

## **The removal of airborne SARS-CoV-2 and other microbial bioaerosols by air filtration on COVID-19 surge units**

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### **Running head**

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### **Competing interests**

Vilas Navapurkar is the founder, Director, and shareholder of Cambridge Infection Diagnostics Ltd. Andrew Conway-Morris, Paul White, Gordon Dougan and Stephen Baker are members of the Scientific Advisory Board of Cambridge Infection Diagnostics Ltd. Theodore Gouliouris has received a research grant from Shionogi. R Andres Floto has received research grants and/or consultancy payments from GSK, AZ, Chiesi, Shionogi, Insmad, Thirty Technology. Effrossyni Gkrania-Klotsas has received a National Institute of Health Research Greenshoots Award

### **Author contributions**

ACM conceptualisation, methodology, data analysis, writing-original draft

KS investigation, supervision, writing-review and editing

RB investigation, supervision, writing-review and editing

LK investigation, data analysis, supervision, writing-review and editing

MM investigation, writing-review and editing

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AK investigation, writing-review and editing

AT investigation, writing-review and editing

PW conceptualisation, provision of resources, writing-review and editing

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## **Summary**

### *Background*

The COVID-19 pandemic has overwhelmed the respiratory isolation capacity in hospitals; many wards lacking high-frequency air changes have been repurposed for managing patients infected with SARS-CoV-2 requiring either standard or intensive care. Hospital-acquired COVID-19 is a recognised problem amongst both patients and staff, with growing evidence for the relevance of airborne transmission. This study examined the effect of air filtration and ultra-violet (UV) light sterilisation on detectable airborne SARS-CoV-2 and other microbial bioaerosols.

### *Methods*

We conducted a crossover study of portable air filtration and sterilisation devices in a repurposed ‘surge’ COVID ward and ‘surge’ ICU. National Institute for Occupational Safety and Health (NIOSH) cyclonic aerosol samplers and PCR assays were used to detect the presence of airborne SARS-CoV-2 and other microbial bioaerosol with and without air/UV filtration.

### *Results*

Airborne SARS-CoV-2 was detected in the ward on all five days before activation of air/UV filtration, but on none of the five days when the air/UV filter was operational; SARS-CoV-2 was again detected on four out of five days when the filter was off. Airborne SARS-CoV-2 was infrequently detected in the ICU. Filtration significantly reduced the burden of other microbial bioaerosols in both the ward (48 pathogens detected before filtration, two after,  $p=0.05$ ) and the ICU (45 pathogens detected before filtration, five after  $p=0.05$ ).

### *Conclusions*

These data demonstrate the feasibility of removing SARS-CoV-2 from the air of repurposed ‘surge’ wards and suggest that air filtration devices may help reduce the risk of hospital-acquired SARS-CoV-2.

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## 1 **Introduction**

2 During the COVID-19 pandemic ‘general’ hospital wards in the UK were rapidly repurposed into  
3 ‘surge’ wards and intensive care units (ICU), which lacked the capacity for high frequency air-  
4 changes. Airborne dissemination is likely an important transmission route for SARS-CoV-2 <sup>1</sup>, with  
5 SARS-CoV-2 RNA being detected in air samples from wards managing COVID-19 patients <sup>2,3</sup>.  
6 Despite the use of appropriate personal protective equipment (PPE) that filter medium and large size  
7 droplets, there are multiple reports of patient-to-healthcare worker transmission of SARS-CoV-2 <sup>4,5,6</sup>,  
8 potentially through the inhalation of viral particles in small (< 5µM) aerosols <sup>7</sup>. Furthermore,  
9 nosocomial acquisition of COVID-19 has continued to blight healthcare systems despite the  
10 systematic introduction of patient and healthcare worker asymptomatic screening programmes<sup>8</sup>. There  
11 is a need to improve the safety for healthcare workers and patients during the pandemic by decreasing  
12 the potential for the airborne transmission of SARS-CoV-2<sup>7</sup>. Engineering solutions that improve  
13 ventilation with provision of UV light sterilisation are considered a more effective intervention in the  
14 hierarchy of controls against transmissible infections compared to enhanced respiratory protective  
15 equipment<sup>9,10</sup>. Portable air filtration systems, that combine high efficiency particulate filtration and  
16 ultraviolet (UV) light sterilisation, may be a scalable solution for removing respirable SARS-CoV-2.  
17 A recent review by the UK Scientific Advisory Group for Emergencies modelling group found  
18 limited data regarding the effectiveness of such devices <sup>11</sup>, which is consistent with findings from two  
19 recent systematic reviews <sup>12,13</sup>. Most of the testing of such systems has been physical device  
20 validation using inorganic particles or removal of bacterial bioaerosols in controlled test environments  
21 <sup>12,13</sup>. Here we present the first data providing evidence for the removal of SARS-CoV-2 and microbial  
22 bioaerosols from the air using portable air filters with UV sterilisation on a COVID-19 ‘surge’ ward  
23 during the ongoing pandemic.

