

1 **Viable SARS-CoV-2 in the air of a hospital room with COVID-19 patients**

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27 **Summary**

28 **Background** There currently is substantial controversy about the role played by SARS-CoV-2 in aerosols
29 in disease transmission, due in part to detections of viral RNA but failures to isolate viable virus from
30 clinically generated aerosols.

31 **Methods** Air samples were collected in the room of two COVID-19 patients, one of whom had an active
32 respiratory infection with a nasopharyngeal (NP) swab positive for SARS-CoV-2 by RT-qPCR. By using
33 VIVAS air samplers that operate on a gentle water-vapor condensation principle, material was collected
34 from room air and subjected to RT-qPCR and virus culture. The genomes of the SARS-CoV-2 collected
35 from the air and of virus isolated in cell culture from air sampling and from a NP swab from a newly
36 admitted patient in the room were sequenced.

37 **Findings** Viable virus was isolated from air samples collected 2 to 4.8m away from the patients. The
38 genome sequence of the SARS-CoV-2 strain isolated from the material collected by the air samplers was
39 identical to that isolated from the NP swab from the patient with an active infection. Estimates of viable
40 viral concentrations ranged from 6 to 74 TCID₅₀ units/L of air.

41 **Interpretation** Patients with respiratory manifestations of COVID-19 produce aerosols in the absence of
42 aerosol-generating procedures that contain viable SARS-CoV-2, and these aerosols may serve as a source
43 of transmission of the virus.

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53 **Research in context**

54 **Evidence before this study**

55 Various studies report detection of SARS-CoV-2 in material collected by air samplers positioned in
56 clinics and in some public spaces. For those studies, detection of SARS-CoV-2 has been by indirect
57 means; instead of virus isolation, the presence of the virus in material collected by air samplers has been
58 through RT-PCR detection of SARS-CoV-2 RNA. However, questions have been raised about the clinical
59 significance of detection of SARS-CoV-2 RNA, particularly as airborne viruses are often inactivated by
60 exposure to UV light, drying, and other environmental conditions, and inactivated SARS-CoV-2 cannot
61 cause COVID-19.

62 **Added value of this study**

63 Our virus isolation work provides direct evidence that SARS-CoV-2 in aerosols can be viable and thus
64 pose a risk for transmission of the virus. Furthermore, we show a clear progression of virus-induced
65 cytopathic effects in cell culture, and demonstrate that the recovered virus can be serially propagated.
66 Moreover, we demonstrate an essential link: the viruses we isolated in material collected in four air
67 sampling runs and the virus in a newly admitted symptomatic patient in the room were identical. These
68 findings strengthen the notion that airborne transmission of viable SARS-CoV-2 is likely and plays a
69 critical role in the spread of COVID-19.

70 **Implications of all the available evidence**

71 Scientific information on the mode of transmission should guide best practices Current best practices for
72 limiting the spread of COVID-19. Transmission secondary to aerosols, without the need for an aerosol-
73 generating procedure, especially in closed spaces and gatherings, has been epidemiologically linked to
74 exposures and outbreaks. For aerosol-based transmission, measures such as physical distancing by 6 feet
75 would not be helpful in an indoor setting and would provide a false-sense of security. With the current
76 surges of cases, to help stem the COVID-19 pandemic, clear guidance on control measures against SARS-
77 CoV-2 aerosols are needed.

78

79 **Introduction**

80 Severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2), genus *Betacoronavirus*, subgenus
81 *Sarbecovirus*, family *Coronaviridae*, is a positive-polarity single-stranded RNA virus that probably
82 originated in bats¹⁻³ and is the causative agent of coronavirus disease of 2019 (COVID-19).⁴ The
83 dynamics of the COVID-19 pandemic have proven to be complex. Many challenges remain pertaining to
84 a better understanding of the epidemiology, pathology, and transmission of COVID-19. For example, the
85 clinical manifestations of COVID-19 range from an asymptomatic infection, mild respiratory illness to
86 pneumonia, respiratory failure, multi-organ failure, and death.⁵⁻⁷ Diarrhea due to gastro-intestinal
87 infection can also occur, and *in vitro* modeling suggests that the virus infects human gut enterocytes.⁸
88 SARS-CoV-2 RNA can be found in rectal swabs and fecal aerosols, even after nasal-pharyngeal testing
89 has turned negative,⁹⁻¹² suggesting that a fecal–oral transmission route may be possible.

