

**FINAL REPORT**

**VIRUCIDAL EFFICACY SUSPENSION TEST**

**Coxsackie Virus B6**

**TEST AGENT**

**3M Avagard™ 9250 Antiseptic Handrub**

**Author**

**Salimatu Lukula, M.S.**

**Performing Laboratory**

**MicroBioTest**

**Division of Microbac Laboratories, Inc.**

**105 Carpenter Drive**

**Sterling, Virginia 20164**

**Laboratory Project Identification Number**

**109-228**

**Sponsor**

**3M Health Care**

**3M Infection Prevention Division**

**222 Tian Lin Rd.**

**Shanghai 200233, China**

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**COMPLIANCE STATEMENT**

This study meets the requirements for 21 CFR § 58 with the following exceptions:

- Information on the identity, strength, purity, stability, uniformity, and dose solution analysis of the test agent resides with the sponsor of the study.

The following technical personnel participated in this study:

George Drakeley, Salimatu Lukula

Study Director: MicroBioTest



Salimatu Lukula, M.S.

2/29/16

Date

**QUALITY ASSURANCE UNIT STATEMENT**

Title: VIRUCIDAL EFFICACY SUSPENSION TEST- Cocksackie Virus B6

The Quality Assurance Unit of MicroBioTest has inspected the Project Number 109-228 in compliance with current Good Laboratory Practice regulations (21 CFR § 58).

The dates that inspections were made and the dates that findings were reported to management and to the study director are listed below.

<u>PHASE INSPECTED</u>	<u>DATE OF INSPECTION</u>	<u>DATE REPORTED TO STUDY DIRECTOR</u>	<u>DATE REPORTED TO MANAGEMENT</u>
Protocol	2/02/16	2/02/16	2/02/16
In-Process	2/02/16	2/02/16	2/02/16
Final Report	2/24/16	2/24/16	2/24/16



Savanna Green  
Quality Assurance Associate II

2-29-16

Date

## **TEST SUMMARY**

**TITLE:** VIRUCIDAL EFFICACY SUSPENSION TEST – Coxsackie Virus B6

**STUDY DESIGN:** This study was performed according to the signed protocol and project sheet(s) issued by the Study Director (See Appendix).

**TEST AGENT:** 3M Avagard™ 9250 Antiseptic Handrub, received at MicroBioTest on 12/08/2015, and assigned DS No. F1071

**SPONSOR:** 3M Health Care  
3M Infection Prevention Division  
222 Tian Lin Rd.  
Shanghai 200233, China

## TEST CONDITIONS

Challenge virus:

Coxsackie Virus B6, strain: Coxsackievirus B6 antiserum, ATCC VR-1037

Host:

LLC-MK2 cells, ATCC CCL-7.1

Active ingredients:

Chlorhexidine Gluconate 0.45-0.55% (w/v) Ethanol 63.1-77% (v/v)

Dilution Medium:

Minimum Essential Medium (MEM) + 2% Fetal Bovine Serum (FBS)

Neutralizer used:

MEM + 10% FBS + 1% Polysorbate 80 + 1% Tamol + 0.5% Lecithin

Contact times:

1 minute and 5 minutes

Contact temperature:

Ambient Room temperature  $20\pm 1^{\circ}\text{C}$  ( $21^{\circ}\text{C}$  actual)

Test agent application:

Suspension test – direct mixing (2.7 mL aliquot of the test agent was spiked with 0.3 mL of the virus suspension and thoroughly mixed by vortex mixing)

Media and reagents:

MEM + 2% FBS

MEM + 10% FBS + 1% Polysorbate 80 + 1% Tamol + 0.5% Lecithin

## STUDY DATES AND FACILITIES

The laboratory phase of this test was performed at MicroBioTest Division of Microbac Laboratories, Inc., 105 Carpenter Drive, Sterling, VA 20164. Testing was laboratory initiated on 02/02/16 and was completed on 02/05/16. The study director signed the protocol on 02/02/16. The study completion date is the date the study director signed the final report.

