When is it Time to Clean for Facility Decontamination?


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In association with sterilizing and disinfecting agents for food processing, the term bioburden is exactly that—a burden. Materials which provide a safe-haven for unwanted microbes by covering and protecting them from decontaminating agents are considered bioburdens. Whether that bioburden is dirt, food remnants, or any other organic load, facilities have always been encumbered by the essential, timely, and overall costly task of its removal. Failure to physically rid focused areas of all bioburden prior to the administration of the decontaminating agent will in all likelihood result in inadequate kill. Due to this constraint, a study was performed to gain a better understanding as to "how clean is clean." The goal was to determine how clean a facility needs to be for a gaseous chlorine dioxide (CD) fumigation to be successful.

Gaseous CD is an ideal sterilizer. CD is a true gas under ambient pressure and temperature and, when paired with its small molecular size, can be easily distributed into an area to reach inside nooks and crannies smaller than a micron. Its unique molecular composition can be advantageous over those of bleach (hypochlorous acid), ozone, and hydrogen peroxide by removing 5 electrons opposed to only 2 when reacting with organic loads. Through this process, gaseous CD's reacting power is sustained for longer periods of time, which in turn, makes it more penetrable.

To understand "how clean is clean," varieties of bioburdens and an indicator to denote the penetrability of gaseous CD needed to be established. In regard to the former, powdered milk, powdered baby formula, protein powder, flour, sugar, grains, and general dust/dirt were selected to simulate various bioburdens. This selection was based upon food material commonly found in food processing facilities that require physical removal prior to any form of decontamination. Whereas the latter, a Tyvek-wrapped biological indicator (BI), was selected to validate CD's penetrability through the aforementioned organic loads while still demonstrating a 6-log sporicidal reduction.

Validation

Unlike antiseptics, germicides, sanitizers, or disinfectants, a sterilizer is the only antimicrobial pesticide that is considered by the U.S.-EPA to eliminate all forms of microbial life, including spores. Spore forming bacteria is amongst the most difficult bacteria to kill; therefore this is the reason why it is used to validate sterilization. In almost all cases of facility decontamination, validity is gauged by the results of BIs or through the practice of swabbing. The advantage of BIs is that they contain a

known amount of organisms and those organisms are in the spore form, which is the most difficult to kill. Generally, a BI used to validate the success of a gaseous decontamination consists of a spore forming bacterium inoculated onto a stainless steel disc or paper strip. Otherwise known as a carrier, the disc or strip is enveloped in either Tyvek or glassine. The population, or amount of individual spores that are inoculated onto the carrier, is critical in determining the logarithmic reduction capabilities of that decontaminating agent.

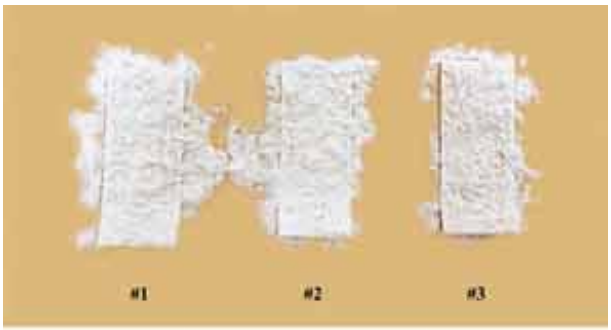
The logarithmic reduction of microorganisms by a decontaminating agent directly reflects its efficacy. Because BIs have a fixed population of microbes, they are an ideal tool to gauge this effectiveness. In regard to gaseous CD, it is easily capable of yielding a 6-log reduction of all forms of microbial life. To better understand this, a 1-log reduction reduces all microbes by 10 times or 90 percent, whereas a 2-log reduction reduces all microbes by 100 times or 99 percent. Therefore, a 6-log reduction reduces all microbes by 1,000,000 times or eliminates 99.9999 percent of all microbes. Of course the population of organisms associated with the BI must be sufficient enough to support its efficacy. For example, a decontaminating agent cannot demonstrate a 6-log reduction by inactivating a BI with a population of less than 1,000,000 microbes.

For this study, a population of 1.3x106 Geobacillus stearothermophilus spores inoculated onto paper strips wrapped in Tyvek were utilized. Tyvek is comprised of flash spun non-directional polyethylene, which makes it not only durable, but also porous. These microscopic pores are too minute for not only the indicator microbes residing inside the Tyvek to escape, but also for any microbes and particulate outside of the Tyvek to penetrate. Gaseous CD molecules and water vapor however, are easily able to maneuver in and out of these pores.

As a result of this combination, this BI is capable of not only validating a 6-log sporicidal reduction, but can also be used as a tool in determining CD's penetrability through organic loads.

Example 1. Organic loading with protein powder.

Example 2. Organic loading with various grains.



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Gaseous CD Decontamination

For this study, a ClorDiSys Minidox-M gaseous CD generator was utilized to automate the five step decontamination process in an effort to reduce human error.

Earlier studies have confirmed that the following cycle ensures a 6-log reduction of spore forming bacteria. Though these studies were conducted under controlled conditions, they indicate the baseline for which to gauge penetration of gaseous CD through the organic loads used in this study.

