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

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3

The study was registered as a service evaluation with Cambridge University Hospitals NHS

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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.

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 [1], with

5 SARS-CoV-2 RNA being detected in air samples from wards managing COVID-19 patients [2,3].

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 [4,5,6,]

8 potentially through the inhalation of viral particles in small ($< 5\mu M$) aerosols [7]. 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[8]. 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[7]. 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[9,10]. 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 [11] , which is consistent with findings from two

19 recent systematic reviews [12, 13]. 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 [12,13]. 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 B1.1.7) comprised >80% of circulating

6

29 SARS-CoV-2[8]. 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 ≥1.5metres 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[12]

46 (Donated by B Lindsley, Centers for Disease Control, Atlanta), operated in accordance with previous

47 studies demonstrating capture of airborne viruses (supplemental methods)[2,14-18]. 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°C until analysis.

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

7

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[19]. 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 ≤ 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 (18th January to 5th 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 (1-4μM particle size) and the

8

84   large (>4μM particle size) particulate fractions (Fig. 2A). SARS-CoV-2 RNA was not detected in the

85   small (<1μM) 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 (1-4μM 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

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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 >1µM. Droplets of

118   1-4µM 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 1-4 µM 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[20]. 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[21].

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[22]. 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 [1]. 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[22].   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[23], 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[24], higher viral

148   loads in the lower rather than upper respiratory tract in critically ill patients[25] and use of respiratory

149   devices, which reduce aerosol generation[20].   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[12,] 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[12]. 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[13], 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[26].

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 [27,28];

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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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^3/hour. The room's volumes are approximately 107 m^3 and 195m^3 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 µm, medium1-4 µm, small <1 µm. 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 µm

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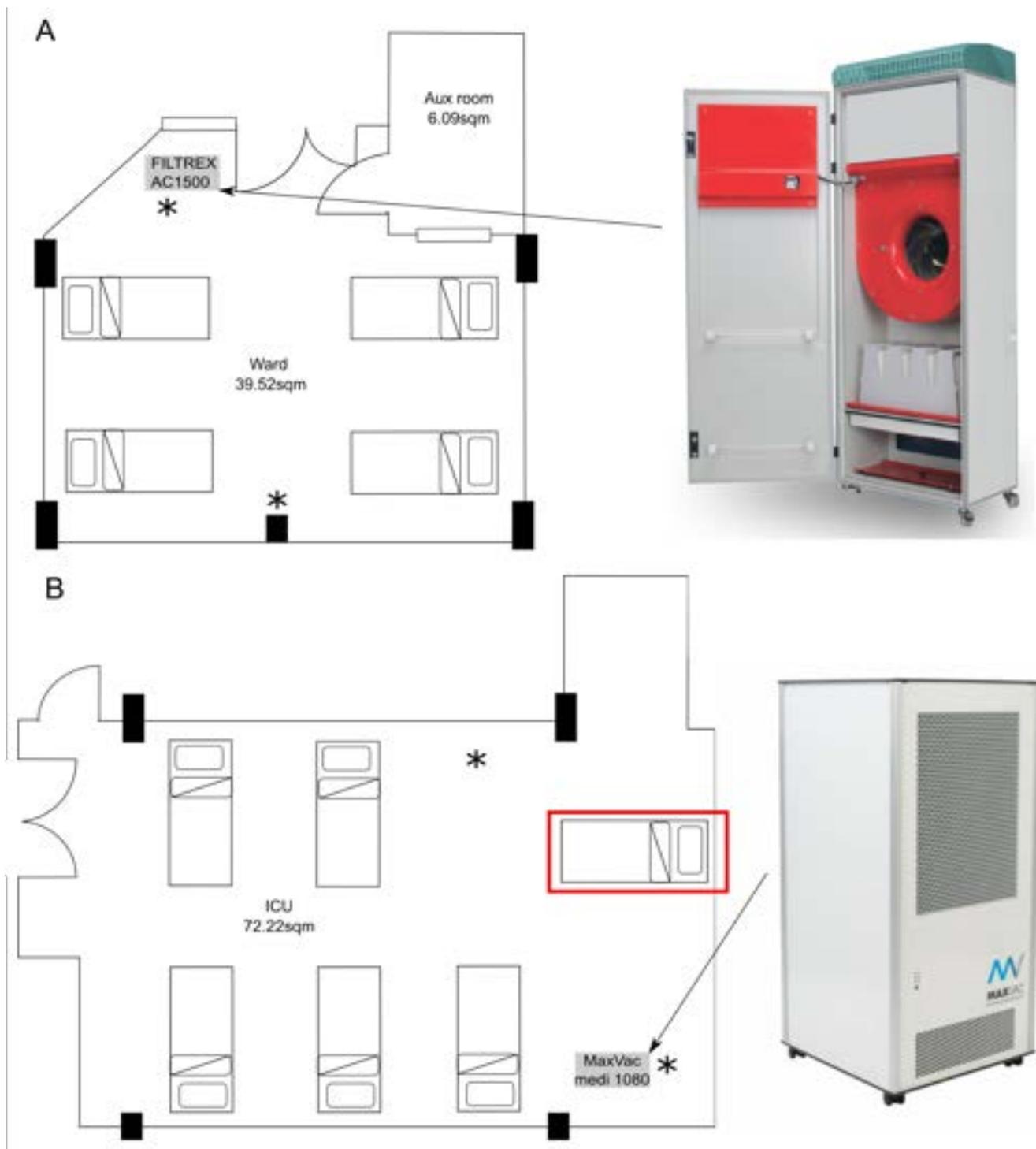
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330   microfluidics technology, and do not reflect higher bioaerosol burdens. C) Stacked bar chart showing

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.

