Volume _____

FINAL REPORT

CONFIRMATORY VIRUCIDAL EFFICACY TEST – Feline calicivirus (Surrogate for Human Norovirus)

Test Agent
EcaFlo® Anolyte

Lot Number
5/30/2012

Test Organism
Feline calicivirus, ATCC VR-782

Test Guideline
EPA Guidelines 810.2200 (f)(2)

Author
S. Steve Zhou Ph.D.

Study Completion Date
06/28/12

Performing Laboratory
MICROBIOTEST
A Division of Microbac Laboratories, Inc.
105 Carpenter Drive
Sterling, Virginia 20164

Laboratory Project Identification Number
657-110

Protocol Identification Number
657.6.04.10.12

Sponsor
Integrated Environmental Technologies
4235 Commerce St.
Little River, SC 29566

STATEMENT OF NO DATA CONFIDENTIALITY

Title: CONFIRMATORY VIRUCIDAL EFFICACY TEST – Feline calicivirus (Surrogate for Human Norovirus)

Performed by: MICROBIOTEST
A Division of Microbac Laboratories, Inc.
105 Carpenter Drive
Sterling, Virginia 20164

No claim of confidentiality is made for any information contained in this study on the basis of its falling within the scope of FIFRA sec. 10(d)(1)(A), (B) or (C).

Submitter signature: _____ Date: _____

Typed Name of Signer: _____

Typed Name of Company: Integrated Environmental Technologies

COMPLIANCE STATEMENT

This study meets the requirements for 40 CFR § 160 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.

Study Director signature: _____ Date: 06/28/2012

Typed Name: S. Steve Zhou Ph.D.

Typed Name of Laboratory: MicroBioTest, a division of Microbac Laboratories, Inc.

Sponsor signature: _____ Date: _____

Typed Name of Signer: _____

Typed Name of Company: Integrated Environmental Technologies

Submitter signature: _____ Date: _____

Typed Name of Signer: _____

Typed Name of Company: Integrated Environmental Technologies

QUALITY ASSURANCE UNIT STATEMENT

Title of Study: CONFIRMATORY VIRUCIDAL EFFICACY TEST – Feline calicivirus (Surrogate for Human Norovirus)

The Quality Assurance Unit of MICROBIOTEST has inspected Project Number 657-110 in compliance with current Good Laboratory Practice regulations, (40 CFR § 160).

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

PHASE INSPECTED	DATE OF INSPECTION	DATE REPORTED TO STUDY DIRECTOR	DATE REPORTED TO MANAGEMENT
Protocol	06/04/12	06/05/12	06/05/12
In Process (Control)	06/05/12	06/05/12	06/05/12
Final Report	06/28/12	06/28/12	06/28/12

Jeanne M. Anderegg
Quality Assurance Associate

6-28-12
Date

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TEST SUMMARY

TITLE: CONFIRMATORY VIRUCIDAL EFFICACY TEST – Feline calicivirus (Surrogate for Human Norovirus)

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

TEST MATERIALS SUPPLIED BY THE SPONSOR OF THE STUDY:

1. EcaFlo® Anolyte; Lot No. 5/30/2012; received at MICROBIOTEST on 05/31/12; and assigned DS No. C370

SPONSOR: Integrated Environmental Technologies
4235 Commerce St
Little River, SC 29566

TEST CONDITIONS

Challenge virus:

Feline calicivirus; ATCC VR-782

Active ingredient in test product:

Hypochlorous Acid (HOCl)

Neutralizer:

Newborn Calf Serum (NCS) + 1% Polysorbate 80 + 0.5% $Na_2S_2O_3$

Dilution medium:

RPMI 1640 + 2% NCS

Contact time:

10 minutes

Contact temperature:

Ambient Room Temperature (21C)

Carriers:

Glass petri dishes

Carrier inoculation/dry time:

2 x 2 inch area of glass carrier inoculated with 0.4 mL of virus and dried for 33 minutes at 21C

Organic load:

≥ 5% serum

Dilution:

205 ppm HOCl (final)

TEST CONDITIONS (continued)

Diluent:

Sterile Deionized Water

Media and reagents:

Newborn Calf Serum (NCS) + 1% Polysorbate 80 + 0.5% $Na_2S_2O_3$
RPMI 1640 + 2% NCS
Sterile Deionized Water
NCS

STUDY DATES AND FACILITIES

The laboratory phase of this test was performed at MICROBIOTEST, 105 Carpenter Drive, Sterling, VA 20164, from 06/05/12 to 06/13/12. The study director signed the protocol 06/04/12. The day of test conduct on 06/05/12, the testing started at 9:30 am and ended at 1:00 pm. The study completion date is the date the study director signed the final report.

