**ORIGINAL PAPER**

# Measurement and Simulation of Biocontamination in an Enclosed Habitat

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**Abstract**
Microbial aerosols can be used as model particles for examining the dispersion and deposition of particles as well as assessing the reliability of the simulation methods. For example, the computational fluid dynamics model (CFD) can be used in the evaluation of indoor microbial contamination and the possible spread of harmful microbes in spaces with high densities of people or in special hermetic environments. The aim of this study was to compare the results of the CFD simulation, which predicts the deposition of biological particles on the surfaces of a spacecraft, and real particle deposition, using *Bacillus licheniformis/aerius* bacterium particles as the model organism. The results showed that the particles were mainly deposited on floor surfaces, but also onto the supply air diffusers, where bacterial concentrations were higher than on the wall and ceiling surfaces. The CFD simulation showed similar trends with actual particle dispersal, conducted in this experiment with *Bacillus* particles.

## 1 Introduction

Bioaerosols such as bacteria, fungi, viruses and all other particles that contain living organisms or which can be released from living organisms are recognized as important negative contributors to the indoor air quality (Reponen et al. 2011). Indoors, bioaerosols can originate from humans, pets, plants or they can enter from the outdoor air. Humans are major

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sources of bacteria and viruses indoors, while moulds mostly originate from human activities such as handling of root vegetable and firewood (Reponen et al. 1992; Nazaroff 2004; National Research Council 2005). In addition, contaminated clothes can act as a source of fungal spores.

Crew members are a major source of microorganisms on spacecraft and station and, although most of the microbes released are generally harmless, there may also be some opportunistic pathogens (Pierson 2001; Checinska et al. 2015; Ichijo et al. 2016). The bioaerosols emitted by the crew members originate mainly from the respiratory tract and skin, although emissions from the gastrointestinal tract in some off-nominal events are also possible (National Research Council 2005). Microorganisms are ubiquitous ecological partners of humans, materials and devices also during manned space flight and in other hermetically sealed environments. Microbes can be transmitted and enter into the human body naturally by inhalation, by direct contact between persons or through indirect contact, e.g. with environmental surfaces or by ingestion through contaminated food and water (Siegel et al. 2007). Furthermore, airborne biocontamination is considered especially important under spaceflight conditions, because microgravity means that the bioaerosol dispersion will be very different from that on

Earth. It is also known that an enclosed environment (such as in ISS space station) can produce resistant microbial and more virulent strains (Wilson et al. 2007, 2008; Parra et al. 2008; Zea et al. 2017). During the envisioned future long-duration flights, it will be mandatory to prevent and control bioaerosol contamination, and this will demand a better understanding about the contamination mechanisms ranging from potential sources to actual deposition of the aerosolized particle.

In a space station, most of the bioaerosols are exhausted and subsequently removed by the high efficiency particulate air (HEPA) filters installed in the recirculation air (Van Houdt et al. 2012; Checinska et al. 2015). The HEPA filters used in a space station retain > 99.97% particles of size 0.3 µm and larger. However, a small fraction of the airborne microbes will still become deposited on surfaces. If one considers microbial contamination deposition under normal gravity, then large particles carrying bacteria settle rapidly and are thus removed from the air onto solid surfaces. Under microgravity conditions, the settling of larger particles is less significant and other removal mechanisms become more dominant. According to Van Houdt et al. (2018), continuous ventilation and efficient particle filtration will remove the majority of these airborne bioaerosols, but imperfect mixing of air may result in areas where there is a risk of unwanted deposition onto surfaces.

Particle deposition on surfaces can be studied using generated bacterial and fungal aerosols. These bacterial and fungal aerosols can be exploited also as model particles when examining the dispersion and deposition of particles as well as when assessing the reliability of the models. The models could be applied to evaluate microbial contamination and the possible spread of harmful microbes in crowded areas, such as transport hubs. Another example would be to determine the spread of pathogens in airports or in special hermetic environments such as submarines or a space station.

CFD models have been widely used to study indoor air quality, thermal comfort, fire safety, particle concentration levels and HVAC system performance (Chen 2009). In addition to particulate matter, CFD has also been used to monitor indoor air quality which has been deteriorated by gaseous contaminants (Chen 2009). Typical environments for these studies have included various buildings (commercial buildings, residential buildings, schools, health-care facilities, institutional buildings, and industrial buildings), underground facilities, public transportation vehicles, greenhouses, animal facilities, etc. Particle deposition has been modelled by many researchers. The deposition of particles on indoor smooth surfaces has been investigated as a function of particle size and density (Lai and Nazaroff 2000; Lai and Chen 2006; Zhao et al. 2008; Zhang and Chen 2009). Also, the behaviour of bioaerosols can be simulated with computational fluid dynamics (CFD) which offers detailed predictions about airflows and particle transport in mechanically ventilated enclosed spaces (Zhao et al. 2004). Advances in computational fluid dynamics (CFD) as well as the underlying algorithms and the developments in computational power have made it possible to better resolve indoor air movements and transport of contaminants. The simulations presented in this article are expected to be performed in computational cluster environments, mainly currently existing in research institutions. The simulations have focused on studying airflow patterns and contaminant concentration dispersion in the air. In addition, by modifying the gravity force in the CFD simulation, the method can be exploited also to predict the deposition of bioaerosols on surfaces in a microgravity environment. However, a suitable Earth analogue of the space ecosystem is not available and the comparison of the simulation results with the experimental results is currently feasible only under normal gravity conditions.

