**Conference for Food Protection**
**2010 Issue Form**

**Internal Number: 071**
**Issue: 2010 III-016**

| | | | | | | |
|---|---|---|---|---|---|---|
| **Council Recommendation:** | Accepted as Submitted | _____ | Accepted as Amended | _____ | No Action | _____ |
| **Delegate Action:** | Accepted | _____ | Rejected | _____ | | |

*All information above the line is for conference use only.*

**Title:**
Sequential Application of Hand Antiseptic for Use in No-Water Situations

**Issue you would like the Conference to consider:**
Effective hand hygiene for situations where soap and water are unavailable remains a challenge for food safety. Under the 2009 FDA Model Food Code, Section 2-301.16, employees may use a hand antiseptic to clean hands when food exposure is limited and handwashing sinks are not conveniently available. In addition, employees may use chemically treated disinfectant towelettes per Section 5-203.11(C).

It has now been found that an effective hand cleansing, equivalent to handwashing with soap and water as specified in Section 5-203.11, can be achieved by sequential use of alcohol-based hand antiseptics, wherein a first application is wiped off with a dry single-use towel, followed immediately by a second application that is allowed to dry as per normal use directions. The latest testing of this hand cleansing/degerming technique shows it to be effective in the presence of organic food soils. This adds an additional safety factor to support incorporation of the method into food safety practices for select situations.

This protocol is not a substitute for handwashing in stationary facilities where cleaning can be accomplished per 2-301.12.

[Note: After the near unanimous vote for adoption by Council III in 2008, this issue was extracted during the Assembly of Delegates, citing the need for additional testing which has now been concluded along with an additional two years of field testing under the guidance of the Southern Nevada Health District. SNHD has also cleared this intervention for school foodservice use during water outages.]

**Public Health Significance:**
Potential contamination of ready-to-eat foods is increased in situations where access to soap and water are limited or simply unavailable. The new proposed option increases the odds of effective hand degerming in those situations, including its use between single-use glove changes.

**Recommended Solution: The Conference recommends...:**
a letter be sent to FDA requesting the following change to the Model Food Code:
5-203.11 Handwashing Sinks
(A)(B)(C)

(D) When food exposure is limited and handwashing sinks are not conveniently located, such as at outdoor events, mobile or temporary food service and some vending machine locations, employees may use a regimen of sequential application of hand antiseptic wherein the first application is treated as a handwash with full scrubbing action for 15 seconds and then, while wet, wiped off with a single-use paper towel, immediately followed by a second application which is allowed to dry per standard label instruction.
(i) Said hand antiseptic shall meet requirements of 2-301.16
(ii) Said hand antiseptic shall have supporting test data indicating statistical equivalence to a standard handwash in hand degerming.

**Submitter Information:**


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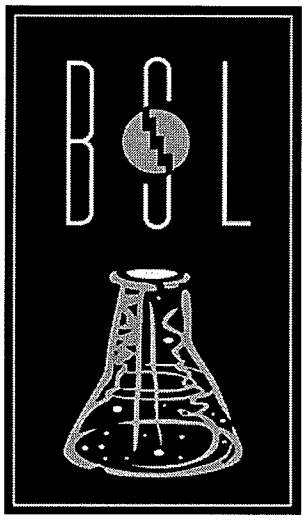
**Attachments:**

- "Determination of the Antimicrobial Efficacy of Three Test Articles (2008)"
- "Determination of the Antimicrobial Efficacy of Three Test Articles (2009)"
- "Sequential Application of Hand Antiseptic for Use in No-Water Situations"
- "SaniTwice: A Hand Hygiene Solution for Food Handlers"
- "Test Results For Heavy Soil Pilot SaniTwice Study"

*It is the policy of the Conference for Food Protection to not accept Issues that would endorse a brand name or a commercial proprietary process.*



January 17, 2008

FINAL REPORT #070723-150

# DETERMINATION OF THE ANTIMICROBIAL EFFICACY OF THREE (3) TEST ARTICLES USING A VARIATION OF THE HEALTH CARE PERSONNEL HANDWASH PROCEDURE

Prepared for:

(SPONSOR)

Prepared by:

**BIOSCIENCE LABORATORIES, INC.** (TESTING FACILITY)
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## TABLE OF CONTENTS





This Study has been approved by the GIRB on _____

## EXECUTIVE SUMMARY

The purpose of this study was to evaluate the antimicrobial efficacy of three (3) test articles using a modification of the Health Care Personnel Handwash evaluation. The indicator microorganism used for hand contaminations was *Escherichia coli* (ATCC #11229). Eleven (11) subjects used each of the three (3) test articles (reference Section 14.0 of this Final Report and a Protocol and/or SOP Deviation Recording Form [Form No. 99-QA-004] in Addendum I of this Final Report), one (1) at a time. Subjects performed two (2) consecutive hand contaminations with the challenge suspension in a beef broth medium, the first followed by a sample for baseline, and the second by a product application. Subjects then decontaminated their hands with a 70% Ethanol rinse and a nonmedicated soap wash, and then used a second Test Article. This procedure was repeated again with the remaining Test Article. The baseline and post-application samples were evaluated for the presence of *Escherichia coli* (ATCC #11229). The testing methods were based on the Food and Drug Administration Tentative Final Monograph (TFM) for *Effectiveness Testing of an Antiseptic Handwash or Health Care Personnel Handwash.* (FR59:116, 17 June 94) and ASTM E1174-06, *Standard Test Method for Evaluation of the Effectiveness of Health Care Personnel Handwash Formulations.*

The critical index for this study was a two (2) $\log_{10}$ reduction in baseline populations after product application.

STATISTICAL ANALYSIS #1

For Test Article #1, Bland Foaming Handwash (Lot Number 275543), the bacterial populations recovered after product application were reduced significantly from baseline populations. Test Article #1 produced a mean $\log_{10}$ reduction of 2.80 after product application and met the critical index of the study.

For Test Article #2, Instant Hand Sanitizer Gel (62% Ethanol; Lot Number 240041 5179), the bacterial populations recovered after product application were reduced significantly from baseline populations. Test Article #2 produced a mean $\log_{10}$ reduction of 2.64 after product application and met the critical index of the study.

For Test Article #3, Sanitizing Hand Wipes (68.15% Ethanol; Lot Number 973-12), followed by Test Article #2, Instant Hand Sanitizer Gel (62% Ethanol; Lot Number 240041 5179), applied per Test Article #3 Application Procedure, the bacterial populations recovered after product application were reduced significantly from baseline populations. Test Article #3 followed by Test Article #2, applied per Test Article #3 Application Procedure, produced a mean $\log_{10}$ reduction of 2.47 after product application and met the critical index of the study.

STATISTICAL ANALYSIS #2

Upon completion of the statistical analysis, Subject #12's data were determined to be outliers. Further investigation revealed that the subject appeared to have a learning disability and needed repeated instruction by the monitoring laboratory technician to be able to perform each of the steps required by the study protocol. The conclusions below results from a statistical analysis excluding data from testing of Subject #12.

For Test Article #1, Bland Foaming Handwash (Lot Number 275543), the bacterial populations recovered after product application were reduced significantly from baseline populations. Test Article #1 produced a mean $\log_{10}$ reduction of 2.93 after product application and met the critical index of the study.

Test Article #2, Instant Hand Sanitizer Gel (62% Ethanol; Lot Number 240041 5179), the bacterial populations recovered after product application were reduced significantly from baseline populations. Test Article #2 produced a mean $\log_{10}$ reduction of 2.83 after product application and met the critical index of the study.

Test Article #3, Sanitizing Hand Wipes (68.15% Ethanol; Lot Number 973-12), followed by Test Article #2, Instant Hand Sanitizer Gel (62% Ethanol; Lot Number 240041 5179), applied per Test Article #3 Application Procedure, the bacterial populations recovered after product application were reduced significantly from baseline populations. Test Article #3 followed by Test Article #2, applied per Test Article #3 Application Procedure, produced a mean $\log_{10}$ reduction of 2.63 after product application and met the critical index of the study.



This Study has been approved by the GIRB on _____

January 17, 2008

FINAL REPORT # 070723-150

**1.0 TITLE:** **DETERMINATION OF THE ANTIMICROBIAL EFFICACY OF THREE (3) TEST ARTICLES USING A VARIATION OF THE HEALTH CARE PERSONNEL HANDWASH PROCEDURE**

**2.0 TESTING FACILITY:** **BIOSCIENCE LABORATORIES, INC.**
300 N. Willson Avenue
Bozeman, Montana 59715

**3.0 STUDY DIRECTORS:**

Robert R. McCormack - Principal Study Director
Kendra F. Drake - Associate Study Director

**4.0 PURPOSE OF STUDY:**

The purpose of this study was to evaluate the antimicrobial efficacy of three (3) test articles for use in the food service industry. Testing was performed per methodology based on the Food and Drug Administration Tentative Final Monograph (TFM) for *Effectiveness Testing of an Antiseptic Handwash or Health Care Personnel Handwash* (FR59:116, 17 June 94, pp. 31448-31450) and ASTM E1174-06, *Standard Test Method for Evaluation of the Effectiveness of Health Care Personnel Handwash Formulations.*

**5.0 SCOPE:**

The purpose of this study was to evaluate the antimicrobial efficacy of three (3) test articles using a modification of the Health Care Personnel Handwash evaluation. The indicator microorganism used for hand contaminations was *Escherichia coli* (ATCC #11229). Eleven (11) subjects used each of the three (3) test articles, one (1) at a time. Subjects performed two (2) consecutive hand contaminations with the challenge suspension in a beef broth medium, the first followed by a sample for baseline, and the second by a product application. Subjects then decontaminated their hands with a 70% Ethanol rinse and a nonmedicated soap wash, and then used a second Test Article. This procedure was repeated again with the remaining Test Article. The baseline and post-application samples were evaluated for the presence of *Escherichia coli* (ATCC #11229). The testing methods were based on the Food and Drug Administration Tentative Final Monograph (TFM) for *Effectiveness Testing of an Antiseptic Handwash or Health Care Personnel Handwash.* (FR59:116, 17 June 94) and ASTM E1174-06, *Standard Test Method for Evaluation of the Effectiveness of Health Care Personnel Handwash Formulations.* The Study Protocol was approved by the Gallatin Institutional Review Board (GIRB) on 12/04/07 (See Addendum I of this Final Report). One (1) deviation from the methodology described in the Study Protocol occurred (reference Section 14.0 of this Final Report), and as is detailed on a Protocol and/or SOP Deviation Recording Form (Form No. 99-QA-004) in Addendum I of this Final Report, it had no adverse effect upon the study outcome. No deviations from BioScience Laboratories, Inc., Standard Operating Procedures occurred during the course of this evaluation.



This Study has been approved by the GIRB on _____

## 6.0 STUDY DATES:

**STUDY INITIATION DATE:** 11/30/07

**EXPERIMENTAL START DATE:** 12/19/07

**EXPERIMENTAL END DATE:** 01/07/08

**STUDY COMPLETION DATE:** 01/17/08

## 7.0 TEST MATERIALS:

The test articles were provided to the Testing Facility by the Sponsor. Responsibility for determination of the identity, strength, purity, composition, stability, and solubility of the test articles, as well as responsibility for retention of the test articles, remained with the Sponsor. All documentation provided with the test articles is included in Addendum IX of this Final Report.

| | |
|---|---|
| Test Article #1: | Bland Foaming Handwash |
| Lot Number: | 275543 |
| Expiration Date: | 01/2010 |
| | |
| Test Article #2: | Instant Hand Sanitizer Gel |
| Active Ingredient: | 62% Ethanol |
| Lot Number: | 240041 5179 |
| Expiration Date: | 06/2008 |
| | |
| Test Article #3: | Sanitizing Hand Wipes |
| Active Ingredient: | 68.15% Ethanol |
| Lot Number: | 973-12 |
| Expiration Date: | 04/19/08 |

## 8.0 TEST ARTICLE APPLICATION PROCEDURES:

Test Period

8.1 Each subject was in testing for approximately four (4) hours on a single day and used each of the three (3) test articles. Prior to being admitted into testing, subjects were questioned regarding their adherence to the Protocol requirements. Subjects clipped their fingernails to a free edge of $\leq 1$ mm, if they had not already done so. All jewelry was removed from the hands and arms prior to washing.

**NOTE:** Each subject used each of the three (3) test articles, one (1) at a time, per specified application procedures. After the Glove Juice Sampling Procedure was performed following test article application and prior to use of another test article, the subjects were required to decontaminate their hands by performing a one (1) minute rinse with 70% Ethanol and an air-dry, followed by a thirty (30) second handwash using a nonmedicated soap. The subjects waited a minimum of twenty (20) minutes following the use of the nonmedicated soap and prior to use of another test article.

8.2 A handwash was performed using a nonmedicated soap to remove dirt and oil from the hands. A technician instructed subjects in the appropriate technique and verified its proper execution by subjects. The temperature of the water used for all wash or rinse procedures was controlled at 40° ± 2°C (see Water Temperature Monitoring Sheets [Form No. 96-CT-017] in Addendum VII of this Final Report]).



This Study has been approved by the GIRB on _____

Inoculum Application Procedure

8.3 Four and one-half (4.5) mLs of the beef broth suspension containing at least 1 x $10^9$ CFU/mL of *Escherichia coli* (ATCC #11229) were transferred into each subject's cupped hands in three (3) aliquant amounts of one and one-half (1.5) mLs.

8.4 The suspension was distributed over the entire surface of the hands (front and back), not reaching above the wrists, for twenty (20) ± five (5) seconds. Following distribution of the inoculum, the hands were held motionless, away from the body, and allowed to air-dry for thirty (30) ± five (5) seconds.

8.5 The procedure in Section 8.4 was repeated.

8.6 A final one and one-half (1.5) mL aliquant amount of the challenge suspension was dispensed into the subject's cupped hands and distributed over the entire surface of the hands (front and back), not reaching above the wrists, for twenty (20) ± five (5) seconds. The hands were allowed to air-dry for ninety (90) seconds.

8.7 After the timed ninety (90) second air-dry, the Glove Juice Sampling Procedure was performed. This first contamination cycle provided the baseline population level. It was followed with a thirty (30) second handwash using nonmedicated soap.

8.8 The challenge suspension was again dispensed into each subject's cupped hands and distributed as described above. After a timed ninety (90) second air-dry, the subjects applied their randomly assigned test article according to the directions below.

Test Article #1 Application Procedure

8.9 The subject wet hands within ten (10) seconds of completing the drying step.

8.10 Two (2) pumps (1.4 mL) of Test Article #1 were placed in the subject's cupped hands.

8.11 The subject lathered Test Article #1 for fifteen (15) seconds, followed by a ten (10) second rinse with water.

8.12 Following the water rinse, the subject used two (2) paper towels to pat-dry hands for ten (10) seconds.

Test Article #2 Application Procedure

8.13 Two (2) pumps (3.0 mL) of Test Article #2 were placed in the subject's cupped hands within ten (10) seconds of completing the drying step.

8.14 The subject rubbed Test Article #2 into the hands in a vigorous manner for fifteen (15) seconds.

8.15 Following Test Article #2 application, the subject used two (2) paper towels to pat-dry hands for ten (10) seconds.

8.16 An additional one (1) pump of Test Article #2 was placed in the subject's cupped hands (1.5 mL), and the hands were rubbed together until dry.

Test Article #3 Application Procedure

8.17 Within ten (10) seconds of completing the drying step, the subject wiped both hands with Test Article #3 in a standardized fashion for twenty-five (25) seconds.

8.18 Following the wiping procedure, one (1) pump of Test Article #2 was placed in the subject's cupped hands, and hands were rubbed together until dry.

