A Novel Broad-Spectrum CB Decontaminant

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Abstract

Current Chemical/Biological Warfare (CBW) threat decontamination technologies for surface cleanup include pH-neutral bleach, commercial formulations based on per-acetic acid (PAA) and peroxide. Sporicidal chemicals often contain a strong reactive oxidant (0.4-0.6%, w/v), and therefore suffer from logistical burden, storage incompatibility issues, and they are detrimental to the user and the environment. Since the active component is a small fraction, much of transport and storage issues pertain to lugging water in bulk. A new powder-based decontaminant, Bioxy-Enviro (Atomes, Inc., Quebec, Canada), generates active components, PAA/peroxide, at a neutral pH. A 5% solution was tested (30 min contact time) against spores of *Bacillus anthracis* (Sterne). A 2% solution was tested (15 min) against vegetative cells of *Burkholderia thailandensis*. In addition, a 0.5% solution was tested (10 min) against Feline calicivirus (FCV) and Vaccinia virus (VACV). All the disinfection testing was highly efficacious. While 6-7 logs live spores/cells/viral particles were recovered from control sets, <1 log was recovered from test samples. When the powder was thoroughly mixed in DeconGel (at 5% strength), the formulated gel was highly effective as a sporicidal technology.

In preliminary experiments, rapid detoxification of 3 mM of the CW agents GD and A-232 was observed in the presence of 5% solution of Bioxy. The leaving group, fluorine atom was rapidly released in the presence of Bioxy. A powder version of CBW decontaminant, i.e. Bioxy, is highly desirable, as it offers three unique advantages: a) reduced logistical burden of transport and special storage conditions; b) the solution is non-corrosive; and c) it is fully degradable. The decontaminant is totally safe for the user and the environment. Applicability of Bioxy in decontamination of other CBW agents and toxins is of great interest. This broad-spectrum decontaminant is expected to be highly superior for CBW decontamination and serve our warfighter, with minimal collateral damage.

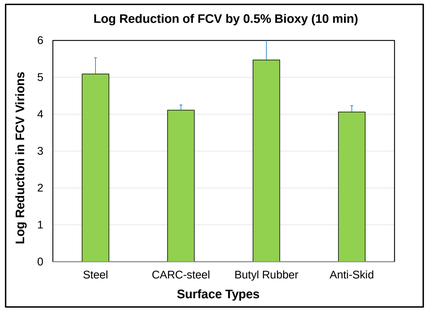
Objectives

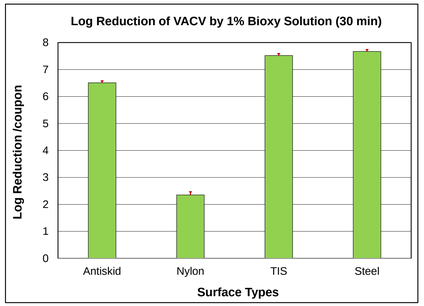
The objective of this effort was to investigate CBW decontamination efficacy and demonstrate broad-spectrum effectiveness of Bioxy as a sporicidal, biocidal, virucidal and CW decontamination potential. Method development and efficacy studies were performed under the auspices of the FTAS program.

Methodology

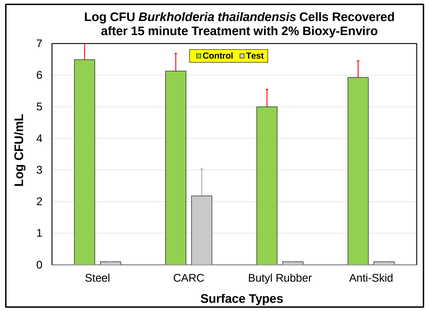
1. *Bacillus anthracis* Sterne Spores were prepared as per Rastogi *et al* (2009), using Lab-Lemko agar plates.
2. Cells of *Burkholderia thailandensis* were grown in Tryptic Soy broth.
3. Feline calicivirus (FCV) is a non-enveloped +ve strand RNA virus, and was grown in CRFK Cells in MEM media supplemented with 10% FBS. FCV exhibits cytopathic effects (CPE).
4. The OECD test method was used for this study, in which an aliquot of 10-µL spore, or cell, or viral suspension was dried on a small coupon for an hour. An aliquot of 50-µL test chemical (5% Bioxy) was layered on top of the inoculum for 10 min. An aliquot of 10-ml appropriate neutralizer (MEM-10% FBS for FCV, PBST+STS for cell and spore) was added to neutralize the active moiety.
5. The coupons were vortexed in neutralizer for one minute, before dilution and plating for cells and spores, or viral infection for FCV.
6. Log Reduction was estimated by subtracting log of spore/cell/viral count recovered from test coupons from the log value of same from control coupons.

Virucidal Efficacy

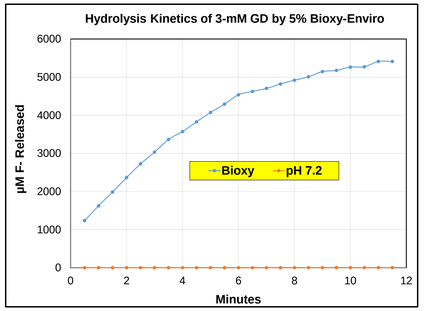




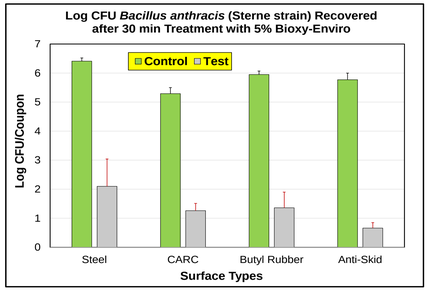
Biocidal Efficacy

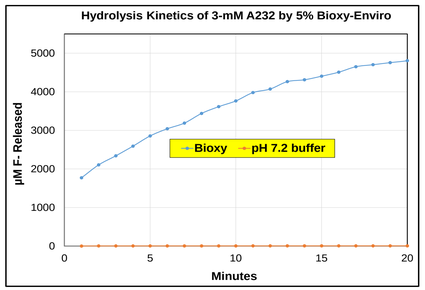


CWA Efficacy



Sporicidal Efficacy





Results and Conclusions

The biocidal, sporicidal, and virucidal effectiveness of Bioxy solution is evident from the log reduction in the number of cells, spores and infective virions, recovered from test coupons after 10-min (for FCV), 15-min (cells of *Burkholderia thailandensis*), and 30-min (spores) contact period. In addition to FCV, the disinfectant was also found to inactivate Vaccinia virus very effectively. The efficacy appears to be material dependent, with CARC being the most challenging surface for decontamination. Clearly, additional optimization is needed to achieve "all-kill" or >6-log reduction in the number of viable test material. A higher efficacy can be achieved by increasing the strength (>5% in case of spores) and/or exposure time.

Interestingly, a 5% solution was found to detoxify 3mM GD and A-232 within minutes. Since, fluorine was directly measured, clearly the P-F bond was susceptible to breakage. It will be of interest to investigate other CW agents. Additional studies are highly desirable to optimize the CW inactivation by Bioxy solution.

Taken together, the results summarized in this presentation clearly demonstrate a broad-level efficacy of Bioxy solution for CB detoxification. Future research should be focused on optimization, stability of the solution under different holding temperatures, and shelf life of powder under extreme conditions. Such a body of data shall lay foundation for a broad-range of applicability of Bioxy as a CB decontaminant on a diverse range of surfaces in the event of a CB release. The non-corrosivity, biodegradability, and significantly reduced logistical burden in transport and storage of this decontaminant potentially makes Bioxy a first choice for decontamination.

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