UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON, DC 20460

OFFICE OF
PREVENTION,
PESTICIDES
AND TOXIC
SUBSTANCES

January 20, 2010

MEMORANDUM

Subject: Efficacy Review for WC Complete;
EPA File Symbol 64240-AL;
DP Barcode: D371362

From: Ayaad Assaad, D.V.M., Ph.D.
Efficacy Evaluation Team
Product Science Branch
Antimicrobials Division (7510P)
1/20/2010

Thru: Stacy Grigsby, PM 34
Regulatory Management Branch II
Antimicrobials Division (7510P)

To: Tajah Blackburn, Ph.D.
Team Leader, Product Science Branch
Antimicrobials Division (7510P)
1/26/10

Applicant: Combat Insect Control Systems
19001 N. Scottsdale Road
Scottsdale, AZ 85255

Formulation from the Label:

Active Ingredient(s)	% by wt.
Lactic acid, 80% solution	4.0%
Inert Ingredients	96.0 %
Total	100.0 %

I BACKGROUND

The product, WC Complete (EPA File Symbol 64240-AL), is a new product. The applicant requested to register the product for use as a disinfectant (bactericide, virucide), sanitizer, mildewstat, and deodorizer on hard, non-porous surfaces in household, commercial, institutional, industrial, food service, animal care, and hospital or medical environments. The label states that the product is a "one-step" disinfectant. Studies were conducted at ATS Labs, located at 1285 Corporate Center Drive, Suite 110, in Eagan, MN 55121.

This data package contained a letter from the applicant's representative to EPA (dated October 14, 2009), EPA Form 8570-1 (Application for Pesticide), EPA Form 8570-4 (Confidential Statement of Formula), EPA Form 8570-27 (Formulator's Exemption Statement), EPA Form 8570-34 (Certification with Respect to Citation of Data), EPA Form 8570-35 (End-Use Data Matrix), EPA Form 8570-36 (Summary of the Physical/ Chemical Properties (PR Notice 98-1)), nine studies (MRID 478898-09 through 478898-17), Statements of No Data Confidentiality Claims for all nine studies, and the proposed label.

Note: EPA Form 8570-4 (Confidential Statement of Formula) contains Confidential Business Information. Data or information claimed by the applicant to be FIFRA confidential has not been included in this report.

II USE DIRECTIONS

The product is designed for disinfecting and sanitizing hard, non-porous surfaces, including: appliances, bathtubs, countertops, doorknobs, faucets, fixtures, floors, shower doors, shower stalls, sinks, toilet bowls, and toilets. The proposed label indicates that the product may be used on hard, non-porous surfaces including: chrome, cultured marble, glass, glazed ceramic, glazed porcelain, glazed tile, plastic laminate, sealed fiberglass, sealed granite, stainless steel, and synthetic marble. Directions on the proposed label provide the following information regarding use of the product:

> As a disinfectant: Apply product and leave on surface for 10 minutes before wiping clean. Heavily soiled surfaces must be pre-cleaned prior to disinfecting.
>
> As a sanitizer: Apply product and leave on surface for 30 seconds before wiping clean. Pre-clean surfaces prior to sanitizing.

III AGENCY STANDARDS FOR PROPOSED CLAIMS

Disinfectants for Use on Hard Surfaces in Hospital or Medical Environments

The effectiveness of disinfectants for use on hard surfaces in hospital or medical environments must be substantiated by data derived using the AOAC Use-Dilution Method (for water soluble powders and liquid products) or the AOAC Germicidal Spray Products as Disinfectants Method (for spray products). Sixty carriers must be tested with each of 3 product samples, representing 3 different product lots, one of which is at least 60 days old, against *Salmonella enterica* (ATCC 10708; formerly *Salmonella choleraesuis*), *Staphylococcus aureus* (ATCC 6538), and *Pseudomonas aeruginosa* (ATCC 15442). To support products labeled as "disinfectants," killing on 59 out of 60 carriers is required to provide effectiveness at the 95% confidence level.

Virucides

The effectiveness of virucides against specific viruses must be supported by efficacy data that simulates, to the extent possible in the laboratory, the conditions under which the product is intended to be used. Carrier methods that are modifications of either the AOAC Use-Dilution Method (for liquid disinfectants) or the AOAC Germicidal Spray Products as Disinfectants Method (for spray disinfectants) must be used. To simulate in-use conditions, the specific virus to be treated must be inoculated onto hard surfaces, allowed to dry, and then treated with the product according to the directions for use on the product label. One surface for each of 2 different product lots of disinfectant must be tested against a recoverable virus titer of at least 10^4 from the test surface for a specified exposure period at room temperature. Then, the virus must be assayed by an appropriate virological technique, using a minimum of four determinations per each dilution assayed. Separate studies are required for each virus. The calculated viral titers must be reported with the test results. For the data to be considered acceptable, results must demonstrate complete inactivation of the virus at all dilutions. When cytotoxicity is evident, at least a 3-log reduction in titer must be demonstrated beyond the cytotoxic level.

