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Appendix

PathO3Gen Solutions™

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Coronavirus Study Summary

General Details

Study title: Assessment of PathO3Gen Solutions Footwear Sanitizing Station for Decontaminating Hard, Non-Porous Environmental Surfaces (Shoes)

Organism tested: Coronavirus 229E (ATCC VR-740) - Human Coronavirus

Performing laboratory: CREM Co. Labs, Ontario, Canada

Date Performed: March 20th, 2020

Summary

The initial challenge on each carrier in **Test 1** was 3.68 log 10. The PathO3Gen Solutions' Footwear Sanitizing Station achieved the maximum attainable result of 3.68 log 10 at both 8 and 10 seconds, leaving behind <u>zero</u> plaque. Similarly, the initial challenge on each carrier in **Test 2** was 3.73 log 10, and the PathO3Gen Solutions FSS also achieved the maximum attainable result at both 8 and 10 seconds leaving <u>zero</u> plaque behind. Lastly, the initial challenge on each carrier in **Test 3** was 3.65 log 10, and the FSS also achieved the maximum attainable result at both 8 and 10 seconds leaving <u>zero</u> plaque behind.

Overall, the results were as follows:

6 seconds: 1 PFU remaining (PFU = Plaque forming unit = pathogen)

8 seconds: 0 PFU remaining 10 seconds: 0 PFU remaining

Concluding statement

"The PathO3Gen Solutions' Footwear Sanitizing Station left 0 Human Coronavirus residue on the bottom of footwear, in 8 seconds."

Modified Quantitative Disk Carrier Test Method to Determine the Virucidal Activity of an Ozone-Generating Whole-Shoe Disinfection Device



STUDY TITLE

Assessment of PathO3Gen Solutions[™] Footwear Sanitizing Station for Decontaminating Hard, Non-Porous Environmental Surfaces: Testing against Human Respiratory Coronavirus 229E (ATCC VR-740) as a representative Healthcare-Associated Pathogen

TEST ORGANISM

Coronavirus 229E (ATCC VR-740)

TEST SAMPLE IDENTITY

PathO3Gen Solutions™ Footwear Sanitizing Station

TEST Method

Modified Quantitative Disk Carrier Test Method (ASTM 2197) to Determine the Virucidal Activity of an Ozone-Generating Whole-Shoe Disinfection Device

AUTHOR

Dr. Syed A. Sattar Study Director

STUDY COMPLETION DATE

March 20, 2020

PERFORMING LABORATORY

CREM Co. Labs. Units 1-2, 3403 American Dr., Mississauga, Ontario, Canada L4V 1T4

SPONSOR

PathO3Gen Solutions™

STUDY NUMBER

PTGS200219-01

Modified Quantitative Disk Carrier Test Method to Determine the Virucidal Activity of an Ozone-Generating Whole-Shoe Disinfection Device



STUDY PERSONNEL

STUDY DIRECTOR: Syed A. Sattar, PhD

PROFESSIONAL PERSONNEL INVOLVED: Bahram Zargar, PhD

Sepideh Khoshnevis, MSC.

Modified Quantitative Disk Carrier Test Method to Determine the Virucidal Activity of an Ozone-Generating Whole-Shoe Disinfection Device



TEST SYSTEM

1. Test Microorganism

Coronavirus 229E (ATCC VR-740): Coronavirus 229E (ATCC VR-740) is an enveloped virus in the genus Coronavirus. Members of this genus can cause acute respiratory infections such as SARS-1 and SARS-2 (19-nCOV). Unlike Coronavirus 229E, SARS-1, SARS-2 and Middle-East Respiratory Syndrome (MERS) virus require Biosafety Level 3 labs. Therefore, Coronavirus 229E is frequently used as surrogate for them to assess the activity of different technologies for infection prevention and control (IPAC).

2. Host Cell Line

L-132 cells were used as hosts to support the replication and quantitation of 229E.

The cells were seeded into 12-well multi-well cell culture plates containing modified Eagle's medium (MEM) supplemented with 10% fetal bovine serum (FBS) and maintained at 36±1°C in a humidified atmosphere of 5% CO₂. Efficacy test was performed when the cell monolayer reached >90% confluency.

3. Preparation of Test Inocula

To prepare the virus for inoculation, the virus stock was mixed directly with the soil load (5% FBS). Dilution of the mixture was prepared using Earle's balanced salt solution (EBSS; pH 7.2-7.4).

TEST METHOD

1. Preparation of Test Substance

The efficacy tests were performed on PathO3Gen Solutions[™] Footwear Sanitizing Station following the instruction in the device's user manual at three exposure times (6, 8 and 10 seconds).

2. Test Procedure

A quantitative test system to closely simulate the field-application of the environmental surface decontamination process (modified quantitative carrier test – Tier 2 or QCT-2 (ASTM 2197)) was applied. The protocol was adapted to test the UV LED-based technology. Disks (2 cm diameter) from croc sole shoe were used as archetypical environmental surfaces. Sterile disks were placed on a small platform which was the same size of a shoe at three different positions (middle, back and front). The platform was taped at the bottom of the shoe. The platforms with the disks were exposed to the UV without touching the glass cover of the device. The disks were retrieved in an eluent/neutralizer immediately at the end of the exposure time. The disks were then eluted and the eluates assayed for viable virus.

