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Capillary column gas chromatography–mass spectrometry and gas chromatography–tandem mass spectrometry detection of chemical warfare agents in a complex airborne matrix

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SUMMARY

The chemical warfare agents sarin, soman and mustard were detected and confirmed during full-scanning gas chromatography (GC)–mass spectrometry (MS) at the nanogram level in spiked extracts of a diesel exhaust environment sampled onto the charcoal of a Canadian C2 respirator canister. This matrix, typical of what might be expected under battlefield conditions, was used for the development of a GC–MS–MS method for the verification of trace levels of sarin, soman and mustard. Chemical interferences associated with this complex sample were virtually eliminated and low-picogram GC–MS–MS detection limits were estimated for these chemical warfare agents in the presence of numerous interfering diesel exhaust and charcoal bed components.

INTRODUCTION

Chemical weapons use, although prohibited by the 1925 Geneva Protocol, has been documented during several armed conflicts, including the Iran/Iraq war[1–4]. Verification of chemical agent use has often been difficult, due in part to inadequate battlefield sampling and identification procedures. Capillary column gas chromatography (GC)–flame ionization detection (FID) may be used for the routine screening of samples for the presence of chemical warfare agents[5,6]. However, it is generally agreed that confirmation of the chemical warfare agents or their degradation products requires identification by mass spectrometry (MS). Electron impact (EI), the traditional MS method of ionization, has gained wide acceptance for the verification of chemical warfare agents, as the EI mass spectra of many organophosphorus[7–12] and

sulfur vesicant[13–17] chemical warfare agents, their decomposition products and related compounds have been published. Comparison of acquired mass spectra with published data, along with supporting chromatographic and/or other spectroscopic data meets suggested verification requirements[18].

The availability of commercial tandem mass spectrometry (MS–MS) systems with triple quadrupole or hybrid (*e.g.*, sector/quadrupole) design has provided researchers with the opportunity to confirm the presence of "target" compounds in a highly specific manner without the need for extensive sample handling. Tandem mass spectrometers offer a number of specific scan functions including parent ion, daughter ion, constant neutral loss and reaction ion monitoring. During reaction ion monitoring, the method of choice for many trace "target" compound applications, the first mass analyser is tuned to allow a desired mass (*e.g.*, $M^{+\bullet}$) into the collisional activated dissociation (CAD) cell while the second mass analyser allows only characteristic ion(s) derived from fragmentation(s) of the ion selected by the first analyser to be detected. The two degrees of selectivity offered by the MS–MS instrument are further enhanced by the use of gas chromatographic sample introduction.

MS–MS has been reviewed recently[19–22], and methodology has been reported for selected organophosphorus pesticides[23–25] and organophosphorus nerve agent standards[26]. Although MS–MS has been suggested as a possible means of chemical warfare agent verification in complex environmental samples[26], there have been no reports of development and application of GC–MS–MS methodology for this purpose.

A capillary column GC study using FID, EI-MS and MS–MS detection was initiated with the principal objective being the development and evaluation of these methods for the detection and confirmation of sarin (isopropyl methylphosphonofluoridate), soman (pinacolyl methylphosphonofluoridate), and mustard [bis(2-chloroethyl)sulfide] in a complex airborne matrix. The air sampled during this study contained the volatile components of diesel exhaust and was very similar in composition to battlefield air sampled onto charcoal during a recent interlaboratory analytical exercise[27]. Charcoal from exposed C2 Canadian respirator canisters was solvent extracted and spiked at several levels to allow evaluation of these analytical methods for the trace detection of the sarin, soman and mustard.

Capillary column GC–FID was of little utility due to the complexity of the sample extract. Sarin, soman and mustard could be detected and confirmed during full-scanning GC–MS at nanogram levels in spiked extracts of the diesel exhaust environment sampled onto the C2 canister charcoal. This airborne matrix, being the most complex of those sampled, was used in the development of a GC–MS–MS approach for the identification of sarin, soman and mustard. Chemical interferences were virtually eliminated and low picogram GC–MS–MS detection limits were estimated for these chemical warfare agents in the presence of numerous interfering diesel exhaust components.

