UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON, DC 20460

OFFICE OF
PREVENTION,
PESTICIDES
AND TOXIC
SUBSTANCES

September 30, 2009

MEMORANDUM

Subject: Efficacy Review for EPA Reg. No. 777-99, Brace
DP Barcode: 368117

From: Tajah L. Blackburn, Ph.D., Microbiologist
Efficacy Evaluation Team
Product Science Branch
Antimicrobials Division (7510P)

9/30/09

To: Sharon Carlisle Acting PM 34/ Renae Whitaker
Regulatory Management Branch II
Antimicrobials Division (7510P)

Applicant: Reckitt Benckiser, Inc.
Morris Corporate Center IV
399 Interspace Parkway
PO Box 225
Parsippany, NJ 07054-0225

Formulation from the Label:

Active Ingredient(s)	% by wt.
Alkyl (50% C_{14}, 40% C_{12}, 10% C_{16})	
Dimethyl benzyl ammonium chloride	0.10%
Ethanol	58.00%
Other Ingredients	41.90%
Total	100.00%

I BACKGROUND

The current submission is in response to the Agency's letter dated November 18, 2008 and the Efficacy Review Letter dated September 30, 2008. According to the registrant's letter (dated July 20, 2009), "this submission includes the confirmatory testing requested in the Agency's letter....These letters request confirmatory data to be submitted using the AOAC Germicidal Spray Products Test method without the coarse filtration step for formula numbers 1178-172, 1338-015, 1338-016, 1338-020, and 1338-027 against *Staphylococcus aureus*, *Salmonella enterica*, and *Pseudomonas aeruginosa*." Studies were conducted at Reckitt Benckiser Inc., Microbiology Laboratory, located at One Philips Parkway, in Montvale, NJ 07645.

II USE DIRECTIONS

The product is designed to be used for disinfecting and sanitizing hard, non-porous surfaces such as appliance exteriors, bathtubs, bed frames and bed springs, cabinets, cat litter boxes, carts, counter tops, cuspidors, furniture, diaper changing tables, diaper pails, dish pails and racks, door knobs, drinking fountains, examination tables, faucets, fixtures, floors, garbage cans, lamps, laundry baskets, light switches, mattress covers, metal blinds, mirrors, outdoor furniture, recycling bins, remote controls, showers, sinks, sports equipment, stretchers, toilets, toys, telephones, tools, urinals, walls, wheelchairs, whirlpool interiors, and windows. The proposed label indicated that the product can be used on surfaces such as crystal, enamel, glass, glazed ceramic tile, glazed porcelain, laminate, linoleum, marble, Marlite, metal (e.g., brass, chrome, copper, stainless steel, and tin), Parquet, plastic, Plexiglas, sealed granite, and vinyl. Directions on the proposed label provided the following information regarding use of the product:

> As a disinfectant: Pre-clean surfaces prior to use. Hold container upright 6-8 inches from surface. Spray for 2-3 seconds until covered with mist. Let stand for 10 minutes to air dry. Food contact surfaces must be thoroughly rinsed with potable water.
>
> As a soft (fabric) surface sanitizer: Spray a light, even coating on fabric until wet. Fabric must remain wet for 30 seconds. Do not saturate. Let air dry. For difficult odors or heavy fabrics, repeat application.

III AGENCY STANDARDS FOR PROPOSED CLAIMS

Disinfectants for Use on Hard Surfaces in Hospital or Medical Environments (Confirmatory Efficacy Data Requirements)

Under certain circumstances, an applicant is permitted to rely on previously submitted efficacy data to support an application or amendment for registration of a product and to submit only minimal confirmatory efficacy data on his own product to demonstrate his ability to produce an effective formation. This includes a minor formulation change (e.g., a change in an inert ingredient) in a registered product. Confirmatory data must be developed on the applicant's own finished product. For hospital disinfectants, 10 carriers on each of 2 samples representing 2 different product

lots must be tested against *Salmonella enterica* (ATCC 10708; formerly *Salmonella choleraesuis*), *Staphylococcus aureus* (ATCC 6538), and *Pseudomonas aeruginosa* (ATCC 15442) using either the AOAC Use-Dilution Method or the AOAC Germicidal Spray Products as Disinfectants Method. Killing on all carriers is required.

IV SYNOPSIS OF SUBMITTED EFFICACY STUDIES

1. MRID 478175-02 "Hospital Type Disinfectant Efficacy Testing in the Presence of Organic Soil" for Formula Number 1338-015, by Kyle T. Smith. Study conducted at Reckitt Benckiser Inc. Study completion date – June 30, 2009. Study Identification Number 2009-0060.