Each disk on the platform was contaminated with 20 uL of the virus inoculum with a soil

Modified Quantitative Disk Carrier Test Method to Determine the Virucidal Activity of an Ozone-Generating Whole-Shoe Disinfection Device



Experimental Design

a) Input

The stock virus utilized in the testing was titrated by 10-fold serial dilutions and plaque assayed for infectivity to determine the starting titer of the virus. The results of this control were for informational purposes only.

b) Cytotoxicity Control

Prior to the test, cytotoxicity control and control for interference with virus infectivity were performed to determine if the shoe material caused any apparent degeneration (cytotoxicity) of the host cell line. Control monolayers received an equivalent volume of EBSS (without any neutralizer) only.

c) Efficacy Test

- 1. Disks (2 cm diameter) from croc sole shoes were used in testing of this method, 3 disks were assessed as control without exposure to UV.
- 2. Disks were left inside an operating BSC to dry.
- 3. Disks were inserted on a platform with the same size of the shoe at three different locations (front, middle and back).
- 4. The platform was taped to the bottom of a shoe.
- 5. The experimenter put on the shoes with the platforms at their bottom.
- 6. The experimenter stepped on the device which was already on for 10 minutes.
- 7. After the specific exposure time, the experimenters stepped out of the device.
- 8. The disks were removed from the platform and each disk was placed into a Nalgene vial containing 2 mL of an eluent.
- 9. The L-132 cells in multi-well culture plates were inoculated with 100 μL of the dilutions prepared from test and control samples. Uninfected indicator cell cultures (cell controls) were inoculated with 100 μL EBSS alone. The cultures were incubated at 33±1°C in a humidified atmosphere of 5% CO₂ for 40-44 hrs before fixing and staining them for counting the plaque-forming units (PFU).
- 10. Three control disks were included in each test to estimate the initial contamination on the platform. The test was initiated with processing one control before the processing test carriers, one in the middle of the test and ended up with the third control. This was done to take into the account the changes in the input level of the test organisms during the experiment.

DATA ANALYSIS

Calculation of Log₁₀ Reduction

Log₁₀ Reduction = Log₁₀ of average PFU from control carriers – log₁₀ of average PFU the test carriers.

Study No.: PTGS200219-01 Modified Quantitative Disk Carrier Test Method to Determine the Virucidal Activity of an Ozone-Generating Whole-Shoe Disinfection Device



TEST RESULTS

The initial challenge on each carrier were 3.68, 3.73 and 3.65 log₁₀ PFU in three different tests performed on PathO3Gen Solutions™ Footwear Sanitizing Station. Table 1 show the result of log₁₀ reduction for each contact time. In this test, the drying time of inoculated disks was reduced to 1 hr. In all contact times the log₁₀ reduction was more than 3. No plaque was detected for 8 and 10 seconds contact times.

Table 1: Virucidal Activity Test of PathO3Gen Solutions™ Footwear Sanitizing Station against Coronavirus 229E (ATCC VR-740) with three different contact times

| Contact times | | Log₁₀ Reduction in PFU | | | | | | | | | |
|---------------|---------|------------------------|---------|---------------------------|--|--|--|--|--|--|--|
| | Test #1 | Test #2 | Test #3 | Average of Three tests | | | | | | | |
| 6 seconds | 3.07 | 3.28 | 3.42 | 3.27 | | | | | | | |
| 8 seconds | 3.68 | 3.73 | 3.65 | 3.69 | | | | | | | |
| 10 seconds | 3.68 | 3.73 | 3.65 | 3.69 | | | | | | | |

Modified Quantitative Disk Carrier Test Method to Determine the Virucidal Activity of an Ozone-Generating Whole-Shoe Disinfection Device



APPENDIX

Result of efficacy test on the device with three different contact times (6, 8 and 10 seconds) against Coronavirus 229E dried on carriers representing shoe soles.

| | Test #1 | | | | | | | | | | | | | | |
|------------------|-----------|--------|-------|-----------|--------|-------|------------|--------|-------|----------|--------------|--------------|--|--|--|
| Contac t Time | 6 seconds | | | 8 seconds | | | 10 seconds | | | Control | | | | | |
| Dilution | Front | Middle | Back | Front | Middle | Back | Front | Middle | Back | C1 | C2 | C3 | | | |
| 10 ⁻⁰ | 0,1,0 | 1,1,0 | 0,1,1 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | TNTC | TNTC | TNTC | | | |
| 10 ⁻¹ | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 21,16,25 | 25,24, 25 | 11,16, 23 | | | |
| 10 ⁻² | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 3,3,3 | 2,2,2 | 2,2,2 | | | |
| 10 ⁻³ | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | | | |
| 10-4 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | | | |

C= Control TNTC= Too numerous to count

| | | | | | | Test #2 | | | | | | | |
|------------------|------------------------------------|-----------|-------|-----------|--------|---------|-------|------------|-------|----------|----------|----------|--|
| Contact Time | | 6 seconds | | 8 seconds | | | | 10 secon | ds | | Control | | |
| Dilution | Front | Middle | Back | Front | Middle | Back | Front | Middl e | Back | C1 C2 | | C3 | |
| 10 ⁻⁰ | 0,1,1 | 1,0,0 | 1,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | TNTC | TNTC | TNTC | |
| 10 ⁻¹ | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 24,25,25 | 21,22,24 | 24,20,25 | |
| 10 ⁻² | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 4,3,2 | 3,2,4 | 3,3,4 | |
| 10 ⁻³ | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | |
| 10 ⁻⁴ | 10 ⁻⁴ 0,0,0 0,0,0 0,0,0 | | | | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | |
| | | | | | | Test #3 | | | | | | | |
| Contact Time | | 6 seconds | | 8 seconds | | | | 10 seconds | | | Control | | |
| Dilution | Front | Middle | Back | Front | Middle | Back | Front | Middl e | Back | C1 | C2 | C3 | |
| 10 ⁻⁰ | 0,0,0 | 0,1,0 | 0,1,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | TNTC | TNTC | TNTC | |
| 10 ⁻¹ | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 20,22,22 | 25,25,17 | 21,19,19 | |
| 10 ⁻² | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 2,2,2 | 2,2,4 | 3,2,2 | |
| 10 ⁻³ | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | |
| 10-4 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | 0,0,0 | |

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PathO₃Gen Solutions is another strategic partner to Crothall Healthcare. They imagine a world where the number of deaths due to preventable, life-threatening infections are significantly reduced with the use of their unique patented Ozone + UVC Footwear Sanitizing Station. NSF Labs in Ann Arbor, Michigan performed an independent study and found that the Footwear Sanitizing Stations eliminated up to 99.99% of deadly infection-causing pathogens on the bottom of footwear in seconds including but not limited to Methicillin-resistant staphylococcus aureus (MRSA), Candida auris and Clostridium difficile (C.diff). In another independent study completed by CREM Co. Labs in Ontario, Canada, the Footwear Sanitizing Station completely eliminated Coronavirus in eight (8) seconds.

Nine of their Footwear Sanitizing Stations are installed at AdventHealth Connerton located in Land O' Lakes, FL. They were under-going construction and saw a 34% reduction in infections from February 2019 – July 2019. They updated their HAI rate graphs and the reduction that they experienced since February 2019 (Footwear Sanitizing Stations implementation date) is 53% through March 2020. They have always had a lower than expected HAI rate at their facility. However, since the inception of the Footwear Sanitizing Stations and other infection control measures, they have reduced their rate by 53%.

An employee satisfaction survey resulted in a 92% overall satisfaction rate. Nurses feel comfortable taking their shoes home after stepping on the stations and the public feels safer for their loved ones in the hospital. Debi Martoccio, Vice President, Chief Operating Officer stated, "The stations allow us to establish new protocols that proactively prevent infections to ensure the best outcomes for patients." They have experienced no slip and falls associated with the stations.

Jeffrey Miley, Pharm. D., CPh., Director of Pharmacy Services, AdventHealth Connerton stated: "To help improve our compliance with USP 797 and minimize the risk of pathogens contaminating our Clean room, we added to our department's action plan your PathO₃Gen Solutions Footwear Sanitizing Station. The stations are now part of our process that each employee uses prior to entering our clean room. Our last Air and Surface samples were negative for any growth in both rooms. We will continue to utilize the PathO₃Gen Solutions Footwear Sanitizing Station in our Ante room because the more tools we have to minimize risk for our patients helps us provide safer patient care."

For more information, visit www.patho3gen.com.

There are multiple studies that show the value in utilizing a footwear sanitizing station within a facility.

- An independent study of a Tampa hospital was done in 2017 by My Lien Dao, Ph.D., Associate
 Professor at the University of South Florida. The results showed a 99.59% reduction in MRSA
 and a 97.5% reduction in C.diff after stepping on the unit for 8 seconds. The study can be found
 here: https://www.patho3gen.com/wp-content/uploads/Dao_Final-Report-on-GEM-SSD-at-Hospital_9-3-2017-.pdf
- In August 2019 a study was conducted by NSF Labs International showing kill rates at 6, 8 and 10 seconds of Ozone + UVC exposure. Below are the following percentage and log reduction results at 10 seconds:

| Type of Pathogen | Pathogen Reduction at 10 Seconds | Log Reduction at 10 Seconds |
|--|-------------------------------------|--------------------------------|
| Candida auris | 99.9993% | 5.16 |
| Escherichia coli (ESBL) | 99.9989% | 4.96 |
| Klebsiella pneumoniae (CRE) | 99.9986% | 4.86 |
| Pseudomonas aeruginosa (MDRO) | 99.9970% | 4.53 |
| Methicillin resistant staphylococcus aureus (MRSA) | 99.9969% | 4.51 |
| Clostridioides difficile (C. diff) | 99.9960% | 4.40 |
| Enterococcus faecalis (VRE) | 99.9863% | 3.86 |
| | | |