## 9.0 EQUIPMENT AND SUPPLIES:

The equipment and supplies used for this study are summarized in the Study Protocol, included in Addendum I of this Final Report, and are also detailed on Clinical Trials Equipment Tracking Forms (Form No. 01-L-009) and Clinical Trials Supplies Tracking Forms (Form 01-L-008) in Addendum VII of this Final Report.

## 10.0 MEDIA:

The growth media and diluting fluids used in this study are as described in the Study Protocol in Addendum I of this Final Report. Additional details are recorded on Media/Diluent Tracking Forms (Form No. 97-L-007) in Addendum VIII of this Final Report.

## 11.0 SUBJECT DEMOGRAPHICS:

Twenty-seven (27) overtly healthy subjects, at least eighteen (18) years of age were admitted into the study. Eleven (11) subjects completed the study (reference Protocol and/or SOP Deviation Recording Form [Form No. 99-QA-004] in Addendum I of this Final Report). Insofar as possible, the group of subjects selected was of mixed sex, age, and race. Hands and forearms were free from clinically evident dermatoses, other injuries to the area, and/or any other disorders that may have compromised the subject and the study. All subjects who participated in the Study signed the Study Description and Informed Consent Form, Subject Confidential Information and Acceptance Criteria, and Authorization to Use and Disclose Protected Health Information Form (Appendix I of Addendum I of this Final Report) and List of Restricted Products (Appendix II of Addendum I of this Final Report) prior to participating in the study. The demographics of the study are presented in the table below.

| DEMOGRAPHIC SUMMARY | ALL SUBJECTS | |
|---|---|---|
| | Recruited | Received Product |
| **AGE** | | |
| Minimum Age | 19 | 19 |
| *Median Age* | *35* | *45* |
| Maximum Age | 69 | 69 |
| **SEX** | | |
| Males (M) | 14 | 5 |
| Females (F) | 13 | 6 |
| *Total* | *27* | *11* |
| **RACE** | | |
| White/Caucasian (C) | 26 | 10 |
| Latino (L) | 1 | 1 |
| *Total* | *27* | *11* |



This Study has been approved by the GIRB on _____

| DID NOT PARTICIPATE IN TESTING | |
|---|---|
| SC = Schedule Conflict | 1 |
| QC = Qualification (Inclusion/Exclusion) Criteria Failure | 10 |
| NS = No Show | 5 |

## 12.0 ADVERSE EVENTS:

No subject experienced an adverse event during or following completion of this study.

## 13.0 NEUTRALIZATION EVALUATION:

The results of a neutralization evaluation (BSLI SOP CT-1006) indicated that the neutralizer(s) used in the recovery medium successfully quenched the antimicrobial activity of the test articles. Study procedures followed guidelines set forth in ASTM E 1054-02, *Standard Test Methods for Evaluation of Inactivators of Antimicrobial Agents*, except that the microorganism was added to the neutralizer prior to the addition of the test articles. *Escherichia coli* (ATCC #11229) was used as the challenge species in the neutralizer validation study. All data resulting from the Neutralization Assay are included in Addendum VI of this Final Report.

## 14.0 DEVIATION FROM PROTOCOL:

Section 12.40 in Protocol 070723-150 states, "Within ten (10) seconds of completing the drying step (Section 12.31), the subject will wipe both hands with Test Article #3 in a standardized fashion for twenty-five (25) seconds." Subject 21 did not use Test Article #3 on both hands in a standardized fashion nor for the full twenty-five (25) seconds. Subject 21 dropped wipe with three (3) seconds left on the rub, continued without wipe, and one (1) pump of Test Article #2 was then placed in the subject's cupped hands. Subject 21 failed to follow applications instructions as directed by the monitoring laboratory technician. Subject 21's data for Test Article #3 were disregarded from the analysis, so there is no effect on the outcome of the study.

## 15.0 RESULTS - TABLES I THROUGH XII:

15.1 Table I presents the statistical summary of the $\log_{10}$ values following performance of Test Article #1 Application Procedure (Bland Foaming Handwash [Lot Number 275543]).

**Table I: Statistical Summary of the $\log_{10}$ Recovery Values following Performance of the Test Article #1 Application Procedure (Bland Foaming Handwash [Lot Number 275543])**

| Sample | Sample Size | Mean | Standard Deviation | 95% Confidence Interval |
|---|---|---|---|---|
| Baseline | 22 | 8.18 | 0.24 | 8.08 to 8.29 |
| Application 1 | 22 | 5.38 | 0.58 | 5.13 to 5.64 |
| Application 1 $\text{Log}_{10}$ Reduction | 22 | 2.80 | 0.68 | 2.50 to 3.10 |



This Study has been approved by the GIRB on _____

15.2 Table II presents the $log_{10}$ values and $log_{10}$ reduction from baseline values, by subject, following performance of the Test Article #1 Application Procedure (Bland Foaming Handwash [Lot Number 275543]).

**Table II: $Log_{10}$ Values and $Log_{10}$ Reduction from Baseline Values, by subject, following Performance of the Test Article #1 Application Procedure (Bland Foaming Handwash [Lot Number 275543])**

| Subject | Side | Baseline $log_{10}$ Values | Application 1 $log_{10}$ Values | Application 1 $log_{10}$ Reduction from Baseline |
|---|---|---|---|---|
| 1 | Left | 8.31 | 5.66 | 2.65 |
| | Right | 8.12 | 5.81 | 2.31 |
| 16 | Left | 8.34 | 4.33 | 4.01 |
| | Right | 8.34 | 5.27 | 3.07 |
| 3 | Left | 8.23 | 5.12 | 3.11 |
| | Right | 8.11 | 5.60 | 2.51 |
| 9 | Left | 8.43 | 4.97 | 3.46 |
| | Right | 8.19 | 5.11 | 3.07 |
| 20 | Left | 8.04 | 4.82 | 3.22 |
| | Right | 8.11 | 4.69 | 3.42 |
| 7 | Left | 8.19 | 4.14 | 4.05 |
| | Right | 8.14 | 4.69 | 3.45 |
| 18 | Left | 8.01 | 6.00 | 2.01 |
| | Right | 8.04 | 6.09 | 1.95 |
| 12 | Left | 7.66 | 5.87 | 1.79 |
| | Right | 7.58 | 6.22 | 1.36 |
| 21 | Left | 8.58 | 5.64 | 2.94 |
| | Right | 8.48 | 5.96 | 2.52 |
| 27 | Left | 8.21 | 5.55 | 2.67 |
| | Right | 8.21 | 5.76 | 2.44 |
| 26 | Left | 8.32 | 5.47 | 2.85 |
| | Right | 8.38 | 5.60 | 2.78 |



This Study has been approved by the GIRB on _____

15.3 Table III presents the statistical summary of the $log_{10}$ values following performance of Test Article #2 Application Procedure (Instant Hand Sanitizer Gel [62% Ethanol; Lot Number 240041 5179]).

**Table III: Statistical Summary of the $log_{10}$ Recovery Values following Performance of the Test Article #2 Application Procedure (Instant Hand Sanitizer Gel [62% Ethanol; Lot Number 240041 5179])**

| Sample | Sample Size | Mean | Standard Deviation | 95% Confidence Interval |
|---|---|---|---|---|
| Baseline | 22 | 8.14 | 0.31 | 8.00 to 8.28 |
| Application 1 | 22 | 5.50 | 0.79 | 5.15 to 5.85 |
| Application 1 $Log_{10}$ Reduction | 22 | 2.64 | 0.89 | 2.24 to 3.03 |

15.4 Table IV presents the $log_{10}$ values and $log_{10}$ reduction from baseline values, by subject, following performance of the Test Article #2 Application Procedure (Instant Hand Sanitizer Gel [62% Ethanol; Lot Number 240041 5179]).

**Table IV: $Log_{10}$ Values and $Log_{10}$ Reduction from Baseline Values by subject following Performance of the Test Article #2 Application Procedure (Instant Hand Sanitizer Gel [62% Ethanol; Lot Number 240041 5179])**

| Subject | Side | Baseline $log_{10}$ Values | Application 1 $log_{10}$ Values | Application 1 $log_{10}$ Reduction from Baseline |
|---|---|---|---|---|
| 1 | Left | 8.14 | 5.96 | 2.18 |
| | Right | 8.23 | 5.80 | 2.43 |
| 16 | Left | 8.28 | 5.99 | 2.29 |
| | Right | 8.25 | 6.45 | 1.80 |
| 3 | Left | 8.02 | 5.36 | 2.67 |
| | Right | 8.21 | 5.34 | 2.87 |
| 9 | Left | 8.43 | 4.96 | 3.47 |
| | Right | 8.39 | 5.56 | 2.84 |
| 20 | Left | 8.11 | 4.74 | 3.37 |
| | Right | 8.03 | 5.82 | 2.21 |
| 7 | Left | 8.29 | 4.19 | 4.10 |
| | Right | 8.20 | 4.85 | 3.35 |
| 18 | Left | 7.74 | 4.33 | 3.40 |
| | Right | 7.88 | 3.66 | 4.22 |
| 12 | Left | 7.25 | 6.47 | 0.78 |
| | Right | 7.45 | 6.90 | 0.55 |
| 21 | Left | 8.44 | 5.85 | 2.59 |
| | Right | 8.27 | 5.90 | 2.37 |
| 27 | Left | 8.27 | 6.21 | 2.06 |
| | Right | 8.31 | 5.61 | 2.69 |
| 26 | Left | 8.42 | 5.77 | 2.66 |
| | Right | 8.42 | 5.34 | 3.08 |



This Study has been approved by the GIRB on _____

15.5 Table V presents the statistical summary of the $log_{10}$ values following performance of Test Article #3 Application Procedure (Sanitizing Hand Wipes [68.15% Ethanol; Lot Number 973-12] followed by Instant Hand Sanitizer Gel [62% Ethanol; Lot Number 240041 5179]).

**Table V: Statistical Summary of the $log_{10}$ Recovery Values following Performance of the Test Article #3 Application Procedure (Sanitizing Hand Wipes [68.15% Ethanol; Lot Number 973-12] followed by Instant Hand Sanitizer Gel [62% Ethanol; Log Number 240041 5179])**

| Sample | Sample Size | Mean | Standard Deviation | 95% Confidence Interval |
|---|---|---|---|---|
| Baseline | 20 | 8.07 | 0.34 | 7.92 to 8.23 |
| Wash 1 | 20 | 5.60 | 0.64 | 5.30 to 5.90 |
| Wash 1 $Log_{10}$ Reduction | 20 | 2.47 | 0.76 | 2.12 to 2.83 |

15.6 Table VI presents the $log_{10}$ values and $log_{10}$ reduction from baseline values, by subject, following performance of the Test Article #3 Application Procedure (Sanitizing Hand Wipes [68.15% Ethanol; Lot Number 973-12] followed by Instant Hand Sanitizer Gel [62% Ethanol; Lot Number 240041 5179]).

**Table VI: $Log_{10}$ Values and $Log_{10}$ Reduction from Baseline Values by subject following Performance of the Test Article #3 Application Procedure (Sanitizing Hand Wipes [68.15% Ethanol; Lot Number 973-12] followed by Instant Hand Sanitizer Gel [62% Ethanol; Log Number 240041 5179])**

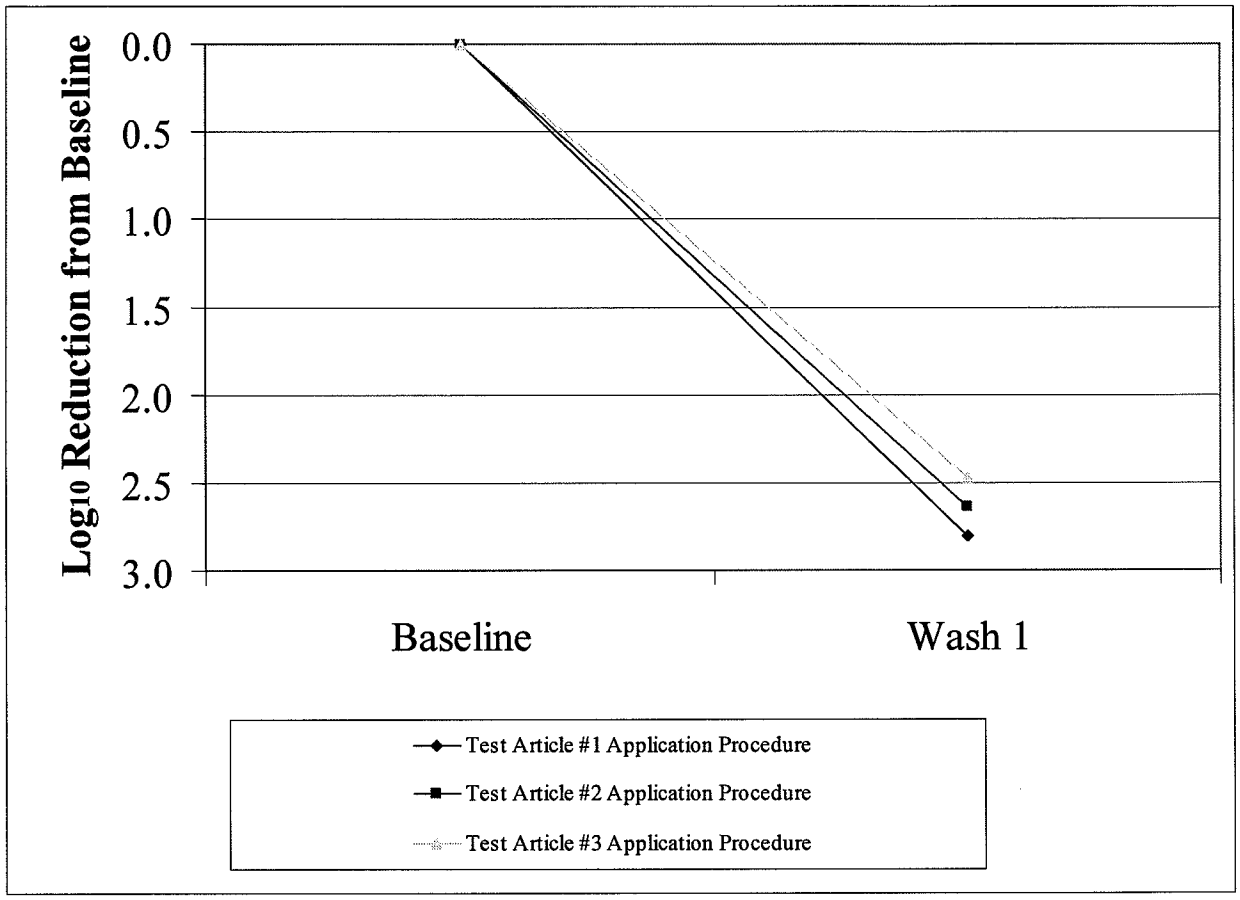
| Subject | Side | Baseline $log_{10}$ Values | Application 1 $log_{10}$ Values | Application 1 $log_{10}$ Reduction from Baseline |
|---|---|---|---|---|
| 1 | Left | 8.37 | 6.20 | 2.17 |
| | Right | 8.33 | 6.37 | 1.97 |
| 16 | Left | 8.13 | 5.31 | 2.82 |
| | Right | 8.29 | 5.81 | 2.48 |
| 3 | Left | 8.09 | 4.88 | 3.21 |
| | Right | 8.22 | 4.14 | 4.08 |
| 9 | Left | 8.41 | 5.18 | 3.23 |
| | Right | 8.35 | 5.33 | 3.02 |
| 20 | Left | 7.69 | 4.75 | 2.94 |
| | Right | 7.70 | 5.48 | 2.22 |
| 7 | Left | 8.16 | 5.07 | 3.09 |
| | Right | 8.23 | 6.32 | 1.91 |
| 18 | Left | 7.79 | 5.65 | 2.14 |
| | Right | 8.25 | 5.18 | 3.07 |
| 12 | Left | 7.23 | 6.21 | 1.03 |
| | Right | 7.35 | 6.30 | 1.05 |
| 21 | Left | * | * | * |
| | Right | * | * | * |
| 27 | Left | 8.16 | 5.33 | 2.83 |
| | Right | 8.23 | 5.86 | 2.38 |
| 26 | Left | 8.24 | 6.28 | 1.96 |
| | Right | 8.27 | 6.39 | 1.88 |



This Study has been approved by the GIRB on _____

15.7 Figure 1 presents the graphical presentation of the mean log10 reductions from baseline from each of the three (3) test article application procedures.

