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Liquid chromatography electrospray tandem mass spectrometric and desorption electrospray ionization tandem mass spectrometric analysis of chemical warfare agents in office media typically collected during a forensic investigation

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Abstract

Most prior analytical studies have dealt with the determination of chemical warfare agents in environmental or biological matrices that would typically be collected following battlefield use or in support of the Chemical Weapons Convention. These methods may be useful for some investigations, but may not be practical for indoor forensic investigations where chemical warfare agent use is suspected. There is a need for analytical methods for chemical warfare agent identification in office media, including flooring, wall surfaces, office fabrics and paper products, which would typically be collected in an office environment during forensic investigations. During this study, typical office environment media were spiked at the 4–20 μg/g level with either a complex munitions grade sample of tabun (GA) or with a standard containing the three nerve agents, sarin (GB), cyclohexyl methylphosphonofluoridate (GF), soman (GD) and the nerve agent simulant, triethyl phosphate (TEP), to evaluate the potentials of liquid chromatography electrospray ionization mass spectrometry (LC-ESI-MS) and liquid chromatography electrospray ionization tandem mass spectrometry (LC-ESI-MS/MS) for forensic purposes. An emerging technique, desorption electrospray ionization (DESI-MS/MS), was also investigated for the direct determination of TEP, GB and GD sampled onto solid phase microextraction (SPME) fibers exposed to spiked office media. The spiked chemical warfare agents were recovered with varying efficiencies during this study, but in all cases sufficient chemical warfare agent was recovered for mass spectrometric identification purposes. Full high resolution mass spectra were acquired for all the chemical warfare agents in the continuum mode, which typically resulted in mass measurement errors of 0.001 Da or less.

Keywords: Liquid chromatography; Electrospray ionization; Mass spectrometry; Tandem mass spectrometry; Desorption electrospray ioniozation; Chemical warfare agents

1. Introduction

The ending of the Cold War and the widespread acceptance of the Chemical Weapons Convention has reduced the likelihood of battlefield chemical weapons use, but there remains a serious concern world-wide that other parties may make use of chemical warfare agents against civilian or military targets. Sarin, a well known nerve agent, was used by the Aum Shinrikyo sect in Japan in 1995 during an attack on the Tokyo underground transit system, during which a dozen people were killed and thousands more were injured. Public concern about the use of chemical warfare agents, or other weapons of mass destruction, reached a new peak following the terrorist events of September 11, 2001 and the subsequent delivery of anthrax-containing letters through the United States postal system. These events heightened security concerns within many countries and considerable resources were expended to improve all facets of response to the perceived terrorist threat.

Within Canada and other nations, laboratories or laboratory networks have been established to deal with the analytical challenges following a terrorist incident. The focus within these laboratories varies from nation to nation but generally deals with the evaluation of existing identification methodologies, the identification of analytical deficiencies and the development of new analytical methods to meet the perceived challenges. Within Canada, the Chemical, Biological, Radiological and Nuclear

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Research and Technology Initiative (CRTI), a research-oriented organization, established four clusters including a Chemical Laboratory Cluster to deal with the analytical (and other) challenges associated with chemical warfare agent use.

Analytical methods for the detection and identification of chemical warfare agents, their degradation products and related compounds have been thoroughly reviewed with different emphases on a number of occasions [1–7]. Much of the analytical methods development was driven by the requirements of the military and their need to be able to detect and identify these compounds in typical battlefield samples. These and other methods covered in the reviews have focused largely on the determination of chemical warfare agents or their degradation products in environmental matrices such as soil [8–11], water [10,12–15], air [16,17], recovered munitions and munition blocks [9,18], decontamination solutions [19–21] and military clothing/gear [8,9]. Newer methods based on solid phase microextraction (SPME) sampling followed by gas chromatography (GC–MS) analysis [22–25] and direct analysis by secondary ion mass spectrometry [26] have been reported for environmental analyses, but most of the literature methods have been based on GC–MS analysis of an extract of the collected medium [8–21].

Organic extracts of chemical warfare agents may be analysed directly by GC–MS, but the hydrolysis products of chemical warfare agents usually require derivatization prior to GC–MS analysis [6]. More recently researchers have demonstrated the value of liquid chromatography-mass spectrometry (LC–MS) as a complementary or replacement method for GC–MS, particularly for the confirmation of hydrolysis products of chemical warfare agents in aqueous extracts or samples [27–35], as the hydrolysis products may be analysed directly without the need for additional sample handling and derivatization. In addition, LC–MS has the added benefit that it may also be utilized for the determination of organophosphorus chemical warfare agents and related compounds in aqueous samples and extracts [31–34].

Methods for biological media, such as urine and blood, received increased attention following the use of mustard during the Iran-Iraq War [36], the Tokyo subway incident [37,38] and continues today given the concern over possible use of chemical warfare agents against civilian populations. Biological methods have been reviewed recently [4] and *J. Anal. Toxicology* (volume 28, 2004) recognized this research emphasis with a special issue dealing with the mass spectrometric determination of chemical warfare agents in biological samples.

The realization that terrorist use of chemical warfare agents against civilians could involve the targeting of enclosed populated spaces,. emphasized the need for the development of analytical methods for these compounds in media that would typically be collected during forensic investigations. The present study focused on the development and application of sample handling and liquid chromatography electrospray tandem mass spectrometry (LC-ESI-MS/MS) analytical methods for contaminated office media, including flooring, wall surfaces, office fabrics and paper products.

Typical office environment media were spiked at the 4–20 μg/g level with either a complex munitions grade sample of tabun (GA) or with a standard containing three nerve agents, sarin (GB), cyclohexyl methylphosphonofluoridate (GF), soman (GD) and the nerve agent simulant, triethyl phosphate (TEP), to evaluate the potentials of LC-ESI-MS and LC-ESI-MS/MS for forensic purposes. Preliminary investigations with spiked office carpet [39] were extended to include other office media including, latex painted drywall, office fabrics, photocopy paper and Dacron sampling swabs. Representative samples of each were spiked with chemical warfare agents and/or related compounds, extracted with water using ultrasonic vibration, centrifuged to reduce the presence of fines and analysed by LC-ESI-MS and LC-ESI-MS/MS. A novel mass spectrometric method for sample ionization and analysis, developed by Cooks' group and referred to as desorption electrospray ionization (DESI) [40]), was also evaluated for the first time for the direct analysis of solid phase microextraction (SPME) fibers used to sample the headspace above office media spiked with TEP [41,42], GB and GD.

The spiked chemical warfare agents were recovered with varying efficiencies, but in all cases sufficient chemical warfare agent was recovered for identification purposes.

