

## ENVIROFOAM TECHNOLOGIES™ AOAC USE DILUTION AND ALLERGEN TEST REPORTS

### EXECUTIVE SUMMARY

1. EnviroFoam Technologies™ (EFT) is a small, privately held company located in Huntsville, Alabama. In 2000, EFT licensed the Sandia National Laboratories decontamination foam technology and created a product named EasyDECON™. The company is focused on providing an effective indoor air remediation solution for the professional remediator.
2. EasyDECON is a unique surfactant, made of components commonly found under ones house hold sink, similar to laundry detergent or shampoo. The surfactant is engineered to absorb the mold spore. Once the mold spore is exposed to EasyDECON the formula uses a hydrogen peroxide activator to oxidize the spore making it permanently harmless. This mild oxidation process chemically attacks the protein backbone of the mold spore's DNA and chemically alters the very composition of the mold spore.

### LABORATORY TESTING

#### 1. Aspergillus Niger.

- a. Recent testing was conducted using EasyDECON Indoor Air Quality Disinfectant against a common mold type Aspergillus. The test was conducted in accordance with the FDA/EPA approved AOAC Use Dilution Test. The following is a summary of the final test report "Laboratory Project Identification Number 479-111, AOAC Use Dilution Test using Aspergillus Niger":
  - i. Challenge Microorganism Aspergillus Niger, ATCC 16404
  - ii. Test Materials Supplied EasyDECON™ 200-5313 & EasyDECON™ 200-8313
  - iii. Neutralizer Used Sabouraud Dextrose Broth containing 0.25% Yeast Extract, 0.1% Sodium Thioglycollate, 0.6% Sodium Thiosulfate, 0.5% Polysorbate 80 and 0.7% Lecithin.
  - iv. Contact Time 60 Minutes
  - v. Contact Temperature 20±2C
- b. Conclusion:
  - i. Results are presented in Tables 1 and 2 of the Final Report Project Identification Number 479-111, AOAC Use Dilution Test using Aspergillus Niger. The challenge microorganism was confirmed by wet mount to be consistent with *A. niger*. The sterility control exhibited no growth. The viability and neutralizer effectiveness controls exhibited growth. An average of 88 colony-forming units (CFU) of *A. niger* were added to the neutralizer effectiveness controls. Due to the turbid nature of the neutralizer, test control tubes were streaked onto SDA for growth

observations. Test tube streaks exhibited no growth, negative fungistasis. Pre-test inoculum counts were  $2.3 \times 10^8$  CFU/mL.

- ii. All *Aspergillus* mold spores introduced to EasyDECON™ 200-5313 & EasyDECON™ 200-8313 were killed within the allotted contact time. Although 60 minutes contact times were designated by the test protocol, kill rates were exhibited within 5 to 10 minutes. There was no re-growth of *Aspergillus* mold spores after contact with EasyDECON™ 200-5313 or EasyDECON™ 200-8313.

## 2. *Penicillium Niger*.

- a. Recent testing was conducted using EasyDECON Indoor Air Quality Disinfectant against a common mold type *Penicillium*. The test was conducted in accordance with the FDA/EPA approved AOAC Use Dilution Test. The following is a summary of the final test report “Laboratory Project Identification Number 479-112, AOAC Use Dilution Test using *Penicillium Digitatum*”:

- i. Challenge Microorganism      *Penicillium Digitatum* ATCC 16404
- ii. Test Materials Supplied      EasyDECON™ 200-5313 &  
EasyDECON™ 200-8313
- iii. Neutralizer Used              Potato Dextrose Agar containing 0.25%  
Yeast Extract, 0.1% Sodium Thioglycollate,  
0.6% Sodium Thiosulfate, 0.5% Polysorbate  
80 and 0.7% Lecithin.
- vi. Contact Time                    60 Minutes
- vii. Contact Temperature      20±2C

- b. Conclusion:

- ii. Results are presented in Tables 1 and 2 of the Final Report Project Identification Number 479-112, AOAC Use Dilution Test using *Penicillium Digitatum*. The challenge microorganism was confirmed by wet mount to be consistent with *P. digitatum*. The sterility control exhibited no growth. The viability and neutralizer effectiveness controls exhibited growth. An average of 89 colony-forming units (CFU) of *P. digitatum* were added to the neutralizer effectiveness controls. Due to the turbid nature of the neutralizer, test control tubes were streaked onto PDYA for growth observations. Test tube streaks exhibited no growth, negative fungistasis. Pre-test inoculum counts were  $8.2 \times 10^6$  CFU/mL.
- ii. All *Penicillium* mold spores introduced to EasyDECON 200-5313 & EasyDECON 200-8313 were killed within the allotted contact time. Although 60 minutes contact times were designated by the test protocol, kill rates were exhibited within 5 to 10 minutes. There was no re-growth of *Penicillium* mold spores after contact with EasyDECON™ 200-5313 or EasyDECON 200-8313.