The objective of this study was to develop and compare a model to predict microbial contamination in an enclosed environment using *Bacillus aerius/licheniformis* bacteria isolated from the ISS: the transportation of bioaerosols and the concurrent spread of biocontamination. A more elaborate aim was to produce a robust modelling tool for predicting airborne microbial contamination transport in manned spacecraft and vehicles, since this can help to develop adequate control programmes and countermeasures.

## 2 Materials and Methods

Experiments were conducted in an environmentally controlled and a hermetically sealed 26.4 m$^3$ chamber (Hydrosphere Habitat (HH) of COMEX, FR), which was divided into three distinct compartments: D = displacement ventilation zone, M = mixing ventilation zone and E = exhaust ventilation zone (Fig. 1).

In zone D, a low-velocity air diffuser was installed to make it possible to study a displacement ventilation configuration. Zone M hosted four separate air diffusers to simulate a mixing ventilation configuration similar to the current one onboard the Columbus module in the International Space Station (ISS). Columbus is a research laboratory module; it is a part of the International Space Station and is the largest single contribution made by the European Space Agency (ESA) to the ISS. Zone E did not contain an air supply system, but instead there was a ventilation system exhausting the air supplied by the displacement and mixing ventilation as transfer air.

The ventilation system of chamber was an HEPA-powered (high efficiency particulate air filter) filtration unit (Oy Lifa Air Ltd, FIN) that produces a sufficient HEPA-filtered airflow. The ventilation was arranged with the closed circulation principle in which the exhaust air was led to a filtration

**Fig. 1** Test volume CAD view describing the three air distribution zones and dimensions in the research chamber hydrosphere habitat
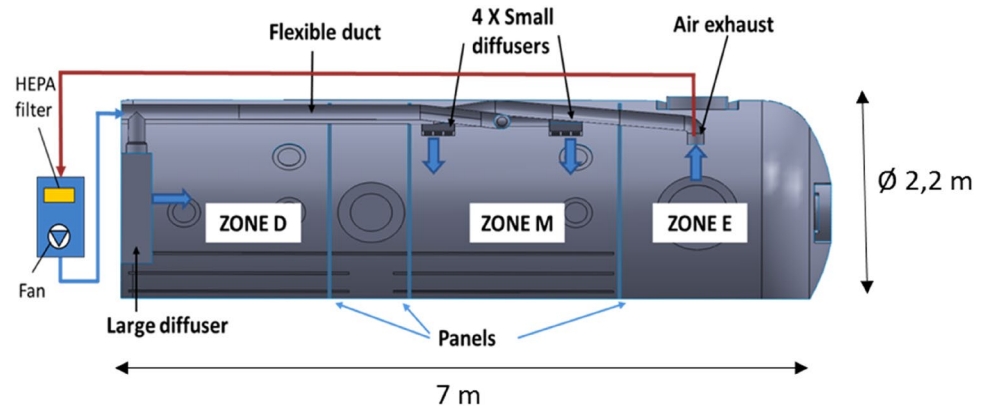


unit and supplied back to the air distribution system. The instrumentation of the airflow and microbial measurements systems has also been incorporated into the chamber and coupled with environmental measurement devices. Airflow rate and air velocities in the empty chamber were measured using a hot wire comfort anemometer, i.e. the omnidirectional thermoanemometer (TSI model 8475-075-1). Temperature and relative humidity were measured (HM70, Vaisala, FIN) also before and after each test period. Inlet air temperature and relative humidity in the chamber varied during the different test days from $20 \pm 4$ °C and from $70 \pm 10\%$, respectively. The corresponding values for outdoor air were $17.5 \pm 2.5$ °C and $65 \pm 25\%$, respectively.

Before the microbial experimental campaign, pre-tests with non-biological diethylhexyl sebacate (DESH) particles and airflow measurements were performed to adjust the air distribution system as well as allowing calibration of the airflow pattern in the model. The concentration of the used DESH particles were much smaller than bioaerosol concentration in experiments, but because particle–particle interactions were not taken into account, the relative results are comparable. A measurement ring and an adjustment

damper were installed upstream of each diffuser to measure and adjust the airflow rate. The flow rates were balanced so that the total supply flow rate of the displacement ventilation was 150 m³/h. The supply flow rate of the mixing ventilation chamber was 160 m³/h, divided equally with the four supply devices, giving 40 m³/h for each. The total airflow rate was thus 310 m³/h. Figure 2 presents the mean velocities in selected planes of interest. Some unsteady non-convergent structures can be seen, but they are mainly due to logarithmic scale used in the figure; deviations from the smooth flow field are insignificant.