All changes or revisions of the protocol were documented and signed by the study director, and maintained with the protocol.

## RECORDS TO BE MAINTAINED

All testing data, protocol, protocol modifications, test agent records, the final report, and correspondence between MicroBioTest Division of Microbac Laboratories, Inc. and the sponsor will be stored in the archives at MicroBioTest Division of Microbac Laboratories, Inc., 105 Carpenter Drive, Sterling, VA 20164, or at a controlled facility off site.

## CALCULATION OF TITER

The 50% tissue culture infectious dose per mL (TCID<sub>50</sub>/mL) was determined using the Spearman-Kärber method using the following formula:

$$m = x_k + \left( \frac{d}{2} \right) - d \sum p_i$$

where:

m = the logarithm of the dilution at which half the wells are infected relative to the test volume

x<sub>k</sub> = the logarithm of the smallest dosage which induces infection in all cultures

d = the logarithm of the dilution factor

p<sub>i</sub> = the proportion of positive results at dilution i

∑p<sub>i</sub> = the sum of p<sub>i</sub> (starting with the highest dilution producing 100% infection)

## RESULTS

Results are presented in Tables 1-3.

The Viral load was determined in the following manner:

Viral Load ( $\log_{10}$  TCID<sub>50</sub>) = Titer ( $\log_{10}$  TCID<sub>50</sub>/mL) +  $\log_{10}$  [Volume (mL) x Volume Correction]

The  $\log_{10}$  Reduction Factor was calculated in the following manner:

$\log_{10}$  Reduction Factor = Initial viral load ( $\log_{10}$ ) – Output viral load ( $\log_{10}$ )

Conversion of  $\log_{10}$  reduction to percent reduction

$\log_{10}$  reduction = A

% reduction = B

$$B = \left[ 1 - \frac{1}{10^A} \right] \times 100$$

Example:

3.5  $\log_{10}$  reduction =  $\{1 - 1/[\text{power}(10, 3.5)]\} \times 100 = 99.97\%$

**RESULTS (continued)****Table 1  
Titer Results**

Sample	Contact time	Replicate	Titer ( $\text{Log}_{10}\text{TCID}_{50}/\text{mL}$ )	Volume (mL)	Volume Correction <sup>a</sup>	Viral Load ( $\text{Log}_{10}\text{TCID}_{50}$ )
Virus Stock Titer Control	N/A	N/A	6.75	-	-	-
Cell Viability Control			no virus was detected, cells remained viable; media was sterile			
Virus Recovery Control	5 minutes	Replicate 1	5.50	3	2	6.28
		Replicate 2	5.25	3	2	6.03
						6.16
3M Avagard™ 9250 Antiseptic Handrub <sup>b</sup>	1 minute	Replicate 1	4.25	3	2	5.03
		Replicate 2	4.00	3	2	4.78
						4.91
	5 minutes	Replicate 1	3.25	3	2	4.03
		Replicate 2	3.00	3	2	3.78
						3.91

<sup>a</sup> Volume correction accounts for the neutralization of the sample post contact time.<sup>b</sup> Cytotoxicity observed at  $10^{-1}$  dilution**Table 2  
Neutralizer Effectiveness/Viral Interference and Cytotoxicity Controls**

Dilutions*	Neutralizer Effectiveness/Viral Interference Control	Cytotoxicity Control
$10^{-1}$	cytotoxicity observed	cytotoxicity observed
$10^{-2}$	virus detected in all inoculated wells	no cytotoxicity observed
$10^{-3}$	virus detected in all inoculated wells	no cytotoxicity observed

\* Dilution refers to the fold of the dilution from the neutralized sample

**Table 3  
Viral Reduction**

Test Agent	Contact Time	Initial Load* ( $\text{Log}_{10}\text{TCID}_{50}$ )	Output Load* ( $\text{Log}_{10}\text{TCID}_{50}$ )	$\text{Log}_{10}$ Reduction*	Percent Reduction*
3M Avagard™ 9250 Antiseptic Handrub	1 minute	6.16	4.91	1.25	94.38
	5 minutes		3.91	2.25	99.44

\* Results represent the average of two replicates.