2. Experimental

2.1. Samples and standards

Samples of six different office media were selected for evaluation (typical sample weights used in the spiking experiments are given in parentheses).

1. 100% (solution dyed) Nylon office carpet (1 g).
2. 20 lb white photocopy paper (0.25 g).
3. Fisherbrand sterile Dacron swab tip (0.1 g).
4. Latex painted drywall (0.17 g).
5. 56% Nylon/44% polyester office fabric treated with Scotchguard (0.23 g).
6. 100% Nylon office fabric treated with Teflon (0.15 g).

Two different standards were used for spiking the office media listed above.

1. The standard containing GB, GD, GF and TEP was prepared at an individual concentration of 0.4 mg/ml in dichloromethane. Triplicate samples of each of the media were spiked with a 5 μl aliquot of the mixture with the exception of the office carpet sample, which was spiked with a 10 μl aliquot.
2. The munitions grade tabun (GA) standard contained GA and a number of other sample components with the concentration of the sum of all the sample components being 1 mg/ml in dichloromethane. The office carpet and the photocopy paper were spiked with 25 and 10 μl aliquots of the munitions grade GA, respectively.

2.2. Sample handling

Weighed samples of the listed office media were placed in individual 20 ml screw capped glass scintillation vials. The samples were then spiked with either 5 μl (or 10 μl) aliquots of the

standard containing GB, GD, GF and TEP or 10 μl (or 25 μl) aliquots of the munitions grade GA sample. The spiked samples were allowed to stand for 30 min. Spiked samples were ultrasonically extracted with 2 ml (photocopy paper, latex painted drywall, Dacron swab) or 5 ml (office carpet and both office fabrics) of water (Aldrich-Sigma) for 10 min. The water volume was sufficient to completely immerse the sample and a narrow glass insert of the correct height was inserted into the scintillation vial to ensure that the sample did not float. The vial was then subjected to ultrasonic vibration for 10 min. A 1 ml aliquot was removed and centrifuged at 14,000 rpm to remove any fines. A 750 μl portion of the centrifuged sample was placed in a 1.8 ml glass autosampler vial for analysis. The same procedures were followed for (unspiked) office media blanks.

2.3. LC-ESI-MS analysis

LC-ESI-MS and LC-ESI-MS/MS data were acquired using a Waters Q-ToF Ultima tandem mass spectrometer equipped with a Z-spray electrospray interface. The electrospray capillary was operated in the 1–3 kV range with a sampling cone voltage of 35 V. The collision energy was maintained at 5 V for LC-ESI-MS operation and was varied from 2 to 10 V (depending on the precursor ion selected) for LC-ESI-MS/MS operation. Argon was continually flowing into the collision cell at 9 psi during both LC-ESI-MS and LC-ESI-MS/MS operation. Nitrogen desolvation gas was introduced into the interface (80 °C) at a flow rate of 300 L/h and nitrogen cone gas was introduced at a flow rate of 50 L/h. ESI-MS data were acquired from 40 to 700 Da (1 s with a 0.1 s interscan delay or 0.3 s with a 0.1 s interscan delay) and ESI-MS/MS (product ion mass spectra) data were acquired for the protonated molecular ions of the spiked compounds (1 s with a 0.1 s interscan delay). All data were acquired in the continuum mode with a resolution of 9000 (V-mode, 50% valley definition).

Chromatographic separations were performed with a Waters CapLC using a 5–75% B gradient over 30 min and a flow rate of 10 μl/min. The following solvent compositions were prepared for the mobile phase: Solvent A (0.1% trifluoroacetic acid in water) and Solvent B (acetonitrile). All LC separations were performed with MicroTech 150 mm × 0.32 mm i.d. fused-silica capillary columns packed with Zorbax C18 SB (5 μm particle size). The CapLC autosampler was used to introduce 1 μl samples of the aqueous extracts.

The retention times varied slightly throughout the course of the study due to the use of a new LC column (same packing material) for the latex painted drywall, Dacron swab and office fabrics sample extract analyses.

2.4. DESI-MS analysis

DESI-MS and DESI-MS/MS data were acquired under mass spectrometric conditions similar to the LC–MS investigations. Initial investigations with TEP [41,42]) were performed with the Z-spray interface glass sleeve removed to allow introduction of the SPME fiber into the region between the electrospray needle and the sampling cone. A laboratory stand was used to hold and position the SPME manual holder so that the fiber could be introduced into an ethanol/water (1:1) mobile phase spraying at 10 μl/min. DESI-MS experiments that vented into the laboratory (without the glass interface sleeve) were not attempted with either chemical warfare agents or the LC mobile phase for safety reasons. A replacement plexiglass sleeve was then machined with a small beveled hole approximately the diameter of the SPME fiber sheath to facilitate the safe introduction of SPME fibers contaminated with chemical warfare agents. The acetonitrile/water LC mobile phase (isocratic, 5% B) was sprayed at 10 μL/min during these analyses.

The office carpet sample was spiked in a similar manner to the LC–MS experiments with TEP, GB or GD (2–4 mg/ml individual standards in dichloromethane). Vials containing the spiked carpet were sampled using a Supelco (65 μm film thickness) polydimethylsiloxane/divinyl benzene (PDMS/DVB) SPME fiber. The SPME fiber was inserted through the septum in the cap and allowed to sample the headspace above the spiked carpet for 3 min at room temperature. It was then inserted into the region between the electrospray needle and the sampling cone, using the new interface sleeve. Product mass spectral data were acquired for the protonated molecular ion of each compound. For GD, the product ion mass spectrum was acquired for *m/z* 183 over a 70–500 Da mass range (1 s with a 0.1 s interscan delay) with a collision energy of 3 V.

3. Results and discussion

3.1. LC-ESI-MS analysis

Analytical methods need to be developed to ensure that suspect samples collected during forensic or other investigations can be analysed for the presence of chemical warfare agents in a timely manner. Increasingly, researchers have utilized LC–MS as a complementary or replacement method for GC–MS, particularly for the confirmation of hydrolysis products of chemical warfare agents in aqueous extracts or samples [27–35]. Recent publications have also demonstrated the application of LC-ESI-MS for the determination of organophosphorus chemical warfare agents and related compounds in water, snow and aqueous extracts [31–34]. Aqueous extracts of sample media offer a number of advantages and disadvantages for chemical warfare agent analysis. Advantages include the fact that aqueous extracts may be used to analyse both organophosphorus chemical warfare agents and their hydrolysis products during a single LC–MS analysis, that aqueous extracts tend to contain less co-extracted chemical interferences than organic extracts and that aqueous extracts do not generally dissolve or degrade the sample media as can be the case for organic solvent extracts. The principal disadvantage to aqueous extraction for the determination of organophosphorus chemical warfare agents is the possibility of chemical warfare agent hydrolysis during sample handling or on prolonged standing.

