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DF-200: An Enhanced Sandia Decontamination Formulation for Neutralization of CBW Agents, Biological Pathogens, and Toxic Industrial Chemicals

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Introduction

Sandia National Laboratories has developed, demonstrated and commercialized an aqueous-based decontamination technology (DF-200) that:

- **is effective for neutralizing chemical (CW) and biological (BW) warfare agents, biological pathogens, and many toxic industrial chemicals;**
- **has low toxicity and corrosivity properties;**
- **can work on a number of anticipated material surfaces;**
- **can be incorporated into a number of carriers (foams, liquid sprays, mists, fogs) that satisfy a wide variety of operational objectives.**

DF-200 is considerably less corrosive than bleach or other potential decontaminants. The photographs below compare unprotected carbon steel coupons that have been exposed to de-ionized water, DF-200 (liquid solution), and a 10% bleach solution for 24 hours.

Deionized Water - 24 Hour Exposure

DF-200 - 24 Hour Exposure

10% Bleach - 24 Hour Exposure

Introduction

DF-200 was procured by the U.S. Military and staged in the Middle East to support Operation Iraqi Freedom. An earlier version of the technology (DF-100) was used to remediate portions of the U.S. Capitol Hill office buildings and office buildings in New York City following the anthrax incidents of October 2001.

Sandia National Laboratories has licensed the DF-200 technology to two private companies for production and sales. These companies are Envirofoam Technologies, Inc. (Huntsville, AL USA) which produces EasyDECON™-200 and Modec, Inc. (Denver, CO USA) which produces MDF-200™. Both products are registered with the U.S. Environmental Protection Agency (EPA).

This poster presents results of DF-200 testing versus CW/BW agents conducted at the Edgewood Chemical Biological Center (ECBC; Edgewood, MD) and the Illinois Institute of Technology Research Institute (IITRI; Chicago, IL) and results of cloud knock-down tests against CBW agent simulants conducted at Sandia. It also presents results of DF-200 testing against biological pathogens performed at Kansas State University (KSU; Manhattan, KS) and against mold spores and toxins performed at MICROBIOTEST, Inc. (Sterling, VA), Environmental Microbiology Laboratory, Inc. (San Bruno, CA), and MycoLogics, Inc. (Aurora, CO). Results of DF-200 testing against toxic industrial chemicals (TICs) conducted at Sandia and at the Southwest Research Institute (SWRI; San Antonio, TX) are also presented.

Test Results Against CW Agents

Live CW agent testing with DF-200 was conducted at ECBC for the U.S. Department of Defense (DOD) against VX, HD, and GD. The tests were conducted in stirred reactors with samples being collected at 10 and 60 minutes followed by analysis by gas chromatography (GC). Results are shown below:

Decontaminant	GD		VX		HD	
	10 Min.	60 Min.	10 Min.	60 Min.	10 Min.	60 Min.
DS2	>99.9	>99.9	>99.9	>99.9	>99.9	>99.9
DF-200	>99.9	>99.9	97.8	>99.9	84.8	99.9

Percent decontamination from kinetic tests against CW agents (US DOD stirred reactor tests using EasyDECON™-200 Lot 3829 at 25˚C).

Test Results Against BW Agents

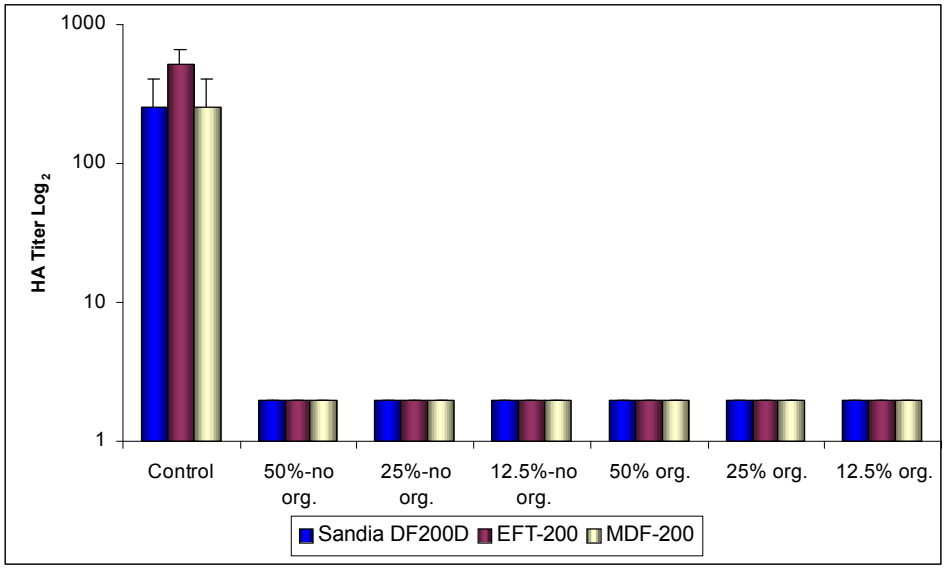
DF-200 is highly effective against BW agents. Kill tests against BW agents were conducted at IITRI using DF-200 against *Bacillus anthracis* spores (Ames-RIID and ANR-1 strains) and *Yersinia pestis.* The tests against these BW agents were conducted in a solution of DF-200 with an initial concentration of ~10^7 CFU/ml and a minimum contact time of 15 minutes.