Disinfectants for Use as Fungicides (Against Pathogenic Fungi, Using the AOAC Germicidal Spray Products as Disinfectants Method)

The effectiveness of liquid disinfectants against specific pathogenic fungi must be supported by efficacy data using an appropriate test. The AOAC Germicidal Spray Products as Disinfectants Method contains procedures for testing fungicidal activity. Ten carriers on each of 2 product samples representing 2 different product lots must be employed in the test. Killing of the specific pathogenic fungi on all carriers is required.

Note: As an interim policy, EPA is accepting studies with dried carrier counts that are at least 10^4 for *Trichophyton mentagrophytes*, *Aspergillus niger*, and *Candida albicans*. EPA recognizes laboratories are experiencing problems in maintaining dried carrier counts at the 10^6 level. This interim policy will be in effect until EPA determines that the laboratories are able to achieve consistent carrier counts at the 10^6 level.

Sanitizers (For Non-Food Contact Surfaces)

The effectiveness of sanitizers for non-food contact surfaces must be supported by data that show that the product will substantially reduce the numbers of test bacteria on a treated surface. The test surface(s) should represent the type(s) of surfaces recommended for treatment on the label, i.e., porous or non-porous. Products that are represented as "one-step sanitizers" should be tested with an appropriate organic soil load, such as 5 percent serum. Tests should be performed with each of 3 product samples, representing 3 different product lots, one of which is at least 60 days old against *Staphylococcus aureus* (ATCC 6538) and either *Klebsiella pneumoniae* (aberrant, ATCC 4352) or *Enterobacter aerogenes* (ATCC 13048 or 15038). Results must show a bacterial reduction of at least 99.9 percent over the parallel control within 5 minutes.

Mildewstats/Fungistats

The effectiveness of mildewstats and fungistats may be supported by efficacy data

derived using the EPA Hard Surface Mildew Fungistatic Test Method. All ten treated tiles must be free of fungal growth after 7 days. To be considered a valid test, untreated control tiles must be at least 50% covered with fungal growth after 7 days. Agency standards are presented in the Pesticide Assessment Guidelines, Subdivision G, Section 93-30, Product Performance, November 1982.

Supplemental Claims

An antimicrobial agent identified as a "one-step" disinfectant or as effective in the presence of organic soil must be tested for efficacy with an appropriate organic soil load, such as 5 percent serum.

IV COMMENTS ON THE SUBMITTED EFFICACY STUDIES

1. MRID 478898-09 "Standard Test Method for Efficacy of Sanitizers Recommended for Inanimate Non-Food Contact Surfaces (Modification for Spray Product Application)," Test Organism: *Escherichia coli* (ATCC 11229), for WC Complete, by Joy Salverda. Study conducted at ATS Labs. Study completion date – June 3, 2009. Project Number A07601.

This study was conducted against *Escherichia coli* (ATCC 11229). Two lots (Lot Nos. 1 and 2) of the product, WC Complete, were tested. The laboratory report referenced the Sanitizer Test from DIS/TSS-10 and the Standard Test Method for Efficacy of Sanitizers Recommended for Inanimate Non-Food Contact Surfaces (ASTM E1153). The product was received ready-to-use, as a trigger spray. The product was not tested in the presence of a 5% organic soil load. Five sterile glass carriers (1 inch x 1 inch) per product lot were inoculated with 20.0 µL of a 48±4 hour old suspension of test organism. The inoculum was spread to within 1/8 inch of the edges of each carrier. The carriers were dried for 20 minutes at 35-37°C at 40% relative humidity. Each carrier was transferred to a plastic Petri dish and was sprayed (3 pumps) until thoroughly wet with the product at a distance of 6-8 inches from the carrier surface. Each carrier remained in contact with the product for 30 seconds at 20°C at 16% relative humidity. Following exposure, each carrier was placed into a sterile vessel containing 20 mL of Letheen Broth with 0.14% Lecithin and 1.0% Tween 80. Following neutralization, the excess liquid from each plastic Petri dish was transferred to the corresponding neutralizer vessel containing the corresponding carrier. The vessels were rotated vigorously on an even plane for ~50 rotations to suspend the surviving organisms. Within 30 minutes of the addition of the neutralizer, 1.00 mL aliquots of the 10^0 and 10^{-1} dilutions were plated in duplicate on tryptic soy agar with 5% sheep's blood. All subcultures were incubated for ~51 hours at 35-37°C. Following incubation, the subcultures were visually enumerated. Controls included those for inoculum count, carrier quantitation, purity, sterility, and neutralization confirmation.

