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# Validation Study for the Use of Hydrogen Peroxide Vapor as a Decontaminant for Biosafety Cabinets in Accordance with the Requirements of NSF/ANSI 49

ABSA

October 22, 2012

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# Presentation Overview

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- Hydrogen Peroxide Validation Studies and NSF 49 Annex K Overview
- Validation Study Criteria
- Validation Study Methods
- Validation Study Results

# Hydrogen Peroxide Studies and Annex K Overview

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- Studies (2011/2012) executed under direction of NSF 49 Joint Committee on Biosafety Cabinetry Hydrogen Peroxide Task Group and in accordance with NSF 49 Annex K, Protocol for the Validation of Alternate Biosafety Cabinet Decontaminating Methods and Agents
- Given widespread and historical use of hydrogen peroxide as a sterilant in biomedical research and manufacturing environments, significant industry interest in incorporation of a validated procedure for BSC decon utilizing HP vapor as per NSF 49



# Hydrogen Peroxide Studies and Annex K Overview (contd.)

- Three separate studies utilizing three separate hydrogen peroxide systems: STERIS Vaporized Hydrogen Peroxide (dry), Bioquell vapor phase hydrogen peroxide (wet), and AeroClave “energized” hydrogen peroxide (wet aerosolized)
- B&V Testing performed validation study for STERIS VHP 1000 ARD



# NSF 49 Annex K Overview

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- Purpose

To establish a protocol for validating alternative decontamination agents and systems

- Objective

To demonstrate the alternative decon agent/system is at least as effective as formaldehyde gas

- Decontamination Procedure

The methodology for the decontamination procedure during the validation study should be clearly specified prior to the study. The protocol should clearly state the method being validated, i.e. measured concentration/fixed mass



# NSF 49 Annex K Overview (contd.)

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- Cabinet Selection Requirements

The study shall include two different models of each Class II Type BSC (A1,A2,B1 and B2) with a minimum of three trials for each BSC model including trials with no internal BSC blower and incorporating material compatibility testing

# NSF 49 Annex K Overview (contd.)

- Establishing Efficacy, Biological Indicator (BI) Criteria

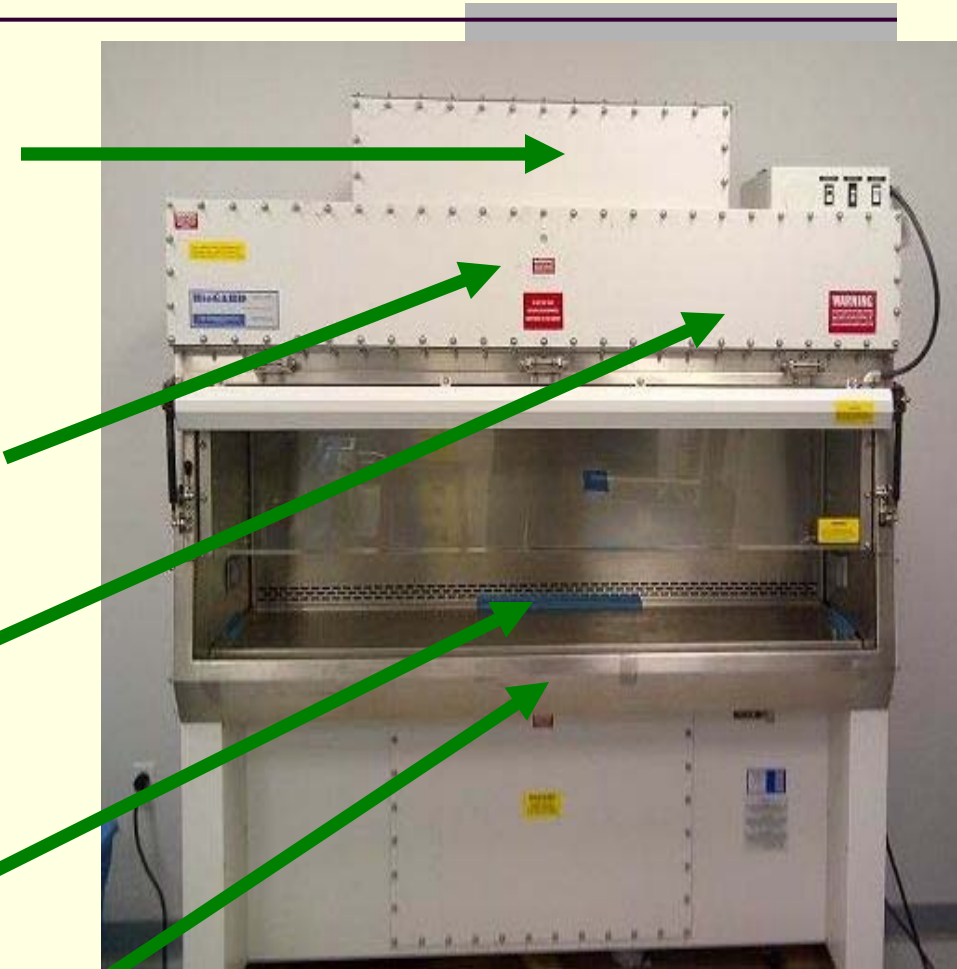
Seven pairs of BIs (A+B sample) consisting of  $\geq 10^6$  log *geobacillus stearothermophilus* placed within BSC