	B. Anthracis - Ames-RIID		*B. anthracis* – ANR-1		*Y. pestis* – (ATCC 11953)	
	Average CFU/ml	**Log Reduction**	**Average CFU/ml**	**Log Reduction**	**Average CFU/ml**	**Log Reduction**
Control	1.21×10^7	0	6.42×10^7	0	6.42×10^7	0
15 Minute Contact	No Growth	7	No Growth	7	No Growth	7
30 Minute Contact	No Growth	7	No Growth	7	No Growth	7
60 Minute Contact	No Growth	7	No Growth	7	No Growth	7

Results of kill tests conducted against BW agents in DF-200 solution.

Inactivation of Viruses (SARS Surrogate)

Tests have also been conducted at KSU against biological pathogens such as the bovine coronovirus (BCV), a member of antigenic group II of the *Coronviridae* that causes both enteric and respiratory disease in cattle and related species. BCV was used as a surrogate of SARS for studying viral inactivation. Chemical inactivation of BCV was conducted using DF-200 with reduced surfactant and peroxide concentrations (designated DF-200D), and with reduced concentrations of EasyDECON™-200 and MDF-200. Quantitative inactivation was assessed using hemagglutinin activity (HA) following infection into human rectal tumor cells (HRT-18). BCV titers of 5-6 log levels as demonstrated by $TCID_{50}$ (neutralization titration in HRT-18 cells) and HA were completely inactivated after 1 and 3 minute exposure to all formulations.



BCV recovered after treatments with and without organic challenge (bovine feces or compost, 10%) by HA titration following infection in HRT 18 cells.

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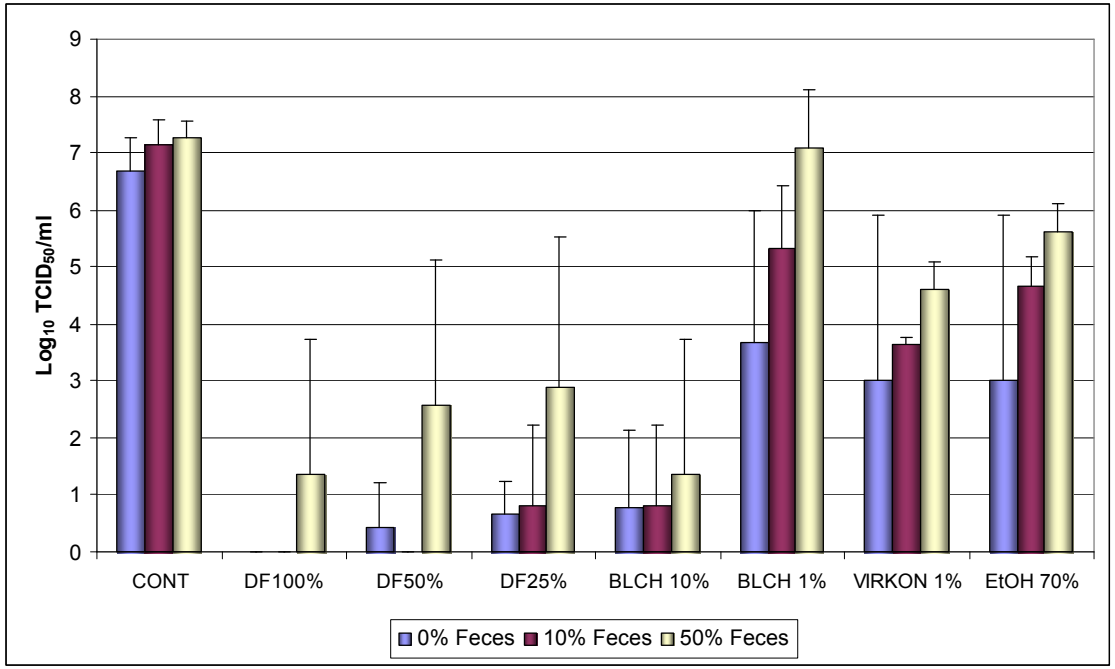
Inactivation of Avian Influenza Viruses