Computational fluid dynamics is determined by solving of the governing equations of the fluid flow; in the case of the incompressible flow, these are the continuity equation and the momentum equation, i.e. Navier–Stokes equations. Equations are presented, e.g. in Ferziger et al. (2020). Together with the boundary conditions and material properties, these equations are all that are needed to obtain time-dependent fluid flow field in every spatial location of the domain. The method based on solving these equations without any simplifications is called a direct numerical simulation (DNS). The commonly used CFD methods are based

**Fig. 2** Mean velocity field in selected planes. Note: logarithmic



U (m/s)

0.00  0.00  0.00 0.01 0.02  0.05 0.10 0.20  0.50 1.00 2.00  8.00
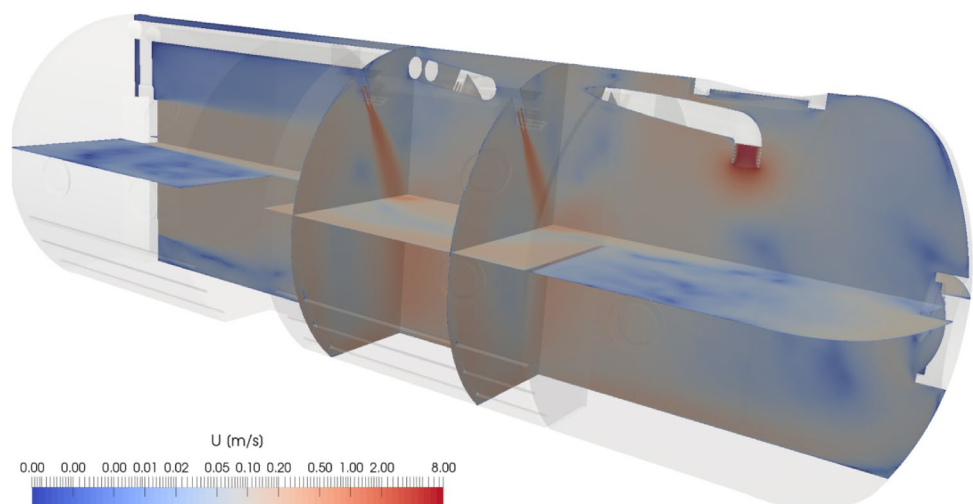
on the domain and equation discretization and the solving of the equations by marching in a time stepwise manner. Apart from rare case of laminar flow, DNS requires a very dense computational grid and small time steps and is, therefore, computationally too expensive to be used as a part of a routine simulation process. The traditional procedure applied to overcome this problem is to conduct time averaging of the governing equations yielding the Reynolds averaged Navier–Stokes (RANS) equations and to assume that there is a time-independent steady-state solution. The nonlinear nature of the momentum equation generates an additional term to the time-averaged equation, which needs to be modelled using some turbulence closure, for example, the well-known $k$–$\epsilon$ model (Launder & Spalding 1974). Turbulence models are always empirical models and require fine-tuning of the parameters in the models. This means that the models usually give good results only for the cases in which they are meant to be used and therefore there is no universal model that works well in every case. The method applied in this study can be considered to lie between the DNS and RANS, both with respect to computational demands and accuracy. The method is called a large eddy simulation (LES). In LES, the case is simulated as an unsteady case, but in contrast to DNS, all the turbulence scales are not simulated but the smallest one is modelled using a sub-grid scale model. This substantially reduces the needed computational power as compared to DNS. In this study, a WALE (wall-adapting local eddy viscosity) model was used for the sub-grid model (Nicoud and Ducros 1999).

The size and quality of the computational grid affect the results as well as influencing the convergence of the simulations. In general, a higher density of grid points is needed where gradients of the simulated variables are large. Special caution is needed when modelling near surfaces. The geometry of the simulated domain should naturally be as close as possible to the real one. The calculation domain was divided into discrete cells so that the grid density was higher in locations where high mean velocity gradients were expected such as in the vicinity of surfaces and supply outlets. The computational grid had about 81 million cells. The typical size of the grid cell in the core region of the domain was 2 cm. We applied a threefold refinement near the wall grids, yielding a grid size of 2.5 mm. Subsequently, the nearest wall cell was divided into three cells using a thickness ratio of 2. The typical distance from the centre point of the wall adjacent cell to the wall of different surfaces of the domain was about 0.35 mm. The computational geometry was devised using open source software, Salome-platform (www.salome-platform.org). A computational grid was produced using utility of the open source CFD package Open-FOAM (www.openfoam.org).

The actual simulation was performed using an LES WALE turbulence model implemented in the open source software OpenFOAM. The case was initially simulated for 300 s to depict the situation where the statistics of the flow field are no longer varying. After that, the flow field was simulated for 180 s and time average field over that period was used in the particle simulation stage. A simulation time using 400 CPU cores of modern Linux cluster was about 3 months in the air velocity flow field simulation phase, whereas the particle simulation required only days using one CPU core.