 CREM Co. Labs, Ontario, Canada, conducted an independent study "Assessment of PathO3Gen Solutions Footwear Sanitizing Station for Decontaminating Hard, Non-Porous Environmental Surfaces (Shoes)" testing Coronavirus 229E (ATCC VR-740) - Human Coronavirus the EPA recommend surrogate standard for testing products to combat COVID-19. They issued a final report on March 20, 2020.

The initial challenge on each carrier in Test 1 was 3.68 log 10. The PathO3Gen Solutions' Footwear Sanitizing Station achieved the maximum attainable result of 3.68 log 10 at both 8 and 10 seconds, leaving behind zero plaque. Similarly, the initial challenge on each carrier in Test 2 was 3.73 log 10, and the PathO3Gen Solutions FSS also achieved the maximum attainable result at both 8 and 10 seconds leaving zero plaque behind. Lastly, the initial challenge on each carrier in Test 3 was 3.65 log 10, and the FSS also achieved the maximum attainable result at both 8 and 10 seconds leaving zero plaque behind.

Overall, the results were as follows:

6 seconds: 1 PFU remaining (PFU = Plaque forming unit = pathogen)

8 seconds: 0 PFU remaining 10 seconds: 0 PFU remaining

Concluding statement

"The PathO3Gen Solutions' Footwear Sanitizing Station left 0 Human Coronavirus residue on the bottom of footwear, in 8 seconds."

In the scientific article, "Evaluation of Hospital Floors as a Potential Source of Pathogen
Dissemination Using a Nonpathogenic Virus as a Surrogate Marker", (Koganti, Donskey et al) a
blinded study approved by the Cleveland Veterans Administration Hospital, demonstrated that a
nonpathogenic virus inoculated onto floors in hospital rooms disseminated rapidly to the hands
of patients and to high-touch surfaces inside and outside the patient rooms as well as on
portable equipment.

https://pdfs.semanticscholar.org/2748/bc5bf40f2215e5bf1772f368b13225b868ac.pdf



Send to: Scott Beal

Hepco Medical, LLC 200 Central Ave Ste 2200 St. Petersburg, FL 33701

Result: COMPLETE Report Date: August 27, 2019

Customer Name: Hepco Medical, LLC

Description: Efficacy of an Ozone-Generating Whole-Shoe Disinfection Device at Three Time Points

Test Type: Test Only

Job Number: J-00340388 Revised

Project Number: 10120011

NSF Corporate: C0484938

Project Manager: S. Hatt

Executive Summary: An efficacy study was performed using a UV-C and ozone-generating device against *Escherichia coli* (ESBL), *Pseudomonas aeruginosa*, Methicillin-resistant *Staphylococcus aureus*, Vancomycin-resistant *Enterococcus faecalis*, Carbapenem-resistant *Klebsiella pneumoniae*, *Candida auris*, *Aspergillus brasiliensis*, and *Clostridioides difficile*. Log and percent reduction were quantified for each microorganism at three exposure times: 6, 8, and 10 seconds.

Thank you for having your product tested by NSF International.

Please contact your Project Manager if you have any questions or concerns pertaining to this report.

Report Authorization:

Jesse Miller – Director, Applied Research Center

TEST REPORT

Experimental Summary:

Challenge microorganisms:

- Extended spectrum beta-lactamase (ESBL) Escherichia coli ATCC BAA-196
- Pseudomonas aeruginosa ATCC BAA- 2108
- Methicillin-resistant Staphylococcus aureus (MRSA) ATCC 33592
- Vancomycin-resistant Enterococcus faecalis (VRE) ATCC 51299
- Carbapenem-resistant Klebsiella pneumoniae (CRE) ATCC BAA-1705
- Candida auris CDC B11903
- Aspergillus brasiliensis ATCC 16404
- Clostridioides difficile ATCC 43598

Test Product:

PathO₃Gen Solutions[™] Footwear Sanitizing Station

Culture Preparation:

- Methicillin-resistant *Staphylococcus aureus* (MRSA), Vancomycin-resistant *Enterococcus faecalis* (VRE), *Klebsiella pneumoniae* (CRE), and *Escherichia coli* (ESBL) were propagated onto Tryptic Soy Agar with 5% Sheep Blood (SBA) and were incubated at 35 ± 2°C for 24 ± 2 hours.
- Candida auris was propagated onto SBA for 18-24 hours at 25 ± 1 °C.
 - O Daily transfers were performed using Sabouraud Dextrose Agar with Letheen (SDA/L). Each daily transfer was incubated at the appropriate temperature for growth for 24 ± 2 hours.
 - After incubation, an isolated colony was picked to SDA/L and incubated at $35 \pm 2^{\circ}$ C for 24 ± 2 hours.
- Aspergillus brasiliensis was propogated on SDA/L for 5 to 7 days. After incubation, the culture was washed with 0.85% saline with tween and filtered with through sterile gauze. The culture was centrifuged at 4,500 rpm, the supernatant removed, and the pellet was rehydrated with Phosphate Buffered Saline (PBS).
- Clostridium difficile spore suspension was prepared using a modification of the U.S. EPA OPP: MB-28 (December 2017)
 Procedure for the Production and Storage of Spores of Clostridium difficile for Use in the Efficacy Evaluation of Antimicrobial Agents based on ASTM Standard E2839-11.