Various test disinfectants were evaluated at KSU for efficacy against isolates of influenza A including a mammalian strain (H1N1, A/WSN/33) and a low pathogenic avian influenza strain (H5N8, isolated from turkey) which are surrogates for the high pathogenic avian influenza strain, H5N1. The disinfectants tested include ethanol (70%), bleach (1, 10%), Virkon® S (potassium peroxymonosulfate, Antec International, Ltd., 1%), and DF-200, diluted by a factor of 2. This formulation was designated as DF-200D and was tested at 25, 50, 100% strength. Infectious titer was determined in $TCID_{50}$ format following treatment with the test disinfectants at various organic loadings.



Infectious titer of mammalian test isolate A/WSN/33 H1N1 determined by TCID50 following 1 minute exposure. CONT, Positive control; DF 100%, DF-200D (100%); DF 50%, DF-200D (50%); DF 25%, DF-200D (25%); BLCH 10%, bleach (10%); BLCH 1%, bleach (1%); VIRKON 1%, Virkon S (1%); EtOH 70%, ethanol (70%)

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Mold Remediation - Efficacy

EasyDECON™-200 (DF-200) was tested against *Stachybotrys* mold spores by MICROBIOTEST, Inc. using the AOAC Use Dilution Test protocol. *S. chartarum*, was dried on stainless steel pennicylinders and exposed to EasyDECON™-200 for 60 minutes at 20±2ºC. Carrier counts were an average of 3.4 x 10^4 CFU/carrier. All of the controls met the criteria established for a valid test. Although a 60 minute contact time was designated by the test protocol, complete kill of the mold spores was exhibited within 5 to 10 minutes.

Microorganism	EasyDECON™-200 (Batch 1)	EasyDECON™-200 (Batch 2)
S. chartarum	0/10	0/10

The capability of EasyDECON™-200 to neutralize the antigenic portion of two specific molds, *Aspergillus fumigatus* and *Alternaria alternata*, for the *Asp f1* and *Alt a1* antigens was tested at Environmental Microbiology Laboratory, Inc. The fungi were grown under controlled conditions, the spores harvested, and then treated with EasyDECON™-200. The subsequent materials were then tested to determine the presence of the antigens. Control samples were run in parallel with the test samples. The following average concentrations of allergens were found: Test *Alt a1* = <0.8 µg/ml, Test *Asp f1*= <0.8 µg/ml, Control *Alt a1* = 32.15 µg/ml, Control *Asp f1* = 19.23 µg/ml. Since 0.8 µg/ml is the detection limit for these allergen analyses, no *Alt a1* and *Asp f1* allergens were considered to be detected in the test samples.

Mold Remediation - Regrowth

A study was conducted by MycoLogics, Inc. to compare the effect of MDF-500™ (a mold remediation formulation based on DF-200 produced by Modec, Inc.) and commercial bleach on oriented strand board (OSB) infected with *Aspergillus versicolor* or *Alternaria alternata*. OSB squares were infected with each fungus. Each square was treated with MDF-500™ or undiluted bleach by spraying to wetness. At various times, the squares were examined for visible growth. Results were: 1) After 3 weeks of treatment, each treatment was efficacious and reduced fungal growth substantially, and 2) After ~60 days of treatment, bleach was inferior to MDF-500™ and regrowth of the fungus was vigorous. MDF-500™ was superior to bleach in preventing regrowth of the fungus.