EXPERIMENTAL

Standards

Sarin (GB), soman (GD) and mustard (H) were provided by our Organic

Chemistry Laboratory. Distilled-in-glass dichloromethane was purchased from BDH (Edmonton, Canada). All samples and standards were stored in PTFE-lined screw-capped vials at 4°C prior to GC analysis.

Sample collection and handling

Air from a diesel exhaust environment was sampled through a Canadian C2 charcoal canister for 4 h at the typical working respiratory rate of 20 l/min. The canister charcoal (108 g) was Soxhlet extracted for 6 h with 250 ml of dichloromethane and concentrated to 10 ml under a gentle stream of nitrogen. Portions of the extract were then spiked at the 50-μg/ml, 5-μg/ml and 500-ng/ml levels with GB, GD and H. Extraction efficiencies were evaluated prior to spiking and found to be in the 10–15% range for GB and GD and 70% for H (based on the extraction of 100 μg of agent spiked onto 10 g of blank charcoal).

Instrumental

GC injection volumes of 0.4 and 1 μl, equivalent to $2 \cdot 10^{-4}$ and $5 \cdot 10^{-4}$ m^3 of air sampled on the charcoal of C2 canisters respectively, were used for all GC analyses of the spiked and unspiked charcoal extracts.

Capillary column GC–FID analyses were performed with a Hewlett-Packard 5890 gas chromatograph equipped with an on-column injector of our own design[5]. A 15 m × 0.32 mm I.D. J&W DB-5 (0.25 μm) capillary column was used for all analyses with the following temperature programme: 40°C (2 min), then 10°C/min to 280°C (5 min).

Capillary column GC–MS analyses were performed at Defence Research Establishment Suffield with a VG 70/70E double-focusing mass spectrometer (VG Analytical, Wythenshawe, U.K.) interfaced to a Varian 3700 gas chromatograph under chromatographic conditions identical to those employed during GC–FID analysis. EI-MS operating conditions were as follows: accelerating voltage, 6 kV; emission, 100 μA; electron energy, 70 eV; source temperature, 200°C; resolution (10% valley definition), 1000; and scan function, 400 to 35 u at 1 s/decade.

Capillary column GC–MS–MS analyses were performed at the Institute of Sedimentary and Petroleum Geology with a VG 70/70SQ hybrid tandem mass spectrometer equipped with a Hewlett-Packard 5890 gas chromatograph. All injections were on-column at 40 or 50°C using a Hewlett-Packard injector. The 15 m × 0.32 mm I.D. J&W DB-5 capillary column was held at this temperature for 2 min and then programmed at 10°C/min to a maximum of 280°C. EI-MS conditions were identical to those employed during GC–MS analysis with the exception of source temperature (250°C) and accelerating voltage (8 kV). The daughter spectrum of m/z 158 for H was obtained under the following conditions: CAD cell, 58 eV (laboratory scale)/air ($5 \cdot 10^{-7}$ Torr) and, quadrupole scan function, 200 to 40 u at 0.5 s/decade. Reaction ion monitoring for H was carried out on the m/z 158 to m/z 109 and m/z 158 to m/z 96 transitions with a 80-ms dwell time and a 20-ms delay. GB and GD daughter spectra were obtained for m/z 99 under similar conditions: CAD cell, 50 eV (laboratory scale)/air ($5 \cdot 10^{-7}$ Torr) and, a quadrupole scan function, 200 to 40 u at 0.5 s/decade. Reaction ion monitoring for GB and GD was carried out on the m/z 99 to m/z 79 transition with a 80-ms dwell time and a 20-ms delay.

RESULTS AND DISCUSSION

The diesel exhaust environment sampled onto Canadian C2 charcoal canisters contained primarily hydrocarbon compounds (Table I) and was similar in composition to the volatile battlefield components extracted from a respirator canister circulated as part of a recent interlaboratory analytical exercise. Charcoal extracts used in this study were however further complicated by the presence of silicon-containing compounds adsorbed onto the charcoal bed of the Canadian C2 respiratory canisters. The development of suitable confirmation methods for chemical warfare agents adsorbed onto charcoal under realistic conditions would be valuable in a chemical weapons convention verification role as charcoal mask canisters represent a possible retrospective sampling device.

