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(510) 887-8885 | info@harrenslab.com

3507 BREAKWATER DRIVE | HAYWARD, CA 94545

Client: LARQ

Invoice Number: IN2024020047 - V2 (Updated the wording for this report)

Sample Number: HR20240290135, HR20240290136

Study: Antimicrobial efficacy of the LARQ Bottles against *Salmonella*

Method: ASTM E2315

Report Date: 02/14/2024

Certificate of Analysis

Experimental Summary:

The objective of this procedure was to test the antimicrobial efficacy of LARQ's UV-C LED technology within the LARQ Bottle against water samples enriched with *Salmonella enterica* subsp. *Typhimurium*. The testing procedure was designed after discussions between LARQ and Harrens Lab Inc. and based on ASTM E2315 ("Standard Guide for Assessment of Antimicrobial Activity Using a Time-Kill Procedure") testing guidelines. The testing procedure was conducted at Harrens Lab Inc. in Hayward, CA.

Methods and Materials:

LARQ provided 2 stainless steel bottles of two different volumes (680mL, 1000mL) and one UV-C LED cap for the testing. All bottles were washed with sterile deionized water prior to testing. *Salmonella enterica* subsp. *Typhimurium* (ATCC 14028) was used as the testing organism in this experiment and spiked into DI water to create a starting solution of about $1.0E+07$ CFU/mL to be treated. Testing was done in 3 replicates for 1-min (Normal Activation Mode), 2-min (Maintenance Mode) and 3-min (Adventure Mode) tests. Inoculated volumes for each run was 600mL to be tested in the 680mL stainless steel bottle with the UV-C cap and 900mL to be tested in the 1000mL stainless steel bottle with the UV-C cap. Pre and post treatment aliquots of the culture suspensions were plated in serial dilutions ranging from 10^{-1} to 10^{-8} on APC media. Plates were incubated for 48 hours at 30-35°C.

Figure 1 and 2: LARQ Bottles with cap on and off



Results:

Table 1: Experimental results using 1-min (Normal Activation Mode) UV-C treatment against *Salmonella*

Bottle	Initial Population (CFU/mL)	T-1 min (CFU/mL)	Log Reduction (T1)	% Reduction (T1)
680 mL	2.5E+07	1.0E+01	6.398	99.9999%
	3.1E+07	1.0E+01	6.491	99.9999%
	3.2E+07	1.0E+01	6.505	99.9999%
1000 mL	3.8E+07	4.0E+02	4.978	99.999%
	4.0E+07	2.9E+02	5.14	99.999%
	4.6E+07	3.5E+02	5.119	99.999%

Comment: No growth was detected on 680 mL plates so a value of 10 was used to indicate the detection limit (< 10 CFU).

Table 2: Experimental Results using 2-min (Maintenance Mode) UV-C treatment against *Salmonella*

Bottle	Initial Population (CFU/mL)	T-2 min (CFU/mL)	Log Reduction (T2)	% Reduction (T2)
680 mL	3.4E+07	1.0E+01	6.531	99.9999%
	3.7E+07	1.0E+01	6.568	99.9999%
	3.3E+07	1.0E+01	6.519	99.9999%
1000 mL	4.1E+07	1.0E+01	6.613	99.9999%
	4.3E+07	1.0E+01	6.633	99.9999%
	3.9E+07	1.0E+01	6.591	99.9999%

Comment: No growth was detected on 2-min (Maintenance Mode) treated plates so a value of 10 was used to indicate the detection limit (< 10 CFU).

Table 3: Experimental Results using 3-min (Adventure Mode) UV-C treatment against *Salmonella*

Bottle	Initial Population (CFU/mL)	T-3 min (CFU/mL)	Log Reduction (T3)	% Reduction (T3)
680 mL	3.9E+07	1.0E+01	6.591	99.9999%
	4.1E+07	1.0E+01	6.613	99.9999%
	4.2E+07	1.0E+01	6.623	99.9999%
1000 mL	5.1E+07	1.0E+01	6.708	99.9999%
	5.5E+07	1.0E+01	6.74	99.9999%
	5.3E+07	1.0E+01	6.724	99.9999%

Comment: No growth was detected on 3-min (Adventure Mode) treated plates so a value of 10 was used to indicate the detection limit (< 10 CFU).

Conclusion:

The purpose of this study was to determine how effective a LARQ Bottle was at killing *Salmonella Typhimurium* at 1-min (Normal Activation Mode), 2-min (Maintenance Mode) and 3-min (Adventure Mode) treatments. Tables 1, 2 and 3 show that the LARQ bottles produced detectable log reductions of *Salmonella Typhimurium* at 1-min (Normal Activation Mode), 2-min (Maintenance Mode), and 3-min (Adventure Mode) treatments. Table 1 shows that at 1-min (Normal Activation Mode) treatments, the LARQ bottles yielded a common log reduction of 5.08, 6.46, and killed 99.999% of *Salmonella Typhimurium*. Table 2 shows that at 2-min (Maintenance Mode) treatments, the LARQ bottles yielded a common log reduction of 6.54, 6.61 and killed 99.9999% of *Salmonella Typhimurium*. Table 3 shows that at 3-min (Adventure Mode) treatments, the LARQ bottles yielded a common log reduction of 6.61, 6.72 and killed 99.9999% of *Salmonella Typhimurium*. The 3-min (Adventure Mode) treatment produced the greatest log reduction and percent reduction against *Salmonella*.

Respectfully Submitted,



Ming Li
 General Manager
 Harrens Lab Inc.

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