Effectiveness of Aerosolized Hydrogen Peroxide in Simultaneous Decontamination of a Laboratory and a Biological Safety Cabinet

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with facilities and equipment donated by The BAKER Company and CURIS System

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SUMMARY

The use of aerosolized hydrogen peroxide in laboratory decontamination is becoming more prevalent due to a need for safe surface decontamination. Aerosolized hydrogen peroxide (AHP) is a proven method of spore sterilization within sealed rooms by combining liquid and vapor phases. Achieving similar outcomes within the plenums and filters of a biological safety cabinet while simultaneously treating the laboratory would provide a safe means of complete decontamination. Operational biosafety cabinets filter out 99.99% of aerosols suggesting decontamination may be compromised, thus the efficacy of AHP in simultaneous treatment of a cabinet and laboratory was studied. A 3000-cubic foot lab was sealed and equipped with an AHP generation system and biosafety cabinet. The generator controlled the injection of 7% hydrogen peroxide and dwell phases. Three decontamination times were tested. The cabinet operated at normal, reduced, and no flow conditions. In each test, vapor monitors and a minimum of 30 Geobacillus Stearothermophilus Biological Indicators (BIs) were placed in critical locations in the laboratory and cabinet. A gaseous phase resulted from the cabinet's internally re-circulated airstream that exhausted back into the laboratory. Gaseous concentration depended on the evaporation rate of the lab aerosol and liquid phase collected on the cabinet's filters. Photographs demonstrate a reduced aerosol concentration in the lab when the cabinet was on. While the biosafety cabinet was operational, all BIs were successfully inactivated on 184 carriers signifying spore sterilization. The only exceptions were 9 BIs in the cabinet's internal plenums when it was off. In conclusion, perceived efficacy challenges were proven unfounded. Biosafety cabinet operation, while reducing aerosol concentration, had no significant effect on gaseous concentration and did not compromise decontamination. An operational cabinet was proven advantageous in decontaminating its plenums. AHP is a viable solution to simultaneous decontamination of a laboratory and biosafety cabinet provided the cabinet is operational.

METHODS

Tools and Equipment

- CURIS® Micron Mist fogger
- CURoxide solution
- The Baker Company Biological Safety Cabinet Class II Type A2, Model SG604 6-Foot
- 3000 ft.3 Laboratory
- H2O2 Sensor (ATI)
- Temperature Gauge (AMP Probe)
- Humidity Gauge (ATI)
- Biological Indicators (Mesa Labs)

Figure 1

Figure 2

Testing Parameters

The aerosol generator injected 7% H_2O_2 into the lab space for 15 minutes in each experiment. Experiments 1-3 consisted of a 15 minute primary injection phase (PIP), a 40 minute secondary Pulse phase (SPP) and a 35 minute
dissipation period before BIs were collected. Experiments 4-5 employed similar PIP and SPP with a dissipation period of 0 minutes. Experiment 6 had a PIP of 15 minutes with a reduced SPP time of 20 minutes and no dissipation before BI collection. All experiment times and cycles were recorded (Fig. 3).

BSC Operational Modes: Normal, Reduced, and Off
Primary Injection Phase (PIP): Initial continuous solution injection as fog into the lab area
Secondary Pulse Phase (SPP): Intermittent solution injection into the lab area to replenish H_2O_2 levels
Starting Average Relative Humidity: 31 percent
Starting Average Temperature: 74 degrees Fahrenheit

