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Identification of Sarin and Related Compounds in Snow by Packed Capillary Liquid Chromatography-Electrospray Mass Spectrometry (LC-ESI-MS)

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Abstract

Packed capillary LC-ESI-MS was used for the analysis of a snow sample that was accidentally contaminated with an organophosphorus chemical warfare agent during the destruction of a chemical munition. Sarin, its hydrolysis products and a number of unique related compounds were identified on the basis of acquired LC-ESI-MS data. Full mass spectra were acquired for all fourteen compounds detected, with all compounds exhibiting MH^+ , $[MH+ACN]^+$ ions and/or protonated dimers that could be used to confirm molecular mass. Sampling cone voltages from 20 to 70 volts were utilized with the higher sampling voltages enhancing formation of structurally important product ions in the ESI interface. All data were acquired with a time-of-flight (TOF) mass spectrometer with a resolution of 5000 (50% valley definition), a resolution that aided in the assignment of elemental composition of the observed ions. The application of LC-ESI-MS to snow analysis appears to be an attractive alternative to the GC-MS methods, since both chemical warfare agents and their hydrolysis products may be analysed directly, eliminating the need for additional sample handling and derivatization steps.

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Executive summary

Introduction: The Canadian Forces (CF) may be called on to perform peacekeeping or battlefield operations in regions of the world where there is a significant threat of chemical/biological (CB) warfare agent use. To operate effectively in these theatres the CF must be able to identify the CB agent used. Mass spectrometry (MS), is a powerful analytical technique for the identification of both known and unknown compounds and DRE Suffield is currently investigating this instrumental technique in fulfillment of CF detection and identification requirements.

Results: Packed capillary LC-ESI-MS was used for the analysis of a snow sample that was accidentally contaminated with an organophosphorus chemical warfare agent during the destruction of a chemical munition. Sarin, its hydrolysis products and a number of unique related compounds were identified on the basis of acquired LC-ESI-MS data. Full mass spectra were acquired for all fourteen compounds detected, with all compounds exhibiting MH^+ , $[MH+ACN]^+$ ions and/or protonated dimers that could be used to confirm molecular mass. Sampling cone voltages from 20 to 70 volts were utilized with the higher sampling voltages enhancing formation of structurally important product ions in the ESI interface. All data were acquired with a time-of-flight (TOF) mass spectrometer with a resolution of 5000 (50% valley definition), a resolution that aided in the assignment of elemental composition of the observed ions. The application of LC-ESI-MS to snow analysis appears to be an attractive alternative to the GC-MS methods, since both chemical warfare agents and their hydrolysis products may be analysed directly, eliminating the need for additional sample handling steps and derivatization steps.

Significance: The CF may be deployed in regions of the world where there is a significant threat of chemical/biological warfare agent use. Identification of the CB agent is of importance since the results of such analyses would contribute to the development of strategic and political positions regarding future Canadian military operations and would facilitate the dissemination of technical advice to in-theatre field commanders and medical personnel.

Future Plans: The reported method would be valuable for the identification of organophosphorus chemical warfare agents and their hydrolysis products in snow samples collected by the Canadian Forces or in support of Chemical Weapons Convention challenge inspections.

D'Agostino, P.A., Chenier, C.L., Hancock, 2001. Identification of Sarin and Related Compounds in Snow by Packed Capillary LC-ESI-MS. DRES TM 2001-044, Defence Research Establishment Suffield.

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Introduction

Canada has ratified the Chemical Weapons Convention and complies with this treaty by maintaining only small stocks of chemical warfare agents at Defence Research Establishment Suffield (DRES) for defensive research and development. On a periodic basis old chemical shells are discovered on the Suffield Military Training Range and need to be destroyed. The disposal of chemical shells frequently involves perforation of the shell, resulting in a small sampling hole that can be used to access the contents for identification purposes. Recently a 1950's 105 mm chemical shell was split apart by the perforating charge and contaminated the surrounding snow. Chemical Agent Monitor readings in the G-mode (8 bars) above the snow indicated the presence of a nerve agent and a sample of the snow was collected for laboratory analysis to confirm the identity of the chemical warfare agent(s). This collection and analysis operation was typical of what might be expected during retrospective analysis of munition or other samples in support of the Canadian Forces. Analyses of this type requires the use of sensitive, specific analytical methods, particularly when unambiguous proof is required for the presence of chemical warfare agents (1). These analytical demands are being actively addressed by DRES through the development and application of new analytical methods for the detection and identification of chemical warfare agents, their hydrolysis products and related compounds.

