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RAPID CB MONITORING BY PYROLYSIS/GC PHOTOIONIZATION MASS SPECTROMETRY

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Abstract

In this paper we report on a rapid chemical/biological (CB) analyzer based on a pyrolysis/gas chromatography (Py/GC) interface to a photoionization mass spectrometer (PI MS) system. The MS system consists of a portable quadrupole ion trap, time-of-flight (QitTof) analyzer with a total system weight of 70 lb. The PI QitTof MS system has previously been shown to be a fast and sensitive identifier of chemical agents. In this work we report on results for biological agent detection. Six surrogates were tested: Bacillus globigii (BG), Erwinia herbicola (EH), E-coli, Ovalbumin, Bovine Serum Albumin, and Bradykinin. The Py/MS spectra are complex but show distinguishing patterns of peaks that can be used to differentiate between the different biological samples. The PI source appears to simplify the spectra relative to electron ionization and chemical ionization. Further improvement in the ability to differentiate biological entities was obtained by including the GC interface. The Py/GC PI MS disperses the total mass spectrum along the GC retention time axis providing 2D separation of signatures. Distinct signature peaks are observed at specific ion masses and retention times and are clearly evident in selected ion monitoring (SIM) traces. We present results that show the differentiation of the above biological samples with the exception of EH and E-coli, which appear essentially identical in their signatures. The recorded SIM signature traces are reproducible from run to run. The particle detection limits ranged from 1 ng to 50 ng depending on sample and signature peaks used. Using standard particle concentrators this suggests the potential to achieve detection limits on the order of 1 ACPLA.

1. Background

a. Introduction

This work describes a successful feasibility demonstration of a new chemical and biological (CB) point-detection system based on pyrolysis gas chromatography (Py/GC) photoionization mass spectrometry (PI MS). The technology has been extensively tested on chemical agents and is showing the potential to discriminate effectively between different biological entities.

A variety of chemical agent sensors, monitors, and point-detection systems have been developed to meet different requirements. Less adequate solutions exist for biological agent detection. The U.S. Army began the Biological Integrated Detection System (BIDS) in 1993. This project employed a battery of instruments, such as a high volume particle sizer, liquid sampler, and flow cytometer in a mobile laboratory mated to a Humvee vehicle. The BIDS vehicle was tested in June 1997 and proved capable of detecting a variety of pathogens in about 5-min, a marked improvement over previous field instruments. Added to this suite of instruments was the Chemical Biological Mass Spectrometer (CBMS). This instrument is an ion trap mass spectrometer capable of biological or chemical detection when connected to an aerosol collector/concentrator or chemical probe, respectively.

The Joint Biological Point Detection System (JBPDS) does not use MS and is currently only capable of BW detection. It employs a fluorescence particle detector for biological aerosols and immunoassay identification. The current version of the JBPDS requires a significant amount of consumables including biological reagents.

b. Competing Methods

Many technologies have been developed for CB detection.[1] Mass spectrometry offers the benefit of molecular identification. For CW detection only MS is a true identifier. Other detectors (e.g., IMS, GC, chemical sensors, etc.) are indicators in that they give a characteristic response indicative of the target compound. For BW detection, there are some viable options to MS detection; however, for continuous real-time monitoring, MS has clear advantages. There are three general technologies for BW detection:

- Mass spectrometry (MS) offers molecular fingerprinting and rapid analysis. Effective MS analysis requires a sampling system that can collect the particles and effectively lyse the microorganisms to release the molecular signature content within.
- DNA sequencing can be used to characterize BWs. As with MS, a sampler and method to release the DNA fragments is needed. DNA sequencing is a slower method than MS and is not well suited to detection of viruses and biotoxins.
- Immunoscreening with antibodies can be used to selectively bind to exposed antigens on BWs. This method has high potential; however, an array of different antibodies is needed to screen for a wide range of BWs. Currently this method is limited to a few biological threats and by false positive rates and high background levels due to antibody binding of non-specific and cross-reacting interferent materials.

Another detector alternative is ion mobility spectrometry (IMS). Though popular for explosives detection in airports and other venues, it is not a very specific detector and is prone to false positives in complex environments. IMS is a very low resolution mass analyzer. MS on the other hand has 10-100x higher resolution, and QitTof MS can achieve an additional dimensionality of analysis by selectively fragmenting target ions to measure for known signature fragments (2D MS). The general consensus is that IMS cannot achieve effective screening for a myriad of threats and maintain a low false positive rate. It is better suited for targeting one or a few threats at a time and even then would not provide definitive molecular identification.

c. MS Methods

Screening at access control points requires; (1) high-throughput, (2) a rapid detection system, and (3) an operational capability to detect CB agents on personnel, baggage and other objects. MS is capable of meeting all three requirements. The PI quadrupole ion trap, time-of-flight (QitTof) MS system offers potentially greater capability compared to competing MS systems. Other competing MS technologies, which are summarized in Table I, are based on the use of conventional ionization sources and mass analyzers. Previous Py/MS analyzers use electron ionization (EI) or chemical ionization (CI) coupled to a conventional ion trap MS (IT MS) analyzer (Bruker and Oak Ridge National Laboratories/Hamilton Sundstand). Another MS approach is based on matrix assisted laser desorption ionization, MALDI/TOF (APL/Johns Hopkins group). This latter method is complex, expensive, and requires significant sample preparation and operator experience. Other potential MS methods include the use of electrospray ionization (ESI). Like MALDI, this method enables analysis of the protein and peptide content of biological entities. However, also like MALDI, the sample preparation is complex and not yet amenable to field applications.

Syagen implemented key performance enhancements across most major subcomponents of a CB MS analyzer (Table I). These are discussed next.

[1] "An Introduction to Biological Agent Detection Equipment for Emergency First Responders," (NIJ Guide 101-00, Dec. 2001).

Table I. Comparison of different MS approaches for CB detection.