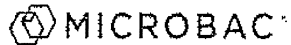
## CONCLUSIONS

3M Avagard™ 9250 Antiseptic Handrub was evaluated for the ability to inactivate Cocksackie Virus B6. MicroBioTest personnel performed the inactivation procedure using Cocksackie Virus B6 to spike the test agent solution. Samples were titrated by 50% tissue culture infectious dose (TCID<sub>50</sub>) endpoint assay using LLC-MK2 cells.

When tested as described, 3M Avagard™ 9250 Antiseptic Handrub (Lot No. 510110D1) inactivated Cocksackie Virus B6 by 1.25 log<sub>10</sub> when the target virus was exposed to the test agent for 1 minute at 21C, and by 2.25 log<sub>10</sub> when the target virus was exposed to the test agent for 5 minutes at 21C.

Table 3 reports the individual Log<sub>10</sub> virus reduction factors for the test agent treatment procedure. All of the controls met the criteria for a valid test. These conclusions are based on observed data.

## **APPENDIX**



MicroBioTest

**MicroBioTest Protocol**  
**VIRUCIDAL EFFICACY SUSPENSION TEST -**  
**Coxsackie Virus B6**

Testing Facility  
**MicroBioTest**  
Division of Microbac Laboratories, Inc.  
105 Carpenter Drive  
Sterling, VA 20164

Prepared for  
**3M Health Care**  
**3M Infection Prevention Division**  
**3M Center, 270-4N-03**  
**St. Paul, MN 55144**

**October 27, 2015**

**Page 1 of 12**

MicroBioTest Protocol: 109.3.10.27.15

MicroBioTest Project: 109-228

*F.H.*

## OBJECTIVE:

This study is designed to measure virucidal effectiveness of a test product. It determines the potential of the test product to inactivate virus in suspension. The test follows the ASTM International test method designated E1052-11 "Standard Test Method to Assess the Activity of Microbicides against Viruses in Suspension".

## TESTING CONDITIONS:

The test products will be evaluated against the challenge virus in suspension. One test product, one lot, will be evaluated for inactivation of Coxsackie Virus B6 at two exposure (contact) time points. Two replicates (N=2) will be performed for each condition. To minimize buffer interference and to minimize reduction of virucidal activity, the volume of virus inoculum added to test material will be kept to equal or less than 10% of the total volume of the test. Aliquots will be removed at the completion of the contact time from the test product/virus reaction mixture; neutralized (quenched); and inoculated onto appropriate host cell system. The inoculated host system will be incubated and read for presence of infectious virus.

## MATERIALS:

- A. Test, control and reference substances will be supplied by the sponsor of the study (see last page).

The test product will be tested as supplied by the sponsor unless directed otherwise. All operations performed on the test product such as dilution or specialized storage conditions must be specified by the sponsor before initiation of testing.

The sponsor assures MicroBioTest testing facility management that the test product has been appropriately tested for identity, strength, purity, stability, and uniformity as applicable.

MicroBioTest will retain all unused test products for a period of one year upon completion of the test, and then discard them in a manner that meets the approval of the safety officer.

B. Materials supplied by MicroBioTest, including, but not limited to:

1. Challenge virus (requested by the Sponsor):
  - **Coxsackie Virus B6**, strain: Schmitt; ATCC VR-155
2. Host: LLC-MK2 cells
3. Laboratory equipment and supplies.
4. Media and reagents:

Media and reagents appropriate to the virus-host system will be used and documented in the data pack and project sheets.

**TEST SYSTEM IDENTIFICATION:**

All dilution tube racks, and host cell-containing apparatus will be labeled with virus identification and project number.

