# Decontamination of Laboratory Animal Research Rooms Using Vaporized Hydrogen Peroxide (VHP®)

### Introduction

Walter Reed Army Institute for Research is a U. S. government laboratory facility located in the Washington D.C. area.



Construction of the state-of-the art facility began in 1994. The institute contains an animal research facility consisting of over 15 animal rooms designed for several different types of laboratory research animals, a BL2 lab and a BL3 lab.

The animal rooms, as well as equipment and animal cages, periodically require decontamination, especially before housing a new group of animals or in the event of suspected contamination. The rooms have been designed and constructed to use a VHP 1000 Biodecontamination System (Figure 1).

Figure 1. VHP 1000 Mobile Biodecontamination System

The focus of this case study is cycle development and decontamination testing of two animal rooms having volumes of 1,875 ft<sup>3</sup> (53 m<sup>3</sup>) and 1,875 ft<sup>3</sup> (53 m<sup>3</sup>) using hydrogen peroxide vapors. The study was completed in April 2001.

## **Room Construction/Room Articles**

Animal rooms of six different sizes exist in this particular block of the building. The smallest room volume is  $900 \text{ ft}^3$  ( $26 \text{ m}^3$ ) while the largest room volume is  $3,025 \text{ ft}^3$  ( $86 \text{ m}^3$ ).

A VHP decontamination cycle has been developed for Room AG104/GW71C (Room C) having a volume of 1,750ft<sup>3</sup> (50 m<sup>3</sup>), and Room AG108/GW71D (Room D) having a volume of 1,875ft<sup>3</sup> (53 m<sup>3</sup>). Both rooms have sealed ceiling tiles and separate external exhausts that can be controlled from outside the room. An automated floor rinse system, which can be turned off and capped during decontamination, has also been constructed. All doors can be sealed with duct tape during decontamination and aeration. Each room is humidity and temperature controlled.

Room C contains a fume hood, floor hood with shaker, incubator, cabinets, refrigerator, freezer and tissue culture incubators, while Room D contains a large stainless steel animal cage that houses eight primates, one laminar flow hood, a plastic floor mat, a mop and a large trash can with lid.

All room construction materials and room articles are compatible with hydrogen peroxide vapor and require no special consideration prior to decontamination.

### **VHP Inlet/Outlet Ports**

All rooms contain at least one pair of inlet/outlet ports to deliver and return hydrogen peroxide vapor (Figure 2). More than one pair of inlet/outlet ports were installed on some of the larger volume



rooms in case more than one VHP 1000 system would be required to provide decontamination.

The hall-side of the VHP ports is constructed of 1.5-inch quick coupler connectors while the room-side of the VHP ports is constructed of three-inch quick coupler connectors.

Figure 2. VHP Inlet/Outlet Ports

The ports are about two inches from each other and are welded to a stainless steel port plate the size of a cinderblock. Port plates are the same size as cinderblocks so that they can easily be added to additional rooms which may require decontamination. All ports are capped when not in use. To promote distribution of the hydrogen peroxide vapor, Room D has a three-inch wide, 12-foot long PVC hose attached to the VHP supply room port. The PVC hose is extended to the center of the room on the floor. Room C has ceiling piping to accommodate the distribution and exchange of hydrogen peroxide vapor (Figure 3).



Figure 3. VHP Inlet/Outlet Ceiling Piping

# **Room Preparation for VHP Decontamination**

Rooms C and D were prepared for decontamination by cleaning the rooms and allowing the rooms to air-dry. The floor drains in both rooms were not sealed with drain covers.

To facilitate hydrogen peroxide vapor distribution, one medium size fan was placed in Room D and one large size fan was placed in Room C.

The VHP ports were uncapped from both inside and outside of the rooms. NB305 VHP Chemical Indicators (CI's) and NA300-P Spordex®-Polyflex VHP Biological Indicators-E5 Bacillus stearothermophilus (BI's) packaged in a \*Tyvek® pouch were hung throughout the rooms. There were 18 CI's and 18 BI's in Room D, and 20 CI's and 20 BI's in Room C. Two unexposed BI's served as positive controls for each room.

The doors to the rooms were closed and cracks around the doors were sealed using duct tape. A switch outside the door turned the external exhaust system off.

# VHP Cycle Parameters

The VHP cycle was developed for Room C and Room D:

#### Dehumidification

Airflow = 20 cfm  $(34 \text{ m}^3/\text{h})$ Time  $= 30 \min$ 

#### Conditioning

Airflow = 20 cfm  $(34 \text{ m}^3/\text{h})$ Time = 45 minInjection Rate = 9.5 g/min.

#### Sterilization

Air Exchange =  $20 \text{ cfm} (34 \text{ m}^3/\text{h})$ Time = 60 minInjection Rate = 8.5 g/min.

#### Aeration

Air Exchange =  $20 \text{ cfm} (34 \text{ m}^3/\text{h})$ Time = 10 min with VHP 1000 followed by overnight aeration using the external exhaust system

The total Vaprox<sup>®</sup> Hydrogen Peroxide Sterilant (H<sub>2</sub>O<sub>2</sub>) used for each room was 937.5 g (840.18 ml). No leak test or pressure control was programmed into the cycle. The rooms were externally aerated overnight. The hoses to the VHP unit were disconnected and capped. The VHP room ports were also capped, releasing the VHP unit to be used to decontaminate additional rooms. The CI's and BI's were collected for analysis.

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Technologies to Prevent Infection and Contamination

### Results

All CI's exhibited uniform color change from blue to gray confirming even distribution of  $H_2O_2$  in the rooms.

All BI's were cultured aseptically in a trypticase soy broth (TSB) media and incubated at 55-60°F (13-16°C) for seven days. All BI's showed no growth, confirming kill. As expected, the two positive controls showed growth.

### Conclusion

A VHP decontamination cycle has been developed for animal rooms having volumes of 1,750 ft<sup>3</sup> (50 m<sup>3</sup>) and 1,875 ft<sup>3</sup> (53 m<sup>3</sup>) at Walter Reed Army Institute for Research. BI and CI results confirmed the effectiveness of VHP 1000 system for room decontamination. Additionally, VHP technology was found to be compatible with room construction materials and room articles.

VHP decontamination cycles are currently being developed for four additional rooms and there are also plans to develop cycles for the remaining rooms.

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