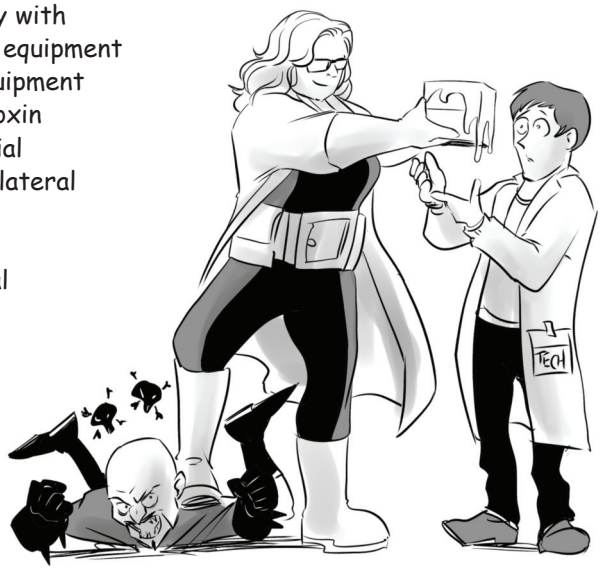
Dr Christina Baxter, of EmergencyResponseTIPS.com and Hazard3.com, offers helpful advice for first responders

Keeping you safe!

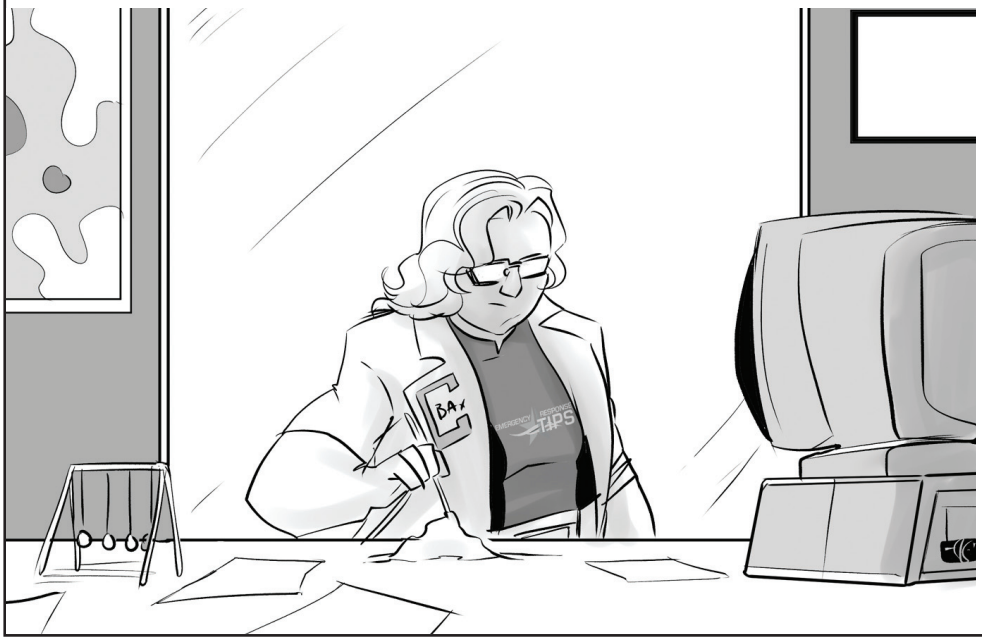
The column aims to provide the hazmat/CBRNE community with operational guidance on the selection and performance of equipment and tactics. This time we are focusing on biodetection equipment for hazmat/CBRNE response. In recent years biological toxin detection has seen rapid growth, however most commercial technologies are still based upon ATP or protein content, lateral flow immunoassays and polymerase chain reaction (PCR).



Where intelligence indicates a material may be a biological threat, it is of the utmost importance to **preserve a sample for laboratory analysis**. All field detection technologies are presumptive for legal purposes, so should be used to minimise operational risk, help inform the selection of sampling sites, and evaluate the effectiveness of decontamination and cleaning, so the site can be safely reoccupied.

Current threat landscape

Recent years have shown just how much havoc a novel bioagent can cause. Besides Covid-19, a variety of biological agents have been seen in areas where they are not considered endemic, with examples including Monkeypox and the polio virus. Biological toxins, and particularly ricin, abrin and nicotine, retain their relevance as regards terrorism, due to factors including ready access to starting products, simple extraction procedures, stability of product in storage, and multiple dissemination methods.