Inoculation:

- Hard rubber carriers of approximately 2" x 2" were sterilized prior to testing.
- The soles of the shoe carriers were inoculated with 0.1 mL aliquot of standardized suspension of the challenge microorganism and allowed to dry for 60 ± 5 minutes.

Exposure Period:

- The disinfection device was sterilized using isopropyl alcohol prior to testing.
- After drying, the sole of the shoe carrier was aseptically placed inoculum side down onto the floor disinfection device using sterilized forceps.
- A volunteer (~150 lb) stood on the shoe carrier (with a sterile barrier between the individual and shoe carrier) for the exposure time. The instrument automatically shut off after the exposure time.
- After the exposure time, the shoe carrier was moved to a saline solution using sterile forceps. Three carriers were tested per each microorganism.
 - For bacteria, dilutions were plated via pour plate method in duplicate on Microbial Content Test Agar and incubated for 48 ± 3 hours at 35 ± 2 °C
 - o For fungi, dilutions were plated via pour plate method in duplicate on Sabouraud Dextrose Agar with Letheen and incubated for 5 to 7 days at 25 ± 2 °C
 - o For spores, dilutions were plated via spread plate method in duplicate on Brucella Blood Agar and incubated for 48 ± 3 hours at 36 ± 2 °C.
- After incubation, colonies were counted, and data recorded. Geometric mean was calculated from the duplicated plates and log and percent reduction were calculated using the positive control counts.

TEST REPORT

Table 1. Geometric mean of the inoculum concentrations on unexposed carriers used for each microorganism. The results shown are the geomean of the inoculum plated in triplicate.

| Organism | Time Point | CFU/mL | Log (CFU/mL) |
|-------------------------------------|------------|----------|--------------|
| EGDI E 1 | 6 seconds | 3.03E+07 | 7.4814 |
| ESBL <i>E. coli</i> ATCC BAA-196 | 8 seconds | 5.90E+07 | 7.7706 |
| ATCC BAA-190 | 10 seconds | 3.26E+07 | 7.5129 |
| Pseudomonas | 6 seconds | 1.52E+06 | 6.1829 |
| aeruginosa ATCC | 8 seconds | 2.21E+06 | 6.3439 |
| BAA- 2108 | 10 seconds | 1.60E+06 | 6.2037 |
| Methicillin-resistant | 6 seconds | 2.79E+07 | 7.4461 |
| Staphylococcus aureus (MRSA) | 8 seconds | 4.36E+07 | 7.6398 |
| ATCC 33592 | 10 seconds | 4.01E+07 | 7.6030 |
| Vancomycin- | 6 seconds | 7.12E+07 | 7.8528 |
| resistant <i>Enterococcus</i> | 8 seconds | 7.40E+07 | 7.8690 |
| faecalis (VRE) | 10 seconds | 4.64E+07 | 7.6664 |
| Klebsiella | 6 seconds | 1.97E+08 | 8.2944 |
| pneumoniae CRE | 8 seconds | 3.36E+08 | 8.5262 |
| ATCC BAA-1705 | 10 seconds | 2.69E+08 | 8.4302 |
| | 6 seconds | 5.16E+06 | 6.7129 |
| Candida auris CDC B11903 | 8 seconds | 1.32E+06 | 6.1219 |
| CDC B11703 | 10 seconds | 1.86E+06 | 6.2692 |
| Aspergillus | 6 seconds | 6.70E+06 | 6.8261 |
| brasiliensis ATCC | 8 seconds | 8.41E+06 | 6.9246 |
| 16404 | 10 seconds | 1.39E+07 | 7.1438 |
| Clostridioides | 6 seconds | 1.15E+07 | 7.0614 |
| difficile ATCC | 8 seconds | 1.37E+07 | 7.1380 |
| 43598 | 10 seconds | 1.24E+07 | 7.0927 |

TEST REPORT

Table 2. Carrier density for each of the carriers inoculated with ESBL *E. coli* and exposed to the disinfection device. The

results shown are the geomean of each of the carriers, which were plated in triplicate.