Particle simulation was performed using time-averaged gas velocity field and the so-called Lagrangian method in which trajectories of the particles are calculated taking into account the forces influencing the particles. Since the influence of turbulence is lost in the time-averaging process, it should be included by incorporating turbulence quantities into the particle tracking process. Traditionally, this is done by modifying the trajectory of the particle using turbulence quantities obtained directly from the turbulence model being used. In a large eddy simulation, however, only part of the turbulence is modelled using a turbulence model and the rest of the turbulence is simulated as a part of the unsteady simulation procedure. In this work, this simulated part of the turbulence is taken into account by calculating the averages of the fluctuating velocity components in the unsteady simulation stage. Hence, both modelled and simulated turbulence of the gas flow field are used in the particle trajectory modification procedure, in which the fluctuation of the particle trajectories is much more vigorous than in the case in which only modelled turbulence is considered.

The microbial experimental campaign was carried out with a wet generated *Bacillus* bacteria bioaerosol. Briefly according Salmela et al. (2017), *Bacillus* suspension was prepared as follows: *B. aerius/licheniformis* was inoculated with loop from stock culture on trypticase soy agar (TSA, Sigma-Aldrich Co., St. Luis, MO, US) and grown for $24 \pm 2$ h at 37 °C. After incubation, bacterial colonies were harvested with an inoculation loop, inoculated in potato agar (PA, Thermo Fisher Scientific–Oxoid Limited, UK) and incubated for $48 \pm 2$ h at 37 °C. After the $48 \pm 2$ h incubation of the PA plates, the incubation was continued at room temperature ($21 \pm 1$ °C) under natural light for 5 days until the amount of spores was > 80%. The number of spores was pre-checked with Schaeffer–Fulton staining. After incubation, the *B. aerius/licheniformis* suspension was prepared by applying 5 ml of sterile deionized water onto the sporulating bacterial culture growing in PA plates. Bacterial spores were suspended into water by swapping the culture with a sterile inoculation loop and decanting the suspension into a test tube ($V = 50$ mL) with the suspension being mixed well. The relative amount of spores in suspension was determined with Schaeffer–Fulton staining. A total of 400 spores and vegetative cells were counted under oil immersion in a light microscope at 1000× magnification and the relative amount

of spores (> 80% of all) was calculated. The concentration of suspension was adjusted to $10^8$ microbes/mL by using a hemacytometer (e.g. Fuchs–Rosenthal: Hirschmann EM Techcolor) (Salmela et al. 2017). A diluted *Bacillus* suspension was generated by using 6-jet Collison Nebulizer (BGI, MA, US) at airflow 6 L/min. Compressed oil and particle-free air were used to generate the aerosol from the bacterial suspension solution. The suspension load was 25 mL and it produced stable aerosols for 2 h. The tests were conducted in triplicate.

From the air of the chamber, both the stability of bioaerosol generation and the total particle concentration were measured with an optical particle counter (OPS 3330, TSI, MN, US). Particle detection is based on light scattering; when a particle passes through a beam of light, some of the light is scattered and thus the number of particles is determined by counting the pulses of scattered light reaching the detector (Burkart et al. 2010, Salmela 2018). OPC counts all the particles in air in a predetermined size range (e.g. 0.3–10 μm). The deposition of bacteria cells and spores was studied at 34 different sampling points (Fig. 3) with contact agar (sampling area = 20 cm²) (Petrifilm AC, 3 M, US) and spread plate (sampling area = 25 cm²) (Quantiswab, Copan, IT) methods on the surface of the sampling sheets (glass material). In the spread plate method, dilution series were done up to $10^{-1}$ and two dilutions of each sample were inoculated onto TSA plates with 0.1 ml per dilution each conducted in duplicate. The detection limit was 0.2 cfu/cm² and 1 cfu/20 cm² for the spread plate method and the contact agar method, respectively. TSA plates and contact agars were incubated at 37 °C for 3 days. The agglomeration stage and the possible presence of other particles in the generated bioaerosol were analysed by using filter sampling. The generated bioaerosols were collected from the air into polycarbonate filters (MB PCB ⌀25 mm, 0.2 μm, Merck KGaA, DE)

with airflow of 1.8 ± 0.2 L/min. The SEM slides were prepared by coating the filters with gold and then visualized in a high-vacuum scanning electron microscope (SEM) (Mensah-Attipoe et al. 2016). From SEM images, the agglomerate stage was evaluated by counting either 40 fields or 400 endospores and cells, whichever came first (Paerl 1978).

Decontamination of surfaces and air was undertaken before the first microbe generation and after every bioaerosol generation. Decontamination was achieved by wiping the glass sampling sheets with 6% hydrogen peroxide solution, followed by hydrogen vapour treatment (generated from 35% aqueous $H_2O_2$ solution). Control samples were collected onto the surfaces after each decontamination processes with contact agars. In addition, air samples using Andersen impactors (sampling time 10 min, 28.3 L/min, LOD 3.5 cfu/m³) (Andersen 1958) were taken after the final decontamination. The decontamination results of contact agars were less than 0.5 cfu/cm², which was below the set trigger value for contamination, i.e. 50 cfu/100 cm²: the criteria are between grades B and C in the EU Guidelines to Good Manufacturing Practice of Sterile Medicinal Products (GMP) (EudraLex 2018). The microbial air concentration after the final decontamination as measured by Andersen impactors was < LOD in compartments D, M and E.