Control

Bleach

MDF-500™

Cloud Knockdown and Neutralization

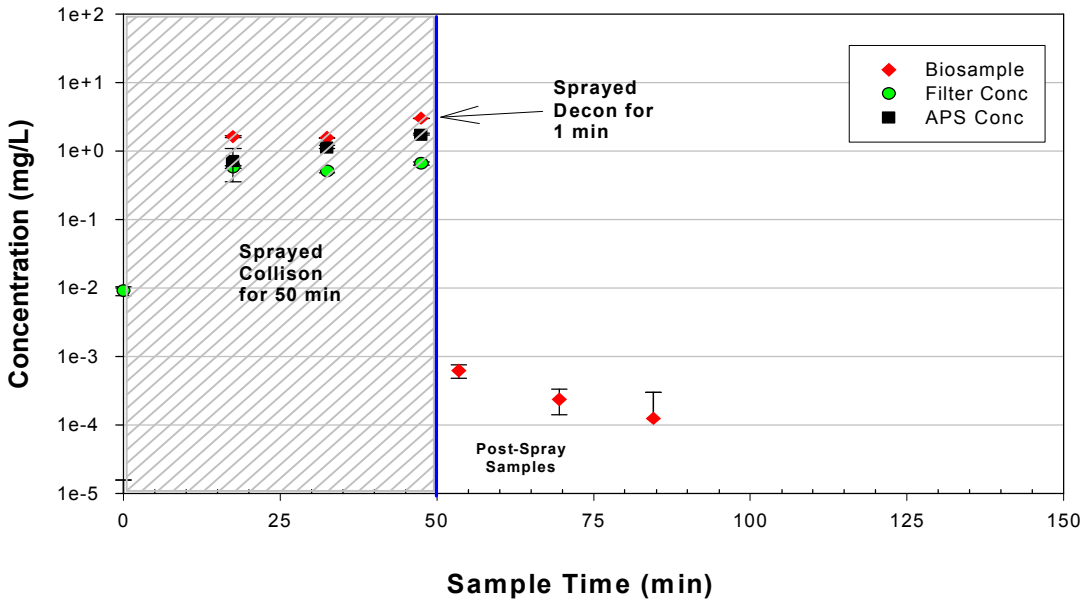
DF-200 can also be used to knock-down and neutralize a cloud of aerosolized CBW agents. An aerosolized cloud of a G-agent simulant (diphenyl chlorophosphate) was released at a concentration of 3.2 g/m^3 in an 8' x 8' x 8' test chamber at Sandia that is shown below. The cloud was well mixed by a series of fans in the test chamber for a period of 50 minutes. At that time, DF-200 was deployed as a spray for one minute through a series of nine ESS (Electrostatic Spray) nozzles that were located in an array at the top of the test chamber as shown in the photograph below (the air pressure supplied to the nozzles was approximately 80 psig).

Aerosol Test Chamber at Sandia National Laboratories

ESS nozzles in the chamber

Cloud Knockdown and Neutralization

The total spray volume deployed was 2 L and the concentration of DF-200 was approximately 138 g/m^3 making the challenge ratio (decontaminant to simulant) approximately 40:1. The G-agent simulant was collected by aerosol sampling and the concentration was determined by GC immediately after the end of the spray period and again at 15 and 30 minutes after the end of the spray period. The results demonstrated a nearly 4 log knock-down and neutralization of the simulant immediately after the spray was stopped. Similar tests conducted with the anthrax simulant demonstrated 5 log knockdown within one minute. These tests indicate the feasibility of using DF-200 to mitigate the release of clouds of CBW agents.



Results of cloud knock-down tests using a mist of DF-200 against a G-agent simulant.

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Results Against TICS

Tests of DF-200 efficacy were conducted against various toxic industrial chemicals (TICs). The challenge ratio was 200:1 for most TICs on a weight/volume basis [except: HCN (1:1); anhydrous ammonia (~2:1); butyl isocyanate (250:1)]. Residual concentration was determined by GC and GC/mass spectroscopy.

TIC	Residual Measured In:	% Decontaminated		
		1 Minute	15 Minutes	60 Minutes
Malathion	Liquid	89	95	Below Detection
Hydrogen Cyanide	Headspace	96	95	96
Sodium Cyanide	Liquid	93	98	>99
Butyl Isocyanate	Liquid	99	Below Detection	Below Detection
Carbon Disulfide	Liquid	>99	>99	Below Detection
Phosgene	Headspace	98	>99	>99
Capsaicin	Liquid	Below Detection	Below Detection	Below Detection
Anhydrous Ammonia	Headspace	>99	>99	>99

Deployment

DF-200 can be deployed as a foam, liquid spray, or mist through both small- and large-scale equipment. Deployment of DF-200 as a foam through the Intelagard Merlin™ (left) and Falcon™ (right) systems are shown below:

DF-200 deployment using the Intelagard Merlin™ System

DF-200 deployment using the Intelagard Falcon™ System

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