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## Bioaccumulation on HVAC Filters in University Buildings in Singapore

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#### **SUMMARY**

Particle filters are important components of HVAC systems. Biomass accumulation on HVAC filters can potentially have adverse effects on building occupants. The research reported in this paper aims to quantitatively describe the content of biomass on HVAC filters from various indoor office environments in Singapore in relation to key governing processes. In this initial phase, filter samples were collected and analysed using DNA-based methods. The collected DNA was directly extracted from the filters and the final concentration was measured with Qubit and qPCR methods. The DNA concentrations were then related to the cumulative indoor occupancy level. The results show good correlations between bacterial DNA and cumulative occupancy. The proportion of total DNA that belongs to fungal and bacterial species decreases as occupancy level increases, probably because of biomass from other sources such as human skin cells.

#### INTRODUCTION

More than 90% of office buildings in Singapore are equipped with a centralized ventilation and air conditioning system. In a tropical climate, such systems are commonly referred to as "air conditioning and mechanical ventilation (ACMV), because heating is not needed. However, to be consistent with common international usage, we will refer to these systems using the common HVAC acronym. Current building practices were developed without detailed knowledge about how accumulation of biological material can happen in various parts of the HVAC system. It is feasible that processes such as microbial colonization and metabolism in HVAC systems contribute to adverse consequences for occupants, such as malodors, allergic reactions, asthma or sick building syndrome symptoms (Crook and Burton, 2010; Xu et al., 2011).

Accumulation on the HVAC filter of particulate matter, which includes particles of biological origin, is inevitable as it is the first line of defence in the HVAC system. Studies have reported the existence on filters of opportunistic pathogens (Noris et al., 2011). Research also indicates the possibility of more resilient organisms growing without additional nutrients (Pigeot-Remy et al., 2014). However, to date only a few studies have reported biological characteristics accumulating on building ventilation system filters. To our knowledge, there has not yet been a study using DNA-based analysis methods that *quantitatively* assesses the biological content of used HVAC filters.

The present study utilizes culture-independent DNA-based analysis applied to extracted material from HVAC filters that were collected from office environments in Singapore. The

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goal is to provide quantitative information about the biological content of the accumulated particulate matter. Total, bacterial and fungal DNA concentration on material directly extracted from the filters was measured. The results are correlated to estimates of cumulative occupancy level in the area serviced by the HVAC system. The results may provide a partial basis to reassess current practices with a view towards improving future building systems.

#### **METHODS**

Ventilation system filters from nine different air-handling units (AHUs) at Nanyang Technological University, Singapore, were acquired for this study. The nine AHUs served rooms containing typical office and library facilities at a university. The occupancy levels of the selected indoor environments were determined by conducting hourly population counts by means of direct observation on five weekdays and four weekend days for each of two months. The rooms were also characterized based on activities levels and sizes.

The filter samples (polyester, MERV 8 rating) were obtained from the AHUs' indoor secondary filter bank, which is positioned after the primary outdoor-air filter in the centralized fan-coil HVAC system. This secondary filter processes a mixture of recirculated indoor and make-up outdoor air, as designed to keep the indoor CO<sub>2</sub> level within allowable limits. In accordance with Singapore regulations, these filters are replaced every three months. In this study, all filters were collected after full three-month periods of service.

Pieces were cut from each filter for DNA extraction and analysis. Specifically, a 2×5 cm size was used for filters that were 2.5-5 cm thick and a 1×5 cm size was used for filters with pleats deeper than 5 cm. The filter pieces were directly inserted into the 5-ml bead beating tubes of the MOBIO Power Water (PW) DNA extraction kit. The process of DNA extraction followed the manufacturer's protocol with some modifications to improve DNA yield. Extraction replicates were taken the same filter panel.

The final DNA solutions were analyzed with a Qubit (Invitrogen) for total DNA and with qPCR (LC480, Roche) using universal probes and primers for specific quantification of bacterial and fungal DNA. The DNA concentrations measured by Qubit and qPCR were scaled up using the ratio of the actual area of the AHU's filter panels to the extracted filter area. Weekly average DNA accumulation rates (ng DNA/week) were then determined, by dividing the total accumulation by the weeks of service for the given filter.

