

Design and Evaluation of the Field-Deployable Electrostatic Precipitator with Superhydrophobic Surface (FDEPSS) with High Concentration Rate

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ABSTRACT

Here we report on further development of an electrostatics-based bioaerosol collector with high concentration rate. We developed a field-deployable version of the electrostatic precipitator with superhydrophobic surface (FDEPSS), which consists of two combined half-cylinder collection chambers and an integrated control box. The collector is made of a static dissipative material and each collection chamber features a 3.2 mm wide collection electrode. The round top part of each chamber contains eight carbon fiber ionizers arranged in two lines of four. The collected particles are removed by a 20 µL rolling water droplet. Sampler's components were integrated into a control box.

The FDEPSS was tested with two bacterial species, *Bacillus atropheus* and *Pseudomonas fluorescens* bacteria, and one fungal spore, *Penicillium chrysogenum* for 10 and 60 min collection times and showed collection efficiency of ~70% at a sampling flow rate of 20 L min⁻¹. The use of a collecting water droplet of 20 μ L per collection chamber achieved sample concentration rates approaching 0.5 × 10⁶ min⁻¹. The FDEPSS was also tested against BioSampler (SKC Inc., Eighty Four, PA) and Button aerosol sampler (SKC Inc.) when sampling bioaerosols outdoors for 60 min. The samples were characterized based on the total airborne adenosine triphosphate (ATP) concentration, which was reported as relative luminescence units (*RLU*). The FDEPSS detected 5.1 × 10⁵ *RLU* m⁻³, while the BioSampler and the Button sampler showed 4.1 × 10⁵ *RLU* m⁻³ and 8.7 × 10⁵ *RLU* m⁻³, respectively. Since ATP analysis can be performed with small sample volumes, and the FDEPSS captures particles into 20 μ L of liquid, resulting in a high concentration rate, we show that this sampler can detect the presence of airborne microorganisms 40× faster than the BioSampler or Button aerosol sampler. This FDEPSS feature could be integrated into bioaerosol detection systems, especially where concentrations are low and time is critical.

Keywords: Field-deployable bioaerosol sampler; Electrostatic collection; Concentration rate; Adenosine triphosphate (ATP); Bioaerosol detection system.

INTRODUCTION

Bioaerosols (airborne microorganisms) comprise approximately ~10% of urban and rural fine aerosols (Monn, 2001) and could be found in almost any indoor or outdoor air environment (Yassin *et al.*, 2010; Prussin *et al.*, 2015). They are known to play an important role in many negative health-related effects (Curtis *et al.*, 2006; Kim *et al.*, 2013), including those associated with atmospheric processes. For example, mineral-dust particles were found to be associated with bioaerosol (Maki *et al.*, 2014). Airborne bacteria carried by dust events increase allergenic burden, causing increased incidences of asthma (Ichinose *et al.*, 2005). Moreover, bioaerosols are thought to influence atmospheric processes by participating in atmospheric chemical reactions and cloud particle formation (Creamean *et al.*, 2013). Therefore, it is important to be able to accurately identify and quantify bioaerosol concentrations indoors and outdoors (Prussin *et al.*, 2015). In addition, selection of bioaerosol samplers with an appropriate analysis method is another important factor determining whether bioaerosols are detected at various environmental conditions (Mandal and Brandl, 2011).

A number of bioaerosol samplers have been developed and evaluated, including comparison of portable versus stationary samplers, or passive versus active samplers (Mehta *et al.*, 1996; Kenny *et al.*, 1999; Bellin and Schillinger, 2001; Agranovski *et al.*, 2002; An *et al.*, 2004; Madsen and Sharma, 2008). Most popular bioaerosol sampler types include impingers and impactors (Macher and Macher, 1997; Kesavan and Sagripanti, 2015). Water-based samplers,

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such as impingers, allow analyzing bioaerosol samples by a variety of methods, including epifluorescence microscopy (Lunau *et al.*, 2005), culture-based methods (Noble *et al.*, 2010), flow cytometry (Veal *et al.*, 2000), quantitative polymerase chain reaction (qPCR) (Noble *et al.*, 2010), adenosine triphosphate (ATP)-based bioluminescence method (Seshadri *et al.*, 2009), and liquid chromatography mass spectrometry (LC-MS) (Ho and Reddy, 2010).

In recent years, several electrostatics-based bioaerosol samplers have been developed (Han and Mainelis, 2008; Madsen and Sharma, 2008; Roux et al., 2013). Han and Mainelis (2008) developed a prototype electrostatic precipitator with superhydrophobic surface (EPSS, Mark I) to capture airborne bacteria and fungal spores and concentrate them into small amounts of liquid (Han et al., 2010; Han et al., 2011). In this sampler, airborne particles are electrostatically deposited onto a narrow electrode (2.1–3.2 mm in width) covered by a superhydrophobic substance and then removed and collected by rolling water droplets $(5-60 \ \mu L)$ to achieve a high sample concentration rate (i.e., the ratio of particle concentration in the collection liquid versus the airborne particle concentration per time unit). The prototype EPSS has also been successfully tested with microscopy, ATP and qPCR sample analysis methods (Han et al., 2010, 2011, 2015). It also preserved integrity of the collected bacteria better than an impactor or an impinger (Zhen et al., 2013). The prototype EPSS model was further developed as a single-stage electrostatic collector with more effective materials for the sampler body and electrode, and optimized ionizer configuration (EPSS, Mark II) (Han et al., 2015). The EPSS Mark II is made of a static dissipative material (e.g., Delrin) and uses a collection electrode made of pressed carbon. The incoming particles are charged using eight carbon fiber brushes. The EPSS Mark II showed an overall collection efficiency of up to 84% when sampling *Escherichia coli* at 10 L min⁻¹. The collected bacteria were captured by a rolling water droplet as small as 20 µL thus achieving concentration rate of up to $4.2 \times 10^5 \text{ min}^{-1}$.

However, this performance of EPSS Mark II was achieved when using bench scale components, such as large power supplies and external pumps. For the application and testing of this technology in the field, the components of the sampler were downsized and integrated into a field-deployable instrument. Thus, the primary objective of this project was to build a field-deployable EPSS and evaluate its performance with commonly available biological particles at different sampling flow rates, and sampling periods. Finally, the sampler's performance was compared with two commercially available biosamplers when sampling in an outdoor environment.

MATERIALS AND METHODS

Design Features of the Field-Deployable Electrostatic Precipitator with Superhydrophobic Surface (FDEPSS)

The field-deployable version of the EPSS (FDEPSS) is shown in Fig. 1. It consists of an electrostatic collector and an integrated control box (Fig. 1(a)); both were fabricated using conventional machining and 3D printing. The latest iteration of the collector consists of two half-cylinder collection chambers joined together, forming a cylinder, thus allowing collection of two samples concurrently (Fig. 1(c)). Two samples could be analyzed by different techniques to obtain more information about the bioaerosols, or they could be combined to improve detection limit.