(note: Annex K indicates six locations; HP task group increased to seven)



# NSF 49, Annex K Efficacy, BI Placement

- 3 pairs placed between the filter pleats on the downstream (clean) side of the exhaust HEPA filter ( center and opposite corners)
- 1 pair placed in the contaminated positive pressure plenum
- 1 pair placed between the filter pleats near the center of the upstream (dirty) side of the downflow HEPA filter
- 1 pair placed on the work surface or sidewall
- 1 pair placed beneath the work surface





# NSF 49, Annex K Efficacy, BI Validation Criteria

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(BI) Site for single trial

- Success if either of 2 samples is negative
- Failure if both samples test positive

Single trial

- Pass if all seven (BI) sites pass
- Conditional pass if six (BI) sites pass
- Fail if 2 or more (BI) sites fail

# NSF 49, Annex K Efficacy, BI Validation Criteria (contd.)

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## Cabinet type/Decontamination method validation

- Minimum of 3 trials
- Pass if all 3 trials pass
- Pass if 3 trials have conditional or full passes with failed sites not coinciding
- Trials may be repeated if cause for failure identified

# VHP Validation Experiments

- Measured concentration methodology validated
- Methodology based on established EPA-registered sterilant validation data and B&V pre-testing experiments
- ❖ STERIS VHP EPA registration validation data
  - 250 PPM @ 90 minutes
  - 400 PPM @ 30 minutes