## 3 Results and Discussion

Optical particle counter (OPC) measurements revealed that the production of the bioaerosol was stable for 2 h, which was a sufficient time to allow us to conduct deposition measurements (Fig. 4). The particles were mostly in the size range of 0.5–3 μm, which is similar to the size of *Bacillus licheniformis/aerius* spores 1.2 μm length and 0.8 μm breadth, respectively (Carrera et al. 2007; Rey et al.
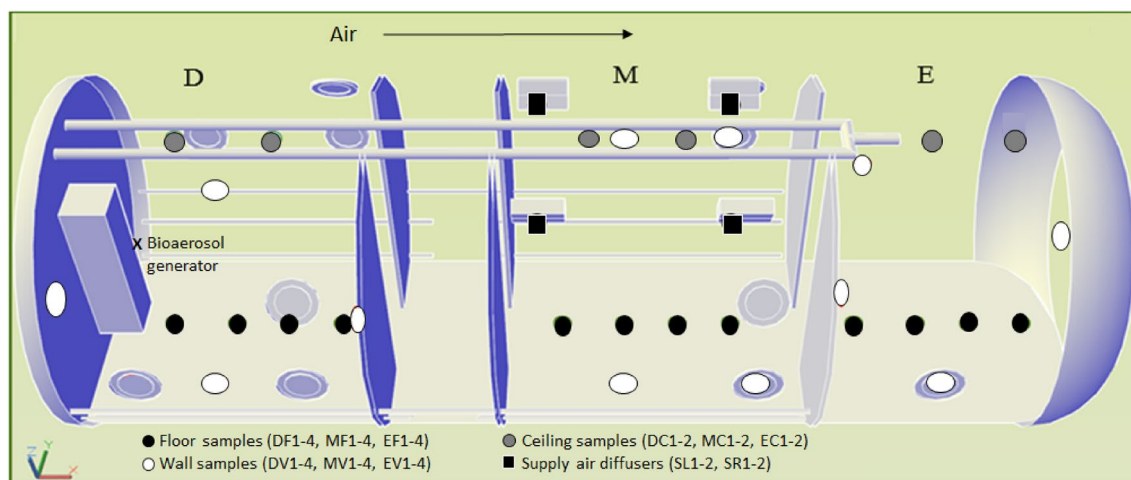


**Fig. 3** Surface sampling points; floors, ceilings, walls and supply air diffusers, in COMEX' Test chamber
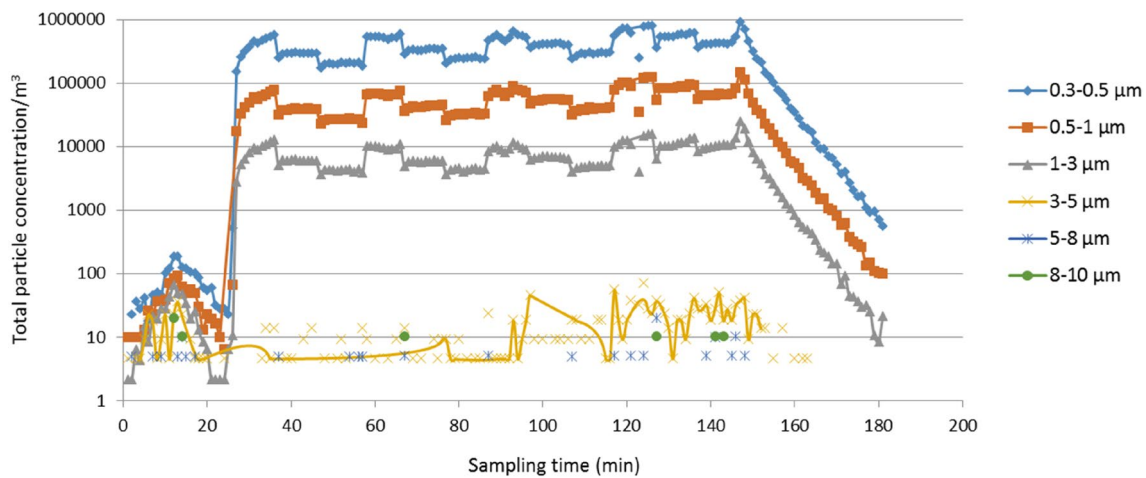
**Fig. 4** Particle concentration during 2 h bioaerosol generation with Collision Nebulizer starting at time point 20 min

2004; Chada et al. 2003). The total particle concentration was on average 63,000 particle/m$^3$ and 9000 particle/m$^3$ in size class 0.5–1 μm and 1–3 μm during 2 h wet generation from 10$^8$ cfu/ml suspension with a Collision Nebulizer, respectively. Changes in the stability were followed in all compartments with OPC by successive samples from D, M and E zones. However, changes between different compartments were rather small.

