IG MicroMed Environmental Inc.
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CERTIFICATE OF ANALYSIS

Attention: Daniel Shin
Monos Travel Ltd
1383 West 8th Avenue
Vancouver, BC
V6H 3W4

16 June, 2020

Phone: [redacted]

Reference No: 399818 Salmonella.

Please find attached the results of the sample received May 28.

RE: CleanPod UVC Sterilizer

Test: Assessment of Antimicrobial Efficacy of the Monos CleanPod UVC Sterilizer against Salmonella typhimurium using Hard Surface Carriers

Method: Non-GLP ASTM E2315

Jim Wilson, B.Sc.
Microbiologist

JW/al

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Assessment of Antimicrobial Efficacy of the Monos CleanPod UVC Sterilizer against *Salmonella typhimurium* using Hard Surface Carriers.

Client: Daniel Shin, Monos Travel Ltd.
1383 West 8th Avenue, Vancouver, BC V6H 3W4

Date: 8th June 2020

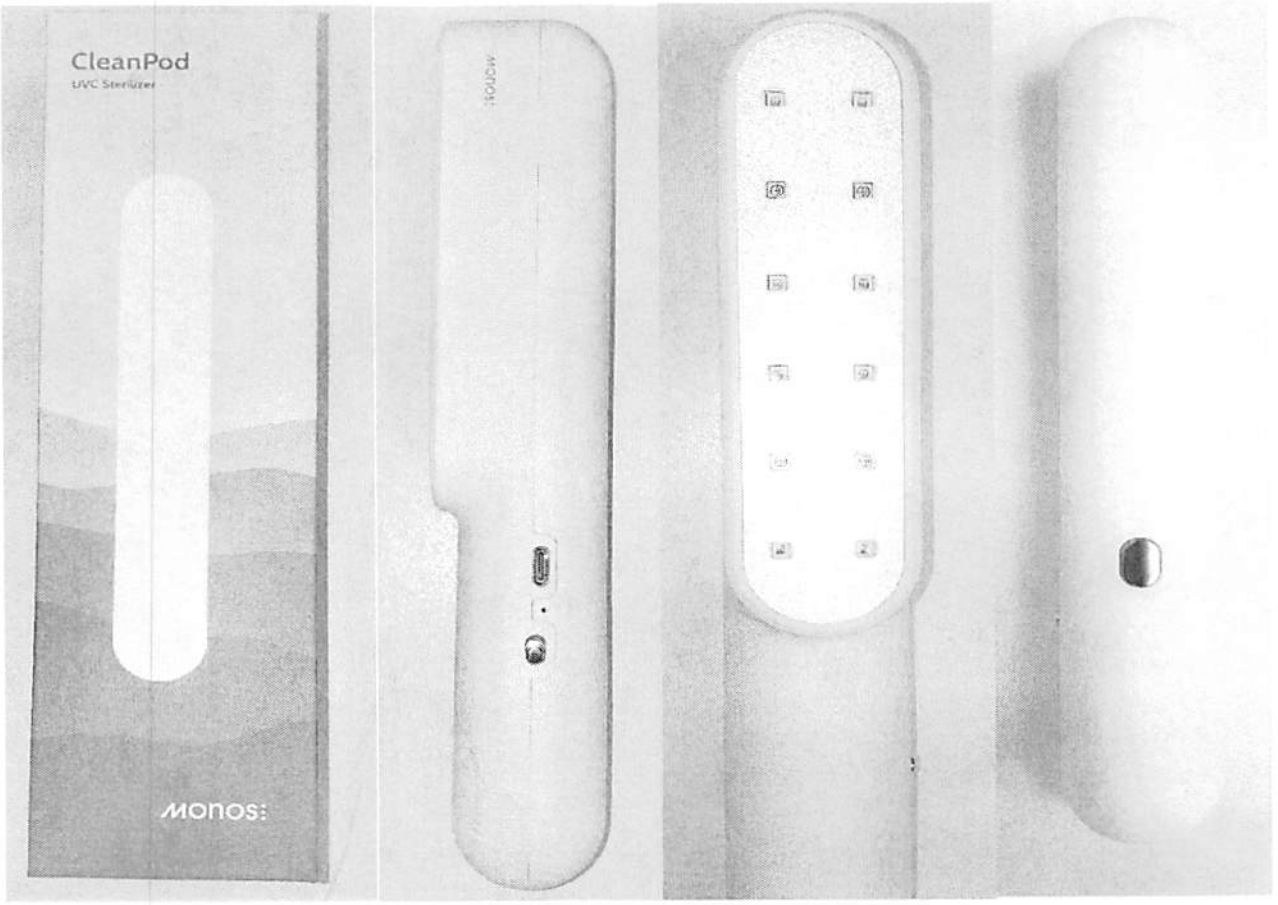
monos:

Method: Non-GLP ASTM E2315.

