

Field Performance of a New Technology with the Potential to Identify Allergy and Asthma Triggers

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Abstract

RATIONALE: The Compact Ionic Capture Device (cICD) is a consumer friendly device that collects aerosol particles for testing. The aim is to evaluate its performance for a range of analytes and field conditions. **METHODS:** Sites were a clean bathroom, a basement with sump drain, and a hay storage room in an equestrian facility. The ICD was run for up to 24 hours at approximately 100 lpm. Reference was 0.4 µm polycarbonate filters pumped at 15 lpm. Analytical procedures were MARIA™ 9-plex immunoassays for allergens (Indoor Biotechnologies), multiplex qPCR for 23 indoor molds (EMLabsP&K), and next generation (Illumina) sequencing with Procrustes analysis for V region of bacterial 16S rRNA. **RESULTS:** Despite the presence of a unique spectrum of analytes in each environment, there was concordance between cICD and filter for presence or absence of 7 allergens and 21 mold species across all environments. In several instances, significant levels of allergens or spore equivalents were found by the cICD and not by filters. The cICD and filters both showed concordant bacterial community compositions dominated by Actinobacteria, Cyanobacteria, Proteobacteria, and Bacteroidetes. The cICD's high flow rate permitted faster detection of analytes than the filter. **CONCLUSIONS:** There was remarkable consistency between the performance of the cICD and filters over a wide range of environmental types and airborne analytes. Therefore, the cICD may be used to measure and discover new aeroallergens. In clinical practice, it may easily and reliably confirm suspected allergen exposure, direct avoidance recommendations and assist individualization of therapy for allergic patients.

Significance

The aerobiome is a new frontier in environmental discovery. Numerous diseases are spread through airborne biological agents and changes in the microbiome are affecting human health in ways previously unimagined. Testing for airborne transmission of disease agents is currently technologically challenging. This work introduces a very simple plug-and-play device that can sample air at a high volume flow rate with no moving parts. Aerosol particles are captured on electrodes and eluted for testing. We show that performance of the device is substantially equivalent to capture by pumping through a filter for immunological and molecular analysis. **Application to allergy patients is in poster number L10.**

Methods

Air sampling was done in three locations representing a range of environments. Sampling was done in parallel with the Inspirotec Sampler (left) and Zefon 37 mm cassette with 0.4 µm polycarbonate filter, pumped with high volume rotary vane pump at 15 lpm. Metal strip electrodes and filters were extracted and analyzed as indicated.



Mold spores and allergens in 3 locations

	Spore equivalents (units per 10,000 liters of air)					
	Bathroom		Basement		Stable	
	Inspiro	Filter	Inspiro	Filter	Inspiro	Filter
<i>Acremonium strictum</i>	<0.4	<3	<0.5	<3	38.00	50.93
<i>Alternaria alternata</i>	<0.5	<3	1.77	3.70	28.83	78.70
<i>Aspergillus flavus</i>	<2	<14	5.24	35.19	4,323.90	9,259.26
<i>Aspergillus fumigatus</i>	0.46	<1	1.57	46.30	8,713.31	14,814.81
<i>Aspergillus niger</i>	<0.1	<0.9	<0.2	<0.9	64.86	208.33
<i>Aspergillus ochraceus</i>	1.64	5.09	41.93	2,500.00	0.39	<1.4
<i>Aspergillus sydowii</i>	<11	<90	<14	<90	1,637.84	<33
<i>Aspergillus ustus</i>	<0.3	<2	<0.3	<2	<1	<1
<i>Aspergillus versicolor</i>	<4	<33	21.62	342.59	1,074.42	2,962.96
<i>Chaetomium globosum</i>	<0.1	<0.9	<0.1	<0.9	7.47	0.93
<i>Cladosporium cladosporioides (Type 1)</i>	1.24	0.93	39.96	370.37	373.43	648.15
<i>Eurotium (Asp.) amstelodami</i>	3.73	<0.5	1,972.62	458.33	262,054.51	555,555.56
<i>Memnoniella echinata</i>	<0.1	<0.5	<0.1	<0.5	<0.2	<0.5
<i>Paecilomyces variotii</i>	<0.1	<0.5	3.87	16.20	6,027.25	10,185.19
<i>Penicillium aurantiogriseum</i>	<0.7	<6	<0.9	<6	222.75	180.56
<i>Penicillium brevicompactum</i>	20.70	92.59	19.85	375.00	1,356.13	925.93
<i>Penicillium chrysogenum (Type 2)</i>	<0.2	<3	1.57	37.96	1,388.89	2,500.00
<i>Penicillium purpurogenum</i>	<0.1	<1.4	<0.3	<1.4	<0.7	<0.5
<i>Penicillium variable</i>	<1	<9	<1	<9	0.98	<3
<i>Scopulariopsis brevicaulis</i>	0.13	<0.9	<0.2	<0.9	176.89	337.96
<i>Stachybotrys chartarum</i>	<0.7	<6	1.83	<6	3.93	12.04
<i>Trichoderma viride</i>	<0.1	<0.5	0.33	<0.6	27.52	60.19
<i>Ulocladium botrytis</i>	<0.2	<1.4	1.31	3.24	18.15	15.28
			Allergens (fg/liter of air)			
Der p 1	<0.4	<3	<0.4	<3	<0.4	<3
Der f 1	<0.4	<3	<0.4	<3	<0.4	<3
MG2	<0.1	<1	<0.1	<1	<0.1	<1
Fel d 1	<0.1	<1	<0.1	<1	17.7	38.9
Can f 1	<0.4	<3	<0.4	<3	4.9	13.5
Rat n 1	<0.1	<1	<0.1	<1	8.4	18.3
Mus m 1	<0.07	<0.5	<0.07	<0.5	276.5	389.7
Alt a 1	<0.13	<1	<0.13	<1	2.8	3.2
Asp f 1	0.7	67.5	<0.3	<2	6.5	<3