Methods involving aqueous extraction followed by LC-ESI-MS and LC-ESI-MS/MS analysis of soil samples contaminated with organophosphorus chemical warfare agents and their hydrolysis products [31] and soil samples containing the hydrol-

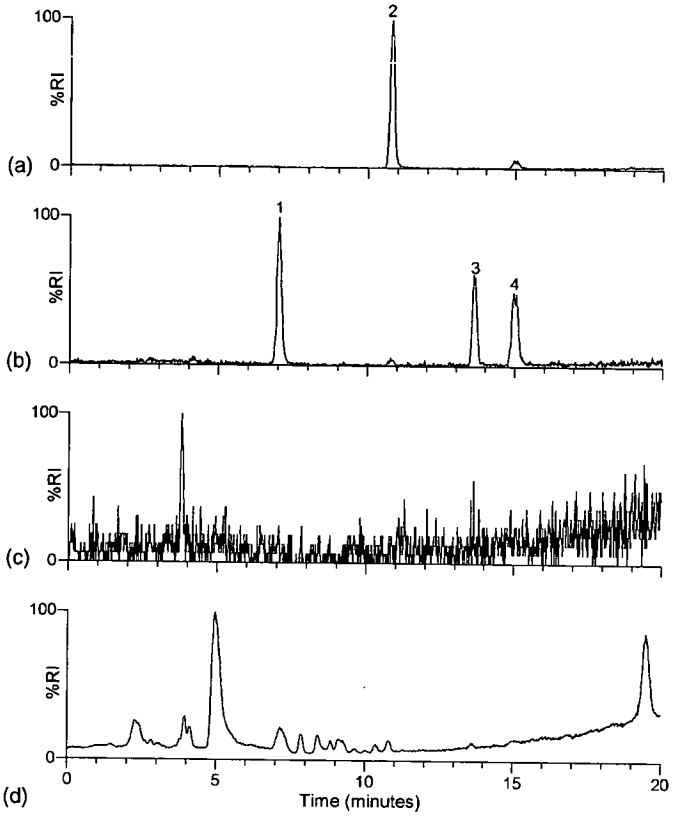

Fig. 1. Reconstructed-ion-current chromatograms for (a) *m/z* 183, (b) *m/z* 99, (c) *m/z* 97 and (d) total-ion-current chromatogram obtained during LC-ESI-MS analysis of an extract of an office carpet sample spiked at the 4 μg/g level with GB (1), TEP (2), GF (3) and GD (4).

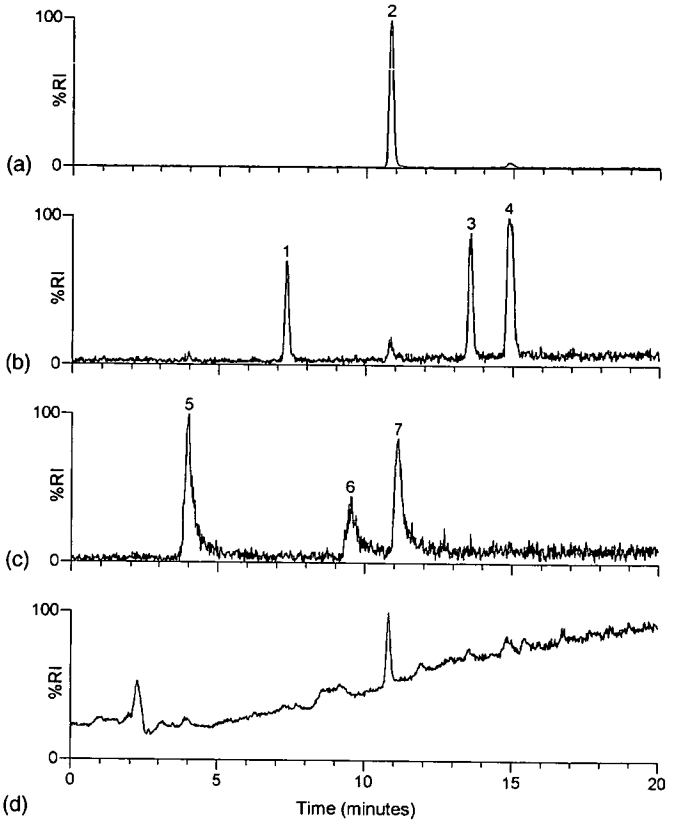

Fig. 2. Reconstructed-ion-current chromatograms for (a) *m/z* 183, (b) *m/z* 99, (c) *m/z* 97 and (d) total-ion-current chromatogram obtained during LC-ESI-MS analysis of an extract of a photocopy paper sample spiked at the 8 μg/g level with GB (1), TEP (2), GF (3) and GD (4). Isopropyl methylphosphonic acid (5), cyclohexyl methylphosphonic acid (6) and pinacolyl methylphosphonic acid (7), the hydrolysis products of GB, GF and GD, respectively, were also observed.

ysis products of munitions grade mustard [34] have been developed at DRDC Suffield. A similar approach for indoor office media that could be collected during a forensic investigation was worth investigating. The present study focused on the development and application of sample handling and LC-ESI-MS and LC-ESI-MS/MS analytical methods for contaminated indoor sample media that might be collected during a forensic investigation, including office flooring, wall surfaces, office fabrics and paper products.

Typical office environment media were spiked at the 4–20 μg/g level with a standard containing three nerve agents, sarin (GB), cyclohexyl methylphosphonofluoridate (GF), soman (GD) and the nerve agent simulant, triethyl phosphate (TEP), to evaluate the potential of LC-ESI-MS and LC-ESI-MS/MS for forensic purposes. Representative samples of the office media were spiked with the standard, extracted with water using ultrasonic vibration, centrifuged to reduce the presence of fines and analysed by both LC-ESI-MS and LC-ESI-MS/MS. The spiked compounds were recovered with varying efficiencies, but in all cases sufficient chemical warfare agent was recovered for identification purposes. Figs. 1 and 2 illustrate the reconstructed-ion-current chromatograms for *m/z* 183, *m/z* 99, *m/z* 97 and the total-ion-current chromatogram obtained during LC-ESI-MS analysis of the aqueous extracts of the office carpet and photocopy paper, respectively. Similar chromatograms were obtained for the Dacron swab, latex painted drywall, and two office fabrics, spiked with this standard. The total-ion-current chromatogram provided a good indication of the complexity of the sample extract. Significant co-extracted components were observed in the office carpet extract (Fig. 1), somewhat less were observed for the office fabrics and other sample extracts and very few were observed in the photocopy paper extract (Fig. 2).