90 To-date, there has been a strong emphasis on the role of respiratory droplets and fomites in the
91 transmission of SARS-CoV-2.^{13,14} Yet SARS-CoV-2 does not appear to be exclusively inhaled as a
92 droplet, and epidemiologic data are consistent with aerosol transmission of SARS-CoV-2.¹⁵⁻¹⁹
93 Furthermore, SARS-CoV-2 genomic RNA has been detected in airborne material collected by air
94 samplers positioned distal to COVID-19 patients.^{9, 20-23} Any respiratory virus that can survive
95 aerosolization poses an inhalation biohazard risk, and van Doremalen *et al.*²⁴ experimentally generated
96 aerosol particles with SARS-CoV-2 and found that the virus remained viable during a three-hour testing
97 period. More recently, Fears *et al.*²⁵ reported that the virus retained infectivity and integrity for up to 16
98 hours in laboratory-created respirable-sized aerosols. Nevertheless, finding virus RNA in material
99 collected by an air sampler may not correlate with risk. Indeed, the air we breathe is full of viruses
100 (animal, plant, bacterial, human, etc.), yet a large proportion of the viruses in air are non-viable due to
101 UV-inactivation, drying, etc., and non-viable viruses cannot cause illnesses. Because efforts to isolate
102 virus in cell cultures in the aforementioned air sampling studies in hospital wards were not made,^{20,22} or
103 failed when they were attempted due to overgrowth by faster replicating respiratory viruses,²³ or provided

104 weak evidence of virus isolation,²¹ uncertainties about the role of aerosols in COVID-19 transmission
105 remain.

106 It is well known that virus particles collected by various air samplers become inactivated during the air
107 sampling process,²⁶ and if such is the case for SARS-CoV-2, this partly explains why it has been difficult
108 to prove that SARS-CoV-2 collected from aerosols is viable. Because we previously collected SARS-
109 CoV-2 from the air of a respiratory illness ward within a clinic but were unable to isolate the virus in cell
110 cultures due to out-competition by other respiratory viruses,²³ we sought to perform air sampling tests in a
111 hospital room reserved for COVID-19 patients, to lessen the probability of collecting other airborne
112 human respiratory viruses. We thus collected aerosols containing SARS-CoV-2 in a room housing
113 COVID-19 patients using our VIVAS air samplers that collect virus particles without damaging them,
114 thus conserving their viability. These samplers operate using a water-vapor condensation mechanism.^{27,28}
115 Air samplings were performed at the University of Florida Health (UF Health) Shands Hospital, which is
116 a 1,050-bed teaching hospital situated in Gainesville, Florida. As of 10 July 2020, > 200 patients have
117 been treated at the hospital for COVID-19. The current study was conducted as part of ongoing
118 environmental investigations by the UF Health infection control group to assess possible healthcare
119 worker exposure to SARS-CoV-2.

120

121 **Methods**

122 Detailed methods are provided in a Technical Appendix. An abbreviated summary of methods is
123 provided below:

124 **Institutional Review Board (IRB) approval and patients**

125 The study protocol was approved by the UF IRB (study IRB202002102). Patient 1 was a person with
126 coronary artery disease and other co-morbidities who had been transferred from a long-term care facility
127 for COVID-19 treatment the evening before our air sampling tests were initiated; he had a positive NP
128 swab test on admission that was positive for SARS-CoV-2 by RT-PCR. Patient 2 had been admitted four
129 days before the air sampling tests with a mid-brain stroke; the patient had a positive NP swab test for

130 SARS-CoV-2 on admission, but a repeat test was negative, and the patient was in the process of being
131 discharged at the time the air sampling was being done.

132 **Hospital room**

133 Air samples were collected in a room that was part of a designated COVID-19 ward (Figure 1). The room
134 had six air changes per hour and the exhaust air underwent triple filter treatment (minimum efficiency
135 reporting value [MERV] 14, 75%-85% efficiency for 0.3 μm particles), coil condensation (to remove
136 moisture), and UV-C irradiation prior to recycling 90% of the treated air back to the room.