Figure 3

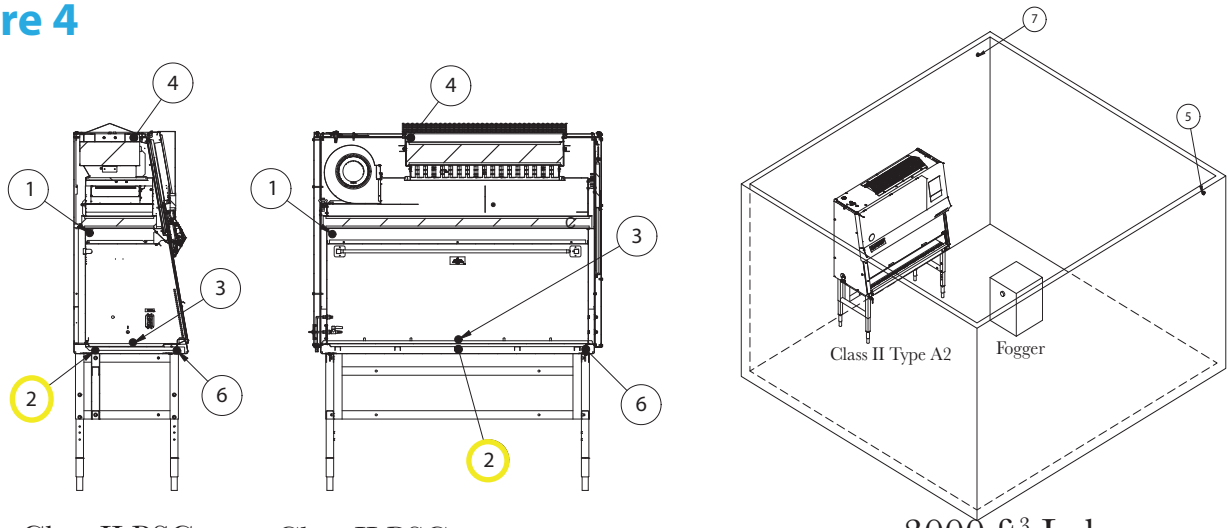
Phase Duration and BSC Modes

	Curis Fogger Phase Duration (minutes)			
Experiment #	Fog	Pulse	Dissipation	Baker BSC Mode
1	15	40	35	Operational
2	15	40	35	Ready Safe
3	15	40	35	Off
4	15	40	0	Operational
5	15	40	0	Operational
6	15	20	0	Operational

Biological Indicators

A total of 193 Geobacillus Stearothermophilus Strain 12980 (1 x 10^6 organisms) were used in 6 different experiments. A combination of 97 stainless steel exposed ribbon coupons (common for aerosolized hydrogen peroxide) and 96 stainless steel discs enclosed in Tyvek/Tyvek pouches (common as markers for gaseous/VHP spore sterilization) were used. These carriers were strategically placed in 2 locations in the lab itself (Fig. 2 A, Fig. 4 B) and also 5 locations (Fig. 2 B, Fig. 4 A) inside the plenums and filter of the cabinet to test efficacy. After each experiment time, BIs were collected and placed in tryptic soy broth media using aseptic techniques in accordance with good laboratory practices. The BIs were incubated for 7 days whereby spore inactivation was determined by turbidity changes (Fig. 2 C) to the soy broth media. Purple indicated inactivated, yellow indicated failure and those results were recorded (Fig. 7 B).

Figure 4



RESULTS

Aerosolization creates a suspended concentration approaching saturation resulting in a simultaneous increase in gaseous concentration. The aerosol then combines to produce a decontaminating effect. The filtration of the biosafety cabinet, when in operation, did not have a negative effect on efficacy and in one test was shown to aid efficacy in the cabinet's internal plenums.