24

## 25 **Methods**

### 26 *Setting*

27 The study was conducted in two repurposed COVID-19 units in Addenbrooke’s Hospital, Cambridge,  
28 UK in January/February 2021 when the alpha variant (lineage B.1.1.7) comprised >80% of circulating

29 SARS-CoV-2<sup>8</sup>. One area was a ‘surge ward’ (ward) managing patients requiring simple oxygen  
30 therapy or no respiratory support, the second was a ‘surge ICU’ (ICU) managing patients requiring  
31 invasive and non-invasive respiratory support. The ward was a fully occupied four-bedded bay (Fig.  
32 1A). The ICU was fully occupied five-bedded bay, with a super-capacity sixth occupied bed used in  
33 week 2 (Fig. 1B).

34

35 In the ward we installed an AC1500 HEPA14/UV steriliser (Filtrex, Harlow, UK), whilst in the ICU  
36 we installed a Medi 10 HEPA13/UV steriliser (Max Vac, Zurich, Switzerland) (supplemental  
37 methods). The air filters were placed in fixed positions before the initiation of the three-week study  
38 period (Fig. 1), switched on at the beginning of week two and run continuously from Sunday to  
39 Sunday for 24 hours per day, providing approximately 5-10 room-volume filtrations per hour in each  
40 location. As the devices do not meet medical device electrical safety standards (EN60601) they were  
41 operated at a distance of  $\geq 1.5$  metres from any patient.

#### 42 *Study design*

43 We performed a crossover evaluation, with the primary outcome being detection of SARS-CoV-2  
44 RNA in the various size fractions of the air samples. Air sampling was conducted using National  
45 Institute for Occupational Safety and Health (NIOSH) BC 251 two-stage cyclone aerosol samplers<sup>12</sup>  
46 (Donated by B Lindsley, Centers for Disease Control, Atlanta), operated in accordance with previous  
47 studies demonstrating capture of airborne viruses (supplemental methods)<sup>2,14-18</sup>. Air samplers were  
48 assembled daily with a sampler left in a sealed bag as a control. Samplers placed adjacent to the air  
49 filter inlet and the other at approximately four meters and no closer than two meters to patients (Fig.  
50 1). In ICU two distant samplers were used, one mounted at head height and one at bed height.

51

52 The samplers were operated on weekdays (0815hrs to 1415hrs) for three consecutive weeks. After  
53 sampling, the samplers were disassembled using sterile technique and the filter papers were  
54 transferred to 15 ml Falcon tubes. The samples were processed then stored at  $-80^{\circ}\text{C}$  until analysis.

55 The samplers were washed with 80% ethanol and demineralised water.

56

57 *Pathogen detection*

58 Nucleic acids were extracted from each NIOSH sampler component (tubes containing large aerosols,  
59 medium aerosols, and filter), as previously described<sup>19</sup>. Details of the RT-qPCR for SARS-CoV-2 and  
60 multiplex qPCR assays for a range of respiratory and other bacterial, viral, and fungal pathogens are  
61 in the supplemental section.

62

63 *Statistical analyses*

64 Differences in the number of pathogens detected when air filter was on and off were compared by  
65 Mann-Whitney U-test. Statistical significance was inferred when  $p$  values were  $\leq 0.05$ . Graphs were  
66 generated in R studio.

67

68 The study was registered as a service evaluation with Cambridge University Hospitals NHS  
69 Foundation Trust (Service Evaluation Number PRN 9798).