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

RECORDS TO BE MAINTAINED

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

CALCULATION OF TITER

The 50% tissue culture infectious dose per mL ($TCID_{50}$/mL) was determined using the Spearman-Karber method using the following formula:

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

where:

m = the logarithm of the titer relative to the test volume
x_k = the logarithm of the smallest dosage which induces infection in all cultures
d = the logarithm of the dilution factor
p_i = the proportion of positive results at dilution i
The values were converted to $TCID_{50}$/mL using a sample inoculum of 0.05 mL.

RESULTS

Data are presented in Tables 1-4.

The Log_{10} Reduction Factor (LRF) was calculated in the following manner:

Log_{10} Reduction = Log_{10} $TCID_{50}$ (Plate Recovery Control) – Log_{10} $TCID_{50}$ (Test)

The Load (Log_{10} $TCID_{50}$) per carrier was calculated in the following manner:

Load (Log_{10} $TCID_{50}$) = Titer (Log_{10} $TCID_{50}$/mL) + Log_{10} [volume per carrier (mL)]

Key (for all tables):

C/y = Cytotoxicity observed in y wells inoculated; no viral cytopathic effect (CPE) could be determined.
X/y = X wells out of y wells inoculated exhibited viral CPE
0/y = 0 wells out of y wells inoculated exhibited viral CPE, no cytotoxicity or bacterial contamination was observed in any of the wells inoculated

Table 1
Test Agent Results

Dilution*	EcaFlo® Anolyte Lot No. 5/30/2012	
	Replicate 1	Replicate 2
10^{-2}	8/8	8/8
10^{-3}	8/8	8/8
10^{-4}	8/8	8/8
10^{-5}	7/8	7/8
10^{-6}	1/8	3/8
10^{-7}	1/8	0/8
Titer (Log_{10} $TCID_{50}$/mL)	**6.93**	**7.05**
Load (Log_{10} $TCID_{50}$) per carrier (0.4 mL challenge)	**6.53**	**6.65**
Log_{10} Reduction	**1.01**	**0.89**

*Dilution refers to the fold of dilution from virus inoculum.

RESULTS (continued)

Table 2
Neutralizer Effectiveness and Cytotoxicity Related Controls

Dilution*	EcaFlo® Anolyte Lot No. 5/30/2012	
	Neutralizer Effectiveness Control	**Cytotoxicity Control**
10^{-2}	8/8	0/8
10^{-3}	8/8	0/8
10^{-4}	8/8	0/8

*Dilution refers to the fold of dilution from mock inoculum.

Table 3
Virus Recovery Controls

Dilution*	**Plate Recovery Control**		**Virus Stock Titer Control**
	Replicate 1	**Replicate 2**	
10^{-3}	8/8	8/8	Not determined
10^{-4}	8/8	8/8	8/8
10^{-5}	8/8	8/8	8/8
10^{-6}	8/8	7/8	8/8
10^{-7}	2/8	1/8	1/8
10^{-8}	0/8	0/8	0/8
10^{-9}	Not determined	Not determined	0/8
Titer (Log_{10} $TCID_{50}$/mL)	**8.05**	**7.80**	**7.93**
Load (Log_{10} $TCID_{50}$) per carrier (0.4 mL challenge)	**7.65**	**7.40**	**NA**
Average Load for Plate Recovery Controls	**7.54**		**NA**

*Dilution refers to the fold of dilution from virus inoculum.
NA = Not Applicable

Table 4
Viability Control Results

Cell Viability Control
0/8
Cells were viable; media was sterile

CONCLUSIONS

According to the regulatory agencies, the test agent passes the Virucidal Efficacy Test if there is complete inactivation of the challenge virus at all dilutions. When cytotoxicity is evident, at least a three-log reduction in titer must be demonstrated beyond the cytotoxic level.