2. MRID 478898-10 "Standard Test Method for Efficacy of Sanitizers Recommended for Inanimate Non-Food Contact Surfaces (Modification for Spray Product Application)," Test Organisms: *Staphylococcus aureus* (ATCC 6538) and *Enterobacter aerogenes* (ATCC 13048), for WC Complete, by Becky Lien. Study conducted at ATS Labs. Study completion date – June 11, 2009. Project Number A07502.

This study was conducted against *Staphylococcus aureus* (ATCC 6538) and

Enterobacter aerogenes (ATCC 13048). Three lots (Lot Nos. 1, 2, and 3) of the product, WC Complete, were tested. The laboratory report referenced the Sanitizer Test from DIS/TSS-10 and the Standard Test Method for Efficacy of Sanitizers Recommended for Inanimate Non-Food Contact Surfaces (ASTM E1153). At least one of the three product lots tested (i.e., Lot No. 3) was at least 60 days old at the time of testing. The product was received ready-to-use, as a trigger spray. The product was not tested in the presence of a 5% organic soil load. Five sterile glass carriers (1 inch x 1 inch) per product lot per microorganism were inoculated with 20.0 µL of a 48±4 hour old suspension of test organism. The inoculum was spread to within 1/8 inch of the edges of each carrier. The carriers were dried for 20 minutes at 36.0°C at 41% relative humidity. Each carrier was transferred to a plastic Petri dish and was sprayed (3 pumps) until thoroughly wet with the product at a distance of 6-8 inches from the carrier surface. Each carrier remained in contact with the product for 30 seconds at 20°C. Following exposure, each carrier was placed into a sterile vessel containing 20 mL of Letheen Broth with 0.28% Lecithin and 2.0% Tween 80. Following neutralization, the excess liquid from each plastic Petri dish was transferred to the corresponding neutralizer vessel containing the corresponding carrier. The vessels were rotated vigorously on an even plane for ~50 rotations to suspend the surviving organisms. Within 30 minutes of the addition of the neutralizer, 1.00 mL aliquots of the 10^0 and 10^{-1} dilutions were plated in duplicate on tryptic soy agar with 5% sheep's blood. *Staphylococcus aureus* cultures were incubated for ~47.75 hours at 35-37°C. *Enterobacter aerogenes* cultures were incubated for 47.75 hours at 25-30°C. Following incubation, the subcultures were visually enumerated. Controls included those for inoculum count, carrier quantitation, purity, sterility, and neutralization confirmation.

Note: Protocol deviations/amendments reported in the study were reviewed.

3. MRID 478898-11 "Standard Test Method for Efficacy of Sanitizers Recommended for Inanimate Non-Food Contact Surfaces (Modification for Spray Product Application)," Test Organism: *Shigella sonnei* (ATCC 25931), for WC Complete, by Joy Salverda. Study conducted at ATS Labs. Study completion date – June 4, 2009. Project Number A07602.

This study was conducted against *Shigella sonnei* (ATCC 25931). Two lots (Lot Nos. 1 and 2) of the product, WC Complete, were tested. The laboratory report referenced the Sanitizer Test from DIS/TSS-10 and the Standard Test Method for Efficacy of Sanitizers Recommended for Inanimate Non-Food Contact Surfaces (ASTM E1153). The product was received ready-to-use, as a trigger spray. The product was not tested in the presence of a 5% organic soil load. Five sterile glass carriers (1 inch x 1 inch) per product lot were inoculated with 10.0 µL of a 48±4 hour old suspension of test organism (which differs from the 18-24 hour old suspension specified in DIS/TSS-10). The inoculum was spread to within 1/8 inch of the edges of each carrier. The carriers were dried for 20 minutes at 35-37°C at 40% relative humidity. Each carrier was transferred to a plastic Petri dish and was sprayed (3 pumps) until thoroughly wet with the product at a distance of 6-8 inches from the carrier surface. Each carrier remained in contact with the product for 30 seconds at 22.5°C. Following exposure, each carrier was placed into a sterile vessel containing 20 mL of Letheen Broth with 0.14% Lecithin and 1.0% Tween 80. Following neutralization, the excess liquid from each plastic Petri dish was transferred to the corresponding neutralizer vessel containing the corresponding carrier. The vessels were rotated vigorously on an even plane for ~50 rotations to suspend the surviving organisms. Within 30 minutes of the addition of the neutralizer, 1.00 mL aliquots of the 10^0 and 10^{-1} dilutions were plated in duplicate on tryptic soy agar with 5% sheep's blood. All cultures were incubated for ~46.75 hours at 35-37°C. The subcultures were stored for 1 day at 2-8°C

prior to examination. Following incubation and storage, the subcultures were visually enumerated. Controls included those for inoculum count, carrier quantitation, purity, sterility, and neutralization confirmation.

