AMENDMENT TO GLP TEST PROTOCOL

ATS LABS

Amendment No.:	1
Effective Date:	6/19/13
Sponsor:	Sherwin-Williams Diversified Brands 101 W. Prospect Ave. Cleveland, OH 44115-1075
Test Facility:	ATS Labs 1285 Corporate Center Drive, Suite 110 Eagan, MN 55121
Protocol Title:	Test Method Two for Determining Efficacy
ATS Labs Protocol Number:	SHE09041613.CCR.2
ATS Labs Project Number:	A14895

Modifications to Protocol:

Per Sponsor's request, the protocol is amended to update the study title and Sponsor address as follows:

Study Title: Original: "Test Method for the Continuous Reduction of Organisms on Antimicrobial Coated Surfaces"
New: "Test Method Two for Determining Efficacy"

Original Sponsor Address: Sherwin Williams Breen Technical Center
601 Canal Road
Cleveland, OH 44113

New Sponsor address: Sherwin-Williams Diversified Brands
101 W. Prospect Ave.
Cleveland, OH 44115-1075

Changes to the protocol are acceptable as noted.

Matthew Dotts
Study Director

6-20-13
Date

EXACT COPY
INITIALS MS DATE 6-24-13

(For Laboratory Use Only)
ATS Labs Project # A14895
6-24-13

ATS LABS

PROTOCOL

Test Method for the Continuous Reduction of Organisms on Antimicrobial Coated Surfaces

Test Organism:

Enterobacter aerogenes (ATCC 13048)

PROTOCOL NUMBER

SHE09041613.CCR.2

PREPARED FOR

Sherwin Williams Breen Technical Center
601 Canal Road
Cleveland, OH 44113

PERFORMING LABORATORY

ATS Labs
1285 Corporate Center Drive, Suite 110
Eagan, MN 55121

PREPARED BY

Matthew Sathe, B.S.
Senior Microbiologist

DATE

April 16, 2013

PROPRIETARY INFORMATION

THIS DOCUMENT IS THE PROPERTY OF AND CONTAINS PROPRIETARY INFORMATION OF ATS LABS. NEITHER THIS DOCUMENT, NOR INFORMATION CONTAINED HEREIN IS TO BE REPRODUCED OR DISCLOSED TO OTHERS, IN WHOLE OR IN PART, NOR USED FOR ANY PURPOSE OTHER THAN THE PERFORMANCE OF THIS WORK ON BEHALF OF THE SPONSOR, WITHOUT PRIOR WRITTEN PERMISSION OF ATS LABS.

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Test Method for the Continuous Reduction of Organisms on Antimicrobial Coated Surfaces

SPONSOR

Sherwin Williams Breen Technical Center
601 Canal Road
Cleveland, OH 44113

TEST FACILITY

ATS Labs
1285 Corporate Center Drive, Suite 110
Eagan, MN 55121

PURPOSE

The purpose of this study is to determine the effectiveness of antimicrobial coated surfaces to continuously reduce test organism contamination after multiple re-inoculations over extended time periods.

TEST SUBSTANCE CHARACTERIZATION

Test substance characterization as to content, stability, etc., (40 CFR, Part 160, Subpart F [160.105]) is the responsibility of the Sponsor. The test substance shall be characterized by the Sponsor prior to the experimental start date of this study. Pertinent information, which may affect the outcome of this study, shall be communicated in writing to the Study Director upon sample submission to ATS Labs.

SCHEDULING AND DISCLAIMER OF WARRANTY

Experimental start dates are generally scheduled on a first-come/first-serve basis once ATS Labs receives the Sponsor approved/completed protocol, signed fee schedule and corresponding test substance(s). Based on all required materials being received at this time, the proposed experimental start date is April 23, 2013. Verbal results may be given upon completion of the study with a written report to follow on the proposed completion date of May 21, 2013. To expedite scheduling, please be sure all required paperwork and test substance documentation is complete/accurate upon arrival at ATS Labs.

If a test must be repeated, or a portion of it, due to failure by ATS Labs to adhere to specified procedures, it will be repeated free of charge. If a test must be repeated, or a portion of it, due to failure of internal controls, it will be repeated free of charge.