TABLE I

MAJOR SAMPLE COMPONENTS IDENTIFIED IN CHARCOAL AIRBORNE SAMPLE EXTRACTS

Chromatogram peak No. (Figs. 1 and 2)	*Molecular weight*	*Compound*
1	128	*n*-C_9 alkane
2	142	*n*-C_{10} alkane
3, 5, 8, 11, 13		Charcoal impurity
4	156	*n*-C_{11} alkane
6	170	*n*-C_{12} alkane
7	184	*n*-C_{13} alkane
9	198	*n*-C_{14} alkane
10	212	*n*-C_{15} alkane
12	226	*n*-C_{16} alkane
14	240	*n*-C_{17} alkane
15	254	*n*-C_{18} alkane

Capillary column GC–FID

The chemical warfare agents GB, GD (two chromatographic components due to diastereoisomeric pairs) and H were easily detected in a standard solution at the 20-ng level during GC–FID analysis (Fig. 1a). However, GD and H were not detected by GC–FID in the presence of the charcoal extract components at the 20-ng level. Only GB was detected at this level, since it eluted prior to most of the sample extract components (Fig. 1c). Clearly, a more specific chromatographic detector, such as a flame photometric detector, would be more suitable for sample screening and tentative identification of these chemical warfare agents[28,29] in the presence of this matrix (Fig. 1b).

Capillary column GC–EI-MS

Fig. 2 illustrates capillary column GC–MS chromatograms obtained for the charcoal extract (Fig. 2a) and chemical warfare agent spikes of this extract at the 20-ng (Fig. 2b) and 2-ng (Fig. 2c) levels. Recognizable full scanning EI mass spectra were

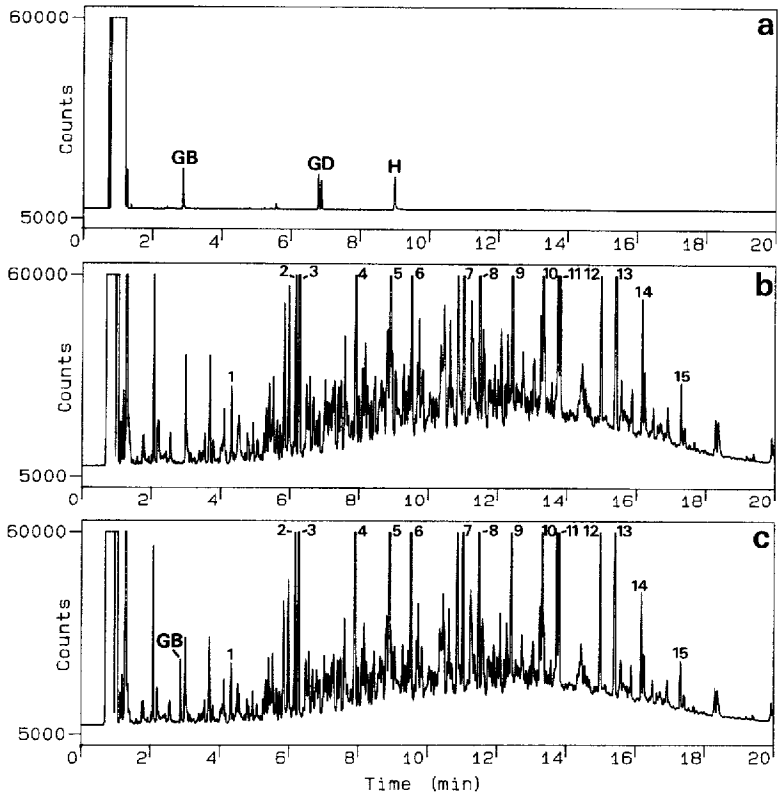

Fig. 1. Capillary column GC–FID chromatograms of (a) 20 ng sarin (GB), soman (GD) and mustard (H), (b) dichloromethane extract of the equivalent of $2 \cdot 10^{-4}$ m^3 of air sampled onto the charcoal of a C2 canister and (c) the previous sample spiked with 20 ng of GB, GD and H. Principal airborne extract sample components are identified in Table I. Column: 15 m × 0.32 mm I.D. J&W DB-5; temperature programme: 40°C (2 min), 10°C/min to 280°C (5 min).

only possible at the 20-ng level for all the spiked chemical warfare agents due to the complexity of this airborne extract.