70

71 **Results**

72 *Removal of SARS-CoV-2 by air filtration on surge ward*

73 For the duration of the study (18<sup>th</sup> January to 5<sup>th</sup> February) the beds in the ward and ICU were at  
74 100% occupancy, with 15 patients admitted to the ward and 14 admitted to the ICU over the three-  
75 week sampling period (7, 4, 4 in weeks 1-3 in the ward and 6, 5, 3 in the ICU, respectively). All  
76 patients were symptomatic and tested positive for SARS-CoV-2 RNA from a respiratory sample  
77 before admission. Patients in the ICU were managed with non-invasive mask ventilation, high flow  
78 nasal oxygen or invasive ventilation via endotracheal tube or tracheostomy. Patients in the ward were  
79 spontaneously ventilating with simple oxygen therapy or no respiratory support and no aerosol-  
80 generating procedures performed.

81

82 In the ward, during the first week whilst the air filter was inactive, we were able to detect SARS-CoV-  
83 2 on all five sampling days; RNA was detected in both the medium (1-4 $\mu$ M particle size) and the



84 large ( $>4\mu\text{M}$  particle size) particulate fractions (Fig. 2A). SARS-CoV-2 RNA was not detected in the  
85 small ( $<1\mu\text{M}$ ) particulate filter. The air filter was switched on in week two and run continuously; we  
86 were unable to detect SARS-CoV-2 RNA in any of the sampling fractions on any of the five testing  
87 days. These initial observations provided evidence for the removal of SARS-CoV-2 via the air filter  
88 system, albeit at high baseline  $C_T$  values. To confirm this observation, we completed the study by  
89 repeating the sampling with an inactive air filter. As in week one, we were able to detect SARS-CoV-  
90 2 RNA in the medium and the large particulate fractions on 3/5 days of sampling (a sample without  
91 tube size indicated tested positive on day 5) (Fig. 2A). We did not detect SARS-CoV-2 RNA from the  
92 control sampler.

93

#### 94 *Removal of additional bioaerosols by air filtration on surge ward*

95 We subjected the extracted nucleic acid preparations to high-throughput qPCR using a Biomark HD  
96 system to detect a range of viral, bacterial, and fungal targets. In the week one samples, we detected  
97 nucleic acid from multiple viral, bacterial, and fungal pathogens on all sampling days (Fig. 2B). In  
98 contrast, when the air filter was switched on, we detected yeast targets only on a single day, with a  
99 significant reduction ( $p=0.05$ ) in microbial bioaerosols when the air filter was operational (Fig. 2C).  
100 Using this high-throughput approach, SARS-CoV-2 RNA was detected on 4/5 days tested in week 1  
101 but was again absent in week 2. We were unable to generate multiplex data for week three due to  
102 sample degradation after storage of sample following SARS-CoV-2 RNA amplification.

103

#### 104 *Effectiveness of air filter on surge ICU*

105 In contrast to the ward, we found limited evidence of airborne SARS-CoV-2 in weeks one and three  
106 (filter off) but detected SARS-CoV-2 RNA in a single sample in the medium ( $1-4\mu\text{M}$  particle size)  
107 particulates on week 2 (filter on) (Fig. 3A). This contrary result did not reflect a general lack of  
108 bioaerosols in the ICU, which demonstrated a comparable quantity and array of pathogen associated  
109 nucleic acids to that seen in the unfiltered ward air on week one (Fig. 3B). Again, the use of the air  
110 filtration device significantly ( $p=0.05$ ) reduced the microbial bioaerosols (Fig 3C); with only three

111 organism types detected on two of the sampling days (Fig 3B). SARS-CoV-2 RNA was only detected  
112 once on the high-throughput qPCR assay, during week one.