Fig. 1(d) presents a 3D view of the entire collector and a 3D view of the flat bottom plate. In each half cylinder, a flat bottom section holds a narrow collection electrode (width (w): 3.2 mm \times length (l): 254 mm) covered by a superhydrophobic substance (HIREC-1450, NTT Corporation Inc., Japan). The round top half-cylinder section houses eight carbon fiber brushes arranged in two lines of four brushes and positioned at 45 and 135 degree angles in the cross-section of the collector. The diameter of the cylinder, d, is 50.8 mm. Since the carbon brushes are connected to high voltage and the collection electrode is grounded, this configuration creates an ion cloud which charges the incoming particles and then deposits them onto the collection electrode. The single-stage EPSS design requires only one single power supply for both charging and collection (Han et al., 2015), thus reducing the cost and complexity of its design and field deployment. The dual-sided collector is made of static dissipative material: homopolymer acetal (Delrin, Professional Plastics Inc., Fullerton, NY). In addition, a bell shape inlet fabricated by 3D printing and an inlet screen (nickel electroformed screen: 20×20 mesh, 65 µm in diameter, 90% fraction of open area, Industrial Netting Inc., Minneapolis, MN) are attached to the collector to prevent unwanted large-sized debris such as insects, plant fragments, and fibers from entering the sampler.

All components necessary to operate the FDEPSS, including DC-to-DC high voltage power converter/supply (F121, EMCO Corp., Sutter Creek, CA), voltage divider (V1G, EMCO), batteries (e.g., 9 V alkaline), voltage regulators (OS-1212CCBA-80W, Oskipower Co., China), power analyzers (130A Watt, Powerwerx Inc., Yorba Linda, CA), and switches (Grainger Inc., South Plainfield, NJ) are integrated in a control box (Fig. 1(b)). The collector is easily connected with the control box by sliding it into the base adaptor which was fabricated by 3D printing. The base adaptor houses an air mover (computer fan, NF-A4×10, Noctua Co., Austria). The benchtop integrated controller is housed in a rugged enclosure with dimensions of width (W): $254 \text{ mm} \times \text{length}$ (L): $254 \text{ mm} \times \text{height}$ (H): 100 mm. Using controls mounted on the front of the control box (Fig. 1(a)), a user can easily turn the instrument on/off, regulate input voltages for the air mover and particle charger/ collector. The high voltage power supply is a DC-to-DC converter, which receives input voltage from two highcapacity 9 V batteries with a regulator (Fig. 1(b)). Voltage applied to the charger/collector is monitored using a power analyzer of the control box or an external voltage meter through a voltage divider (1000:1). Power to the air mover is provided by a separate battery and can be easily adjusted to achieve the desired sampling flow rates.

After completing the sampling, the collector is removed from the base adaptor and then the collection electrode is

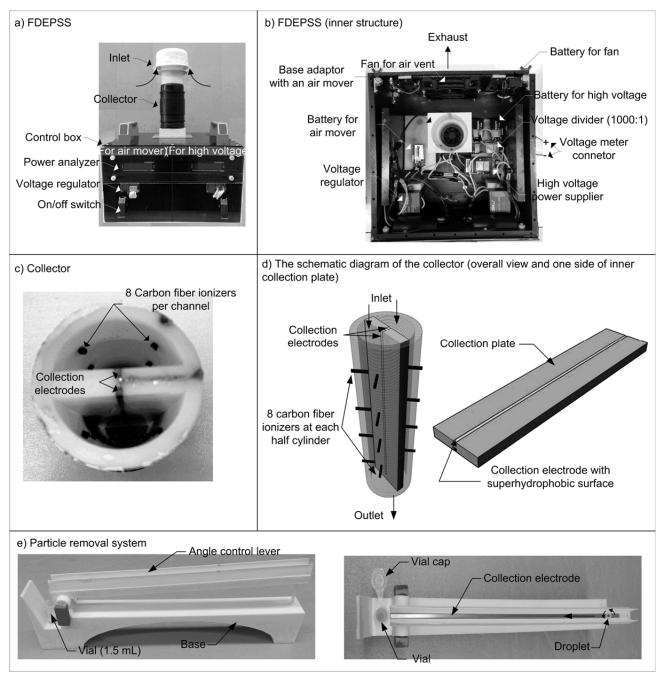


Fig. 1. A prototype of the field-deployable version of the electrostatic precipitator with superhydrophobic surface (FDEPSS) made of static dissipative material by machining and 3D printing. The sampler incorporates a particle charger with eight carbon brushes (400 fibers/brush) arranged in two rows, stainless steel electrodes ($254 \times 3.2 \times 2.5$ mm). All components are assembled in the control box: a) FDEPSS, b) FDEPSS (inner structure), c) collector, d) the schematic diagram of the collector, and e) particle removal system.

moved to the particle removal system (Fig. 1(e)), which was fabricated by 3D printing. A droplet (e.g., 20 μ L) is manually injected at the top of the collection electrode. Due to the inclination of the electrode holder the liquid droplet rolls down picking up the deposited particles and is then collected in a vial located at the bottom of the removal system. Inclination of the electrode holder can be easily adjusted with a lever. We found that ~5–10° angle is most effective (Han and Mainelis, 2008).

Experimental Setup for Testing FDEPSS in Laboratory

The FDEPSS was tested with green fluorescent polystyrene latex (PSL) particles (Duke Scientific Corp., Palo Alto, CA), two species of bacteria and one species of fungi. The test system is shown in Fig. 2, and it consisted of a flow control system, a particle generation system, an air-particle mixing system, and a particle monitoring system. The system was housed inside a Class II Biosafety cabinet (NUAIRE Inc., Plymouth, MN).

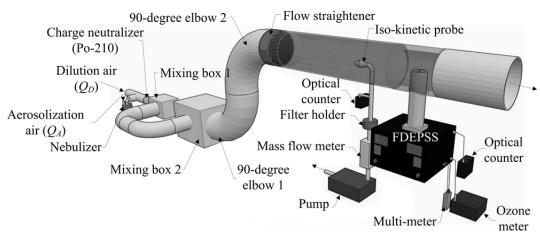


Fig. 2. Schematic diagram of the experimental setup.

