1	UV-C light completely blocks highly contagious Delta SARS-CoV-2 aerosol transmission in
2	hamsters
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24 Abstract:

- 25 Behavioral and medical control measures are not effective in containing the spread of SARS-
- 26 CoV-2. Here we report on the effectiveness of a preemptive environmental strategy using UV-C
- 27 light to prevent airborne transmission of the virus in a hamster model and show that UV-C
- 28 exposure completely prevents airborne transmission between individuals

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31 Introduction

The COVID-19 pandemic has officially caused more than 5.4 million deaths worldwide as of 32 December 28, 2021.¹ Epidemiological and experimental data suggests that the primary mode of 33 transmission of the virus is through airborne particles.²⁻⁴ Medical countermeasures, such as 34 35 vaccines and monoclonals antibody therapies were rapidly developed, but have had limited impact on the overall control of the pandemic. While the developed vaccines are highly 36 effective against preventing sever COVID-19 and hospitalization, their transmission-blocking 37 potential on population level appears limited. Currently, 44.65% of the global population are 38 fully vaccinated and an estimated 285 million people have been infected with SARS-CoV-2.1 39 This has drastically changed SARS-CoV-2 immune landscape and likely promoted the 40 emergence of Variants of Concern (VoC) escaping antibody immunity, fueling the current 41 global spikes in infection rates.⁵ These rapid increases in SARS-CoV-2 prevalence prompt crude 42 control measures such as: travel restrictions, large-scale quarantining and "lock downs" of entire 43 populations leading to economic and public health burden.⁶ The inability to control the ongoing 44 SARS-CoV-2 pandemic has put the focus on the development of pathogen agnostic non-medical 45 46 intervention strategies. These non-medical intervention strategies should ideally be practical, 47 effective under multiple conditions, not depend on the cooperation of individuals, not contribute to virus evolution and prove efficacious for multiple epidemic and pandemic pathogens. One 48 49 measure that has the potential to decrease the concentration of infectious airborne pathogens in enclosed spaces is ultraviolet (UV) light. Ultraviolet light, in particular UV-C light (wavelengths 50 in the range of 200 nm - 280 nm) has germicidal properties. Several studies have shown that 51 UV-C light can be used to inactivate SARS-CoV-2 on surfaces using a UV-C germicidal lamp.⁷⁻⁹ 52

Here we report on the effectiveness of UV-C light in blocking transmission of airborne SARSCoV-2 in a hamster model.

55 **Results**

56 To test the ability of UV-C light to prevent infection of naïve hamsters by naturally aspirated

aerosols we employed a modified version of an aerosol transmission system described

previously.⁴ In this system two cages are separated by a 1250 mm X 73 mm tube resulting in a

size exclusion of airborne particles $\geq 10 \,\mu m$. The central portion of the tube is quartz enclosed in

a HDPE box containing a UV-C light source (Figure 1). The length of the tube inside the box is

61 66.2 cm and the air traveling from the infected animals to the naïve animals had a residence time

of 10.7 seconds in the tube. A 934.5 L/hr airflow, approximately 30 cage air exchanges per hour,

is maintained throughout the experiment resulting in a UV-C dose exposure of the pathogen-

64 containing airborne particles of approximately 21.4 mJ/cm^2 .

Briefly, for each trial, 2 hamsters were inoculated intranasally (IN) with 8 X 10^4 TCID₅₀ SARS-

66 CoV-2 strain nCoV-WA1-2020 (EPI_ISL_404895) (prototype lineage A SARS-CoV-2) or

67 hCoV-19/USA/KY-CDC-2-4242084/2021 (EPI_ISL_1823618) (VoC Delta). At 1 day post

68 infection (dpi) 2 infected hamsters were placed in the upstream (donor) cage and 2 naïve

hamsters were placed in the downstream (naïve) cage. After a 4-hour exposure the exposed

naïve hamsters were moved to individual cages and the donor hamsters were euthanized after an

71 oropharyngeal swab was collected.

72 To determine whether the naïve exposed sentinel hamsters became infected, oropharyngeal

swabs were collected on days 1, 2 and 3 post exposure (DPE) and analyzed for the presence of

subgenomic viral RNA (sgRNA, marker for active SARS-CoV-2 replication) and genomic viral

75 RNA (gRNA) by qRT-PCR. The experiment was repeated 4 times for the following conditions:

76]	UV-C light treatment, no	UV-C light treatment	with variant nC	oV-WA1-2020 c	or hCoV-
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- 19/USA/KY-CDC-2-4242084/2021 (Delta). When testing under UV-C conditions, the light was
- turned on 1 hour prior to introducing the animals to the system.
- All the animals in the no UV-C treatment groups became infected as early as 1 DPE. gRNA was
- 80 detected in all animals as early as 1 DPE for both the lineage A and the Delta VOC and
- continued through DPE 3 (Figure 2, A & C). No gRNA was detected in either of the UV-C
- groups (Figure 2, A &C). sgRNA was also detected on DPE 1 3 in the no UV-C treatment
- groups, but not in any of the animals in the UV-C groups (Figure 2, B &D). To conclusively
- 84 demonstrate absence of transmission of SARS-CoV-2 in both UV-C treatment groups the
- binding antibody titers against the SARS-CoV-2 spike protein (S) were determined on sera
- obtained at 14 DPE. Both no UV-C light treatment groups had high antibody titers (\geq 52,000 in
- all animals, n = 16), but both no UV-C light treatment groups displayed a complete lack of
- binding antibody titers against SARS-CoV-2 S (<400 in all animals, n = 16).
- 89 **Discussion**