If the Sponsor requests a repeat test, they will be charged for an additional test.

Neither the name of ATS Labs nor any of its employees are to be used in advertising or other promotion without written consent from ATS Labs.

The Sponsor is responsible for any rejection of the final report by the regulatory agencies concerning report format, pagination, etc. To prevent rejection, Sponsor should carefully review the ATS Labs final report and notify ATS Labs of any perceived deficiencies in these areas before submission of the report to the regulatory agency. ATS Labs will make reasonable changes deemed necessary by the Sponsor, without altering the technical data.

JUSTIFICATION FOR SELECTION OF THE TEST SYSTEM

The U.S. Environmental Protection Agency requires that specific antibacterial claims be supported by appropriate scientific data demonstrating the efficacy of the product, in this case an antimicrobial surface, against the claimed bacteria. This is accomplished in the laboratory by inoculating and reinoculating the antimicrobial surface with the target bacteria under conditions which simulate as closely as possible, the actual conditions under which the product is designed to be used. In this instance, the coating is intended to be applied to wall surfaces. The test system to be used in this study will follow a modification of the EPA approved protocol entitled "Test Method for the Continuous Reduction of Bacterial Contamination on Copper Alloy Surfaces". It should be noted that these protocols are typically reviewed on a case by case basis for regulatory compliance by the chosen regulating agencies. It is the Sponsor's responsibility to ensure compliance.

TEST PRINCIPLE

Antimicrobial coated surfaces will be inoculated and re-inoculated with the test organism multiple times over a 24 hour period. Inoculations will be performed at time zero, 3, 6, 9, 12, 15, 18 and 22 hours. At five times within this 24 hour period, the carriers will be neutralized and assayed for survivors. Test organism recovery and enumerations will be performed at 2, 5, 11, 17 and 24 hours. Appropriate controls including: culture purity, organic soil load sterility, initial suspension (inoculum count), neutralizer sterility, carrier sterility, stainless steel control (viability control), neutralization confirmation, and carrier quantitation will be performed.

TEST METHOD

Chart 1

Test Organisms (ATCC #)	Growth Medium	Recovery Medium	Incubation Parameters
Enterobacter aerogenes (ATCC 13048)	Synthetic Broth	Tryptic Soy Agar + 5% Sheep's Blood	25-30°C, aerobic

The test organism to be used in this study was obtained from the American Type Culture Collection (ATCC), Manassas, VA.

Carriers

The Sponsor supplied treated test samples and the Sponsor supplied untreated control samples will be cut into squares approximately 1" x 1" in size. The carriers will be placed in the biosafety hood and exposed to UV light for 15±2 minutes on each side in order to decontaminate the surface. After decontamination, each carrier will be placed into a sterile plastic Petri dish matted with two pieces of sterile filter paper.

Stainless steel squares (1" x 1"), used as the organism viability control material, will be provided by ATS Labs. The stainless steel carriers will be prepared by removing the adhesive protective backing, if applicable. Each carrier will be cleaned by dipping in ethyl alcohol and rinsing thoroughly in deionized water. After cleaning, the carriers will be decontaminated by autoclave sterilization or by dipping in absolute ethanol and allowing the carriers to dry aseptically in a bio-safety hood. After decontamination, each carrier will be placed into a sterile plastic Petri dish matted with two pieces of sterile filter paper.

Preparation of Test Organism

From a stock slant, an initial tube (10 mL) of culture broth will be inoculated. This culture is termed the "initial broth suspension." From this initial broth suspension, a minimum of three daily transfers using 1 loopful (10 µL) of culture into 10 mL of culture media will be performed on consecutive days prior to use in testing procedure. For each test organism, the appropriate growth medium will be subcultured using a daily transfer (more than 3, but less than 18 transfers) of the test organism. Use 48±4 hour cultures as the inocula for this test.

Thoroughly mix each 48±4 hour culture on a "vortex" mixer and allow culture to settle for ≥15 minutes. Aspirate the upper two thirds of this suspension and use this as the inoculum for testing. An organic soil load and/or Triton X-100 (to aid in spreading of the test culture) may be added to the test culture per Sponsor's request or if hydrophobicity is a concern. (Example: 0.25 mL serum + 0.05 mL Triton X-100 + 4.70 mL bacteria suspension.) Following preparation, the test culture will be held at room temperature until the testing is completed.