**Figure 1: Graphical Presentation of the Mean $log_{10}$ Reductions from Baseline From the Three (3) Test Article Application Procedures**




15.8 Table VII presents the statistical summary of the $log_{10}$ values following performance of Test Article #1 Application Procedure (Bland Foaming Handwash (Lot Number 275543)) excluding data from Subject 12.

**Table VII: Statistical Summary of the $log_{10}$ Recovery Values following Performance of the Test Article #1 Application Procedure (Bland Foaming Handwash [Lot Number 275543]) excluding Data from Subject #12**

| Sample | Sample Size | Mean | Standard Deviation | 95% Confidence Interval |
|---|---|---|---|---|
| Baseline | 20 | 8.24 | 0.16 | 8.17 to 8.31 |
| Application 1 | 20 | 5.32 | 0.56 | 5.05 to 5.58 |
| Application 1 $Log_{10}$ Reduction | 20 | 2.93 | 0.58 | 2.66 to 3.19 |



This Study has been approved by the GIRB on _____

15.9 Table VIII presents the $log_{10}$ values and $log_{10}$ reduction from baseline values, by subject, following performance of the Test Article #1 Application Procedure (Bland Foaming Handwash [Lot Number 275543]) excluding data from Subject #12.

**Table VIII: $Log_{10}$ Values and $Log_{10}$ Reduction from Baseline Values, by subject, following Performance of the Test Article #1 Application Procedure (Bland Foaming Handwash [Lot Number 275543]) excluding Data from Subject #12**

| Subject | Side | Baseline $log_{10}$ Values | Application 1 $log_{10}$ Values | Application 1 $log_{10}$ Reduction from Baseline |
|---|---|---|---|---|
| 1 | Left | 8.31 | 5.66 | 2.65 |
| | Right | 8.12 | 5.81 | 2.31 |
| 16 | Left | 8.34 | 4.33 | 4.01 |
| | Right | 8.34 | 5.27 | 3.07 |
| 3 | Left | 8.23 | 5.12 | 3.11 |
| | Right | 8.11 | 5.60 | 2.51 |
| 9 | Left | 8.43 | 4.97 | 3.46 |
| | Right | 8.19 | 5.11 | 3.07 |
| 20 | Left | 8.04 | 4.82 | 3.22 |
| | Right | 8.11 | 4.69 | 3.42 |
| 7 | Left | 8.19 | 4.14 | 4.05 |
| | Right | 8.14 | 4.69 | 3.45 |
| 18 | Left | 8.01 | 6.00 | 2.01 |
| | Right | 8.04 | 6.09 | 1.95 |
| 12 | Left | * | * | * |
| | Right | * | * | * |
| 21 | Left | 8.58 | 5.64 | 2.94 |
| | Right | 8.48 | 5.96 | 2.52 |
| 27 | Left | 8.21 | 5.55 | 2.67 |
| | Right | 8.21 | 5.76 | 2.44 |
| 26 | Left | 8.32 | 5.47 | 2.85 |
| | Right | 8.38 | 5.60 | 2.78 |



This Study has been approved by the GIRB on _____

15.10 Table IX presents the statistical summary of the $log_{10}$ values following performance of Test Article #2 Application Procedure (Instant Hand Sanitizer Gel [62% Ethanol; Lot Number 240041 5179]) excluding data from Subject #12.

**Table IX: Statistical Summary of the $log_{10}$ Recovery Values following Performance of the Test Article #2 Application Procedure (Instant Hand Sanitizer Gel [62% Ethanol; Lot Number 240041 5179]) excluding Data from Subject #12**

| Sample | Sample Size | Mean | Standard Deviation | 95% Confidence Interval |
|---|---|---|---|---|
| Baseline | 20 | 8.22 | 0.19 | 8.13 to 8.30 |
| Application 1 | 20 | 5.38 | 0.73 | 5.05 to 5.72 |
| Application 1 $Log_{10}$ Reduction | 20 | 2.83 | 0.66 | 2.53 to 3.14 |

15.11 Table X presents the $log_{10}$ values and $log_{10}$ reduction from baseline values, by subject, following performance of the Test Article #2 Application Procedure (Instant Hand Sanitizer Gel [62% Ethanol; Lot Number 240041 5179]) excluding data from Subject #12.

**Table X: $Log_{10}$ Values and $Log_{10}$ Reduction from Baseline Values by subject following Performance of the Test Article #2 Application Procedure (Instant Hand Sanitizer Gel [62% Ethanol; Lot Number 240041 5179]) excluding Data from Subject #12**

| Subject | Side | Baseline $log_{10}$ Values | Application 1 $log_{10}$ Values | Application 1 $log_{10}$ Reduction from Baseline |
|---|---|---|---|---|
| 1 | Left | 8.14 | 5.96 | 2.18 |
| | Right | 8.23 | 5.80 | 2.43 |
| 16 | Left | 8.28 | 5.99 | 2.29 |
| | Right | 8.25 | 6.45 | 1.80 |
| 3 | Left | 8.02 | 5.36 | 2.67 |
| | Right | 8.21 | 5.34 | 2.87 |
| 9 | Left | 8.43 | 4.96 | 3.47 |
| | Right | 8.39 | 5.56 | 2.84 |
| 20 | Left | 8.11 | 4.74 | 3.37 |
| | Right | 8.03 | 5.82 | 2.21 |
| 7 | Left | 8.29 | 4.19 | 4.10 |
| | Right | 8.20 | 4.85 | 3.35 |
| 18 | Left | 7.74 | 4.33 | 3.40 |
| | Right | 7.88 | 3.66 | 4.22 |
| 12 | Left | * | * | * |
| | Right | * | * | * |
| 21 | Left | 8.44 | 5.85 | 2.59 |
| | Right | 8.27 | 5.90 | 2.37 |
| 27 | Left | 8.27 | 6.21 | 2.06 |
| | Right | 8.31 | 5.61 | 2.69 |
| 26 | Left | 8.42 | 5.77 | 2.66 |
| | Right | 8.42 | 5.34 | 3.08 |



This Study has been approved by the GIRB on _____

15.12 Table V presents the statistical summary of the $log_{10}$ values following performance of Test Article #3 Application Procedure (Sanitizing Hand Wipes [68.15% Ethanol; Lot Number 973-12] followed by Instant Hand Sanitizer Gel [62% Ethanol; Lot Number 240041 5179]) excluding data from Subject #12.

**Table XI: Statistical Summary of the $log_{10}$ Recovery Values following Performance of the Test Article #3 Application Procedure (Sanitizing Hand Wipes [68.15% Ethanol; Lot Number 973-12] followed by Instant Hand Sanitizer Gel [62% Ethanol; Log Number 240041 5179]) excluding Data from Subject #12**

| Sample | Sample Size | Mean | Standard Deviation | 95% Confidence Interval |
|---|---|---|---|---|
| Baseline | 18 | 8.16 | 0.22 | 8.05 to 8.27 |
| Wash 1 | 18 | 5.53 | 0.63 | 5.21 to 5.85 |
| Wash 1 $Log_{10}$ Reduction | 18 | 2.63 | 0.61 | 2.33 to 2.93 |

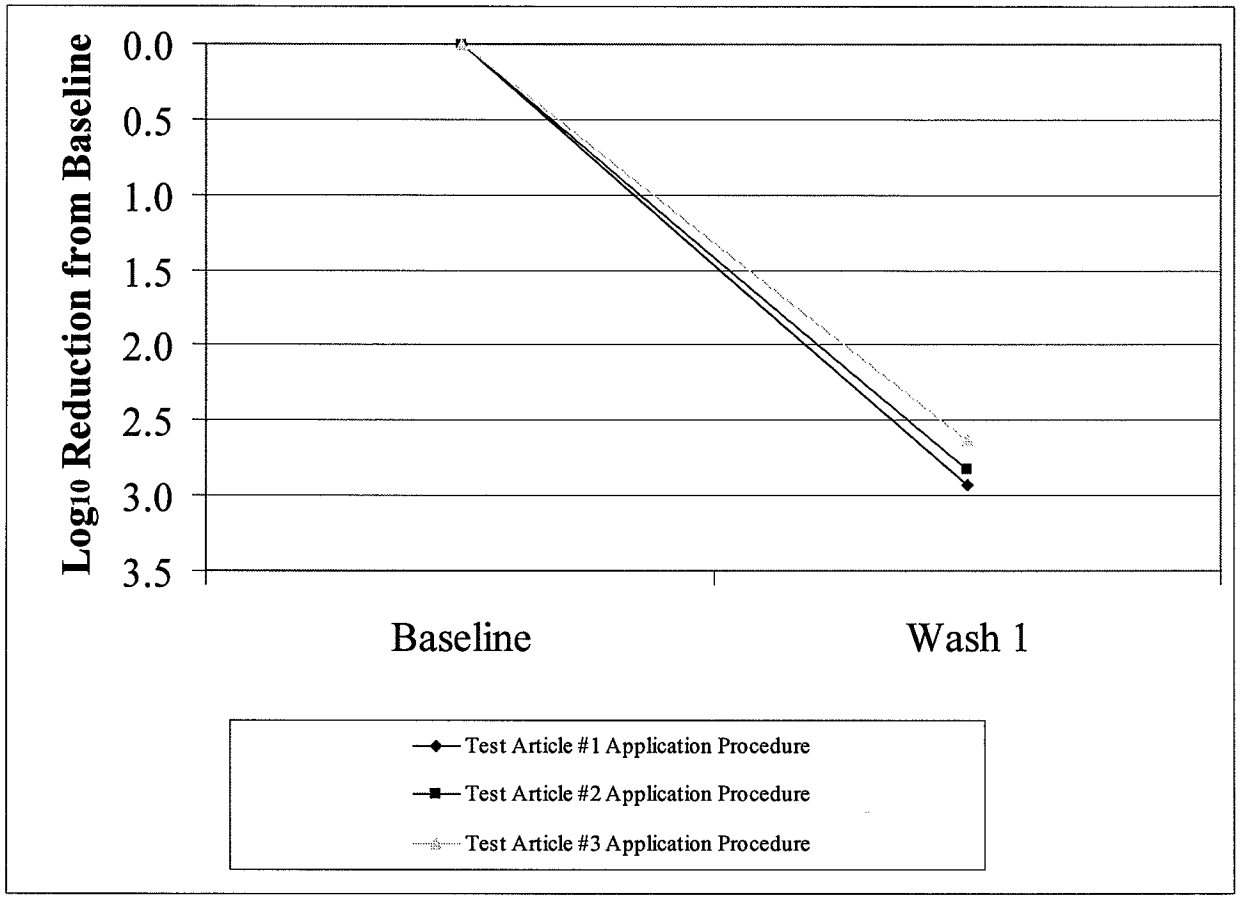
15.13 Table XII presents the $log_{10}$ values and $log_{10}$ reduction from baseline values, by subject, following performance of the Test Article #3 Application Procedure (Sanitizing Hand Wipes [68.15% Ethanol; Lot Number 973-12] followed by Instant Hand Sanitizer Gel [62% Ethanol; Lot Number 240041 5179]) excluding data from Subject #12.

**Table XII: $Log_{10}$ Values and $Log_{10}$ Reduction from Baseline Values by subject following Performance of the Test Article #3 Application Procedure (Sanitizing Hand Wipes [68.15% Ethanol; Lot Number 973-12] followed by Instant Hand Sanitizer Gel [62% Ethanol; Log Number 240041 5179]) excluding Data from Subject #12**

| Subject | Side | Baseline $log_{10}$ Values | Application 1 $log_{10}$ Values | Application 1 $log_{10}$ Reduction from Baseline |
|---|---|---|---|---|
| 1 | Left | 8.37 | 6.20 | 2.17 |
| | Right | 8.33 | 6.37 | 1.97 |
| 16 | Left | 8.13 | 5.31 | 2.82 |
| | Right | 8.29 | 5.81 | 2.48 |
| 3 | Left | 8.09 | 4.88 | 3.21 |
| | Right | 8.22 | 4.14 | 4.08 |
| 9 | Left | 8.41 | 5.18 | 3.23 |
| | Right | 8.35 | 5.33 | 3.02 |
| 20 | Left | 7.69 | 4.75 | 2.94 |
| | Right | 7.70 | 5.48 | 2.22 |
| 7 | Left | 8.16 | 5.07 | 3.09 |
| | Right | 8.23 | 6.32 | 1.91 |
| 18 | Left | 7.79 | 5.65 | 2.14 |
| | Right | 8.25 | 5.18 | 3.07 |
| 12 | Left | * | * | * |
| | Right | * | * | * |
| 21 | Left | * | * | * |
| | Right | * | * | * |
| 27 | Left | 8.16 | 5.33 | 2.83 |
| | Right | 8.23 | 5.86 | 2.38 |
| 26 | Left | 8.24 | 6.28 | 1.96 |
| | Right | 8.27 | 6.39 | 1.88 |

15.14 Figure 2 presents the graphical presentation of the mean log10 reductions from baseline from each of the three (3) test article application procedures excluding data from Subject #12.

**Figure 2: Graphical Presentation of the Mean $log_{10}$ Reductions from Baseline From the Three Test Article Application Procedures excluding Data from Subject #12**




## 16.0 CONCLUSION:

The critical index for this study was a two (2) $log_{10}$ reduction in baseline populations after product application.

STATISTICAL ANALYSIS #1

For Test Article #1, Bland Foaming Handwash (Lot Number 275543), the bacterial populations recovered after product application were reduced significantly from baseline populations. Test Article #1 produced a mean $log_{10}$ reduction of 2.80 after product application and met the critical index of the study.

For Test Article #2, Instant Hand Sanitizer Gel (62% Ethanol; Lot Number 240041 5179), the bacterial populations recovered after product application were reduced significantly from baseline populations. Test Article #2 produced a mean $log_{10}$ reduction of 2.64 after product application and met the critical index of the study.



This Study has been approved by the GIRB on _____

For Test Article #3, Sanitizing Hand Wipes (68.15% Ethanol; Lot Number 973-12), followed by Test Article #2, Instant Hand Sanitizer Gel (62% Ethanol; Lot Number 240041 5179), applied per Test Article #3 Application Procedure, the bacterial populations recovered after product application were reduced significantly from baseline populations. Test Article #3 followed by Test Article #2, applied per Test Article #3 Application Procedure, produced a mean $\log_{10}$ reduction of 2.47 after product application and met the critical index of the study.

STATISTICAL ANALYSIS #2

Upon completion of the statistical analysis, Subject #12's data were determined to be outliers. Further investigation revealed that the subject appeared to have a learning disability and needed repeated instruction by the monitoring laboratory technician to be able to perform each of the steps required by the study protocol. The conclusions below results from a statistical analysis excluding data from testing of Subject #12.

For Test Article #1, Bland Foaming Handwash (Lot Number 275543), the bacterial populations recovered after product application were reduced significantly from baseline populations. Test Article #1 produced a mean $\log_{10}$ reduction of 2.93 after product application and met the critical index of the study.