# VHP Validation Experiments (contd.)

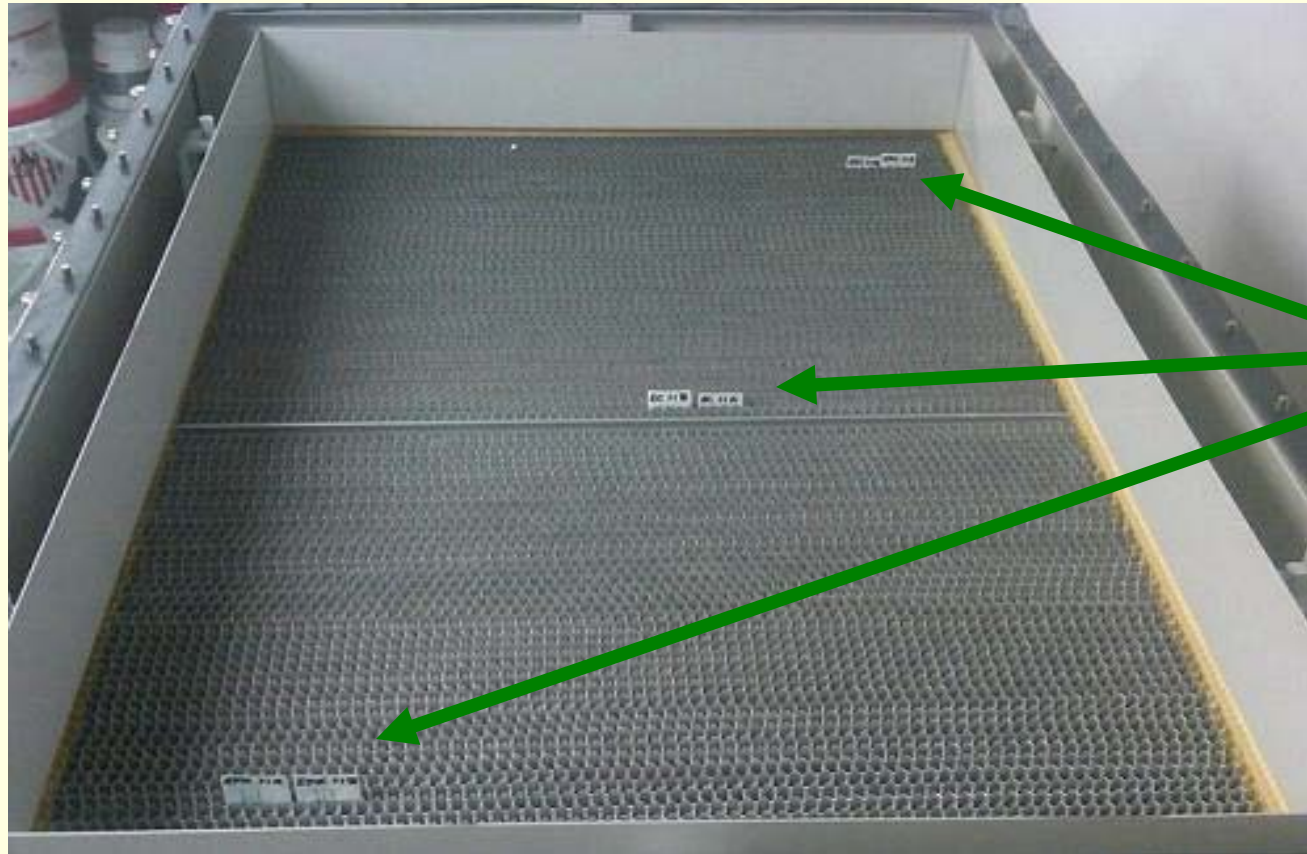
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- Experiments at B&V lab Waltham, MA and NuAire lab Plymouth, MN
- Independent microbiology laboratory utilized for BI testing