Sample Prep	*Ionizer*	*Mass Analyzer*	*Comments*	*Developer*
Pyrolysis	EI, CI	IT MS	>3 min analysis time; requires consumables	Bruker
Pyrolysis	EI, CI	IT MS	2 min analysis time; requires consumables	ORNL/Hamilton Sundstrand
Lysing	MALDI	TOFMS	Requires laser; complicated sample prep; CW not efficiently detected; requires consumables	APL/ Johns Hopkins
Pyrolysis/GC	PI	QitTof MS	65-90 lb with concentrator; no consumables needed	Syagen

2. Methodology

a. Portable QitTof MS

In this work we employed our novel QitTof MS as it provides powerful performance in a compact package. QitTof MS is a hybrid mass spectrometer[2] that combines the best features of an ion trap and a TOF system. Compared to a conventional ion trap MS (IT MS) it has the following advantages:

- Higher repetition rate (60 Hz vs. 3-10 Hz) because the total collection of ions are pulsed into the TOFMS for mass analysis rather than sequentially scanned out of the trap as is the case for IT MS.
- Higher dynamic range because (i) space-charge repulsion is much less a detriment for QitTof MS compared to IT MS and (ii) QitTof MS operates at about 10x higher repetition rate thereby processing more ions per unit time than IT MS. These two factors account for a theoretical improvement in dynamic range of about 100x.
- Lower power need because RF amplitude doesn't have to be scanned to high values

Compared to orthogonal TOFMS, QitTof MS has the following advantages:

- Capability to perform MS/MS (and MS^n through software upgrade)
- Higher dynamic range because of use of a digitizer rather than a counter for data acquisition

Syagen has achieved the highest levels of performance with regard to sensitivity, specificity, speed, and size of any detection system currently available for monitoring CW and explosives compounds.[3,4] Syagen has adapted this technology for the detection of BW under an Army/SBCCOM contract. The key subassemblies of this system are a detector consisting of a compact mass spectrometer and a high-volume vapor/particle collection system. The MS subassembly is shown in Figure 1. Despite the sophisticated nature of the instrument, the total parts count is relatively low (Figure 1B) and uses small-size pumps.

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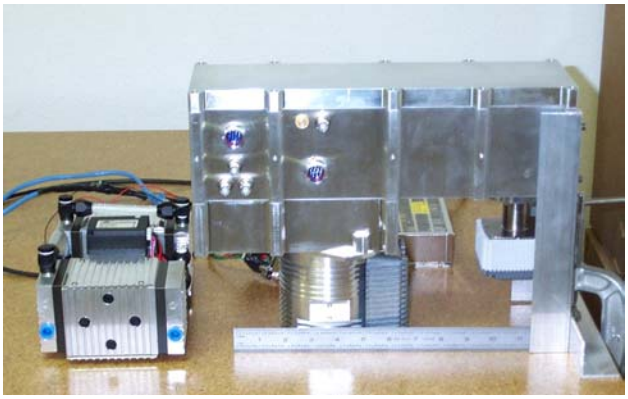
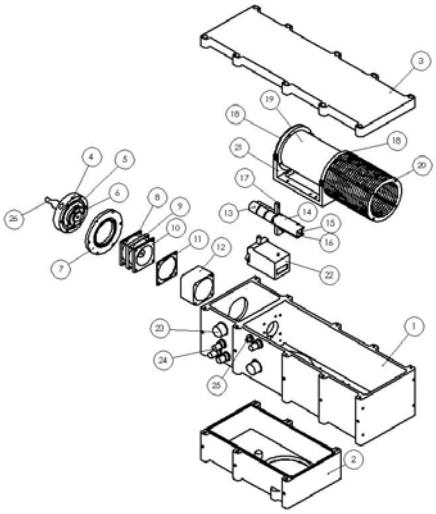
[2] S. M. Michael, B. M. Chien, and D. M. Lubman, *Anal. Chem*. **65**, 2614 (1993); S. M. Michael, M. Chien, and D. M. Lubman, *Rev. Sci. Instrum*. **63**, 4277 (1992).

[3] J. A. Syage, B. J. Nies, M. D. Evans, and K. A. Hanold, "Field-Portable, High-Speed GC/TOFMS," *J. Am. Soc. Mass Spectrom*. **12**, 648 (2001).

[4] J. A. Syage, M. A. Hanning-Lee, and K. A. Hanold, "A Man-Portable, Photoionization, Time-of-Flight Mass Spectrometer," *Field Anal. Chem. & Tech.*, **4**, 204-215 (2000).



(A)



(B)

Figure 1. (A) Picture of QitTof MS detector and pumps, (B) and exploded view of internal parts. The detector measures 14x5x8 in.

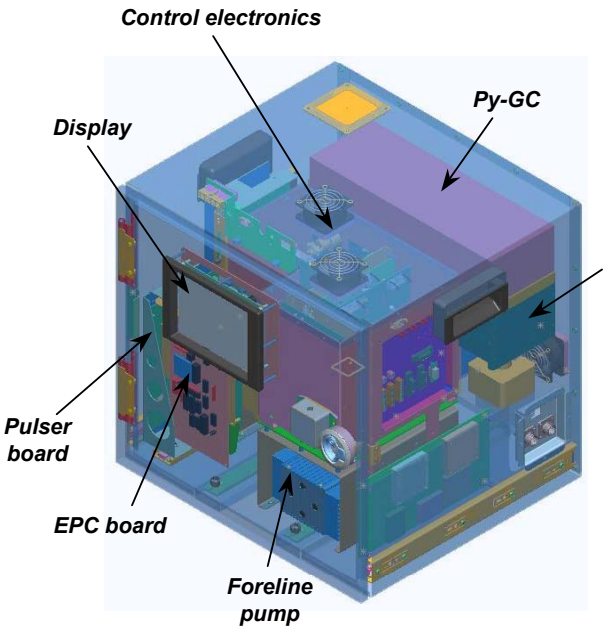


Figure 2. Prototype of CB detector delivered to Army/SBCCOM.

The current prototype detector has been miniaturized to about 65-lb system for CB defense. The Army application is for battlefield air sampling for early warning of a toxic release. The instrument can serve as a core detector for other sampling applications such as for screening of concealed CB by detecting surface contamination. The delivered system to the Army is pictured in Figure 2. This feasibility system demonstrated capability as a BIDS replacement in terms of dimensions (3 cu ft), front door access, and totally self-contained analyzer subassemblies (e.g., pumps, microprocessor, etc.). With further engineering and better form factoring, the analyzer could be reduced to about 50 lb and 2 cu ft without reduction in performance.