Test Article #2, Instant Hand Sanitizer Gel (62% Ethanol; Lot Number 240041 5179), the bacterial populations recovered after product application were reduced significantly from baseline populations. Test Article #2 produced a mean $\log_{10}$ reduction of 2.83 after product application and met the critical index of the study.

Test Article #3, Sanitizing Hand Wipes (68.15% Ethanol; Lot Number 973-12), followed by Test Article #2, Instant Hand Sanitizer Gel (62% Ethanol; Lot Number 240041 5179), applied per Test Article #3 Application Procedure, the bacterial populations recovered after product application were reduced significantly from baseline populations. Test Article #3 followed by Test Article #2, applied per Test Article #3 Application Procedure, produced a mean $\log_{10}$ reduction of 2.63 after product application and met the critical index of the study.

## 17.0 LABORATORY PERSONNEL:

The following employees of BioScience Laboratories, Inc., were involved in the testing or ancillary support of this Study. The laboratory personnel have been appropriately trained, and their training records are on-file in the Quality Assurance Unit at the Testing Facility.

| | |
|---|---|
| STUDY DIRECTOR: | Robert R. McCormack<br>Microbiologist |
| Sabrina Bakich<br>Marketing Manager/Product Handling | Paul O' Brien<br>Clinical Laboratory Technician |
| Amanda Berry<br>Subject Recruitment | Alicia Pfile<br>Microbiologist |
| Stephanie Cebulla<br>Laboratory Support Technician | Christine Roath<br>Microbiologist |
| Kendra F. Drake<br>Associate Study Director, Microbiologist | Lori Schlotfeldt<br>Supervisor of Laboratory Support |



This Study has been approved by the GIRB on _____

**LABORATORY PERSONNEL (Continued)**

Collette Duley
Microbiologist

Erika Ecton
Subject Recruitment

Amanda Henry
Microbiologist

August Grace Johnson
Microbiologist

Jacqueline Joyner
Subject Recruitment

Lisa Lehman
Microbiologist

Ron Neibauer
Manager of Clinical Laboratories

Jessica Sheehy
Laboratory Support Technician

Carl Schmidt
Microbiologist

Brian Stancil
Clinical Laboratory Technician

Robert H. Stancil
Microbiologist

Clare Wilson
Microbiologist

Annette C. Woods
Microbiologist

Kristy Wuebber
Microbiologist

## 18.0 QUALITY ASSURANCE PERSONNEL:

Liv Graving
Quality Assurance Associate

Amy Juhnke
Manager of Quality Assurance/Document Control

Scott McCommon
Manager of Quality Control

John A. Mitchell, Ph.D.
Director of Quality Assurance

Janis Smoke
Quality Assurance Associate

## 19.0 DOCUMENTATION AND RECORD-KEEPING:

All documentation and records were compiled, analyzed, and will be retained by BioScience Laboratories, Inc., at its facility in Bozeman, Montana. All raw data for this study, as well as the Final Report, will be retained in safe storage by the Testing Facility for a period of at least three (3) years.



This Study has been approved by the GIRB on _____

## 20.0 ACCEPTANCE:

### QUALITY ASSURANCE STATEMENT:

This study was inspected by the Quality Assurance Unit, and reports were submitted to the Study Director and Management in accordance with Standard Operating Procedures, as follows:

| Phase | Date |
|---|---|
| Neutralization Assay | 01/04/08 |
| Product Testing | 12/19/07 and 12/26/07 |
| Data Audit | 01/14/08 |
| Final Report Review | 01/17/08 |
| Reports to Study Director and Management | 12/19/07, 12/26/07, 01/04/08, and 01/17/08 |

This study was conducted in compliance with Good Laboratory Practices standards, as described by the FDA (reference CFR 21 Parts 50, 56, 312, and 314), with the following exception: test article preparations were not analyzed at BioScience Laboratories, Inc., to confirm concentration, stability, or homogeneity.



This Study has been approved by the GIRB on _____

## INDEX OF ADDENDA

I GIRB-Approved Protocol #070723-150
Protocol and/or SOP Deviation Recording Form (Form No. 99-QA-004)

II Qualification Criteria for Study 070723-150

III Sampling Data Sheets for Healthcare Personnel Handwash Study 070723-150
Irritation Evaluations for Study 070723-150

IV Q-Count™ Plate Counter Data Sheets (Form No. 00-L-009)
Q-Count™ Plate Count Data and Calculations

V Statistical Analysis

VI Neutralization Evaluation
- Project Notes (Form No. 95-G-001) for Neutralization Assay
- Neutralization Evaluation Data Sheets for Study 070723-150
- Neutralization Statistics

VII Study Notes and General Records
- Age Calculation and Demographics Worksheet
- Project Notes (Form No. 95-G-001)
- Protocol 070723-150 Randomization Scheme
- Clinical Trials Equipment Tracking Forms (Form No. 01-L-009)
- Clinical Trials Supplies Tracking Forms (Form No. 01-L-008)
- Water Temperature Monitoring Sheets (Form No. 96-CT-017)
- Incubator Log Forms (Form No. 96-L-008)
- Refrigerator Log Form (Form No. 96-L-015)
- Inoculum Preparation Tracking Forms - Flask Preparation (Form No. 07-CT-001)
- Inoculum Preparation Tracking Forms - Solid Media Preparation (Form No. 07-CT-002)
- Autoplate® 4000 Data Sheets for Healthcare Personnel Handwash Study 070723-150

VIII Media/Diluent Tracking Forms (Form No. 97-L-007)

IX Product Information
- Product Receipt Log (Form No. 92-L-023)
- Sample Submission Form and Document Compliance Statement (Form No. 94-G-007)
- Material Safety Data Sheets (MSDS)
- Product-Tracking Forms (Form No. 93-L-029)



This Study has been approved by the GIRB on _____

BIOSCIENCE
LABORATORIES•INC

March 12, 2009

FINAL REPORT #081211-150

# DETERMINATION OF THE ANTIMICROBIAL EFFICACY OF THREE (3) TEST ARTICLES USED IN FOUR (4) APPLICATION CONFIGURATIONS USING A VARIATION OF THE HEALTH CARE PERSONNEL HANDWASH PROCEDURE

Prepared for:

**GOJO INDUSTRIES, INC.** (SPONSOR)
One GOJO Plaza, Suite 500
Akron, Ohio 44311

Prepared by:

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300 N. Willson Avenue
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## TABLE OF CONTENTS

## EXECUTIVE SUMMARY

Twelve subjects used each of the four test article configurations over the course of two consecutive hand contaminations with *Escherichia coli* (ATCC #11229) in a beef broth medium as the indicator microorganism for each configuration. The four test configurations were assigned randomly according to an incomplete crossover design; that is, the order of use of each configuration was randomly determined. The first was followed by a sample for baseline, and the second by a product application. The subject then decontaminated their hands with a 70% ethanol rinse and a nonmedicated soap wash, and used a second test article configuration. This procedure was repeated twice more with the remaining test article configurations. The baseline and all post-application samples were evaluated for the presence of *Escherichia coli* (ATCC #11229). Testing was performed per a modification of the methodology in the Food and Drug Administration Tentative Final Monograph (TFM) for *Effectiveness Testing of an Antiseptic Handwash or Health Care Personnel Handwash* (FR59:116, 17 June 94, pp. 31448-31450).

The critical index for this study was a 2.0 $\log_{10}$ reduction in baseline populations after product application.

For Test Article #1, GOJO Luxury Foam Handwash (5400-520; Lot Number 322503), applied per Test Article Configuration #1 Application Procedure (two pumps [approximately 1.4 mL] of Test Article #1 into wet hands, 15-second lather, 10-second rinse, pat-dry hands with paper towels), the bacterial populations recovered after product application were reduced significantly from baseline populations. Test Article #1 produced a mean $\log_{10}$ reduction of 2.92 after product application and met the critical index of the study.

For Test Article #2, PURELL® Hand Sanitizing Gel VF481 (9900-501; Lot Number 306273), applied per Test Article Configuration #2 Application Procedure (one pump [approximately 1.5 mL] of Test Article #2 rubbed until dry), the bacterial populations recovered after product application were reduced significantly from baseline populations. Test Article #2, applied per Test Article Configuration #2 Application Procedure, produced a mean $\log_{10}$ reduction of 4.44 after product application and met the critical index of the study.

For Test Article #2, PURELL® Hand Sanitizing Gel VF481 (9900-501; Lot Number 306273), applied per Test Article Configuration #3 Application Procedure (two pumps [approximately 3.0 mL] of Test Article #2, pat-dry hands with paper towels, then an additional one pump [approximately 1.5 mL] rubbed until dry), the bacterial populations recovered after product application were reduced significantly from baseline populations. Test Article #2, applied per Test Article Configuration #3 Application Procedure, produced a mean $\log_{10}$ reduction of 4.61 after product application and met the critical index of the study.

For Test Article #3, PURELL® Instant Hand Sanitizer Foam (9800-502; Lot Number 320887), applied per Test Article Configuration #4 Application Procedure (four pumps [approximately 2.8 mL] of Test Article #3, pat-dry hands with paper towels, then an additional two pumps [approximately 1.4 mL] rubbed until dry), the bacterial populations recovered after product application were reduced significantly from baseline populations. Test Article #3 produced a mean $\log_{10}$ reduction of 3.64 after product application and met the critical index of the study.

March 12, 2009

FINAL REPORT #081211-150

**1.0 TITLE:** **DETERMINATION OF THE ANTIMICROBIAL EFFICACY OF THREE (3) TEST ARTICLES USED IN FOUR (4) APPLICATION CONFIGURATIONS USING A VARIATION OF THE HEALTH CARE PERSONNEL HANDWASH PROCEDURE**

**2.0 SPONSOR:** **GOJO INDUSTRIES, INC.**
One GOJO Plaza, Suite 500
Akron, Ohio 44311

**3.0 TESTING FACILITY:** **BIOSCIENCE LABORATORIES, INC.**
300 N. Willson Avenue
Bozeman, Montana 59715

**4.0 STUDY DIRECTORS:**

Robert R. McCormack - Principal Study Director
Kendra Drake - Associate Study Director

**5.0 PURPOSE OF STUDY:**

The purpose of this study was to evaluate the antimicrobial efficacy of three test articles used in four application configurations using a modification of the Health Care Personnel Handwash evaluation for use in the food service industry.

**6.0 SCOPE:**

A total of twelve subjects used each of the four test article configurations over the course of two consecutive hand contaminations with *Escherichia coli* (ATCC #11229) in a beef broth medium as the indicator microorganism for each configuration. The first was followed by a sample for baseline, and the second by a product application. The subject then decontaminated their hands with a 70% ethanol rinse and a nonmedicated soap wash, and used a second test article configuration. This procedure was repeated twice more with the remaining test article configurations. The baseline and all post-application samples were evaluated for the presence of *Escherichia coli* (ATCC #11229). The four test configurations were assigned randomly according to an incomplete crossover design; that is, the order of use of each configuration was randomly determined. Testing was performed per a modification of the methodology in the Food and Drug Administration Tentative Final Monograph (TFM) for *Effectiveness Testing of an Antiseptic Handwash or Health Care Personnel Handwash* (FR59:116, 17 June 94, pp. 31448-31450). The Study Protocol was approved by the Gallatin Institutional Review Board (GIRB) on 02/10/09. No deviations from the methodology described in the Study Protocol or from BioScience Laboratories, Inc., Standard Operating Procedures occurred during the course of this evaluation.

**7.0 STUDY DATES:**

**STUDY INITIATION DATE:** 02/06/09

**EXPERIMENTAL START DATE:** 02/13/09

**EXPERIMENTAL END DATE:** 02/23/09

**STUDY COMPLETION DATE:** 03/12/09

## 8.0 TEST MATERIALS:

The test articles were provided to the Testing Facility by the Sponsor. Responsibility for determination of the identity, strength, purity, composition, stability, and solubility of the test articles, as well as responsibility for retention of the test articles, remained with the Sponsor.

| | |
|---|---|
| Test Article #1: | GOJO Luxury Foam Handwash (5400-520) |
| Active Ingredient: | N/A |
| Lot Number: | 322503 |
| Expiration Date: | 11/2011 |

| | |
|---|---|
| Test Article #2: | PURELL® Hand Sanitizing Gel VF481 (9900-501) |
| Active Ingredient: | 70% ethanol |
| Lot Number: | 306273 |
| Expiration Date: | 04/2010 |

| | |
|---|---|
| Test Article #3: | PURELL® Instant Hand Sanitizer Foam (9800-502) |
| Active Ingredient: | 62% ethanol |
| Lot Number: | 320887 |
| Expiration Date: | 11/2010 |

## 9.0 EQUIPMENT AND SUPPLIES:

The equipment and supplies used for this study are summarized in the Study Protocol, included as Addendum I of this Final Report, and are also detailed on the Clinical Trials Equipment Tracking Forms (Form No. 01-L-009) and the Clinical Trials Supplies Tracking Forms (Form 01-L-008) in Addendum VII of this Final Report.

## 10.0 MEDIA:

The growth media and diluting fluids used in this study are as described in the Study Protocol in Addendum I of this Final Report. Additional details are recorded on the Media/Diluent Tracking Forms (Form No. 97-L-007) in Addendum VII of this Final Report.

## 11.0 SUBJECT DEMOGRAPHICS:

Twenty overtly healthy subjects at least 18 years of age were admitted into the study. Twelve subjects received product during testing. Insofar as possible, the group of subjects selected was of mixed sex, age, and race. Hands and forearms were free from clinically evident dermatoses, injuries, and/or any other disorders that may have compromised the subject and the study. All subjects who participated in the Study signed the Study Description and Informed Consent Form, Subject Confidential Information and Acceptance Criteria, and Authorization to Use and Disclose Protected Health Information Form (Appendix I of Addendum I of this Final Report) and List of Restricted Products (Appendix II of Addendum I of this Final Report) prior to participating in the study. The demographics of the study are presented in the table below.

| DEMOGRAPHIC SUMMARY | ALL SUBJECTS | |
|---|---|---|
| | Recruited | Received Product |
| **AGE** | | |
| Minimum Age | 18 | 19 |
| Median Age | 42 | 49 |
| Maximum Age | 71 | 71 |
| **SEX** | | |
| Males (M) | 8 | 3 |
| Females (F) | 12 | 9 |
| *Total* | *20* | *12* |
| **RACE** | | |
| White/Caucasian (C) | 20 | 12 |
| *Total* | *20* | *12* |
| **DID NOT PARTICIPATE IN TESTING** | | |
| QC = Qualification (Inclusion/Exclusion) Criteria Failure | 6 | |
| NS = No Show | 2 | |

**12.0 ADVERSE EVENTS:**

No subject experienced an adverse event during or following completion of this study.

**13.0 NEUTRALIZATION:**

A neutralization assay was performed to assure that the neutralizer(s) used in the recovery medium quenched the antibacterial properties of the test articles. Study procedures were based on guidelines set forth in ASTM E 1054-08, *Standard Test Methods for Evaluation of Inactivators of Antimicrobial Agents. Escherichia coli* (ATCC #11229) was used as the challenge species in the neutralizer validation study.

**14.0 TEST METHODS:**

14.1 Each subject was in testing for approximately five hours on a single day and used each of the four test article configurations. Prior to being admitted into testing, subjects were questioned regarding their adherence to protocol requirements. Subjects clipped their fingernails to a free edge of $\leq 1$ mm, if they had not already done so. All jewelry was removed from the hands and arms prior to washing.

14.2 A handwash was performed using a nonmedicated soap to remove dirt and oil from the hands. A technician instructed subjects in the appropriate technique and verified its proper execution. The temperature of the water used for this and any subsequent wash or rinse procedures was controlled at 40° ± 2°C.