### 3. Stachybotrys Chartum

a. Recent testing was conducted using EasyDECON Indoor Air Quality Disinfectant against a common mold type Stachybotrys Chartum. The test was conducted in accordance with the FDA/EPA approved AOAC Use Dilution Test. The following is a summary of the final test report “Laboratory Project Identification Number 479-113, AOAC Use Dilution Test using Stachybotrys Chartum”:

- i. Challenge Microorganism      Stachybotrys Chartum ATCC 9182
- ii. Test Materials Supplied      EasyDECON™ 200-5313 &  
EasyDECON™ 200-8313
- iii. Neutralizer Used              Potato Dextrose Agar containing 0.25%  
Yeast Extract, 0.1% Sodium Thioglycollate,  
0.6% Sodium Thiosulfate, 0.5% Polysorbate  
80 and 0.7% Lecithin.
- iv. Contact Time                    60 Minutes
- v. Contact Temperature      20±2C

b. Conclusion:

- i. Results are presented in Tables 1 and 2 of the Final Report Project Identification Number 479-113, AOAC Use Dilution Test using Stachybotrys Chartum. The challenge microorganism was confirmed by wet mount and morphology to be consistent with *S.chartum*. The sterility control exhibited no growth. The viability and neutralizer effectiveness controls exhibited growth. An average of 4 colony-forming units (CFU) of *S.chartum* were added to the neutralizer effectiveness controls. Due to the turbid nature of the neutralizer, test control tubes were streaked onto Potato Dextrose Agar containing 0.5% Yeast Extract PDYA for growth observations. Test tube streaks exhibited no growth, negative fungistasis. Pre-test inoculum counts were  $2.6 \times 10^6$  CFU/mL
- ii. All Stachybotrys Chartum mold spores introduced to EasyDECON 200-5313 & EasyDECON 200-8313 were killed within the allotted contact time. Although 60 minutes contact times were designated by the test protocol, kill rates were exhibited within 5 to 10 minutes. There was no re-growth of Stachybotrys Chartum mold spores after contact with EasyDECON 200-5313 or EasyDECON 200-8313.

#### 4. Allergen Testing

- a. **Scope** – The purpose of the following test was to determine if the product, EasyDECON 200 Indoor Air Quality Disinfectant, was capable of neutralizing the antigenic portion of two specific molds, *Aspergillus fumigatus* and *Alternaria alternata*, for the *Asp fl* and *Alt a1* antigens.
- b. **Terminology**
  - i. Antigen-A chemical or compound produced by the body in response to exposure to an allergen or material causing an allergic response in a susceptible individual. A chemical or compound that reacts with a monoclonal or polyclonal antibody specifically developed for the detection of the chemical or compound.
  - ii. q.s.-Abbreviation for Quantum Sufficit. The process of adding a solute or reagent to achieve a final desired volume.
- c. **Significance and Use** – The fungi were grown under controlled conditions, the spores harvested, and then treated with the product following the manufacturer s instructions. The subsequent materials were then tested to determine if the antigens, *Asp fl* and *Alt a1* were present in the sample. Control samples were run in parallel with the test samples to insure that the antigenic properties of the organisms were not affected by any other procedures during the test.

If the test product works according to the manufacturer s expectations, then there should be no detectable allergens in the test solutions for either of the fungi. The samples must be adequately neutralized after exposure to the product to stop all activity by the product and prior to the allergen analysis to invalid contact time data with the allergen analysis. The allergen data are reported in micrograms of allergen per milliliter of spore solution. The spore solution was quantified and data are reported in Colony Forming Units per milliliter of spore solution.

- d. **Results:** The following spores concentration were used for the tests:
  - i. *Alternaria alternata*-5ml x (3.2 x 10<sup>8</sup> CFU/ml)=1.6 x 10<sup>9</sup> spores per test
  - ii. *Aspergillus fumigatus*-5ml x (2.2 x 10<sup>9</sup> CFU/ml)=1.1 x 10<sup>10</sup> spores per test
  - iii. The following average concentrations of allergens were found:
    - I. Test Alt a 1 = < 0.8 µg/ml (Below detection Level)
    - II. Test Asp f 1 = < 0.8 µg/ml (Below detection Level)
    - III. Control Alt a 1 = 32.15 µg/ml
    - IV. Control Asp f 1 = 19.23 µg/ml
    - V. 0.8 µg/ml is the detection limit for these allergen analyses, and so therefore no Alt a 1 and Asp f 1 allergens were detected in the test samples using the EMLab ELISA methodology.

## **CONCLUSION**

EasyDECON 200 Indoor Air Quality Disinfectant demonstrated its effectiveness in killing *Aspergillus*, *Penicillium* and *Stachybotrys Chartum* mold spores. It was further shown that the dead mold spores posed no allergic threat to humans or animals.

Additional case studies have demonstrated little or no collateral damage on common substrates using EasyDECON. EasyDECON 200 Indoor Air Quality Disinfectant is a mild skin and mucus membrane irritant. EasyDECON products have proven to be inherently biodegradable.