# VHP Validation Experiments (contd.)

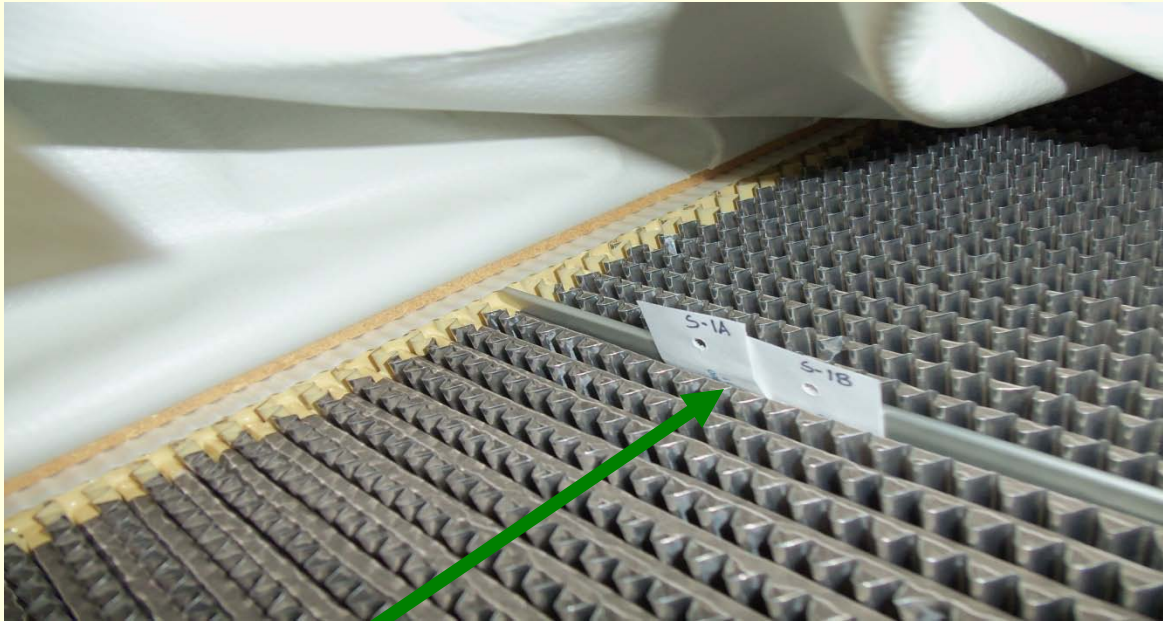
## Cabinet preparation



Exhaust Filter BI  
Placement

# VHP Validation Experiments (contd.)

## Cabinet preparation



Supply  
Filter BI  
Placement



H<sub>2</sub>O<sub>2</sub>  
sensor

# VHP Validation Experiments (contd.)

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Pre-testing experiments performed on multiple BSC models  
20+ runs executed to :

- Establish environmental conditions (Temp/RH)
- Establish target VHP concentration and identify VHP monitoring location
- Optimize vapor distribution by identifying injection and return port locations
- Establish internal blower operation cycle, as applicable