4. MRID 478898-12 "AOAC Germicidal Spray Method," Test Organisms: *Staphylococcus aureus* (ATCC 6538), *Salmonella enterica* (ATCC 10708), and *Pseudomonas aeruginosa* (ATCC 15442), for WC Complete, by Becky Lien. Study conducted at ATS Labs. Study completion date – June 11, 2009. Project Number A07415.

This study was conducted against *Staphylococcus aureus* (ATCC 6538), *Salmonella enterica* (ATCC 10708), and *Pseudomonas aeruginosa* (ATCC 15442). Three lots (Lot Nos. 1, 2, and 3) of the product, WC Complete, were tested using the AOAC Germicidal Spray Products as Disinfectants Method as described in the AOAC Official Methods of Analysis, 17th Edition, 2000. At least one of the product lots tested (i.e., Lot No. 3) was at least 60 days old at the time of testing. The product was received ready-to-use, as a trigger spray. Cultures of the challenge microorganisms were prepared in accordance with the published AOAC method, with the following exceptions: (1) the *Pseudomonas aeruginosa* culture was incubated for 48-54 hours at 35-37°C (which differs from the AOAC method specification of 18-24 hours); and (2) the cultures for *Staphylococcus aureus* and *Salmonella enterica* were incubated for 48-54 hours at 35-37°C (which differs from the AOAC method specification of 48 hours for all bacterial cultures except *Pseudomonas aeruginosa*). Fetal bovine serum was added to each culture to achieve a 5% organic soil load. Sixty (60) glass slide carriers (18 mm x 36 mm) per product lot per test organism were inoculated with 10.0 µL of a 48-54 hour old suspension of test organism. Inoculum was uniformly spread over the entire surface of each carrier. The carriers were dried for 30 minutes at 35-37°C at 41% relative humidity. For each lot of product, separate carriers were sprayed (3 pumps) until thoroughly wet with the product at a distance of 6-8 inches from the carrier surface. The carriers were allowed to remain wet for 10 minutes at 21°C at 26% relative humidity. Following the exposure period, the remaining liquid was drained from each carrier. Individual carriers were transferred to 20 mL of Letheen Broth with 0.07% Lecithin and 0.5% Tween 80 to neutralize. [It is unknown whether tubes containing neutralizer were shaken thoroughly after addition of the carriers, as specified in the AOAC method.] All subcultures were incubated for 47.5 hours at 35-37°C (which differs from the AOAC method specification of 48 hours at 37°C). Following incubation, the subcultures were examined for the presence or absence of visible growth. Controls included those for carrier population, purity, sterility, viability, and neutralization confirmation.

5. MRID 478898-13 "Virucidal Efficacy of a Disinfectant for Use on Inanimate Environmental Surfaces, Virus: Rhinovirus type 39" for WC Complete, by Mary J. Miller. Study conducted at ATS Labs. Study completion date – June 15, 2009. Project Number A07598.

This study was conducted against Rhinovirus type 39 (Strain 209; ATCC VR-340), using WI-38 cells (ATCC CCL-75; propagated in-house) as the host system. Two lots (Lot Nos. 1 and 3) of the product, WC Complete, were tested according to ATS Labs Protocol No. DIA01040109.R39 (copy provided). The product was received ready-to-use, as a trigger spray. The stock virus culture contained 1% fetal bovine serum as the organic soil load. Films of virus were prepared by spreading 0.2 mL of virus inoculum uniformly over the undersides of separate

sterile glass Petri dishes. The virus films were air-dried for 20 minutes at 20.0°C at 51% relative humidity. One dried virus film per product lot was tested. For each lot of product, separate dried virus films were sprayed (3 pumps) until thoroughly wet with the product at a distance of 6-8 inches from the carrier surface. The carriers were allowed to remain wet for 30 seconds at 20.0°C. Following exposure, the plates were scraped with a cell scraper to re-suspend the contents. The virus-disinfectant mixtures were passed immediately through individual Sephadex columns, and diluted serially in Minimum Essential Medium supplemented with 10% heat-inactivated fetal bovine serum, 10 µg/mL gentamicin, 100 units/mL penicillin, and 2.5 µg/mL amphotericin B. WI-38 cells in multi-well culture dishes were inoculated in quadruplicate with 0.1 mL of the dilutions. The cultures were incubated at 31-35°C in a humidified atmosphere of 5-7% CO_2. The cultures were scored periodically for 7 days for the presence or absence of unspecified cytopathic effects, cytotoxicity, and viability. Controls included those for dried virus count, cytotoxicity, and neutralization. Viral and cytotoxicity titers were calculated by the method of Spearman Karber.