A three jet Collison nebulizer (BGI Inc., Waltham, MA) was used to aerosolize test particles from a liquid suspension at a flow rate (Q_A) of 5 L min⁻¹ (pressure of 12 psi). A HEPAfiltered dilution air flow, $Q_D(105 \text{ Lmin}^{-1})$, provided by an in-house compressor was used to dilute the particle stream; it was controlled by a pressure regulator and monitored by a mass flowmeter (TSI Inc., Shoreview, MN). The dilution air and aerosolized particle stream were combined $(Q_D + Q_A)$ = 110 L min⁻¹) and passed through a 2-mCi Po-210 charge neutralizer (Amstat Industries Inc., Glenview, IL) to reduce aerosolization-related particle charges to Boltzmann charge equilibrium. The electrically neutralized particles then passed through the first mixing box (Han et al., 2005) which improved the uniformity of particle distribution across the flow cross-section. A second mixing box (Han et al., 2005) and a U-type duct connector further improved particle mixing. A well-mixed flow stream then entered a raised test duct (0.152 m (6 inches) in diameter and a 0.91 m (36 inches) in length), as shown in Fig. 2. A raised test duct allowed the FDEPSS collector to be perpendicularly oriented relative to the air stream simulating a realistic field sampling scenario. A flow straightener was placed at the exit of the second elbow to eliminate large scale turbulence and flow swirl generated by the mixing boxes and the 90-degree elbows. The FDEPSS was vertically elevated by an adjustable-height support stand and positioned four duct diameters downstream of the exit of the flow straightener in order to provide uniform crosssectional profile of test particles. The coefficient of variation (i.e., the ratio of the unbiased standard deviation of a set of measurements to the mean of the set of measurements, COV) of 0.5 µm PSL concentration across the test duct was about 2.7% at the measurement location. The COV was measured over five equally distributed sampling points in the cross-sectional area of the duct in triplicate. The reference aerosol concentration was determined by the following methods: a) an optical particle counter (OPC) (model 1.108, Grimm Technologies Inc., Douglasville, GA) through a probe positioned downstream of the collector with its power OFF, b) a filter (e.g., glass fiber filter for PSL particles and membrane filter for bioaerosols) through an isokinetic probe positioned 1 duct diameter upstream of the FDEPSS,

or c) an OPC through an isokinetic probe positioned 1 duct diameter upstream of the FDEPSS. The FDEPSS sampling flow rate was provided by a computer fan; its flow rate as a function of input operating voltage was calibrated using a hot wire anemometer (TSI Inc., Shoreview, MN). The air velocity at input voltages from 4.2 to 10.3 V was measured inside the collector, close to its inlet (about 0.381 m away from the fan). The ozone concentration was measured using a UV photometric ozone monitor (MODEL202, 2B Technologies Inc., Boulder, CO) at the outlet of the collector in the FDEPSS system (Fig. 2).

Tests with PSL Particles

The sampler's performance was tested with fluorescent PSL particles of 0.5, 1.0, and 3.0 μ m in geometric diameter and at sampling flow rates, Q_s , of 20, 40, and 60 L min⁻¹ for 3 min sampling time (by the OPC) or 10 min (by the filter). The airborne concentration of fluorescent PSL particles was approximately 10^3-10^4 Liter⁻¹. The collector was tested at charging/collection voltages of -8, -9, and -10 kV, and an uncoated stainless steel collection electrode was used.

The overall collection efficiency of the FDEPSS was determined by comparing particle concentration downstream of the collector with its power ON and OFF using a Grimm OPC. The actual collection efficiency was determined by comparing the number of particles deposited on each FDEPSS collection electrode and removed by a 20 μ L autoclaved deionized (DI) water droplet with the number of particles isokinetically sampled onto a reference filter (type A/E, 47 mm, Pall Inc., East Hills, NY). PSL particle concentration in each sample was determined by measuring its fluorescence intensity using a digital filter fluorometer (Turner Quantech model FM109515, Barnstead/Thermolyne Corp., Dubuque, IA) as described previously (Han and Mainelis, 2008). The overall collection efficiency from the collection electrode, $\eta_{OVERALL, PSL}$, was determined as follows:

when OPC was used:

$$\eta_{OVERALL, PSL} = 1 - \frac{C_{ON}}{C_{OFF}} \tag{1}$$

where C_{ON} and C_{OFF} are airborne particle concentrations downstream of FDEPSS with its voltage ON and OFF, respectively; this metric does not take into account particle losses inside the collector.

The actual collection efficiency from a water droplet, $\eta_{ACTUAL, PSL}$, was determined as follows:

when fluorometry was used:

$$\eta_{ACTUAL, PSL} = \frac{C_{droplet}}{C_{reference filter}}$$
(2)

where $C_{droplet}$ and $C_{reference filter}$ are aerosol concentrations in a water droplet and isokinetically sampled onto a reference filter, respectively, based on fluorometer reading.

Tests with Biological Particles

The efficiency of the FDEPSS when collecting biological particles was determined with gram-positive Bacillus atrophaeus bacterial cells (ATCC 49337, American Type Culture Collection, MD), gram-negative Pseudomonas fluorescens (ATCC 13525), and fungal spores Penicillium chrysogenum (ATCC 10135). These species of bacteria and fungal spores have been widely used in bioaerosol studies (Hill et al., 1999; Johnson et al., 1994; Nadkarni et al., 2002). The bacteria were cultured in nutrient broth (Becton, Dickinson and Company, Sparks, MD) for 18 hours at 30°C (B. atrophaeus) or 26°C (P. fluorescens) (Han et al., 2010). The cells were harvested by centrifugation at 7000 rpm (6140g) for 5 min at 4°C (BR4, Jouan Inc., Winchester, VA) and then washed 4 times with sterile DI water (Millipore Corp., Billerica, MA). The final liquid suspension was diluted with sterile DI water to obtain a target airborne cell concentration of $\sim 10^3$ to 10^4 cells Liter⁻¹ as determined by the Grimm OPC. P. chrysogenum was plated onto Sabouraud dextrose agar (Becton Dickinson Microbiology Systems, Cockeysville, MD, USA) and incubated at room temperature (approximately 26°C) for seven days following an established protocol (Han et al., 2011). After incubation, about 3 ml of sterile deionized water was added to each plate of P. chrysogenum and the spores were gently harvested from mycelium using a cell spreader. The volume of the resulting spore suspension was then increased to 50 mL.

The FDEPSS was tested at sampling flow rates $Q_S = 20$, 40, and 60 L min⁻¹, and for 10 and 60 minute sampling times. Fresh liquid suspension (30 mL) of each species was prepared for each test, and was aerosolized using a Collison nebulizer, operated at flow rate of 5 L min⁻¹ (pressure of 12 psi).

The total number of cells removed by each 20 μ L droplet was determined by acridine orange epifluorescence microscopy (AOEM) using the Axioskop 20 (Carl Zeiss MicroImaging Inc., Thornwood, NY), according to a previously published method (Han *et al.*, 2010). Briefly, formaldehyde (100 μ L) and sterile DI water (880 μ L) were added to 20 μ L droplet to increase sample volume to 1 mL. The 1 mL sample was serially diluted in 10-fold dilutions with sterilized water to achieve a cell concentration that could be comfortably counted using a microscope (i.e.,

~20 cells per microscope view field). Each glass slide for microscope counting was prepared by filtering a sample through a 25 mm black polycarbonate filter (Thermo Fisher Scientific Inc., Suwanee, GA) and staining it with 1 mL of 0.1 μ g mL⁻¹ Acridine Orange solution (Becton Dickinson Microbiology System, Sparks, MD) for 15 minutes.

The total cell number in each sample, C_{sample} , was calculated as follows:

$$C_{sample} = N \times X \times D \tag{3}$$

where N is the average cell count in each microscope view field, X is the number of fields for the entire filter (X = 6125), and D is the dilution factor.