- Establish VHP injection rates
- Establish VHP exposure time

# VHP Validation Experiments (contd.)

## Methodology Conditions:

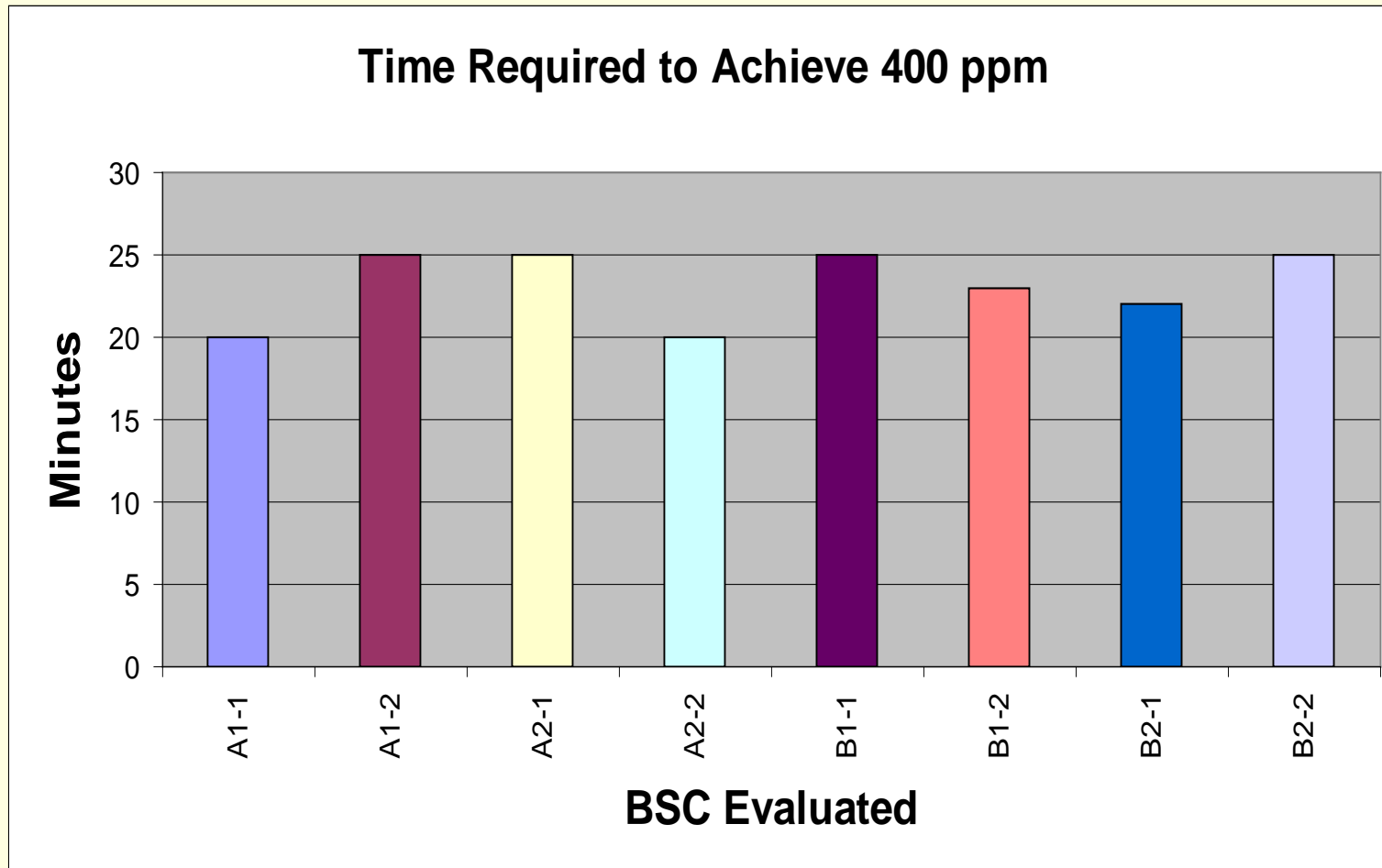
- Start temperature+humidity:  $\geq 60^{\circ}$  F/ $\leq 70\%$  Rh
- VHP injected into the downstream (clean) side of the exhaust HEPA filter with return through BSC access opening for A1,A2 and B1 and via cabinet supply intake on B2.
- VHP injection rate of 3g/minute, 59% H<sub>2</sub>O<sub>2</sub>
- VHP injection to  $\geq 400$  ppm @ center work surface + 30 minutes continuous VHP injection, reversing injection/return pathway halfway through
- Where operational operate the BSC blower at a minimum of one minute for every 15 minutes of VHP injection