Note: The laboratory reported a failed study set up on April 20, 2009. In that study, a 4 log_{10} of infectivity from the dried virus control was not achieved. The laboratory did not accept the assay. These data were not used to evaluate efficacy of the product. Testing was repeated on May 27, 2009. See page 8 and Attachment 1 of the laboratory report.

6. MRID 478898-14 "Virucidal Efficacy of a Disinfectant for Use on Inanimate Environmental Surfaces, Virus: Influenza A virus" for WC Complete, by Mary J. Miller. Study conducted at ATS Labs. Study completion date – June 15, 2009. Project Number A07597.

This study was conducted against Influenza A virus (Strain Hong Kong; ATCC VR-544), using RMK cells (Rhesus monkey kidney cells; obtained from ViroMed Laboratories, Inc., Cell Culture Division) as the host system. Two lots (Lot Nos. 1 and 3) of the product, WC Complete, were tested according to ATS Labs Protocol No. DIA01040109.FLUA (copy provided). The product was received ready-to-use, as a trigger spray. The stock virus culture contained 1% fetal bovine serum as the organic soil load. Films of virus were prepared by spreading 0.2 mL of virus inoculum uniformly over the undersides of separate sterile glass Petri dishes. The virus films were air-dried for 20 minutes at 20.0°C at 50% relative humidity. One dried virus film per product lot was tested. For each lot of product, separate dried virus films were sprayed (3 pumps) until thoroughly wet with the product at a distance of 6-8 inches from the carrier surface. The carriers were allowed to remain wet for 30 seconds at 21.0°C. Following exposure, the plates were scraped with a cell scraper to re-suspend the contents. The virus-disinfectant mixtures were passed immediately through individual Sephadex columns, and diluted serially in Minimum Essential Medium supplemented with 1% heat-inactivated fetal bovine serum, 10 µg/mL gentamicin, 100 units/mL penicillin, and 2.5 µg/mL amphotericin B. RMK cells in multi-well culture dishes were inoculated in quadruplicate with 0.1 mL of the dilutions. The cultures were incubated at 36-38°C in a humidified atmosphere of 5-7% CO_2. The cultures were scored periodically for 7 days for the presence or absence of unspecified cytopathic effects, cytotoxicity, and viability. Controls included those for dried virus count, cytotoxicity, and neutralization. Viral and cytotoxicity titers were calculated by the method of Spearman Karber.

7. MRID 478898-15 "Virucidal Efficacy of a Disinfectant for Use on Inanimate Environmental Surfaces, Virus: Rotavirus" for WC Complete, by Mary J. Miller. Study conducted at ATS Labs. Study completion date – June 15, 2009. Project Number A07596.

This study was conducted against Rotavirus (Strain WA; obtained from the University of Ottawa), using MA-104 cells (Rhesus monkey kidney cells; obtained from Diagnostic Hybrids Inc., Athens, OH; propagated in-house) as the host system. Two lots (Lot Nos. 1 and 3) of the product, WC Complete, were tested according to ATS Labs Protocol No. DIA01040109.ROT (copy provided). The product was received ready-to-use, as a trigger spray. The stock virus culture was adjusted to contain 1% fetal bovine serum as the organic soil load. Films of virus were prepared by spreading 0.2 mL of virus inoculum uniformly over the undersides of separate sterile glass Petri dishes. The virus films were air-dried for 20 minutes at 20.0°C at 50% relative humidity. One dried virus film per product lot was tested. For each lot of product, separate dried virus films were sprayed (3 pumps) until thoroughly wet with the product at a distance of 6-8 inches from the carrier surface. The carriers were allowed to remain wet for 30 seconds at 20.0°C. Following exposure, the plates were scraped with a cell scraper to re-suspend the contents. The virus-disinfectant mixtures were passed immediately through individual Sephadex columns, and diluted serially in serum-free Minimum Essential Medium supplemented with 10 µg/mL gentamicin, 100 units/mL penicillin, 2.5 µg/mL amphotericin B, 0.5 µg/mL trypsin, and 2.0 mM L-glutamine. MA-104 cells in multi-well culture dishes were inoculated in quadruplicate with 0.1 mL of the dilutions. The inoculum was allowed to adsorb for 60 minutes at 36-38°C in a humidified atmosphere of 5-7% CO_2. Following adsorption, the cultures were re-fed and incubated at 36-38°C in a humidified atmosphere of 5-7% CO_2. The cultures were scored periodically for 7 days for the presence or absence of unspecified cytopathic effects, cytotoxicity, and viability. Controls included those for dried virus count, cytotoxicity, and neutralization. Viral and cytotoxicity titers were calculated by the method of Spearman Karber.

