UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON, DC 20460

OFFICE OF PREVENTION, PESTICIDES
AND TOXIC SUBSTANCES
Antimicrobials Division

August 22, 2003

MEMORANDUM:

Subject: Efficacy Review EPA Reg. No. 73478-1 *Bleach Wipe*
DP Barcode 289796
Case No. 069069

rom: Nancy Whyte, Microbiologist NWW
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Applicant: Infection Control Ventures:
1900 N. Austin Avenue, Suite 39E
Chicago, IL 60639

Formulation Label:

Active Ingredient(s)	**%/wt**
Sodium hypochlorite	0.525%
Other ingredients	99.475%
Total	100.000%

I. Background:

This data package is a re-submission of information in response to an EPA letter dated April 2, 2003. The product, Bleach-Wipe™ (EPA Reg. No. 73478-1), is a

disinfectant (bactericide, tuberculocide) for use on hard, non-porous surfaces, including use in patient care, institutional, and commercial environments. All studies were conducted at MicroBioTest, Inc., located at 105B Carpenter Drive in Sterling, VA 20164.

This data package contained EPA correspondence dated April 2, 2003, the applicant's response (date not specified), EPA Form 8570-1 (Application for Pesticide), two studies (MRID Nos. 459173-01 and 459173-02), Statements of No Data Confidentiality Claims for both studies, and the proposed label.

In December 2002, the contractor Dyncorp Systems & Solutions (DSS) reviewed two laboratory reports (i.e., MRID Nos. 457830-01 and 457830-02) submitted by the applicant for the product, Bleach-Wipe™. DSS found the studies to be deficient at that time. EPA concurred that the efficacy data were not acceptable to support label claims for the product as a disinfectant and tuberculocidal product. The studies were conducted on an insufficient number of product lots. One of the studies was conducted on an insufficient number of carriers. In addition, the studies were not performed in the presence of an organic soil load, which was required because the label directions did not include a pre-cleaning step. The two studies discussed in this report are the applicant's effort to correct the identified deficiencies.

II. Use Directions:

The product is designed to be used for disinfecting hard, non-porous surfaces on medical, dental and laboratory counters, exam tables, carts, point-of-care equipment, telephones, and sink tops. Directions on the proposed label rear panel provided the following information regarding preparation and use of the ready-to-use product: Using personal protective equipments (gloves), open wipe packet. Remove pre-moistened 8"x10" towelette. Apply towelette and wipe desired surface. Allow surface to air dry. Discard used towelette. The front panel of the label, in part, reads:

1 minute minimum contact time
Tuberculocidal & Bactericidal
Maximum surface area that can be disinfected per towelette is 3' x 3'

III. Agency Standards for Proposed Change:

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 Test (for spray products). Sixty carriers must be tested with each of 3 product samples, representing 3 different batches, one of which is at least 60 days old, against *Salmonella choleraesuis* (ATCC 10708), *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. The above Agency standards are presented in DIS/TSS-1.

Disinfectants for Use as Tuberculocides (Using the AOAC Tuberculocidal Activity Test Method or the AOAC Germicidal Spray Products Test Method)

Disinfectants may bear additional label claims of effectiveness as tuberculocides when supported by appropriate tuberculocidal effectiveness data. Certain chemical classes (i.e., glutaraldehyde and quaternary ammonium compounds) are required to undergo validation testing in addition to basic testing. Products that are formulated with other chemical groups do not require validation testing. Products may be tested using one of four recommended methods: the AOAC Tuberculocidal Test Method, Tuberculocidal Activity of Disinfectants Test Method with significant modification of the standard test conditions of contact time and/or temperature, Quantitative Tuberculocidal Activity Test Method, and AOAC Germicidal Spray Products Test Method.

When using the existing or modified AOAC Tuberculocidal Activity Test Methods, or the AOAC Germicidal Spray Products Test Method, 10 carriers for each of 2 samples, representing 2 different batches of product, must be tested against *Mycobacterium bovis* BCG (a member of the *Mycobacterium tuberculosis* species complex). When using the existing or modified AOAC Tuberculocidal Activity Test Method, or the AOAC Germicidal Spray Products Test Method, killing on all carriers/slides as demonstrated in Modified Proskauer-Beck Broth, and no growth in any of the inoculated tubes of two additional media (i.e., Middlebrook 7H9 Broth Difco B, Kirchners Medium, and/or TB Broth Base) is required. Agency standards are presented in EPA DIS/TSS-6, Subdivision G Guidelines, and "EPA Data Call-in Notice for Tuberculocidal Claims," dated June 13, 1986.