# VHP BSC Validation Experiments (contd.)

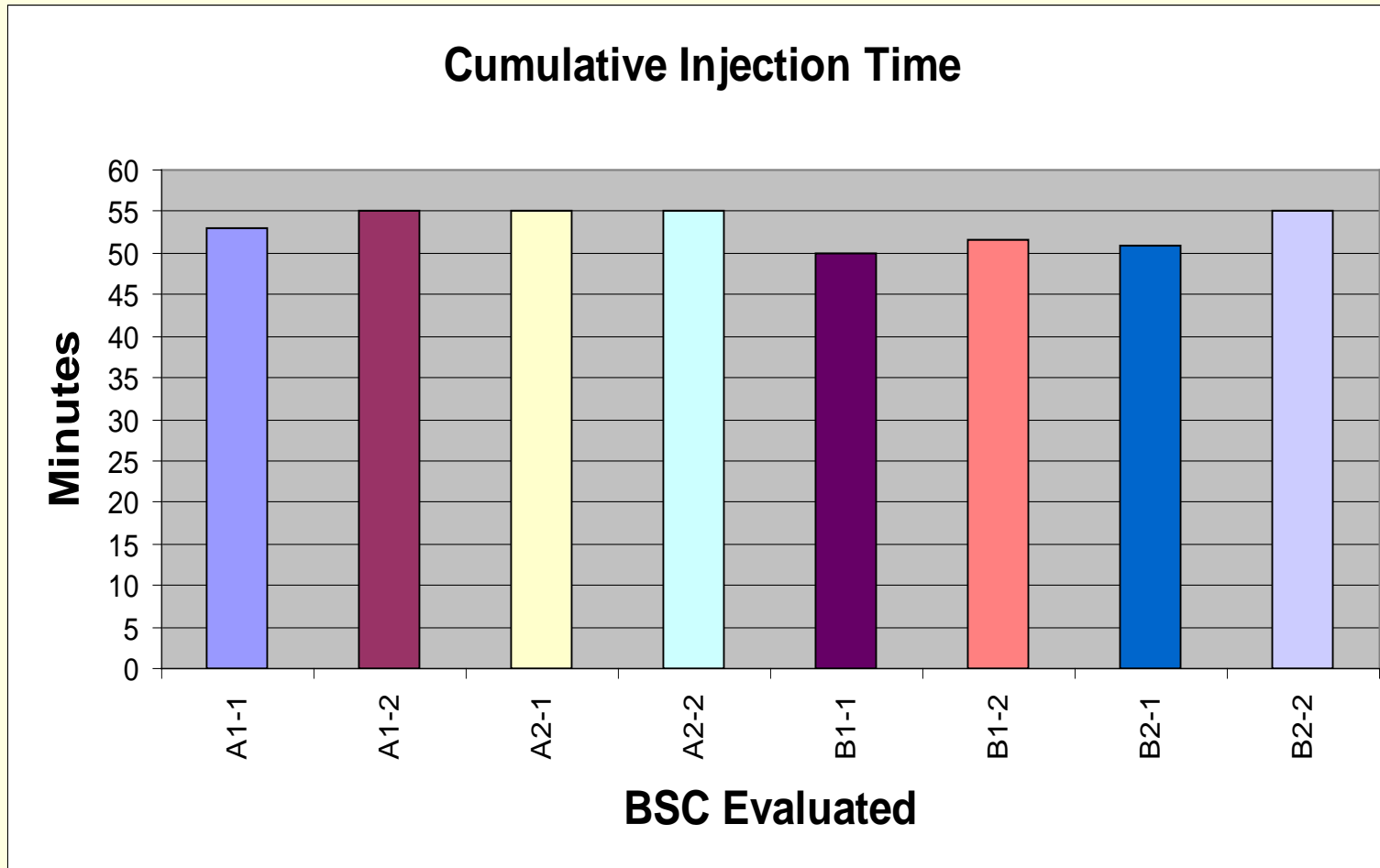
## Study Data



Average Time to reach 400 ppm:  
23 Minutes

# VHP BSC Validation Experiments (contd.)

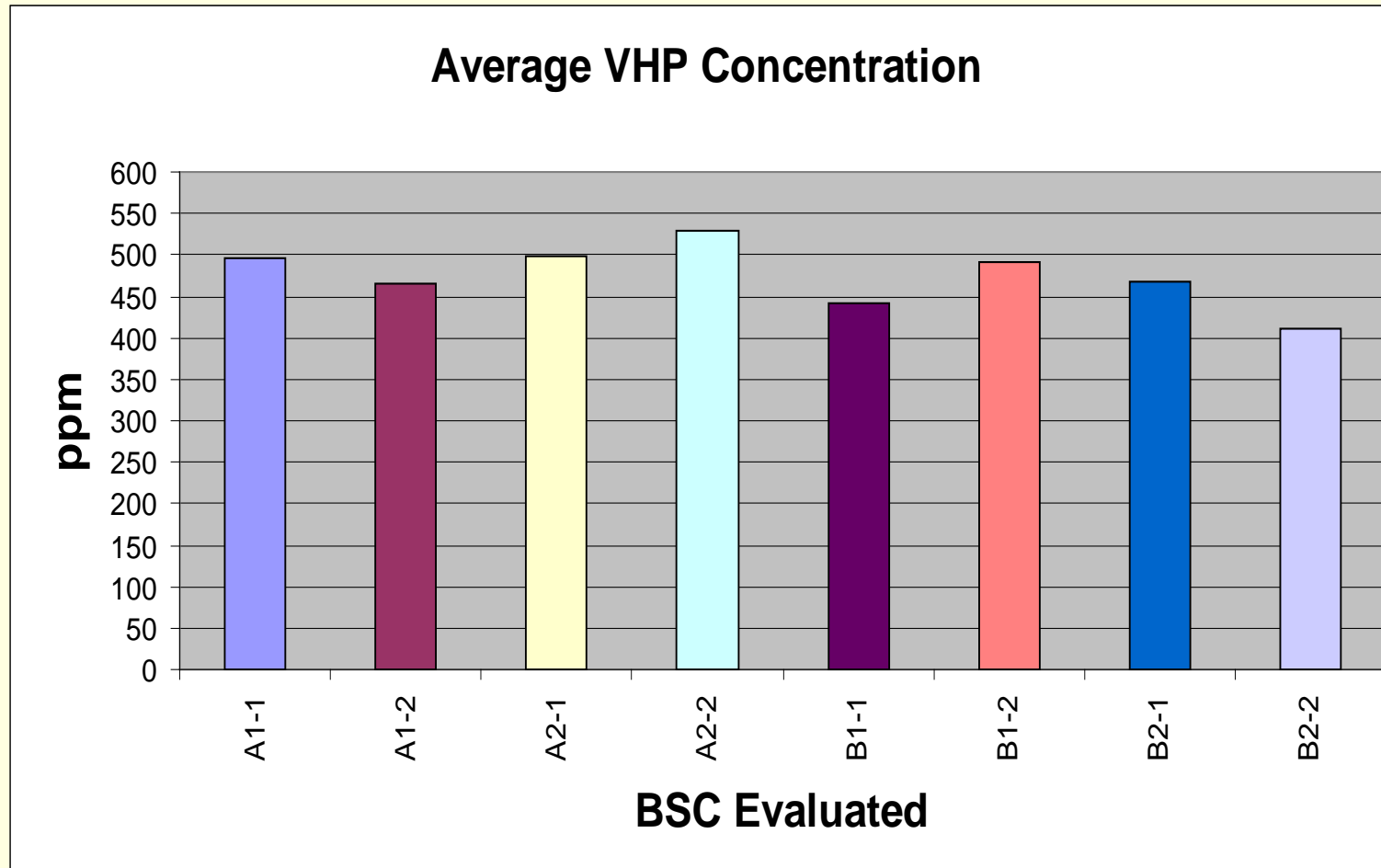
## Study Data



Average Total  
Injection Time :  
53 Minutes

# VHP BSC Validation Experiments (contd.)

## Study Data



Average VHP  
Concentration:  
475 ppm

# VHP BSC Validation Study BI Sites and Trial Results

<u>BSC</u>	<u>Trial #</u>	<u>BI Site Result</u>	<u>Trial Result</u>
A1-1	1	All 14 BIs Negative	Pass
A1-1	2	All 14 BIs Negative	Pass
A1-1	3	All 14 BIs Negative	Pass
A1-2	1	All 14 BIs Negative	Pass
A1-2	2	All 14 BIs Negative	Pass
A1-2	3	All 14 BIs Negative	Pass

# VHP BSC Validation Study BI Sites and Trial Results (contd.)

<u>BSC</u>	<u>Trial #</u>	<u>BI Site Result</u>	<u>Trial Result</u>
A2-1	1	All 14 BI Negative	Pass
A2-1	2	All 14 BI Negative	Pass
A2-1	3	All 14 BI Negative	Pass
A2-2	1	Work Surface BIs A+B Positive	Conditional Pass
A2-2	2	All 14 BI Negative	Pass
A2-2	3	All 14 BI Negative	Pass

# VHP BSC Validation Study BI Sites and Trial Results (contd.)

<u>BSC</u>	<u>Trial #</u>	<u>BI Site Result</u>	<u>Trial Result</u>
