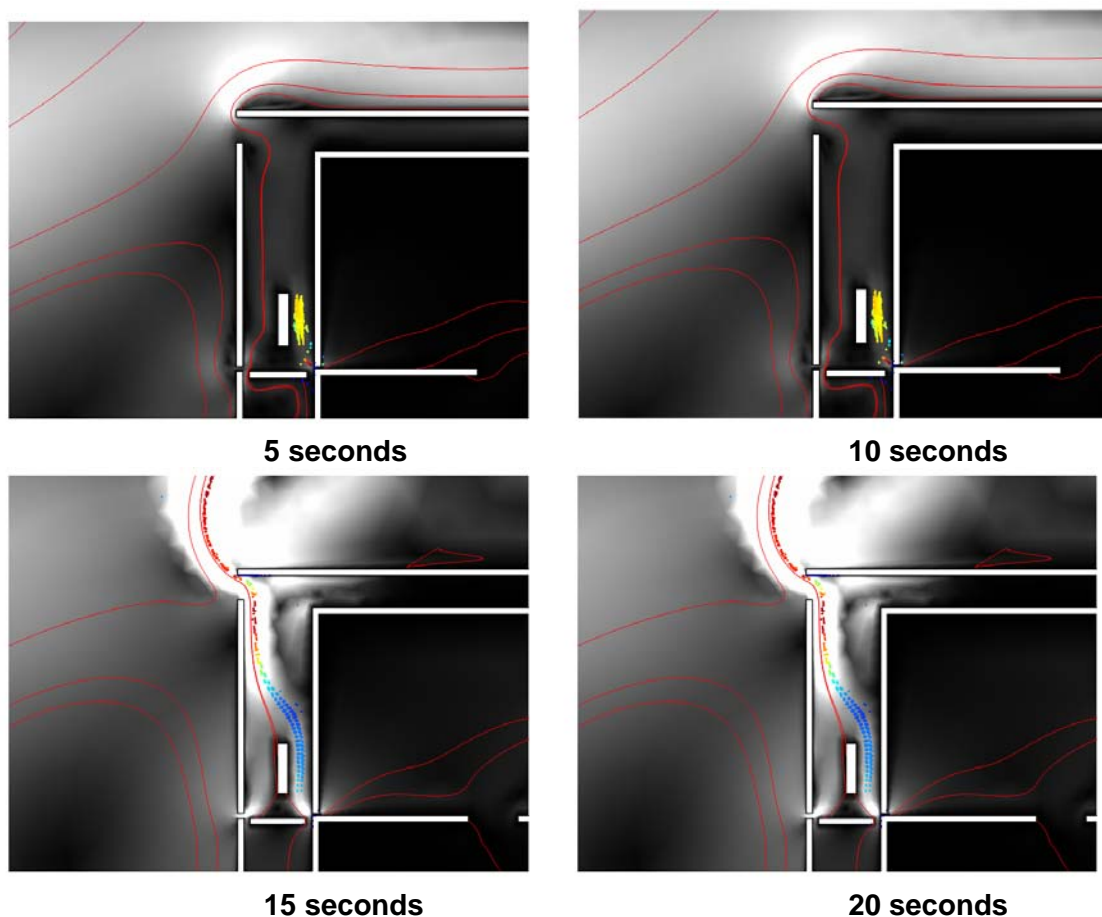
15 seconds   **20 seconds**
Figure 55: Trajectory of biocontaminant cloud with wind velocity of -1 m/s

It can be seen that the biocontaminant cloud is dispersing after 10 seconds when the external velocity is 2 m/s but for an external velocity of -1 m/s dispersion begins 5 seconds later at 15 seconds. At 2 m/s the cloud exits at the right end of the canopy, but at -1 m/s it exits to the left. This is consistent with data shown in Figure 41 which shows that frequently particulate count consist of several consecutive peaks, each sampled over two seconds, implying exposure to biocontaminant clouds between 2 to perhaps 6 or 8 seconds.

Results for an external velocity of 0.1 m/s, not reported here, are very similar to the results for -1 m/s but for different reasons. The cloud takes to about 15 seconds to begin to disperse in both cases due to low air velocities around the remediator, but exits to the left in the -1 m/s cased because the wind blows from right to left, but in the 0.1 m/s case because of convective air currents driven by temperature differences and the thermal plume from the remediator. The modelling suggests that from the instance of release the remediator is exposed to a cloud of biocontaminants for around 10 seconds for relatively high air velocities on the same face of the building as the remediator, to 15 seconds if the air velocities are very low or coming from the opposite face of the building.