113

## 114 **Discussion**

115 Our study represents the first report of successful removal of airborne SARS-CoV-2 in a hospital  
116 environment using combined air filtration and UV sterilisation technology. Specifically, we provide  
117 evidence for the circulation of SARS-CoV-2 in a ward within airborne droplets of  $>1\mu\text{M}$ . Droplets of  
118  $1\text{--}4\mu\text{M}$  are likely a key vehicle for SARS-CoV-2 transmission, as they can remain airborne for a  
119 prolonged period. They are also readily respirable and can deposit in the distal airways. Recent data  
120 has shown that exertional respiratory activity, such as that seen in patients with COVID-19, increases  
121 the release of  $1\text{--}4\mu\text{M}$  respiratory aerosols, whilst conventionally defined ‘aerosol generating  
122 procedures’ such as high flow nasal oxygen and non-invasive ventilation actively reduce aerosol  
123 generation during exertion<sup>20</sup>. These data are consistent with our observations, suggesting that  
124 precautions to remove aerosolisation may be more important in conventional wards than in well  
125 defined ‘aerosol risk areas’. We also found a low burden of SARS-CoV-2 in the air on the ICU. This  
126 observation, combined with the higher level of aerosol protection worn by ICU staff, may explain  
127 why staff in these areas appear to be at significantly lower risk of acquiring COVID than those  
128 working on wards<sup>21</sup>.

129

130 The sampling and detection of airborne viruses poses several technological challenges, and although  
131 several approaches have been developed, there remains no agreed standard for their use or  
132 interpretation<sup>22</sup>. However, the detection of SARS-CoV-2 RNA by RT-qPCR (albeit at a high  $C_T$   
133 value), and the lack of detection during use of an air filtration/UV sterilisation system, adds to a  
134 growing body of evidence implicating the airborne transmission of SARS-CoV-2<sup>1</sup>. The detection of  
135 SARS-CoV-2 RNA in the air of a ward managing patients with COVID-19 intimates that this is a key  
136 mechanism by which healthcare professionals could become infected during patient care. The removal  
137 of airborne viral particles and other pathogens may help reduce the likelihood of hospital-acquired

138 respiratory infections. This reduction may be by both decreasing the load of respirable particles and  
139 by removal of larger droplets that can facilitate fomite-associated spread<sup>22</sup>. The clearance of  
140 bioaerosol was not restricted to SARS-CoV-2. A range of bacteria, yeasts, and other respiratory  
141 viruses with pathogenic potential were detected in the air of both rooms in the first week, with their  
142 burden significantly reduced during air filtration. Although the impact of air filtration on nosocomial  
143 infection is uncertain<sup>23</sup>, the broad range of pathogens removed in this study suggests potential for  
144 benefit beyond removal of SARS-CoV-2.

145

146 There are several potential explanations for the lower detection of SARS-CoV-2 in air of an ICU.  
147 These include a later stage of disease during which viral replication is less pronounced<sup>24</sup>, higher viral  
148 loads in the lower rather than upper respiratory tract in critically ill patients<sup>25</sup> and use of respiratory  
149 devices, which reduce aerosol generation<sup>20</sup>. The reduction in microbial bioaerosols found in ICU  
150 during the week of the air filtration system provides confidence that the device was similarly effective  
151 to that used on the ward, despite the infrequent detection of SARS-CoV-2.

152

153 A recent systematic review of both static and portable air filtration, which also assessed relevant  
154 building codes and guidelines<sup>12</sup>, identified no robust studies of air filtration. Although multiple  
155 building codes propose air filtration to protect vulnerable patients and to reduce risks of transmission  
156 of airborne diseases, these have not been updated in light of COVID-19<sup>12</sup>. Mousvai and colleagues  
157 identified several studies demonstrating the capacity of air filtration to reduce inert, fungal, and  
158 bacterial bioaerosols in experimental and clinical contexts. These findings are consistent with our  
159 report, but previous data originate from fixed rather than portable air filtration devices. No reports of  
160 SARS-CoV-2 removal were identified. A further recent review focussed solely on portable air  
161 filters<sup>13</sup>, with studies demonstrating the removal of inert particles and deliberately aerosolised  
162 bacteria, again no reports of SARS-CoV-2 removal were identified. The Centres for Disease Control  
163 recommend the consideration of portable HEPA-based air filters; this recommendation applies only to  
164 dental facilities where there is deemed to be a high risk of aerosol generation<sup>26</sup>.

