ANTIMICROBIAL TEST LABORATORIES

Microbiology Study Report NG4569

Client Information

Company Name:	CitroBio, Inc.	Sponsor:	R. Maguire
Phone:	(941) 359-1647	E-mail:	lina@citrobio.com

Test Information

Test(s) Performed:	Suspension Time Kill (ASTM E2315)		
SOP Followed:	Testing Facility Operation 026.1	Performed by:	D. Sowersby

Test Substance Information

Date Received:	04 OCT 2013	Test Substances:	CB Original Formula

Parameters

Microorganism :	*S. enterica* ATCC 10708 (SE); *L. monocytogenes* ATCC 15313 (LM); *E. coli* ATCC 8739 (EC)		
Subculture #:	1- overnight culture (18-24 hours)	Exposure Temp.	Room Temperature
Growth Medium:	Tryptic Soy Broth (TSB) (SE & EC)	Type of Sample:	Liquid (10 mL)
	Brain-Heart Infusion (BHI) (LM)	Neutralizer Used:	Dey/Engley (D/E) Broth, 9.0 mL
Inoculum Volume:	25 µL (SE); 50 µL (LM); 12 µL (EC)	# of Replicates:	Singlet
Enumeration Medium:	Tryptic Soy Agar (TSA)	Plate Incubation:	36.0 ± 1
Incubation Time:	24 hours (SE & EC); 48 hours (LM)	Contact Times:	15 minutes

Controls

Neutralized:	N/A	Growth Control:	N/A
Broth Sterility:	Passed	Agar Sterility:	Passed

Test Results

Test(s) Valid?:	See Results	Confirmation:	Morphology on TSA

Notes: The test substance was diluted with water to a final concentration of 400 ppm before use. A 10 ml volume of diluted test substance was aliquoted into a sterile vessel which was then inoculated with respective microorganisms directly from overnight working cultures. The resulting mixture was vortexed thoroughly and incubated at room temperature for the contact time described above whereupon a 1.0 ml aliquot was then harvested in 9.0 ml of D/E broth. Surviving test organisms were enumerated per standard dilution and plating techniques. CFU reductions were compared to an RO water control at time zero.

Tests Completed:	10 OCT 2013	**Report Sent:**	11 OCT 2013

Antimicrobial Test Laboratories
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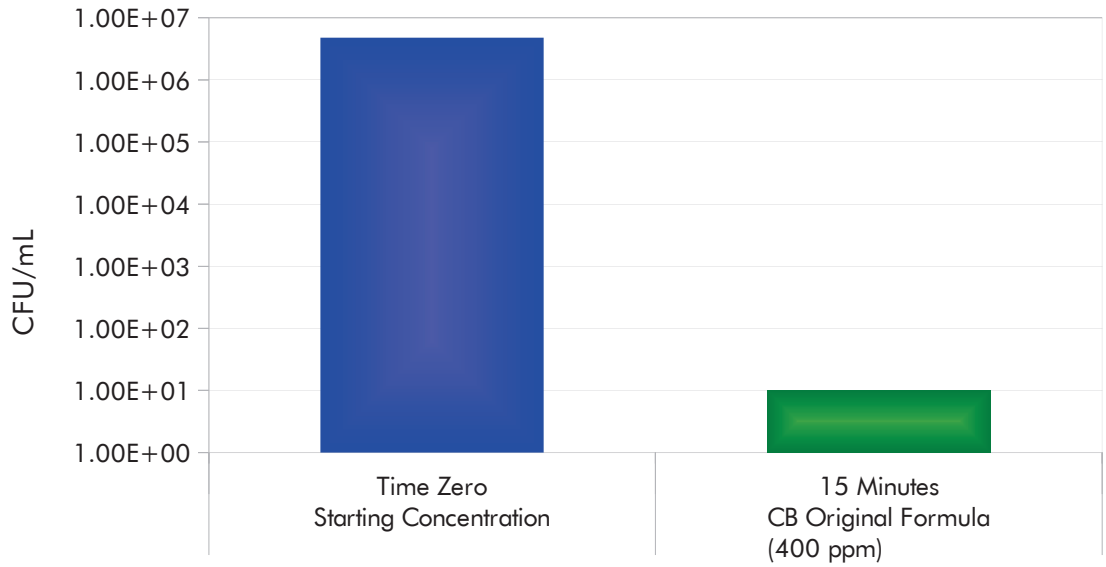
Results (*S. enterica*)

Microorganism	Sample ID	Time Point	CFU/ml	Percent Reduction	Log_{10} Reduction
S. enterica ATCC 10708	Starting Concentration	Time Zero	4.75E+06	N/A	
	CB Original Formula (400 ppm)	15 Minutes	1.00E+01	99.9998%	5.68