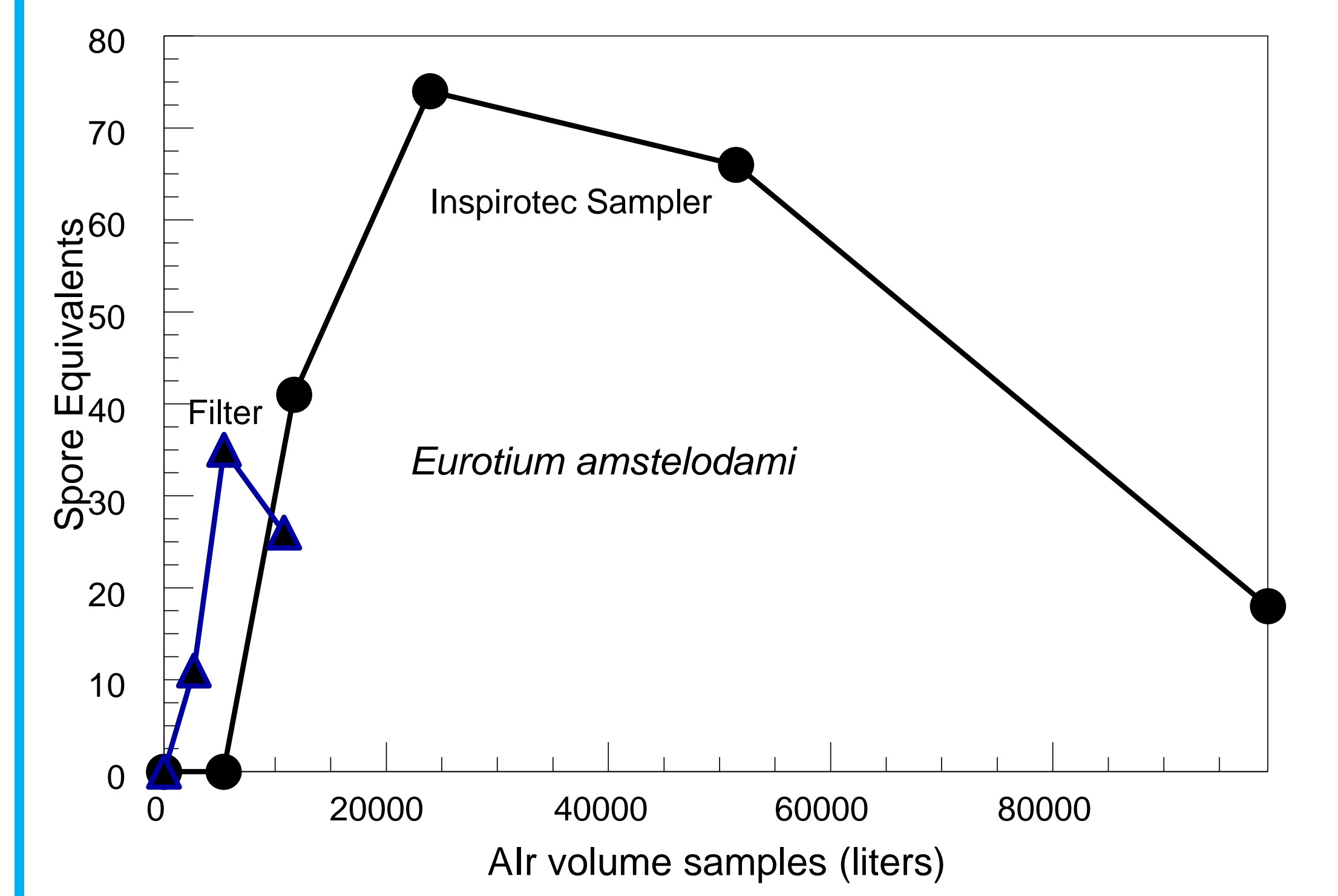
KEY:

Nosocomial significant	Yellow
Missed by filter	Blue
Consistency	Green
Below detection limit	Grey

In each location, 2 of each, Inspirotec samplers and filters, were run in parallel for 24 hrs. One of each pair was sent to EMLabs for their qPCR for 23 important Indoor Molds, and the other to Indoor Biotechnologies for their MARIA™ 9 plex for common household allergens.

In other runs the mouse *Mus m 1* allergen capture had cv of ±20% with 20 parallel replicates. The qPCR values have considerably higher cv's because of the nature of the method. Relative capture efficiency with fluorescent microparticles in a controlled environment was 22% (performed by Alburty Labs, Drexel, MO). Results are all expressed as nominal concentration in the air based on air volume flow. 17 of the 22 molds and 6 of the 9 allergens were detected by both types of sampler. Eight molds were detected by the Inspirotec sampler and not by the filter. This may be accounted for by the larger volume of air sampled by the Inspirotec Sampler in the given time.

Time course of mold spore capture in basement



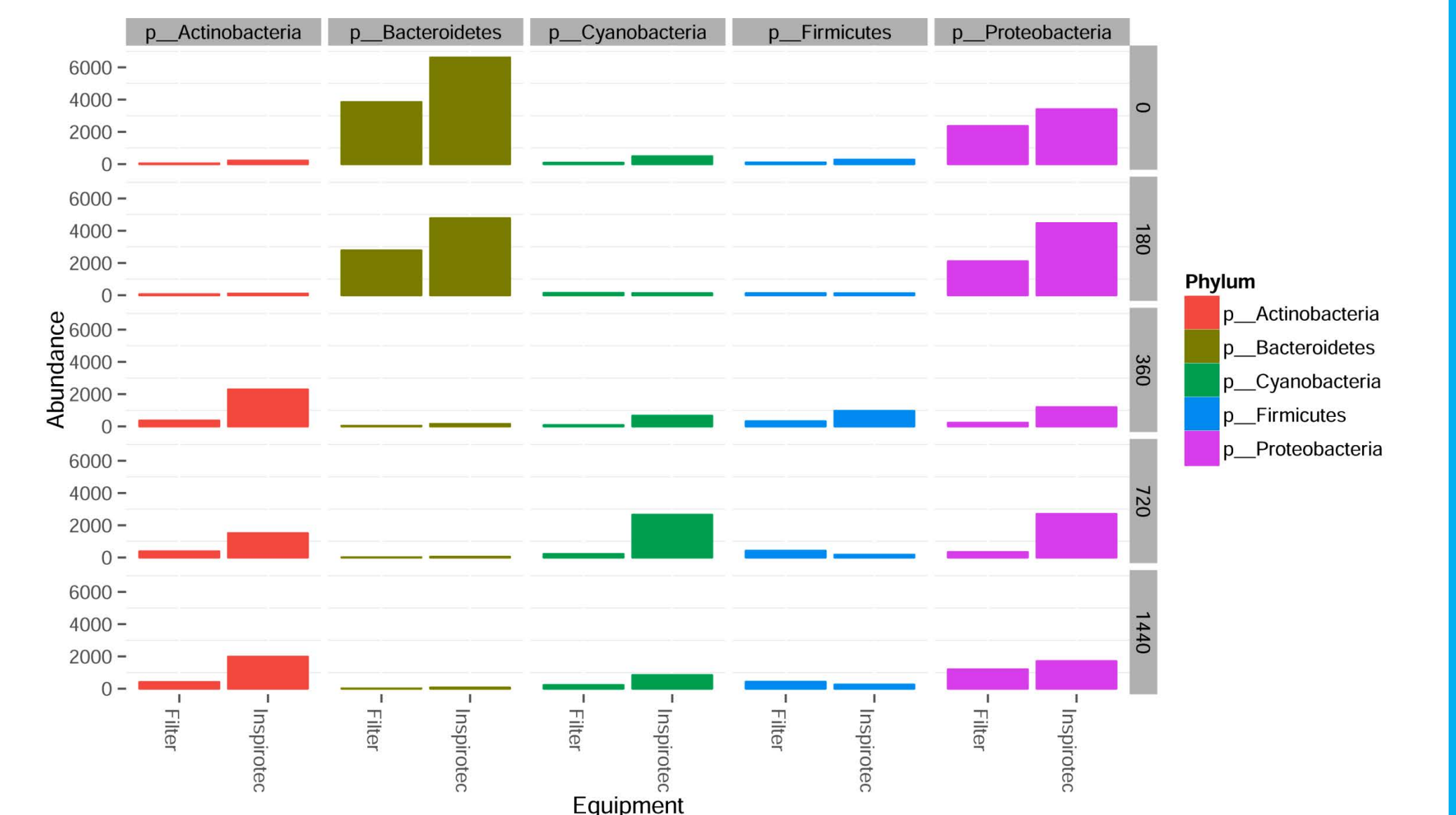
Five Inspirotec samplers and 3 filters with pumps were run in parallel in the basement as in the preceding table, and removed for analysis at timed intervals up to 24 hrs. The results are plotted as volume of air sampled during the time interval. Amounts are number of spore equivalents captured per sample. For both, the spore equivalents determined by qPCR became detectable at 6 hrs of sampling.