Separate runs were done for particles that made there way indoors. These had a residence time of several minutes before they exited the room.

Discussion

This project examined the potential for aerosolising a biocontaminant cloud from heavily contaminated building materials in the walls of "leaky buildings", examining the nature of that cloud, especially assess the resulting fungal exposure to those who are remediating the buildings, and determining how the cloud disperses.

Stachybotryotoxicosis

Of note was the well established abundance of *Stachybotrys chartarum* contaminating a wide range of building materials in this study. This fungus has raised concerns in New Zealand since its widespread prevalence has been reported from a large number of leaky buildings (Waipara 2010) and its deleterious link to occupant health reported internationally (Miller et al. 2003). Despite its ubiquitous presence in water damaged buildings in New Zealand little research has been conducted to elucidate its local ecology and impact.

Most interest of *S. chartarum* has been due to its capacity to form wet rot, deteriorating organic fabric fibres including cellulose, and its ability to produce the highly cytotoxic macrocyclic trichothecenes which have given significant problems in mouldy straw in Eastern and Northern Europe. Stachybotryotoxicosis was first described in the 1930s by Russian researchers, and later by Forgacs in the English written literature (Neilsen 2002). He assisted Russian veterinarians during World War II, where stachybotryotoxicosis in horses was a severe problem for the Russian army. Russian researchers showed that horses were very susceptible to Stachybotrys infested straws (Neilsen 2002). Forgacs describes two types of toxicosis: i) A typical form with inflammation of the stomach, and the mouth as well as necrosis of some areas, followed by a decreasing number of thrombocytes and a decreases in the blood clotting, followed by a decreasing number of leukocytes and an almost total absence of blood clotting; ii) An atypical form, characterised by nervous disorders, as loss of reflexes, elevated body temperatures, and altered cardiac action.

The link to indoor air appeared in 1986, when it was described a household in Chicago, where the occupants suffered from symptoms similar to stachybotryotoxicosis. Filters from air sampling in this home were black after sampling due to the many *Stachybotrys* spores (Jarvis et al. 1999). Since these reports international research into causation linkages between human exposure to this fungus, its mycotoxins and illness is ongoing (Miller et al. 2003). However, it is universally accepted that a precautionary approach to avoid both chronic and acute exposure to this fungus should be undertaken at all times. Workers were clearly observed in this study to be potentially susceptible to a range of exposure types (including potential for ingestion) and levels to this and other toxigenic species. We recommend that urgent and immediate research and monitoring into worker exposures to *Stachybotrys* and other associated toxigenic building rot assemblages is undertaken to further inform safe work practices and procedures, particularly as increased remediation may be stimulated in response to central and local governments funding partnership and resolution service announced in 2011 to help building owners remediate a large and outstanding leaky building stock.

Nature of the biocontaminant cloud

The experimental data and CFD modelling enable us to describe the nature of the biocontaminant cloud that showers the remediator as the cladding is removed.

The cloud was found to be a complex mix of biocontaminants, including very high levels of *Stachybotrys*, *Cladosporium*, *Pencilium* sp., *Aspergillus* sp., *Alternaria/Ulocladium*, *Epicoccum*, gram positive and gram-negative bacteria including *Bacillus*, *Pseudomonas* and others; spore clusters, hyphal fragments, *Basidiomycete*; and particulates consisting of bacterial clusters, siliceous particles and amorphous particles. Especially concerning was the dangerously high concentrations of *Stachybotrys* in the cloud.

The particulate data shows many short duration very sharp peaks consistent with episodes of biocontaminant showering as the cladding is torn off. It seems clear that there is not one cloud but many episodic clouds suggests that biocontaminants are released frequently but episodically as sections of the cladding are torn off and wall biocontaminants are entrained into the external air. The episodic nature of the contaminant levels is evidenced by elevated biocontaminant levels varying widely from minute to minute. Consistently levels of particulates, both PM_{10} and $PM_{2.5}$, were also found to peak in a sporadic way during sampling. Consistent also are the CFD results which show that the remediator is exposed to a cloud episode for between 2 and 10 seconds, after which the cloud quickly disperses.

Because of the episodic nature of the biocontaminant levels which vary widely from minute to minute, even the smallest sampling timeframes of 30 seconds could lead to an overloaded agar plate containing 400 spores. Therefore this suggests that the upper levels observed in this study would be a minimum of exposure, and the maximum could be thousands of spores per litre higher. The types of culturable fungi obtained during RTU counts on the collected agar plates are evidence that large spore clouds are being generated during the removal of building material.