165

166 This study has limitations, being conducted rapidly in active wards during an ongoing pandemic. The  
167 evaluation was conducted in two rooms and there are no data defining the optimal air changes  
168 required to remove detectable pathogens with the specified devices. Given the large volume of air  
169 within the room and the stability of viruses in the sampling fluid, it was predictable that the amount of  
170 SARS-CoV-2 detected via qPCR would be minimal, as evidenced by high  $C_T$  values. Therefore, we  
171 cannot categorically state that there was circulating infectious virus. RNA is sufficient to suggest the  
172 virus was present and it has been shown that aerosolised virus can remain infectious for >3 hours<sup>27,28</sup>;  
173 additionally, air sampling devices can artefactually reduce the apparent viability of sampled virus.  
174 Negative results from the control sampler, and the striking but reversible effect of the air filtration  
175 devices, suggest these are not false positive detections and we cannot exclude the risk of airborne  
176 infection. Future studies should examine whether air filtration devices, such as those used here, have  
177 an impact on healthcare professional and patient focussed outcomes, including measuring  
178 infection/exposure as an endpoint, as well as assessing potential harm, such as noise, reduced ambient  
179 humidity or impact on delivery of care.

180

181 In conclusion, we were able to detect airborne SARS-CoV-2 RNA in a repurposed COVID-19 ‘surge  
182 ward’ and found that air filtration can remove SARS-CoV-2 RNA below the limit of qPCR detection.  
183 SARS-CoV-2 was infrequently detected in the air of a ‘surge ICU’; however, the device retained its  
184 ability to reduce microbial bioaerosols. Our data is highly indicative of aerosolised SARS-CoV-2  
185 circulating in areas that are not classically considered ‘aerosol risk areas’. Furthermore, portable air  
186 filtration devices can mitigate the reduced availability of airborne infection isolation facilities when  
187 surges of COVID-19 patients overwhelm healthcare resources. The use of such systems may provide  
188 additional safety for those that are of high exposure risk to respiratory pathogens such as SARS-CoV-  
189 2.

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303

304 **Figure legends**

305

306 **Figure 1.** Location of the air filters and room layout.

307 A) Layout of the room on the ‘surge’ ward with four beds. B) Layout on the ‘surge’ ICU with six beds  
308 including the addition of a further bed to increase occupancy (labelled with red box). Locations of the  
309 NIOSH air samplers indicated by \*. The air filters were installed in the marked locations and set to  
310 operate at 1000 m<sup>3</sup>/hour. The room’s volumes are approximately 107 m<sup>3</sup> and 195m<sup>3</sup> respectively.  
311 Fresh air was not supplied or extracted in these areas.

312

313 **Figure 2.** Bioaerosol detection in specific air sampler fractions over the three-week testing period on  
314 the ‘surge’ ward.

315 A)  $C_T$  values for SARS-CoV-2 qPCR on air sample fractions collected daily from the ward. Colours  
316 indicate the specific component of the sampler where SARS-CoV-2 was detected. Components  
317 collected aerosols dependent on size fractions; large >4  $\mu\text{m}$ , medium 1-4  $\mu\text{m}$ , small <1  $\mu\text{m}$ . B) Daily  
318 detection of fungal, bacterial and viral bioaerosols detected by high-throughput qPCR collected during  
319 weeks one (filter off) and two (filter on). The differences in  $C_T$  values between the regular qPCR (A)  
320 and high-throughput qPCR (B) are a function of the microfluidics technology, and do not reflect  
321 higher bioaerosol burdens. C) Stacked bar chart showing collated total number of bioaerosol  
322 detections during weeks one (filter off) and two (filter on). \* $p=0.05$  by Mann-Whitney U test.

323

324 **Figure 3.** Bioaerosol detection in specific air sampler fractions over the three-week testing period on  
325 the ‘surge’ ICU.

326 A)  $C_T$  value for the single qPCR SARS-CoV-2 detection on day 9 (week 2) in the medium (1-4  $\mu\text{m}$   
327 particle size) fraction. B) Daily detection of fungal, bacterial and viral bioaerosol detected by high-  
328 throughput qPCR collected during weeks one (filter off) and two (filter on). The differences in  $C_T$   
329 values between the regular qPCR (A) and high-throughput qPCR (B) are a function of the



330 microfluidics technology, and do not reflect higher bioaerosol burdens. C) Stacked bar chart showing