Amounts of 200–500 pg of chemical warfare agent standard were routinely detected during full scanning operation, but the presence of the diesel exhaust components severely hampered trace confirmation of the chemical warfare agents. Selected ion monitoring under EI conditions typically improves sensitivity by about two orders of magnitude over full scanning so that low-picogram levels may be verified. However, this technique was not applicable at a resolution of 1–2000 due to chemical noise. For this reason no detection limits were estimated. Higher-resolution (*e.g.*, 10 000) selected ion monitoring under capillary column GC–MS conditions, while not readily achieved on our instrument, may reduce the matrix chemical noise so that lower levels of the chemical warfare agents may be confirmed during capillary column GC–EI-MS analysis. This approach to the reduction of chemical noise has been used for the confirmation of compounds such as dioxins[30]. However, recent reports of interferences even at high resolution in some sample matrices suggest that an alternative approach such as MS–MS be considered[30].

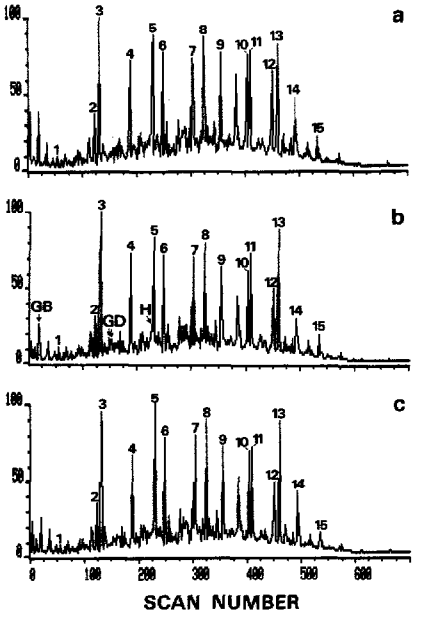

Fig. 2. Capillary column GC–EI-MS chromatograms of (a) dichloromethane extract of the equivalent of $2 \cdot 10^{-4}$ m^3 of air sampled onto the charcoal of a C2 canister, and the previous sample spiked at the (b) 20 ng and (c) 2 ng level with sarin (GB), soman (GD) and mustard (H). Principal airborne extract sample components are identified in Table I. Column: 15 m × 0.32 mm I.D. J&W DB-5; temperature programme: 40°C (2 min), 10°C/min, 280°C (5 min).

Capillary column GC–MS–MS

Hesso and Kostiainen[26] reported the daughter spectra for the pseudo-molecular ions formed during ammonia chemical ionization of GB, GD, tabun and VX. The utility of reaction ion monitoring for the detection of chemical warfare agents in

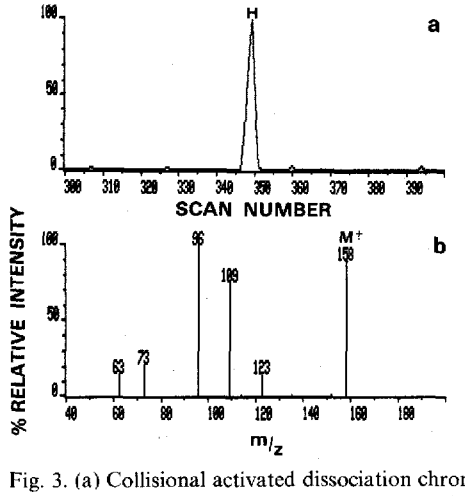

Fig. 3. (a) Collisional activated dissociation chromatogram obtained for the daughters of *m/z* 158 during GC–MS–MS analysis of mustard (H). (b) Daughter spectrum of H. Column: 15 m × 0.32 mm I.D., J&W DB-5; temperature programme: 50°C (2 min), 10°C/min, 280°C (5 min).

a complex or environmental matrix, while mentioned, was not demonstrated. The inability of conventional capillary column GC–EI-MS to confirm trace levels of chemical warfare agents in a real environmental matrix prompted investigation into application of MS–MS instrumentation for the trace detection of chemical warfare agents in a complex matrix.