8. MRID 478898-16 "Fungicidal Germicidal Spray Method, Test Organism: *Trichophyton mentagrophytes* (ATCC 9533)" for WC Complete, by Becky Lien. Study conducted at ATS Labs. Study completion date – June 9, 2009. Project Number A07599.

This study was conducted against *Trichophyton mentagrophytes* (ATCC 9533). Two lots (Lot Nos. 2 and 3) of the product, WC Complete, were tested using the AOAC Germicidal Spray Products as Disinfectants Method as described in the AOAC Official Methods of Analysis, 17th Edition, 2000. The product was received ready-to-use, as a trigger spray. A culture of the challenge microorganism was prepared in accordance with the published AOAC method, with the following exception: the culture was not standardized (which differs from the AOAC method specification of diluting the stock suspension using physiological NaCl solution so that it contains 5×10^6 conidia/mL (for *Trichophyton mentagrophytes*)). The product was not tested in the presence of a 5% organic soil load. Ten (10) glass slide carriers (18 mm x 36 mm) were inoculated with 10.0 µL (i.e., 0.01 mL) of a 10 day old suspension of test organism. The inoculum was uniformly spread over the surface of each carrier. The carriers were dried for 30 minutes at 35-37°C at 42% relative humidity. For each lot of product, separate carriers were sprayed (using 3 pumps) until thoroughly wet with the product at a distance of 6-8 inches from the carrier surface. The carriers were allowed to remain wet for 30 seconds at 22.84°C. Following the exposure period, the remaining liquid was drained from each carrier. Individual carriers were transferred to 20 mL of Sabouraud Dextrose Broth with 0.07% Lecithin and 0.5%

Tween 80 to neutralize. Carriers were transferred from the primary subcultures into individual secondary subcultures containing 20 mL of Sabouraud Dextrose Broth with 0.07% Lecithin and 0.5% Tween 80 at least 30 minutes following the first transfer. [It is unknown whether tubes containing neutralizer were shaken thoroughly after addition of the carriers, as specified in the AOAC method.] All subcultures were incubated for 10 days at 25-30°C. Following incubation, the subcultures were examined for the presence or absence of visible growth. Controls included those for carrier population, purity, sterility, viability, and neutralization confirmation.

9. MRID 478898-17 "EPA Hard Surface Mildew-Fungistatic Test, Test Organism: *Aspergillus niger* (ATCC 6275)" for WC Complete, by Becky Lien. Study conducted at ATS Labs. Study completion date – June 9, 2009. Project Number A07600.

This study was conducted against *Aspergillus niger* (ATCC 6275). Two lots (Lot Nos. 2 and 3) of the product, WC Complete, were tested using the EPA Hard Surface Mildew Fungistatic Test Method. The product was received ready-to-use, as a trigger spray. Testing was not conducted in the presence of an organic soil load. A culture of the challenge microorganism was prepared in accordance with the EPA method, with the following deviations: the culture was incubated for 11 days at 25-30°C (which differs from the EPA method specification of 7-10 days at 25°C). A 1.00 mL aliquot of the conidial suspension was added to 20.0 mL of sterile Czapek's solution. Sterile 1 x 1 inch glazed ceramic tiles (10 per treatment) were used (which complies with the EPA method specification of 25 mm square tiles with a glazed surface). The tiles were sprayed (3 pumps) until thoroughly wet with the product at a distance of 6-8 inches from the carrier surface. After treatment, tiles were placed in a vertical or near vertical position to allow excess liquid to drain. Following treatment, the tiles were dried for 49 minutes at 36.0°C. Following the drying period, the surfaces of each test tile and each untreated control tile were sprayed with the *Aspergillus niger* conidia-Czapek suspension using the DeVilbiss #151 Atomizer. The tiles were returned to a 36.0°C incubator and dried for 4 hours and 45 minutes. Each tile (treated side up) was placed in an individual Petri dish containing hardened sterile water agar. The plates were incubated for 7 days at 25-30°C at a minimum of 95% relative humidity. The tiles were examined for the presence or absence of fungal growth after 7 days of incubation. When no growth was visually observed, a magnified examination was performed (magnification specification not specified). Controls included those for purity and sterility.