The reconstructed-ion-current chromatograms for *m/z* 97, the common product ion observed for alkyl methylphosphonic acids formed after hydrolysis of GB, GF and GD, were presented for each extract to give an indication of hydrolysis, either during sample handling or on standing prior to analysis. Hydrolysis products were only noted during analysis of the photocopy paper extracts. At a collision energy of 4 V the three acids initially formed after hydrolysis of GB, GF and GD exhibited the $[M+H]^+$ precursor and a significant product ion at *m/z* 97 due to loss of the alkene associated with the alkoxy group of GB, GF and GD. Hydrolysis products were not evident in the extracts of the other office media provided the analyses were completed within approximately an hour of sample handling. Over increasing hours some hydrolysis of the spiked chemical warfare agents was evident, consistent with earlier findings [43].

The reconstructed-ion-current chromatograms for *m/z* 183 ($[M+H]^+$ for TEP and GD) and *m/z* 99 (a common product ion due to loss of the alkene associated with the alkoxy group

Table 1
Recovery of GB, TEP, GF and GD from office media

Spiking Agent	Percentage recovery from office media (spiking levels indicated)					
	Office carpet (4 μg/g)	Photocopy paper (8 μg/g)	Dacron swab (20 μg/g)	Latex painted drywall (12 μg/g)	56% Nylon/44% polyester office fabric (9 μg/g)	100% office fabric Nylon (13 μg/g)
GB	79	27	60	54	71	83
	86	30	57	56	91	72
	100	39	54	57	96	90
TEP	72	80	64	32	76	86
	82	88	72	36	94	82
	73	83	66	37	81	89
GF	43	23	56	20	69	69
	48	30	57	22	85	65
	49	37	47	26	75	76
GD	46	48	63	20	77	73
	53	52	62	27	89	67
	47	62	53	23	83	78

of GB, GF and GD) were used to indicate the presence of the four spiked compounds. ESI-MS data were similar to previously published data acquired with a ToF mass spectrometer [44], with the principal differences being in the relative abundances of the sodiated and protonated acetonitrile adducts.

A semi-quantitative estimate of the recovery efficiency of each compound from each of the extracts was calculated by comparing the area under the reconstructed-ion-current chromatogram profiles for *m/z* 99 + 141 (GB), *m/z* 183 (TEP), *m/z* 99 + 181 (GF) and *m/z* 99 + 183 (GD) with data obtained during analysis of a standard solution. Table 1 lists the recovery estimates for the office media in triplicate. Recoveries varied with both media and compound, with TEP, a compound that resists hydrolysis, generally being recovered with the greatest efficiency. Recoveries were for the most part in the 50–85% range for the spiked chemical warfare agents. The lowest recoveries, about 25%, were for GF and GD from the latex painted drywall, a surface material that absorbs chemical warfare agents more easily than the chemically resistant paints used for military applications. At the lower recovery levels full, interpretable, mass spectra (ESI-MS or ESI-MS/MS) were readily acquired for all the spiked compounds leading to the conclusion that GB, TEP, GF and GD could be identified in contaminated office media of the type described at or below the μg/g level. Recovery variabilities of this order were also observed by Tørnes, Opstad and Johnsen during a study investigating the recovery of GA, GB, GD, mustard and diisopropyl methylphosphonate from typical battlefield media including water, grass, soil, sand, protective suit materials and paper [8] and by D'Agostino, Hancock and Provost during the analysis of soils spiked with GB and GD [31].

In some cases, the aqueous extracts of the spiked office media contained numerous co-extracted sample components that complicated LC-ESI-MS analysis and hampered identification. These interferences were minimized during LC-ESI-MS/MS analysis, where each of the chemical warfare agents was identified on the basis of acquired product ion mass spectra. Fig. 3 illustrates the marked difference between the LC-ESI-MS total-ion-current chromatogram and the LC-ESI-MS/MS product ion chromatogram for the extract of a spiked office carpet sample. The GB was effectively masked by the other organic components and was not observable as a distinct chromatographic peak during LC-ESI-MS. LC-ESI-MS/MS was used to resolve the GB from the other extract components. The chromatogram obtained for the product ions of *m/z* 141 ($[M+H]^+$ for GB) during LC-ESI-MS/MS resulted in a significantly less complex chromatogram where GB was completely resolved as a significant chromatographic component.

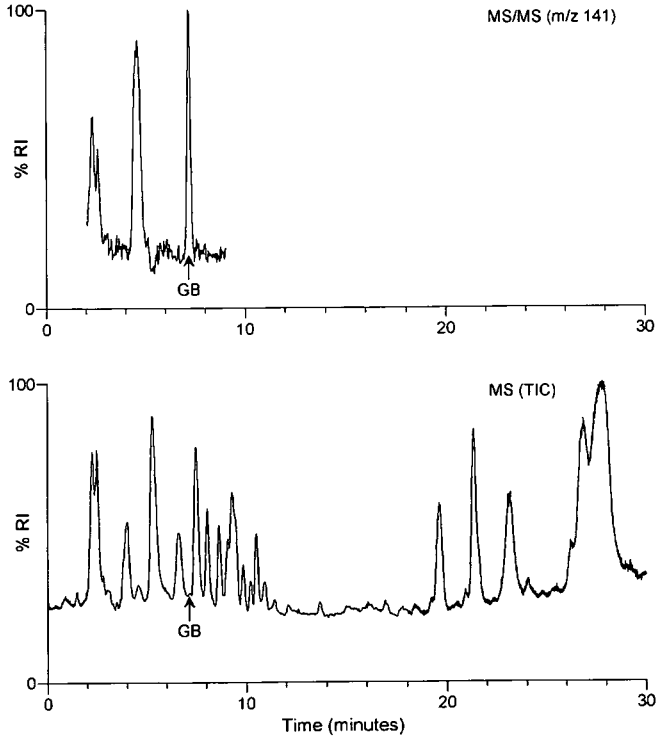

Fig. 3. LC-ESI-MS total-ion-current chromatogram (lower) and LC-ESI-MS/MS chromatogram for the product ions of *m/z* 141 (upper) obtained during analysis of an extract of a office carpet sample spiked at the 4 μg/g level with GB, TEP, GF and GD.