This study was conducted against *Pseudomonas aeruginosa* (ATCC 15442), *Salmonella enterica* (ATCC 10708), and *Staphylococcus aureus* (ATCC 6538). Two lots (Lot Nos. 1367-064 and 1367-068) of the product, Formula Number 1338-015, 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. Each test substance was ≥ 60 days at the time of testing. Cultures of the challenge microorganisms were prepared in accordance with the published AOAC methods. Horse serum was added to the culture to achieve a 5% organic soil load. Ten (10) glass slide carriers per product lot per microorganism were inoculated with 0.01 mL of a test organism suspension. The inoculum was spread evenly over the carrier surface. The carriers were dried for 40 minutes at 33.6-35.7°C (which is slightly cooler temperature than the 37°C specified in the AOAC method). Each carrier was sprayed with the product for 2-3 seconds from a distance of 6-8 inches from the carrier surface, and allowed to remain wet for 5 minutes at 22.6-23.0°C. Following exposure, individual carriers were transferred to 20 mL of Letheen Broth to neutralize. Subcultures were gently shaken (see protocol) as specified in the AOAC method. All subcultures were incubated for 46 hours at 32.5-37.5°C (which is a slightly shorter time than the 48 hours specified in the AOAC method). Following incubation, the subcultures were examined for the presence or absence of visible growth. Controls included those for inoculum count, dried recovery carrier count, test system verification (i.e., purity and identity), neutralizer efficacy, and sterility.

Note: Protocol amendments were cited.
Note: Some of the incubation durations exceed 70 hours.
Appendix B: Assay repeated on June 2nd, 2009 against 60 carriers to further evaluate this test substance batch against *P. aeruginosa*.

2. MRID 478175-03 "Hospital Type Disinfectant Efficacy Testing in the Presence of Organic Soil" for Formula Number 1338-016, by Kyle T. Smith. Study conducted at Reckitt Benckiser Inc. Study completion date – June 30, 2009. Study Identification Number 2009-0061.

This study was conducted against *Pseudomonas aeruginosa* (ATCC 15442), *Salmonella enterica* (ATCC 10708), and *Staphylococcus aureus* (ATCC 6538). Two lots (Lot Nos. 1367-073 and 1367-076) of the product, Formula Number 1338-016, 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. Each test substance was ≥ 60 days at the time of testing. Cultures of the challenge microorganisms were prepared in accordance with the published AOAC methods. Horse serum was added to the culture to achieve a 5% organic soil load. Ten (10) glass slide carriers per product lot per microorganism were inoculated with 0.01 mL of a test organism suspension. The inoculum was spread evenly over the carrier surface. The carriers were dried for 40 minutes at 33.6-36.9°C (which is slightly cooler temperature than the 37°C specified in the AOAC method). Each carrier was sprayed with the product for 2-3 seconds from a distance of 6-8 inches from the carrier surface, and allowed to remain wet for 5 minutes at 23.2-25.4°C. Following exposure, individual carriers were transferred to 20 mL of Letheen Broth to neutralize. Subcultures were gently shaken (see protocol) as specified in the AOAC method. All subcultures were incubated for 46 hours at 32.5-37.5°C (which is a slightly shorter time than the 48 hours specified in the AOAC method). Following incubation, the subcultures were examined for the presence or absence of visible growth. Controls included those for inoculum count, dried recovery carrier count, test system verification (i.e., purity and identity), neutralizer efficacy, and sterility.

Note: Protocol amendments were cited.
Note: Incubation duration for *S. aureus* is 67 hours.
Appendix C: The result of this testing were concluded to be inconclusive. There were no countable plates (30-300) for the dilutions series for 2 of the 3 Dried Recovery Control counts. Testing against *Salmonella enterica* was re-tested on June 16th, 2009. The results of this testing were concluded to be inconclusive. There were no recovery counts on the Neutralization Inoculum Verification and Inoculum Count Agar plates. Testing against *Pseudomonas aeruginosa* was re-tested on June 2nd, 2009.

3. MRID 478175-04 "Hospital Type Disinfectant Efficacy Testing in the Presence of Organic Soil" for Formula Number 1338-020, by Kyle T. Smith. Study conducted at Reckitt Benckiser Inc. Study completion date – June 30, 2009. Study Identification Number 2009-0062.