137 **Air samplers and sampling parameters**

138 Three serial 3-hr air samplings were performed using our prototype VIVAS air sampler,^{23, 27, 28} as well as
139 a BioSpot-VIVAS BSS300P, which is a commercial version of the VIVAS (available from Aerosol
140 Devices Inc., Ft. Collins, CO). These samplers collect airborne particles using a water-vapor
141 condensation method.^{23, 27, 28} Two samplers were used so that air could be collected/sampled at different
142 sites of the same room during a given air sampling period. For each sampler, the second of the three
143 samplings was performed with a high efficiency particulate arrestance (HEPA) filter affixed to the inlet
144 tube, a process we implement to reveal whether virus detected in consecutive samplings reflect true
145 collection and not detection of residual virus within the collector. The air-samplers were stationed from 2
146 to 4.8 m away from the patients (Figure 1).

147 **Detection of SARS-CoV-2 genomic RNA (vRNA) in collection media**

148 vRNA was extracted from virions in collection media and purified by using a QIAamp Viral RNA Mini
149 Kit (Qiagen, Valencia, CA, USA). Twenty-five μL (final volume) real-time reverse-transcription
150 polymerase chain reaction (rtRT-PCR) tests were performed in a BioRad CFX96 Touch Real-Time PCR
151 Detection System using 5 μL of purified vRNA and rtRT-PCR primers and the probe listed in Table 1
152 that detect a section of the SARS-CoV-2 N-gene.²³ The N-gene rRT-PCR assay that was used was part of
153 a dual (N- and RdRp-gene) rRT-PCR assay designed by J. Lednicky and does not detect common human
154 alpha- or beta-coronaviruses. Using this particular N-gene rRT-PCR detection system, the limit of
155 detection is about 1.5 SARS-CoV-2 genome equivalents per 25 μL rRT-PCR assay.

156 **Cell lines for virus isolation**

157 Cell lines used for the isolation of SARS-CoV-2 were obtained from the American Type Culture
158 Collection (ATCC) and consisted of LLC-MK2 (Rhesus monkey kidney cells, catalog no. ATCC CCL-7)
159 and Vero E6 cells (African green monkey kidney cells, catalog no. ATCC CRL-1586).

160 **Isolation of virus in cultured cells**

161 Cells grown as monolayers in a T-25 flask (growing surface 25 cm²) were inoculated when they were at
162 80% of confluency. First, aliquots (100 µL) of the concentrated air sampler collection media were filtered
163 through a sterile 0.45 µm pore-size PVDV syringe-tip filter to remove bacterial and fungal cells and
164 spores. Next, the spent LLC-MK2 and Vero E6 cell culture medium was removed and replaced with 1 mL
165 of cell culture medium, and the cells inoculated with 50 µL of cell filtrate. When virus-induced cytopathic
166 effects (CPE) were evident, the presence of SARS-CoV-2 was determined by rRT-PCR.

167 **Quantification of SARS-CoV-2 genomes in sampled air**

168 The number of viral genome equivalents present in each sample was estimated from the measured
169 quantification cycle (Cq) values. To do so, a 6-log standard curve was run using 10-fold dilutions of a
170 calibrated plasmid containing an insert of the SARS-CoV-2 N-gene that had been obtained from IDT
171 Technologies, Inc. (Coralville, Iowa). The data was fit using equation (eq.) 1:

172 Eq. 1. $y = (\log_{10}GE)(a) + b$, where $y = Cq$ value, $a =$ slope of the regression line, $\log_{10}GE$ is the base
173 10 log genome equivalents, and b is the intercept of the regression line.