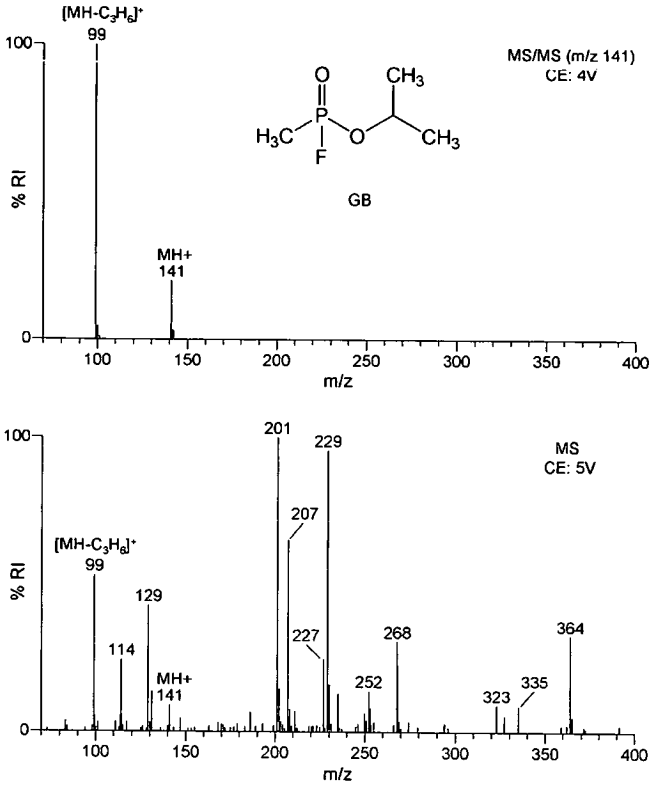

Fig. 4. ESI-MS (lower) and ESI-MS/MS (upper) data obtained at the retention time for GB. Data were obtained during analysis of an extract of an office carpet sample spiked at the 4 μg/g level with GB, TEP, GF and GD (CE: collision energy).

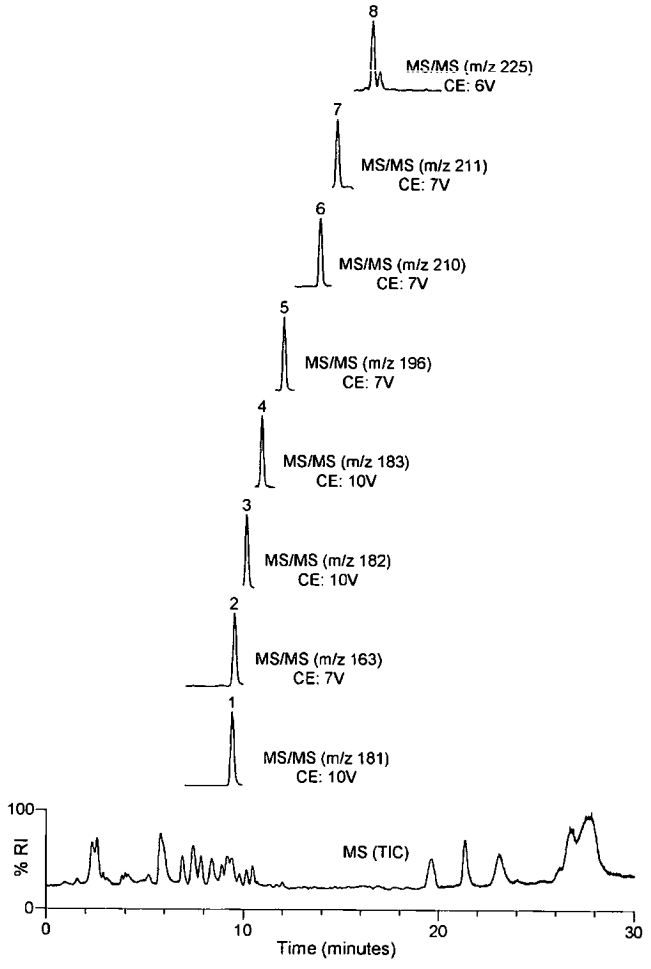

Fig. 5. LC-ESI-MS (lowest) and LC-ESI-MS/MS chromatograms (above) of an extract of an office carpet spiked with munitions grade GA (0.5–5 μg/g per component). Components 1–8 identified in Table 2 (CE: collision energy).

The selectivity afforded by ESI-MS/MS was evident following comparison of the mass spectra acquired during LC-ESI-MS and LC-ESI-MS/MS analysis of the spiked office carpet extract (Fig. 4). During ESI-MS analysis, the characteristic ions for GB, at *m/z* 141 and *m/z* 99, due to $[M+H]^+$ and $[M+H\text{-}C_3H_6]^+$, respectively, were much less significant than the background ions. However the ESI-MS/MS mass spectrum for GB (collision energy 4 V) contained only a precursor ion at *m/z* 141, due to $[M+H]^+$, and a product ion at *m/z* 99, due to loss of the alkene associated with the alkoxy group. Similar product ion mass spectra for the $[M+H]^+$ precursor ion of GF and GD were acquired with collision energies of 4 and 2 V, respectively. Like GB, a common product ion was observed at *m/z* 99, due to loss of the alkene associated with the alkoxy group of GF and GD. GD also exhibited an additional product ion at *m/z* 85 due to $[C_6H_{13}]^+$ and an ion at *m/z* 117 due to $[m/z\ 99+H_2O]^+$. The $[M+H]^+$ precursor ion for TEP produced three product ions at *m/z* 155, *m/z* 127 and *m/z* 99, due to loss(es) of ethylene from the ethoxy substituents. Higher collision energies (8–10 V) resulted in the best product ion mass spectra for TEP identification purposes.

Terrorist use of chemical warfare agents may involve the use of crude or munitions grade chemical warfare agent that contains not only the chemical warfare agent but also related co-synthetic, degradation or other products. Extraction and identification of these additional sample components could be helpful in establishing a link between the agent used in the incident and a source, or provide an indication of synthetic route used to prepare the chemical warfare agent. A munitions grade sample of GA containing numerous related compounds [43] was selected to evaluate the applicability of the developed approach for the identification and characterization of related organophosphorus compounds. Two office media, office carpet and photocopy paper, were spiked with a munitions grade GA standard at the μg/g level (approximately 0.5–5 μg/g per sample component), extracted and analysed by LC-ESI-MS and LC-ESI-MS/MS. Recoveries ranged between 65 and 92% for GA and seven related organophosphorus compounds in the aqueous extracts, with GA being recovered with 65% efficiency from the photocopy paper and 75% efficiency from the office carpet. Some hydrolysis products of GA were observed in the photocopy paper extracts, consistent with earlier spiking results with GB, GF and GD with this medium.