Inoculation of Treated Test and Untreated Control Carriers

Five sets of Sponsor supplied treated test and untreated control (either Sponsor provided or provided by ATS Labs) carriers will be used in this study for each test organism. At time zero, all carrier sets will be inoculated. At 3, 6, 9, 12, 15, 18 and 22 hours, carrier sets not removed for quantitative recovery will be reinoculated (see Neutralization and Subculture section for additional clarification on recovery time points) as outlined in Chart 2. Inoculate each Sponsor supplied treated test carrier at staggered intervals with 40 µL of culture using a calibrated pipettor. At a low angle of incidence, slowly expel the inoculum across the surface of the carrier moving back and forth across the carrier to facilitate spreading. Replace the lids on each Petri dish. Carrier inoculation for each individual carrier is completed in ≤20 seconds. The carriers will be held covered under the conditions indicated by the Sponsor for the duration of the exposure. For consistency, the exposure time will begin when the inoculum is first applied to the initial carriers.

Test System Recovery
Sets of Sponsor supplied treated test and untreated control (either Sponsor provided or provided by ATS Labs) carriers will be removed for quantitative recovery two hours after their corresponding inoculation point as outlined in Chart 2.

Chart 2: Test System Inoculation & Recovery

Carrier Set	Inoculation Time(s)	Recovery Time	Total Number of Inoculations
1	Time Zero	2 hr	1
2	Time Zero, and 3 hrs	5 hrs	2
3	Time Zero, 3 hrs, 6 hrs, and 9 hrs	11 hrs	4
4	Time Zero, 3 hrs, 6 hrs, 9 hrs, 12 hrs, and 15 hrs	17 hrs	6
5	Time Zero, 3 hrs, 6 hrs, 9 hrs, 12 hrs, 15 hrs, 18 hrs and 22 hrs	24 hrs	8

At each recovery time, the carriers will be transferred to 20 mL of an appropriate neutralizer. Each neutralized carrier will be sonicated for 5 minutes to suspend any survivors from the carriers and rotated to mix. Prepare ten-fold serial dilutions and plate duplicate 1.0 mL aliquots of the 10^0 –10^{-4} dilutions using standard spread plate technique and appropriate agar. If swarming is a concern, plate duplicate 1.0 mL aliquots of 10^0 and duplicate 0.1 mL aliquots of 10^0 through 10^{-3}.

Incubation and Observation
Incubate the subculture plates for 48±4 hours at the temperature indicated in Chart 1 prior to observation and enumeration. (Alternate incubation conditions may be needed for certain organisms. Incubation conditions will be modified to suit the test organism if needed.) If necessary, the subcultures may be refrigerated at 2-8°C for up to three days prior to examination. Following incubation, the subcultures will be visually examined for growth. If possible, count plates containing between 30 and 300 Colony Forming Units (CFU). Representative test subcultures will be stained and/or biochemically assayed to confirm or rule out the presence of the test organism.

STUDY CONTROLS

Purity Control
A "streak plate for isolation" will be performed on the organism culture(s) and following incubation examined in order to confirm the presence of a pure culture. The acceptance criterion for this study control is a pure culture demonstrating colony morphology typical of the test organism.

Organic Soil Load Sterility Control
The solution(s) used for the soil load will be cultured, incubated as in the test, and visually examined. The acceptance criterion for this study control is lack of growth.

Initial Suspension Control (Inoculum Count)
This control will be performed in parallel with the time zero, 12 hour and 22 hour inoculations. Prepare and plate serial dilutions of the cultures used as the inocula and incubate the plates as in the test. Count the colonies to determine the number of organisms per milliliter of inoculum present at the time of testing. This control is for informational purposes only and therefore has no acceptance criterion.

Neutralizer Sterility Control
A representative sample of uninoculated neutralizing subculture medium will be incubated as in the test and visually examined. The acceptance criterion for this study control is lack of growth.

Carrier Sterility Control
Representative uninoculated Sponsor provided treated test and untreated control (either Sponsor provided or provided by ATS Labs) carriers will be added to the neutralizing subculture medium. A 1.0 mL sample will be plated onto an appropriate agar and will be incubated and examined. This control is for informational purposes only and therefore, has no acceptance criterion.