Gas chromatography (GC) has been used for the separation and identification of chemical warfare agents, with gas chromatography-mass spectrometry (GC-MS) being used most frequently for the characterization of these compounds (1,2). Organophosphorus chemical warfare agents have been studied extensively by electron impact and chemical ionization mass spectrometry, as the use of these complementary ionization techniques facilitates the acquisition of molecular and fragmentation ion information that may be used for identification (3-10). GC separation, while suitable for the direct analysis of organophosphorus chemical warfare agent in organic extracts, is usually not preferred for the direct analysis of aqueous samples or extracts. Aqueous samples or extracts containing organophosphorus chemical warfare agents and/or their nonvolatile hydrolysis products normally require additional sample handling steps and derivatization prior to analysis (11-13). Water samples containing chemical warfare agents have been analysed by GC-MS following solid-phase microextraction (14) and by microcolumn liquid chromatography (LC) with flame photometric detection (15). Increasingly, researchers have developed LC-MS analytical methods to deal with the analysis of aqueous samples containing these nonvolatile hydrolysis products (16-22).

Atmospheric pressure ionization (e.g., electrospray (ESI), ionspray and atmospheric pressure chemical ionization) techniques (20-27) have been investigated for the analysis of organophosphorus chemical warfare agent hydrolysis products as these techniques may be interfaced to LC for component separation. DRES recently published several LC-ESI-MS papers on the direct determination of chemical warfare agents, their hydrolysis products and related compounds in aqueous samples or extracts during a single analysis (27-30). LC-ESI-MS was an attractive alternative to GC-MS for these analyses, as both the organophosphorus chemical warfare agents and/or their hydrolysis products could be analysed directly without

the need for additional sample handling and derivatization. A similar advantage should be possible during the analysis of contaminated snow samples.

Few reports deal with the analysis of chemical warfare agents and their hydrolysis products from snow samples. The Norwegian Defence Research Establishment conducted a series of experiments in the 1980's involving the analysis of chemical warfare agents, including sarin, soman, tabun and VX, in contaminated snow samples (31-33). Snow samples were melted, extracted with chloroform and analysed by GC-MS for the presence of chemical warfare agent(s). A second extraction and analysis by GC-MS was required to determine the presence of the chemical warfare agent hydrolysis products as their diazomethane derivatives. Chemical warfare agent verification work continues at the Norwegian Defence Research Establishment, with the most recent report in the series containing additional references to prior investigations dealing with water, soil and other media (34).

This paper focuses on the application of a packed capillary LC-ESI-MS method (29,30) for the identification of organophosphorus chemical warfare agents and/or their hydrolysis products observed during analysis of a contaminated snow sample. Sarin (isopropyl methylphosphonofluoridate) and its initial hydrolysis product, isopropyl methylphosphonic acid, were identified as the principal sample components in the melted snow sample using this approach. A number of other previously uncharacterized organophosphorus compounds were also observed during this analysis, the identities of which were determined by interpreting high resolution ESI-MS data collected at several different sampling cone voltages.

Experimental

Samples and sample handling

A sample of the snow from near a split 105 mm chemical shell was collected for laboratory analysis to confirm the identity of the chemical warfare agent(s). The snow sample was allowed to melt and an aliquot was diluted 1:10 with distilled water prior to LC-ESI-MS analysis. The remaining snow sample was refrozen at -20°C. Organophosphorus standards, including sarin, were provided by the Canadian National Single Small Scale Facility at DRES.

Instrumental analysis

ESI-MS data were acquired using a Micromass LCT time-of-flight mass spectrometer equipped with the Z-spray electrospray interface. The electrospray capillary was operated at 3.2 kV with sampling cone voltages in the 20 to 70 volt range. Nitrogen desolvation gas was introduced into the interface (80 °C) at a flow rate of 480 L/h. Nitrogen nebulizer gas was introduced at a flow rate of 66 L/h. ESI-MS data were acquired from 600 to 70 Da (1 sec) in the continuum mode with a resolution of 5000 (50% valley definition).

LC separations were performed using an Applied Biosystems Model 140B dual syringe pump equipped with a Micro-Tech 150 mm × 0.32 mm I.D. C₁₈ (Zorbax, 5 µm) packed fused-silica capillary column and a Rheodyne 8125 injector with a 5 µL sample loop. The following solvent compositions were prepared for LC separation: Solvent A (0.1% trifluoroacetic acid in water) and Solvent B (0.1% trifluoroacetic acid in acetonitrile (ACN)/water, 95:5). Chromatographic separations were performed using a 1% to 75%B gradient program over 30 minutes. In order to minimize dead volume effects and ensure reproducible mixing, the mobile phase was delivered at 200 µL/min and split prior to the injector such that the flow through the column was 16 µL/min.

Results and Discussion

Unexploded chemical shells, from exercises held more than forty years ago, are periodically discovered on the Suffield Military Training Area. Disposal of chemical shells frequently involves perforation of the shell, resulting in a small sampling hole that can be used to access the contents for identification purposes. Recently an old chemical shell was accidentally split apart by the perforating charge and the contents contaminated the surrounding snow. Chemical Agent Monitor readings in the G-mode (8 bars) above the snow indicated the presence of a nerve agent. A sample of the snow was collected for laboratory analysis to confirm the identity of the chemical warfare agent(s).