The levels of biocontaminant indoors do not fall quickly as indicated by the elevated levels indoors even 30 minutes after work had stopped on Unit 2, which the CFD results slow clearly is because strong air currents do not enter or leave the indoors, as required for rapid dispersal.

Exposure to and protection from the biocontaminant cloud

Results from the nasal samplers suggest that the spores which are generated are capable of being inhaled into the nasal cavity, and concerningly, that these may still be inhaled while wearing protective masks. A larger study is needed to examine whether this was a problem with the fitting of the respirator masks, or whether the filters were indeed letting fungal materials through into the mask.

The dispersion of the cloud

The dispersion of the cloud is determined by the nature of the temporary canopy and curtain erected around the building, and by the wind patterns and temperature distribution around the buildings. The CFD modelling suggests that the remediator will be exposed to these high levels for a few seconds, then to much lower levels as the cloud disperses upwards under the canopy and then exits the building, the details of which are determined by wind velocity and direction. The particulate data is consistent with these CFD results.

Indoor levels

Indoor concentrations of biocontaminants were already elevated before remediation, particular with *Stachybotrys* levels being unacceptably high. Unsurprisingly these indoor levels became very high during remediation. Of more concern for Unit 2 these levels did not fall after remediation. One would hope these high levels are due to dust and detritus entering the Unit during cladding removal which had not been cleaned from the building, or that the repaired buildings were closed up and left quite airtight before handing back to the owner.

Mycotoxins

Results from the mycotoxin sampling indicate that mycotoxins are released into the air during remediation and that they are also present in high levels on the building materials, and importantly, that touching the material is enough to transport the mycotoxins onto safety gloves. This raises the question as to whether mycotoxins are also present on the builder's overalls, on their skin and on their footwear, and also what happens to this equipment at the end of the builders day - are mycotoxins further transported into cars and homes, or left onsite.

The current study only allowed for mycological measurement of three Units, since the study is essentially a pilot study to document potential exposures, examine current PPE practice and affirm techniques in the assessment of fungal spore exposure of remediators in New Zealand. Therefore the results from the study are indicative and descriptive rather than quantitative. To unequivocally establish the industry wide mean exposure levels would require a much larger number of sampling days during remediation. A larger study is essential to confirm some of the findings from this pilot study, and to capture some potentially new exposure routes, that the findings of this study have suggested may be present, such as the direct transfer of mycotoxins to clothing. If the co-operation of builders could be obtained then detailed health markers could also be taken, such as urine samples to test for presence of further mycotoxin presence in the body.

Personal Protective Equipment

What was particularly concerning to us over the course of this project, and certainly not expected from the overseas literature, was that there was no standard practice for the use of Personal Protective Equipment (PPE) amongst the workers remediating leaky buildings.

Overseas literature (e.g. Kemp & Neumeister-Kemp 2010) would suggest that PPE would include respirators, gloves, eye protection, ear protection and hooded disposable suites – rather than standard overalls and gloves which appeared to be worn in the New Zealand setting. When placing our observations in context with what we know from the overseas literature, we would therefore appear to have a far less stringent approach to health and safety in this instance, when compared to our overseas counterparts.

Contractors that we are working with in this study appear unaware that some of the practices they are undertaking could be a potential hazard, and are unsure just what part of the process would result in the spore disturbance (many believed that the removal of building paper was the critical spore release part – which we clearly demonstrated is untrue).

Also, while overseas standards require the use of respirators for remediation of mouldy materials, interestingly we have found that these may not provide full protection. Our results suggest that respirators will not sufficiently protect workers from all spore types, particularly the smaller fungal spore exposures e.g. *Penicillium* and *Aspergillus* spores. These types of spores were found in large quantities in our nasal samplers through the respirator apparatus. This is a finding that requires further investigation to establish whether this is due to an error in the fitting of the respirator masks, or whether the spores are able to enter through the filters.

Effectiveness of piloted techniques

The techniques chosen in this study are a mix of internationally established techniques and new techniques. The pilot study has identified a number of methods which worked for exposure assessment, and a number that did not.

Firstly, air sampling using agar plates was able to pick up the changing levels of spores in the air, although frequent sampling was needed to minimise plate overloading (after starting with 500 and 100 L samples on Unit 1, this was modified to 50 L samples on Unit 2 and 3).