Carrier Quantitation Control
Three inoculated Sponsor provided control material carriers per organism will be used to determine the number of test organisms per carrier at each quantitative recovery time point. The control carriers will be transferred to neutralizing subculture media and sonicated as in the test. Ten-fold serial dilutions of the neutralizing subculture medium will be prepared and 1.0 mL or 0.1 mL of the appropriate dilutions will be plated in duplicate to yield countable numbers. The plates will be incubated as in the test and enumerated. The results of this control will be used to calculate the reduction of test organism achieved by the test carriers. There is no acceptance criterion for this study control.

Stainless Steel Control (Viability Control)
Three inoculated stainless steel control carriers will be used to verify the test organism's ability to survive on inert surfaces under the chosen test conditions at each quantitative recovery time point. The control carrier will be transferred to neutralizing subculture media and sonicated as in the test. Ten-fold serial dilutions of the neutralizing subculture medium will be prepared and 1.0 mL or 0.1 mL of the appropriate dilutions will be plated in duplicate to yield countable numbers. The plates will be incubated as in the test and enumerated. The acceptance criterion for this study control is a minimum geometric mean of 2.0×10^4 CFU/carrier.

Neutralization Confirmation Control
The neutralization of the Sponsor supplied treated carriers will be confirmed by neutralizing the carrier as in the test procedure. (If multiple concentrations of treated test carriers are being evaluated, only the lot with the highest level of active ingredient needs to be evaluated in this control.) Transfer a 1.0 mL aliquot of a diluted suspension of the test organism to target approximately 100 CFU/mL of neutralized solution to the vessel. Sonicate each neutralized vessel containing the test carrier and test organism suspension for 5 minutes. Plate 1.0 mL of this mixed solution in duplicate.

Determine the concentration of inoculum used in the neutralization confirmation assay as a numbers control. Perform the numbers control utilizing untreated neutralizer. Transfer a 1.0 mL aliquot of a diluted suspension of the test organism to target approximately 100 CFU/mL of neutralized solution to the vessel. Mix the numbers control vessel (do not sonicate) and plate 1.0 mL of the mixed solution in duplicate. Incubate the resulting plates as in the test and enumerate. The acceptance criterion for this study control is growth within 1 $\log_{10}$ of the numbers control.

Note: If swarming is a concern, 0.1 mL aliquots may be spread plated. In this case, approximately 1000 CFU/mL will be targeted when adding organism to the neutralized solution.

STUDY ACCEPTANCE CRITERIA

Test Substance Performance Criteria
To support a claim for continuously reducing test organisms on a treated surface, a minimum of a 90% reduction in numbers of the test organism(s) on the test surface compared to the number of test organism(s) on the control surface must be achieved at all recovery times over the 24 hour inoculation and exposure period.

Control Acceptance Criteria
The study controls must perform according to the criteria detailed in the study controls description section. If any control fails to meet the acceptance criteria, the test may be repeated under the same protocol.

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Timing Schematic for the Continuous Reduction Test of Antimicrobial Treated Surfaces

(The timing example is for one lab technician testing one lot of product using a staggered interval of 30 seconds. Additional technicians are utilized for extra lots and controls.)