The daughter spectrum of the molecular ion (*m/z* 158) for H was acquired under CAD conditions, which, while perhaps not optimal, did provide significant lower mass ions for use in a reaction ion monitoring experiment. The use of a higher mass ion, such as the molecular or higher mass fragmentation ion, for the acquisition of daughter spectra is usually preferred to minimize potential interferences. Daughter ions of *m/z* 158 at *m/z* 63, 73, 96, 109 and 123 were observed by scanning the quadrupole after CAD at 58 eV using air (Fig. 3). Both the *m/z* 158 to 109 (loss of CH_2Cl) and *m/z* 158 to 96 (loss of C_2H_3Cl) transitions were considered suitable and monitored during reaction ion monitoring of H in the diesel exhaust extract. Fig. 4 illustrates the reaction ion monitoring chromatograms obtained for the *m/z* 158 to 109 transition for 500 pg of H, the charcoal extract, and the charcoal extract spiked with 500 pg of H. H was easily confirmed without any interference in the presence of more than twice as much sample extract (*i.e.*, $5 \cdot 10^{-4}$ m^3 of diesel exhaust air) as was used during GC–FID and GC–EI-MS evaluation. The signal-to-noise ratio for 500 pg of H (greater than 80:1) was independent of the matrix and virtually identical for both the standard (Fig. 4a) and spiked extract (Fig. 4c). A conservative method detection limit of 30 pg (signal-to-noise ratio 5:1) was estimatd for H based on these findings.

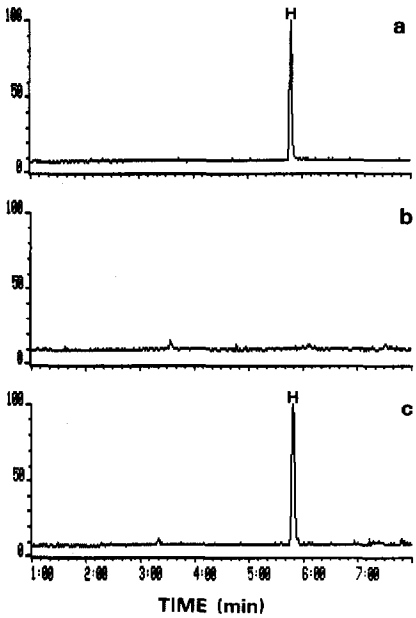

Fig. 4. Reaction ion monitoring chromatogram for *m/z* 158 to *m/z* 109 obtained during GC–MS–MS analysis of (a) 500 pg of mustard (H), (b) dichloromethane extract of the equivalent of $5 \cdot 10^{-4}$ m^3 of air sampled onto the charcoal of a C2 canister and (c) the previous sample spiked with 500 pg of mustard (H). Column: 15 m × 0.32 mm I.D., J&W DB-5; temperature programme: 50°C (2 min), 10°C/min, 280°C (5 min).