When tested as described, EcaFlo® Anolyte did not pass the Confirmatory Virucidal Efficacy Test when Feline calicivirus (Surrogate for Human Norovirus), with ≥5% organic load added, was exposed to the test agent for 10 minutes at 21C. All of the controls met the criteria for a valid test. These conclusions are based on observed data.

APPENDIX

MICROBIOTEST
A Division of Microbac Laboratories, Inc.
105-B Carpenter Drive
Sterling, VA 20164

MICROBIOTEST PROTOCOL

CONFIRMATORY VIRUCIDAL EFFICACY TEST -

Feline calicivirus
(Surrogate for Human Norovirus)

Testing Facility
MICROBIOTEST
A Division of Microbac Laboratories, Inc.
105 Carpenter Drive
Sterling, VA 20164

Prepared for
Integrated Environmental Technologies
4235 Commerce Street
Little River, SC 29566

April 10, 2012

OBJECTIVE:

This test is designed to substantiate virucidal effectiveness claims for a product to be labeled as a virucide. It determines the potential of the test agent to disinfect hard surfaces contaminated with viruses. The test is designed to simulate consumer use and conforms to EPA Guidelines 810.2200 (2012) and DIS/TSS-7 (1981), and follows the procedure outlined in the American Society for Test Materials (ASTM) test method designated ASTM E1053.

TESTING CONDITIONS:

Virus will be dried on a sterile glass Petri dish at room temperature. One lot of the test agent will be used to treat the dried virus according to the label claims. Two replicates will be performed for each lot of the product at one exposure (contact) time. After a defined exposure period, as specified by the sponsor in the miscellaneous section of the protocol, the neutralized test agent-virus mixture will be scraped from the surface, serially diluted and assayed for the 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 agent will be tested as supplied by the sponsor unless directed otherwise. All operations performed on the test agent 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 agent has been appropriately tested for identity, strength, purity, stability, and uniformity as applicable.

MICROBIOTEST will retain all unused test agents for a period of at least three months after 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 of the study): Feline calicivirus (Surrogate for Human Norovirus)

2. Host cell line: CrFK 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 Petri dishes, dilution tube racks, and host-containing apparatus will be labeled with virus identification and project number.

EXPERIMENTAL DESIGN:

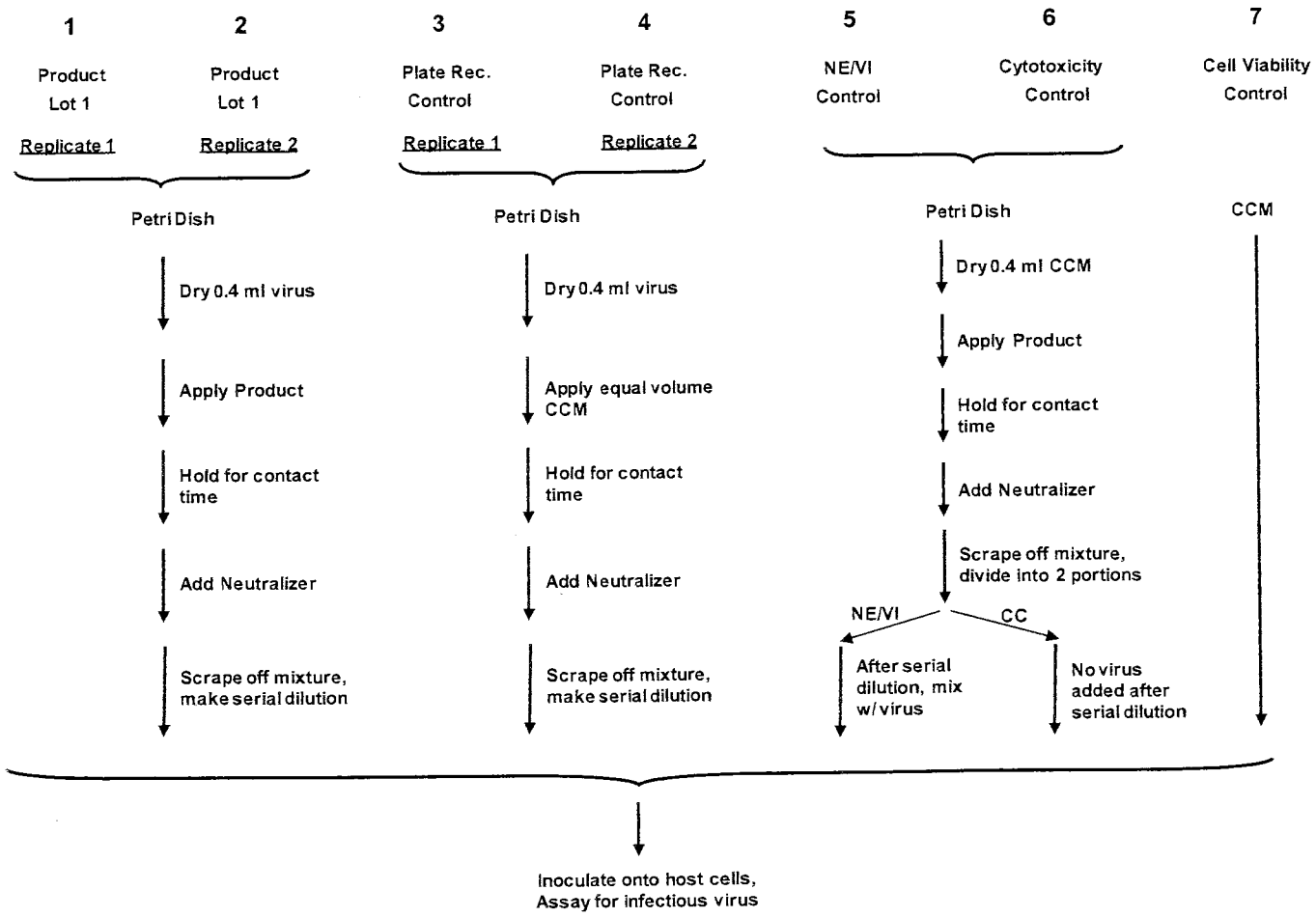
All of the procedures involved in performance of this study are described in a detailed series of SOPs that are maintained at MICROBIOTEST. SOPs and Logs are referred to in the raw data and are required as part of GLP regulations. The procedures used in different phases of the study will be documented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

