



## Brief Report

# Advanced photohydrolysis technology demonstrates rapid inactivation of aerosolized SARS-CoV-2 and efficacy against other respiratory viral pathogens

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Efficient, rapid means of air decontamination are needed against widespread respiratory pathogens such as SARS-CoV-2, the virus that causes COVID-19. This study demonstrated the efficacy of advanced photohydrolysis technology in significantly reducing infectious, aerosolized SARS-CoV-2, achieving over 99% viral inactivation. Proof-of-concept assessments for respiratory syncytial virus and monkeypox virus showed similar results, suggesting broad applicability. These findings highlight the potential of the novel technology to enhance air purification and infection control strategies against multiple airborne viral pathogens.

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## BACKGROUND

SARS-CoV-2, respiratory syncytial virus (RSV), and monkeypox virus (MPXV) pose major public health threats due to their modes of transmission and disproportionate impact on vulnerable populations. Aerosol transmission of SARS-CoV-2, the virus responsible for COVID-19, has been widely recognized as the primary route of infection, contributing to severe respiratory complications.<sup>1,2</sup> RSV is a leading cause of bronchiolitis and pneumonia in infants and poses a significant risk to the elderly, especially those with underlying health conditions.<sup>3</sup> MPXV, while historically associated with close contact transmission, possesses the potential for respiratory transmission and consequently has sparked new public health concerns.<sup>4,5</sup>

Advanced photohydrolysis technology (APHT) was previously shown to significantly reduce microbial loads, including pathogens like methicillin-resistant *Staphylococcus aureus*, in health care settings.<sup>6</sup> It operates through a unique photocatalytic process that harnesses a specialized matrix material activated by UV light. The process results in the continuous generation and delivery of a range of reactive oxygen species, including hydroxyl radicals, super oxygen ions, and hydrogen peroxide molecules, into the surrounding air where they interact with and inactivate environmental pathogens.<sup>6,7</sup> Unlike conventional air purification systems that rely on passive filtration, APHT continuously reduces contamination levels in real time, providing air and surface disinfection, without requiring air to pass directly through the device.<sup>6</sup> Here, we evaluated the ability of APHT to rapidly inactivate aerosolized SARS-CoV-2 and explored its effectiveness against RSV and MPXV, providing insights into applicability for improving indoor air quality and infection control.

## METHODS

The SARS-CoV-2 strain USA\_WA1/2020 used for this study was prepared from frozen seed stock (NR-596, BEI Resources) cultured in

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Vero E6 cells and harvested at 50% cytopathic effects (CPE). The RSV long strain was propagated in HEp-2 cells (ATCC VR-26) and concentrated using ultrafiltration. The MPXV strain MXV\_HUG\_2 was cultivated in Vero E6 cells from material provided by the World Reference Center for Emerging Viruses and Arboviruses (TVP 23377) and was harvested at 50% CPE.

SARS-CoV-2 was quantified using a TCID<sub>50</sub> assay.<sup>8</sup> Briefly, low-passage Vero E6 cells were used to seed 96-well plates, and serial 1:10 dilutions of viral samples were added. Cultures were incubated for 72 hours at 37 °C with 5% CO<sub>2</sub>, and CPE were observed microscopically to determine TCID<sub>50</sub>/mL, with a lower limit of detection (LLOD) of 63 TCID<sub>50</sub>/mL.

RSV and MPXV were quantified using plaque assays in HEp-2 and Vero E6 cells, respectively.<sup>9</sup> Briefly, serial 10-fold dilutions of samples were incubated at 37 °C with 5% CO<sub>2</sub> under carboxymethyl cellulose overlay. Plates were stained with crystal violet to calculate plaque-forming units (PFU/mL), with LLODs of 10 and 4 PFU/mL for RSV and MPXV, respectively.

Two devices manufactured by ActivePure Technologies, LLC were used for each pathogen: an experimental device equipped with APHT and a control device identical in design but without the photohydrolysis technology. Aerosol runs for SARS-CoV-2 were conducted in triplicate for each device, and single runs for RSV and MPXV.

Aerosols were generated using a Biaera Aero3G control platform (Biaera Technologies, LLC) fitted with a 150-L custom-made chamber. Viral aerosols were produced using a 6-jet Collision nebulizer at 14.0 LPM. Fresh 10 mL viral suspensions ( $8.3 \times 10^6$  TCID<sub>50</sub>/mL for SARS-CoV-2,  $1 \times 10^7$  PFU/mL for RSV, and  $8 \times 10^4$  PFU/mL for MPXV) were aerosolized for 15 minutes, and chamber air was sampled the last 5 minutes of aerosolization using SKC Biosamplers (SKC, Inc) at 12.5 LPM. This initial air sample was collected before the device was turned on; therefore, it gave the initial viral concentration before the device was on. Each device was turned on immediately following completion of aerosolization and collection of the initial air sample. Air samples were collected again after 1-minute with the devices on for SARS-CoV-2 and RSV, and after 5 minutes with the devices on for MPXV. The initial viral concentration and the 1-minute (for SARS-CoV-2 and RSV) or 5-minute (for MPXV) viral concentrations were used to calculate the overall reduction in viral concentration. The relative humidity was continuously monitored and ranged from 74% to 93%.