However, for identification of all of the species that were present in the air, culturable sampling was not ideal. Even though *Stachybotrys* was clearly present on building materials and found in Biodet's total spore assessments, no *Stachybotrys* spores were observed on the plates, as this species can be difficult detect by most routine air sampling methods (Tucker et al 2007). A combination of both air sampling, potentially total spore sampling, and direct sampling of building materials and surfaces is therefore recommended for future surveys to enable detection of all microbial contaminants. Additionally some agar plates were heavily contaminated by fast growing species, preventing further assessment of RTU levels. Rapidly growing saprophytes, such as *Chrysolinia* and Zygomycete species, not only bias against the growth of slower growing fungi, but also restrict development of diagnostic morphological characters of many other fungi that are present. We concur with previous surveys that the range of 25 - 75 colonies per plate is optimal for countable plates and to enable cultural identification. Future microbial surveys should ensure isolation methods to achieve these levels. Total spore samples may be an area to investigate in future, but any spores that adhered to the microscope slides could well be swamped by the high level of dust particles that would be expected to be generated during removal of building materials, in addition to the spores.

This is the first study to trial nasal sampling for fungal exposure in New Zealand, and the results suggest this could be a valuable tool to examine exactly what is breathed into the nose during building remediation. The techniques were refined through the pilot study, and it would now be recommended that elastic straps be added to each sampler to secure it in place, and for this type of spore identification work, that an adhesive tape film be used, rather than the Vaseline layer.

The trial of a new technique to examine whether fungal spores and other contaminants could be found on the interior of the respirator and disposable mask, using swabbing was also found to be successful. Further examination of this on a wider range of samples would be useful, in combination perhaps with identifying what is already present on faces and in saliva samples from the person wearing the mask.

Testing of building materials and air samples for trichothecene mycotoxins was also trialled in this pilot study. Air sampling prior to remediation for 24 hours did not result in enough measurable mycotoxin on filters in Units 1 and 2, suggesting that longer

time frames may be needed if this is to be used in future assessments, unless they are at high levels, as per Unit 3. However, short term air sampling during removal of materials did result in measureable levels of trichothecene in the two Units where this was measured, suggesting this would be a good measure to continue to collect for exposure. Additionally transference of mycotoxins onto clothing and safety equipment from building materials showed promise, and would be a very good method to start to quantify exposure risk in subsequent studies, with higher dilutions needed than used in the current pilot study to capture the exact quantities.

The study also demonstrated that the placement of the samplers, particularly outdoors, can result in vastly different readings, particularly noticed on Unit 3 when researchers A and B were following the builders round. This suggests that wind direction and other factors could quickly disperse the spore clouds outside, in line with the CFD results, and also that it is important to take measurements as close to the builders as possible, in order to establish exposure risk. The ideal study would have remediators wearing some of the samplers, particularly those that do not require changing of agar plates, such as those measuring PM levels, and mycotoxin air sampling.

Long term health

The fungal exposure observed during remediation in this study, both that from contaminated materials and that which is aerosolised during remediation, suggest that builders, particularly those without adequate safety equipment, could be exposed to large quantities of fungi and mycotoxins. There is every chance that these builders could be exposed to multiple remediation jobs over their careers (particularly as there are so few building companies involved in remediation, and so many houses to remediate). The potential for a cumulative life time exposure for the builders is therefore possible. Based on the results from our pilot study, we believe this is an important potential issue which requires further investigation and the further development of safety recommendations to avoid deleterious health effects.

Observations of potential biocontaminant exposures to workers

A number of qualitative observations were made of workers during the time the researchers were onsite during the collection of samples. These are areas that should be followed up with contractors, and with further research, could form the basis of some recommendations on best practice for dealing with building materials contaminated with fungi and mycotoxins.

- Overalls, safety boots, gloves, helmets and eye protection were sometimes worn during removal of building material, which was good to see. We would suggest re-emphasizing the importance of this to the workers as some workers were without eye or glove protection.
- Workers who remove cladding, paper, batts, contaminated building materials did not always wear respirators (asked one worker if his respirator was uncomfortable or difficult to wear, replied that this was the first time he had ever worn one – so it is possible that our presence on-site was also modifying regular work practices). One worker said that they usually only wore the respirators during building paper removal, believing this to be the time of highest spore exposure. Our observations would suggest that all stages of contaminated building material removal lead to an increased risk of

fungal exposure.