**EXPERIMENTAL DESIGN:**

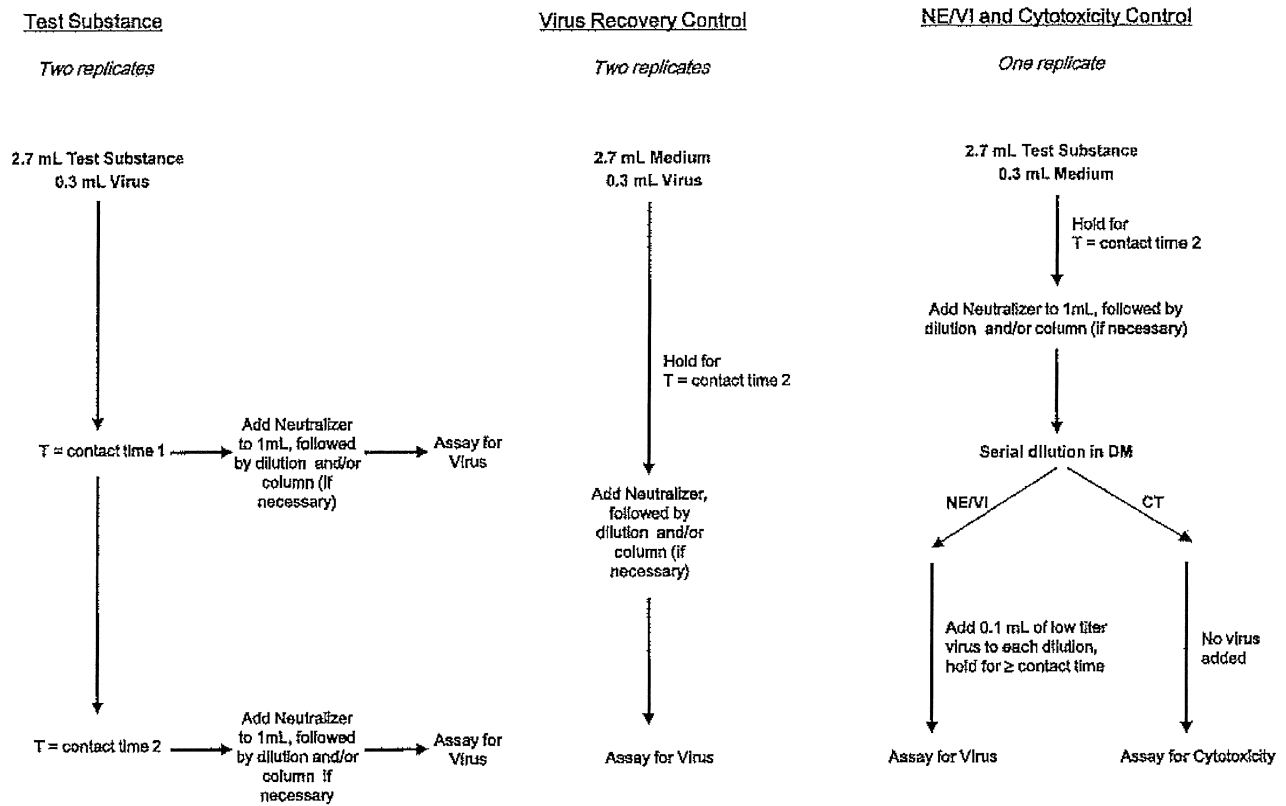
All of the procedures involved in performance of this study are described in a detailed series of SOPs that are maintained at MicroBioTest. SOPs and Logs are referred to in the raw data and are required as part of GLP regulations. The procedures used in different phases of the study will be documented in the data pack.

The study flow diagram is summarized in Figure 1, with details described below.

F.H.

FIGURE 1

Title: VIRUCIDAL EFFICACY SUSPENSION TEST – Coxsackie Virus B6



DM: Dilution Medium

NE/VI: Neutralizer Effectiveness/Viral Interference

CT: Cytotoxicity Control

Note: One test product, one lot, will be evaluated at one concentration and two contact times, in duplicate runs (N=2). The NE/VI and CT controls will be performed at N=1.