174 **Sanger sequencing of SARS-CoV-2 genomes in material collected by air samplers**

175 To obtain the virus consensus sequence prior to possible changes that might occur during isolation of the
176 virus in cell cultures, a direct sequencing approach was used. Because the amount of virus present in the
177 samples was low and thus unsuitable for common next-generation sequencing approaches, Sanger
178 sequencing based on a gene-walking approach with over-lapping primers was used to obtain the virus
179 sequence.²³

180 **Next-generation sequencing the genome of SARS-CoV-2 isolated from NP swab**

181 The vRNA extracted from virions in spent Vero E6 cell culture medium served as a template to generate a
182 cDNA library using a NEBNext Ultra II RNA Library Prep kit (New England Biolabs, Inc.). Sequencing
183 was performed on an Illumina MiSeq sequencer using a 600-cycle v3 MiSeq Reagent kit. Following the
184 removal of host sequences (*Chlorocebus sabaues*; GenBank assembly accession number
185 GCA_000409795.2) using Kraken 2,²⁹ *de novo* assembly of paired-end reads was performed in SPAdes
186 v3.13.0 with default parameters.³⁰

187 **Results**

188 SARS-CoV-2 genomic RNA (vRNA) was detected by real-time reverse transcriptase quantitative
189 polymerase chain reaction (rRT-qPCR) in material collected by air samplings 1-1, 1-3, 2-1, and 2-3,
190 which had been performed without a HEPA filter covering the inlet tube. In contrast, in the presence of a
191 HEPA filter, no SARS-CoV-2 genomes were detected in air samplings 1-2 and 2-2 (Table 1).

192 Virus-induced CPE were observed in LLC-MK2 and Vero E6 cells inoculated with material extruded
193 from the NP specimen of patient 1 and from liquid collection media from air samples 1-1, 1-3, 2-1, and 2-
194 3. Early CPE in both LLC-MK2 and Vero E6 cells consisted of the formation of cytoplasmic vacuoles
195 that were apparent within 2 days post-inoculation (dpi) of the cells with material extruded from the NP
196 swab and 4 to 6 dpi with aliquots of the liquid collection media from the air samplers. At later times (4
197 days onwards after inoculation of cell cultures with material from the NP swab, and 6 – 11 dpi of the cells
198 with material collected by air samplers), rounding of the cells occurred in foci, followed by detachment of
199 the cells from the growing surface. Some of the rounded cells detached in clumps, and occasional small
200 syncytia with 3 -5 nuclei were observed. Apoptotic and necrotic cells were also observed. A
201 representative collage showing the progressive development of CPE in Vero E6 cells inoculated with
202 material collected during air sampling 1-1 is shown in Figure 2. Cytopathic effects were not observed and
203 virus was not detected or isolated from the culture medium of samples 1-2 and 2-2, wherein HEPA filters
204 had been affixed to the inlet nozzles of the air samplers, and were not observed in mock-inoculated cells
205 which were maintained in parallel with the inoculated cell cultures.

206 SARS-CoV-2-specific rRT-PCR tests were performed and the results indicated that the LLC-MK2 and
207 Vero E6 cultures inoculated with collection media from air samplings 1-1, 1-3, 2-1, and 2-3 contained
208 SARS-CoV-2 (data not shown). No other respiratory virus was identified in the samples using a BioFire
209 FilmArray Respiratory 2 Panel (BioMérieux Inc., Durham, North Carolina), following the manufacturer's
210 instructions.

211 Whereas the concentration of SARS-CoV-2 genome equivalents per liter of air were estimated (Table
212 2), determination of the specific infectivity (ratio of SARS-CoV-2 genome equivalents present for every
213 one able to infect a cell in culture) required performance of a plaque assay or a standard 50% endpoint
214 dilution assay (TCID₅₀ assay). Plaque assays could not be performed due to a nationwide non-availability
215 of some critical media components (due to COVID-19 pandemic-related temporary lockdown of
216 production facilities), so TCID₅₀ assays were performed in Vero E6 cells to estimate the percentage of the
217 collected virus particles that were viable. Estimates ranged from 2 to 74 TCID₅₀ units/L of air (Table 3).