Figure 5 shows the averages of culturable concentration of deposited particles on the sampling points of the three repetitions measured with the spread plate method and with contact agar. Culturable surface concentrations analysed by the spread plate method varied from < LOD to 110 cfu/cm$^2$ and in contact agar samples from < LOD to 44 cfu/cm$^2$. The results showed that particles became deposited mostly on the

floors, at sampling points DF1–4, MF1–4 and EF1–4, where deposition concentrations ranged from 25 to 110 cfu/cm$^2$ and from 20 to 44 cfu/cm$^2$ in TSA plate samples and in contact agars, respectively. More than 90% of viable particles were deposited onto floor surfaces. In addition, sampling points on the wall, points DV1–4, were also emphasized. The deposition concentrations on the walls varied from < LOD to 6 cfu/cm$^2$ and < LOD–9 cfu/cm$^2$ by the spread plate and contact agar methods, respectively. There was only minor deposition on ceiling surfaces (< LOD to 2 cfu/cm$^2$). In contrast, there were also high deposition concentrations detected on the supply air diffusers (SL1–2, SR1–2) (4–18 cfu/cm$^2$). Due to their small surface area, only swab sampling with the spread plate method was done in the supply air diffusers. The highest culturable concentrations were observed in zone D.
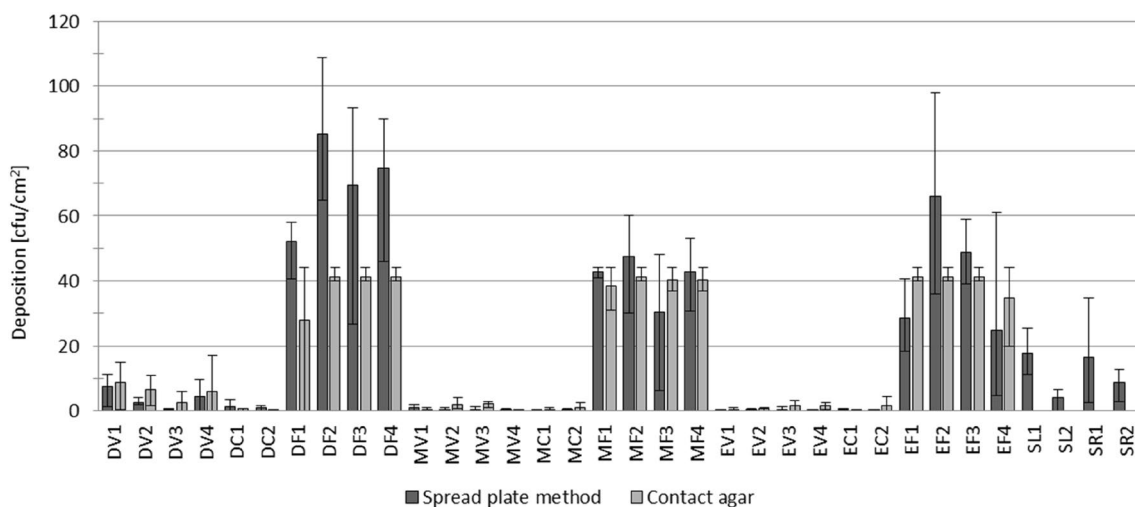


**Fig. 5** Averages and ranges of culturable concentrations of surface densities of cultivation *Bacillus* aerosols (*n* = 3) with swab and contact agar (*n* = 3) samples from all sampling points

However, no significant differences were detected between the compartments.

In general, the deposition of biological particles was highest on upward-facing surfaces. Gravitational settling is likely to have primarily resulted on deposition onto floors, whereas turbulent and Brownian diffusions were dominant processes when particles were deposited on walls and ceilings (Lai 2002; Kohli and Mittal 2009). Other external factors which can affect particle deposition were not studied. It is known that in addition to particle size, the deposition of bioaerosols onto surfaces depends also on the characteristics of the surface such as moisture, hydrophilic interactions, electrostatic forces and physical configuration, i.e. smooth, rough, porous or irregular surface (Tamburini et al. 2015). Also temperature may have an effect (thermophoretic force) (Han et al. 2011). Moreover, air turbulences and physical disturbances play a role in whether a particle will adhere or be re-aerosolized. However, the size of a bioaerosol is the most important parameter that determines its deposition properties. Bioaerosols with a size less than 5.0 µm generally remain in the air, and thus they will follow the air flow that carries them, whereas larger particles (> 10 µm) are soon deposited on surfaces (Mohr 2001; Ghosh et al. 2015).

The bioaerosols selected in this study, *Bacillus licheniformis* cells and endospores, were of a size less than 2 µm and their movements were influenced by the airflow pattern (Lu et al 1996; Carrera et al. 2007; Hurst et al. 2007; Wong et al. 2010) and that is the reason why more particles were in air than deposited on surfaces.

The comparison between the two different surface sampling methods revealed that one obtains similar results with the cultivation method and Petrifilm contact agar methods. The manufacturer of Quantiswabs (Biomerieux S.A.) has claimed that their nylon flocked swab used in this study should be able to recover 60% of microorganisms from a surface, whereas traditional fibre wound swabs achieve only

around 20% recovery (Dalmaso et al. 2007). The Petrifilm manufacturer has not provided comparable information about sampling efficiency, but several studies (e.g. Salo et al. 2000, Nelson et al. 2013, Warren et al. 2015) have compared the Petrifilm contact agar method to the conventional spread plate method and the data have indicated that the Petrifilm method is as sensitive as the conventional spread plate method. In addition, the Petrifilm AC method is simpler and less time-consuming than the conventional spread plate method as noted also by Kudaka et al. (2010).