Carrier Description	Initial Inoculation	Subculture Time (2 hour)	Reinoculation 1 (3 hours)	Subculture Time (5 hours)	Reinoculation 2 (6 hours)	Reinoculation 3 (9 hours)	Subculture Time (11 hours)	Reinoculation 4 (12 hours)	Reinoculation 5 (15 hours)	Subculture Time (17 hours)	Reinoculation 6 (18 hours)	Reinoculation 7 (22 hours)	Subculture Time (24 hours)
Lot A, 2 hour, carrier 1	0:00:00	2:00:00											
Lot A, 2 hour, carrier 2	0:00:30	2:00:30											
Lot A, 2 hour, carrier 3	0:01:00	2:01:00											
Lot A, 2 hour, carrier 4	0:01:30	2:01:30											
Lot A, 2 hour, carrier 5	0:02:00	2:02:00											
Lot A, 5 hour, carrier 1	0:02:30		3:02:30	5:02:30									
Lot A, 5 hour, carrier 2	0:03:00		3:03:00	5:03:00									
Lot A, 5 hour, carrier 3	0:03:30		3:03:30	5:03:30									
Lot A, 5 hour, carrier 4	0:04:00		3:04:00	5:04:00									
Lot A, 5 hour, carrier 5	0:04:30		3:04:30	5:04:30									
Lot A, 11 hour, carrier 1	0:05:00		3:05:00		6:05:00	9:05:00	11:05:00						
Lot A, 11 hour, carrier 2	0:05:30		3:05:30		6:05:30	9:05:30	11:05:30						
Lot A, 11 hour, carrier 3	0:06:00		3:06:00		6:06:00	9:06:00	11:06:00						
Lot A, 11 hour, carrier 4	0:06:30		3:06:30		6:06:30	9:06:30	11:06:30						
Lot A, 11 hour, carrier 5	0:07:00		3:07:00		6:07:00	9:07:00	11:07:00						
Lot A, 17 hour, carrier 1	0:07:30		3:07:30		6:07:30	9:07:30		12:07:30	15:07:30	17:07:30			
Lot A, 17 hour, carrier 2	0:08:00		3:08:00		6:08:00	9:08:00		12:08:00	15:08:00	17:08:00			
Lot A, 17 hour, carrier 3	0:08:30		3:08:30		6:08:30	9:08:30		12:08:30	15:08:30	17:08:30			
Lot A, 17 hour, carrier 4	0:09:00		3:09:00		6:09:00	9:09:00		12:09:00	15:09:00	17:09:00			
Lot A, 17 hour, carrier 5	0:09:30		3:09:30		6:09:30	9:09:30		12:09:30	15:09:30	17:09:30			
Lot A, 24 hour, carrier 1	0:10:00		3:10:00		6:10:00	9:10:00		12:10:00	15:10:00		18:10:00	22:10:00	24:10:00
Lot A, 24 hour, carrier 2	0:10:30		3:10:30		6:10:30	9:10:30		12:10:30	15:10:30		18:10:30	22:10:30	24:10:30
Lot A, 24 hour, carrier 3	0:11:00		3:11:00		6:11:00	9:11:00		12:11:00	15:11:00		18:11:00	22:11:00	24:11:00
Lot A, 24 hour, carrier 4	0:11:30		3:11:30		6:11:30	9:11:30		12:11:30	15:11:30		18:11:30	22:11:30	24:11:30
Lot A, 24 hour, carrier 5	0:12:00		3:12:00		6:12:00	9:12:00		12:12:00	15:12:00		18:12:00	22:12:00	24:12:00

PROCEDURE FOR IDENTIFICATION OF THE TEST SYSTEM

ATS Labs maintains Standard Operating Procedures (SOPs) relative to efficacy testing studies. Efficacy testing is performed in strict adherence to these SOPs which have been constructed to cover all aspects of the work including, but not limited to, receipt, log-in, and tracking of biological reagents including test organism strains for purposes of identification, receipt and use of chemical reagents. These procedures are designed to document each step of efficacy testing studies. Appropriate references to medium, batch number, etc. are documented in the raw data collected during the course of each study.

Additionally, each efficacy test is assigned a unique Project Number when the protocol for the study is initiated by the Study Director. This number is used for identification of the test subcultures, etc. during the course of the test. Test subcultures are also labeled with reference to the test organism, experimental start date, and test product. Microscopic and/or macroscopic evaluations of positive subcultures are performed in order to confirm the identity of the test organism. These measures are designed to document the identity of the test system.

METHOD FOR CONTROL OF BIAS: NA

REPORT

The report will include, but not be limited to, identification of the sample, date received, initiation and completion dates, identification of the bacterial strains used, description of media and reagents, description of the methods employed, tabulated results and conclusion as it relates to the purpose of the test, and all other items required by 40 CFR Part 160.185.

PROTOCOL CHANGES

If it becomes necessary to make changes in the approved protocol, the revision and reasons for changes will be documented, reported to the Sponsor and will become a part of the permanent file for that study. Similarly, the Sponsor will be notified as soon as possible whenever an event occurs that may have an effect on the validity of the study.