| Organism | Time Point | Replicate A (CFU/mL) | Replicate B (CFU/mL) | Replicate C (CFU/mL) | Geomean (CFU/mL) | Log CFU/mL | Percent Reduction | Log Reduction |
|-------------------------------------|------------|-------------------------|-------------------------|-------------------------|---------------------|---------------|----------------------|------------------|
| ESBL <i>E. coli</i> ATCC BAA-196 | 6 seconds | 3.96E+05 | 3.59E+05 | 1.29E+05 | 2.64E+05 | 5.4211 | 99.1297% | 2.06 |
| | 8 seconds | 2.95E+04 | 4.14E+04 | 3.51E+03 | 1.62E+04 | 4.2107 | 99.9725% | 3.56 |
| | 10 seconds | 1.23E+02 | 4.11E+02 | 9.23E+02 | 3.60E+02 | 2.5563 | 99.9989% | 4.96 |

Table 3. Carrier density for each of the carriers inoculated with *Pseudomonas aeruginosa* and exposed to the disinfection

device. The results shown are the geomean of each of the carriers, which were plated in triplicate.

| Organism | Time Point | Replicate A (CFU/mL) | Replicate B (CFU/mL) | Replicate C (CFU/mL) | Geomean (CFU/mL) | Log CFU/mL | Percent Reduction | Log Reduction |
|-----------------|------------|----------------------|----------------------|-------------------------|---------------------|---------------|----------------------|------------------|
| Pseudomonas | 6 seconds | 1.89E+03 | 2.84E+03 | 3.29E+03 | 2.60E+03 | 3.4157 | 99.8287% | 2.77 |
| aeruginosa ATCC | 8 seconds | 4.32E+01 | 5.15E+01 | 6.53E+01 | 5.26E+01 | 1.7207 | 99.9976% | 4.62 |
| BAA- 2108 | 10 seconds | 5.90E+01 | 6.39E+01 | 2.82E+01 | 4.74E+01 | 1.6755 | 99.9970% | 4.53 |

Table 4. Carrier density for each of the carriers inoculated with MRSA and exposed to the disinfection device. The results

shown are the geomean of each of the carriers, which were plated in triplicate.

| Organism | Time Point | Replicate A (CFU/mL) | Replicate B (CFU/mL) | Replicate C (CFU/mL) | Geomean (CFU/mL) | Log CFU/mL | Percent Reduction | Log Reduction |
|-----------------------------|------------|----------------------|----------------------|-------------------------|---------------------|---------------|----------------------|------------------|
| Methicillin Resistant | 6 seconds | 2.73E+04 | 5.25E+04 | 3.75E+04 | 3.77E+04 | 4.5768 | 99.8647% | 2.87 |
| Staphylococcus | 8 seconds | 4.90E+03 | 5.78E+03 | 1.15E+04 | 6.88E+03 | 3.8376 | 99.9842% | 3.80 |
| aureus (MRSA) ATCC 33592 | 10 seconds | 5.18E+02 | 1.15E+03 | 3.30E+03 | 1.25E+03 | 3.0978 | 99.9969% | 4.51 |

Table 5. Carrier density for each of the carriers inoculated with VRE and exposed to the disinfection device. The results

shown are the geomean of each of the carriers, which were plated in triplicate.

| Organism | Time Point | Replicate A (CFU/mL) | Replicate B (CFU/mL) | Replicate C (CFU/mL) | Geomean (CFU/mL) | Log CFU/mL | Percent Reduction | Log Reduction |
|--------------|------------|----------------------|----------------------|----------------------|---------------------|---------------|----------------------|------------------|
| Enterococcus | 6 seconds | 1.08E+06 | 1.16E+06 | 8.14E+05 | 1.01E+06 | 6.0028 | 98.5863% | 1.85 |
| faecalis VRE | 8 seconds | 5.83E+03 | 1.28E+04 | 1.29E+04 | 9.87E+03 | 3.9945 | 99.9867% | 3.87 |
| ATCC 51299 | 10 seconds | 7.62E+03 | 5.88E+03 | 5.77E+03 | 6.37E+03 | 3.8042 | 99.9863% | 3.86 |

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Table 6. Carrier density for each of the carriers inoculated with CRE and exposed to the disinfection device. The results

shown are the geomean of each of the carriers, which were plated in triplicate.

| Organism | Time Point | Replicate A (CFU/mL) | Replicate B (CFU/mL) | Replicate C (CFU/mL) | Geomean (CFU/mL) | Log CFU/mL | Percent Reduction | Log Reduction |
|----------------|------------|----------------------|----------------------|----------------------|---------------------|---------------|----------------------|------------------|
| Klebsiella | 6 seconds | 7.26E+05 | 3.86E+05 | 1.29E+06 | 7.12E+05 | 5.8527 | 99.6384% | 2.44 |
| pneumoniae CRE | 8 seconds | 8.75E+04 | 1.20E+05 | 2.72E+05 | 1.42E+05 | 5.1519 | 99.9578% | 3.37 |
| ATCC BAA-1705 | 10 seconds | 3.91E+03 | 1.32E+03 | 9.85E+03 | 3.70E+03 | 3.5687 | 99.9986% | 4.86 |