Figure 1 illustrates the total-ion-current chromatogram acquired for the melted snow sample diluted 1:10 with distilled water. Sarin and its initial hydrolysis product, isopropyl methylphosphonic acid were identified as major components in the snow sample, based on the acquired chromatographic and ESI-MS data (29,30). An additional twelve sample components, many of which have not been previously characterized by LC-ESI-MS, were also detected. ESI-MS data were acquired with a resolution of 5000 (50% valley definition), with all compounds exhibiting MH^+ , $[MH+ACN]^+$, $[MH+HN(CH_3)_2]^+$ ions and/or protonated dimers that were used to confirm molecular mass. Sampling cone voltages from 20 to 70 volts were utilized with the higher sampling voltages enhancing formation of structurally important product ions in the ESI interface. Interpretation of all the acquired high resolution ESI-MS data resulted in the identification or provisional identification of all fourteen compounds listed in Table 1. ESI-MS characterization and identification of these additional sample components will prove valuable during future analyses, since their presence could help verify the prior presence of sarin in collected samples, indicate the synthetic procedure used for munition production or suggest the origin of the munition.

Methylphosphonofluoridic acid, methylphosphonic acid and dimethylphosphinic acid, eluted at or near the dead volume of the LC column and were identified on the basis of MH^+ data acquired using a sampling cone voltage of 20 volts. MH^+ ions were observed at m/z 97.0099, m/z 99.0038 and m/z 95.0247 for the above three compounds, respectively (Table 1). Errors associated with the observed measurements were usually within 0.005 Da of theoretical values, consistent with the errors typically observed during TOF-MS operation at a resolution of 5000 (50% valley definition). Phosphoric acid (theoretical mass of MH^+ ion is 98.9847 Da), an organophosphorus compound with the same nominal molecular mass as methylphosphonofluoridic acid, was not considered as a possible sample component due to the large mass measurement error (0.019 Da) that would be associated with this assignment. Product ions, which have been observed during prior negative ion ESI-MS/MS (24) of phosphoric acid and methylphosphonofluoridic acid, were not significant in this investigation.

Table 1. Compounds identified in the snow sample by high resolution LC-ESI-MS (Sampling cone voltage 20 volts).

Peak # ¹	Compound Name	Ion	Observed Mass (Da) (mean \pm SD; n = 5)	Theoretical Mass (Da)	Average Error (Da)
1	Methylphosphonic acid	MH ⁺	97.0099 \pm 0.0044	97.0055	0.0044
2	Methylphosphonofluoridic acid	MH ⁺	99.0038 \pm 0.0007	99.0011	0.0027
3	Dimethylphosphinic acid ²	MH ⁺	95.0247 \pm 0.0014	95.0262	0.0015
4	Isopropyl methylphosphonic acid	MH ⁺	139.0455 \pm 0.0019	139.0524	0.0069
		[MH-C ₃ H ₆] ⁺	97.0071 \pm 0.0015	97.0055	0.0016
5	Isopropyl dimethylphosphinate ²	MH ⁺	137.0711 \pm 0.0005	137.0731	0.0020
		[MH-C ₃ H ₆] ⁺	95.0311 \pm 0.0010	95.0262	0.0049
6	Sann (Isopropyl methylphosphonofluoridate)	MH ⁺	141.0444 \pm 0.0021	141.0481	0.0037
		[MH-C ₃ H ₆] ⁺	99.0036 \pm 0.0014	99.0011	0.0025
7	Isopropyl methyl methylphosphonate ²	MH ⁺	153.0665 \pm 0.0028	153.0681	0.0016
		[MH-C ₃ H ₆] ⁺	111.0267 \pm 0.0020	111.0211	0.0056
8	Diisopropyl methylphosphonate	MH ⁺	181.1004 \pm 0.0028	181.0994	0.0010
		[MH-C ₃ H ₆] ⁺	139.0547 \pm 0.0019	139.0524	0.0023
9	Pentyl dibutylamine ²	MH ⁺	200.2376 \pm 0.0033	200.2378	0.0002
10	Tributylamine	MH ⁺	186.2221 \pm 0.0026	186.2222	0.0001
11	Diisopropyl chloromethylphosphonate ²	MH ⁺	215.0642 \pm 0.0016	215.0604	0.0038
		[MH-C ₃ H ₆] ⁺	173.0191 \pm 0.0013	173.0134	0.0057
12	Diisopropyl phosphorofluoridate ²	MH ⁺	185.0747 \pm 0.0003	185.0743	0.0004
		[MH-C ₃ H ₆] ⁺	143.0285 \pm 0.0010	143.0273	0.0012
13	Trisopropyl phosphate	MH ⁺	225.1246 \pm 0.0027	225.1256	0.0010
		[MH-C ₃ H ₆] ⁺	183.0814 \pm 0.0005	183.0786	0.0028
14	Diisopropyl dichloromethylphosphonate ²	MH ⁺	249.0216 \pm 0.0006	249.0214	0.0002
		[MH-C ₃ H ₆] ⁺	206.9768 \pm 0.0038	206.9745	0.0023

¹ Refer to Figure 1

² Identification based on interpretation of acquired mass spectrometric data.