Log<sub>10</sub>-fold reductions in viral concentrations were compared between the experimental and control devices using a 1-tailed paired t test, and were expressed as base-10 (log<sub>10</sub>) values. Statistical significance was set at  $P < .05$ . Analyses were conducted using GraphPad Prism version 10.3.1.

## RESULTS

Table 1 summarizes the mean log<sub>10</sub>-fold reductions for SARS-CoV-2 for the experimental and control devices. The experimental device significantly reduced viral concentrations below or at the assay's LLOD of 63 TCID<sub>50</sub>/mL ( $1.8 \log_{10}$ ), reductions ranging from 2.37 to  $> 2.77 \log_{10}$ , corresponding to an efficiency of 99.57% to  $> 99.83\%$ . The control device showed a mean log reduction of  $0.64 \log_{10}$  which was significantly ( $P = .012$ ) less than that of the experimental device. The net log<sub>10</sub> reduction (difference between the experimental and control devices) ranged from 1.56 to  $> 2.46 \log_{10}$ , with a net efficiency of 97.37% to  $> 99.65\%$ ; however, since no virus was detected in nearly every run using the

**Table 1**

Effect of APHT on log<sub>10</sub> reductions in SARS-CoV-2 viral concentrations at T = 1 min from T = 0 min (in triplicate)

Control device	Experimental device	P-value*
0.81	$> 2.37$	$P = .012$
0.78	2.37	
0.32	$> 2.77$	

APHT, advanced photohydrolysis technology; SARS, severe acute respiratory syndrome.

\*P-value based on paired t test. Statistical significance at  $P \leq .05$ .

**Table 2**

Effect of APHT on log<sub>10</sub> reductions in RSV and MPXV viral concentrations at T = 1 or 5 min, respectively, from T = 0 min

Virus	Control device	Experimental device
RSV	1.16	$> 3.72$
MPXV	0.70	$> 2.41$

APHT, advanced photohydrolysis technology; MPXV, monkeypox virus; RSV, respiratory syncytial virus.

experimental device, the true percent reduction likely exceeded the calculated percentages.

Table 2 provides the log<sub>10</sub> reduction for RSV and MPXV (single aerosol runs for both pathogens). The experimental device reduced RSV concentrations below the assay's LLOD of 10 PFU/mL ( $1.00 \log_{10}$ ), achieving a reduction  $> 3.72 \log_{10}$  ( $> 99.98\%$  efficiency), compared to  $1.16 \log_{10}$  for the control device. The net log reduction for RSV was  $> 2.56 \log_{10}$  ( $> 99.72\%$  efficiency). In the case of MPXV, the experimental device reduced viral concentrations below the assay's LLOD of 4 PFU/mL ( $0.60 \log_{10}$ ), achieving a reduction of  $> 2.43 \log_{10}$  ( $> 99.63\%$  efficiency), compared to  $0.70 \log_{10}$  for the control. The net log reduction for MPXV was  $> 1.73 \log_{10}$  ( $> 98.14\%$ ). Similar to the SARS-CoV-2 test, the true percent reductions for RSV and MPXV likely exceeded the calculated percentages since no virus was detected after use of the experimental device.

## DISCUSSION

This study demonstrates the effectiveness of APHT in rapidly reducing ( $> 99.83\%$ ) infectious SARS-CoV-2 concentrations. The proof-of-concept studies also show its efficacy in reducing RSV and MPXV infectious titers by  $> 99.98\%$  and  $> 99.63\%$ , respectively. These results highlight APHT's potential as a public health tool for mitigating airborne pathogens, particularly in high-risk settings such as hospitals, schools, and long-term care facilities.

## CONCLUSIONS

APHT's continuous air disinfection is especially valuable in environments vulnerable to viral outbreaks, where sustained pathogen control is crucial to reducing transmission and protecting vulnerable populations. These findings are particularly relevant given the epidemiological challenges posed by airborne respiratory viruses in densely populated indoor spaces.<sup>10</sup> APHT's ability to continuously reduce infectious viral concentrations makes it a promising adjunct to existing infection control protocols, improving air quality and lowering transmission risks for at-risk populations.

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