The reference concentration was determined by measuring the concentration of bioaerosols by a Grimm OPC through an isokinetic probe positioned 1 duct diameter upstream of the FDEPSS. The accuracy of using Grimm OPC to measure the reference concentration was examined by comparing the concentration of cells measured by the Grimm OPC with that measured using a reference filter (25-mm membrane filter, Pall Inc., East Hills, NY). Here, particles collected on the filter were eluted into sterile DI water as described elsewhere (Wang et al., 2001). The number of particles in the resulting suspension was determined by AOEM and compared with the Grimm OPC reading. Tests with each species were performed in triplicate. The two number concentrations agreed within $6.8 \pm 3.5\%$. Since bioaerosol measurement with the Grimm OPC is far less time-consuming than microscopy, it was used to determine reference concentration with biological particles.

Thus, the FDEPSS actual collection efficiency for bioaerosols (based on microorganisms in a water droplet), $\eta_{ACTUAL, BIO}$, was determined as:

$$\eta_{ACTUAL, BIO} = \frac{C_{sample}}{C_{Grimm} \times Q_S \times t}$$
(4)

where C_{Grimm} is the average airborne cell concentration (#/Liter) as measured by the Grimm OPC connected to the isokinetic probe every 6 s, Q_s (L min⁻¹) is the sampling flow rate of the FDEPSS, and t (min) is the sampling time. Here, if the sampling flow rate through the both FDEPSS sampling is used (e.g., 20, 40 or 60 L min⁻¹) then the C_{sample} is based on the two combined 20 µL droplets (one for each collection electrode).

In addition to collection efficiency, the sampler's concentration rate, R_C (min⁻¹), was calculated using the following operational parameters (Han and Mainelis, 2008):

$$R_C = \frac{Q_S}{V_{WD}} \times \eta_{ACTUAL, BIO}$$
(5)

where Q_s (L min⁻¹) is the sampling flow rate, V_{WD} (L) is the volume of the collecting sterile deionized water droplet, and $\eta_{ACTUAL,BIO}$ is the actual collection efficiency for biological particles.

Relative Performance of FDEPSS against Other Bioaerosol Samplers When Collecting Samples Outdoors

The FDEPSS was also pilot-tested when collecting samples outdoors on the Rutgers University Cook campus in New Brunswick, NJ, in March of 2015. Its performance was compared against a Button aerosol sampler (SKC Inc., Eighty Four, PA) and a BioSampler (SKC Inc.) when sampling bioaerosols for 60 min. The Button sampler was operated with a 0.2 µm pore size polycarbonate filter (Millipore, Billerica, MA) and its nominal sampling flow rate of 4 L min⁻¹. For the BioSampler, 5 mL of collection fluid was placed in an appropriate sampling cup and it was operated at a nominal sampling flow rate of 12.5 L min⁻¹. The FDEPSS was operated at a flow rate of 20 L min⁻¹ (10 L min⁻¹ per chamber) with the charging/collection voltage of -9 kV. Once the sampling by the three devices was completed, the particles collected on the Button sampler's filter were eluted into sterile deionized water (5 mL) using a previously described procedure (Wang et al., 2001). The collection liquid remaining in the BioSampler cup was transferred to a 50 mL centrifuge tube. Particles collected by each chamber of the FDEPSS were removed by two 20 µL rolling water droplets (one per chamber) and combined. Liquid samples from each device were transferred into a centrifuge tube (50 mL) and then sterile DI water was added to increase sample volume to 5 mL for subsequent analysis. Bioaerosol content of the samples was analyzed using the adenosine triphosphate (ATP)-based bioluminescence method (Venkateswaran et al., 2003; Seshadri et al., 2009; Han et al., 2011). All types of viable cells have a basic energy molecule, ATP (Karl, 1980), which, when combined with appropriate reagents, produces luminescence. The amount of light emitted during the reaction is directly proportional to the ATP content, i.e., viable bioaerosol mass. Here, 100 µL from each sample was combined with an equal volume of Bactiter-Glo reagent (Promega Corp., Madison, WI). The contents were briefly vortexed and then left at room temperature for 1 min. The luminescence intensity of the resulting suspension was measured by a luminometer (model 20/20ⁿ, Turner Biosystems Inc., Sunnyvale, CA) and recorded as relative luminescence units (RLU). The total airborne ATP concentration, C_{ATP} (RLU m⁻³), was determined for each tested device:

$$C_{ATP} = \frac{RLU \times V \times 1000}{0.1 \times Q_S \times t} \tag{6}$$

where *RLU* is the ATP concentration per 0.1 mL, *V* is the total volume of liquid sample (5 mL), Q_s is the sampling flow rate (L min⁻¹), *t* is the sampling period (60 min), and 1000 is a conversion factor from L into m³. Background *RLU* values for sterile deionized water (typically 1.5–2.0 × 10³ *RLU*/100 µL) were subtracted from *RLU* readout (Seshadri *et al.*, 2009).

RESULTS AND DISCUSSION

Fig. 3 presents overall collection efficiency, $\eta_{OVERALL, PSL}$, of

the FDEPSS as a function of collection/charging voltage for different PSL particle sizes (0.5, 1.0, and 3.0 µm) and sampling flow rates (20, 40, and 60 L min⁻¹). As the particle size increased from 0.5 to 1 μ m and then to 3 μ m, the collection efficiency for the same voltage and sampling flow rate also increased, since the electrical charges acquired by a particle in the field charging regime is proportional to its diameter squared (Hinds, 1999). When averaged over the three sampling flow rates and three collection/charging voltages, the increase for 1.0 µm PSL was 6.3% and the increase for 3.0 µm PSL was 26.2%, compared to the average collection efficiency for 0.5 µm PSL. As could be expected, the collection efficiency for all particle sizes and collection voltages decreased with increasing sampling flow rate, since particles were retained for shorter time periods in the collection chamber.

When carbon fibers are used to charge aerosol particles they are known to produce lower concentrations of ozone compared to often-used corona discharger (Han et al., 2009), likely due to the small diameter of the carbon fibers (~7 µm): Boelter and Davidson (1997) determined that smaller diameter of a discharge electrode produce less ozone. Overall, ozone production depends on the sampling flow rate, operational high voltage, polarity, relative humidity, and size and material of a charging electrode (Goheen et al., 1984; Kulkarni et al., 2002). Thus, we investigated ozone emissions by the FDEPSS as a function of sampling flow rate and collection/charging voltage (Fig. 4). During each test, the temperature in the test chamber stayed in the range of 25–28°C and the relative humidity (RH) ranged from 35% to 40%. Ozone concentration at the outlet of the collector in the FDEPSS increased non-linearly with increasing operating voltage, but decreased with increasing flow rate, when the same amount of ozone was diluted in a larger air volume. As the operating voltage increased, the ozone concentration averaged over all three sampling flow rates (20, 40, and 60 L min⁻¹) increased from approximately 7 ppb at -8kV, to 39 ppb at -9 kV, and then to and 113 ppb at -10 kV. The average overall collection efficiency at -9 kV was 56% (Fig. 3), i.e., only 19% lower than that at -10 kV (69%), but the average ozone concentration at -9 kV was about 66%lower than that at -10 kV. Since a -9 kV operating voltage yielded relatively good collection efficiency while resulting in a lower ozone concentration, it was utilized for the remaining experiments with biological particles.