This study was conducted against *Pseudomonas aeruginosa* (ATCC 15442), *Salmonella enterica* (ATCC 10708), and *Staphylococcus aureus* (ATCC 6538). Two lots (Lot Nos. 1367-080 and 1367-083) of the product, Formula Number 1338-020, 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. Cultures of the challenge microorganisms were prepared in accordance with the published AOAC methods. Horse serum was added to the culture to achieve a 5% organic soil load. Ten (10) glass slide carriers per product lot per microorganism were inoculated with 0.01 mL of a test organism suspension. The inoculum was spread evenly over the carrier surface. The carriers were dried for 41-42 minutes at 34.2-36.7°C (which is slightly cooler temperature than the 37°C specified in the AOAC method). Each carrier was sprayed with the product for 2-3 seconds from a distance of 6-8 inches from the carrier surface, and allowed to remain wet for 5 minutes at 23.0-24.4°C. Following exposure, individual carriers were transferred to 20 mL of Letheen Broth to neutralize. Subcultures were gently shaken (see protocol) as specified in the AOAC method. All subcultures were incubated for 46 hours at 32.5-37.5°C (which is a

slightly shorter time than the 48 hours specified in the AOAC method). Following incubation, the subcultures were examined for the presence or absence of visible growth. Controls included those for inoculum count, dried recovery carrier count, test system verification (i.e., purity and identity), neutralizer efficacy, and sterility.

Note: Protocol deviations were cited.
Note: Incubation durations >60 hours.
Appendix B: Results were inconclusive. The Dried Recovery Control counts did not meet the acceptance criteria of ≥104 organisms per carrier. Testing against *P. aeruginosa* was repeated on June 4, 2009.

4. MRID 478175-05 "Hospital Type Disinfectant Efficacy Testing in the Presence of Organic Soil" for Formula Number 1178-172, by Ann Marie De Luca. Study conducted at Reckitt Benckiser Inc. Study completion date – June 30, 2009. Study Identification Number 2009-0063.

This study was conducted against *Pseudomonas aeruginosa* (ATCC 15442), *Salmonella enterica* (ATCC 10708), and *Staphylococcus aureus* (ATCC 6538). Two lots (Lot Nos. 1325-046 and 1325-069) of the product, Formula Number 1178-172, 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. Each test substance was ≥ 60 days at the time of testing. Cultures of the challenge microorganisms were prepared in accordance with the published AOAC methods. Horse serum was added to the culture to achieve a 5% organic soil load. Ten (10) glass slide carriers per product lot per microorganism were inoculated with 0.01 mL of a test organism suspension. The inoculum was spread evenly over the carrier surface. The carriers were dried for 40 minutes at 33.6-35.3°C (which is slightly cooler temperature than the 37°C specified in the AOAC method). Each carrier was sprayed with the product for 2-3 seconds from a distance of 6-8 inches from the carrier surface, and allowed to remain wet for 5 minutes at 23.6-24.7°C. Following exposure, individual carriers were transferred to 20 mL of Letheen Broth to neutralize. Subcultures were gently shaken (see protocol) as specified in the AOAC method. All subcultures were incubated for 46 hours at 32.5-37.5°C (which is a slightly shorter time than the 48 hours specified in the AOAC method). Following incubation, the subcultures were examined for the presence or absence of visible growth. Controls included those for inoculum count, dried recovery carrier count, test system verification (i.e., purity and identity), neutralizer efficacy, and sterility.

Note: Protocol deviations/amendments were cited.
Note: Incubation times were > 90 hours.
Note: Data was inconclusive on the May 22, 2009 test date. The test system was shown not to be pure culture. Results were therefore not recorded and testing against *S. enterica* was repeated on June 3, 2009.

5. MRID 478175-06 "Hospital Type Disinfectant Efficacy Testing in the Presence of Organic Soil" for Formula Number 1338-027, by Ann Marie De Luca. Study conducted at Reckitt Benckiser Inc. Study completion date – June 30, 2009. Study Identification Number 2009-0063.

This study was conducted against *Pseudomonas aeruginosa* (ATCC 15442), *Salmonella enterica* (ATCC 10708), and *Staphylococcus aureus* (ATCC 6538). Two lots (Lot Nos. 1325-047 and 1325-045) of the product, Formula Number 1178-172, 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. Each test substance was ≥ 60 days at the time of testing. Cultures of the challenge microorganisms were prepared in accordance with the published AOAC methods. Horse serum was added to the culture to achieve a 5% organic soil load. Ten (10) glass slide carriers per product lot per microorganism were inoculated with 0.01 mL of a test organism suspension. The inoculum was spread evenly over the carrier surface. The carriers were dried for 40-42 minutes at 33.6-36.9°C (which is slightly cooler temperature than the 37°C specified in the AOAC method). Each carrier was sprayed with the product for 2-3 seconds from a distance of 6-8 inches from the carrier surface, and allowed to remain wet for 5 minutes at 23.2-24.1°C. Following exposure, individual carriers were transferred to 20 mL of Letheen Broth to neutralize. Subcultures were gently shaken (see protocol) as specified in the AOAC method. All subcultures were incubated for ~97* hours at 34.1-37.0°C (which is a longer time than the 48 hours specified in the AOAC method). Following incubation, the subcultures were examined for the presence or absence of visible growth. Controls included those for inoculum count, dried recovery carrier count, test system verification (i.e., purity and identity), neutralizer efficacy, and sterility.