Reference No: 399818 Salmonella

Analyst: Usman Qazi, *Senior Laboratory Technician.*
Jim Wilson, *Laboratory Manager.*

Product: CleanPod UVC Sterilizer



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Experimental Report:-

A Non GLP Time-based study using the ASTM E2315 Method was conducted on 2nd June 2020 to determine the Antibacterial efficacy of the MONOS CleanPod UVC Sterilizer against *Salmonella typhimurium* ATCC 14028. Glass Slide Carriers were used to represent non porous hard surfaces. On the clients request, 6 replicates were tested at Time Points 30 seconds, 1 minute and 3 minutes. 3 replicates were tested at 2 minutes.

Inoculum Preparation: -

24 Hour cultures of *S. typhimurium* were grown in sterile Tryptic Soy Broth at 35°C. The cultures was enumerated by serial dilution with 0.1% peptone up to 10^{-7}. The bacterial titre was determined to be approximately 1 x 10^9 CFU/ml.
The Glass slide carriers were sterilized by wiping dry slides with 70% Iso-Propyl Alcohol and allowing them to dry. Inoculation was performed by pipetting a 30 µl inoculum of bacteria from the stock culture bag on to a 25 mm x 75 mm glass slide placed on a piece of Whatman filter paper in a petri dish. The inoculum was spread evenly on the slide surface using a sterile inoculating loop, and dried by incubating for 45 minutes inside a 35°C incubator.

Method Verification: -

The use of glass slides as an acceptable hard surface carrier was verified by inoculating 4 slides each with cultures of *S. typhimurium* using the method described above. The slides were divided into 3 groups, Untreated Blank Slides, 2.5 min UV Treated Slides and Untreated Inoculated slides. Following treatment the slides were submerged in 100 mL 0.1% Sterile Peptone Diluent in Sterile Plastic Bags and Incubated at Room Temperature for 30 minutes with constant shaking. The carriers were enumerated by serially diluting up to 10^{-2} and plating 1 mL of each dilution with Tryptic Soy Agar (TSA). The plates were incubated at 35°C for 48 hours. No colonies were observed on the plates corresponding to the Untreated Blank Slides, and UV Treated Slides. Excess colonies were obtained for the Untreated Inoculated Slides. The results of this trial verified the experimental design and the sterilization procedure for the slides.

Procedure: -

Glass slide carriers were prepared according to the procedure provided above under *Method Verification*. 7 slide carriers, 1 positive control and 6 experimental, were prepared for the 30 second, 1 minute, and 3

minute time points. 4 slide carriers, 1 positive control and 3 experimental, were prepared for the 2 minute time point. UV treatment was performed by holding the UVC Sterilizer 3 cm above the slide carriers and gradually moving it back and forth for the duration of the exposure. The slides were transferred to sterile bags containing 80 mL of 0.1% Peptone using flame sterilized forceps and incubated at room temperature for 45 minutes with constant shaking at 60 rpm. The 30 second, and 1 minute treatment groups were serially diluted up to 10^{-2} and plated with TSA. The 2 minute and 3 minute treatment groups were diluted to 10^{-1} and plated. The plated were incubated at 35°C for 48 hours.

Results:

The results of the UV treatment of *S. typhimurium* glass carriers using the UVC sterilizer at 30 seconds, 1 minute, 2 minute, and 3 minute are provided in Table 1. The detection limit of the procedure was <1 CFU/mL as countable colonies were also obtained from the undiluted 1 mL aliquots.

Conclusion:

The results of the study demonstrate that there is a progressive reduction in the counts of *S. typhimurium* on the glass slide carriers with increased exposure time to the UVC Sterilizer. A percentage reduction greater than 99% was observed for both organisms at 1 minute, 2 minute and 3 minutes UVC exposure. The greatest reduction for *S. typhimurium* carriers was observed at 1 minute. A count of 11 CFU was obtained for *S. typhimurium* Replicate 1 at 2 minutes. This count was greater than the Replicate 1 count at 1 minute, and can possibly be attributed to contamination.

Reviewed by:

Jim Wilson, B.Sc.
Laboratory Manager.
I.G.MicroMed Environmental Inc.

GE

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Table 1 - Effect of UV Exposure on the growth of *Salmonella typhimurium* on Glass Slide Carriers at 4 time points.

Exposure Time	30 seconds			1 minute			2 minute			3 minute		
	CFU	Log Reduction	% Reduction	CFU	Log Reduction	% Reduction	CFU	Log Reduction	% Reduction	CFU	Log Reduction	% Reduction
Replicate 1	<1	2.90	99.99	<1	3.0	99.99	11	2.56	99.72	<1	2.60	99.99
Replicate 2	<1	2.90	99.99	<1	3.0	99.99	<1	3.60	99.98	<1	2.60	99.99
Replicate 3	4	2.30	99.50	<1	3.0	99.89	<1	3.60	99.99	<1	2.60	99.99
Replicate 4	4	2.30	99.50	<1	3.0	99.99				<1	2.60	99.99
Replicate 5	30	1.43	96.25	<1	3.0	99.99				<1	2.60	99.99
Replicate 6	30	1.43	96.25	<1	3.0	99.99				<1	2.60	99.99
Positive Control	8×10^2 CFU/ml			1×10^3 CFU/ml			4×10^3 CFU/ml			4×10^2 CFU/ml		