Standard operating procedures used in this study will be the correct effective revision at the time of the work. Any minor changes to SOPs (for this study) or methods used will be documented in the raw data and approved by the Study Director.

TEST SUBSTANCE RETENTION

It is the responsibility of the Sponsor to retain a sample of the test substance. All unused test substance will be discarded following study completion unless otherwise indicated by Sponsor.

RECORD RETENTION

Study Specific Documents

All of the original raw data developed exclusively for this study shall be archived at ATS Labs. These original data include, but are not limited to, the following:

1. All handwritten raw data for control and test substances including, but not limited to, notebooks, data forms and calculations.
2. Any protocol amendments/deviation notifications.
3. All measured data used in formulating the final report.
4. Memoranda, specifications, and other study specific correspondence relating to interpretation and evaluation of data, other than those documents contained in the final study report.
5. Original signed protocol.
6. Certified copy of final study report.
7. Study-specific SOP deviations made during the study.

Facility Specific Documents

The following records shall also be archived at ATS Labs. These documents include, but are not limited to, the following:

1. SOPs which pertain to the study conducted.
2. Non study-specific SOP deviations made during the course of this study which may affect the results obtained during this study.
3. Methods which were used or referenced in the study conducted.
4. QA reports for each QA inspection with comments.
5. Facility Records: Temperature Logs (ambient, incubator, etc.), Instrument Logs, Calibration and Maintenance Records.
6. Current curriculum vitae, training records, and job descriptions for all personnel involved in the study.

REFERENCES

1. U.S. Environmental Protection Agency, Registration Division, Office of Pesticide Programs, Efficacy Data Requirements Sanitizer Test (for inanimate, non-food contact surfaces), DIS/TSS-10, January 7, 1982.

2. U.S. Environmental Protection Agency Pesticide Assessment Guidelines, Subdivision G, Section 91-2; Item j Sanitizers (for non-food contact surfaces).

3. ASTM Test Method, Standard Test Method for Efficacy of Sanitizers Recommended for Inanimate Non-Food Contact Surfaces, E1153.

4. Association of Official Analytical Chemists (AOAC) Official Method 961.02 Germicidal Spray Products as Disinfectants. In Official Methods of Analysis of the AOAC, 2009 Edition.

5. EPA Protocol: "Test Method for the Continuous Reduction of Bacterial Contamination on Copper Alloy Surfaces." Website link: *http://www.epa.gov/oppad001/pdf_files/test_meth_contin_reduc_surfaces.pdf*

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CALCULATIONS

Initial Suspension

$$\text{CFU/mL} = \frac{\text{(average number of colonies/plate@dilution) x (dilution factor)}}{\text{(volume plated in mL)}}$$

Number of Organisms Surviving per Carrier

$$\text{CFU/carrier} = \frac{\text{(average number colonies/plate @ dilution) x (dilution factor) x (volume neutralized solution)}}{\text{(volume plated)}}$$

The carrier population will be calculated and reported using data from the most appropriate dilution(s).

Geometric Mean of Number of Organisms Surviving on the Treated Test or Untreated Control Carriers

$$\text{Geometric Mean} = \text{Antilog of } \frac{\text{Log}_{10}X_1 + \text{Log}_{10}X_2 + \ldots \text{Log}_{10}X_N}{N}$$

where X equals CFU/carrier and N equals the number of replicates tested

Percent Reduction Achieved by the Test Carriers

% reduction = [(a - b) / a] x 100

where:

a = geometric mean of the number of organisms surviving on the Sponsor supplied untreated control carriers
b = geometric mean of the number of organisms surviving on the Sponsor provided treated test carriers

Log_{10} Reduction Achieved by the Test Carriers

(Average Log_{10} Sponsor supplied untreated control carriers) – (Average Log_{10} Sponsor provided treated test carriers)

Neutralization Control Recovery Log_{10} Difference = (Log_{10} Numbers Control) – (Log_{10} Neutralization Results)
Used for the neutralization confirmation control only.

Statistical Analysis

None used.

Three digits will be used when calculating Log_{10}, Average Log_{10}, Geometric Mean and Percent Reduction values.