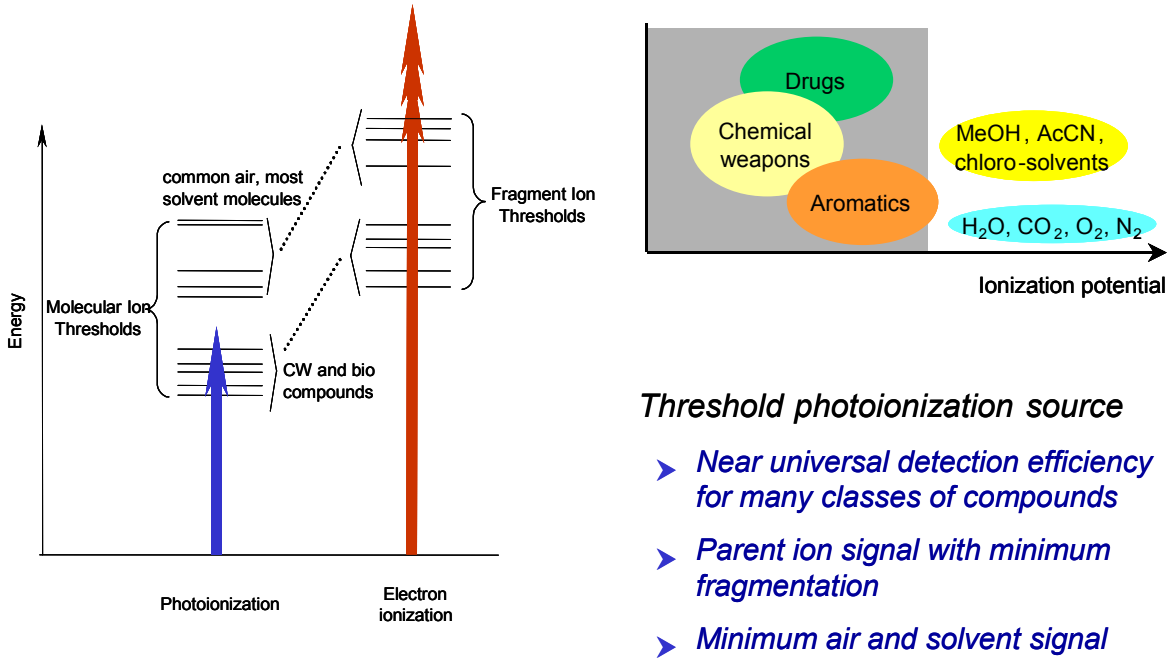


Figure 3. Energy diagram showing PI process vs. EI process.

b. Photoionization

Photoionization (PI) is a key enabling technology that provides a clear differentiation and a major advantage for CB detection compared to EI and CI methods. PI is being adopted rapidly into the commercial MS market and Syagen is providing the PI technology to Agilent, Bruker, Thermo Electron, Waters/Micromass, and Shimadzu for their line of MS instruments. Syagen has pioneered PI developments both experimentally and commercially and is generating the key knowledge to optimize its use for a wide range of applications.[5,6]

Figure 3 illustrates the attributes of PI that provide key benefits for analysis of complex mixtures. [7,8] PI is a threshold ionization process that exceeds the ionization potentials of nearly all relevant compounds, yet typically does not energize the ions to the point of dissociation. This leads to very clean parent ion mass spectra that are very easy to analyze, an important property when analyzing complex mixtures. Furthermore, small molecules such as all common air constituents and most common solvents have high ionization potentials and are not ionized by PI. This enables trace levels of molecules to be detected without interference from the abundant air. PI enables the proverbial "needle" to be seen by making the "haystack" invisible.

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[5] K. A. Hanold, S. M. Fischer, P. Cormia, C. E. Miller, and J. A. Syage, "Atmospheric Pressure Photoionization (APPI): I. General Properties for LC/MS," Anal Chem. In print (2004).

[6] J. A. Syage, K. A. Hanold, T. C. Lynn, J. A. Horner, and R. A. Thakur, "Atmospheric Pressure Photoionization (APPI): II. Dual Source Ionization," J. Chromatog. A., submitted.

[7] J. A. Syage, M. D. Evans, and K. A. Hanold, "Photoionization Mass Spectrometry", *Amer. Lab* **32**, 24 (2000).

[8] J. A. Syage and M. D. Evans, " Photoionization Mass Spectrometry - A Powerful New Tool for Drug Discovery," *Spectroscopy* **16**, 14 (2001).

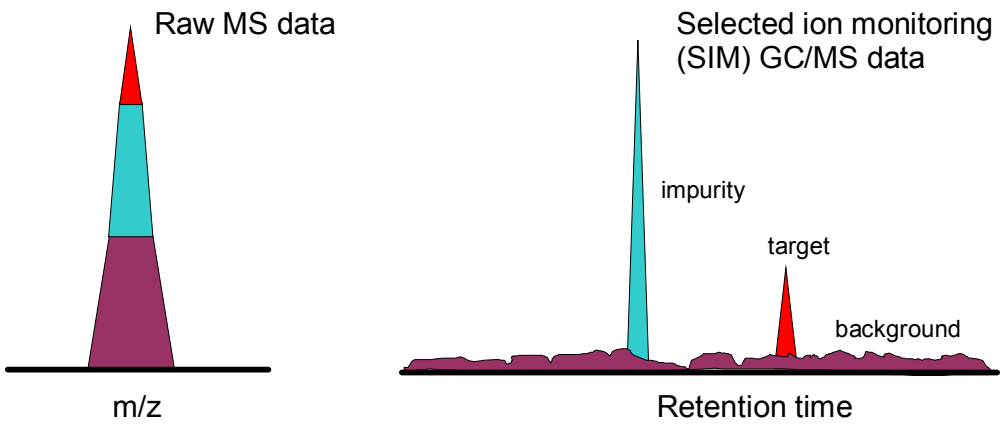

Figure 4. Diagram illustrating benefit of GC/MS analysis vs. MS alone.

c. Pyrolysis/GC

Pyrolysis is based on a thermal degradation of a complex media, and is a convenient way to break down biological material, such as spores, bacteria, toxins, to molecular fragments that can be analyzed directly by MS or other detectors. The pyrolysis surface may be a strip of metal, a silica frit or a silica felt. One can use metal alloys to control temperature in a process called Curie Point pyrolysis; however, we find that this is as limiting as it is useful. The pyrolysis temperature is ramped to about 500 C in about 1-2 s. This temperature gives a good balance between fragmentation from polysaccharides (carbohydrates) and protein fractions, both of which are abundant in bacteria.[9]

A tremendous benefit of pyrolysis is that it is an extremely convenient method of sample preparation. However the break-down process is harsh and reduces the biological material into a large variety of small molecular fragments that can produce a very cluttered spectrum. The Bruker and ORNL groups analyze the pyrolysis mixtures directly into their detector using CI or EI for ionization and IT MS for analysis. The multivariate data generated by the Bruker and ORNL Py/MS analyzers is reduced by multivariate algorithms using principal components analysis (PCA).[10,11,12] PCA is capable of reducing the dimensionality of multivariate data and preserving most of the variance, and can be an effective technique for observing the *natural* relationships between samples. However, the strong statistical nature of this analysis introduces uncertainties that can lead to false identifications.