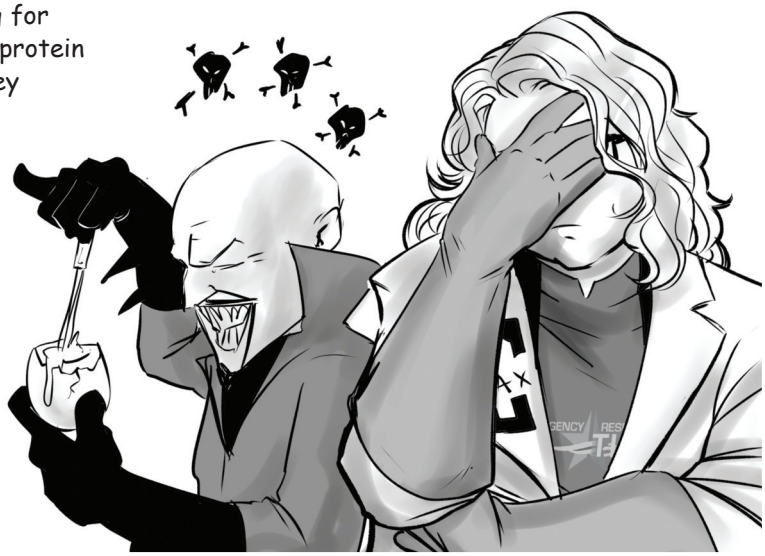
Sampling

Biological agents can be dispersed as powders, liquids or aerosols. Bulk powders and liquids can easily be sampled and tested using available equipment, although care should be taken to avoid aerosolising them. Due to the high inhalation threat associated with biological threats, it is prudent to consider technologies that minimise the potential to generate aerosols, such as vacuum-based sampling.

Sample screening
There are several are commonly used methods for quick sample screening that manage risk levels while awaiting presumptive and field confirmatory test results. Such methods are not definitive, however, and should be used cautionsly: the information is not selective for individual biothreats nor are they sensitive enough to measure at toxicological levels of interest. Operationally, a quick protein or alkaloid test can be used to manage risks while awaiting immunoassay (15 minutes) or PCR (hours) results. These methods should also be considered in the context of the limits of detection and the criteria at which the biological agent is no longer considered a threat to responders or the community.

Adenosine triphosphate (ATP) is present in all living organisms. But while viruses are not alive, ATP is often present in enzymes and host cells required for the virus life cycle. Also, most spore-based materials cannot be detected unless a spore germination step is incorporated prior to testing. Commercially available ATP tests seek out ATP in a sample and correlate the amount present with an index of cleanliness using relative light units (RLUs) on a luminometer. This system is simple and easy to use, but many factors affect operational utility, including the ability to detect biological materials, differentiating between naturally occurring and threat materials, and whether something is bioavailable or denatured. Other materials like disinfectants can also affect results. For these reasons, ATP testing is generally used to determine if a surface has been wiped clean, but should not be used to indicate the presence of a threat material or to determine that the threat material has been destroyed. Note that ATP tests require both swabs with the appropriate buffer solutions and a luminometer to read the results.

Protein tests enable you to identify protein containing materials in minutes, while waiting for more definitive test results. Unfortunately, protein tests, like ATP tests, are non-specific as they are cannot identify a specific threat nor determine whether the biological material is a threat material (ie, protein powders and eggs test positive). Unlike ATP tests, protein tests can differentiate between potentially viable and non-viable threats, as denatured proteins cannot be detected. Therefore, these tests can be used to confirm denaturing of proteins during decontamination, as well as providing rapid identification for the presence of proteins at the beginning of a testing scheme.



Protein tests are often used in conjunction with pH paper to try and rule out potential false positives. Unfortunately, this approach is flawed as biological materials, while often neutral, can also be viable in extreme corrosive (high and low pH) environments, so pH should not be used to rule out the presence of a biological threat material.