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STUDY INFORMATION

(All sections must be completed prior to submitting protocol)

Test Substance (Name and Batch Number - exactly as it should appear on final report):
See Attachment

NOTE: Recommended carrier size is 1" x 1"

Sponsor Supplied Untreated Lot (charged as an additional lot)
☑ Untreated Lot 2011-130:109
☐ Not Applicable

☑ Perform calculations based on untreated lot
☐ Perform calculations based on stainless steel control

Expiration Date: NA

Test Substance Active Concentration (upon submission to ATS Labs): Barquat MB-80, n-Alkyl (approximately C14-50%, C12-40%, C16-10%) Dimethyl Benzyl-Ammonium Chloride

Product Description:
☐ Copper ☐ Silver ☑ Other Sanitizer #1

Sample Preparation:
☐ Pre-cleaning needed:
☑ No pre-cleaning needed

Sample Decontamination:
☑ UV sterilization
☐ Dipping in ethanol and drying aseptically
☐ Autoclave sterilization (dry cycle)
☐ No sterilization is necessary

Neutralization/Subculture Broth:
☐
☑ ATS Labs' Discretion. By checking, the Sponsor authorizes ATS Labs, at their discretion, to perform neutralization confirmation assays at the Sponsor's expense prior to testing to determine the most appropriate neutralizer. (See Fee Schedule).

Storage Conditions:
☑ Room Temperature
☐ 2-8°C
☐ Other:

Hazards:
☑ None known: Use Standard Precautions
☐ Material Safety Data Sheet, Attached for each product
☐ As Follows:

Test Organism(s): ☑ *Enterobacter aerogenes* (ATCC 13048)

Carrier Number: 5 treated carriers and 3 control carriers, per exposure time

Exposure Time: 2 hours following inoculation with recovery to occur at 2, 5, 11, 17, and 24 hours

Exposure Temperature: 25±2°C and 60±2% RH

Organic Soil Load:
☑ Minimum 5% Organic Soil Load (Fetal Bovine Serum) and a final concentration of 0.01% Triton X-100
☐ Minimum 5% Organic Soil Load (Fetal Bovine Serum)
☐ Include Triton X-100 to a final concentration of 0.01% Triton X-100 only, to aid in spreading of the test organism
☐ No Organic Soil Load Required
☐ Other

TEST SUBSTANCE SHIPMENT STATUS

☑ Has been used in one or more previous studies at ATS Labs .
☐ Has been shipped to ATS Labs (but has not been used in a previous study).
Date shipped to ATS Labs:_____ Sent via *overnight* delivery? ☐ Yes ☐ No
☐ Will be shipped to ATS Labs.
Date of expected receipt at ATS Labs:_____
☐ Sender (if other than Sponsor):_____

COMPLIANCE

Study to be performed under EPA Good Laboratory Practice regulations (40 CFR Part 160) and in accordance to standard operating procedures.
☑ Yes
☐ No (Non-GLP Study)

PROTOCOL MODIFICATIONS

☑ Approved without modification
☐ Approved with modification

PROTOCOL ATTACHMENTS

Supplemental Information Form Attached - ☑ Yes ☐ No

APPROVAL SIGNATURES

SPONSOR:

NAME: Mrs. Stephanie Magnani TITLE: Chemist II
SIGNATURE: Stephanie Magnani DATE: 4/19/2013
PHONE: (216) 566 - 3235 FAX: (216) 515 - 5824 EMAIL: stephanie.magnani@sherwin.com

For confidentiality purposes, study information will be released only to the sponsor/representative signing the protocol (above) unless other individuals are specifically authorized in writing to receive study information.

Other individuals authorized to receive information regarding this study: ☐ See Attached

ATS Labs:

NAME: Matthew Sathe
Study Director

SIGNATURE: Matthew Sathe DATE: 4-19-13
Study Director

Attachment for SHE09041613.CCR.2 MS 4-19-13

Protocol Attachment 1 of 1 - Test Substance Name and Batch Numbers

PROTOCOLS : SHE09041613.CCR.1,
SHE09041613.CCR.2

PRODUCT ID : Sanitizer #1

UNTREATED BATCH : 2011-130:109
BATCH 1 : 2011-130:124
BATCH 2 : 2011-130:125
BATCH 3 : 2011-130:126

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