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

JAE

FIGURE 1

Title: CONFIRMATORY VIRUCIDAL EFFICACY TEST- Feline calicivirus



CCM: Cell Culture Medium
NE/VI: Neutralizer Effectiveness/Viral Interference
CC: Cytotoxicity Control

Note: the volume of inoculum applied onto each carrier may be changed depending on the titer of the virus. This volume will remain consistent among all test and control samples.

ASC

A. Inoculum preparation:

Viral stocks are purchased from reputable sources that identify them by scientifically accepted methods and are propagated at MICROBIOTEST. 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 (fresh stock cultures may be used at the discretion of the Study Director). The organic soil concentration will be adjusted to at least 5% for the virus unless otherwise directed by the Sponsor.

B. Carrier preparation:

One lot of the test agent will be tested. Two replicates will be performed for each lot of the product.

For each lot of the sample, an aliquot of 0.4 mL of stock virus will be spread, with the cell scraper, over an area of approximately 4 in^2 that has been marked on the underside of pre-sterilized Petri dishes. Note: the volume of inoculum applied onto each carrier may be changed depending on the titer of the virus. This volume will remain consistent among all test and control samples. Then the virus will be allowed to dry at ambient temperature. The drying time and temperature will be recorded.

Two carriers will be prepared for the test product. Two carriers will be prepared for the plate recovery control. Additionally, one carrier will be prepared for the neutralizer effectiveness control using cell culture medium (CCM) challenge in lieu of the virus.

C. Test agent preparation:

The agent will be prepared according to the sponsor's directions or proposed label claims. The test agent will be allowed to come to testing temperature for at least ten minutes before testing.

D. Test:

One lot of test product will be evaluated at two replicates at one contact time.

For each run, after the inoculum has dried, 2.0 mL of the test agent will be applied (for spray type agents, see below). The plates will remain at the temperature and for the time specified by the sponsor. After the contact period, the test agent will be neutralized with an equal volume of appropriate neutralizer and the mixture will be scraped from the surface of the dish with a cell scraper. This will be considered approximately one $\log_{10}$ dilution.