One concern when using superhydrophobic surface as a coating for the collection electrode is its usability over multiple consecutive samples (Boinovich *et al.*, 2010). The data presented in Fig. 5 show that once the electrode is coated by the superhydrophobic substance, the particles can be collected and removed from the electrode multiple times without a loss in collection efficiency. The average collection efficiency, $\eta_{ACTUAL, PSL}$, based on a 20 µL water droplet was 63 ± 3.8% when collecting 1.0 µm PSL at 10 L min⁻¹ sampling flow rate per chamber with an -8 kV operating voltage and a 10 min sampling time. The coefficient of variation (*COV*) of the collection efficiency over the 10 replicate experiments was ~6.0%. Based on the one-way ANOVA test results, the difference in collection efficiency

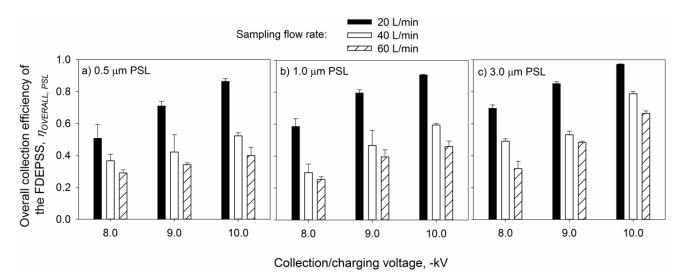
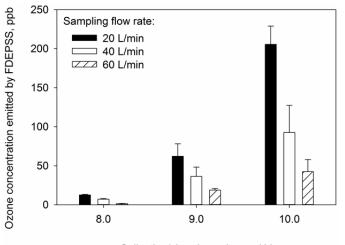


Fig. 3. Overall collection efficiency of the field-deployable version of the electrostatic precipitator with superhydrophobic surface (FDEPSS) as a function of collection/charging voltage (-8, -9, and -10 kV) when collecting polystyrene latex particles of a) 0.5 µm, b) 1.0 µm, and c) 3.0 µm at three sampling flow rates: $Q_S = 20$, 40, and 60 L min⁻¹. The data represent averages and standard deviations from at least three repeats.



Collection/charging voltage, -kV

Fig. 4. Average ozone concentration emitted by the field-deployable version of the electrostatic precipitator with superhydrophobic surface (FDEPSS) as a function of collection/charging voltage (-8, -9, and -10 kV) when collecting polystyrene latex particles (0.5, 1.0, and 3.0 µm) at three sampling flow rates, $Q_S = 20$, 40, and 60 L min⁻¹. The data represent averages and standard deviations from at least three repeats.

among subsequent experiments was not statistically significant (p = 0.119). Therefore, the superhydrophobic coating could be used for multiple collections.

For many bioaerosol sampling applications sampling times longer than 10 min are needed. Therefore, performance of the FDEPSS was tested for sampling times of 10 and 60 min. Fig. 6 presents actual collection efficiency, $\eta_{ACTUAL, BIO}$, of the FDEPSS when collecting *B. atrophaeus*, *P. fluorescens*, and *P. chrysogenum* as a function of sampling time (10 and 60 min) at two different flow rates (20 and 60 L min⁻¹) at –9 kV operating voltage. Concentrations of airborne *B. atropheus* and *P. fluorescens* bacteria were ~10⁶ m⁻³, and concentration of airborne *P. chrysogenum* fungal spores was ~10⁵ m⁻³. These concentrations are much higher than typically observed in indoor and outdoor environments (Prussin *et al.*, 2015) and were selected to simulate a high bioaerosol load.

The collection efficiency for *B. atropheus* (Fig. 6(a)) when sampling for 10 and 60 min was 69.1 \pm 2.2% and 40.2 \pm 0.5%, respectively, when collected at 20 L min⁻¹ total sampling flow rate (10 L min⁻¹ per sampling chamber). At 60 L min⁻¹ total sampling flow rate (30 L min⁻¹ per collection chamber), the collection efficiency was 32.7 \pm 1.8% and 14.8 \pm 1.7% for 10 and 60 min sampling times, respectively. The collection efficiency for *P. fluorescens* (Fig. 6(b)) was 58.7 \pm 4.9% and 28.8 \pm 2.7% when sampling for 10 and 60 min, respectively, at 20 L min⁻¹ total sampling flow rate. At 60 L min⁻¹ total sampling flow rate, the collection efficiency was 24.8 \pm 2.1% and 10.4 \pm 1.1% for 10 and 60

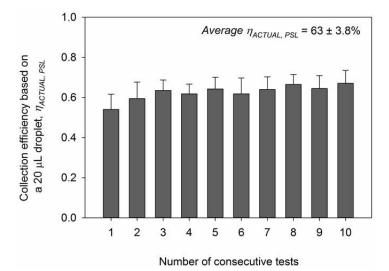


Fig. 5. Collection efficiency of the field-deployable version of the electrostatic precipitator with superhydrophobic surface (FDEPSS) when the same coating of the collection electrode is used multiple times. The tests were performed with polystyrene latex particles of 1.0 μ m at a 10 L min⁻¹ sampling flow rate and -8 kV collection/charging voltage. The data are averages and standard deviations based on five repeats.

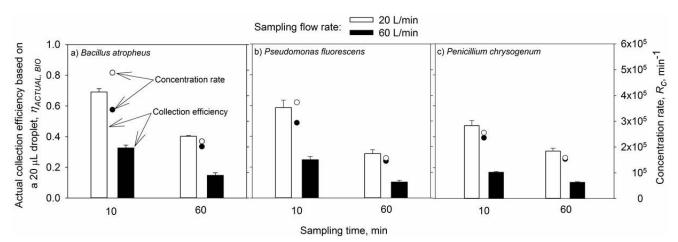


Fig. 6. Actual collection efficiency and concentration rate of the field-deployable version of the electrostatic precipitator with superhydrophobic surface (FDEPSS) as a function of sampling time (10 and 60 min) when collecting biological particles: a) *Bacillus atropheus*, b) *Pseudomonas fluorescens*, and c) *Penicillium chrysogenum* at two sampling flow rates, $Q_S = 20$ and 60 L min⁻¹ and at -9 kV collection/charging voltage. The concentrations of bioaerosols were ~10⁵-10⁶ m⁻³. The bars represent collection efficiency and the circles represent a concentration rate. The data represent averages and standard deviations from ate least three repeats.

min sampling times, respectively. For the fungal spores of *P. chrysogenum*, the average collection efficiency (Fig. 6(c)) for 10 and 60 min was $47.2 \pm 3.4\%$ and $30.7 \pm 1.9\%$ at 20 L min⁻¹ total sampling flow rate, respectively. The average collection efficiency for 10 and 60 min was $17.0 \pm 0.6\%$ and $10.5 \pm 0.4\%$ at 60 L min⁻¹, respectively.