Inoculum Application Procedure

14.3 A total of 4.5 mL of the beef broth suspension containing at least $1 \times 10^9$ CFU/mL of *Escherichia coli* (ATCC #11229) was transferred into each subject's cupped hands in three aliquant amounts.

14.3.1 A 1.5 mL aliquot of the challenge suspension was dispensed into the subject's cupped hands. The suspension was distributed over the entire surface of the hands (front and back), not reaching above the wrists, for 20 ± 5 seconds. Following distribution of the inoculum, the hands were held motionless, away from the body, and allowed to air-dry for 30 ± 5 seconds.

14.3.2 The procedure in Section 14.3.1 was repeated.

14.3.3 A final 1.5 mL of the challenge suspension was dispensed into the subject's cupped hands and distributed over the entire surface of the hands (front and back), not reaching above the wrists, for 20 ± 5 seconds. The hands were allowed to air-dry for 90 seconds.

14.4 After the timed 90-second air-dry, the Glove Juice Sampling Procedure (Section 12.45 of the Study Protocol) was performed. This first contamination cycle provided the baseline population level. It was followed with a 30-second handwash using nonmedicated soap.

14.5 The challenge suspension was again dispensed into each subject's cupped hands and distributed. After a timed 90-second air-dry, the subjects applied their randomly assigned test article configuration according to the directions below.

14.6 Test Article Configuration #1 Application Procedure

14.6.1 Subject wet their hands within 10 seconds of completing the drying step (Section 14.5).

14.6.2 Two pumps (approximately 1.4 mL) of Test Article #1 were placed in the subject's cupped hands.

14.6.3 Subject lathered Test Article #1 for 15 seconds, followed by a 10-second rinse with water.

14.6.4 Following the water rinse, the subject used two paper towels to pat-dry hands for 10 seconds.

14.7 Test Article Configuration #2 Application Procedure

14.7.1 Within 10 seconds of completing the drying step (Section 14.5), one pump (approximately 1.5 mL) of Test Article #2 was placed in the subject's cupped hands, and the hands were rubbed together until dry.

14.8 Test Article Configuration #3 Application Procedure

14.8.1 Two pumps (approximately 3.0 mL) of Test Article #2 were placed in the subject's cupped hands within 10 seconds of completing the drying step (Section 14.5).

14.8.2 Subject rubbed Test Article #2 into the hands in a vigorous manner for 15 seconds.

14.8.3 Subject then used two paper towels to pat-dry hands for 10 seconds.

14.8.4 An additional one pump (approximately 1.5 mL) of Test Article #2 was placed in the subject's cupped hands, and the hands were rubbed together until dry.

14.9 Test Article Configuration #4 Application Procedure

14.9.1 Four pumps (approximately 2.8 mL) of Test Article #3 were placed in the subject's cupped hands within 10 seconds of completing the drying step (Section 14.5).

14.9.2 Subject rubbed Test Article #3 into the hands in a vigorous manner for 15 seconds.

14.9.3 Subject then used two paper towels to pat-dry hands for 10 seconds.

14.9.4 An additional two pumps (approximately 1.4 mL) of Test Article #3 were placed in the subject's cupped hands, and the hands were rubbed together until dry.

14.10 Each subject used each of the four test article configurations, one at a time, per specified application procedures. After the Glove Juice Sampling Procedure was performed following test article application, and prior to use of another test article configuration, subjects were required to perform a 1-minute rinse with 70% ethanol and an air-dry, followed by a 30-second handwash using a nonmedicated soap. Subjects waited a minimum of 20 minutes following the use of the nonmedicated soap before using another test article.

## 15.0 STATISTICAL ANALYSIS:

15.1 Minitab® Statistical Software package was used for all statistical calculations. All statistical tests were calculated using the 0.05 level of significance for Type I ($\alpha$) error.

15.2 Descriptive statistics and confidence intervals were calculated using the 0.05 level of significance for Type I ($\alpha$) error. Statistical calculations of means and standard deviations were generated on the $\log_{10}$ recovery data from baseline samples, post-product application samples, and the $\log_{10}$ differences between baseline and post-application samples.

15.3 The critical index for this study was a 2.0 $\log_{10}$ reduction after product application.

## 16.0 RESULTS - TABLES I THROUGH VIII AND FIGURE 1:

16.1 Table I presents a statistical summary of the $\log_{10}$ recovery values following application of Test Article #1, GOJO Luxury Foam Handwash (5400-520; Lot Number 322503), per Test Article Configuration #1 Application Procedure (two pumps [approximately 1.4 mL] into wet hands, 15-second lather, 10-second rinse, pat-dry hands with paper towels).

16.2 Table II presents the $\log_{10}$ values and $\log_{10}$ reduction from baseline values, by subject and hand, following application of Test Article #1, GOJO Luxury Foam Handwash (5400-520; Lot Number 322503), per Test Article Configuration #1 Application Procedure (two pumps [approximately 1.4 mL] into wet hands, 15-second lather, 10-second rinse, pat-dry hands with paper towels).

16.3 Table III presents a statistical summary of the $\log_{10}$ recovery values following application of Test Article #2, PURELL® Hand Sanitizing Gel VF481 (9900-501; Lot Number 306273), per Test Article Configuration #2 Application Procedure (one pump [approximately 1.5 mL] rubbed until dry).

16.4 Table IV presents the $\log_{10}$ values and $\log_{10}$ reduction from baseline values, by subject and hand, following application of Test Article #2, PURELL® Hand Sanitizing Gel VF481 (9900-501; Lot Number 306273), per Test Article Configuration #2 Application Procedure (one pump [approximately 1.5 mL] rubbed until dry).

16.5 Table V presents a statistical summary of the $\log_{10}$ recovery values following application of Test Article #2, PURELL® Hand Sanitizing Gel VF481 (9900-501; Lot Number 306273), per Test Article Configuration #3 Application Procedure (two pumps [approximately 3.0 mL], pat-dry hands with paper towels, then an additional one pump [approximately 1.5 mL] rubbed until dry).

16.6 Table VI presents the $\log_{10}$ values and $\log_{10}$ reduction from baseline values, by subject and hand, following application of Test Article #2, PURELL® Hand Sanitizing Gel VF481 (9900-501; Lot Number 306273), per Test Article Configuration #3 Application Procedure (two pumps [approximately 3.0 mL], pat-dry hands with paper towels, then an additional one pump [approximately 1.5 mL] rubbed until dry).

16.7 Table VII presents a statistical summary of the $\log_{10}$ recovery values following application of Test Article #3, PURELL® Instant Hand Sanitizer Foam (9800-502; Lot Number 320887), per Test Article Configuration #4 Application Procedure (four pumps [approximately 2.8 mL], pat-dry hands with paper towels, then an additional two pumps [approximately 1.4 mL] rubbed until dry).

16.8 Table VIII presents the $\log_{10}$ values and $\log_{10}$ reduction from baseline values, by subject and hand, following application of Test Article #3, PURELL® Instant Hand Sanitizer Foam (9800-502; Lot Number 320887), per Test Article Configuration #4 Application Procedure (four pumps [approximately 2.8 mL], pat-dry hands with paper towels, then an additional two pumps [approximately 1.4 mL] rubbed until dry).

16.9 Figure 1 presents a graphical presentation of the mean $\log_{10}$ reductions from baseline following application of each test article per the four test article application procedures.

**Table I: Statistical Summary of the $log_{10}$ Recovery Values Following Application of Test Article #1, GOJO Luxury Foam Handwash (5400-520; Lot Number 322503), per Test Article Configuration #1 Application Procedure (two pumps [approximately 1.4 mL] into wet hands, 15-second lather, 10-second rinse, pat-dry hands with paper towels)**

| Sample | Sample Size | Mean | Standard Deviation | 95% Confidence Interval |
|---|---|---|---|---|
| Baseline | 24 | 7.77 | 0.31 | 7.64 to 7.90 |
| Application 1 | 24 | 4.85 | 0.53 | 4.63 to 5.07 |
| Application 1 $Log_{10}$ Reduction | 24 | 2.92 | 0.61 | 2.66 to 3.18 |

**Table II: $Log_{10}$ Values and $Log_{10}$ Reduction from Baseline Values by Subject and Hand Following Application of Test Article #1, GOJO Luxury Foam Handwash (5400-520; Lot Number 322503), per Test Article Configuration #1 Application Procedure (two pumps [approximately 1.4 mL] into wet hands, 15-second lather, 10-second rinse, pat-dry hands with paper towels)**

| Subject | Side | Baseline $log_{10}$ Values | Application 1 $log_{10}$ Values | Application 1 $log_{10}$ Reduction from Baseline |
|---|---|---|---|---|
| 1 | Left | 7.91 | 4.82 | 3.09 |
| | Right | 7.87 | 4.96 | 2.91 |
| 2 | Left | 7.55 | 5.11 | 2.43 |
| | Right | 7.72 | 5.17 | 2.56 |
| 3 | Left | 7.82 | 4.69 | 3.13 |
| | Right | 7.77 | 4.96 | 2.81 |
| 6 | Left | 8.01 | 3.49 | 4.52 |
| | Right | 7.99 | 3.18 | 4.82 |
| 7 | Left | 8.07 | 5.08 | 2.99 |
| | Right | 8.11 | 5.38 | 2.73 |
| 8 | Left | 7.99 | 4.90 | 3.09 |
| | Right | 7.96 | 5.25 | 2.72 |
| 10 | Left | 6.92 | 4.51 | 2.41 |
| | Right | 6.93 | 4.76 | 2.16 |
| 11 | Left | 7.64 | 5.28 | 2.36 |
| | Right | 7.71 | 5.44 | 2.28 |
| 13 | Left | 7.78 | 4.70 | 3.08 |
| | Right | 7.98 | 5.04 | 2.94 |
| 14 | Left | 7.47 | 4.75 | 2.72 |
| | Right | 7.56 | 4.96 | 2.60 |
| 15 | Left | 7.84 | 4.81 | 3.04 |
| | Right | 7.86 | 4.80 | 3.06 |
| 16 | Left | 8.00 | 5.02 | 2.98 |
| | Right | 7.99 | 5.29 | 2.70 |

**Table III: Statistical Summary of the $log_{10}$ Recovery Values Following Application of Test Article #2, PURELL® Hand Sanitizing Gel VF481 (9900-501; Lot Number 306273), per Test Article Configuration #2 Application Procedure (one pump [approximately 1.5 mL] rubbed until dry)**

| Sample | Sample Size | Mean | Standard Deviation | 95% Confidence Interval |
|---|---|---|---|---|
| Baseline | 24 | 7.88 | 0.29 | 7.76 to 8.00 |
| Application 1 | 24 | 3.44 | 0.47 | 3.24 to 3.64 |
| Application 1 $Log_{10}$ Reduction | 24 | 4.44 | 0.47 | 4.24 to 4.64 |

**Table IV: $Log_{10}$ Values and $Log_{10}$ Reduction from Baseline Values by Subject and Hand Following Application of Test Article #2, PURELL® Hand Sanitizing Gel VF481 (9900-501; Lot Number 306273), per Test Article Configuration #2 Application Procedure (one pump [approximately 1.5 mL] rubbed until dry)**

| Subject | Side | Baseline $log_{10}$ Values | Application 1 $log_{10}$ Values | Application 1 $log_{10}$ Reduction from Baseline |
|---|---|---|---|---|
| 1 | Left | 7.99 | 3.18 | 4.81 |
| | Right | 7.99 | 3.18 | 4.81 |
| 2 | Left | 7.89 | 3.18 | 4.71 |
| | Right | 7.95 | 3.18 | 4.76 |
| 3 | Left | 7.86 | 3.18 | 4.69 |
| | Right | 7.95 | 3.18 | 4.78 |
| 6 | Left | 7.99 | 3.18 | 4.81 |
| | Right | 8.02 | 3.18 | 4.84 |
| 7 | Left | 8.13 | 4.36 | 3.77 |
| | Right | 8.19 | 4.72 | 3.48 |
| 8 | Left | 7.98 | 3.18 | 4.80 |
| | Right | 7.98 | 3.18 | 4.80 |
| 10 | Left | 6.99 | 3.18 | 3.82 |
| | Right | 7.05 | 3.18 | 3.88 |
| 11 | Left | 8.03 | 4.14 | 3.89 |
| | Right | 8.02 | 4.39 | 3.63 |
| 13 | Left | 7.87 | 3.49 | 4.39 |
| | Right | 8.05 | 3.49 | 4.57 |
| 14 | Left | 7.63 | 3.49 | 4.14 |
| | Right | 7.63 | 3.66 | 3.97 |
| 15 | Left | 7.87 | 3.18 | 4.69 |
| | Right | 8.05 | 3.18 | 4.87 |
| 16 | Left | 8.03 | 3.18 | 4.86 |
| | Right | 7.98 | 3.18 | 4.80 |

**Table V: Statistical Summary of the $log_{10}$ Recovery Values Following Application of Test Article #2, PURELL® Hand Sanitizing Gel VF481 (9900-501; Lot Number 306273), per Test Article Configuration #3 Application Procedure (two pumps [approximately 3.0 mL], pat-dry hands with paper towels, then an additional one pump [approximately 1.5 mL] rubbed until dry)**

| Sample | Sample Size | Mean | Standard Deviation | 95% Confidence Interval |
|---|---|---|---|---|
| Baseline | 24 | 7.79 | 0.33 | 7.65 to 7.93 |
| Wash 1 | 24 | 3.18 | 0.00 | 3.18 to 3.18 |
| Wash 1 $Log_{10}$ Reduction | 24 | 4.61 | 0.33 | 4.47 to 4.75 |

**Table VI: $Log_{10}$ Values and $Log_{10}$ Reduction from Baseline Values by Subject and Hand Following Application of Test Article #2, PURELL® Hand Sanitizing Gel VF481 (9900-501; Lot Number 306273), per Test Article Configuration #3 Application Procedure (two pumps [approximately 3.0 mL], pat-dry hands with paper towels, then an additional one pump [approximately 1.5 mL] rubbed until dry)**

| Subject | Side | Baseline $log_{10}$ Values | Application 1 $log_{10}$ Values | Application 1 $log_{10}$ Reduction from Baseline |
|---|---|---|---|---|
| 1 | Left | 7.90 | 3.18 | 4.73 |
| | Right | 7.96 | 3.18 | 4.78 |
| 2 | Left | 7.58 | 3.18 | 4.40 |
| | Right | 7.71 | 3.18 | 4.53 |
| 3 | Left | 7.95 | 3.18 | 4.78 |
| | Right | 7.98 | 3.18 | 4.79 |
| 6 | Left | 7.99 | 3.18 | 4.82 |
| | Right | 7.95 | 3.18 | 4.77 |
| 7 | Left | 8.19 | 3.18 | 5.02 |
| | Right | 8.23 | 3.18 | 5.06 |
| 8 | Left | 7.89 | 3.18 | 4.71 |
| | Right | 7.90 | 3.18 | 4.73 |
| 10 | Left | 7.34 | 3.18 | 4.16 |
| | Right | 7.21 | 3.18 | 4.03 |
| 11 | Left | 7.95 | 3.18 | 4.77 |
| | Right | 7.89 | 3.18 | 4.71 |
| 13 | Left | 7.75 | 3.18 | 4.58 |
| | Right | 7.85 | 3.18 | 4.68 |
| 14 | Left | 7.07 | 3.18 | 3.89 |
| | Right | 6.98 | 3.18 | 3.81 |
| 15 | Left | 7.80 | 3.18 | 4.62 |
| | Right | 7.73 | 3.18 | 4.55 |
| 16 | Left | 8.01 | 3.18 | 4.83 |
| | Right | 8.16 | 3.18 | 4.98 |