333

334

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

A) Layout of the room on the 'surge' ward with four beds. B) Layout on the 'surge' ICU with six beds including the addition of the additional bed to increase occupancy (labelled with rad box). Locations of the NIOSH air samplers indicated by *. The air filters were installed in the marked locations and set to operate at 1000 m³/hour. The rooms volumes are approximately 107 m³ and 195m³ respectively. Fresh air was not supplied or extracted in these areas.
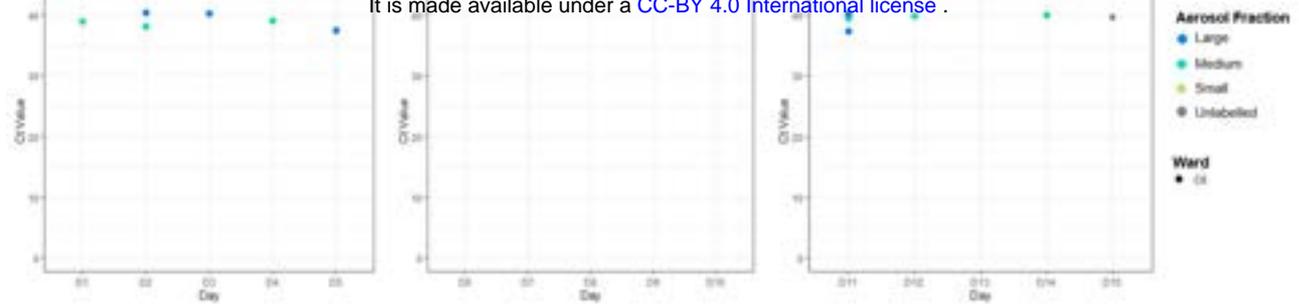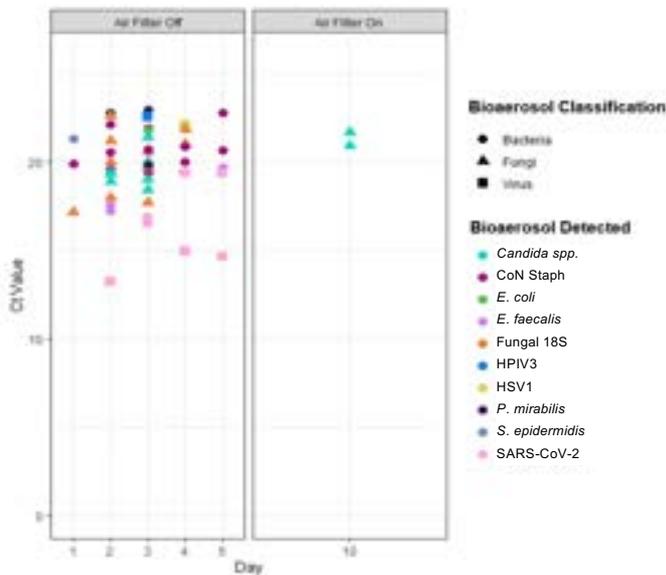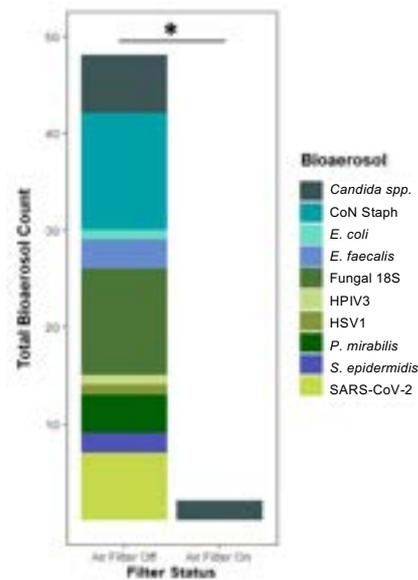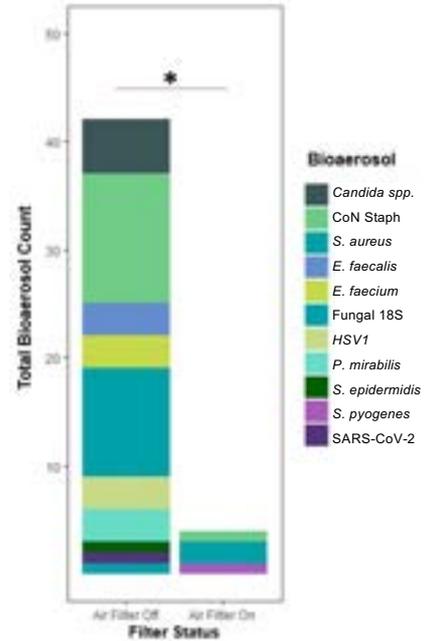
Figure 2. Bioaerosol detection in specific air sampler fractions over the three-week testing period on the 'surge' ward.

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 μm, medium1-4 μm, small <1 μm. 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 μm 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.