For all three test microorganisms, the collection efficiency was statistically significantly affected by both sampling time and flow rate (p < 0.001). At a higher sampling flow rate, particles spend less time inside the FDEPSS and thus their chance of being deposited on the collection electrode decreases, especially if they carry a relatively lower electrical charge. When the sampling time increased from 10 to 60 min, the collection efficiency decreased by approximately 35–55%, depending on the test microorganism. The observed decrease may have been due to a less efficient removal of the deposited particles by the water droplet due to the increased adhesion forces between the collected particles and the electrode as a result of their prolonged contact; these findings are similar to our previous study (Han *et al.*, 2010).

Fig. 6 also shows sampler concentration rates, R_c , based on the presented collection efficiency data, the 20 µL volume of the collection droplet for each chamber, and sampling flow rates of 20–60 L min⁻¹ (10–30 L min⁻¹ per chamber). Depending on the sampling time and flow rate, the concentration rates ranged from $2.0 \times 10^5 \text{ min}^{-1}$ to $4.9 \times 10^5 \text{ min}^{-1}$ for *B. atrophaeus*, from $1.4 \times 10^5 \text{ min}^{-1}$ to $3.7 \times 10^5 \text{ min}^{-1}$ for *P. fluorescens*, and from $1.5 \times 10^5 \text{ min}^{-1}$ to 2.6 × 10^5 min^{-1} for *P. chrysogenum*. Even when the concentration rate is calculated based on a 20 µL droplet per chamber and the lowest observed collection efficiency (about 10% at 60 min), the concentration rate still exceeds 10^5 min^{-1} . For short sampling times, the concentration rate approaches $0.5 \times 10^6 \text{ min}^{-1}$.

The data presented in Fig. 6 show that the FDEPSS achieved satisfactory collection efficiency and concentration rate when challenged in laboratory with three different airborne microorganisms. In the next step, the FDEPSS was tested against a BioSampler and a Button aerosol sampler when sampling outdoor microorganisms for 60 min, and the results are presented in Fig. 7. The total concentration of bioaerosol collected by each sampler is expressed as concentration of airborne ATP (RLU m⁻³). The ATP concentration determined by the FDEPSS was $(5.1 \pm 1.1) \times 10^5 RLU \text{ m}^{-3}$, while $(4.1 \pm 1.7) \times 10^5 RLU \text{ m}^{-3}$ was measured by the BioSampler and $(8.7 \pm 1.9) \times 10^5$ $RLU \text{ m}^{-3}$ was measured by the Button aerosol sampler (Fig. 7(a)). Since the Button aerosol sampler uses filter as its collection medium, we can assume its collection efficiency to be ~100%, and use it as a reference sampler (Burton et al., 2007). In this case, collection efficiency of the FDEPSS relative to the Button aerosol sampler was $59.1 \pm 2.5\%$ and that of the BioSampler was $46.1 \pm 13.6\%$. The difference between these two samplers was not statistically different (p = 0.065).

Another important outcome of the field experiment is applicability of ATP to measure total bioaerosol burden. The three used samplers (FDEPSS, BioSampler, and Button aerosol sampler) have different sampling flow rates (i.e., 20, 12.5, and 4 L min⁻¹), volumes of collection liquid (0.04,

5, and 5 mL) and relative collection efficiency as shown in Fig. 7(a). Since ATP was used as a metric to determine airborne microorganism concentration, one can estimate the sampling time needed by each sampler to exceed ATP quantification threshold in its collection liquid for different airborne ATP concentrations (Fig. 7(b)). A similar approach was used for optimal colony density in agar-plate-based impactors (Nevalainen et al., 1992). In the case presented in Fig. 7(b), sample analysis is based on 100 µL of collection liquid and the sample quantification threshold of 6,000 RLU/100 µL (3× RLU of sterile DI water background level) was used. Since FDEPSS samples are recovered in a 20 µL droplet in each chamber, the total combined sample volume is 40 µL. For sample analysis this volume would be increased to 100 µL, i.e., diluted by 2.5×. For Button sampler and BioSampler, 100 µL aliquot would be taken from the 5 mL elution volume and 5 mL collection liquid volume, respectively, i.e., only 1/50th of the sample is analyzed. The sampling time needed to exceed the sample quantification threshold is a function of the sampler concentration rate, R_C , and is inversely proportional to the sampling flow rate and directly proportional to the sample volume. Therefore, for any particular airborne ATP concentration, the FDEPSS needs a much shorter sampling time compared to other two samplers to accumulate sufficient amount of ATP in its collection fluid owing to its high sample concentration rate (Fig. 7(b)). For the airborne ATP concentration of $10^6 RLU m^{-3}$ (similar to the concentration observed in our experiments), the FDEPSS would need only 1.3 min to exceed the sample quantification threshold in its collection fluid. For the Button sampler and the BioSampler, 75 and 51 min would be needed, respectively,

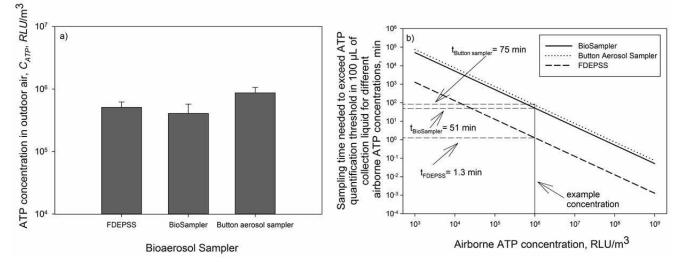


Fig. 7. (a) Average adenosine triphosphate (ATP) concentration (*RLU* m⁻³) outdoors determined by the field-deployable electrostatic precipitator with superhydrophobic surface (FDEPSS), BioSampler (SKC Inc., Eighty Four, PA), and Button aerosol sampler (SKC Inc.) when sampling for 60 min. The FDEPSS was operated at a flow rate of 20 L min⁻¹ with a –9 kV voltage; samples from each chamber were removed by 20 μ L droplets. The BioSampler was operated with 5 mL of collection fluid and at a sampling flow rate of 12.5 L min⁻¹. The Button aerosol sampler was operated at a flow rate of 4 L min⁻¹. The data are averages and standard deviations based on five repeats. (b) A comparison of sampling times needed by the three samplers to collect airborne ATP above quantification threshold in 100 μ L liquid as a function of airborne ATP concentration. Sampling conditions are the same as in Fig. 7(a).

assuming that their samples are collected or eluted in 5 mL of liquid. Thus, the FDEPSS can detect bioaerosol presence at least 40× faster than the other two samplers. The minimum time needed to quantify ATP by the FDEPSS could be decreased even further (by 2.5×), if one used 40 μ L for ATP analysis, instead of 100 μ L, which was selected for convenience. Thus, this estimate shows that the FDEPSS is a tool allowing a rapid detection of bioaerosol presence due to its high concentration rate. This feature would be especially useful in low concentration environments.