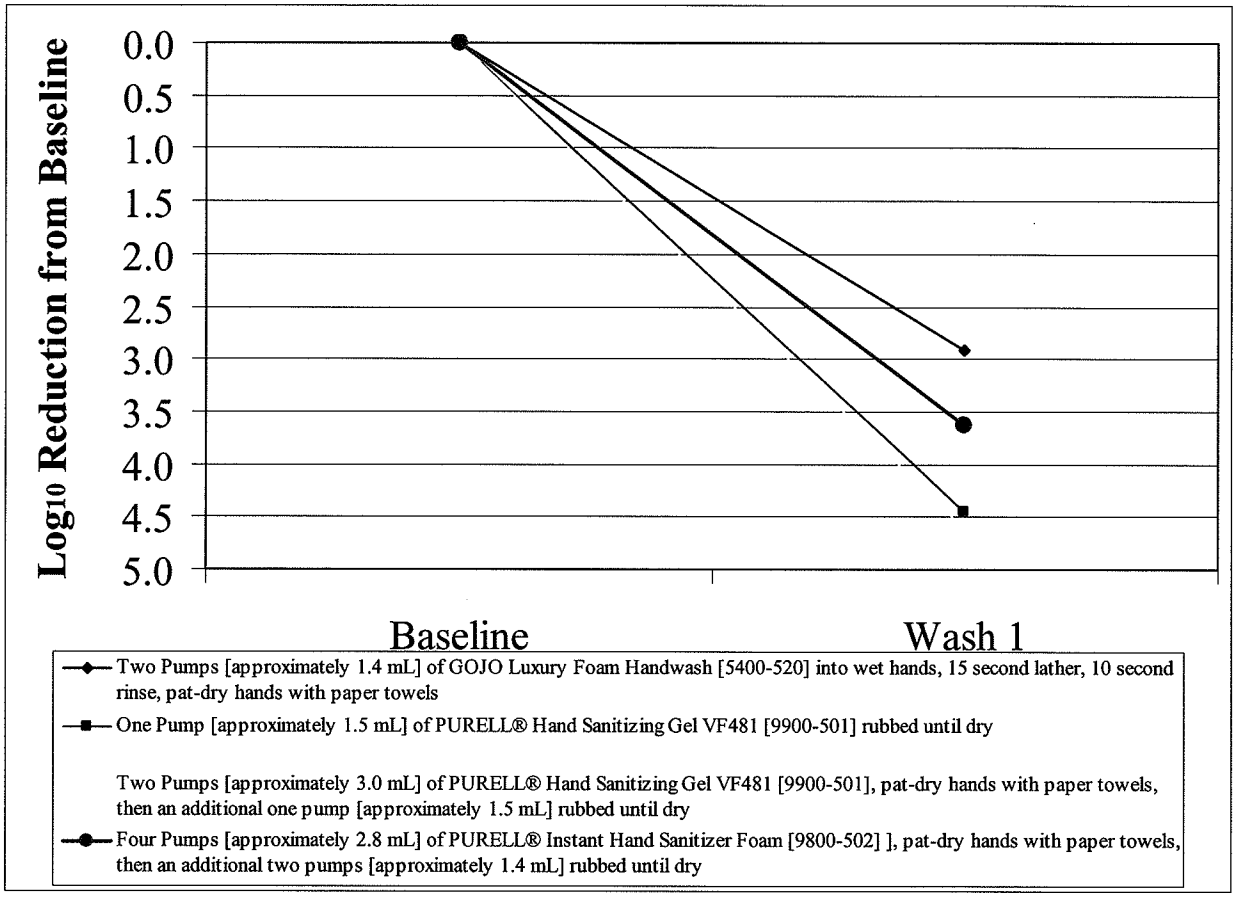
**Table VII: Statistical Summary of the $log_{10}$ Recovery Values Following Application of Test Article #3, PURELL® Instant Hand Sanitizer Foam (9800-502; Lot Number 320887), per Test Article Configuration #4 Application Procedure (four pumps [approximately 2.8 mL], pat-dry hands with paper towels, then an additional two pumps [approximately 1.4 mL] rubbed until dry)**

| Sample | Sample Size | Mean | Standard Deviation | 95% Confidence Interval |
|---|---|---|---|---|
| Baseline | 24 | 7.80 | 0.34 | 7.66 to 7.94 |
| Wash 1 | 24 | 4.16 | 0.56 | 3.93 to 4.40 |
| Wash 1 $Log_{10}$ Reduction | 24 | 3.64 | 0.57 | 3.40 to 3.88 |

**Table VIII: $Log_{10}$ Values and $Log_{10}$ Reduction from Baseline Values by Subject and Hand Following Application of Test Article #3, PURELL® Instant Hand Sanitizer Foam (9800-502; Lot Number 320887), per Test Article Configuration #4 Application Procedure (four pumps [approximately 2.8 mL], pat-dry hands with paper towels, then an additional two pumps [approximately 1.4 mL] rubbed until dry)**

| Subject | Side | Baseline $log_{10}$ Values | Application 1 $log_{10}$ Values | Application 1 $log_{10}$ Reduction from Baseline |
|---|---|---|---|---|
| 1 | Left | 7.98 | 4.19 | 3.79 |
| | Right | 8.03 | 4.55 | 3.48 |
| 2 | Left | 7.89 | 4.53 | 3.37 |
| | Right | 7.79 | 4.51 | 3.28 |
| 3 | Left | 7.64 | 3.79 | 3.85 |
| | Right | 7.73 | 4.23 | 3.51 |
| 6 | Left | 7.79 | 4.72 | 3.08 |
| | Right | 7.80 | 4.81 | 2.99 |
| 7 | Left | 8.08 | 3.18 | 4.90 |
| | Right | 8.10 | 4.03 | 4.07 |
| 8 | Left | 8.18 | 4.23 | 3.95 |
| | Right | 8.19 | 5.03 | 3.16 |
| 10 | Left | 7.32 | 3.18 | 4.14 |
| | Right | 7.02 | 3.18 | 3.84 |
| 11 | Left | 7.96 | 4.57 | 3.39 |
| | Right | 7.88 | 4.58 | 3.29 |
| 13 | Left | 7.91 | 4.27 | 3.64 |
| | Right | 7.80 | 4.58 | 3.22 |
| 14 | Left | 7.21 | 4.85 | 2.36 |
| | Right | 7.03 | 3.79 | 3.24 |
| 15 | Left | 7.84 | 3.96 | 3.87 |
| | Right | 7.94 | 3.18 | 4.75 |
| 16 | Left | 8.06 | 4.23 | 3.83 |
| | Right | 8.11 | 3.79 | 4.32 |

**Figure 1: Graphical Presentation of the Mean $log_{10}$ Reductions from Baseline Following Application of Each Test Article per the Four Test Article Application Procedures**




## 17.0 CONCLUSION:

The critical index for this study was a 2.0 $log_{10}$ reduction in baseline populations after product application.

For Test Article #1, GOJO Luxury Foam Handwash (5400-520; Lot Number 322503), applied per Test Article Configuration #1 Application Procedure (two pumps [approximately 1.4 mL] of Test Article #1 into wet hands, 15-second lather, 10-second rinse, pat-dry hands with paper towels), the bacterial populations recovered after product application were reduced significantly from baseline populations. Test Article #1 produced a mean $log_{10}$ reduction of 2.92 after product application and met the critical index of the study.

For Test Article #2, PURELL® Hand Sanitizing Gel VF481 (9900-501; Lot Number 306273), applied per Test Article Configuration #2 Application Procedure (one pump [approximately 1.5 mL] of Test Article #2 rubbed until dry), the bacterial populations recovered after product application were reduced significantly from baseline populations. Test Article #2, applied per Test Article Configuration #2 Application Procedure, produced a mean $log_{10}$ reduction of 4.44 after product application and met the critical index of the study.

For Test Article #2, PURELL® Hand Sanitizing Gel VF481 (9900-501; Lot Number 306273), applied per Test Article Configuration #3 Application Procedure (two pumps [approximately 3.0 mL] of Test Article #2, pat-dry hands with paper towels, then an additional one pump [approximately 1.5 mL] rubbed until dry), the bacterial populations recovered after product application were reduced significantly from baseline populations. Test Article #2, applied per Test Article Configuration #3 Application Procedure, produced a mean $\log_{10}$ reduction of 4.61 after product application and met the critical index of the study.

For Test Article #3, PURELL® Instant Hand Sanitizer Foam (9800-502; Lot Number 320887), applied per Test Article Configuration #4 Application Procedure (four pumps [approximately 2.8 mL] of Test Article #3, pat-dry hands with paper towels, then an additional two pumps [approximately 1.4 mL] rubbed until dry), the bacterial populations recovered after product application were reduced significantly from baseline populations. Test Article #3 produced a mean $\log_{10}$ reduction of 3.64 after product application and met the critical index of the study.

## 18.0 LABORATORY PERSONNEL:

The following employees of BioScience Laboratories, Inc., were involved in the testing or ancillary support of this Study. The laboratory personnel have been appropriately trained, and their training records are on file in the Quality Assurance Unit at the Testing Facility.

| | |
|---|---|
| STUDY DIRECTOR: | Robert R. McCormack<br>Microbiologist |
| ASSOCIATE STUDY DIRECTOR: | Kendra Drake<br>Microbiologist |
| Tammy Anderson<br>IRB Coordinator | Nathan Nash<br>Microbiologist |
| Jessica Baumgartner<br>Microbiologist | Ron Neibauer<br>Manager of Clinical Laboratories |
| Amanda Berry<br>Supervisor of Subject Recruitment | Jeana Paulson<br>Microbiologist |
| Stephanie Cebulla<br>Laboratory Support Technician | Stephanie Scarff<br>Laboratory Support Technician |
| Collette Duley<br>Microbiologist | Amanda Shaffer<br>Microbiologist |
| B. Cole Irvin<br>Microbiologist | Jessica Sheehy<br>Microbiologist |
| Patricia A. Mays Suko<br>Supervisor of Laboratory Support | Clare Wilson<br>Microbiologist |

## 19.0 QUALITY ASSURANCE PERSONNEL:

Alicia Bogert
Quality Assurance Associate/Product Handling

Scott D. Ferraro
Manager of Quality Control

Amy L. Juhnke
Manager of Quality Assurance/Document Control

John A. Mitchell, Ph.D.
Director of Quality Assurance

Janis Smoke
Quality Assurance Associate

## 20.0 DOCUMENTATION AND RECORD-KEEPING:

All documentation and records were compiled, analyzed, and will be retained by BioScience Laboratories, Inc., at its facility in Bozeman, Montana. All raw data for this study, as well as the Final Report, will be retained in safe storage by the Testing Facility for a period of at least three years. BioScience Laboratories, Inc. will notify the Sponsor before any documents or records are destroyed.

## 21.0 ACCEPTANCE:

**BIOSCIENCE LABORATORIES, INC.** (TESTING FACILITY)
300 N. Willson Avenue
Bozeman, Montana 59715

President & CEO: Daryl S Paulson — 03-12-09
Daryl S. Paulson, Ph.D. — Date

Principal Study Director: Robert R. McCormack — 03/12/09
Robert McCormack — Study Completion Date

Associate Study Director: Kendra Drake — 03/12/09
Kendra Drake — Date

Senior Clinical Director: Christopher M. Beausoleil — 03/12/09
Christopher M. Beausoleil, CCRP — Date

**QUALITY ASSURANCE STATEMENT:**

This study was inspected by the Quality Assurance Unit, and reports were submitted to the Study Director and Management in accordance with Standard Operating Procedures, as follows:

| Phase | Date |
|---|---|
| Neutralization Assay | 02/13/09 |
| Product Testing | 02/18/09 |
| Data Audit | 03/03/09 and 03/04/09 |
| Final Report Review | 03/11/09 |
| Reports to Study Director and Management | 02/13/09, 02/18/09, and 03/12/09 |

This study was conducted in compliance with Good Laboratory Practices standards, as described by the FDA (reference CFR 21 Parts 50, 56, 312, and 314), with the following exception: test article preparations were not analyzed at BioScience Laboratories, Inc., to confirm concentration, stability, or homogeneity.

Quality Assurance/ Associate: Janis E. Smoke — 03/12/09
Janis Smoke — Date

## INDEX OF ADDENDA

I GIRB-Approved Protocol #081211-150

II Qualification Criteria Questionnaire for Study 081211-150
Qualification Criteria of the Subject for Study 081211-150

III Sampling Data Sheets for Healthcare Personnel Handwash Study 081211-150
Irritation Evaluations for Study 081211-150

IV Neutralization Evaluation
- Neutralization Evaluation Results for Study 081211-150
- Project Notes (Form No. 95-G-001)
- Neutralization Evaluation Data Sheets for Protocol 081211-150
- Neutralization Statistics

V Q-Count™ Plate Counter Data Sheets (Form No. 00-L-009)
Q-Count™ Plate Count Data

VI Statistical Analysis

VII Study Notes and General Records
- Project Notes (Form No. 95-G-001)
- Age Calculation and Demographics Worksheet
- Study 081211-150 Randomization Scheme
- Clinical Trials Equipment Tracking Forms (Form No. 01-L-009)
- Clinical Trials Supplies Tracking Forms (Form No. 01-L-008)
- Water Temperature Monitoring Sheet (Form No. 96-CT-017)
- Incubator Log Forms (Form No. 96-L-008)
- Refrigerator Log Form (Form No. 96-L-015)
- Inoculum Preparation Tracking Form - Solid Media Preparation (Form No. 07-CT-002)
- Inoculum Preparation Tracking Form - Flask Preparation (Form No. 07-CT-001)
- Autoplate® 4000 Data Sheet for Healthcare Personnel Handwash Study 081211-150
- Media/Diluent Tracking Forms (Form No. 97-L-007)

VIII Product Information
- Product Receipt Log (Form No. 92-L-023)
- Sample Submission Form and Document Compliance Statement (Form No. 94-G-007)
- Material Safety Data Sheet (MSDS)
- Product-Tracking Forms (Form No. 93-L-029)

# Sequential Application of Hand Antiseptic for Use in No-Water Situations (dubbed SaniTwice)

## A New Hand Hygiene Option

**Robert R. McCormack**
**BioScience Laboratories, Inc.**
**March 25, 2009**

# Background

- Current FDA Model Food Code requires food handlers to wash with soap and water to maintain clean hands.
- A reliable method of hand sanitization is needed for remote locations where water is not readily available. Among the many situations is the need to cleanse hands between changes of single-use gloves in no-water locations.

# Background

- To meet this need, the “Sequential Application of Hand Antiseptic for Use in No-Water Situations” was developed:
  - A method of cleansing and sanitizing light to moderately soiled hands when soap and water are unavailable
  - Purpose is the removal and reduction of transient microorganisms from the hands

# Study Objectives

- To demonstrate the antimicrobial effectiveness of this method as compared to standard handwashing with soap and water
- To evaluate the comparative effectiveness of various hand sanitizers for the reduction of bacteria when used in this methodology.