We obtained even more definitive signatures by incorporating a GC after the Py. This combination was developed at the U.S. Army Edgewood Chemical and Biological Center (ECBC). They obtained successful tests using an IMS detector. The first Py/GC/MS tests using Syagen's PI/QitTof MS technology proved very successful. Figure 4 illustrates the advantages of incorporating an orthogonal dispersive component to the detection scheme. Typical Py/MS data contain a multitude of peaks and overlapping signals. Figure 4 considers the detection of three different constituents of the same mass m/z. The MS spectrum alone is not able to distinguish the target compound signal (in red) from impurity (blue) or background (brown). However when dispersed by GC

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[9] Windig, W., P.G. Kistemaker, J. Haverkamp and H.L.C. Meuzelaar: Factor analysis on the influence of changes in experimental conditions in pyrolysis mass spectrometry. *J. Anal. Appl. Pyrol.* **2** (1980) 7-18

[10] Chatfield, C. and A.J. Collins: *Introduction to Multivariate Analysis*, pp 57-81. Chapman & Hall, London (1980).

[11] Causton, D.R.: *A Biologist's Advanced Mathematics*, pp 48-72. Allen & Unwin, London (1987).

[12] Everitt, B.S.: *Cluster Analysis*. Edward Arnold, London (1993).

while monitoring the target m/z, it is clear that these three compounds of the same mass are easily distinguished. The orthogonal GC separation becomes even more important as the complexity of the mixture increases.

The GC units we use are termed flash GC for their rapid separations. The units we use (either from RVM Scientific, Santa Barbara, from ECBC, or that we make ourselves) are inexpensive, long-lived, require minimal maintenance, and require replacement only every few months. We use short columns that are fast. Furthermore we operate with room air and use the low pressure of the PI source to generate the pressure flow, thus obviating the need for a separate gas canister and pump.

3. Results

a. CW Identification

Syagen's detector technology provides molecular fingerprinting of CW and BW agents. Figure 5 shows the distinct spectral signatures for the nerve agent VX and the mustard agent HN1. The MS spectrum is typically sufficient to serve as a molecular identifier. However for definitive confirmation, the instrument can perform a rapid MS/MS. This process uses a waveform excitation to dissociate a specific parent ion to give a fragment ion. A confirmation is made if the fragment ion matches the known fragment signature ion. For example, the detection of an ion mass at m/z 268 in the MS spectrum is strongly indicative of VX; however, the presence of a m/z 268 ion that dissociates to a m/z 128 ion can be assigned to VX with near absolute certainty. Table II lists all CW relevant compounds that have been tested using a PI/QitTof MS instrument. The instrument detected 46 of 48 compounds; only phosgene and chloropicrin were undetected, since their ionization potentials are greater than the photon energy.

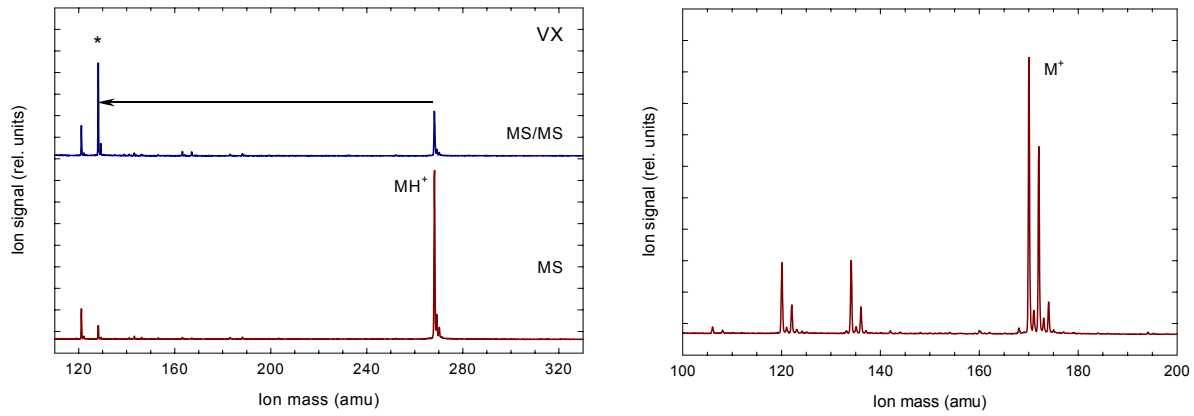

Figure 5

Distinct MS signatures for VX (left) and HN1 (right). The VX spectrum also shows selective ion fragmentation (MS/MS) to give definitive fragment signature peaks. (The MH^+ ion signal arises by formation of M^+ followed by hydrogen abstraction from the i-PrOH solvent used to introduce the sample.) The HN1 spectrum shows the isotope distribution for 2 chlorines.

Table II. CW related compounds tested with Syagen.

Schedule-1 compounds	Schedule-2 compounds	Schedule-3 compounds
Toxic	***Toxic***	***Precursors***
VX	BZ	Phosphorus oxychloride
GA (Tabun)		Dimethyl phosphite
GB (Sarin)	***Precursors***	Trimethyl phosphite
GD (Soman)	Diisopropyl methylphosphonate	Diethyl phosphite
GF	Diethyl methylphosphonate	Triethyl phosphite
	Dimethyl methylphosphonate	Triethanolamine
HD (sulfur mustard)	Diethyl ethylphosphonate	N-ethyldiethanolamine
HN-1 (Nitrogen mustard)	Diethyl methylphosphonothioate	N-methyldiethanolamine
HN-3	Isopropyl methylphosphonic acid	
	Cyclohexyl methylphosphonic acid	
CR	Pinacolyl methylphosphonic acid	**Decomposition Products**
CS	Methyl phosphonyl dichloride	
	3-quinuclidinol	Methylphosphonic acid
Precursors (Binary)	Benzilic acid	Ethylmethylphosphonic acid
QL	Pinacolyl alcohol	EMPTA
DF	Thiodiglycol	2-chlorodiethylsulfide
	N,N-diethylethanolamine	
	1,4-dithiane	**Other relevant compounds**
	Thiodiglycol sulfoxide	
	Methylamine	VX disulfide
	Isopropylamine	YL
	Thioxane	

Not detected: Phosgene, chloropicrin

Live agents detected at ECBC Surety Lab. Other compounds were detected at ECBC or at Syagen.

b. BW Detection and Classification

In this section we review our success in adapting a Py and Py/GC inlet system developed by the U.S. Army ECBC to our high-performance portable MS system. As discussed earlier, pyrolysis is not an elegant method of breaking down biological particles; however, it produces a rich composition of chemical fragments that give distinguishing signatures in PI MS and in GC / PI MS. Figure 6 shows the MS spectrum obtained by Py/MS for a variety of biological entities. Visually, they look more similar than different. The current CB MS