According to use of the cultivation method for determination of deposited particles, a well-known limitation of cultivation is that it detects only a small portion (< 1–10%) of the microorganisms present in different environmental samples and it has a short sampling period (Amann et al. 1995; Nazaroff 2004). Heidelberg et al. (1997) and Torsvik et al. (1994) have found that only a very small proportion, i.e. 10%, of the microorganisms present in the environment could be cultured and identified and less than 1% of total microbial species in any environmental sample could be analysed by cultivation (Hugenholtz 2002). According to Oliver (2005), the other microorganisms were naturally viable but were non-culturable in laboratory conditions remaining in the "viable but non-culturable" stage.

The filter samples collected from the air of the chamber showed that the *B. aerius/licheniformis* spores were released mostly as single spores, which were sized < 2 µm, during the wet generation (Fig. 6). The image analysis detected also some agglomerates, but these made up less than 20% of the total counts and there were also particles of smaller sizes, < 0.5 µm. The number of particles under 0.5 µm was approximately ten times more than particles of spore size. These particles may be tissue fragments such as cell walls or cytoplasmic material (Terzieva et al. 1996; Reponen et al. 1997; Després et al. 2012). An explanatory factor may be an aerosolization stress. The aerosolization stress evoked
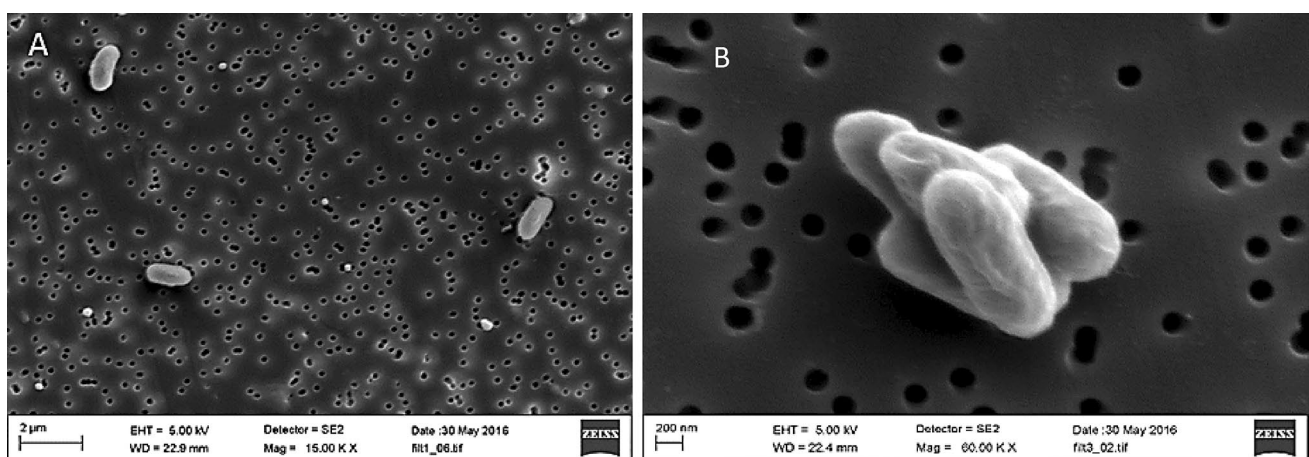


**Fig. 6** Spores and nanoparticles (**a**) and agglomerate (**b**) (the size of the agglomerate $1.2 \times 1.8$ mm), on the filter

by the Collison Nebulizer may induce structural changes in cells as well as cumulative metabolic injuries in the bacteria and thus increase the fragment concentration emitted from the Collison Nebulizer (Mainelis et al. 2005; Ibrahim et al. 2015). Furthermore, this process may affect the survivability of microorganisms, which is shown to depend on growth conditions prior to aerosolization, environmental conditions during aerosolization, methods of aerosolization and methods of collection and enumeration (Marthi et al. 1990). Nevertheless, survivability can also depend on the species of microorganism being aerosolized. However, spore forming *Bacillus* should be stress-resistant bacteria (Marthi et al. 1990, Griffits et al. 1996, Reponen et al. 1997, Thorne et al. 1992, Johansson et al. 2011, Ibrahim et al. 2015).
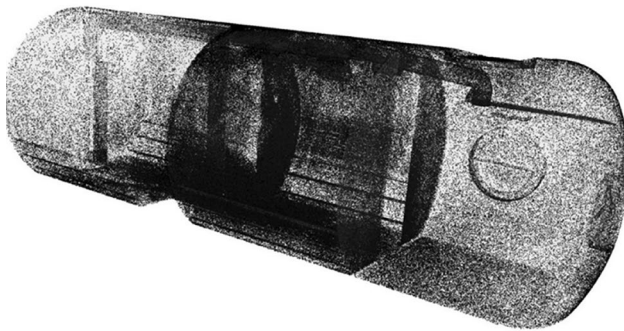