For spray type agents, the test agent will be sprayed at the time and distance as directed by the sponsor or the label instructions after the inoculum has dried. The dried virus film should be completely covered. The plates will remain at the temperature and for the exposure time as specified by the sponsor. During the contact period, the volume dispensed will be measured. Upon completion of the contact time, an equal volume of neutralizer will be used. The mixture will be scraped from the surface of the dish with a cell scraper. This will be considered approximately a one $\log_{10}$ dilution.

If gel filtration columns (e.g. Sephacryl columns) are utilized, 0.8 mL of each sample will be loaded into individual pre-spun Sephacryl columns. The columns will be spun for 3 minutes at 1000 rpm. Following passage through the columns, the eluates will be aseptically collected and tenfold serially diluted in CCM. If columns are not used, serial tenfold dilutions of neutralized inoculum/test agent mixture(s) virus will be prepared in CCM.

E. Infectivity assay:

The residual infectious virus in both test and controls will be detected by viral-induced cytopathic effect (CPE). Selected dilutions of the neutralized inoculum/test agent mixture will be added to cultured cell monolayers. The host cell culture may be washed twice with phosphate buffered saline (PBS) prior to inoculation. At least four wells per dilution will be added to the host cell monolayers and incubated at 36±2C in 5±1% CO_2 for 7-9 days. Post incubation the infectious virus will be scored microscopically by observing CPE produced by replicating infectious virus.

AQC

F. 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. Cell viability control:

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

2. Plate recovery control (PRC):

Two replicates will be performed for his control.

An equal amount of CCM as is used for the test agent analysis will be added to the dried virus. Post-contact time, the virus/CCM mixture will be subjected to the identical neutralization procedure as the test agent. If columns are used, a portion of the virus/CCM/neutralizer mixture will be used for the column titer control (see section F3).

The results from the PRC will be compared with the test results to confirm recovery of at least four $\log_{10}$ of infectious virus following drying and neutralization. Its titer will be used to compare with the titers of the test results to reach the acceptable test criteria (see below).

3. Column titer control:

This control will be performed only if Sephacryl columns are used. It is performed to determine any effects of Sephacryl columns on infectious virus titer while passing through the columns.

The sample for this control will be acquired from a portion of the Plate recovery control prior to passing through the columns. This sample is used to make direct ten-fold serial dilutions in CCM. Then it will be processed in the same manner as the rest of the test and controls.

4. Neutralizer effectiveness/viral interference control:

This control will determine if residual active ingredient is present after neutralization and if the neutralized test agent interferes with virus infectivity.

This control will be processed exactly as the test procedure but instead of viral inoculum, dried CCM will be exposed to the test agent and assayed as previously described. After treatment and neutralization, this control will be divided into two portions, one for cytotoxicity control, the other for the neutralizer effectiveness/viral interference control, and processed as the test.

If gel filtration columns (e.g. Sephacryl columns) are used, each portion will be passed through individual columns and the eluate will be serially diluted ten-fold in CCM. If columns are not used, the neutralizer effectiveness sample will be diluted using serial ten-fold dilutions in CCM.

Following serial dilution of the reaction mixture in CCM, 100 µL of a low titered (10^{-2} - 10^{-3}) virus stock will be added to 4.5 mL of each dilution and held for a period greater than or equal to the contact time. Then these selected dilutions will be used to inoculate host cells as described for the test procedure.

5. Cytotoxicity Control:

The cytotoxicity samples, acquired from the neutralizer effectiveness control, will be diluted and have no virus added. Selected dilutions will be inoculated and incubated in the same manner as the rest of the test and control samples. These effects are distinct from virus-specific cytopathic effects, which will be evident in the stock titer and plate recovery control cultures.

ACE

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.

G. Calculation:

The 50% tissue culture infectious dose per mL ($TCID_{50}$/mL) will be determined using the method of Spearman-Karber or other appropriate methods. The test results will be reported as the reduction of the virus titer due to treatment with test agent expressed as log_{10}.

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.

- The infectious virus recovered from the PRC control must be $\geq$ 4-log_{10}.
- Viral-induced cytopathic effect (if any) must be distinguishable from test agent induced cytotoxic effects.
- Virus must be recovered from the neutralizer effectiveness/viral interference controls (not exhibiting cytotoxicity).
- Virus must not be detected in the cell viability control.