Since the main focus of this study was field investigation of FDEPSS with sample analysis by ATP, culturability of the collected bioaerosol was not investigated. Recovery of the culturable bioaerosol fraction by FDEPSS will be determined in the upcoming studies. In addition, the charging section of the FDEPSS will be re-designed to minimize ozone production.

The Button aerosol sampler features an omnidirectional sampling head which makes it less sensitive to wind direction and speed (Görner *et al.*, 2010). Therefore, variability in relative collection efficiency of samplers run side-by-side can offer estimate of their sensitivity to wind direction and speed. The coefficient of variation of the relative collection efficiency of the FDEPSS and the BioSampler were 21% and 41%, respectively. This suggests that the FDEPSS is less sensitive to wind direction and speed, most likely due to its omnidirectional inlet. The BioSampler, on the other hand, has a unidirectional inlet. The results for the wind sensitivity are based on a limited number of samples, and thus the apparent wind-sensitivity differences should be examined more thoroughly in the future.

CONCLUSIONS

The field-deployable electrostatic precipitator with superhydrophobic surface (FDEPSS) was developed based on the EPSS Mark II (Han et al., 2015) so that it could be operated as a stand-alone device in the field. The device showed satisfactory collection efficiency when challenged with laboratory-generated bioaerosols. When tested in an outdoor environment, the FDEPSS showed ability to rapidly detect airborne microorganisms using ATP-based detection method. Moreover, given its high concentration rate, the device allows detecting the presence of airborne microorganisms much quicker compared to the other two tested samplers (i.e., BioSampler and Button sampler). This feature indicates that the FDEPSS could be used to measure bioaerosols in low concentration environments; it could also serve as part of bioaerosol detection systems, where time is of the essence. These applications will be explored in further research.

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REFERENCES

- Agranovski, I.E., Agranovski, V., Reponen, T., Willeke, K. and Grinshpun, S.A. (2002). Development and Evaluation of a New Personal Sampler for Culturable Airborne Microorganisms. *Atmos. Environ.* 36: 889– 898, doi: 10.1016/s1352-2310(01)00488-5.
- An, H.R., Mainelis, G. and Yao, M. (2004). Evaluation of a High-volume Portable Bioaerosol Sampler in Laboratory and Field Environments. *Indoor Air* 14: 385–393, doi: 10.1111/j.1600-0668.2004.00257.x.
- Bellin, P. and Schillinger, J. (2001). Comparison of Field Performance of the Andersen N6 Single Stage and the SAS Sampler for Airborne Fungal Propagules. *Indoor Air* 11: 65–68, doi: 10.1034/j.1600-0668.2001.011001065.x.
- Boelter, K.J. and Davidson, J.H. (1997). Ozone Generation by Indoor, Electrostatic Air Cleaners. *Aerosol Sci. Technol.* 27: 689–708, doi: 10.1080/02786829708965505.
- Boinovich, L., Emelyanenko, A.M. and Pashinin, A.S. (2010). Analysis of Long-Term Durability of Superhydrophobic Properties under Continuous Contact with Water. *ACS Appl. Mater. Interfaces* 2: 1754–1758, doi: 10.1021/am100241s.
- Burton, N.C., Grinshpun, S.A. and Reponen, T. (2007). Physical Collection Efficiency of Filter Materials for Bacteria and Viruses. *Ann. Occup. Hyg.* 51: 143–151.
- Creamean, J.M., Suski, K.J., Rosenfeld, D., Cazorla, A., DeMott, P.J., Sullivan, R.C., White, A.B., Ralph, F.M., Minnis, P., Comstock, J.M., Tomlinson, J.M. and Prather, K.A. (2013). Dust and Biological Aerosols from the Sahara and Asia Influence Precipitation in the Western U.S. *Science* 339: 1572–1578.
- Curtis, L., Rea, W., Smith-Willis, P., Fenyves, E. and Pan, Y. (2006). Adverse Health Effects of Outdoor Air Pollutants. *Environ. Int.* 32: 815–830, doi: 10.1016/j.envint. 2006.03.012.
- Goheen, S.C., Larkin, E.C. and Bissell, M.G. (1984). Ozone Produced by Corona Discharge in the Presence of Water. *Int. J. Biometeorol.* 28: 157–161, doi: 10.1007/BF 02191728.
- Görner, P., Simon, X., Wrobel, R., Kauffer, E. and Witschger, O. (2010). Laboratory Study of Selected Personal Inhalable Aerosol Samplers. *Ann. Occup. Hyg.* 54: 165–187.
- Han, B., Hudda, N., Ning, Z., Kim, H.J., Kim, Y.J. and Sioutas, C. (2009). A Novel Bipolar Charger for Submicron Aerosol Particles Using Carbon Fiber Ionizers. *J. Aerosol Sci.* 40: 285–294, doi: 10.1016/j.jaerosci.200 8.11.005.
- Han, T., O'Neal, D.L., McFarland, A.R., Haglund, J. and Ortiz, C.A. (2005). Evaluation of Mixing Elements in an L-Shaped Configuration for Application to Single-Point Aerosol Sampling in Ducts. *HVAC&R Res.* 11: 657– 672, doi: 10.1080/10789669.2005.10391160.
- Han, T. and Mainelis, G. (2008). Design and Development of an Electrostatic Sampler for Bioaerosols with High Concentration Rate. J. Aerosol Sci. 39: 1066–1078, doi: 10.1016/j.jaerosci.2008.07.009.
- Han, T., An, H.R. and Mainelis, G. (2010). Performance of an Electrostatic Precipitator with Superhydrophobic

Surface when Collecting Airborne Bacteria. *Aerosol Sci. Technol.* 44: 339–348, doi: 10.1080/02786821003649352.