218 A nearly complete SARS-CoV-2 genome sequence was obtained by next-generation sequencing (NGS)
219 of RNA purified from cell culture medium of Vero E6 cells 7 dpi with NP swab material from patient 1.
220 The nearly complete genome sequence (and the virus isolate) were designated SARS-CoV-2/human/UF-
221 19/2020, and this genome sequence has been deposited in GenBank (accession no. MT668716) and in
222 GISAID (accession no. EPI_ISL_480349). Because the amount of virus RNA was below the threshold
223 that could be easily sequenced by our NGS methods, Sanger sequencing was used to sequence SARS-
224 CoV-2 RNA purified from the collection media of air samplers 1-1, 1-3, 2-1, and 2-3. One complete
225 SARS-CoV-2 sequence was attained for RNA purified in the material collected by air sampling 1-1, and
226 three nearly complete sequences for 1-3, 2-1, and 2-3, respectively. After alignment, comparisons of the
227 three partial sequences with the complete sequence of SARS-CoV-2 in air sampling 1-1 indicated that the
228 same consensus genome sequence were present in the virions that had been collected in all the air
229 samplings. Moreover, they were an exact match with the corresponding sequences of the virus isolated
230 from patient 1. This complete genome sequence of the virus collected by the air samplers (and the virus
231 therein) were considered the same isolate and designated SARS-CoV-2/Environment/UF-20/2020, and

232 this genome sequence has been deposited in GenBank (accession no. MT670008) and in GISAID
233 (accession no. EPI_ISL_477163). The virus' genomic sequence currently falls within GISAID clade
234 B.1(GH), which is characterized by mutations C241T, C3037T, A23403G, G25563T, S-D614G, and
235 NS3-Q57H relative to reference genome WIV04 (GenBank accession no. MN996528.1). As of 10 July
236 2020, SARS-CoV-2 clade B.1(GH) was the predominant virus lineage in circulation in the USA.

237

238 **Discussion**

239 There are substantial epidemiologic data supporting the concept that SARS-CoV, which is highly related
240 to SARS-CoV-2,³ was transmitted via an aerosol route.³¹⁻³³ For SARS-CoV-2, there have also been two
241 epidemiologic reports consistent with aerosol transmission.^{15,34} However, despite these reports,
242 uncertainties remain about the relative importance of aerosol transmission of SARS-CoV-2, given that so
243 far, only one study has provided weak evidence of virus isolation from material collected by air
244 samplers.²¹ In other reports, attempts to isolate the virus were not successful. The current study takes
245 advantage of a newer air sampling technology that operates using a water-vapor condensation mechanism,
246 facilitating the likelihood of isolating the virus in tissue culture.

247 As reported in air sampling tests performed by others^{9-11,21} and in our previous report,²³ airborne
248 SARS-CoV-2 was present in a location with COVID-19 patients. The distance from the air-samplers to
249 the patients (≥ 2 m) suggests that the virus was present in aerosols. Unlike previous studies, we have
250 demonstrated the virus in aerosols can be viable, and this suggests that there is an inhalation risk for
251 acquiring COVID-19 within the vicinity of people who emit the virus through expirations including
252 coughs, sneezes, and speaking.

253 The amount of airborne virus detected per liter of air was small, and future studies should address (a)
254 whether this is typical for COVID-19, (b) if this represented virus production relative to the phase of
255 infection in the patient, (c) if this was a consequence of active air flow related to air exchanges within the

256 room, (d) or if the low number of virus was due to technical difficulties in removing small airborne
257 particles from the air.²⁶

258 Our findings reveal that viable SARS-CoV-2 can be present in aerosols generated by a COVID-19
259 patient in a hospital room in the absence of an aerosol-generating procedure, and can thus serve as a
260 source for transmission of the virus in this setting. Moreover, the public health implications are broad,
261 especially as current best practices for limiting the spread of COVID-19 center on social distancing,
262 wearing of face-coverings while in proximity to others and hand-washing. For aerosol-based
263 transmission, measures such as physical distancing by 6 feet would not be helpful in an indoor setting,
264 provide a false-sense of security and lead to exposures and outbreaks. With the current surges of cases, to
265 help stem the COVID-19 pandemic, clear guidance on control measures against SARS-CoV-2 aerosols
266 are needed, as recently voiced by other scientists.³⁵