A. Inoculum preparation:

Viral stocks are purchased from reputable sources that identify them by scientifically accepted methods. Records are maintained that demonstrate the origin of the virus. The virus stocks are stored at an ultra-low temperature.

Frozen viral stocks will be thawed on the day of the test.

B. Test product preparation:

The test product is a liquid. It will be tested at one concentration (See Miscellaneous Information page for details).

The prepared liquid test product should be pre-equilibrated to the test temperature as applicable.

C. Test

One test product, one lot, will be evaluated at one concentration and two contact times, in duplicate runs (N=2).

For each run, a 2.7-mL aliquot of the test product will be spiked with 0.3 mL of the virus suspension and mixed thoroughly by vortexing. Upon completion of each contact time, a 1.0-mL aliquot of the reaction mixture will be pulled and immediately mixed with an equal volume of neutralizer. The neutralized sample may be further quenched by dilution with dilution medium to remove cytotoxicity. The quenched sample will be serially tenfold diluted with dilution medium (DM) and selected dilutions will be inoculated onto host cells to assay for infectious virus.

If Sephacryl columns are used to further remove cytotoxicity, each sample will be loaded onto separate pre-spun columns. The eluates will be collected and serially ten-fold diluted. If columns are not used, serial tenfold dilutions of the neutralized virus-test product mixture will be directly conducted using an appropriate diluent medium.

F.H.

D. Controls:

All controls will be performed at the same time as the test, incubated under the same conditions and assayed in the same manner as the test (see above).

1. Virus recovery control:

This control will be performed in duplicate runs (N=2). A 2.7-mL aliquot of the medium will be spiked with 0.3 mL of virus suspension and thoroughly mixed by vortexing. After the contact time (the longest contact time point will be used for this control as a worst case), an aliquot of the reaction mixture will be mixed with an equal volume of neutralizer. The neutralized sample will be serially tenfold diluted with dilution medium and selected dilutions will be inoculated onto host cells to assay for infectious virus. The virus control results from this control will be used as the input viral load and compared with the test product treatment results to evaluate viral reduction by the test product.

2. Neutralizer effectiveness/viral interference control:

This control will determine if residual active ingredient is present after neutralization and if the neutralized test product interferes with virus infectivity. This control will be performed in a singlet run using the longer contact time as the worst case scenario.

A 2.7-mL aliquot of the test product will be mixed thoroughly with 0.3 mL of medium by vortexing, held for contact time (the longer contact time), and then neutralized by adding equal volume of neutralizer. The neutralized mixture will then be serially tenfold diluted using dilution medium. Each dilution will be divided into two portions, one for Neutralizer effectiveness/viral interference control, and the other for cytotoxicity control

For the Neutralizer effectiveness/viral interference control, 100  $\mu$ L of a low titered ( $10^{-2}$  to  $10^{-3}$ ) virus stock will be added to 4.5 mL of selected dilutions of the solution and held for a period equivalent or greater than the longer contact time. The virus-spiked solution will be used to inoculate host cells as described for the test procedure.

N.H.



3. Cytotoxicity control:

This control will be performed in a singlet run using the longer contact time as the worst case scenario.

Selected dilutions of the sample obtained from the Neutralizer effectiveness/viral interference control run will be inoculated onto host cells and incubated together with other test and control samples as described for the test procedure. The condition of the host cells will be recorded at the end of the incubation period. The cytotoxic effects should be distinct from virus-specific cytopathic effects, which will be evident in the stock titer and virus recovery control cultures.

4. Column titer control (to be performed only if a Sephacryl column is used):

This control will be performed to determine any affect the columns may have on infectious virus titer.

The sample for this control will be acquired from a portion of the PRC, prior to passing through the columns and will be serially diluted in CCM, then processed in the same manner as the test.

5. Cell viability control:

At least four wells will be inoculated with an appropriate media during the incubation phase of the study. This control will demonstrate that cells remain viable throughout the course of the assay period. In addition, it will confirm the sterility of the media employed throughout the assay period.