Upon loading and executing the standard decontamination cycle on the Minidox-M generator, "Pre-condition" is initiated. During this step, the chamber's relative humidity (RH) is raised by a humidifier inside the chamber. Through continuous monitoring, via an RH/temperature probe, the Minidox-M effectively regulates humidification until the predetermined RH set point is reached. Once satisfied, the generator initiates "Condition," whereby the 65 percent RH residing inside the chamber

is maintained and resupplied accordingly for 30 minutes. “Condition” is critical in promoting the susceptibility of bacterial spores to the gaseous CD.

Subsequent to “Condition”, the Minidox-M initiates the CD gas injection step referred to as “Charge.” CD gas is injected, sampled, and monitored in real-time until it reaches its predetermined concentration of 1 milligram/liter (mg/L). Upon reaching its set point, injection ceases and “Exposure” begins. Just as this step’s name implies, all contents located inside the chamber are exposed to the recently injected CD gas. During “Exposure,” humidity and CD concentration are continuously monitored in real-time and respectively supplied to the chamber when either falls under their set points. This phase persists until 720 ppm-hrs (parts per million-hours) has accumulated, or 120 minutes of 1 mg/L contact time has lapsed.

Procedure

Powdered milk, powdered baby formula, protein powder, flour, sugar, grain, and general dust/dirt were selected to simulate organic loads that are commonly seen in food processing facilities. A set of three Geobacillus stearothermophilus populated Tyvek-wrapped BIs consisting of 1.3 x 106 spores were assigned to each of these six varieties. Each set was dusted so that the Tyvek side of each BI was covered not only in its entirety, but also generously enough for the identifying text to no longer be visible.

A 17.0 foot3 polypropylene isolator was utilized as the chamber to conduct this study. The isolator was equipped with various ports and cables for the Minidox-M generator and a carbon scrubber to interface with. Each covered set of three BIs and a single set of three uncovered control BIs were placed inside the isolator along with a small fan, a humidifier, and a probe that monitored both RH and temperature. The RH/temperature probe was connected to an interfacing cable inside the isolator, which was then connected outside to the generator. Similarly, the humidifier was connected to a relay that sat just outside of the isolator, which was then connected to the generator for humidification control. The small fan was plugged into an outlet located inside the isolator and energized to speed up gas distribution. A 0.375-inch CD gas injection tube and a 0.25- inch gas sample tube were then connected on opposite sides of the isolator to avert any false sample readings during the cycle.

The decontamination cycle was started and the Minidox-M successfully raised the chamber’s RH to 65 percent, whereby both the chamber and its contents were held at 65 percent RH for 30 minutes. At the completion of this dwell period, the single set of three control BIs were extracted via BI ports on the isolator to avoid any contact with CD. These BIs were immediately incubated in modified soybean casein digest broth for seven days at 57 degrees Celsius.

Following “Condition,” the Minidox-M stepped into “Charge” and injected CD gas until its concentration reached 1.0 mg/L. Upon satisfying its set point, “Exposure” began and the CD gas was held inside the chamber for exactly 720 ppm-hrs. At the completion of “Exposure” the carbon scrubber was energized and any gas inside the chamber was evacuated within a matter of a few minutes. Once concentrations were reduced to 0.0 mg/L, the Minidox-M prompted for cycle completion, at which time the 18 experimental BIs were retrieved. The six sets of three BIs were then immediately incubated, just as the control BIs removed earlier, in modified soybean casein digest broth for seven days at 57 degrees Celsius.

Results

After the seventh day of incubation, the set of three control BIs resulted in positive growth as expected; indicating that the specific lot of BIs used for this study were viable prior to any testing.

Each set of three BIs covered with powdered milk, powdered baby formula, protein powder, flour, sugar, and grain, of that same lot, indicated no growth. The set of three BIs covered with the general dust/dirt also indicated no growth. This confirms that gaseous CD was able to penetrate all seven of the organic loads and still obtain a 6-log sporicidal reduction.

Conclusion

Bioburdens such as those tested have a notorious nature of providing refuge and sustenance for unwanted microbes. In a perfect world, any bioburden formed in a facility would be immediately and completely removed. However that is never the case, as it is nearly impossible for facilities to sufficiently clean every crack and crevasse on every wall, ceiling, and floor. As such, there is always some degree of buildup of bioburden somewhere in a facility. This buildup of bioburden creates a more difficult location to clean, as most decontamination methods would be impaired by the existence of bioburden.

Findings from this study did not provide a specific answer regarding how much bioburden needs to be removed prior to administering a decontaminating agent, or "how clean is clean." The results do indicate visually however that gaseous CD is powerful enough to penetrate bioburden to some degree and still achieve a 6-log sporicidal reduction. See photo Examples 1 and 2 for an indication of how soiled a surface can be, with a select choice of bioburden, and still be successfully decontaminated with gaseous CD utilizing the standard cycle dosage. Consequently, the physical removal of significant bioburden remains a necessity while complete removal does not. Thus, even though the impracticality of cleaning every crack, crevasse, and cranny still persists, gaseous CD can be an ideal choice for combating bacteria living amongst overlooked bioburden.

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