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269 **Contributors**

270 JAL, ML, ZHF, AJ, AEF, KC, JGM Jr, and C-YW conceived and designed the study. JAL, ML, KC, JG
271 M Jr, and C-YW curated the data. JAL, ML, JGM Jr, and C-YW performed formal analyses of the data.
272 JAL, ML, ZHF, AJ, JGM Jr. obtained funding for the work; JAL, TBT, MG, MU, SNS, KM, CJS, MMA,
273 MAE, JCL, KS, and TBW performed experiments; JAL, M L, TBT, SNS, CJS, JCL, KS, TBW, JGM Jr,
274 and C-YW established methods; JAL, ML, JCL, JGM Jr, and C-YW administered the project; JAL, ML,
275 ZHF, AJ, JGM Jr, and C-YW provided resources; JAL, ML, JCL, JGM Jr, and C-YW supervised the
276 project; JAL, JGM Jr., and C-Y Wu wrote the original manuscript draft; all authors revised the
277 manuscript critically. All authors read and approved the final version of the manuscript.

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279 **Declaration of interests**

280 The authors proclaim they have no conflicts of interest to report.

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384 **Tables**

Table 1. SARS-CoV-2 N-gene rRT-PCR primers and probe.

| Primer/probe name | Description | Oligonucleotide sequence (5' to 3') | Label |
|--------------------------|-----------------------------|--|--------------|
| Led-N-F | SARS CoV-2 N Forward Primer | 5'-GGGAGCAGAGGCGGCAGTCAAG-3' | None |
| Led-N-R | SARS CoV-2 N Reverse Primer | 5'-CATCACCGCCATTGCCAGCCATTC-3' | None |
| Led-N-Probe ^a | SARS CoV-2 N Probe | 5' FAM-CCTCATCACGTAGTCGCAACAGTTC- BHQ1-3' | FAM, BHQ1 |

385 ^aThis TaqMan® probe is 5'-end labeled with the reporter molecule 6-carboxyfluorescein (FAM) and with
386 quencher Black Hole Quencher 1 (BHQ-1) at the 3'- end.

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405 Table 2. Results of rRT-qPCR tests of materials collected by air samplers.

| Sample ID | Approx. distance (m) from head of patient 1 ^b | Approx. distance (m) from head of patient 2 ^b | rRT-qPCR test | Cq value | SARS-CoV-2 genome equivalents/25 µL rtRT-PCR test | SARS-CoV-2 genome equivalents/L of air |
|--|--|--|---------------|----------|---|--|
| 1-1 BioSpot | 2 | 4.6 | + | 36.02 | 2.82E+03 | 94 |
| 1-2 BioSpot + HEPA | 2 | 4.6 | - | - | - | - |
| 1-3 BioSpot | 2 | 0 (PD ^b) | + | 37.69 | 9.12E+02 | 30 |
| 2-1 VIVAS | 4.8 | 3 | + | 37.42 | 1.15E+03 | 44 |
| 2-2 VIVAS+ HEPA | 4.8 | 3 | - | - | - | - |
| 2-3 VIVAS | 4.8 | 0 (PD ^d) | + | 38.69 | 4.68E+02 | 16 |
| SARS-CoV-2 vRNA | N/A ^c | N/A | + | 29.53 | 2.20E+05 | N/A |
| N-gene ^a DNA control - 1 | N/A | N/A | + | 26.56 | 1.00E+06 | N/A |
| N-gene DNA control - 2 | N/A | N/A | + | 31.21 | 1.00E+05 | N/A |
| N-gene DNA control - 3 | N/A | N/A | + | 34.71 | 1.00E+04 | N/A |
| N-gene DNA control -4 | N/A | N/A | + | 37.74 | 1.00E+03 | N/A |
| N-gene DNA control - 5 | N/A | N/A | + | 40.41 | 1.00E+02 | N/A |
| N-gene DNA control - 6 | N/A | N/A | + | - | 1.00E+01 | N/A |
| Known positive (NP swab ^e) | N/A | N/A | + | 24.12 | 8.36E+06 | N/A |
| Negative (no RNA) control | N/A | N/A | N/A | - | 0 | N/A |

406 ^aN-gene, N-gene plasmid (positive control template).

407 ^bDistance from sampler inlet nozzle to patient's head.

408 ^cN/A, Not applicable.

409 ^dPD, patient discharged.