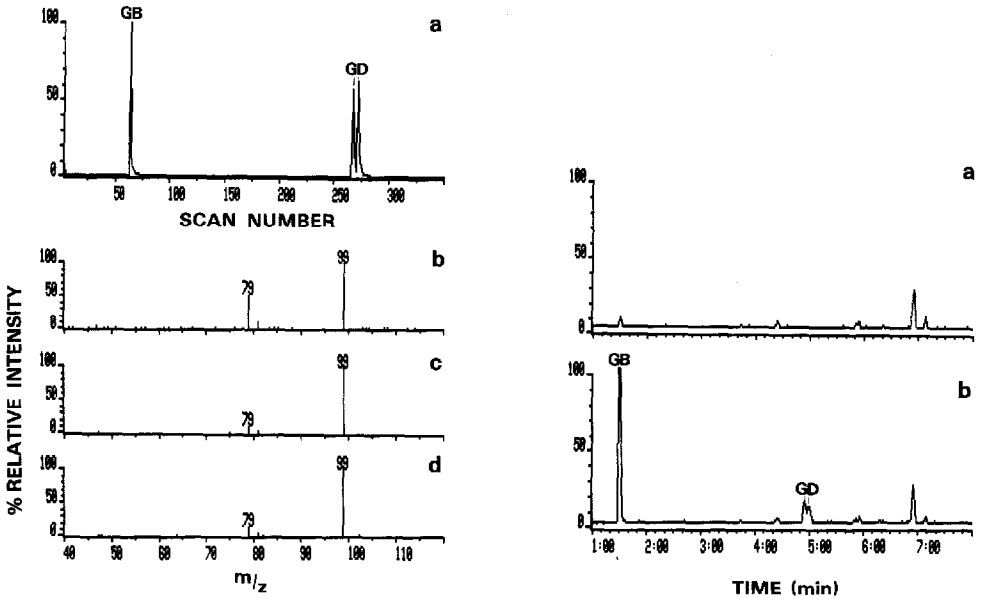

Fig. 5. (a) Collisional activated dissociation chromatogram for the daughters of *m/z* 99 during GC–MS–MS analysis of sarin (GB) and soman (GD). Daughter spectra of GB (b) and chromatographic peaks for GD (c and d). Column: 15 m × 0.32 mm I.D., J&W DB-5; temperature programme: 40°C (2 min), 10°C/min, 280°C (5 min).

Fig. 6. Reaction ion monitoring chromatogram for *m/z* 99 to *m/z* 79 obtained during GC–MS–MS analysis of (a) dichloromethane extract of the equivalent of $5 \cdot 10^{-4}$ m^3 of air sampled onto the charcoal of a C2 canister and (b) the previous sample spiked with 500 pg of sarin (GB) and soman (GD). Column: 15 m × 0.32 mm I.D., J&W DB-5; temperature programme: 40°C (2 min), 10°C/min, 280°C (5 min).

Unlike H, molecular ions are not observed for GD or GB following EI ionization. Both these compounds and other methylphosphonofluoridates do however form a diagnostic EI fragmentation ion at *m/z* 99 due to $[(CH_3)(F)P(OH)_2]^+$. CAD (50 eV in the presence of air) of *m/z* 99 resulted in the detection of a daughter ion at *m/z* 79 for both GB and GD (Fig. 5). Reaction ion monitoring of the *m/z* 99 to 79 transition, due to loss of HF, should be highly specific to methylphosphonofluoridates. Both GB and GD were readily detected at 500 pg in the preseance of $5 \cdot 10^{-4}$ m^3 of diesel exhaust air (Fig. 6). A minor interference (approx. 3% the height of the GB peak) was observed at the retention time of GB, while no interferences were detected at the retention times of the GD peaks. The method detection limit for GB was estimated to be 70 pg in the presence of this interference and 5 pg in matrices that do not contain this interference (signal-to-noise ratio 5:1). A GD method detection limit of 60 pg (signal-to-noise ratio 5:1) was estimated based on these findings.

CONCLUSIONS

The chemical warfare agents sarin, soman and mustard were detected and confirmed during capillary column GC–EI-MS conditions at nanogram levels in spiked extracts of diesel exhaust environment sampled onto the charcoal of Canadian

C2 respiratory canisters. Capillary column GC–FID was of little utility due to the complexity of the sample extract.

Daughter spectra, obtained during capillary column GC–MS–MS of the chemical warfare agents, suggested the use of the m/z 158 to 109 or 96 collisional activated dissociation processes for the detection of H and the m/z 99 to 79 collisional activated dissociation process for the identification of GB and GD. Reaction ion monitoring of these collisional activated processes during GC–MS–MS proved to be the most sensitive of the methods evaluated for the confirmation of the chemical warfare agents GB, GD and H in the presence of components commonly found in an airborne battlefield environment. GC–MS–MS detection limits in the 30–70-pg range were estimated for each of the chemical warfare agents in the presence of sample component concentrations levels two to three orders of magnitude greater than the spiked agents. Application of MS–MS for the detection of chemical warfare agents, or other compounds of chemical defence interest, appears to be an attractive approach for the verification of "target" compounds in complex environmental matrices such as those that may be encountered during airborne sampling of battlefield emissions.