6. Virus Stock Titer control (VST)

An aliquot of the virus used in the study will be directly serially diluted and inoculated onto the host cells to confirm the titer of the stock virus. This control will demonstrate that the titer of the stock virus is appropriate for use and that the viral infectivity assay is performed appropriately.

T.H.

E. Infectivity assay:

The residual infectious virus in the test and controls will be detected by viral-induced cytopathic effect (CPE).

Selected dilutions of the neutralized inoculum/test product mixture will be added to cultured cell monolayers at a minimum of four wells per dilution per sample. The inoculated plates will be incubated at  $36\pm 2^{\circ}\text{C}$  in  $5\pm 1\%$   $\text{CO}_2$  for 3-5 days. The host cells may be washed twice with phosphate buffered saline (PBS) prior to inoculation. The host cell cultures will be observed and refed, as necessary, during the incubation period. These activities, if applicable, will be recorded. Then the host cells will be examined for presence of infectious virus. The resulting virus-specific CPE and test product-specific cytotoxic effects will be scored by examining both test and controls. These observations will be recorded.

F. Calculation:

The 50% tissue culture infective dose per mL ( $\text{TCID}_{50}/\text{mL}$ ) will be determined using the method of Spearman-Kärber (Kärber G. Arch. Exp. Pathol. Pharmacol. 1931, 162:480-483) or other appropriate methods such as Reed and Muench (Am. J. of Hyg. 1938, 27:493). In the case where a sample contains no detectable virus, a statistical analysis may be performed based on Poisson distribution (International Conference On Harmonization, 1999, Topic Q5A:24-25) to determine the theoretical maximum possible titer for that sample. These analyses will be described in detail in the final report.

The test results will be reported as the reduction of the virus load by the test product expressed as  **$\log_{10}$  and percent** reduction.

The Virus Load will be calculated in the following manner:

Virus Load ( $\log_{10} \text{TCID}_{50}$ ) = Virus Titer ( $\log_{10} \text{TCID}_{50}/\text{mL}$ ) +  $\log_{10}$  [Volume (mL)]

The  $\log_{10}$  Reduction Factor (LRF) will be calculated in the following manner:

$\log_{10}$  Reduction Factor = Virus Recovery Control ( $\log_{10} \text{TCID}_{50}$ ) – Test ( $\log_{10} \text{TCID}_{50}$ )

The percentage of virus inactivation was calculated in the following manner:

$[1 - \text{Output Viral Load}/\text{Initial Viral Load}] \times 100 = [1 - 10^{(-\log_{10} \text{Reduction Factor})}] \times 100$

F.H.

#### **TEST ACCEPTANCE CRITERIA:**

The test will be acceptable for evaluation of the test results if the criteria listed below are satisfied. The study director may consider other causes that may affect test reliability and acceptance.

- Virus must be recovered from the neutralizer effectiveness/viral interference control (not exhibiting cytotoxicity).
- Viral-induced CPE must be distinguishable from test product induced toxicity.
- The cell viability control must remain viable throughout the course of the assay period and exhibit absence of virus.

#### **PERSONNEL AND TESTING FACILITIES:**

A study director will be assigned prior to initiation of the test. Resumes are maintained and are available on request. This study will be conducted at MicroBioTest, 105 Carpenter Drive, Sterling, Virginia 20164.

#### **REPORT FORMAT:**

MicroBioTest employs a standard report format for each test design. Each final report will provide the following information:

- Sponsor identification
- Test product identification
- Type of assay and project number
- Interpretation of results and conclusions
- Test results presented in tabular form
- Methods and evaluation criteria, if applicable
- Dates of study initiation and completion (GLP studies only)
- Signed Quality Assurance and Compliance Statements (GLP studies only)

F.H.

## RECORDS TO BE MAINTAINED:

All raw data, protocol, protocol modifications, test product records, final report, and correspondence between MicroBioTest and the sponsor will be stored in the archives at MicroBioTest, 105 Carpenter Drive, Sterling, Virginia 20164 or in a controlled facility off site.