410 ^eNP, Nasal-pharyngeal swab from a person screened for SARS-CoV-2 at the UF EPI High-Throughput

411 COVID-19 Research Testing Facility.

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413 Table 3. Estimate of viable virus counts based on TCID₅₀ tests.

| Sample ID | Virus genome equivalents/L of air ^a | TCID ₅₀ /100 µl | Viable virus count/L air |
|--------------------|--|----------------------------|--------------------------|
| 1-1 BioSpot | 94 | 2.68E+04 | 74 |
| 1-2 BioSpot + HEPA | - | 0 | 0 |
| 1-3 BioSpot | 30 | 6.31E+03 | 18 |
| 2-1 VIVAS | 44 | 1.00E+04 | 27 |
| 2-2 VIVA S+ HEPA | - | 0 | 0 |
| 2-3 VIVAS | 16 | 2.15E+03 | 6 |

414 ^aFrom Table 2.

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417 **Figure legends**

418 Figure 1. Schematic diagram of room with depiction of patient bed and air-sampler locations.

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420 Figure 2. Cytopathic effects in Vero E6 cells inoculated with material collected from the air during air
421 sampling 1-1. [A] Mock-infected Vero E6 cells, 10 days post-inoculation with sterile collection medium.
422 [B]. Large cytoplasmic vacuoles in Vero E6 cells inoculated with collection medium from BioSpot
423 sample 1-1 at 4 dpi. [C] Early focus of infection 7 dpi. [D] Focus of infection 10 dpi. Rounded cells that
424 are detaching, some in clumps, are present. Attached cells remaining in this focus of infection have dark
425 cytoplasm, some have large cytoplasmic inclusion bodies, and some cells are elongated. Original
426 magnifications at 400X.

