MITIGATION - SCIENCE AND TECHNOLOGY ADVANCES FOR CHEMICAL AND BIOLOGICAL CONTAMINATION MITIGATION

Decontamination Of Ricin And Abrin With Detection By Quantitative Activity Assay

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Ricin and abrin are highly potent toxalbumins that can be extracted from the castor bean and rosary pea respectively. Both are ribosome inactivating proteins that exert their toxic activity through inhibiting cell protein synthesis by depurination. Ricin and abrin are listed as Tier 1 (highest concern) security sensitive biological agents according to the Australian National Health Security Act 2007. This assessment is made using three factors, namely: terrorist or criminal interest, potential impact, and feasibility of obtaining and disseminating each specific agent.

We are interested in effective techniques that decontaminate abrin and ricin from a variety of substrates and have investigated the efficacy of a range of decontaminants in the laboratory. The following decontaminants were used: commercial bleach diluted to 0.4% hypochlorite concentration, 1.0% aqueous sodium dichloroisocyanurate, a polyethylene glycol monomethyl ether formulation with a nucleophilic monoxime, a peracetic acid solution (~4%) and 0.5 M aqueous sodium hydroxide (for abrin only).

A quantitative activity-based assay, based on a previously reported qualitative assay for ricin, was developed to determine abrin and ricin activity in post decontaminated samples. This type of assay was chosen for two reasons:

he assay determined ricin and abrin activity as a measure for decontamination efficacy

The assay both acted to halt the decontaminating reaction and provide a suitable matrix for analysis through buffer exchanging

The assay presented here quantifies the adenine released from the enzymatic activity of the ribosome inactivating protein (i.e. abrin and ricin). After raising the challenge, a sub sample of the test matrix is buffer exchanged using molecular weight cut off filtering. A mimic messenger deoxyribonucleic acid substrate is added to the buffer exchanged sample, where the mDNA contains a guanine-adenine-guanine-adenine tetra loop active site. Active abrin and ricin A-chain depurinates the substrate releasing adenine. The amount of adenine in the post-reacted sample is then determined using liquid chromatography-mass spectrometry (LC-MS) operating in multiple reaction monitoring (MRM) mode with transitions specific to adenine. This amount can then be compared to the amount of adenine released by abrin and ricin that was not subjected to decontamination to determine decontamination efficacy.

All decontaminants tested were effective in reducing ricin and abrin activity under the conditions used in this evaluation. The developed quantitative activity assay was successful in both halting the decontaminating reaction and detecting abrin and ricin activity in complex post-test matrices. The results of decontaminant testing, along with analytical figures of merit for the quantitative activity assay will be discussed.