Note: Protocol deviations/amendments were cited.
Note: Incubation duration were >97 hours.

Appendix C: The result of this testing were concluded to be inconclusive. There were no countable plates (30-300) for the dilutions series for 2 of the 3 Dried Recovery Control counts. Testing against *Salmonella enterica* was retested on June 16th, 2009.

V RESULTS

MRID Number	Organism	No. Exhibiting Growth/ Total No. Tested		Dried Carrier Count (CFU/ carrier)
		Lot No. 1367-064	**Lot No. 1367-068**	
478175-02	*Staphylococcus aureus*	0/10	0/10	$\geq 1.8 \times 10^6$
	Salmonella enterica	0/10	0/10	$\geq 6.3 \times 10^4$
	Pseudomonas aeruginosa Test Date: May 15, 2009 Test Date: June 2, 2009	 1/10 0/60	 0/10 --	 $\geq 1.33 \times 10^6$ $\geq 1.16 \times 10^6$
		Lot No. 1367-073	**Lot No. 1367-076**	
478175-03	*Staphylococcus aureus* Test Date: May 15, 2009 *Salmonella enterica* Test Date: May 15, 2009 Test Date: June 16, 2009 *Pseudomonas aeruginosa* Test Date: May 15, 2009 Test Date: June 2, 2009	 0/10 0/10 0/10 0/10 0/10	 0/10 0/10 0/10 0/10 0/10	 $\geq 2.80 \times 10^6$ Not Acceptable $\geq 2.26 \times 10^4$ Not Acceptable $\geq 1.82 \times 10^5$
		Lot No. 1367-080	**Lot No. 1367-083**	
478175-04	*Staphylococcus aureus* *Salmonella enterica* *Pseudomonas aeruginosa* Test Date: May 22, 2009 Test Date: June 4, 2009	0/10 0/10 0/10 0/10	0/10 0/10 0/10 0/10	$\geq 1.61 \times 10^6$ $\geq 3.4 \times 10^4$ Not Acceptable $\geq 7.5 \times 10^5$
		Lot No. 1325-046	**Lot No. 1325-069**	
478175-05	*Staphylococcus aureus* *Salmonella enterica* Test Date: May 22, 2009 Test Date: June 3, 2009 *Pseudomonas aeruginosa*	0/10 Inconclusive 0/10 0/10	0/10 Inconclusive 0/10 0/10	$\geq 3.9 \times 10^6$ Inconclusive $\geq 1.34 \times 10^5$ $\geq 4.6 \times 10^5$
		Lot No. 1325-047	**Lot No. 1325-045**	
478175-06	*Staphylococcus aureus* *Salmonella enterica* Test Date: May 22, 2009 Test Date: June 16, 2009 *Pseudomonas aeruginosa*	0/10 0/10 0/10 0/10	0/10 0/10 0/10 0/10	$\geq 2.29 \times 10^6$ Not Acceptable $\geq 4.8 \times 10^4$ $\geq 7.0 \times 10^5$

VI CONCLUSIONS

1. The submitted efficacy studies are acceptable regarding the use of the product, Brace, as a disinfectant with bactericidal claims against *Staphylococcus aureus*, *Salmonella enterica*, and *Pseudomonas aeruginosa* on hard, non-porous surfaces at a contact time of 5 minutes in the presence of organic soil. The registrant needs to provide a rationale for the excessive incubation time periods observed in several of the submitted studies. These incubation periods included time periods >90 hours. Complete killing was observed in the accepted studies for the required number of carriers tested against the required number of product lots. Repeat testing, due to failures, was conducted for *Pseudomonas aeruginosa*. Inconclusive data results were re-evaluated as documented in the review. Neutralization confirmation/neutralizer effectiveness testing for the studies against was acceptable for evaluated studies. Viability controls were positive for growth. Purity controls were reported as pure for evaluated studies. Sterility controls did not show growth for evaluated studies.

VII RECOMMENDATIONS

1. Upon receipt of the rationale explaining extended incubation time, the proposed claims for the product, Brace, as a disinfectant against *Pseudomonas aeruginosa*, *Salmonella enterica*, and *Staphylococcus aureus* on hard, non-porous surfaces for a contact time of 5 minutes in the presence of organic soil will be accepted.

2. Proposed label was not included in data package.