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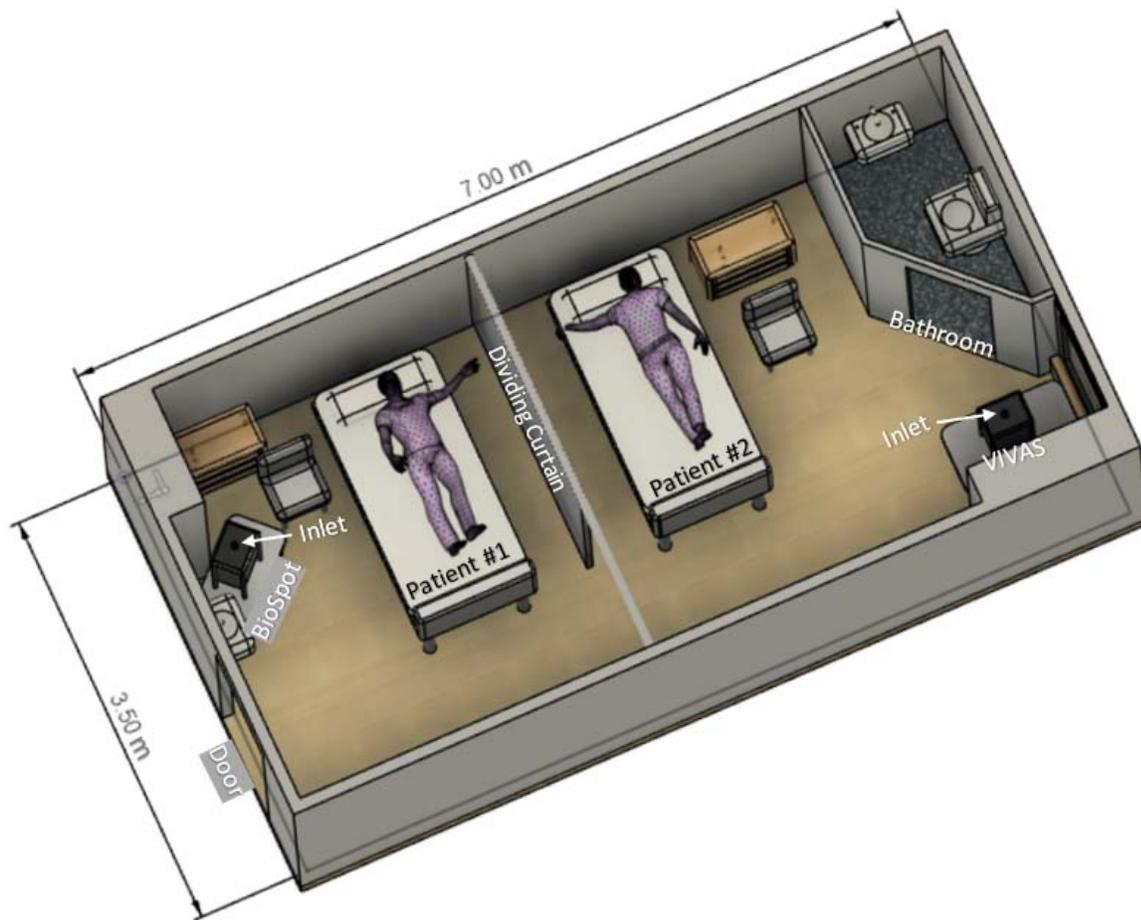
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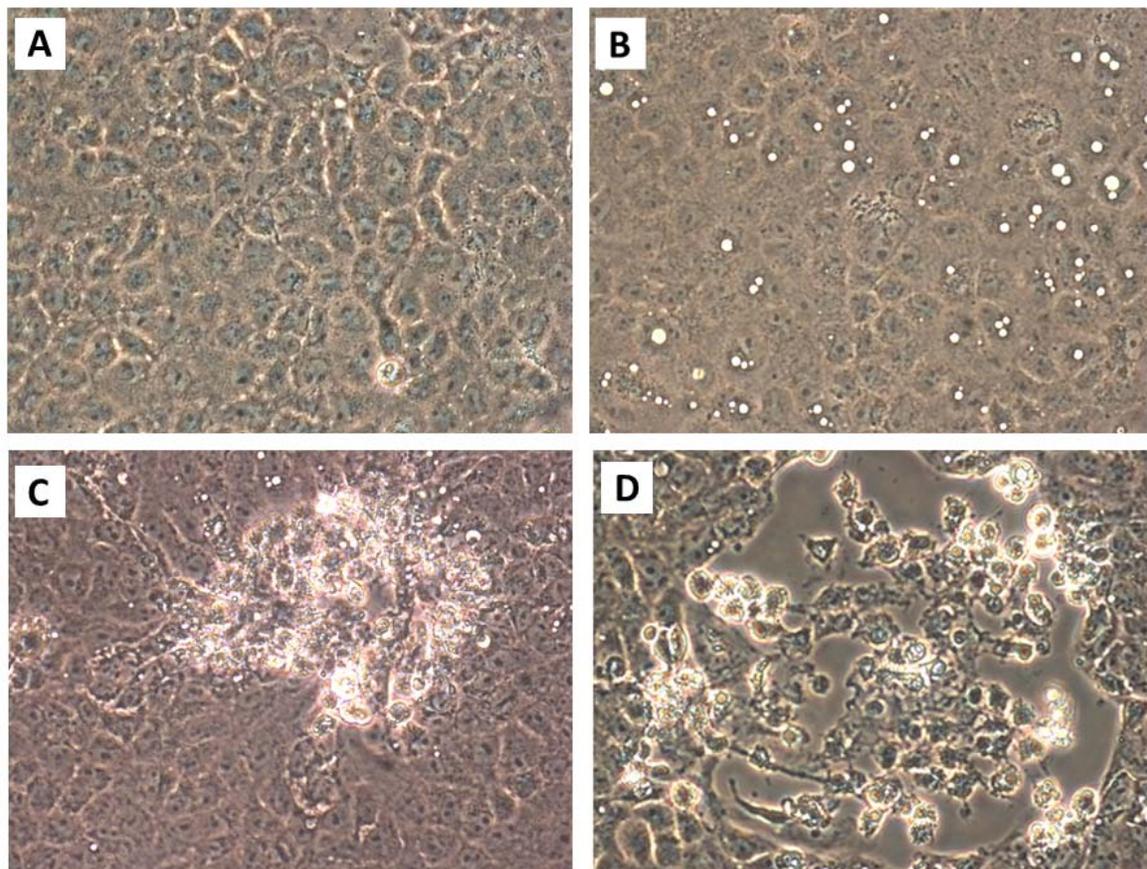
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443 Figure 1.



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