Got Gas? Gas and Vapor Cycle Optimization and Validation Using Biological and Chemical Indicators in a High Containment Facility

Megan Sawyer, Joy Pierzynski, and Megan Trapp

Biosecurity Research Institute, Kansas State University, Manhattan, KS

ABSTRACT

In an industry with many standards and regulations there seems to be a lack of standards on how to optimize or validate gas decontamination cycles in high containment research facilities. While the manufacturers have recommendations on how to set up a space for decontamination, the amount of biological and chemical indicators added to a space during optimization is essentially up to the user. The objective of this poster is to discuss how the Biosecurity Research Institute (BRI) optimizes gas decontamination cycles so that the reader may use this information in their own facility.

OBJECTIVES

- Discuss and give background information about the BRI and its unique research spaces.
- Describe how biological and chemical indicators are placed throughout a space during cycle development.
- Discuss the advantages and disadvantages of each method based on experiences in the BRI facility.

INTRODUCTION

The BRI is located on the Kansas State University campus in Manhattan, KS. It was designed and constructed to meet or exceed BSL-3 and BSL-3Ag standards and provides scientists and collaborators with a safe and secure location to conduct infectious disease and food safety research programs on high-consequence animal, plant, and human pathogens. The BRI has a unique combination of biocontainment areas that are not commonly found together under the same roof. These include: BSL-3 laboratories and preparation laboratories and spaces, BSL-3Ag livestock holding rooms and support areas, a food safety and security space with a food processing area, an insectary, a vivarium, plant and cell culture areas, and walk-in cold rooms and freezers. There is also an interactive training suite for researchers and support staff.

Because of the broad range of high consequence pathogens that are studied at the BRI, it is important to have a reliable way to decontaminate the laboratory and animal spaces between project occupancies or as needed to eliminate the possible transmission of infectious agents to researchers, the general public, and the environment. This is done at the BRI using Vaporized Hydrogen Peroxide (VHP) or Chlorine Dioxide gas (CD gas). For VHP decontamination, the BRI utilizes the Steris VHP 1000-ARD System (Fig. 1) and the Bioquell Z-2 unit (Fig. 2) to decontaminate BSL-3 lab spaces with volumes of 2000 ft^3-5000 ft^3 (57 m^3-141 m^3). Optimization trials have not been conducted at this time in BSL-3Ag spaces with the Bioquell Z-2 unit. In the BSL-3Ag animal holding rooms and necropsy suite, CD gas is generated using the ClorDiSys Minidox-M system (Fig. 3).

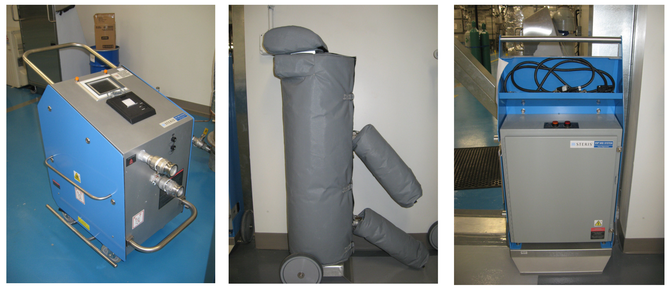

Figure 1. Steris VHP 1000-ARD System with dryer unit and dryer regeneration unit.

Figure 2. Bioquell Z-2 Hydrogen peroxide gas generator.

Figure 3. ClorDiSys Minidox-M Decontamination Unit and external blower set-up.

While the two technologies are vastly different, the way each space is set up for optimization trials is essentially the same. The general standard for biological (BI) and chemical (CI) indicators is to place one of each (BI/CI pair) in the space for every 100 ft^2 of floor space. Some BSL-3 labs at the BRI are only 200 ft^2, which by the standard would require only two BI/CI pairs placed in the room during decontamination. The BRI staff determined it is not sufficient to place such a low number of BI/CI pairs in a space of that size because it doesn't give you an accurate reading of whether the sterilant is reaching all areas of the laboratory for the proper contact time. The BRI staff (Laboratory Coordinators) use architectural drawings of each space to determine the size of the room and room volume. This is also checked by measuring the room with a laser for accuracy. The Laboratory Coordinators then go in and survey each space, noting the placement of key equipment and laboratory furniture to ensure proper placement of biological and chemical indicators. BIs and CIs are placed in pairs throughout the space. The BRI's rule of thumb is to place the pairs in sets of 3 or 4, depending on room design, in the following locations throughout the room: about two inches off the floor, midway between the floor and ceiling, at ceiling height, and as high up as possible in the interstitial space between the false ceiling and true ceiling (if present in laboratory spaces). BI/CI pairs are also placed in any critical equipment that may contain biological agents, such as fridges, freezers, incubators, biosafety cabinets, centrifuges, or other specialized equipment. All doors to cabinets and equipment are opened and BI/CI pairs are placed in a "hard to reach" spots, which is generally the back wall or corners of the cabinet or piece of equipment. A pair is also placed close to the door or on a window, if present, to allow the operator to observe the room during decontamination. If the operator observes a discrepancy in the chemical indicator color, they can easily abort the cycle without having to wait until full cycle completion. Any equipment that does not require decontamination is locked and taped shut. The BI/CI pairs are labelled with their corresponding number (determined by the map) and are hung using easily removable blue painter's tape to fix the BI/CI pairs to their location. The room is prepared for decontamination using the manufacturer's recommended suggestions.