**Fig. 7** Predicted particle deposition (black points) after 30 min simulation onto indoor surfaces in the habitat

In general, the calculated deposition of particles was highest on upward-facing surfaces (floor), which is similar to the measurements (Figs. 5, 7 and 8). The highest deposition rates were predicted to be in zone M, whereas the experimental study resulted in more equal deposition rates to the floor of all compartments. The calculated deposition on vertical surfaces was in general clearly lower than that on upward-facing surfaces, which was also confirmed by measurements. However, in contrast to the experiments, there were quite high-predicted deposition rates on the ceiling in the mixing ventilation and exhaust zones. Overall, the experimental results were mostly less than ten times higher than predicted deposition. However, there was also up to 200 times higher experimental deposition than calculated results (mixing ventilation zone). Although the deposition rates were not predicted exactly, the calculated trends follow clearly the measured ones. There are also uncertainties in the measured values because of the indoor air microbe concentration as a result of adjusted *Bacillus* suspension, and also because varying surface sampling efficiency.

## 4 Conclusions

The development of a reliable model to predict contaminant dispersion and deposition in an enclosed space requires considerable effort. It requires the development of the model itself and then high-quality experimentation to validate the results. The experimentation had several phases which had to
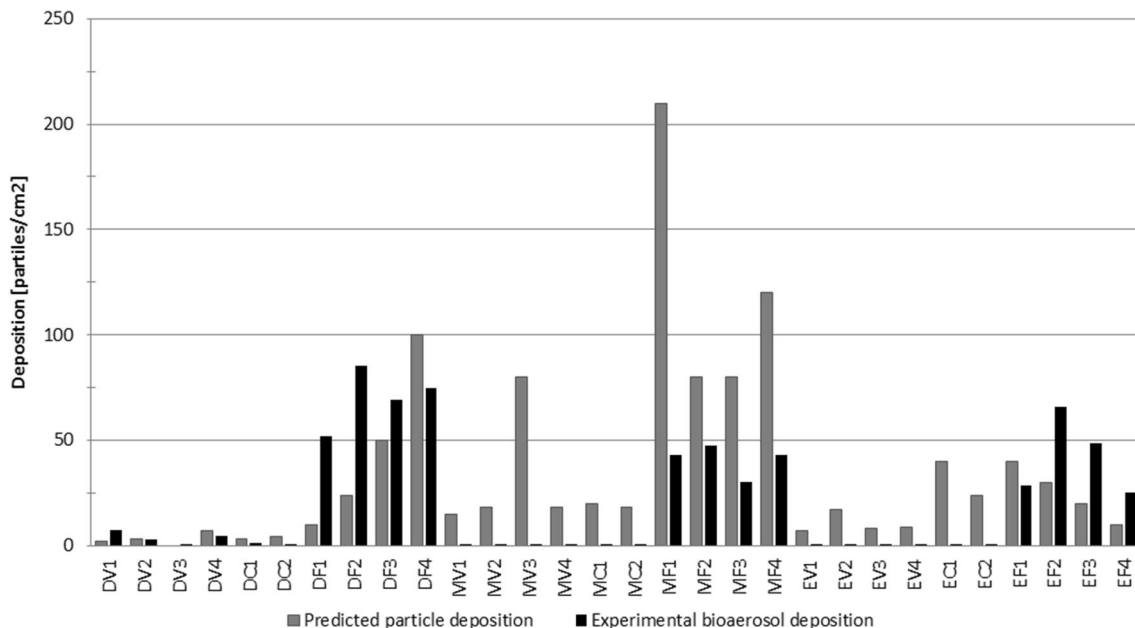


**Fig. 8** Predicted particle deposition after 30 min simulation onto indoor surfaces in the habitat as compared to experimental bioaerosol deposition (spread plate method) (particle/cm$^2$)

be properly performed from the bioaerosol generation, sampling to handling, cultivation and counting of the samples. Careful actions and efficient decontamination of the measurement environment between experiments were needed to avoid contamination which could ruin the experiments.

In conclusion, the results from this study proved that a *Bacillus* aerosol is a useful material when one wishes to assess bioaerosol deposition; the method produced results that were comparable with those obtained by mathematical modelling. However, the dispersion of generated particles was not very well predicted, perhaps because the aerosol generator was placed in the region where the flow field was not properly predicted. However, the predicted deposition rates showed similar trends with experimentation.

In spite of these results, the CFD model can be still used to predict the aerosol particle dispersion and deposition in ventilated enclosed multi-zone spaces. Computational fluid dynamics model can provide detailed information about the airflow and particle movements and deposition in microgravity spacecraft environment, information that would not otherwise be possible to collect under Earth conditions.

This work increases our knowledge about indoor bioaerosol transport and deposition; this will be useful in design, construction, operation and maintenance operations. In addition to spaceflight applications, it may be possible to exploit these results in situations where the spread of airborne pathogens is of interest, e.g. hospitals, operating theatres, airborne infection isolation rooms, pharmaceutical industry, clean rooms, airplanes and other mass transport vehicles.

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