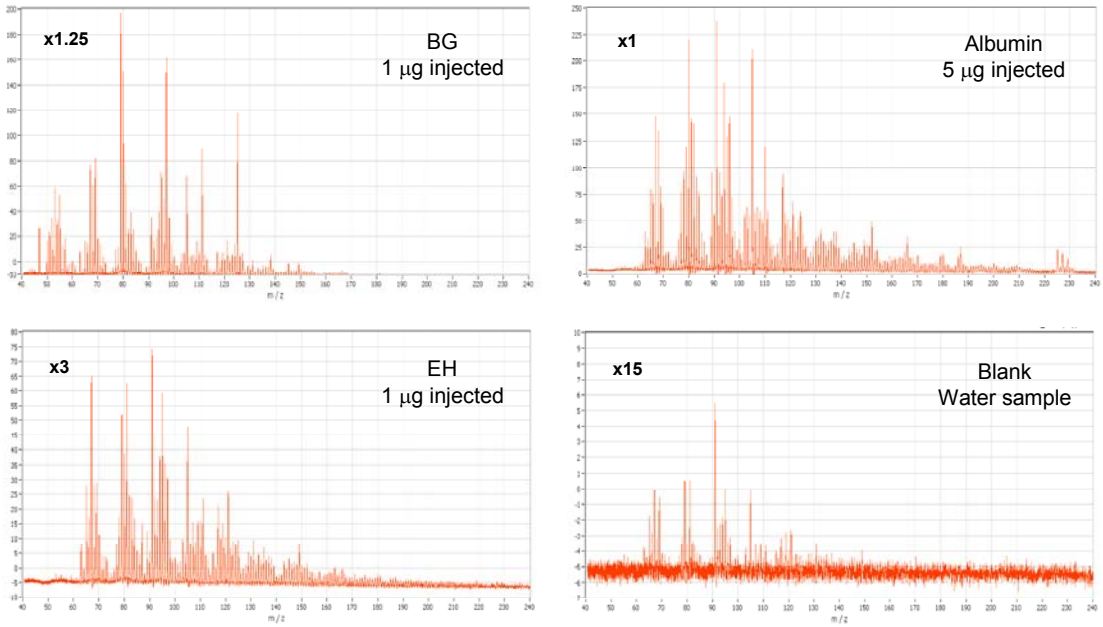

Figure 6. Py/GC PI/QitTof MS summed mass spectrum for four three biological samples and a water blank. Each spectrum is on a different intensity scale (e.g., water blank which shows carry over is <1/20 the intensity of the BG spectrum.

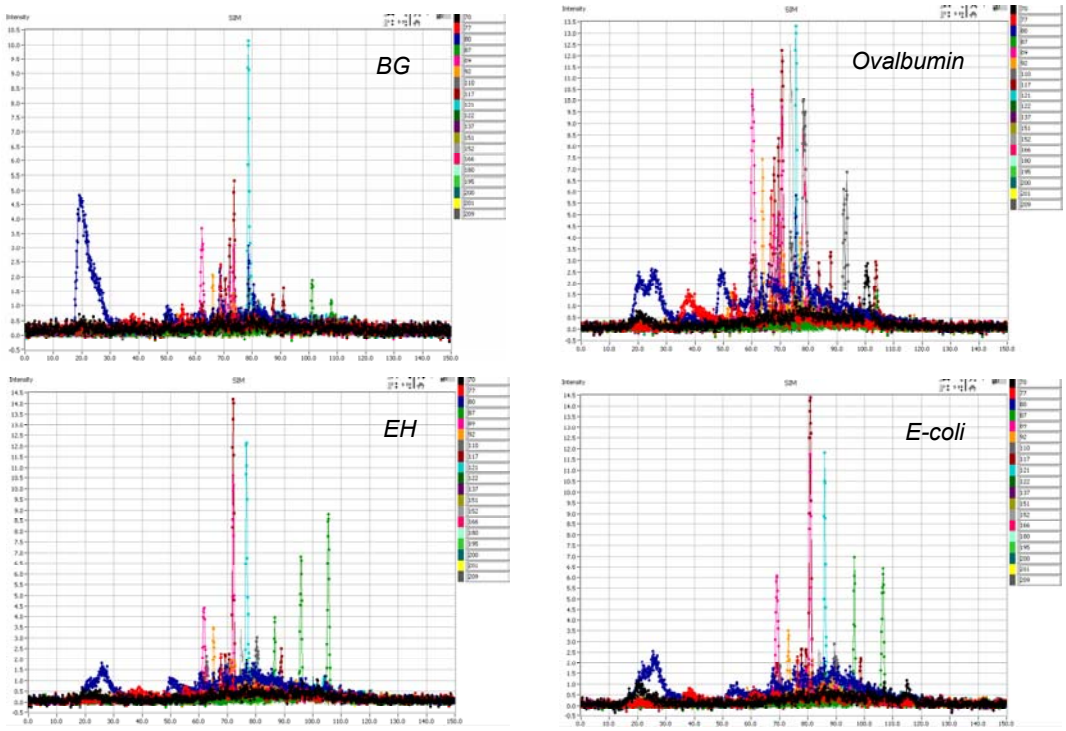

Figure 7. SIM traces showing specific ion signatures for a variety of biological samples. Note also the similarity of signals for EH and E-coli. Each SIM trace corresponds to a different m/z (different shades).

Block I and Block II systems (Table I) are based on analyzing Py/MS spectra using statistical methods to differentiate between the MS spectra of different biological entities. This method has achieved marginal success in discriminating between different biological entities. Our strategy is to recognize that by improving key areas of performance we could improve the success of a Py/MS approach from marginal to significant. The three principal improvements to the MS analyzer itself (details in Section 2a and 2b) are:

- Superior ionizer PI (vs. CI or EI)
- QitTof MS rather than conventional scanning IT MS
- Ported, differential pumping for higher sample throughput

These enhancements, particularly the use of PI, generated total Py/MS spectra (Figure 6) that appear to give better differentiation than the other CB MS methods. Differentiation is visually apparent in Figure 6. However, the real test is to expand the compound list and to determine how well the biological mass spectral information manifests itself in a complex environment with strong chemical background.