Fig. 5 illustrates LC-ESI-MS and LC-ESI-MS/MS chromatograms obtained during an analysis of aqueous extract of the office carpet spiked with munitions grade GA. Both ESI-MS and ESI-MS/MS data were obtained for each sample component, with the ESI-MS/MS data being acquired at collision energies that resulted in product ion mass spectra containing both the precursor $[M+H]^+$ ion and abundant, structurally informative

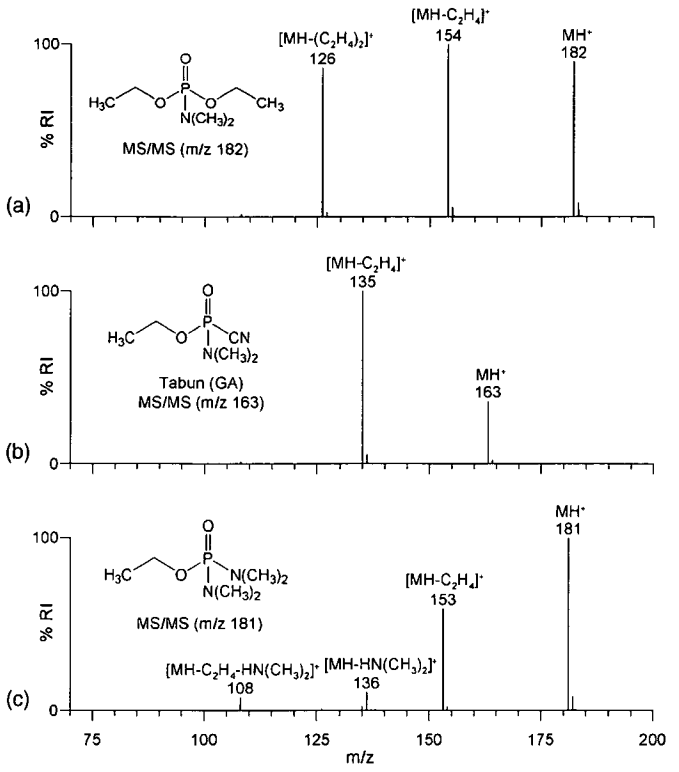

Fig. 6. Product ion mass spectra obtained for (a) diethyl dimethylphosphoramidate (*m/z* 182, collision energy: 10 V), (b) GA (*m/z* 163, collision energy: 7 V) and (c) ethyl tetramethylphosphoramidate (*m/z* 181, Collision energy: 10 V) during LC-ESI-MS/MS analysis of an office carpet sample spiked with munitions grade GA (0.5–5 μg/g per component).

product ions. Fig. 6 illustrates typical product ion mass spectra for tabun and two other related organophosphorus sample components. High resolution data acquired for these and the other related compounds were acquired for identification purposes and have been compiled in Table 2. Errors associated with the mass measurement of the ions were generally less than 0.001 Da, supporting the proposed identities.

The developed method involving aqueous extraction followed by LC-ESI-MS and LC-ESI-MS/MS analysis was successfully applied to the analysis of six different indoor office media contaminated with common chemical warfare agents or a complex munitions grade GA sample. In all cases the spiked components were readily identified in the extracts on the basis of acquired high resolution ESI-MS and/or ESI-MS/MS data, making this method appropriate for these types of sample media and likely applicable to other media that could be collected in support of forensic investigations.

3.2. DESI-MS analysis

Sampling with SPME fibers has received increased attention within the defence and homeland security communities since sampling of contaminated media may be conducted without the need for sample extraction or other steps normally associated with GC–MS or LC–MS analyses. SPME fibers contaminated with common chemical warfare agents would typically be analysed by GC–MS, an approach that has been successfully demonstrated in both laboratory and field settings [45]. Recently, Cooks' group described a novel mass spectrometric for direct sample ionization and analysis of surfaces and referred to this method as desorption electrospray ionization (DESI) [40]. Analysis of SPME fibers using this technique seemed a natural extension of this novel analytical approach, with potential application within the forensic, defence and homeland security communities.

SPME sampling followed by direct analysis of fibers by DESI-MS and DESI-MS/MS was first applied to the sampling and analysis of the headspace above office media spiked with TEP [41,42]. The acquired DESI-MS/MS data for TEP was identical to that obtained by LC-ESI-MS/MS, with the product mass spectrum containing the precusor ion at *m/z* 183, due to $[M+H]^+$, and product ions due to loss of the C_2H_4 from the three ethoxy groups associated with TEP.

Modification of the Z-spray interface sleeve enabled safe DESI-MS/MS analysis of SPME fibers contaminated with the chemical warfare agents GB and GD. Investigations are ongoing, but presentation of the first application of DESI-MS/MS to SPME analysis seemed appropriate as this method compliments and could in some cases replace LC-ESI-MS methods. Preliminary data have been obtained for the DESI-MS/MS analysis of a SPME fiber used to sample the headspace above office carpet spiked at the 20 μg/g level with GB or GD. Fig. 7 illustrates the positioning of the SPME fiber in the Z-spray interface during a typical analysis and Fig. 8 illustrates typical DESI-MS/MS data obtained for a SPME fiber used to sample the headspace above office carpet spiked with GD. Recovery data was not estimated but sufficient GD was sampled onto the fiber in 3 min. to permit acquisition of a full product mass spectrum with a S/N ratio exceeding 20:1 in the total-ion-current chromatogram. High resolution data were acquired for the precusor ion at *m/z* 183 and for the product ions observed at *m/z* 85, *m/z* 99 and *m/z* 117, due to $[C_6H_{13}]^+$, $[M+H\text{-}C_6H_{12}]^+$ and the water adduct of *m/z* 99, respectively. Errors associated these mass measurements were similar to previous LC-ESI-MS/MS data with errors of 0.0007, 0.0007, 0.0018 and 0.0010 Da being observed for *m/z* 183, *m/z* 117, *m/z* 99 and *m/z* 85, respectively.

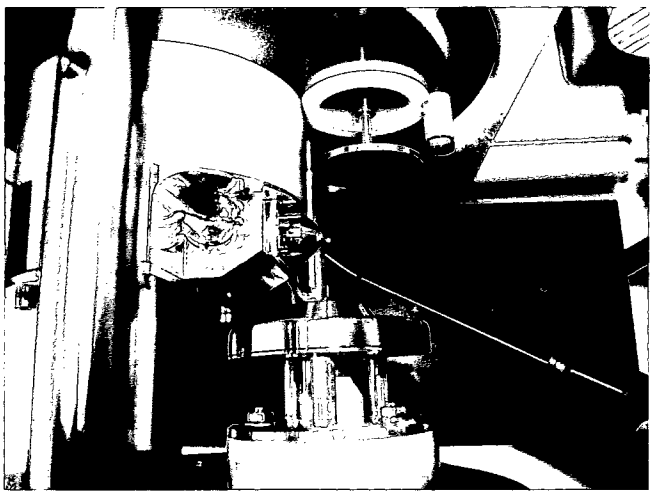

Fig. 7. Top view of SPME fiber in the Z-spray interface.