Disinfectants for Use on Hard Surfaces Using Presaturated or Impregnated Towelettes

Towelette products represent a unique combination of antimicrobial chemical and applicator, pre-packaged as a unit in fixed proportions. As such, the complete product, as offered for sale, should be tested according to the directions for use to ensure the product's effectiveness in disinfecting hard surfaces. The standard test methods available for hard surface disinfectants (i.e., AOAC Use-Dilution Method, AOAC Germicidal Spray Products Test), if followed exactly, would not closely simulate the way a towelette product is used. Agency guidelines recommend that a simulated-use test be conducted by modifying the AOAC Germicidal Spray Products Test. Agency guidelines further recommend that instead of spraying the inoculated surface of the glass slide, the product should be tested by wiping the surface of the glass slide with the saturated towelette, and then subculturing the slides after a specified holding time. Liquid expressed from the used towelette should also be subcultured and is expected to be negative for growth. Sixty carriers must be tested with each of 3 product samples, representing 3 different batches, one of which is at least 60 days old. The towelette should be removed from its container and subsequently handled with sterile gloves. To support products labeled as "disinfectants," killing on 59 out of 60 carriers (for both the slide subculture and the expressed liquid from the towelette) is required to provide effectiveness at the 95% confidence level. The above Agency standards are presented in DIS/TSS-1 and EPA Pesticide Assessment Guidelines, Subdivision G, §91-2(h), Pre-saturated or impregnated towelettes.

Supplemental Claims

An antimicrobial agent identified as a "one-step" cleaner-disinfectant, cleaner-sanitizer, or one intended to be effective in the presence of organic soil must be tested for efficacy with

an appropriate organic soil load, such as 5 percent serum. These Agency standards are presented in DIS/TSS-2.

IV. Summary of Submitted Studies:

1. MRID 459173-01 "Testing Pre-Saturated or Impregnated Towelettes for Hard Surface Disinfection" for Bleach Wipe, by Diane M. LeClercq. Study conducted at MicroBioTest, Inc. Study completion date – December 30, 2002. Laboratory Project Identification Number 480-103.

This study was conducted against *Staphylococcus aureus* (ATCC 6538), *Pseudomonas aeruginosa* (ATCC 15442), and *Salmonella choleraesuis* (ATCC 10708) in the presence of a 5% organic soil load (heat-inactivated fetal bovine serum). Three lots (Lot Nos. 11152002, 10152002, and 09152002, identified by the applicant as at least 60 days old) of the product, Bleach Wipe, were tested using the MicroBioTest Inc. protocol "Testing Pre-Saturated or Impregnated Towelettes for Hard Surface Disinfection," November 13, 2002 (copy provided). This protocol is based on the AOAC Germicidal Spray Products as Disinfectants Method, as described in the AOAC Official Methods of Analysis, 16th Edition, 1995. Sixty (60) glass slide carriers were tested against each lot of product. Aliquots of 0.01 mL of 48-54 hour cultures were transferred onto sterile carriers. The inoculum was spread uniformly over a one square inch area. The carriers were dried for 20-40 minutes at 37±2°C. One towelette was used to wipe ten glass slide carriers. Each carrier was wiped 3 times horizontally and 3 times vertically. Between carriers, the towelette was rotated to expose the maximum amount of unused surface. After 1 minute at 20-22°C, the carriers were transferred to 20 mL of Letheen Broth containing 0.1% $Na_2S_2O_3$ (LB+). Following the wiping of the tenth carrier, the towelette was held in a sterile Petri dish for 1 minute at 20-22°C. Liquid remaining in the towelette was expressed into a sterile Petri dish, and a 0.1 mL aliquot was cultured into 20 mL of LB+. The tubes were incubated for 48±2 hours at 37±2°C. All observations were recorded as growth or no growth. Controls included viability, dried carrier counts, neutralizer effectiveness, sterility, confirmation of the challenge microorganisms, and bacteriostasis. This study was contained in one document, MRID No 450172-01.
Note: Protocol deviations/amendments reported in the study were reviewed and found to be acceptable. One protocol deviation described the inoculum amount added during the neutralization effectiveness control. Instead of fewer than 200 CFU/tube specified in the laboratory's protocol, 330, 369, or 547 CFU was added to each tube of neutralizing broth (depending on the organism).

2. MRID 459173-02 "Testing Pre-Saturated or Impregnated Towelettes for Tuberculocidal Effectiveness" for Bleach Wipe, by Diane M. LeClercq. Study conducted at MicroBioTest, Inc. Study completion date – March 20, 2003. Laboratory Project Identification Number 480-104.