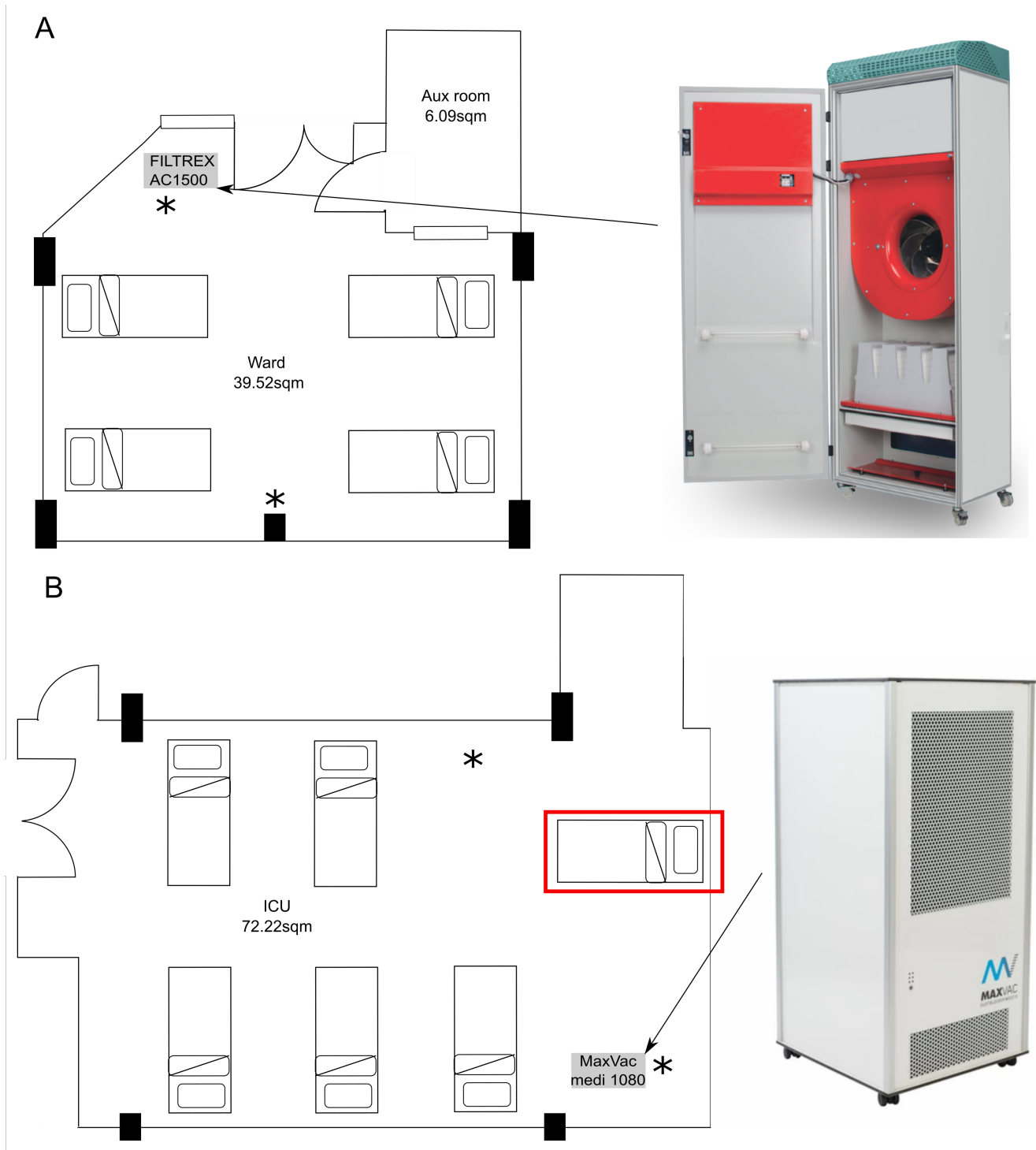
331 collated total number of bioaerosol detections during weeks one (filter off) and two (filter on).

332 \* $p=0.05$  by Mann-Whitney U test.