B1-1	1	All 14 BI Negative	Pass
B1-1	2	All 14 BI Negative	Pass
B1-1	3	Exhaust HEPA Center BI A Positive	Pass
B1-2	1	All 14 BI Negative	Pass
B1-2	2	All 14 BI Negative	Pass
B1-2	3	Downflow HEPA BI A Positive	Pass

# VHP BSC Validation Study BI Sites and Trial Results (contd.)

<u>BSC</u>	<u>Trial #</u>	<u>BI Site Result</u>	<u>Trial Result</u>
B2-1	1	All 14 BI Negative	Pass
B2-1	2	Exhaust HEPA Front Right A Positive	Pass
B2-1	3	All 14 BI Negative	Pass
B2-2	1	All 14 BI Negative	Pass
B2-2	2	All 14 BI Negative	Pass
B2-2	3	All 14 BI Negative	Pass

# VHP BSC Validation Study, Methodology and Cabinet Results

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- VHP successfully validated for decontamination of Class II type A1, A2, B1 and B2 cabinets including with non-operational BSC blower
- Material compatibility studies showed no adverse impact to BSC materials
- NSF task force to review data for inclusion in NSF 49



# Acknowledgements

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- NuAire, Inc.
- The Baker Company
- Esco Technologies, Inc.
- STERIS Corporation