As the SARS-CoV-2 pandemic approaches its third-year, additional non-medical intervention 90 strategies are urgently needed. Especially in areas and locations where there is a higher risk of 91 92 SARS-CoV-2 transmission, such as hospitals, COVID-19 testing centers, schools and other indoor areas effective preemptive environmental intervention measures are needed to protect 93 health care workers and people at risk of developing severe COVID19. Non-medical 94 95 intervention such as social distancing rely on the assumption that small airborne respiration droplets will settle to the ground within about 2 meters from the source. However, true aerosols 96 $(< 10 \,\mu\text{m})$ in diameter will remain suspended, floating on air currents for an extended amount of 97 98 time, can travel more than 2 meters and remain suspended for minutes to hours. In addition,

other non-medical countermeasures, such as mask wearing, are highly dependent on compliance
and as such have had varying levels of effectiveness across different cultural, political, and
religious environments.

102 Here we have demonstrated that a preemptive environmental intervention measure, using UV-C irradiation, can prevent the aerosol transmission of SARS-CoV-2 between hamsters. This work 103 suggests that UV-C could be used to decrease the concentration of viable air-borne virus in 104 105 various environments used in conjunction with existing control measures and where other 106 methods are less likely to work. Extensive literature is available for pathogen inactivation, using either bacterial spore inactivation tests, bacteria or respiratory viruses by UV-C treatment.^{10,11} 107 There are several UV-C systems that have been developed and are already being employed.^{12,13} 108 The experiments described here recapitulate a system in which ducted air is treated and returned 109 110 to the room; the efficiency of this type of system is dependent on the number of room-air 111 exchanges per hour and the ventilation system processes. Another UV-C system that has been employed in areas with a high incidence of tuberculosis (TB) is upper-room ultraviolet 112 germicidal irradiation.¹⁴ Upper-room ultraviolet germicidal irradiation has the potential to treat 113 114 up to 24 room air changes per hour equivalents where comfort level ventilation systems handle between 1 and 2 room air exchanges per hour.¹⁵ 115

Preemptive environmental interventions in public spaces, that are not dependent on the compliance of the at-risk population, would potentially be a highly cost-effective non-medical countermeasure to help control the current pandemic. In addition, given the pathogen agnostic nature of UV-C germicidal irradiation it has the potential to curb airborne transmission of fungal, bacterial, and viral pathogens and even everyday maladies like the common cold.

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180	Figure 1. Experimental aerosol transmission with UV-C irradiation setup. Two cages are separated
181	with a 1250 mm X 73 mm i.d. tube. The center portion of the tube is 662 mm of UV transparent quartz
182	surrounded by a HDPE box housing a UV-C light source. Two donor hamsters, infected intranasally with
183	$8 \times 10^4 \text{ TCID}_{50} \text{ SARS-CoV-2}$ of either lineage A or the Delta variant one day prior to the experiment,
184	were placed in the upstream cage and two naïve sentinel hamsters were placed in the downstream cage
185	with a 934.5 L/hr airflow for 4 hours. The arrow indicates the direction of the airflow.
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188	Figure 2. UV-C irradiation blocks SARS-CoV-2 aerosol transmission in hamsters. A & B) Boxplot
189	(minimum to maximum) of genomicRNA and subgenomicRNA Lineage A SARS-CoV-2 RNA in
190	oropharyngeal swabs collected on 1-, 2- and 3-days post exposure. Blue dots represent the no UV-C
190 191	oropharyngeal swabs collected on 1-, 2- and 3-days post exposure. Blue dots represent the no UV-C treatment group ($n = 8$) and grey dots represent the UV-C treatment group ($N=8$). C & D) Boxplot
190 191 192	oropharyngeal swabs collected on 1-, 2- and 3-days post exposure. Blue dots represent the no UV-C treatment group ($n = 8$) and grey dots represent the UV-C treatment group ($N=8$). C & D) Boxplot (minimum to maximum) of genomicRNA and subgenomicRNA Delta SARS-CoV-2 RNA in
190 191 192 193	oropharyngeal swabs collected on 1-, 2- and 3-days post exposure. Blue dots represent the no UV-C treatment group (n = 8) and grey dots represent the UV-C treatment group (N=8). C & D) Boxplot (minimum to maximum) of genomicRNA and subgenomicRNA Delta SARS-CoV-2 RNA in oropharyngeal swabs collected on 1-, 2- and 3-days post exposure. Pink dots represent the no UV-C
190 191 192 193 194	oropharyngeal swabs collected on 1-, 2- and 3-days post exposure. Blue dots represent the no UV-C treatment group ($n = 8$) and grey dots represent the UV-C treatment group ($N=8$). C & D) Boxplot (minimum to maximum) of genomicRNA and subgenomicRNA Delta SARS-CoV-2 RNA in oropharyngeal swabs collected on 1-, 2- and 3-days post exposure. Pink dots represent the no UV-C treatment group ($n = 8$) and light-blue dots represent the represent the UV-C treatment group ($N=8$).

196 Figure 1.





198 Figure 2.



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