The Inspirotec sampler apparently captures with a higher efficiency than the reference filter. It is not obvious why both fall off at the longest sampling times. Conditions optimized for extraction of allergens may not be optimal for spores as biomass increases.

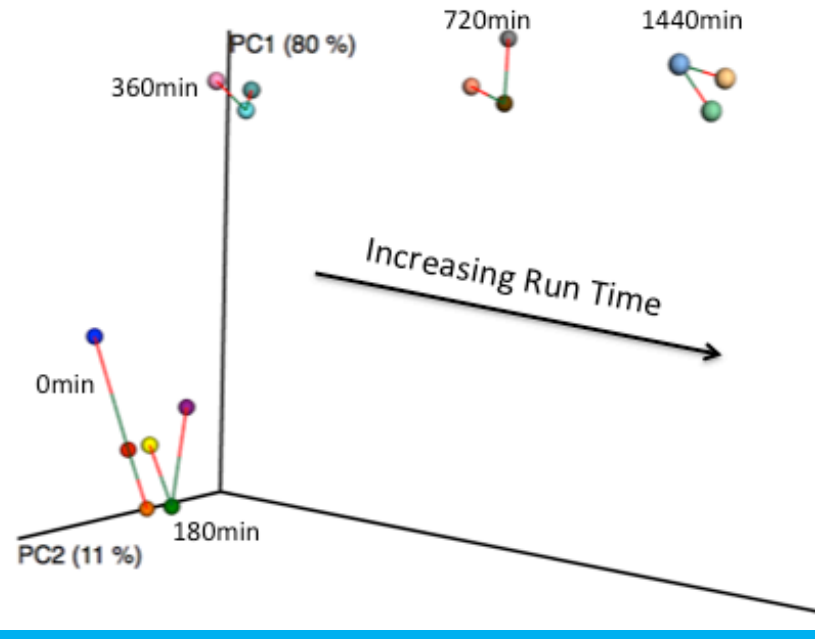
General Conclusion

The Inspirotec Sampler collects biomass from the air and analysis shows species equivalent to those captured by a polycarbonate filter reference method, for allergens, molds and bacteria. The device is simpler to operate and samples at a higher volume flow rate than the reference method. Currently, there is interest in interaction between the microbiome and allergic disease. This opens the way to the discovery of interaction between the aerobiome and allergic disease. **It is also the underpinning for the patient data in poster L10.**

Next generation sequence analysis of bacteria captured in basement



Experimental design was the same and samples were run in parallel with those of the preceding. Bacterial 16S rRNA amplicon sequencing generated a total of 1,294,310 sequences from 22 samples. Left and right electrodes were run separately effectively as duplicates. These data were processed to a rarefaction depth of 9,800 sequences per sample, which comprised 385,076 OTUs (Operational Taxonomic Units; 97% identity). No significant difference in microbial community structure was observed between the technologies (weighted or unweighted UniFrac distance (ANOVA, p > 0.05, R = 0.06). The phylum-level community profile generated by both technologies comprised predominantly Proteobacteria (40%), Bacteroidetes (23%), Actinobacteria (10%) and Firmicutes (9%).



By Procrustes analysis, a Principal Coordinates Analysis (PCoA) plot – shows unifracs distance space. There was no significant difference between either sampler duplicates nor the air filter at later times (Monte Carlo, p > 0.05). Highest variability was for the blank at zero time and for the 180 min sampling time.

Acknowledgements

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