V RESULTS

MRID Number	Organism	No. Exhibiting Growth/ Total No. Tested			Carrier Population (CFU/ Carrier)
		Lot No. 1	Lot No. 2	Lot No. 3	
10-Minute Exposure Time					
478898-12	*Staphylococcus aureus*	0/60	0/60	1/60	1.78×10^6
	Salmonella enterica	0/60	0/60	0/60	3.1×10^4
	Pseudomonas aeruginosa	1/60	0/60	0/60	4.0×10^6

MRID Number	Organism	No. Exhibiting Growth/ Total No. Tested			Carrier Population (CFU/ Carrier)
		Lot No. 1	Lot No. 2	Lot No. 3	
30-Second Exposure Time					
478898-16	*Trichophyton mentagrophytes*	--- ---	1°=10/10 2°=10/10	1°=10/10 2°=10/10	9.9×10^5

MRID Number	Organism	Results			Dried Virus Count ($TCID_{50}$/0.1 mL)
			Lot No. 1	Lot No. 3	
478898-13	Rhinovirus type 39	10^{-1} dilution	Cytotoxicity	Cytotoxicity	$10^{5.0}$
		10^{-2} to 10^{-6} dilutions	Complete inactivation	Complete inactivation	
		$TCID_{50}$/0.1 mL	$\leq 10^{1.5}$	$\leq 10^{1.5}$	
		Log reduction	$\geq 3.5 \log_{10}$	$\geq 3.5 \log_{10}$	
478898-14	Influenza A virus	10^{-1} dilution	Cytotoxicity	Complete inactivation	$10^{5.5}$
		10^{-2} to 10^{-7} dilutions	Complete inactivation		
		$TCID_{50}$/0.1 mL	$\leq 10^{1.5}$	$\leq 10^{0.5}$	
		Log reduction	$\geq 4.0 \log_{10}$	$\geq 5.0 \log_{10}$	
478898-15	Rotavirus	10^{-1} to 10^{-2} dilutions	Cytotoxicity	Cytotoxicity	$10^{5.75}$
		10^{-3} to 10^{-7} dilutions	Complete inactivation	Complete inactivation	
		$TCID_{50}$/0.1 mL	$\leq 10^{2.5}$	$\leq 10^{2.5}$	
		Log reduction	$\geq 3.25 \log_{10}$	$\geq 3.25 \log_{10}$	

MRID Number	Organism	Lot No.	Total No. Surviving	Microbes Initially Present	Percent Reduction
			(CFU/carrier)		
478898-09	*Escherichia coli*	1	$<2.00 \times 10^1$	1.70×10^7	>99.9
		2	$<2.00 \times 10^1$	1.70×10^7	>99.9
478898-10	*Staphylococcus aureus*	1	$<2.75 \times 10^1$	1.00×10^7	>99.9
		2	$<2.00 \times 10^1$	1.00×10^7	>99.9
		3	$<2.00 \times 10^1$	1.00×10^7	>99.9
	Enterococcus aerogenes	1	$<3.55 \times 10^1$	2.75×10^7	>99.9
		2	$<3.80 \times 10^1$	2.75×10^7	>99.9
		3	$<2.00 \times 10^1$	2.75×10^7	>99.9

478898-11	*Shigella sonnei*	1 2	<2.00 x 10^1 <2.00 x 10^1	3.98 x 10^6 3.98 x 10^6	>99.9 >99.9

MRID Number	Organism	No. Exhibiting Growth/ Total No. Tested		Control Tiles
		Lot No. 2	Lot No. 3	
478898-17	*Aspergillus niger*	0/10	0/10	10/10*

* At least 50% fungal growth on each untreated control tile was observed.

VI CONCLUSIONS

1. The submitted efficacy data support the use of the product, WC Complete, as a disinfectant with bactericidal activity against the following microorganisms on hard, non-porous surfaces in the presence of a 5% organic soil load for a 10-minute contact time:

Staphylococcus aureus	MRID 478898-12
Salmonella enterica	MRID 478898-12
Pseudomonas aeruginosa	MRID 478898-12

Killing was observed in the subcultures of at least 59 of the 60 carriers tested against the required number of product lots. At least one of the product lots tested was at least 60 days old at the time of testing. Neutralization confirmation testing showed positive growth of the microorganisms. Purity controls were reported as pure. Viability controls were positive for growth. Sterility controls did not show growth.