Infrared spectroscopy and **Raman spectroscopy** are broadly useful for detecting biological threats in operational settings, especially in relation to toxins. Unfortunately, few instruments have the libraries necessary for detection, and some commercial instruments use generic algorithms to highlight a result as potentially biological due to protein content. These techniques also have the drawback of being unable to identify contents comprising under 10% of the sample matrix. Considerable efforts are underway to evaluate the use of surface enhanced Raman spectroscopy to detect trace levels of biological threats and to validate decontamination, however, there are no solutions currently available.

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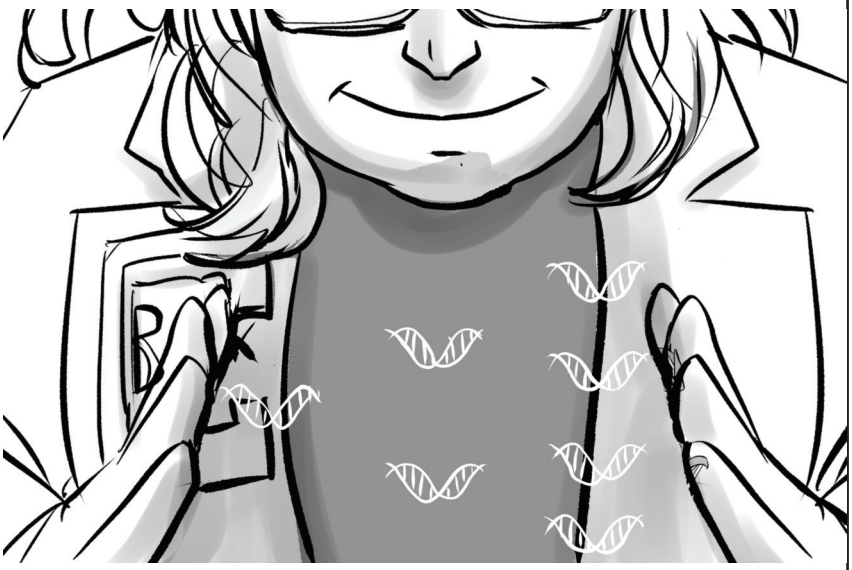
Trigger/cue systems
Intentional or naturally occurring biological releases are often detected retrospectively, as the indicators are less obvious than those in chemical or radiological releases, and the symptoms of exposure generally take days to appear. Continuous monitoring employs trigger technologies to recognise changes in the environmental background using particle sizing, typically looking at particles within the range 0.5 - 30 microns. Cue systems incorporate the trigger system with a second non-specific detection system such as fluorescence or flame spectrophotometry. While the second detection systems do not identify the biological agent, they reduce false positives and negatives by discriminating biological aerosols from other airborne particulates. Both trigger and cue systems activate the gathering of a sample for field presumptive or lab analysis. Most trigger/cue systems are at fixed locations and used to inform the response community about an environmental change that may warrant further investigation.

Presumptive tests
Handheld immunoassays (HHAs) are widely used in field operations. These tests are relatively rapid (15 minutes) and are based on a specific reaction between an antigen and an antibody which amplifies the reaction. Various types of HHAs are commercially available including enzyme-linked immunosorbent assays, lateral flows assays, liquid microsphere based assays, colloidal gold particles and electrochemiluminescence. HHAs typically offer high selectivity and low limits of detection. While our knowledge on these HHAs is growing, the information available regarding performance and cross-sensitivities in operational environments is limited. Newer systems to market incorporate multiplexed immunoassays with simplified sampling and sample processing, further increasing operational utility.

Field-based PCR based assays are the most accurate and sensitive presumptive technology for identifying biological materials. PCR assays have high specificity and low limits of detection (10x lower than HHAs), however, the systems are far more complex to manage and operate. PCR assays amplify small segments of DNA by first denaturing the DNA into two strands, and then duplicating the strands in a thermocycler.



Significant sample preparation is required prior to PCR analysis, so sample costs are higher and assay times longer (30 minutes – hours). Commercial systems are now available which automate sample preparation, minimising the chance for errors and cross contamination. Drawbacks to field PCR testing include the potential for environmental sources to impair results and the inability to distinguish between viable and non-viable biological threats. PCR, however, is the one biodetection technology used in field operations, which can identify threats containing DNA or RNA.
Regardless of the methods you have available, the overarching goal is to minimise the risk to your personnel, the population, and the environment. Each of the tools described above can be used within its operational context to assist with overall risk reduction.

Stay safe!

Images are courtesy of Phil Buckenham
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