# Modified Handwash Method

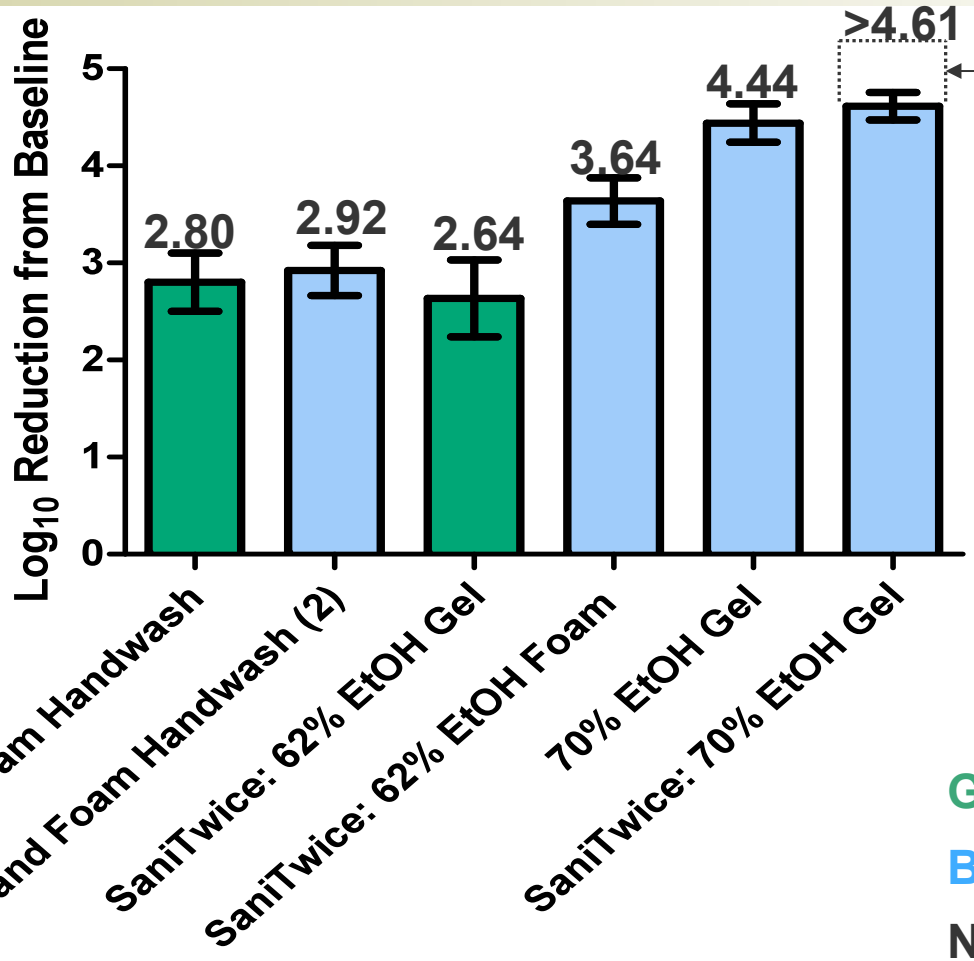
## ASTM E1174

**Step 1:** Inoculate hands with about $1x10^9$ *Escherichia coli* (ATCC #11229) suspended in beef broth

(moderate soil conditions)

**Step 2:** Apply test product according to label application instructions

# Results




**Actual LR value is likely higher**

(LR value reached detection limit of study)

**Green: Study #1**

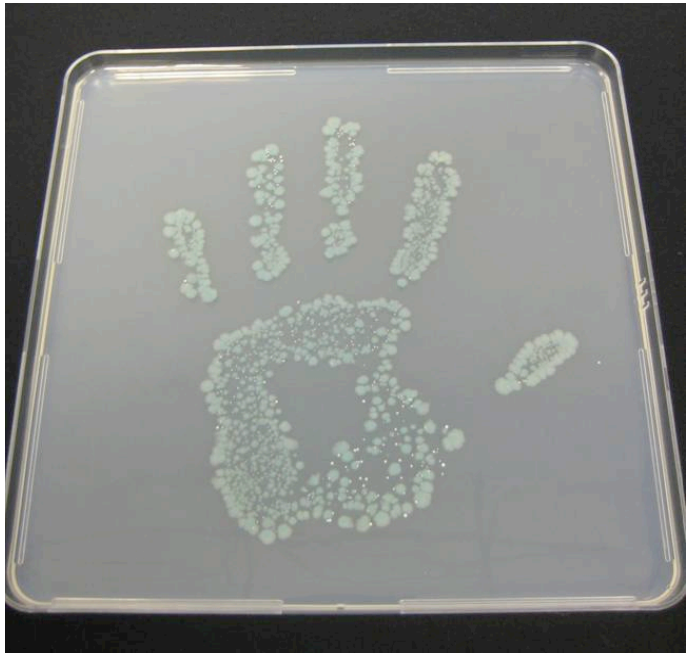
**Blue: Study #2**

**N ≥ 10**

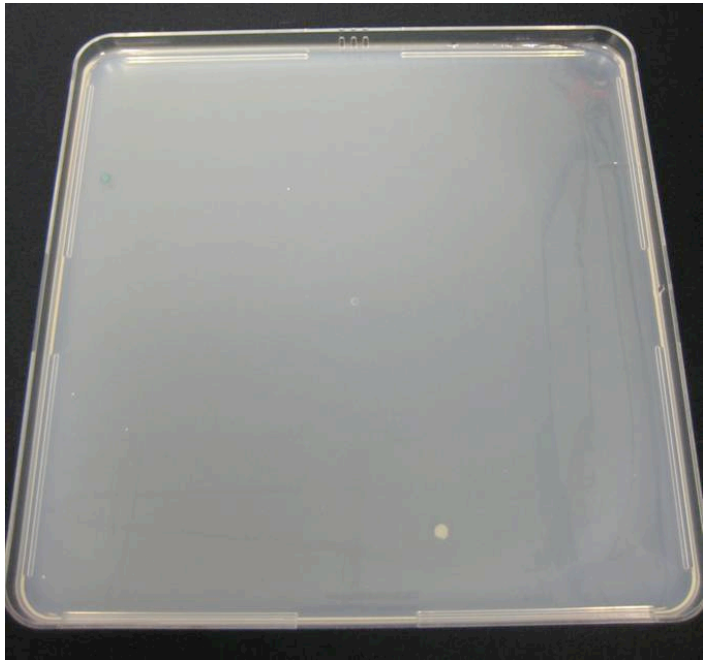
# Statistical Analysis

- The antimicrobial efficacy of SaniTwice with 62% EtOH gel is equivalent to a typical bland handwash product
- SaniTwice with 62% EtOH foam is significantly better at reducing microorganisms on the hands than a typical bland handwash product
- SaniTwice with a high efficacy 70% EtOH gel is significantly better at reducing microorganisms on the hands than a typical bland handwash and the SaniTwice with 62% EtOH

# What Does Your Hand Look Like After SaniTwice?



Hand contaminated with *E. coli*



Hand after performing SaniTwice with 70% EtOH gel

# Conclusions

- Sequential Application of Hand Antiseptic for Use in No-Water Situations (SaniTwice) is an acceptable alternative to handwashing with soap and water
  - All SaniTwice regimens tested were equivalent or better at reducing the number of microorganisms on the hands than standard washing with soap and water

# Conclusions

- There are statistically differentiated SaniTwice options based on antimicrobial efficacy requirements:
  - Good (62% EtOH gel)
  - Better (62% EtOH foam)
  - Best (High efficacy 70% EtOH gel)

# Conclusions

- Use of a high efficacy product (70% EtOH gel) with the SaniTwice method results in superior reduction of bacteria on the hands
  - Complete kill of microorganisms (>4.61 LR)
  - SaniTwice is effective at cleansing and sanitizing the hands whereas using the product according to label instructions only sanitizes and does not clean the hands

# Why Use SaniTwice?

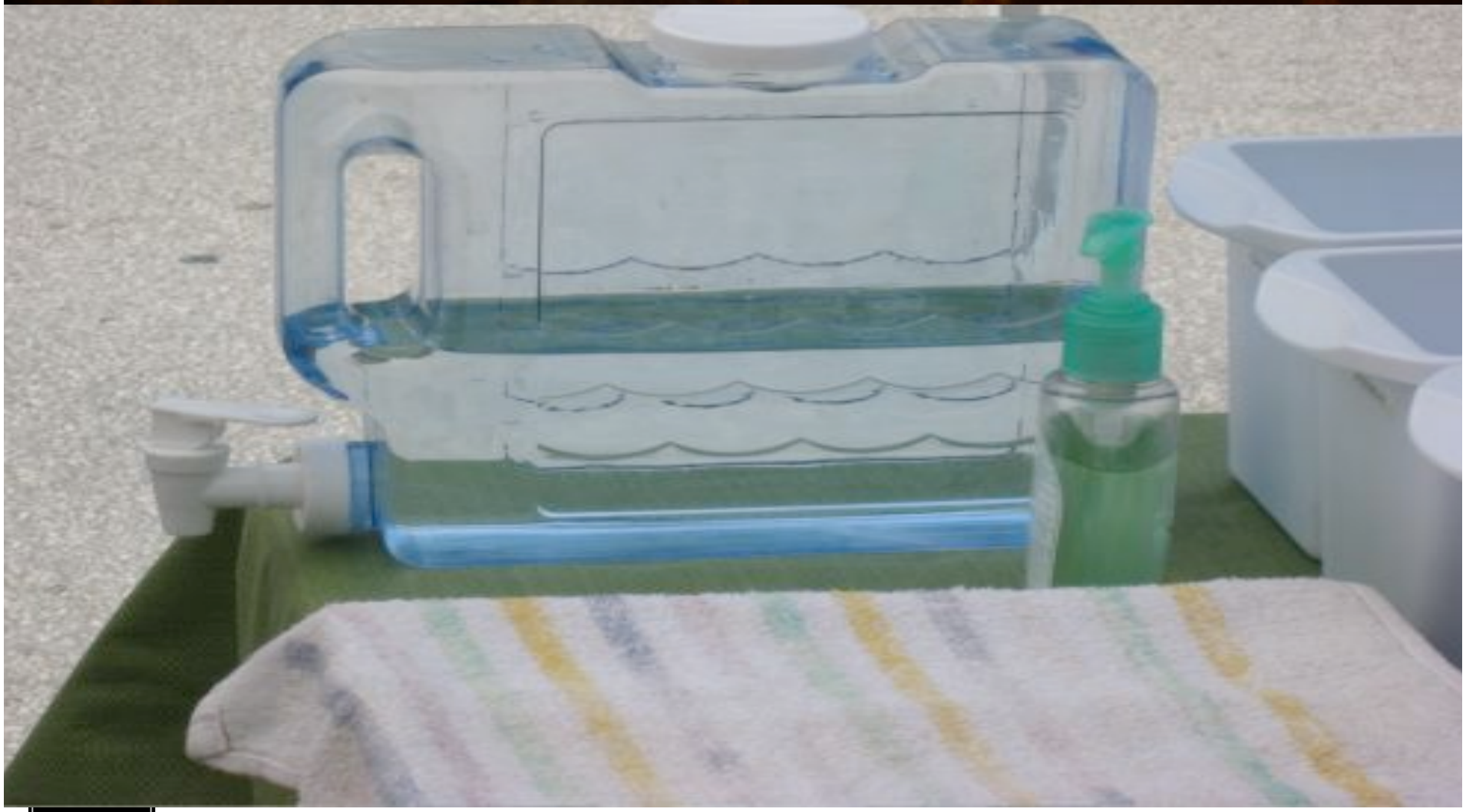
- SaniTwice is a simple method that requires only a supply of hand sanitizer and paper towels
- Use of the SaniTwice method in remote locations is an acceptable alternative to handwashing
- Use of the SaniTwice method with a high efficacy hand sanitizer will result in improved sanitization over soap and water alone

# Why Use SaniTwice?

- It is actually used as confirmed by extended field testing under the guidance of the Southern Nevada Health District
- Superior to currently approved hand hygiene interventions for no-water situations as seen in the following two photos taken in Illinois
- (Other photos are available from the 2008 CFP venue in San Antonio Texas.)

# Food Code Approved Intervention

# Food Code Approved Intervention



# Contact Information

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Principal Study Director in Clinical Trial Laboratories

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# SaniTwice®: A Hand Hygiene Solution for Food Handlers When Water is Unavailable

Sarah Edmonds[1], Cara Bondi[1], Robert McCormack[3], David Macinga[1], James Arbogast[1], James Mann[2], Michael Dolan[1]

1. GOJO Industries, Inc.
2. Handwashing for Life
3. BioScience Laboratories

IAFP 13July09
T2-08

# Agenda

- Why “Sink-less” Hand Hygiene
- The SaniTwice® Solution
- FDA Model Food Code Considerations
- Conclusions

# “Sink-less” Hand Hygiene

- Definition: hand hygiene (degerming) performed in settings where water is not available or is in limited supply
- Historically, a challenge without practical and effective solutions

# Food Safety Challenge: Portable Bars

# Food Safety Challenge: Military Buffet Line

# Food Safety Challenge: Community Event

# Food Safety Challenge: Cookoff

# Food Safety Challenge: The Picnic

# Food Safety Challenge: Symposium Serving Line

# “Sink-less” Hand Hygiene

- Why not just have portable hand washing (i.e., the current paradigm)?

# Food Safety Reality: Trickle Handwashing

# Food Safety Reality: Trickle Handwashing

# Is This Effective Hand Hygiene?

# The SaniTwice® Solution

# The SaniTwice Solution

- A reliable method of hand sanitization for (remote) locations where water is not available or in short supply
- A two stage method, “clean and kill”, for cleansing and sanitizing light to moderately soiled hands when soap and water are unavailable
- Purpose is the removal and reduction of transient microorganisms from the hands
- Benefit is reduction in risk of foodborne illness due to inadequate hand hygiene

# SaniTwice Method

## Step 1:

Apply excess of hand sanitizer (about 3 mL) and "wash" hands vigorously for 15 seconds


## Step 2:

Remove remaining hand sanitizer and soil forcefully with paper towel while hands are still wet

## Step 3:

Rub recommended amount (about 1.5 mL) of hand sanitizer on hands until dry

# The SaniTwice Solution

**Performance Study:**

- *In vivo* microbiological efficacy

**Study Objectives:**

- Determine the effectiveness of the SaniTwice method as compared to standard handwashing with soap and water
- Compare effectiveness of various hand sanitizers when used in the SaniTwice methodology

# Test Product Configurations

| Test Product | Active | Application Method |
|---|---|---|
| Non-Antimicrobial Foam Handwash | N/A | Wash (Apply ~1.5ml, wash for 15s, rinse for 10s, towel dry) |
| Instant Hand Sanitizer Gel | 62% ethanol | SaniTwice |
| Instant Hand Sanitizer Foam | 62% ethanol | SaniTwice |
| Advanced Formula Instant Hand Sanitizer Gel | 70% ethanol | Sanitize (Apply ~1.5ml, rub until dry) |
| Advanced Formula Instant Hand Sanitizer Gel | 70% ethanol | SaniTwice |

*Two studies were conducted at BioScience Laboratories (2008-09)*

# Modified Handwash Method

**ASTM E1174**

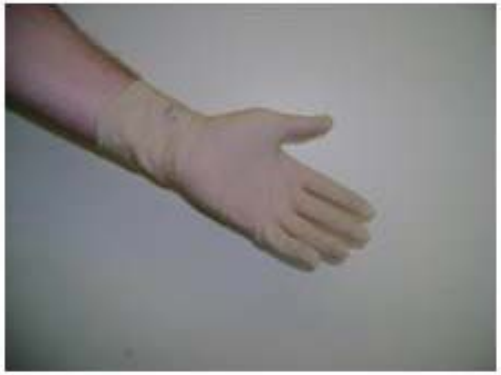
**Step 1:** Contaminate hands with about $1x10^9$ *Escherichia coli* (ATCC #11229) suspended in beef broth

(moderate soil conditions)

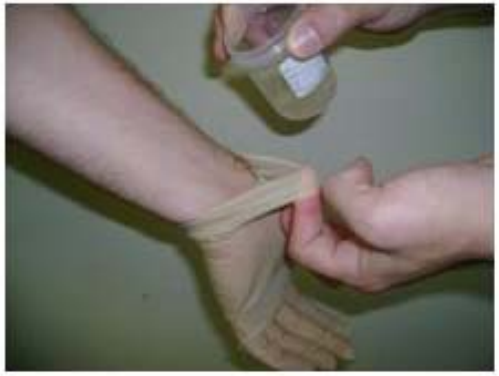
**Step 2:** Apply test product according to application instructions