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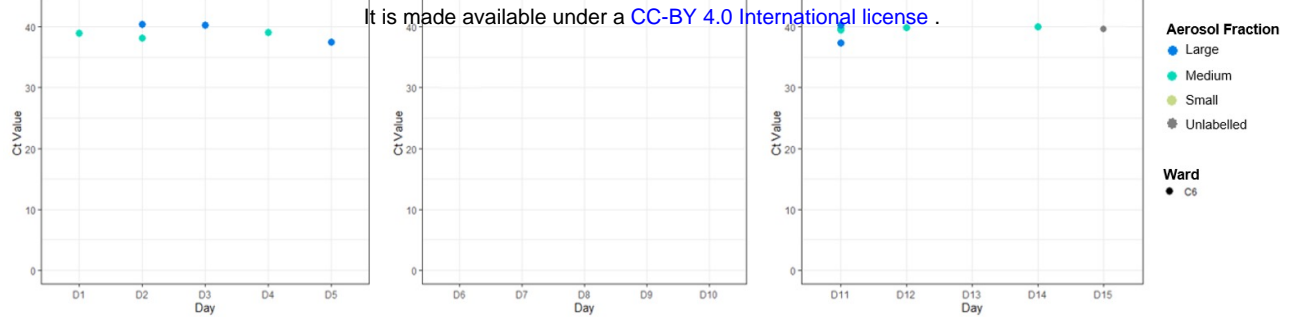
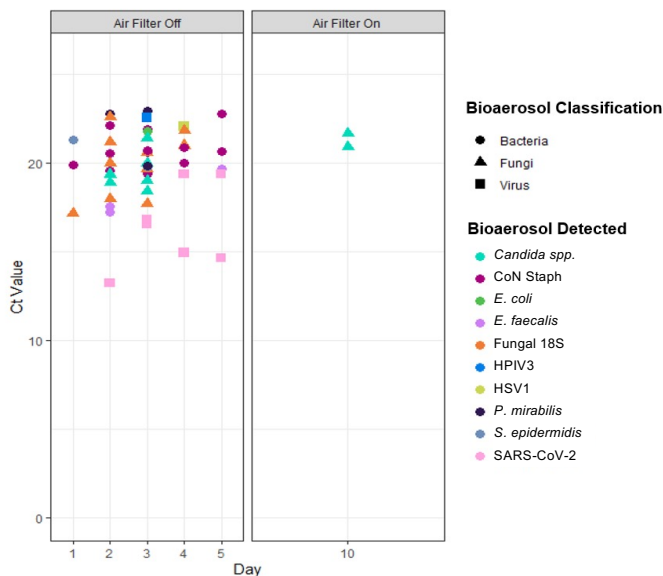
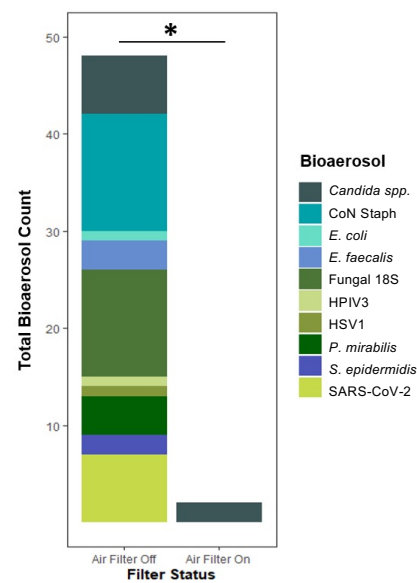