MATERIALS AND METHODS

CD gas and VHP are delivered to each space in a closed-loop system as shown in Figure 4. Both sterilants are delivered to the room via decontamination ports installed in the supply and exhaust duct work (Figure 5A and 5B). Air is circulated in the room with oscillating and/or stationary fans (Figure 6A and 6B). *B. atrophaeus* (CD) and *G. stearothermophilus* (VHP) BIs and sterilant-specific CIs are labelled and hung in pairs throughout the room using blue painter's tape (Figure 7A and 7B). An example of a room schematic used to determine BI/CI placement is shown in Figure 8A and 8B.

After the successful completion of a gas decontamination cycle with VHP or CD gas, the space is aerated until the sterilant is no longer detected using the appropriate monitor. The BI/CI pairs are collected and processed per manufacturer's instructions. BIs are incubated at the manufacturer's recommended temperature for the appropriate length of time and results are recorded. CIs are collected and the results are recorded.

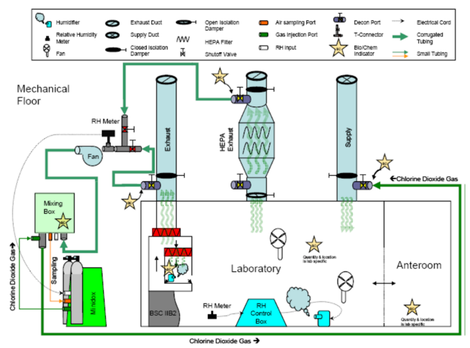

Figure 4. Closed loop system used at BRI (CD gas set-up shown).

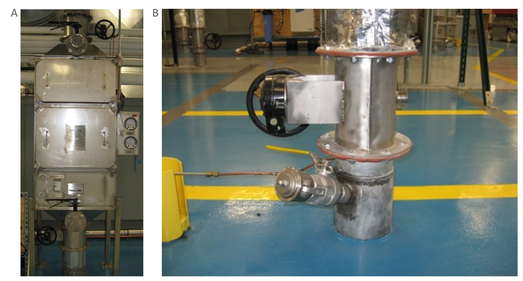

Figure 5. Decontamination ports on room exhaust HEPA filter housing (A) and room supply air (B).

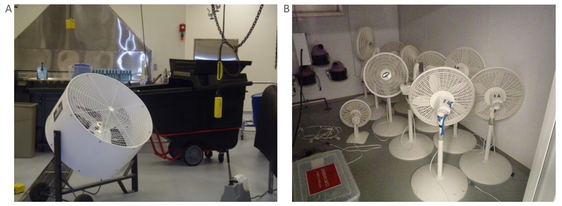

Figure 6. Oscillating fans (A) and stationary fans (B) used at BRI during gas decontamination.

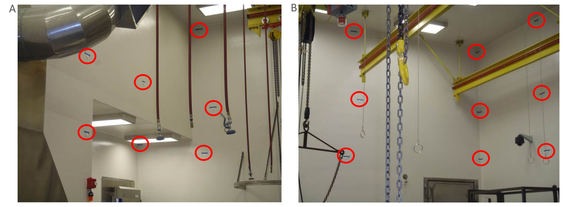

Figure 7. BI and CI pairs hung throughout the room with blue painter's tape.

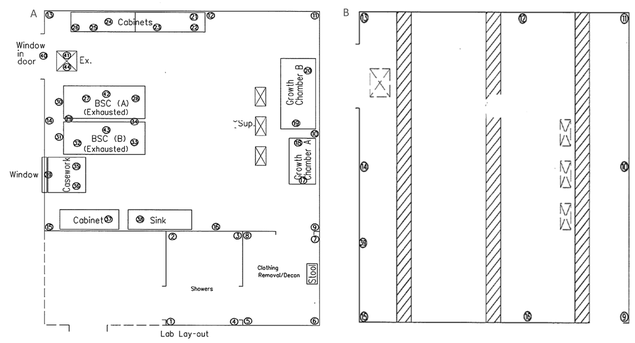

Figure 8. Typical room schematic of BI and CI placement in a BSL-3 lab during cycle optimization. The figure on the left shows BI and CI placement in the room (A). The figure on the right shows BI and CI placement above the false ceiling (B).

RESULTS AND CONCLUSIONS

The BRI's standard for a successful decontamination cycle is a 100% kill rate on all BIs placed in the space. In the event that all BIs come back negative on the first trial run, the process is repeated up to three times to ensure consistent and repeatable results. If the results are maintained for three trial runs, the cycle is considered optimized and validated. All cycle parameters are recorded so subsequent decontamination cycles mirror the same conditions as the optimization cycles. Fan placement and any special notes or procedures are recorded on the map of the space to be used in the future.

A cycle failure is considered any cycle in which one or more BIs comes back positive after the appropriate incubation period. If this result occurs, the Lab Coordinators analyze the results to determine where the positive BIs were located in the room. There are several factors that can influence the results so adjustments to room set-up and cycle parameters must be carefully considered before being changed. If all the BIs in a certain area of the room fail, it may be due to poor air circulation in which the oscillating and stationary fans must be rearranged. If BI failure is random, adjustments are usually made to the cycle parameters (humidification, gassing time, etc). BI placement is never changed, however, as the goal is to decontaminate all "hard to reach" areas. Each new cycle is repeated and parameters are adjusted until a 100% kill rate is obtained. While the CIs are a visual tool to determine if gas or vapor was present in the room during decontamination, they are not used at the BRI as an indicator of proper decontamination because results can vary due to many different factors. If, however, all CI strips maintain their original color and no color change is detected, this could mean there is a problem with the generator and it may need to be serviced.

The BRI goes far above the industry standard to confirm all decontaminated rooms are safe to enter and to eliminate the possible transmission of infectious agents to staff, researchers, or the environment. By using an easily duplicable system, the BRI can decontaminate any space in the building with confidence and reliability.