Table 2
ESI-MS/MS data acquired for significant munitions grade tabun components identified in a spiked office carpet extract

Peak number[a]	Compound name	Ion	Observed mass (Da)[b]	Theoretical mass (Da)	Error (Da)
1	Ethyl tetramethylphosphorodiamidate	MH^+	181.1108	181.1106	0.0002
		$[MH-C_2H_4]^+$	153.0795	153.0793	0.0002
		$[MH-HN(CH_3)_2]^+$	136.0533	136.0527	0.0006
		$[MH-C_2H_4-HN(CH_3)_2]^+$	108.0215	108.0214	0.0001
2	Ethyl dimethylphosphoramidocyanidate (Tabun, GA)	MH^+	163.0628	163.0636	0.0008
		$[MH-C_2H_4]^+$	135.0316	135.0323	0.0007
3	Diethyl dimethylphosphoramidate	MH^+	182.0950	182.0946	0.0024
		$[MH-C_2H_4]^+$	154.0637	154.0633	0.0004
		$[MH-(C_2H_4)_2]^+$	126.0322	126.0320	0.0002
4	Triethyl phosphate	MH^+	183.0805	183.0786	0.0019
		$[MH-C_2H_4]^+$	155.0470	155.0473	0.0003
		$[MH-(C_2H_4)_2]^+$	127.0153	127.0160	0.0007
		$[MH-(C_2H_4)_3]^+$	98.9836	98.9847	0.0011
5	Ethyl isopropyl dimethylphosphoramidate	MH^+	196.1109	196.1102	0.0007
		$[MH-C_3H_6]^+$	154.0630	154.0633	0.0003
		$[MH-C_3H_6-C_2H_4]^+$	126.0327	126.0320	0.0007
6	Diisopropyl dimethylphosphoramidate	MH^+	210.1282	210.1259	0.0023
		$[MH-C_3H_6]^+$	168.0790	168.0789	0.0001
		$[MH-(C_3H_6)_2]^+$	126.0316	126.0320	0.0004
7	Diisopropyl ethyl phosphate	MH^+	211.1109	211.1099	0.0010
		$[MH-C_3H_6]^+$	169.0647	169.0629	0.0018
		$[MH-(C_3H_6)_2]^+$	127.0172	127.0160	0.0012
8	Triisopropyl phosphate	MH^+	225.1273	225.1255	0.0018
		$[MH-C_3H_6]^+$	183.0791	183.0786	0.0005
		$[MH-(C_3H_6)_2]^+$	141.0324	141.0316	0.0008
		$[MH-(C_3H_6)_3]^+$	98.9842	98.9847	0.0005

[a] Refer to Fig. 6.
[b] Average of scans across the chromatographic peak (lock mass used).

SPME headspace sampling followed by DESI-MS/MS analysis reduces the sample preparation and analysis time over aqueous extraction and LC-ESI-MS or LC-ESI-MS/MS analysis and might be a good approach for rapid screening of a larger number of samples. However, as with any method not involving a chromatographic step, chemical interferences and ion suppression may be an issue during analysis. Future DESI-MS/MS experiments will include more complex samples, such as the munitions grade GA sample, to study these issues.

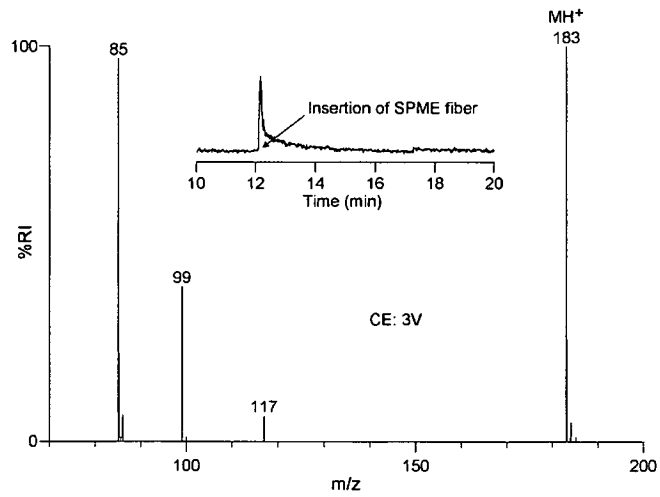

Fig. 8. DESI MS/MS product mass spectrum of GD (*m/z* 183) obtained from a SPME fiber exposed to office carpet spiked at the 20 μg/g level (CE: collision energy). Chromatogram obtained during DESI-MS/MS analysis (inset).

4. Conclusions

An analytical method involving aqueous ultrasonic extraction followed by LC-ESI-MS and LC-ESI-MS/MS analysis was developed and applied to the analysis of six different spiked office sample media that might be collected as part of a indoor forensic investigation where chemical warfare agent use was suspected. Aqueous extraction resulted in co-extraction of other chemicals from the office media samples, but was not destructive to any of the sample media studied. Recovery efficiencies from the six different media studied were generally in the 50–85% range, more than sufficient for the acquisition of full scanning ESI-MS and ESI-MS/MS high resolution data that could be used to confirm identification. In some instances the aqueous extracts contained numerous co-extracted sample components that complicated LC-ESI-MS analysis and hampered identification. These interferences were minimized during LC-ES-MS/MS analysis, where each of the chemical warfare agents was identified on the basis of acquired product ion mass spectra. MS data for all the spiked compounds in the nerve agent

standard and the munitions grade GA were acquired in the continuum mode with a resolution of 9000, which typically resulted in mass measurement errors of 0.001 Da or less.

A recently described method, DESI-MS, was used for the first time for the direct analysis of SPME fibers exposed to office media contaminated with TEP and the chemical warfare agents GB and GD. High resolution DESI-MS/MS data were obtained for all three spiked compounds following SPME sampling of the headspace above office carpet samples spiked at the μg/g level. Application of both the DESI-MS/MS and LC-ESI-MS/MS sample handling and analysis methodology is anticipated during future forensic investigations where evidence of chemical warfare agent use is required for criminal prosecution or to assess remediation/restoration efforts following a chemical incident.

