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**FINAL REPORT**

**AOAC Germicidal Spray Test**

***Escherichia coli***

Test Substance

SANITIZEIT RD286

Lot Numbers

05252021LAB1

05252021LAB2

Test Organism

*Escherichia coli*, ATCC 11229

Test Guidelines

EPA (2018) Guidelines 810.2000 and 810.2200 (E)

Author

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Study Completion Date

09/08/21

Performing Laboratory

Microbac Laboratories, Inc.

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Laboratory Project Identification Number

1080-132

Protocol Identification Number

1080.M.21.013

Sponsor

Aseptic Health, LLC

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## RESULTS

Results are presented in Tables 1 – 5. The challenge microorganism was confirmed by Gram Stain and colony morphology using the Viability Control streaks to be consistent with *Escherichia coli*. The Sterility Control exhibited no growth. The Neutralizer Effectiveness Control exhibited growth.

**Table 1**  
**Test Results**

Results Expressed as Number of Tubes Exhibiting Growth / Total Number of Tubes

Microorganism	Lot No.	Results
<i>Escherichia coli</i>	05252021LAB1	0/10
	05252021LAB2	0/10

**Table 2**  
**Neutralizer Effectiveness**

Results Expressed as Growth (+) or No Growth (0) and Colony-Forming Units (CFU)/mL

Microorganism	Lot No.	Tube Result		Average CFU/Tube
		Rep. 1	Rep. 2	
<i>Escherichia coli</i>	05252021LAB1	+	+	88
	05252021LAB2	+	+	

**Table 3**  
**Viability Control**

Results Expressed as Growth (+) or No Growth (0)

Microorganism	Replicate	Result
<i>Escherichia coli</i>	1	+
	2	+

## RESULTS (continued)

**Table 4**  
**Sterility Controls**

Results Expressed as Growth (+) or No Growth (0)

Source	Replicate	Result
Neutralizer + Carrier	1	0

**Table 5**  
**Carrier Counts**  
*Escherichia coli*

Results Expressed as Average Colony Forming Units (CFU) / Carrier and Mean Log<sub>10</sub> Density

Microorganism	Phase	Rep.	CFU/ Carrier	Mean Test CFU/Carrier	Log Density (Log <sub>10</sub> CFU/Carrier)	Mean Test Log <sub>10</sub> Density
<i>Escherichia coli</i>	Pre- Test	1	1.36 x 10 <sup>6</sup>	1.1 x 10 <sup>6</sup>	6.13	6.0
		2	1.64 x 10 <sup>6</sup>		6.21	
		3	1.26 x 10 <sup>6</sup>		6.10	
	Post- Test	1	7.40 x 10 <sup>5</sup>		5.87	
		2	8.00 x 10 <sup>5</sup>		5.90	
		3	5.60 x 10 <sup>5</sup>		5.75	

## PRODUCT EVALUATION CRITERIA

According to the US Environmental Protection Agency, the test substance passes the test if no visible growth is observed in any of the 10 replicates for each lot and all controls must meet the test acceptance criteria.

## CONCLUSIONS

When tested as described, SANITIZEIT RD286, Lot Nos. 05252021LAB1 and 05252021LAB2 passed the AOAC Germicidal Spray Test when *Escherichia coli* was exposed to the test substances for 15 seconds at 20°C and 70% RH.

All of the controls met the criteria established for a valid test. These conclusions are based on observed data.

## REFERENCES

1. U.S. Environmental Protection Agency, Office of Chemical Safety and Pollution Prevention, Product Performance Test Guidelines, OCSPP 810.2000: General Considerations for Testing Public Health Antimicrobial Pesticides, Guidance for Efficacy Testing, February 2018.
2. U.S. Environmental Protection Agency, Office of Chemical Safety and Pollution Prevention, Product Performance Test Guidelines, Frequent Questions (FAQ) for the 2018 Series 810 – Product Performance Guidelines: Antimicrobial Efficacy Test Guidelines, August 2019.
3. *Official Methods of Analysis of the AOAC International*, Chapter 6, Disinfectants, Official Method 961.02, Germicidal Spray Products as Disinfectants, Eighteenth edition, Revised 2013. AOAC International, Suite 500, 481 North Frederick Avenue, Gaithersburg, MD 20877-2417
4. U.S. Environmental Protection Agency, Office of Chemical Safety and Pollution Prevention, Product Performance Test Guidelines, OCSPP 810.2200: Disinfectants for Use on Environmental Surfaces - Guidance for Efficacy Testing, February 2018.
5. U.S. Environmental Protection Agency, Office of Pesticide Programs, Microbiology Laboratory, Environmental Science Center, Ft. Meade, MD, Standard Operating Procedure for Germicidal Spray Products as Disinfectants (GSPT): Testing of *Staphylococcus aureus*, *Pseudomonas aeruginosa*, and *Salmonella enterica*. SOP Number: MB-06-09. Date Revised: 09-29-17.