#### RESULTS AND DISCUSSION

Figure 1 displays the weekly fungal and bacterial DNA accumulation rates and also the cumulative occupancy level (person-h/week) of each room studied.

The results show that the bacterial DNA accumulation rate has a stronger relation to occupancy level than its fungal counterpart. It can be observed that filters collected from locations with lower occupancy levels tend to be dominated by fungal DNA (Office 1-6 and Classroom). Conversely, bacterial DNA dominates at locations with higher occupancy, such as the library office with occupancy of 6575 person-h/week and the library reading hall with an occupancy of 15,400 person-h/week. A linear regression of bacterial DNA and cumulative occupancy also displayed a high correlation (R<sup>2</sup>=0.69), whereas the correlation between fungal DNA and cumulative occupancy was weak (R<sup>2</sup>=0.05). This finding is broadly consistent with previous studies showing that humans are an important source of indoor

airborne bacteria and that ventilation-system filter dust is influenced by indoor emissions (Hospodsky et al., 2012; Noris et al., 2011).

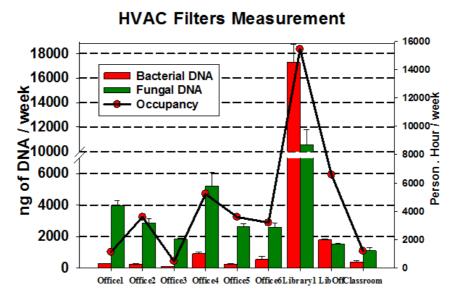


Figure 1. Weekly bacterial and fungal DNA accumulation rates on HVAC filters coupled with occupancy levels (in person-hour/week) for nine AHUs in university buildings.

The main source of fungal DNA on the HVAC filters may be the outdoor air. Outdoor air is known to contain several fungal taxa that may also have reached the filters (Noris et al., 2011). Nevertheless, although weak, some association can be seen between fungal DNA and cumulative occupancy. Occupancy could be a contributor to fungal bioaerosols through several mechanisms, such as resuspension from floors or shedding from clothes. People may be vectors for transport from outdoors, rather than the primary source (Qian et al., 2012).

The amount of non-microbial DNA on the filters also shows a strong relation with occupancy level. The non-microbial DNA was evaluated as the difference between the Qubit determination of total DNA and the qPCR evaluation of fungal plus bacterial DNA. Figure 2 displays the occupancy level and the total DNA accumulation rates on the same filters segmented based on the DNA origin.

Although the accumulation rate of bacterial DNA shows a closer relationship to occupancy level than fungal or total DNA, bacterial DNA only comprises 2-16% of total DNA in these samples. Fungal DNA, on the other hand, comprises a larger share: 5-37% of total DNA. Taken together, bacterial and fungal DNA comprises 12-40% of total DNA on these samples. This evidence suggests that nonmicrobial biomass, which may originate from humans, animals or plants, also are important contributors to total biomass collected on HVAC filters.

#### **CONCLUSIONS**

This study presents baseline information for biomass accumulation on HVAC filters that serve typical office environments in buildings in Singapore's tropical climate. Bacterial-based biomass abundance on the filters correlates more closely with occupancy as compared to fungal-based biomass. Other sources of biomass, possibly associated with human, animal or plant cells, contribute the majority of total accumulation on the filters.

#### **Total DNA Accumulation Rate**

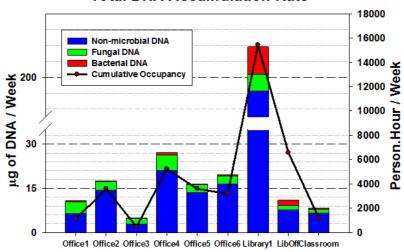


Figure 2. Weekly Total DNA accumulation rates on HVAC filters coupled with occupancy levels (in person-h/week) for nine AHUs in university buildings.

The findings in this study contribute to the foundation needed to further evaluate HVAC filter usage and operational practices, such as filter changing frequency or choice of filter performance rating. Bioaerosol analysis utilizing DNA-based techniques is a promising tool for further studies both to quantitatively estimate concentrations of various biological entities and also to investigate biodiversity on filters by means of sequencing.

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