**Figure 1.** Location of the air filters and room layout.

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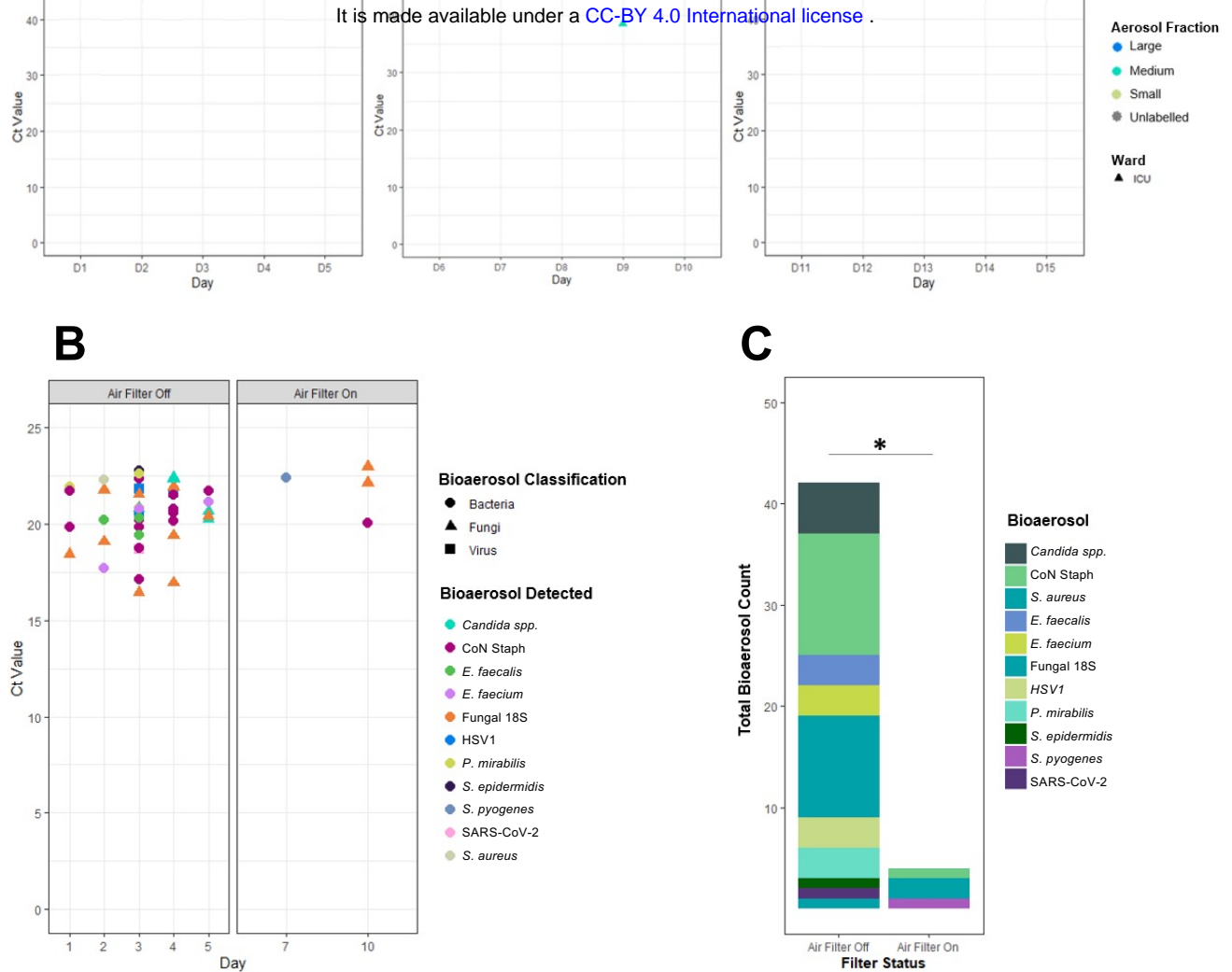
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**B****C**

**Figure 2.** Bioaerosol detection in specific air sampler fractions over the three-week testing period on the ‘surge’ ward.

A)  $C_T$  values for SARS-CoV-2 qPCR on air sample fractions collected daily from the ward. Colours indicate the specific component of the sampler where SARS-CoV-2 was detected. Components collected aerosols dependent on size fractions; large  $>4 \mu\text{m}$ , medium  $1-4 \mu\text{m}$ , small  $<1 \mu\text{m}$ . B) Daily detection of fungal, bacterial and viral bioaerosols detected by high-throughput qPCR collected during weeks one (filter off) and two (filter on). The differences in  $C_T$  values between the regular qPCR (A) and high-throughput qPCR (B) are a function of the microfluidics technology, and do not reflect higher bioaerosol burdens. C) Stacked bar chart showing collated total number of bioaerosol detections during weeks one (filter off) and two (filter on).  $*p=0.05$  by Mann-Whitney U test.



**Figure 3.** Bioaerosol detection in specific air sampler fractions over the three-week testing period on the ‘surge’ ICU.

A)  $C_T$  value for the single qPCR SARS-CoV-2 detection on day 9 (week 2) in the medium (1-4  $\mu\text{m}$  particle size) fraction. B) Daily detection of fungal, bacterial and viral bioaerosol detected by high-throughput qPCR collected during weeks one (filter off) and two (filter on). The differences in  $C_T$  values between the regular qPCR (A) and high-throughput qPCR (B) are a function of the microfluidics technology, and do not reflect higher bioaerosol burdens. C) Stacked bar chart showing collated total number of bioaerosol detections during weeks one (filter off) and two (filter on). \* $p=0.05$  by Mann-Whitney U test.