In order to better resolve the biological signature information from the pyrolate mixture, we first separated the pyrolate mixture by GC and then recorded PI mass spectra as a function of GC retention time. The dispersion of the Py products by GC gives a very identifiable set of signature peaks as shown in Figure 7. These Py/GC PI MS selected ion monitoring (SIM) chromatograms show some of the similar biological signatures that enable the detector to positively perform as a cuing/trigger (differential biological from non-biological). Different biological samples can be differentiated in most cases. Figure 8 compares the SIM chromatograms for BG (bacillus globigii) vs. EH (erwinia herbicola) that highlight some of the signatures that are specific to biological materials. Several other distinct signature peaks have been observed for these different biological samples. Py/GC/PI MS signatures were recorded for a total of 6 biological samples (GC, EH, E-coli, Ovalbumin, Bovine Serum Albumin, and Bradykinin), and differentiation was observed in all cases except for EH and E-coli, which give nearly identical signatures even for the weakest features as shown in Figure 9. These results show the po-

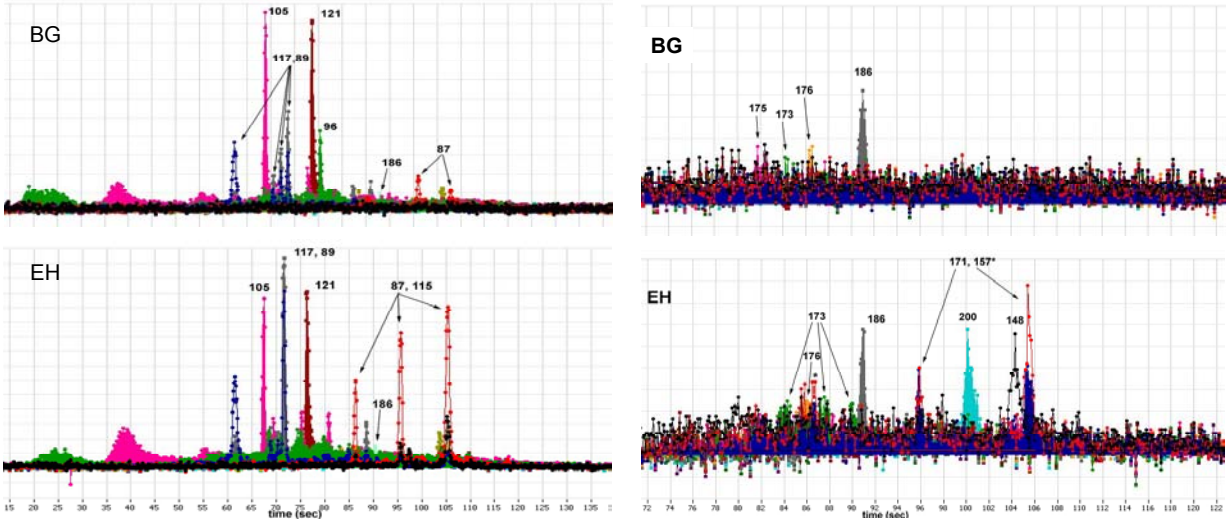

Figure 8. SIM chromatograms showing specific ion signatures for BG and EH. Several other distinct signatures have been identified for these and other biological samples. Each SIM trace corresponds to a different m/z (different shades). Chromatograms showing strong (left) and weak (right) signatures. Spectra were recorded for 1 μg particle weight injected as slurry in water onto pyrolyzer. The total GC run was 150 s.

tential to classify biologicals within groups (e.g., gram positive vs. gram negative bacteria and spores, toxins, proteins, etc.). These results also provide evidence for the potential to obtain identification in some cases. These feasibility measurements demonstrate the potential of the current CB MS configuration to greatly exceed other MS technologies for the combined capability to:

- Collect and measure with minimal sample handling/preparation
- Achieve short cycle times
- Make definitive classifications of biologicals
- Monitor simultaneously a wide range of BW and other threats.

These capabilities are in addition to the CW identification capability described earlier.

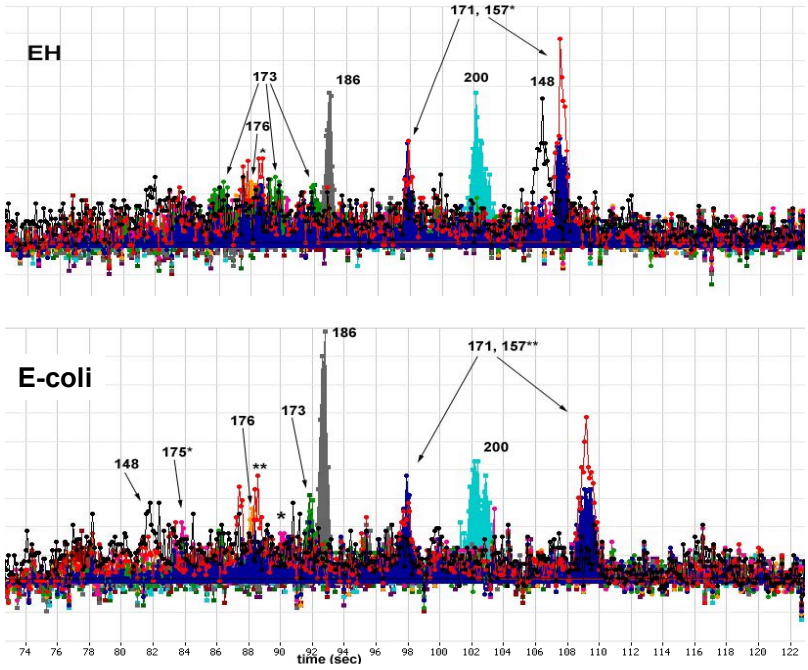

Figure 9. SIM chromatograms showing virtually identical signatures for EH and E-coli even for the weakest features. Spectra were recorded for 1 μg particle weight injected as slurry in water onto pyrolyzer. The total GC run was 150 s.

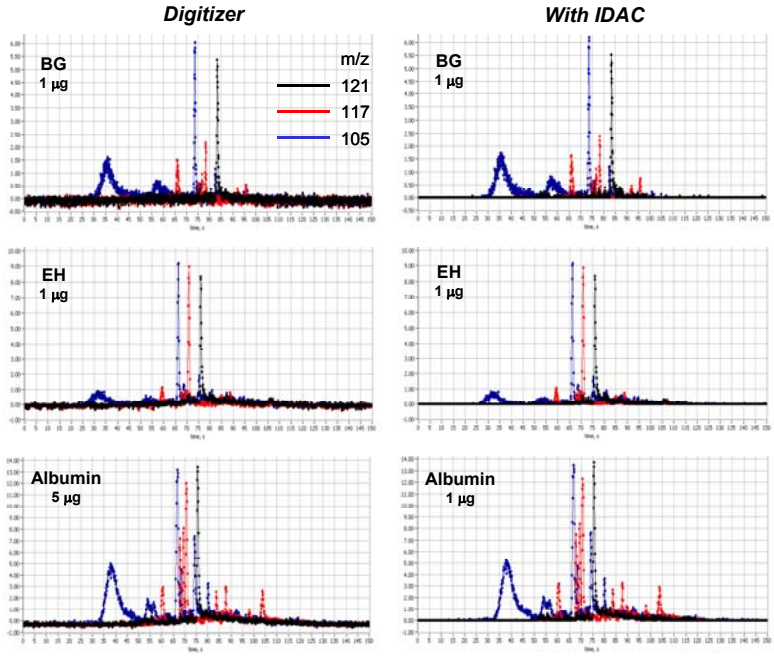

Figure 10. SIM chromatograms for biological detection for conventional digitizer vs. IDAC data acquisition. These data show results for strong signatures.