This study was conducted against *Mycobacterium bovis* BCG in the presence of a 5% organic soil load (heat-inactivated horse serum). Two lots (Lot Nos. 11152002 and 10152002) of the product, Bleach Wipe, were tested using the MicroBioTest, Inc. protocol "Testing Pre-Saturated or Impregnated Towelettes for Tuberculocidal Effectiveness," November 13, 2002 (copy provided). This protocol is based on the AOAC Confirmative in vitro Test for Determining Tuberculocidal Activity and EPA Efficacy Data Requirements for Pre-Saturated or Impregnated

Towelettes for Hard Surface Disinfection. Ten (10) glass slides were tested against each lot of product. Aliquots of 0.01 mL of inoculum were transferred onto the sterile carriers. The inoculum was spread uniformly over a one square inch area. The carriers were dried for 30-40 minutes at 37±2°C. One towelette was used to wipe ten glass slide carriers. Each carrier was wiped 3 times horizontally and 3 times vertically. Between carriers, the towelette was rotated to expose the maximum amount of unused surface. Each slide was held for 1 minute at 20±1°C and then was transferred into tubes of 20 mL of neutralizing broth of Modified Proskauer-Beck Medium (MPBM) containing 0.1% $Na_2S_2O_3$ (MPBM+). From each tube of neutralizer, 2.0 mL was subcultured to a tube containing 20 mL of Kirchner's Medium and a tube containing 20 mL of Middlebrook 7H9 Broth. After 10 minutes, the slides were transferred to 20 mL of MPBM. Following the wiping of the tenth carrier, the towelette was held for 1 minute at 20±1°C. Liquid remaining in the towelette was expressed into a sterile Petri dish, and a 0.1 mL aliquot was cultured into 20 mL of MPBM+. The tubes used for primary and secondary transfers were incubated at 37±2°C for 60 days and, if after 60 days, there were no signs of visible culture growth, the tubes were incubated for an additional 30 days. All observations were recorded as growth or no growth. Controls included viability, neutralizer effectiveness, sterility, carrier counts, and confirmation of the challenge microorganism. This sturdy was contained in one document, MRID No. 459172-02

Note: Protocol deviations/amendments reported in the study were reviewed and found to be acceptable.

Results

MRID Number	Organism	No. Exhibiting Growth/Total No. Tested						Dried Carrier Count (Average CFU/ carrier)
		Lot No. 11152002		Lot No. 10152002		Lot No. 09152002		
		C*	EL*	C	EL	C	EL	
459173-01	*Staphylococcus aureus*	0/60	0/6	0/60	0/6	0/60	0/6	1.4×10^6
459173-01	*Pseudomonas aeruginosa*	0/60	0/6	0/60	0/6	0/60	0/6	1.1×10^6
459173-01	*Salmonella choleraesuis*	0/60	0/6	0/60	0/6	0/60	0/6	4.0×10^4

*C=Carriers
EL=Expressed Liquid

MRID Number	Organism/ Recovery Medium	No. Exhibiting Growth/Total No. Tested				Dried Carrier Count (Avg. CFU/ carrier)
		Carriers (90 days)		Expressed Liquid (90 days)		
		Lot No. 11152002	Lot No. 10152002	Lot No. 11152002	Lot No. 10152002	
459173-02	*Mycobacterium bovis* BCG			0/1	0/1	5.4×10^4
	MPBM+	0/10	0/10			
	MPBM	0/10	0/10			
	Kirchner's Medium	0/10	0/10			
	Middlebrook 7H9 Broth	0/10	0/10			

V. Labeling:

1. Although requested by the Agency in its letter dated January 30, 2003 that the available chlorine amount should be listed in the label ingredient claims statement, that statement does not appear in the label, dated April 24, 2003 which was resubmitted by the registrant..

2. Other labeling items noted by the Agency have been added. The maximum surface area that can be covered by one towelette is listed, and the minimum contact time has been added in directions for use.

VI. Comments and Recommendations:

1 The submitted efficacy data (MRID No. 459173-01) demonstrates the effectiveness of the product Bleach-Wipe™ as a disinfectant with bactericidal activity when tested against *Pseudomonas aeruginosa* ATCC 15442 *Staphylococcus aureus* ATCC 6538, and *Salmonella choleraesuis* ATCC 10708in the presence of a 5% organic soil load (heat-inactivated fetal bovine serum) on hard, non-porous surfaces for a contact time of 1 minute. The laboratory report indicates that one towelette was used to wipe 10 glass slide carriers. No growth was observed in the subcultures of the carriers tested against three lots of the product or in the liquid expressed from the towelettes used. Dried carrier counts were at least 10^4. The laboratory report indicates that neutralization effectiveness and viability controls exhibited growth. The laboratory protocol states that fewer than 200 CFU of the challenge microorganism will be added to each tube of neutralizing broth.

2. The submitted efficacy data (MRID No. 459173-02) support the use of the product, Bleach-Wipe™, as a disinfectant with tuberculocidal activity when tested against *Mycobacterium bovis* BCG in the presence of a 5% organic soil load (heat-inactivated horse serum) on hard, non-porous surfaces for a contact time of 1 minute. No growth

was observed in the subcultures of the carriers tested against two lots of the product or in the liquid expressed from the towelettes used in any of the following media: MPBM, MPBM+, Middlebrook 7H9 Broth, and Kirchner's Medium. The dried carrier count was at least 10^4. According to the laboratory report, all of the controls met the criteria established for a valid test. Because the product, Bleach-Wipe™, is a sodium hypochlorite product, validation testing is not required.