# Bacterial Measurement Steps

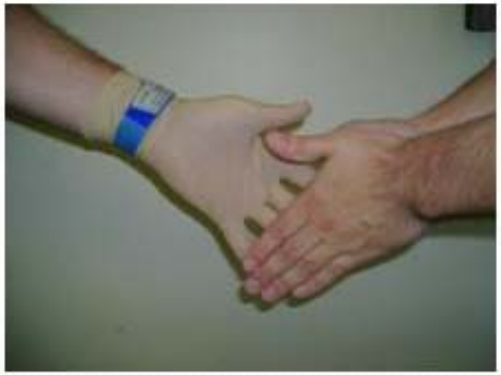
Step 3. Placement of sterile, latex glove



Step 4. Addition of sterile sampling fluid (GJ)



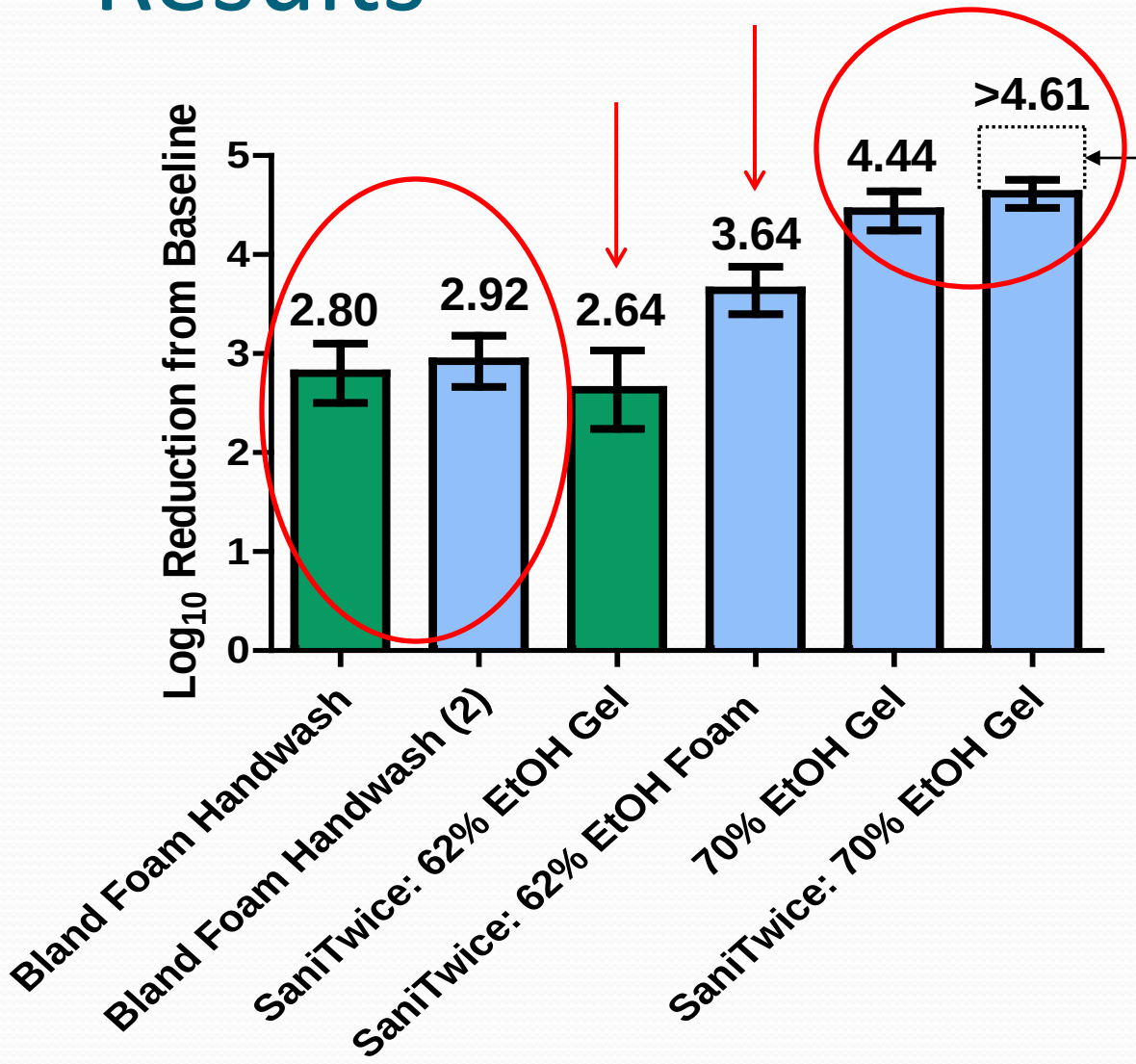
Step 5. Massage hand for 60 seconds



Step 6. Remove sample of glove-juice

**Step 7:** Serially dilute in neutralizing solution, plate on MacConkey Agar, grow overnight and compare to baseline values to calculate log reductions

# Results




**Actual LR value is likely higher**

(LR value reached detection limit of study)

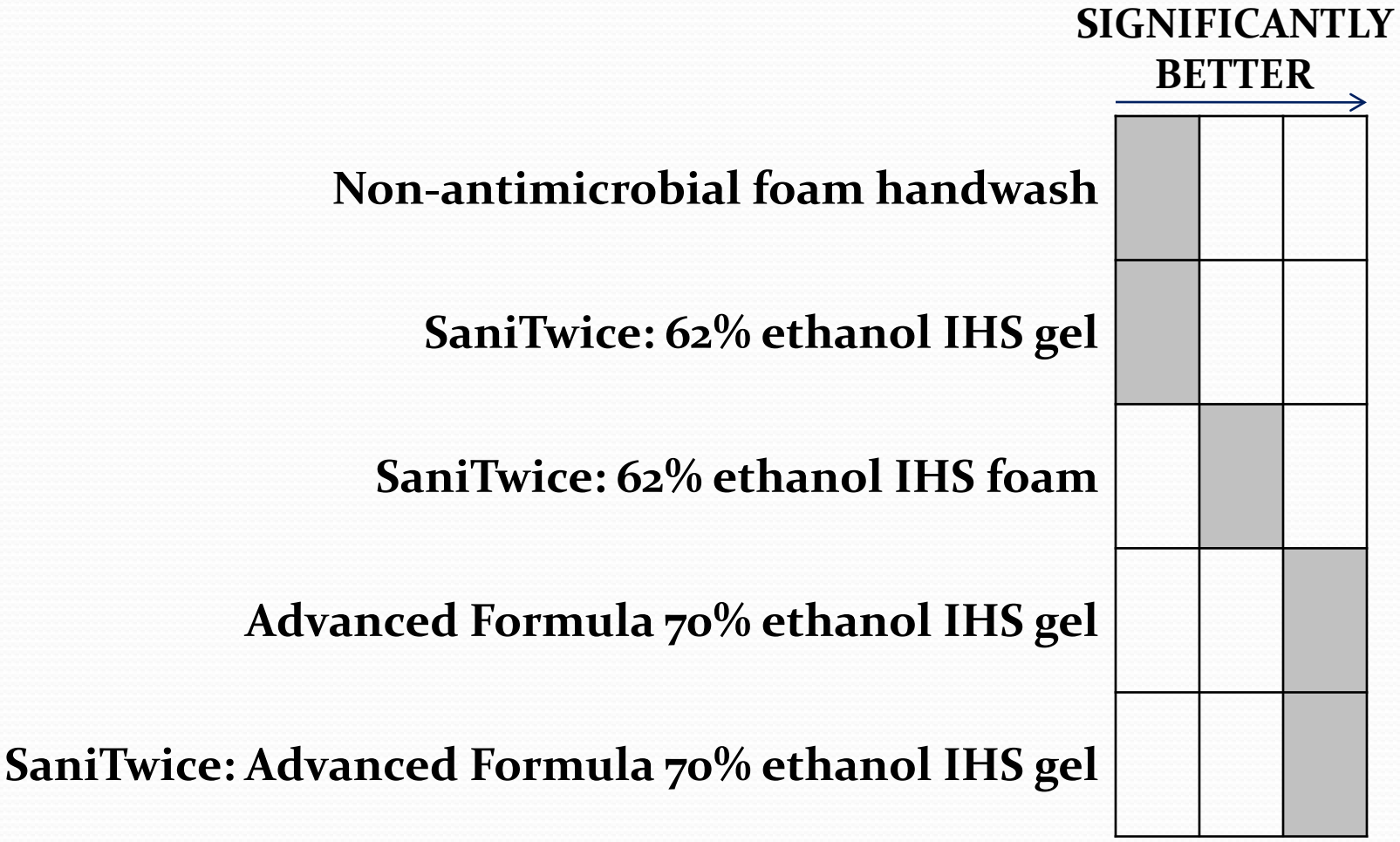
**Green: Study #1**

**N=11**

**Blue: Study #2**

**N=12**

# Statistical Analysis




1 way ANOVA followed by post-hoc analysis ($p<0.05$)

# What Does Your Hand Look Like After SaniTwice?

**Hand contaminated with *E. coli***

**Hand after performing SaniTwice with Advanced Formula 70% ethanol gel**

# Conclusions

- SaniTwice is an acceptable alternative to handwashing with soap and water
  - All SaniTwice regimens tested were equivalent or better than standard washing with soap and water
- SaniTwice is a good substitute to trickle handwashing for “sink-less” food handling situations

# Conclusions

- There are statistically differentiated SaniTwice options based on efficacy results:
  - Good (62% ethanol IHS gel)
  - Better (62% ethanol IHS foam)
  - Best (Advanced Formula 70% ethanol IHS gel)

# Conclusions

- Use of the Advanced Formula 70% ethanol gel with the SaniTwice method resulted in superior reduction of bacteria on the hands compared to washing with a non-antimicrobial handwash and water alone
  - Complete kill of microorganisms (>4.61 LR)

- The Advanced Formula 70% ethanol gel was highly effective at reducing bacteria on the hands when used alone; however, the SaniTwice method has the additional benefit of skin cleansing and soil removal

# SaniTwice Field Research Study:
## The Venetian Portable Bars

High User Acceptability and Compliance to the SaniTwice Regimen

# FDA Model Food Code (2005)

- Section 2-301.16 outlines parameters for hand antiseptics:
  - "applied only to hands that are cleaned as specified under § 2-301.12."

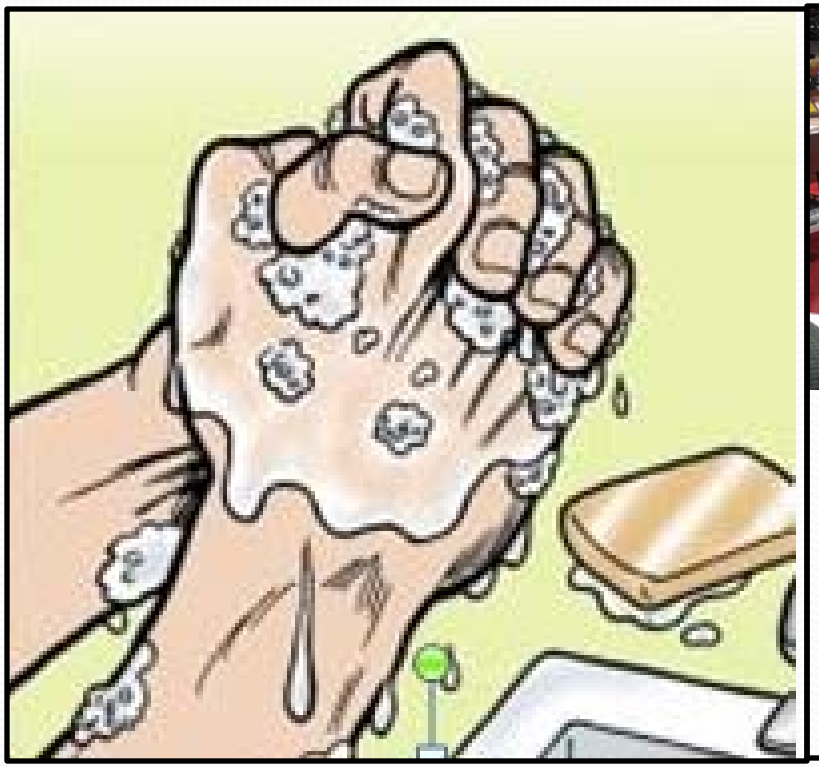
**There is now clear scientific and practical rationale for including the SaniTwice approach in the Food Code**

- SaniTwice has been shown to be an effective hand hygiene regimen, equivalent in degerming to handwashing with soap and water as specified in Section 2-301.12(B)

# In Conclusion…

Prayer is Good;

SaniTwice, Even Better!



# GOJO INDUSTRIES

Internal Communication

**CONFIDENTIAL**

**TO:** Geo Money, Chris Fricker, Amy Stokes

**FYI:** Dave Macinga, Jim Arbogast, Mike Dolan, Jim Mann

**FROM:** Sarah Edmonds

**SUBJECT:** **TEST RESULTS FOR HEAVY SOIL PILOT SANITWICE STUDY**

**DATE:** December 2, 2009

**STUDY OBJECTIVES:**

- **Preliminary evaluation of whether SaniTwice is as effective as handwashing for reducing bacteria on heavily soiled hands**
- **Determine optimal soil type for full heavy soil SaniTwice study**

| TEST PRODUCT CONFIGURATIONS: | ACTIVE: |
|---|---|
| GOJO Luxury Foam Handwash (5200-502)<br>Wash for 15s with 2 pumps (~1.4 ml), rinse for 10s, towel dry | N/A |
| SaniTwice with PURELL Foam (9800-504)<br>Apply 4 pumps (~2.8 ml), towel dry, apply 2 pumps (~1.4 ml) and rub until dry | 62% ethanol |

**TEST METHOD:** A modification of the USFDA Tentative Final Monograph for: *Effectiveness Testing of an Antiseptic Handwash or Health-Care Personnel Handwash* (FR59:116, 17 June 94, pp.31448-31450) using *Escherichia coli* (ATCC #11229) suspended in either chicken chunks or raw hamburger patties

**TESTING LAB:** Bioscience Laboratories, Bozeman, Montana, Study #091010-150

**RESULTS:**

| Test Configuration | LR | SD | 95% CI |
|---|---|---|---|
| Chicken Chunk Contamination | | | |
| GOJO Luxury Foam Handwash | 2.96 | 0.48 | 2.62-3.30 |
| SaniTwice with PURELL Foam | 3.32 | 0.43 | 3.01-3.63 |
| Raw Hamburger Contamination | | | |
| GOJO Luxury Foam Handwash | 2.58 | 0.41 | 2.28-2.87 |
| SaniTwice with PURELL Foam | 2.69 | 0.34 | 2.45-2.93 |
| Results Below from Previous SaniTwice Study # 081211-150 (beef broth as soil load) | | | |
| GOJO Luxury Foam Handwash | 2.92 | 0.61 | 2.66-3.18 |
| SaniTwice with PURELL Foam | 3.64 | 0.57 | 3.40-3.88 |

LR=log reduction from baseline; SD=standard deviation; CI=confidence interval; N=10

**CONCLUSIONS:**

- **SaniTwice was as effective as handwashing for reducing bacteria on heavily soiled hands**
  - **Effective with chicken and raw beef soils**
- **As expected the raw beef appears to be a more difficult soil to penetrate**
  - **Both the handwash and PURELL Foam SaniTwice achieved about a 0.5 higher log reduction with the chicken than the beef**

**NEXT STEPS:**

- Design and conduct full SaniTwice study with raw hamburger to represent "worst-case" heavy soils found in foodservice (SE)