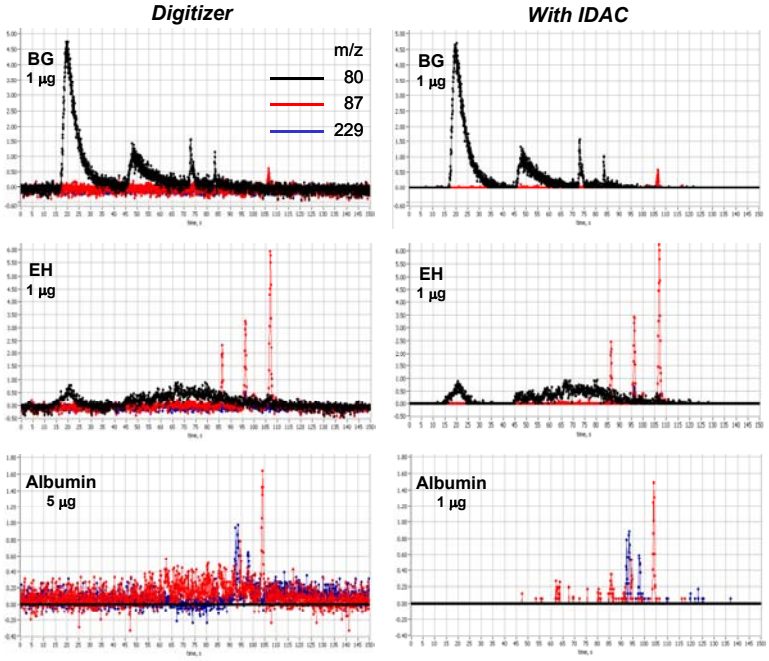

Figure 11. SIM chromatograms for biological detection for conventional digitizer vs. IDAC data acquisition. These data show results for weak signatures.

c. Sensitivity Enhancement using IDAC

Syagen has developed a patented technology to remove analog noise from TOFMS spectra that significantly increases the signal-to-noise and improves detection limits for Py/GC/PI TOFMS analysis of biological particles. The technology essentially acts as a combined ion counter and digitizer and is called IDAC (Integrated Digitizer and Counter). Using fast processing and the IDAC algorithm, it is possible to process each individual TOF spectrum (typically 80,000 pts at 8-bits and 60 Hz) to distinguish intensity down to the single ion count level from noise. By setting a threshold intensity and retaining all signals above the threshold and setting all other signal to a baseline level, each TOFMS spectrum is reduced to only a signal spectrum. The sum of these spectra contain virtually no noise. The SIM chromatograms for specific ion masses also show significant reduction in noise.

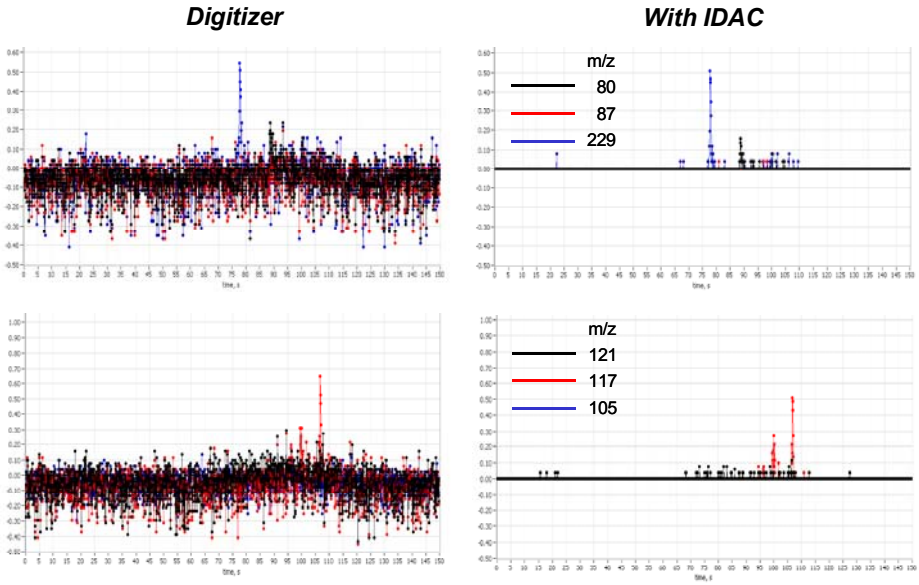

Figure 12. SIM chromatograms for a water blank for conventional digitizer vs. IDAC data acquisition.

Examples of the benefit of IDAC on the biological detection by Py/GC/PI TOFMS are shown in Figures 10 and 11. Figure 10 compares the strong signatures for BG, EH, and ovalbumin in the conventional digitizer mode and for the IDAC mode. The noise reduction is more evident for weaker signatures as shown in Figure 11.

The improvement in sensitivity using IDAC is considerable. Figure 12 shows SIM chromatograms for six key signature peaks for BG, EH, and Ovalbumin differentiation. The IDAC chromatograms are essentially devoid of noise. The weak features are actually real ion signals, which in this water blank case are due to background ions. Individual ion counts are clearly evident. The limit of detection (LOD) for the three biological entities are summarized in Table III. Differentiation of these biologicals is possible with just three signature peaks as shown in Figure 13. These represent the so-called weak signatures in Figure 11. Improved sensitivity can be obtained using the strong signatures in Figure 10, however, these signatures do not discriminate as well. A strategy may be to use the strong signatures to detect and the weak signatures to differentiate.

Using IDAC the mass LOD is considerably reduced for BG (m/z 80 signature) and for Ovalbumin (m/z 229 signature). The EH LOD was only moderately improved (m/z 87 signature) due to a chemical background in this particular case, that limited the low end detection limit. The LODs are also summarized in terms of number of particles. Based on a mass-weighed median particle distribution size of 5 μ diameter, the particle LOD is as low as 8 for BG. However, realistically, we expect particle LODs of 100-1000 particles. Using an aerosol collector to sample upwards of 1000 L with 50% particle collection efficiency translates to potential LODs of about 1 ACPLA (agent containing particle for liter air).

Table III. Limit of detection (LOD) using weak identifier signatures.