REFERENCES

1 *Report of the Specialists Appointed by the Secretary-General to Investigate Allegations by the Islamic Republic of Iran Concerning the Use of Chemical Weapons, S/16433*, United Nations Security Council, New York, 26 March 1984.
2 *Report of the Mission Dispatched by the Secretary-General to Investigate Allegations of the Use of Chemical Weapons in the Conflict between the Islamic Republic of Iran and Iraq, S/17911*, United Nations Security Council, New York, 12 March 1986.
3 G. Andersson, *NBC Defence and Technology International*, April (1986) 62–65.
4 *Report of the Mission Dispatched by the Secretary-General to Investigate Allegations of the Use of Chemical Weapons in the Conflict between the Islamic Republic of Iran and Iraq, S/20060*, United Nations Security Council, New York, 20 July 1988.
5 P. A. D'Agostino and L. R. Provost, *J. Chromatogr.*, 331 (1985) 47–54.
6 P. A. D'Agostino and L. R. Provost, *J. Chromatogr.*, 436 (1988) 399–411.
7 *Chemical and Instrumental Verification of Organophosphorus Warfare Agents*, Ministry of Foreign Affairs of Finland, Helsinki, 1977.
8 S. Sass and T. L. Fisher, *Org. Mass Spectrom.*, 14 (1979) 257–264.
9 P. A. D'Agostino, A. S. Hansen, P. A. Lockwood and L. R. Provost, *J. Chromatogr.*, 347 (1985) 257–266.
10 E. R. J. Wils and A. G. Hulst, *Org. Mass Spectrom.*, 21 (1986) 763–765.
11 P. A. D'Agostino, L. R. Provost and J. Visentini, *J. Chromatogr.*, 402 (1987) 221–232.
12 P. A. D'Agostino, L. R. Provost and K. M. Looye, *J. Chromatogr.*, 465 (1989) 271–283.
13 *Systematic Identification of Chemical Warfare Agents, B.3, Identification of Non-Phosphorus Warfare Agents*, Ministry of Foreign Affairs of Finland, Helsinki, 1982.
14 E. Ali-Mattila, K. Siivinen, H. Kenttamaa and P. Savolahti, *Int. J. Mass Spectrom. Ion Phys.*, 47 (1983) 371–374.
15 E. R. J. Wils and A. G. Hulst, *Fresenius' Z. Anal. Chem.*, 321 (1985) 471–474.
16 P. A. D'Agostino and L. R. Provost, *Biomed. Environ. Mass Spectrom.*, 15 (1988) 553–564.
17 P. A. D'Agostino, L. R. Provost, A. S. Hansen and G. A. Luoma, *Biomed. Environ. Mass Spectrom.*, 18 (1989) 484–491.
18 *Handbook for the Investigation of Allegations of the Use of Chemical or Biological Weapons*, Department of External Affairs, Ottawa, November 1985.
19 R. G. Cooks and G. L. Glish, *Chem. Eng. News*, Nov. (1981) 40–52.
20 R. W. McLafferty, *Tandem Mass Spectrometry*, Wiley, New York, 1983.
21 J. V. Johnson and R. A. Yost, *Anal. Chem.*, 57 (1985) 758A–768A.
22 G. L. Glish and S. A. McLuckey, *Anal. Instrum.*, 15 (1986) 1–36.

23 S. V. Hummel and R. A. Yost, *Org. Mass Spectrom.*, 21 (1986) 785–791.
24 J. A. Roach and L. J. Carson, *J. Assoc. Off. Anal. Chem.*, 70 (1987) 439–442.
25 T. Cairns and E. G. Seigmund, *J. Assoc. Off. Anal. Chem.*, 70 (1987) 858–862.
26 A. Hesso and R. Kostiainen, *Proc. 2nd Int. Symp. Protection Against Chemical Warfare Agents, Stockholm, Sweden, June 15–19, 1986*, National Defence Research Institute, Umeå, pp. 257–260.
27 J. R. Hancock, P. A. D'Agostino and L. R. Provost, *The Analysis of a Respirator Canister: Fourth International Training Exercise*, Defence Research Establishment Suffield, Medicine Hat, June 1986, internal document (available on request).
28 S. S. Brody and J. E. Chaney, *J. Gas Chromatogr.*, 4 (1966) 42–46.
29 S. Sass and G. A. Parker, *J. Chromatogr.*, 189 (1980) 331–349.
30 D. Fraisse, presented at *International Workshop on Hybrid Tandem Mass Spectrometry, Lake Louise, November 17–19, 1988.*