References

[1] Z. Witkiewicz, M. Mazurek, J. Szulc, J. Chromatogr. 503 (1990) 293.
[2] A.F. Kingery, H.E. Allen, Toxicol. Environ. Chem. 47 (1995) 155.
[3] Ch.E. Kientz, J. Chromatogr. A 814 (1998) 1.
[4] D. Noort, H.P. Benschop, R.M. Black, Toxicol. Appl. Pharm. 184 (2002) 116.
[5] E.W.J. Hooijschuur, C.E. Kientz, U.A.T. Brinkman, J. Chromatogr. A 982 (2002) 177.
[6] R.M. Black, B. Muir, J. Chromatogr. A 1000 (2003) 253.
[7] M. Mesilaakso (Ed.), Chemical Weapons Convention Chemicals Analysis. Sample Collection, Preparation and Analytical Methods, Wiley, Chichester, 2005.
[8] J.A. Tørnes, A.M. Opstad, B.A. Johnsen, Int. J. Environ. Anal. Chem. 44 (1991) 227.
[9] R.M. Black, R.J. Clarke, R.W. Read, M.T.J. Reid, J. Chromatogr. A 662 (1994) 301.
[10] E.W.J. Hooijschuur, A.G. Hulst, A.L. de Jong, L.P. de Reuver, S.H. van Krimpen, B.L.M. van Baar, E.R.J. Wils, C.E. Kientz, U.A.T. Brinkman, TRAC-Trend. Anal. Chem. 21 (2002) 116.
[11] M. Noami, M. Kataoka, Y. Seto, Anal. Chem. 74 (2002) 4709.
[12] G.A. Sega, B.A. Tomkins, W.H. Griest, J. Chromatogr. A 790 (1997) 143.
[13] D.K. Rohrbaugh, E.W. Sarver, J. Chromatogr. A 809 (1998) 141.
[14] M. Kataoka, K. Tsuge, Y. Seto, J. Chromatogr. A 891 (2000) 295.
[15] B.A. Tomkins, G.A. Sega, J. Chromatogr. A 911 (2001) 85.
[16] J.R. Hancock, J.M. McAndless, R.P. Hicken, J. Chromatogr. Sci. 29 (1991) 40.
[17] I.N. Stan'kov, A.A. Sergeeva, V.B. Sitnikov, I.D. Derevyagina, O.T. Morozova, S.N. Mylova, V.B. Forov, J. Anal. Chem. 59 (2004) 447.
[18] M. Mazurek, Z. Witkiewicz, S. Popiel, M. Sliwakowski, J. Chromatogr. A 919 (2001) 133.
[19] P.A. D'Agostino, L.R. Provost, J. Chromatogr. 598 (1992) 89.
[20] W.R. Creasy, J.R. Stuff, B. Williams, K. Morrissey, J. Mays, R. Duevel, H.D. Durst, J. Chromatogr. A 774 (1997) 253.
[21] J.R. Stuff, R.L. Cheicante, K.M. Morrissey, H.D. Durst, J. Microcolumn Sep. 12 (2000) 87.
[22] H.-A. Lakso, W.F. Ng, Anal. Chem. 69 (1997) 1866.
[23] M.T. Sng, W.F. Ng, J. Chromatogr. A 832 (1999) 173.
[24] J.F. Schneider, A.S. Boparai, L.L. Reed, J. Chromatogr. Sci. 39 (2001) 420.
[25] G.L. Hook, C. Jackson Lepage, S.I. Miller, P.A. Smith, J. Sep. Sci. 27 (2004) 1017.
[26] G.L. Gresham, G.S. Groenewold, A.D. Appelhans, J.E. Olson, M.T. Benson, M.T. Jeffery, B. Rowland, M.A. Weibel, Int. J. Mass Spectrom. 208 (2001) 135.
[27] P.A. D'Agostino, L.R. Provost, J.R. Hancock, J. Chromatogr. A 808 (1998) 177.
[28] R.W. Read, R.M. Black, J. Chromatogr. A 862 (1999) 169.
[29] J.-P. Mercier, P. Morin, M. Dreux, Chimia 53 (1999) 511.
[30] E.W.J. Hooijschuur, C.E. Kientz, A.G. Hulst, Anal. Chem. 72 (2000) 1199.
[31] P.A. D'Agostino, J.R. Hancock, L.R. Provost, J. Chromatogr. A 912 (2001) 291.
[32] P.A. D'Agostino, C.L. Chenier, J.R. Hancock, J. Chromatogr. A 950 (2002) 149.
[33] P.A. D'Agostino, J.R. Hancock, C.L. Chenier, Eur. J. Mass Spectrom. 9 (2003) 609.
[34] P.A. D'Agostino, J.R. Hancock, C.L. Chenier, J. Chromatogr. A 1058 (2004) 97.
[35] Q. Liu, X.Y. Hu, J.W. Xie, Anal. Chim. Acta 512 (2004) 93.
[36] H.P. Benschop, G.P. van der Schans, D. Noort, A. Fidder, R.H. Mars-Groenendijk, L.P.A. de Jong, J. Anal. Toxicol. 21 (1997) 249.
[37] M. Nagao, T. Takatori, Y. Matsuda, M. Nakajima, H. Iwase, K. Iwadate, Toxicol. Appl. Pharm. 144 (1997) 198.
[38] Y. Matsuda, M. Nagao, T. Takatori, H. Niijima, M. Nakajima, H. Iwase, M. Kobayashi, K. Iwadate, Toxicol. Appl. Pharm. 150 (1998) 310.
[39] P.A. D'Agostino, C.L. Chenier, J.R. Hancock, Analysis of chemical warfare agents in contaminated indoor sample media by high resolution LC-ESI-MS/MS analysis, in: Proceedings of the 17th Sanibel Conference of Mass Spectrometry: Mass Spectrometry in Forensic Science and Counterterrorism, January 28–February 1, 2005, Clearwater Beach, United States.
[40] Z. Takats, J.M. Wiseman, B. Gologan, R.G. Cooks, Science 306 (2004) 471.
[41] J.R. Hancock, P.A. D'Agostino, C.L. Chenier, C.R. Jackson Lepage, Defence against terrorism—the role of liquid chromatography electrospray mass spectrometry in the analysis of chemical warfare agents, in: Proceedings of the 3rd Conference on Mass Spectrometry Applied to Chemical and Biological Warfare Agents, April 17–20, 2005, Noordwijkerhout, The Netherlands.
[42] P.A. D'Agostino, C.L. Chenier, J.R. Hancock, Development of high resolution LC-ESI-MS/MS methodology for the determination of chemical warfare agents and related compounds in an office environment, in: Proceedings of the 53th Annual Conference on Mass Spectrometry and Allied Topics, June 5–9, 2005, San Antonio, Texas, United States.
[43] P.A. D'Agostino, J.R. Hancock, L.R. Provost, Adv. Mass Spectrom. 15 (2001) 297.
[44] P.A. D'Agostino, J.R. Hancock, L.R. Provost, J. Chromatogr. A 840 (1999) 289.
[45] J. Pawliszyn, Sampling and sample preparation for field and laboratory, Elsevier, Amsterdam, 2002.

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