- One worker was observed wearing a handkerchief around his mouth/nose to avoid inhalation while disposing large accumulated piles of contaminated materials into skip rubbish bins
- Respiratory equipment was not worn by other workers in the vicinity of the cladding removal.
- Overalls continued to be worn over lunch breaks but it is likely that spores and mycotoxins are to be adhering to these.
- We were unsure of what practices of glove removal and subsequent hand washing are in place for the workers during breaks and after work.
- We were unsure of whether overalls and safety boots are taken home with the workers or stay onsite. We would suggest that the latter would be preferable, to minimize spore and mycotoxin transfer through to other areas (vehicles, workers' homes etc).
- Spores and mycotoxins also likely to be adhering to workers hair and the unprotected areas of their faces (cheeks, ears, neck etc). It would be desirable to provide some clean 'decontamination area' and methods for allowing builders to remove these – e.g. onsite showers, or basins to wash, prior to leaving the site.
- Worryingly some workers sat inside a remediated Unit to eat lunch maybe thinking that this was outside the contamination area. Our results show clearly that levels of spores were elevated inside the Unit, and is a site of potential additional exposure.
- Some workers were also observed to have their lunch and breaks in on-site locations that were in close vicinity to areas with piles of contaminated materials, or where contaminated materials were being transported past, and or disposed of into, refuse bins.
- Workers involved with disposal were also observed to be susceptible to exposure events; e.g. truck drivers tasked with removing skip bins full of contaminated materials were present during loading and unloading bins and were not wearing any protective clothing or respirators during this task. All workers involved with cleaning and disposal should also be assessed for potential exposure events, see Figure 56.

Figure 56: (Photo 23) Workers place building materials at top of rubbish shoot without respirators

- We would suggest that an information sheet and Health and Safety training/induction be given to all workers on site regarding possible health risk from spores, and how best to avoid exposure to them at all phases of their working day, not just during the removal of contaminated materials.

- Potential exposure risks observed while undertaking remediation were via inhalation, direct and indirect dermal contact, and ingestion. Exposure events could of both acute and chronic in nature.
- A future health survey of workers undertaking remediation could elucidate if there is a prevalence, or not, of reports of illnesses consistent with previous overseas records of sick building syndromes

Conclusions

The primary aim of this work was to characterise the biocontaminant cloud emitted as the cladding is removed, assess its content and toxicity, assess the exposure of remediators to this cloud, and understand its dispersion.

- The evidence suggests that there is not one cloud but many episodic clouds created as bits of the cladding are torn off by the remediators and the biocontaminants in the wall below are released and entrained into the outdoor air currents. We found, with CFD modelling and from the particulate data, that the presence of a temporary canopy and heavy plastic curtaining surrounding the building for containment purposes, has the effect of retaining each cloud episode in the vicinity of the remediator at high concentrations for a few seconds, and then dispersing it under the eaves of the canopy.
- The clouds were found to be a complex, allergenic, pathogenic and toxigenic, mix of biocontaminants, including very high levels of *Stachybotrys*, *Cladosporium*, *Pencilium* sp., *Aspergillus* sp., *Alternaria*/*Ulocladium*, *Epicoccum*, gram positive and gram-negative bacteria including *Bacillus*, *Pseudomonas* and others; spore clusters, hyphal fragments, *Basidiomycete*; and particulates consisting of bacterial clusters, siliceous particles and amorphous particles. Potentially pathogenic species such as *Aspergillus fumigatus* was frequently detected. Other toxigenic species present included*; Aspergillus flavus, Paecilomyces, Pencilium chrysogenum* and *Pithomyces chartarum*. Especially concerning was the dangerously high concentrations of *Stachybotrys* in the cloud.

Towards our secondary aims we found:

- Indoors we found high concentrations of biocontaminants before remediation and very high levels during remediation. Our results suggested that post-remediation, before handing the building back to its occupants the building should be well cleaned to eliminate dust and detritus containing biocontaminants that have entering the building during cladding removal.
- Using nasal samplers and measuring mycotoxins within the protective equipment showed that dust masks and respirators were only moderately effective in protecting the remediator from this toxic cloud, but our methods here are indicative only and more sophisticated techniques would be required to confirm or otherwise these indications.
- There were many opportunities for mycotoxins on equipment and clothing to be spread around the worksite, and even beyond, most of which could be

mitigated by appropriate work place protocols and hygiene procedures when remediators stop for breaks or finish for the day.

- New techniques trialled that worked well included nasal sampling, swabbing the interiors of respirators and disposable masks and testing for the presence of spores and bacterial level, and rapid serial air sampling using agar plates to pick up changing levels of spores in the air.

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