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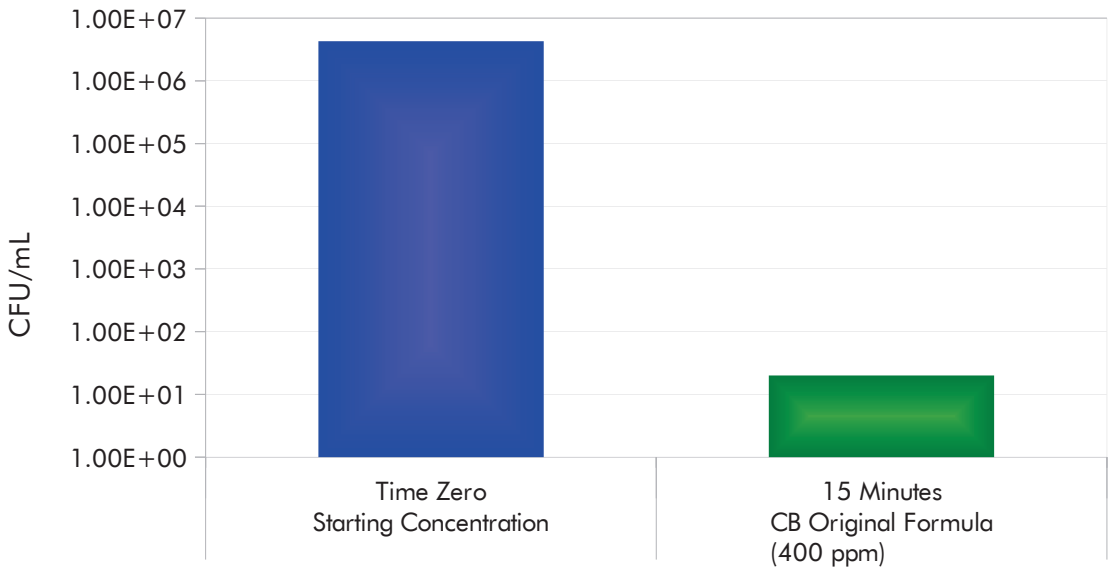
Results (*L. monocytogenes*)

Microorganism	Sample ID	Time Point	CFU/ml	Percent Reduction	Log_{10} Reduction
L. monocytogenes ATCC 15313	Starting Concentration	Time Zero	4.30E+06	N/A	
	CB Original Formula (400 ppm)	15 Minutes	2.00E+01	99.9995%	5.33



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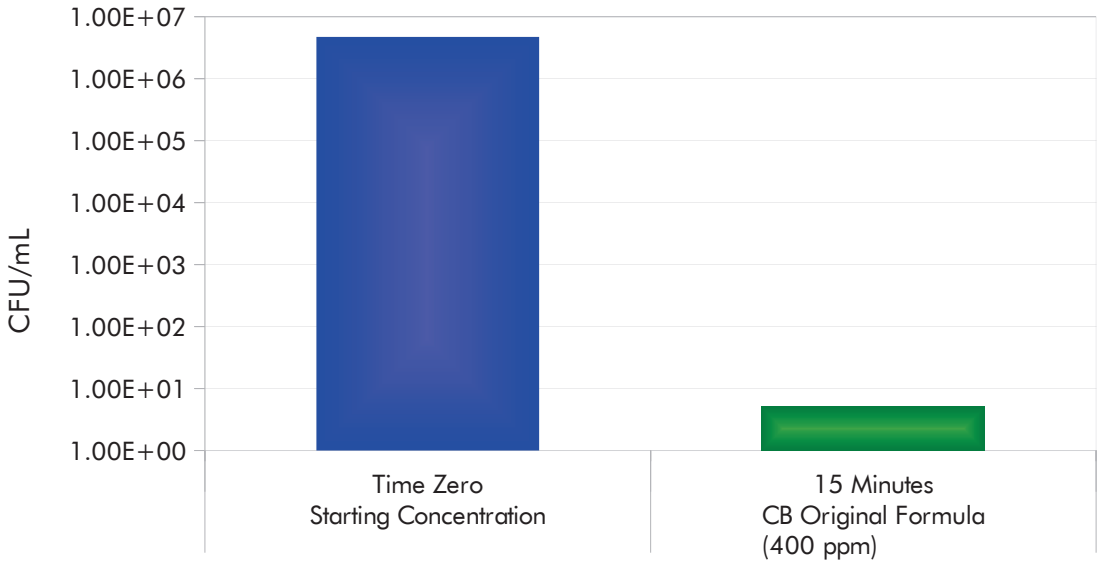
Results (*E. coli*)

Microorganism	Sample ID	Time Point	CFU/ml	Percent Reduction	Log_{10} Reduction
E. coli ATCC 8739	Starting Concentration	Time Zero	4.75E+06	N/A	
	CB Original Formula (400 ppm)	15 Minutes	5.00E+00	99.9999%	5.98



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Additional Information

Sample Calculations

% Reduction = 100 x (B-A)/B

Where:

A = Number of viable test microorganisms in sample after the contact time

B = Number of viable test microorganisms in the untreated sample after immediately after inoculation

Log Reduction = log(B-A)

Where:

A = Number of viable test microorganisms in sample after the contact time

B = Number of viable test microorganisms in the untreated sample after immediately after inoculation

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