2. The submitted efficacy data support the use of the product, WC Complete, as a disinfectant with virucidal activity against the following microorganisms on hard, non-porous surfaces in the presence of a 1% organic soil load for a 30-second contact time:

Rhinovirus type 39	MRID 478898-13
Influenza A virus	MRID 478898-14
Rotavirus	MRID 478898-15

Recoverable virus titers of at least 10^4 were achieved. In studies against Rotavirus, cytotoxicity was observed in the 10^{-1} and 10^{-2} dilutions. Complete inactivation (no growth) was indicated in all higher dilutions tested. In studies against the other viruses, cytotoxicity was observed in the 10^{-1} dilution. Complete inactivation (no growth) was indicated in all higher dilutions tested. At least a 3-log reduction in titer was demonstrated beyond the cytotoxic level. Testing in 1% organic soil load does not support "one-step" claims.

3. The submitted efficacy data (MRID 478898-16) do not support the use of the product, WC Complete, as a disinfectant with fungicidal activity against *Trichophyton mentagrophytes* on hard, non-porous surfaces for a 30-second contact time. Acceptable killing was not observed in the subcultures of the required number of carriers tested against the required number of product lots. Neutralization confirmation testing showed positive growth of the microorganism. Purity controls were reported as pure. Viability controls were positive for growth. Sterility controls did not show growth.

4. The submitted efficacy data support the use of the product, WC Complete, as a sanitizer against the following microorganisms on pre-cleaned, hard, non-porous, non-food contact surfaces at a 30-second contact time:

Escherichia coli	MRID 478898-09
Staphylococcus aureus	MRID 478898-10
Enterococcus aerogenes	MRID 478898-10
Shigella sonnei	MRID 478898-11

Bacterial reductions of at least 99.9 percent over the parallel control were observed within 5 minutes (i.e., 30 seconds specifically). In testing against *Staphylococcus aureus* and *Enterobacter aerogenes*, at least one of the product lots tested was at least 60 days old at the time of testing. The control count demonstrated an average of at least 7.5×10^5 surviving organisms, which is the criterion set forth in ASTM 1153. Neutralization confirmation testing met the acceptance criterion of growth within 1 $\log_{10}$ of the numbers control. Purity controls were reported as pure. Sterility controls did not show growth.

5. The submitted efficacy data (MRID 478898-17) support the use of the product, WC Complete, as a mildewstat against *Aspergillus niger* on hard, non-porous surfaces. No growth was observed 7 days after treatment. Testing was conducted on 2 product lots. Untreated control tiles exhibited growth of *Aspergillus niger* on 75% to 90% of each untreated tile surface. Purity controls were reported as pure. Sterility controls did not show growth.

VII RECOMMENDATIONS

1. The proposed label claims that the product, WC Complete, is an effective "one-step" disinfectant against the following microorganisms on hard, non-porous surfaces for a 10-minute contact time:

Staphylococcus aureus
Salmonella enterica
Pseudomonas aeruginosa

These claims are acceptable as they are supported by the submitted data.

2. The proposed label claims that the product, WC Complete, is an effective "one-step" disinfectant against the following microorganisms on hard, non-porous surfaces for a 30-second contact time:

Influenza A virus
Rhinovirus type 39
Rotavirus

These claims are acceptable as they are supported by the submitted data. Note that testing was conducted in the presence of a 1% organic soil load (not a 5% organic soil load).

√3. The proposed label claims that the product, WC Complete, is an effective disinfectant against H1N1 virus on hard, non-porous surfaces for a 30-second contact time. As this is a new product, there exist no claims against any Influenza A viruses, to support claims against H1N1 (see guidance http://www.epa.gov/oppad001/h1n1-guide.html. References to the H1N1 virus

must be deleted from the proposed label.

4. The proposed label claims that the product, WC Complete, is an effective sanitizer against the following microorganisms on pre-cleaned, hard, non-porous, non-food contact surfaces at a 30-second contact time:

Escherichia coli
Staphylococcus aureus
Enterobacter aerogenes
Shigella sonnei

These claims are acceptable as they are supported by the submitted data.

5. The proposed label claims that the product, WC Complete, inhibits the growth of mold and mildew. This claim is acceptable as it is supported by the submitted data. The proposed label must be revised to include directions for use of the product as a mildewstat.

6. The product is not a "One-Step" disinfectant. Data against viruses was not conducted in the presence of 5% organic soil.

7. Germ claims must be qualified, as data against T. mentagrophytes was unacceptable.