National Biofilms Innovation Centre

Q&A: Bioaerosol Characterisation, the Air Microbiome and Covid-19 Transmission

Robert Ferguson

1. **How do filter/ impingement collection methods compare in relation to survival and viability of microbes rather than for molecular analysis?** Answered Live: filtration risks desiccation of the sample so I would recommend impingement for this.

2. **Did you get the chance to test a space that was not clean and then apply a cleaning product to understand what effect this has on the microbes identified?** Answered Live: Not yet, I have been working on outdoor bioaerosols. It is something we are interested in for future work especially with increased indoor cleaning during the pandemic.

3. **Very interesting, thank you. Is it possible to carry out metagenomic sequencing on the air?** Yes, we have done metagenomes as well as metabarcoding/amplicon sequencing. Data is not published yet. It is hard to get enough DNA for metagenomes and a random PCR amplification step is a good idea. When it works, we have got nice results, e.g., assembling lots of quality MAGs etc. We give some advice in this paper. We have struggled with mRNA for transcriptomes.

4. **What about virus detection in aerosols?** Yes, it can be done. I am not an expert in viruses myself but we have a virus researcher in the department and have been looking at possibilities. The LAMP would be perfect for viruses, including RNA viruses which have become topical. Also, in our metagenome data it is likely that we have DNA virus data that we have not looked at yet as I have been looking at bacteria/fungi.

5. **Do you remove free DNA in your samples before analysis?** Not deliberately. But both main collection methods we use would bias against anything that small, so I would think that it would be hard for us to collect free DNA.

6. **Is there any rule of thumb that we can use to guess how much of an air sample do we need when doing indoor sampling? How do you decide your sampling size for outdoor?** I don't know for indoor, but I think our recommendations here would still be valid for indoor.

7. **How do you know that the air samples collected were enough to represent the outdoor environmental sample?** It is probably unknowable, but: One thing we do is sampling over a full day (e.g., 27 samples over 8 hrs) over multiple days so that it is representative of temporal variation. We can also look at this in the data by looking for saturation in alpha-diversity. We have also looked at diversity over different sampling time periods. But mainly this comes down to how long do we need to sample to get enough DNA.

8. **Why is the short sample period has more biomass than long sample periods?** This topic requires a webinar of its own! When comparing filters (long time period) and impingement (short time period). The longer time period (filters) got more biomass, but lower diversity. The impingement collects at a faster rate, 300 lpm v 28 lpm (for filters). So, impingement collects a more representative sample, but over a shorter time. Note that when we compare different time periods with the same sampler, we get more biomass with time, but it is not linear. More info can be found here.

9. **Would it possible to discriminate the biological and nonbiological particles? fluorescence intensity, size etc**. Yes, the SIBS and WIBS can do this with fluorescence. In practice particles will be agglomerations of biological and nonbiological components

Jiayu Li

1. **Thanks for a great talk. Did your sampling strategy permit the study of a continuum from the outdoors into the indoor bedroom area e.g., were there more larger particles near the window that decreased spatially at the sampling points within the room?** Sorry if I missed this information during the talk. No, we haven't studied the spatial variation of bioaerosols within the room. In our setup in the bedroom, we treated the room as a well-mixed environment, which means we assume that the particle characteristics within the room were equal.

Emma Jane Goode

1. **Who sets the limits and do PHE influence changes in these?** Answered live.

All

1. **Has anyone got any recommendations and/or methodologies for Covid Air detection?** Awaiting feedback.