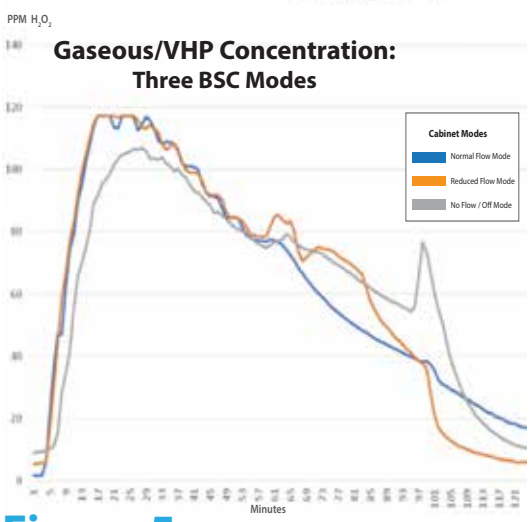


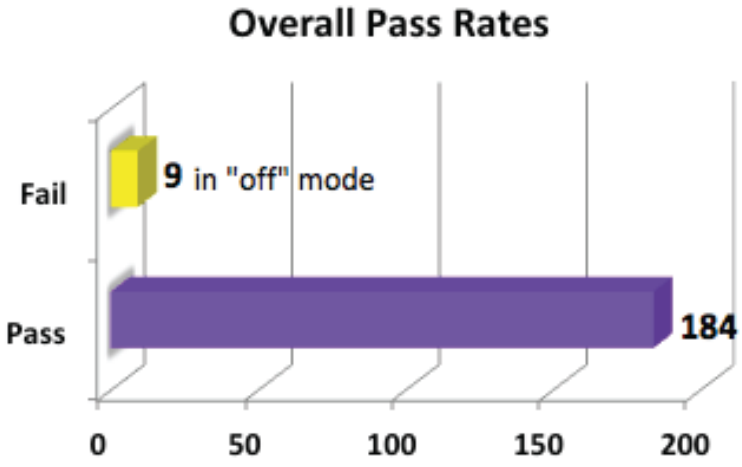
Figure 5

During testing, minimal differences in gaseous concentration were recorded between all three BSC operational modes: Normal flow, Reduced flow, and Off. The monitors also noted where the Pulse phase replenished the H_2O_2 in the peaks following the initial injection phase.

Figure 6

Photographs demonstrate visible fog while cabinet was in Off mode (Fig. 6 A) versus slight visible fog while cabinet was in operational modes (Fig. 6 B).

Figure 7
A



A total of 193 biological indicators were tested in 6 different experiments (Fig. 7 A, B). The results showed inactivation of the spores on 184 carriers of 1.0×10^6 organisms demonstrating a six log reduction. The failures only occurred within the internal plenums of the BSC (Fig. 4 A) when the cabinet was in Off mode (Fig. 3).

Figure 7

B

Test Results

Location	Indicator Type	Duplicate	Experiment # 1	2	3	4	5	6
1	Ribbon Exposed, AHP	1	✔	✔	✖	✔	✔	✔
		2	✔	✔	✔	✔	✔	✔
		3	✔	✔	✔	✔		✔
	Ribbon Sealed in Tyvek/Tyvek Pouch, VHP	1	✔	✔	✖	✔	✔	✔
		2	✔	✔	✖	✔	✔	✔
		3	✔	✔	✔	✔		✔
2	Ribbon Exposed, AHP	1	✔	✔	✖	✔	✔	✔
		2	✔	✔	✖	✔	✔	✔
		3	✔	✔	✖	✔		✔
	Ribbon Sealed in Tyvek/Tyvek Pouch, VHP	1	✔	✔	✖	✔	✔	✔
		2	✔	✔	✖	✔	✔	✔
		3	✔	✔	✖	✔		✔
3	Ribbon Exposed, AHP	1	✔	✔	✔	✔	✔	✔
		2	✔	✔	✔	✔	✔	✔
		3	✔	✔	✔	✔	✔	✔
	Ribbon Sealed in Tyvek/Tyvek Pouch, VHP	1	✔	✔	✔	✔	✔	✔
		2	✔	✔	✔	✔	✔	✔
		3	✔	✔	✔	✔	✔	✔
4	Ribbon Exposed, AHP	1	✔	✔	✔	✔	✔	✔
		2	✔	✔	✔	✔	✔	✔
		3	✔	✔	✔	✔	✔	
	Ribbon Sealed in Tyvek/Tyvek Pouch, VHP	1	✔	✔	✔	✔	✔	✔
		2	✔	✔	✔	✔	✔	✔
		3	✔	✔	✔	✔	✔	
5	Ribbon Exposed, AHP	1	✔	✔	✔	✔	✔	✔
		2	✔	✔	✔	✔	✔	✔
		3	✔	✔	✔	✔	✔	
	Ribbon Sealed in Tyvek/Tyvek Pouch, VHP	1	✔	✔	✔	✔	✔	✔
		2	✔	✔	✔	✔	✔	✔
		3	✔	✔	✔	✔	✔	
6	Ribbon Exposed, AHP	1				✔	✔	✔
		2				✔	✔	✔
		3				✔	✔	✔
	Ribbon Sealed in Tyvek/Tyvek Pouch, VHP	1				✔	✔	✔
		2				✔	✔	✔
		3				✔		
7	Ribbon Exposed, AHP	1						✔
		2						✔
		3						
	Ribbon Sealed in Tyvek/Tyvek Pouch, VHP	1						✔
		2						✔
		3						✔

In conclusion, perceived efficacy challenges were proven unfounded. The biosafety cabinet operation, while reducing aerosol concentration, had no significant effect on gaseous concentration and did not compromise decontamination. In addition, an operational cabinet was proven advantageous in decontaminating its internal plenums. AHP is a viable solution to simultaneous decontamination of a laboratory and a biosafety cabinet provided a cabinet is in operational mode.

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