Table 7. Carrier density for each of the carriers inoculated with *Candida auris* and exposed to the disinfection device. The results shown are the geomean of each of the carriers, which were plated in triplicate. For plate count geomeans below 10

CFU/mL were input as 10 to calculate percent and log reduction.

| Organism | Time Point | Replicate A (CFU/mL) | Replicate B (CFU/mL) | Replicate C (CFU/mL) | Geomean (CFU/mL) | Log CFU/mL | Percent Reduction | Log Reduction |
|-----------------------------|------------|----------------------|----------------------|-------------------------|---------------------|---------------|----------------------|------------------|
| | 6 seconds | 1.09E+03 | 1.82E+02 | 6.08E+02 | 4.94E+02 | 2.6938 | 99.9904% | 4.02 |
| Candida auris CDC B11903 | 8 seconds | 1.46E+02 | <1.00E+01 | 2.93E+01 | 3.50E+01 | 1.5437 | 99.9974% | 4.58 |
| | 10 seconds | 2.11E+01 | <1.00E+01 | <1.00E+01 | 1.28E+01 | 1.1081 | 99.9993% | 5.16 |

Table 8. Carrier density for each of the carriers inoculated with Aspergillus brasiliensis and exposed to the disinfection

device. The results shown are the geomean of each of the carriers, which were plated in triplicate.

| Organism | Time Point | Replicate A (CFU/mL) | Replicate B (CFU/mL) | Replicate C (CFU/mL) | Geomean (CFU/mL) | Log CFU/mL | Percent Reduction | Log Reduction |
|---|------------|----------------------|----------------------|-------------------------|---------------------|---------------|----------------------|------------------|
| Aspergillus brasiliensis ATCC 16404 | 6 seconds | 1.18E+06 | 1.13E+06 | 1.08E+06 | 1.13E+06 | 6.0528 | 83.1453% | 0.77 |
| | 8 seconds | 5.29E+05 | 1.00E+06 | 1.16E+06 | 8.50E+05 | 5.9293 | 89.8956% | 1.00 |
| | 10 seconds | 3.69E+05 | 1.25E+06 | 3.26E+05 | 5.32E+05 | 5.7257 | 96.1744% | 1.42 |

Table 9. Carrier density for each of the carriers inoculated with *C. difficile* and exposed to the disinfection device. The

results shown are the geomean of each of the carriers, which were plated in triplicate.

| Organism | Time Point | Replicate A (CFU/mL) | Replicate B (CFU/mL) | Replicate C (CFU/mL) | Geomean (CFU/mL) | Log CFU/mL | Percent Reduction | Log Reduction |
|---|------------|----------------------|----------------------|----------------------|---------------------|---------------|----------------------|------------------|
| Clostridioides difficile ATCC 43598 | 6 seconds | 1.21E+05 | 8.61E+04 | 1.87E+05 | 1.25E+05 | 5.0965 | 98.9140% | 1.96 |
| | 8 seconds | 9.65E+03 | 9.62E+03 | 4.86E+03 | 7.67E+03 | 3.8848 | 99.9440% | 3.25 |
| | 10 seconds | 2.26E+03 | 1.47E+03 | 3.63E+01 | 4.94E+02 | 2.6938 | 99.9960% | 4.40 |

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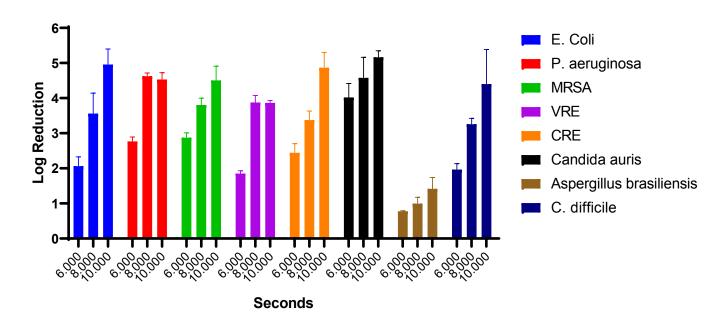
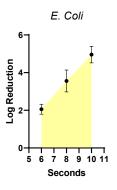
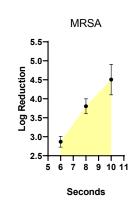
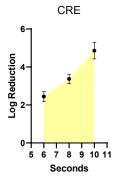


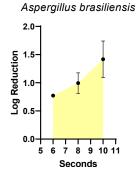
Figure 1. Summary bar plot of mean log reduction at each time point (in seconds) by microorganism tested.

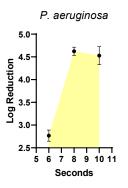
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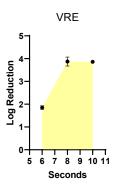


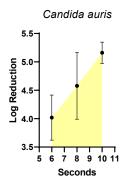












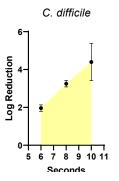


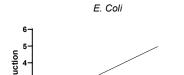


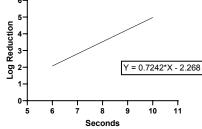
Table 10. Linear regression analysis results assessing the relationship of log reduction by time. The slopes for each line are significantly different from zero.