	IDAC			Analog		
	80	***87***	***229***	***80***	***87***	***229***
BG	**501**	0.1	0.0	**520**	-1.7	0.1
EH	21.7	**20.5**	0.3	31.7	**19.4**	0.589
Albumin	196.0	0.0	**13.9**	196.0	-0.6	**15.7**
LOD (ng particle)	**0.8**	**46.3**	**4.0**	**4.7**	**63.7**	**43**
LOD (# particles)	**8**	**463**	**40**	**47**	**637**	**434**

based on typical 5 μ particle size

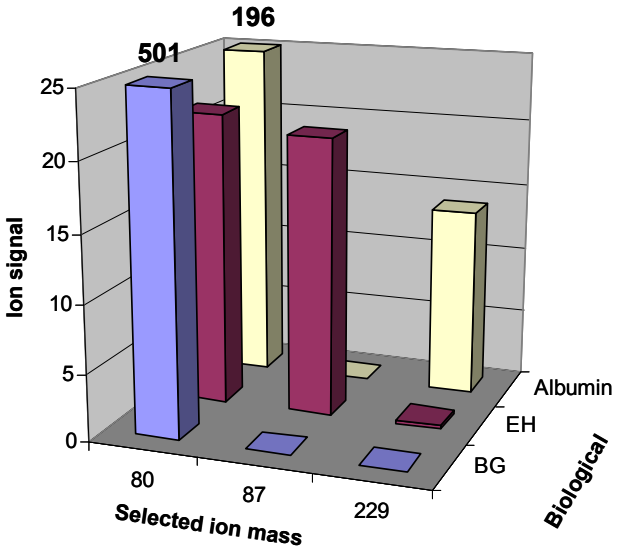

Figure 13. Histogram representation of BG, EH, and Ovalbumin discrimination by just three signature peaks. It should be noted that more than three signatures may be needed to discriminate against a much wider range of biological entities.

d. Biological Detection Strategy

No biological detection system will meet the requirements for all applications and it is arguable that no biological detection system will meet the requirements of even a single application. Rather a combination or suite of detectors may be required. The Syagen CB MS system is ideal for "detect to warn" applications. It provides identification capability for CW agents and classification capability for BW agents. The system is most effective for fixed location, rapid screening applications, but can also be adapted for mobile applications.

Table IV summarizes the key functional categories of an effective BW analyzer. The more of these categories that are provided for by a single subsystem, the more efficient the overall system will operate. For major BW analyzer systems, such as Joint Biological Point Detection System (JBPDS), there is generally a separate device for each category. Table IV summarizes current devices. The major point of the Syagen CB MS is to provide multiple functionality as shown in Table IV. The current CB MS serves as a trigger, detector and in some cases a BW identifier. More conservatively it can classify BW entities into biological groups (e.g., gram positive vs. negative bacteria, viruses, biotoxins, etc.). On a per analysis basis, the CB MS operates rapidly and inexpensively with very low false positive and negative rates. The CB MS should be used for continuous monitoring and only when a positive detection of a BW agent is detected should a biological identifier, such as immunoassay or PCR, be used. Because the probability of a positive event is very low it is not sensible to use an expensive and slow identification technology, rather it should be used in a confirmatory role.

Table IV. Functional categories of a BW analyzer

Category	***Function***	***Current devices***	***Syagen strategy***	***Comments***
Collector	Collect aerosol and vapor	Aerosol collector only	Aerosol and vapor collector	
Trigger	Particle trigger	Light scattering	Syagen MS detector (Py/GC/ PI MS) - Classifies BWs - Identifies CWs	Benefit of Syagen MS: - Combined trigger, detector, and classifier - Dual BW and CW - BW classifier narrows candidates for identifier
Detector	BW vs. non-BW aerosol	BAWS, MS		
Classifier	Identify BW class			
Identifier	Identify BW	Immunoassay	Immunoassay or PCR	- Not all threats identifiable - Time consuming

e. Relevance to DOD and DHS Requirements

The CB MS system described herein provides automated, unattended screening and serves as a continuous early warning system detecting toxic releases. The combined Py/GC PI MS system achieves high specificity, high sensitivity, and low-false positive rate. It also achieves low operating and maintenance costs. We foresee this monitoring system as having the potential for unmatched capability for deployment at airports, post office distribution systems, buildings (in ventilation system), VIP and public venues. The CB monitoring system can be compatible with existing emergency response systems within U.S. cities. This application leverages the Army/SBCCOM requirement for a battlefield early warning system for CB attack.

With increasing governmental regulations, environmental concerns, and military and civilian defense needs, not to mention the heightened concerns from 9/11, the demand for real-time multispecies monitoring systems is growing rapidly. The specifications of the proposed instrument meet the requirements for a broad range of military and civilian applications, including:

- Military force protection from threat of CB terrorism.
- Chemical demilitarization, biological/toxin weapons convention verification and counterproliferation (collateral effects).
- Emergency first response for hazardous materials and portable detectors for contamination in potable water, decontamination/water treatment monitoring and general contamination avoidance.
- Law enforcement for crime scene investigation requires portable detection systems.
- Air quality monitoring of large public venues for civilian defense and environmental pollution monitoring in municipal water facilities and public waterways.
- Medical diagnostics for infectious disease monitoring and for biomolecular analysis.

Based on feasibility results to date and reasonable engineering targets we anticipate that a deployable CB MS system will have the specifications summarized in Table V.

Table V. Target specifications for Syagen's CB MS screening system.

Performance Characteristics		*Operational Characteristics*	
Sensitivity – vapor	<10 ppt (2 min response) <10 ppb (10 s response),	Startup time	10 min
Sensitivity – particle	1 ACPLA for BWA		
Specificity	> 10^4 clutter suppression (e.g., air molecules)	Environment	-5°C to +60°C
Cycle time	0.2 – 2 min (CW) 2 – 3 min (BW)	Flexibility	- CW and BW detection - Targeted compound detection
Dynamic range	4-decade		list is easily software modified
System Characteristics		*Physical Characteristics*	
Sampler	High-volume vapor/particle concentrator	Size	14x16x18 in (without concentrator)
Collection flow	700 L/min	Weight	50 lbs (without concentrator)
Mass analyzer	QitTof	Power	24 VDC, 6 A
Mass range	20-1000 amu		115 VAC, 2 A
Mass accuracy	$\Delta m < 0.1$ amu		

CONCLUSIONS

Chemical and biological agents are weapons of mass destruction that are accessible to terrorists and rogue organizations because of their simplicity and low cost. In order to develop better means to counter these threats, economical, high-resolution, field-portable biological and chemical detection systems are needed. High speed is needed for rapid response in threat situations and for high throughput analysis for treaty compliance and nonproliferation monitoring.

In this paper we have described Syagen's field-portable detection systems for rapid and reliable chemical and biological (CB) agent monitoring. Effective sensors must integrate the requirements for efficient recovery of microorganisms, processing of microorganisms for detection, and molecular analysis for accurate pathogen identification. For in-field detection, the sensors must be compact, rugged, reliable, and low maintenance while achieving exceptional levels of sensitivity, specificity, and speed. The instrument criteria that this work addresses are:

- Combined CW and BW detection
- Continuous, automated, real-time screening
- Molecular identification giving low false positive rate
- Low consumables, operational simplicity, and low recurring costs