All changes or revisions to this approved protocol will be documented, signed by the study director, dated and maintained with this protocol. The sponsor will be notified of any change, resolution, and impact on the study as soon as practical.

The proposed experimental start and termination dates; additional information about the test product, challenge virus, and host cell line monolayers used and the type of neutralizers employed in the test will be addressed in a project sheet issued separately for each study. The date the study director signs the protocol will be the study initiation date. All project sheets issued will be forwarded to the study sponsor for appropriate action.

L.H.

**MISCELLANEOUS INFORMATION:**

The following information is to be completed by Sponsor prior to initiation of the study  
(please check off all applicable boxes):

- A. Name and address: 3M Health Care  
3M Infection Prevention Division  
3M Center, 270-4N-03  
St. Paul, MN 55144
- B. Test product name: 3M Avagard 9250 Antiseptic Hand Rub  
Active ingredient: chlorhexidine gluconate 0.45-1.55% w/w Ethanol 63.1-77.0% v/v  
Lot No.: 5611001
- Exposure time #1: ☒ 1 minute or ☐ \_\_\_\_\_  
Exposure time #2: ☒ 5 minutes or ☐ \_\_\_\_\_
- Exposure temperature: ☒ Room temperature (20±2C)
- Dilution to be tested: ☒ Ready to use or ☐ \_\_\_\_\_
- Diluent: ☒ Not applicable (Product is ready-to-use)  
☐ Sterile deionized water  
☐ Other \_\_\_\_\_
- C. Precautions/storage conditions: refer to MSDS or certificate of analysis  
☒ provided ☐ not provided
- D. Organic Load in inoculum: ☒ Not applicable or ☐ \_\_\_\_\_

Continued on next page

F. H.

**MISCELLANEOUS INFORMATION (Continued):**

**REPORT HANDLING:**

The sponsor intends to submit the final report to: Customer

**STUDY CONDUCT:** ☒ GLP

**PROTOCOL APPROVAL BY SPONSOR**

Sponsor Signature: Fiona Hao Date: 2015.11.16

Printed Name: Fiona Hao

**STUDY DIRECTOR APPROVAL (MicroBioTest):**

Study Director Signature: Salimatu Lukula Date: Oct 2/2/16

Printed Name: Salimatu Lukula

DEESU  
2/2/16

Date Issued: 02/02/16 Project Sheet No. 1 Page No. 1 Laboratory Project Identification No. 109-228

**STUDY TITLE:** VIRUCIDAL EFFICACY  
SUSPENSION TEST- Cocksackie Virus B6

**STUDY DIRECTOR:** Salimatu Lukula, M.S.

*Salimatu Lukula* 2/2/16  
Signature Date

**TEST MATERIAL(S):**  
3M Avagard™9250 antiseptic Handrub

**LOT NO.**  
510110D1

**DATE RECEIVED:**  
12/08/15

**DS NO.**  
F1071

**PERFORMING DEPARTMENT(S):**  
Virology and Molecular Biology Department

**STORAGE CONDITIONS:** Location: H4  
☐ Dark ☒ Ambient Room Temperature  
☐ Desiccator ☐ Freezer ☐ Refrigerator ☐ Other:

**PROTECTIVE PRECAUTION REQUIRED:** MSDS ☒ Yes / ☐ No

**PHYSICAL DESCRIPTION:** ☐ Solid ☒ Liquid ☐ Aerosol ☐ Other:

**PURPOSE:** See attached protocol. **AUTHORIZATION:** See client signature.

**PROPOSED EXPERIMENTAL START DATE:** 02/02/16 **TERMINATION DATE:** 02/08/16

**CONDUCT OF STUDY:** ☐ FDA ☐ EPA ☐ R&D ☒ GLP ☐ GCP ☒ Other: Customer

**SPONSOR:** 3M Health Care  
3M Infection Prevention Division  
3M Center, 270-4N-03  
St. Paul, MN 55144

**CONTACT PERSON:** Fiona Hao  
Email: [fhao@mmm.com](mailto:fhao@mmm.com)

**TEST CONDITIONS:**

**Challenge organism:** Cocksackie Virus B6, strain: Cocksackievirus B6 antiserum [V-033-501-563], ATCC VR-1037

**Host cell line:** LLC-MK2, ATCC CCL-7.1

**Active Ingredient(s):** Chlorhexidine Gluconate 0.45-0.55% (w/v) Ethanol 63.1-77% (w/v)

**Dilution medium:** Minimum Essential Medium (MEM) + 2% Fetal Bovine Serum (FBS)

**Neutralizer:** MEM + 10% FBS + 1% Polysorbate 80 + 0.5% Lecithin + 1% Tamol

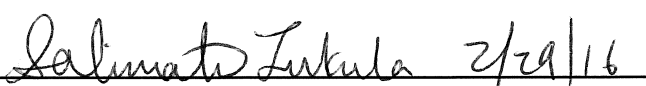
**Dilution:** Ready to use **Organic load:** Not Applicable

**Contact times:** 1 minute and 5 minutes **Contact temperature:** Room temperature (20±2C)

**Incubation time:** 3 - 5 days **Incubation temperature:** 36±2°C with 5±1% CO<sub>2</sub>

**PROTOCOL AMENDMENT:**

- Page 7, section 4 of the protocol states "...a portion of the PRC...serially diluted in CCM". This should read as "a portion of the VRC...serially diluted in DM". This amendment serves to correct this typographical error in the protocol.
- Protocol page 3 states the strain and source of the challenge virus as "strain: Schmitt; ATCC VR-155". The strain and source of the challenge virus used in testing is "strain: Cocksackievirus B6 antiserum [V-033-501-563], ATCC VR-1037". This amendment serves to correct the typographical error in the protocol.

Date Issued: 02/26/16    Project Sheet No. 2    Page No. 1    Laboratory Project Identification No. 109-228			
<b>STUDY TITLE:</b> VIRUCIDAL EFFICACY SUSPENSION TEST – Cocksackie Virus B6		<b>STUDY DIRECTOR:</b> Salimatu Lukula M.S.  Signature _____ Date 2/29/16	
<b>TEST MATERIAL(S):</b> 3M Avagard™ 9250 Antiseptic Handrub		<b>LOT NO.</b> 510110D1	<b>DATE RECEIVED:</b> 12/8/15
<b>PERFORMING DEPARTMENT(S):</b> Virology and Molecular Biology		<b>STORAGE CONDITIONS:</b> Location: H4 <input checked="" type="checkbox"/> Dark <input checked="" type="checkbox"/> Ambient Room Temperature <input type="checkbox"/> Desiccator <input type="checkbox"/> Freezer <input type="checkbox"/> Refrigerator <input type="checkbox"/> Other:	
<b>CONDUCT OF STUDY:</b> <input type="checkbox"/> FDA <input type="checkbox"/> EPA <input type="checkbox"/> R&D <input checked="" type="checkbox"/> GLP <input type="checkbox"/> GCP <input type="checkbox"/> Other:			
<b>SPONSOR:</b> 3M Health Care 3M Infection Prevention Division 222 Tian Lin Road, Shanghai 200233   China		<b>CONTACT PERSON:</b> Fiona Hao E-mail: fhao@mmm.com	
<b>PROTOCOL AMENDMENTS:</b> <ol style="list-style-type: none"> <li>Project sheet No. 1 lists the active ingredient name as "Chlorhexidine Gluconate 0.45-0.55% (w/v) Ethanol 63.1 1-77% (w/v)". The correct listing is "Chlorhexidine Gluconate 0.45-0.55% (w/v) Ethanol 63.1 1-77% (v/v)". This amendment serves to clarify the typographical error on project sheet No.1.</li> <li>Project Sheet No.1 and Protocol states the Sponsor address as 3M Center, 270-4N-03, St. Paul, MN 55144. The correct Sponsor address is 222 Tian Lin Road, Shanghai 200233, China. This amendment serves to correct the sponsor address in Project sheet No.1 and Protocol.</li> </ol>			