

CLEANING LABORATORY EVALUATION SUMMARY

SCL #: 2021
 DateRun: 03/24/2021
 Experimenters: Aditi Patel
 ClientType: Cleaner Manufacturer
 ProjectNumber: Project #2
 Substrates: Stainless Steel
 PartType: Coupon
 Contaminants: Staphylococcus aureus ATCC 6539, E.coli ATCC 29214, Bacteria - Gram Negative, Bacteria - Gram Positive
 Cleaning Methods: Low Pressure Spray
 Analytical Methods: Organism count
 Purpose: To test the disinfection efficacy of Eco Green Heavy Duty Degreaser on stainless steel.

Experimental Procedure: Spread Plating Method - E.coli 29214 and S. aureus 6538
 Twenty-four hours prior to one run (26 plates), bacteria were sub-cultured into three milliliters of tryptic soy broth (TSB) screw-cap tubes and incubated at 37°C (98.6°F). Four glass Petri dishes, each containing one stainless steel coupon, were autoclaved. The biosafety cabinet (BSC) was sprayed with 70% v/v isopropyl alcohol using a paper towel before spraying any items going into the BSC. After autoclaving, the four glass Petri dishes were marked using a black sharpie to designate the positive (P+), negative (N-), Test 1 (T1), and Test 2 (T2). Ten microliters of the organism were pipetted onto the P+, T1, and T2 stainless steel coupons and air-dried for 15 minutes. A motorized pipette with 10ml tips was used to pipet 15 ml of D/E neutralizing broth into four separate 50ml conical tubes labeled P+, N-, T1, and T2. Once the bacteria dried on the coupons, the P+ coupon was placed into the conical tube. The N-, T1 and T2 were pipetted with 1000µl of the cleaner onto each coupon for 10 minutes before immediately placing them in the conical tube with an autoclaved forceps. The conical tubes were then placed on the shaker for 10 minutes. During this time, using the 1000µl pipette, 900µl of was pipetted into nine autoclaved dilution tubes, and serial dilutions were made for P+, T1, and T2 up to 10⁻⁴ using 100µl of the shaken D/E broth. Using a pipette, 100µl of the stock and serial dilutions were each plated and spread evenly over the surface of the solid TSA petri dish plate using a metal spreader. Finished plates were placed into a clean labeled zip lock bag that was partially closed and incubated at 37°C overnight. Isolated colonies were counted the following day to calculate log reduction and percent removal.

Results:	Organism	Positive Plates Average Log	Average Log Reduction	Average % Reduction
	E.coli 29214	6.44	6.44	100
	S. aureus 6538	5.43	3.765	99.5979

Summary:	Substrates:	Stainless Steel				
	Contaminants:	Staphylococcus aureus ATCC 6539, E.coli ATCC 29214, Bacteria - Gram Negative, Bacteria - Gram Positive				
	Company Name:	Product Name:	Conc.:	Efficiency:	Effective:	Observations:
	A & C Green Cleaner LLC	A & C Eco Green Heavy Duty Degreaser	100%	100.00	<input checked="" type="checkbox"/>	E.coli 29214
	A & C Green Cleaner LLC	A & C Eco Green Heavy Duty Degreaser	100%	99.60	<input type="checkbox"/>	S. aureus 6538

Conclusion: Eco Green Heavy-Duty Degreaser was effective at disinfecting E.coli 29214 within 10 minutes, but it was not effective at disinfecting S.aureus 6538 within 10 minutes on a hard, clean surface.