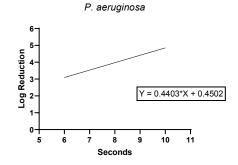
| | E. Coli | P. aeruginosa | MRSA | VRE | CRE | Candida auris | Aspergillus brasiliensis | C. difficile |
|--------------------------------------|-------------------------|--|--------------------------|--------------------------|-------------------------|-------------------------|-----------------------------|-------------------------|
| Best-fit values | | | | | | | | |
| Slope | 0.7242 | 0.4403 | 0.409 | 0.5031 | 0.605 | 0.2854 | 0.1612 | 0.6087 |
| Y-intercept | -2.268 | 0.4502 | 0.4534 | -0.8288 | -1.281 | 2.303 | -0.2276 | -1.664 |
| X-intercept | 3.132 | -1.023 | -1.109 | 1.647 | 2.117 | -8.067 | 1.412 | 2.734 |
| 1/slope | 1.381 | 2.271 | 2.445 | 1.988 | 1.653 | 3.503 | 6.204 | 1.643 |
| Std. Error | | | | | | | | |
| Slope | 0.08503 | 0.1099 | 0.05277 | 0.1138 | 0.06902 | 0.0799 | 0.04195 | 0.111 |
| Y-intercept | 0.6942 | 0.8973 | 0.4309 | 0.929 | 0.5635 | 0.6524 | 0.3425 | 0.9065 |
| 95% Confidence Intervals | | | | | | | | |
| Slope | 0.5231 to 0.9253 | 0.1804 to 0.7002 | 0.2842 to 0.5338 | 0.2340 to 0.7721 | 0.4418 to 0.7682 | 0.09649 to 0.4744 | 0.06200 to 0.2604 | 0.3462 to 0.8712 |
| Y-intercept | -3.910 to - 0.6265 | -1.672 to 2.572 | -0.5654 to 1.472 | -3.025 to 1.368 | -2.613 to 0.05183 | 0.7600 to 3.845 | -1.038 to 0.5822 | -3.808 to 0.4792 |
| X-intercept | 1.187 to 4.264 | -14.10 to 2.413 | -5.151 to 1.065 | -5.766 to 3.972 | -0.1165 to 3.427 | -39.58 to - 1.613 | -9.251 to 4.045 | -1.366 to 4.428 |
| Goodness of Fit | | | | | | | | |
| R square | 0.912 | 0.6963 | 0.8956 | 0.7364 | 0.9165 | 0.6458 | 0.6784 | 0.8111 |
| Sv.x | 0.4165 | 0.5384 | 0.2585 | 0.5574 | 0.3381 | 0.3914 | 0.2055 | 0.5439 |
| Is slope significantly non- zero? | | | | | | | | |
| F | 72.54 | 16.05 | 60.08 | 19.55 | 76.83 | 12.76 | 14.77 | 30.06 |
| DFn, DFd | 1, 7 | 1, 7 | 1, 7 | 1, 7 | 1, 7 | 1, 7 | 1, 7 | 1, 7 |
| P value | < 0.0001 | 0.0051 | 0.0001 | 0.0031 | < 0.0001 | 0.0091 | 0.0064 | 0.0009 |
| Deviation from zero? | Significant | Significant | Significant | Significant | Significant | Significant | Significant | Significant |
| Equation I-00340388 1 Rev | Y = 0.7242*X - 2.268 | Y = 0.4403*X + 0.4502 Page 9 of 11 | Y = 0.4090*X + 0.4534 | Y = 0.5031*X - 0.8288 | Y = 0.6050*X - 1.281 | Y = 0.2854*X + 2.303 | Y = 0.1612*X - 0.2276 | Y = 0.6087*X - 1.664 |

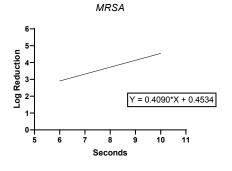
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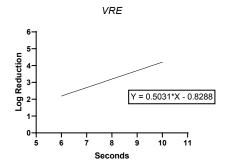


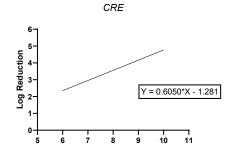


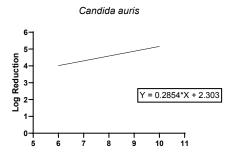


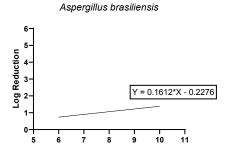


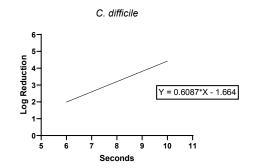












All work performed at:

TEST REPORT

Testing Laboratories:

Lab ID

Approved Subcontract

Note GLP, non-GLP compliant