- Han, T., Nazarenko, Y., Lioy, P.J. and Mainelis, G. (2011). Collection Efficiencies of an Electrostatic Sampler with Superhydrophobic Surface for Fungal Bioaerosols. *Indoor Air* 21: 110–120, doi: 10.1111/j.1600-0668.2010.00685.x.
- Han, T., Fennell, D. and Mainelis, G. (2015). Development and Optimization of the Electrostatic Precipitator with Superhydrophobic Surface (EPSS) Mark II for Collection of Bioaerosols. *Aerosol Sci. Technol.* 49: 210–219, doi: 10.1080/02786826.2015.1017040.
- Hinds, W.C. (1999). *Aerosol Technology*, Wiley, New York.
- Hill, S.C., Pinnick, R.G., Niles, S., Pan, Y.L., Holler, S., Chang, R.K., Bottiger, J., Chen, B.T., Orr, C.S. and Feather, G. (1999). Real-time Measurement of Fluorescence Spectra from Single Airborne Biological Particles. *Field Anal. Chem. Technol.* 3: 221–239, doi: 10.1002/(SICI)1520-6521(1999)3:4/5<221::AID-FACT2>3.0.CO;2-7.
- Ho, Y.P. and Reddy, P.M. (2010). Identification of Pathogens by Mass Spectrometry. *Clin. Chem.* 56: 525–536.
- Ichinose, T., Nishikawa, M., Takano, H., Sera, N., Sadakane, K., Mori, I., Yanagisawa, R., Oda, T., Tamura, H., Hiyoshi, K., Quan, H., Tomura, S. and Shibamoto, T. (2005).
 Pulmonary Toxicity Induced by Intratracheal Instillation of Asian Yellow Dust (Kosa) in Mice. *Environ. Toxicol. Pharmacol.* 20: 48–56, doi: 10.1016/j.etap.2004.10.009.
- Johnson, B., Martin, D.D. and Resnick, I.G. (1994). Efficacy of Selected Respiratory Protective Equipment Challenged with Bacillus *subtilis subsp. niger. Appl. Environ. Microbiol.* 60: 2184–2186.
- Karl, D.M. (1980). Cellular Nucleotide Measurements and Applications in Microbial Ecology. *Microbiol. Rev.* 44: 739–796.
- Kenny, L.C., Bowry, A., Crook, B. and Stancliffe, J.D. (1999). Field Testing of A personal Size-selective Bioaerosol Sampler. Ann. Occup. Hyg. 43: 393–404.
- Kesavan, J. and Sagripanti, J.L. (2015). Evaluation Criteria for Bioaerosol Samplers. *Environ. Sci. Processes Impacts* 17: 638–645, doi: 10.1039/C4EM00510D.
- Kim, K.H., Jahan, S.A. and Kabir, E. (2013). A Review on Human Health Perspective of Air Pollution with Respect to Allergies and Asthma. *Environ. Int.* 59: 41–52, doi: 10.1016/j.envint.2013.05.007.
- Kulkarni, P., Namiki, N., Otani, Y. and Biswas, P. (2002). Charging of Particles in Unipolar Coronas Irradiated by in-situ soft X-rays: Enhancement of Capture Efficiency of Ultrafine Particles. J. Aerosol Sci. 33: 1279–1296, doi: 10.1016/S0021-8502(02)00067-8.
- Lunau, M., Lemke, A., Walther, K., Martens-Habbena, W. and Simon, M. (2005). An Improved Method for Counting Bacteria from Sediments and Turbid Environments by Epifluorescence Microscopy. *Environ. Microbiol.* 7: 961–968, doi: 10.1111/j.1462-2920.2005.00767.x.
- Macher, J.M. and Macher, J.M. (1997). Evaluation of Bioaerosol Sampler Performance. *Appl. Occup. Environ. Hyg.* 12: 730–736, doi: 10.1080/1047322X.1997.10387755.

- Madsen, A.M. and Sharma, A.K. (2008). Sampling of High Amounts of Bioaerosols Using a High-Volume Electrostatic Field Sampler. *Ann. Occup. Hyg.* 52: 167– 176.
- Maki, T., Puspitasari, F., Hara, K., Yamada, M., Kobayashi, F., Hasegawa, H. and Iwasaka, Y. (2014). Variations in the Structure of Airborne Bacterial Communities in a Downwind Area during an Asian Dust (Kosa) Event. *Sci. Total Environ.* 488–489: 75–84, doi: 10.1016/j. scitotenv.2014.04.044.
- Mandal, J. and Brandl, H. (2011). Bioaerosols in Indoor Environment - A Review with Special Reference to Residential and Occupational Locations. *Open Environ. Biol. Monit. J.* 4: 83–96.
- Mehta, S.K., Mishra, S.K. and Pierson, D.L. (1996). Evaluation of Three Portable Samplers for Monitoring Airborne Fungi. *Appl. Environ. Microbiol.* 62: 1835–1838.
- Monn, C. (2001). Exposure Assessment of Air Pollutants: a Review on Spatial Heterogeneity and Indoor/Outdoor/ Personal Exposure to Suspended Particulate Matter, Nitrogen Dioxide and Ozone. *Atmos. Environ.* 35: 1–32, doi: 10.1016/S1352-2310(00)00330-7.
- Nadkarni, M.A., Martin, F.E., Jacques, N.A. and Hunter, N. (2002). Determination of Bacterial Load by Real-time PCR Using a Broad-range (Universal) Probe and Primers Set. *Microbiology* 148: 257–266.
- Nevalainen, A., Pastuszka, J., Liebhaber, F. and Willeke, K. (1992). Performance of Bioaerosol Samplers: Collection Characteristics and Sampler Design Considerations. *Atmos. Environ.* 26: 531–540, doi: 10.1016/0960-1686(92)90166-I.
- Noble, R.T., Blackwood, A.D., Griffith, J.F., McGee, C.D. and Weisberg, S.B. (2010). Comparison of Rapid Quantitative PCR-Based and Conventional Culture-Based Methods for Enumeration of Enterococcus spp. and Escherichia coli in Recreational Waters. *Appl. Environ. Microbiol.* 76: 7437–7443.
- Prussin, A.J., Garcia, E.B. and Marr, L.C. (2015). Total Concentrations of Virus and Bacteria in Indoor and Outdoor Air. *Environ. Sci. Technol. Lett.* 2: 84–88, doi: 10.1021/acs.estlett.5b00050.
- Roux, J.M., Kaspari, O., Heinrich, R., Hanschmann, N. and Grunow, R. (2013). Investigation of a New Electrostatic Sampler for Concentrating Biological and Non-Biological Aerosol Particles. *Aerosol Sci. Technol.* 47: 463–471, doi: 10.1080/02786826.2013.763896.
- Seshadri, S., Han, T., Krumins, V., Fennell, D.E. and Mainelis, G. (2009). Application of ATP Bioluminescence Method to Characterize Performance of Bioaerosol Sampling Devices. J. Aerosol Sci. 40: 113–121, doi: 10.1016/j.jaerosci.2008.10.002.
- Veal, D.A., Deere, D., Ferrari, B., Piper, J. and Attfield, P.V. (2000). Fluorescence Staining and Flow Cytometry for Monitoring Microbial Cells. *J. Immunol. Methods* 243: 191–210, doi: 10.1016/S0022-1759(00)00234-9.
- Venkateswaran, K., Hattori, N., La Duc, M.T. and Kern, R. (2003). ATP As a Biomarker of Viable Microorganisms in Clean-room Facilities. J. Immunol. Methods 52: 367– 377, doi: 10.1016/S0167-7012(02)00192-6.

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- Wang, Z., Reponen, T.A., Grinshpun, S.L., Górny, R. and Willeke, K. (2001). Effect of Sampling Time and Air Humidity on the Bioefficiency of Filter Samplers for Bioaerosol Collection. J. Aerosol Sci. 32: 661–674, doi: 10.1016/S0021-8502(00)00108-7.
- Yassin, M.F. and Almouqatea, S. (2010). Assessment of Airborne Bacteria and Fungi in an Indoor and Outdoor Environment. *Int. J. Environ. Sci. Technol.* 7: 535–544, doi: 10.1007/BF03326162.
- Zhen, H., Han, T., Fennell, D. E. and Mainelis, G. (2013). Release of Free DNA by Membrane-Impaired Bacterial Aerosols Due to Aerosolization and Air Sampling. *Appl. Environ. Microbiol.* 79: 7780–7789.

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