PRODUCT EVALUATION CRITERIA:

According to the regulatory agencies, the test agent passes the test if there is complete inactivation of the virus at all dilutions. When cytotoxicity is evident, at least a three-log reduction in titer must be demonstrated beyond the cytotoxic level.

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 agent 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)

RECORDS TO BE MAINTAINED:

All raw data, protocol, protocol modifications, test agent 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 agent; 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.

AOC

MISCELLANEOUS INFORMATION:

The following information is to be completed by sponsor before initiation of study:

A. Name and address: Integrated Environmental Technologies
4235 Commerce Street
Little River, SC 29566

B. Test agent: Ecaflo® Anolyte

Active ingredient(s): hypochlorous acid

Lot No.: 5/30/2012

Contact time: 10 (must be ≤10 minutes)

Exposure temperature: Ambient room temperature 20±1C

Dilution to be tested:
☐ Ready to use
☒ 205 (____ part test agent + ____ parts diluent)
test pH, titrate & dilute w/ DI water to 205 ppm HOCl

Diluent:
☐ Not applicable
☐ _____ ppm ± 2.9% AOAC hard water
☒ Other: DI water

Spray application: ☒ Until thoroughly wet or ☐ _____

Spraying distance: ☒ 6"-8" or ☐ _____

C. Organic load: ☒ ≥ 5% serum ☐ no

D. Precautions/storage – MSDS or certificate of analysis provided: ☒ yes ☐ no

REPORT HANDLING:

The sponsor intends to submit this information to:
☒ US EPA ☐ Health Canada ☒ CAL DPR ☐ ARTG ☐ other: Internal Purposes

MISCELLANEOUS INFORMATION: (continued)

STUDY CONDUCT: ☒ GLP ☐ non-GLP

PROTOCOL APPROVAL BY SPONSOR:

Sponsor Signature: Stuart O Emmons Date: 5/30/2012

Printed Name: Stuart A Emmons

PROTOCOL APPROVAL BY STUDY DIRECTOR (MICROBIOTEST):

Study Director Signature: _____ Date: 06/04/2012

Printed Name: Steve Short

Date Issued: 06/04/12 Project Sheet No. 1 Page No. 1 Laboratory Project Identification No. 657-110

STUDY TITLE: CONFIRMATORY VIRUCIDAL EFFICACY TEST – Feline calicivirus (Surrogate for Human Norovirus)

STUDY DIRECTOR: S. Steve Zhou, Ph.D.

Signature 06/04/2012 Date

TEST MATERIAL(S):	**LOT NO.:**	**DATE RECEIVED:**	**DS NO.:**
EcaFlo® Anolyte	05/30/12	05/31/12	C370

PERFORMING DEPARTMENT(S): Virology and Molecular biology

STORAGE CONDITIONS: Location: C4
■ 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: 06/05/12 **TERMINATION DATE:** 06/18/12

CONDUCT OF STUDY: ☐ FDA ■ EPA ☐ R&D ■ GLP ☐ GCP ■ Other: CAL DPR

SPONSOR:
Integrated Environment Technologies
4235 Commerce Street
Little River, SC 29556

CONTACT PERSON: Stuart A Emmons, PE
Telephone No. 1.843.390.2500, Ext. 208
E-mail: stuart.emmons@ietltd.net

TEST CONDITIONS:

Challenge organism: Feline calicivirus, ATCC VR-782

Host cell line: CrFK, ATCC CCL-94

Active ingredient(s): Hypochlorous acid (HOCl)

Neutralizer(s): Newborn Calf Serum (NCS) + 1% Polysorbate 80 + 0.5% $Na_2S_2O_3$

Organic load: ≥ 5% Serum

Cell culture medium: RPMI 1640 + 10% NCS Dilution medium: RPMI 1640 + 2% NCS

Dilution to be tested: 205 ppm HOCl (final)

Diluent: Sterile Deionized (DI) Water

Contact time(s): 10 minutes Contact temperature(s): Ambient Room (20±1C)

Incubation time(s): 7-9 days Incubation temperatures(s): 36±2C

Spraying application: Until thoroughly wet Spraying distance: 6 – 8 inches

Comment(s): The original test agent will be titrated for total free available chlorine (FAC), pH measured and diluted with sterile deionized water to the target final HOCl. The test agent will be allowed to come to the testing temperature for at least ten minutes before testing.