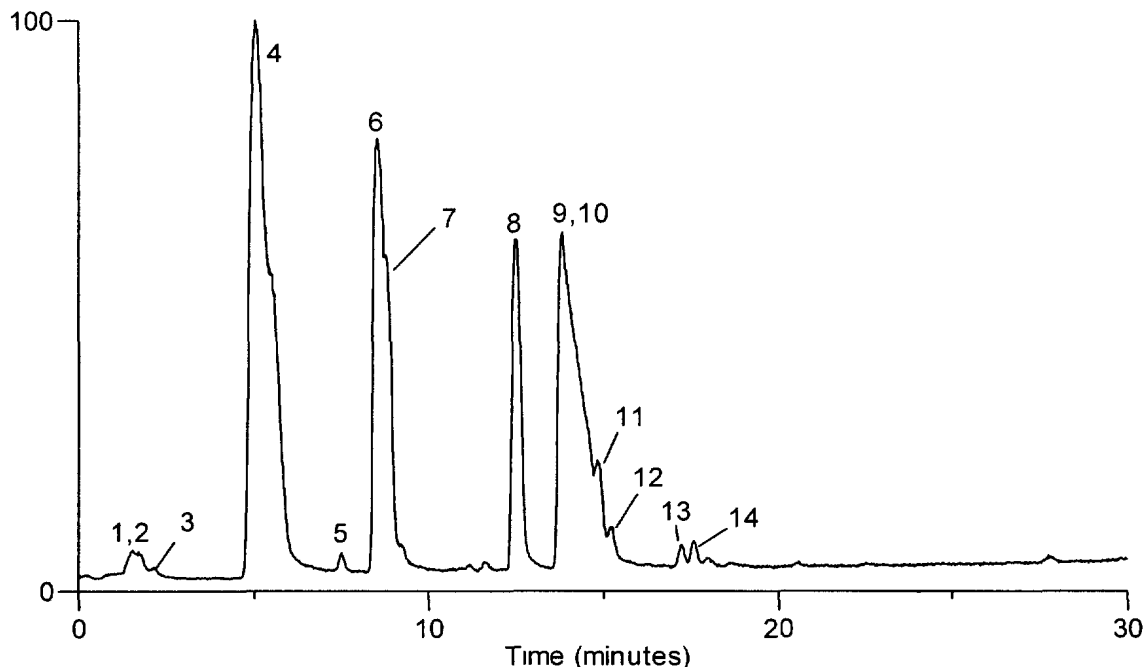


Figure 1. LC-ESI-MS total-ion-current (600 to 85 Da) chromatogram obtained for the snow sample with a sampling cone voltage of 20 volts. Numbered peaks are identified in Table 1.

Figure 2 illustrates typical ESI-MS data acquired for a) isopropyl methylphosphonic acid, b) sarin, c) diisopropyl methylphosphonate and d) triisopropyl phosphate during analysis of the snow sample. All four compounds, characterized during prior LC-ESI-MS analyses of sarin, exhibited MH^+ ions and product ions due to alkene (e.g., C_3H_6) loss from the corresponding alkoxy substituent (e.g., $-OC_3H_6$). Identification was strengthened by comparing the observed masses of the MH^+ and $[MH-C_3H_6]^+$ ions for each compound with theoretical masses (Table 1). Dimers (and a trimer for sarin) were more significant in the acquired mass spectra due to the relatively high concentration of isopropyl methylphosphonic acid, sarin and diisopropyl methylphosphonate. Higher mass adduct ions, due to the presence of acetonitrile and dimethylamine, were also observed 41 Da and 45 Da higher in mass than the protonated adducts (or product ions).

Additional structural information was acquired by promoting product ion formation in the ESI interface. A lower sampling cone voltage of 20 volts resulted in the formation of only one significant product ion, due to loss of C_3H_6 from the MH^+ ion, for both diisopropyl methylphosphonate and triisopropyl phosphate (Figure 2d). Increasing the sampling cone voltage to 30 volts greatly increased product ion formation and resulted in sequential loss of C_3H_6 from the isopropoxy substituents of both compounds. Product ions at m/z 139 and m/z 97 (and their acetonitrile adducts), due to sequential loss of C_3H_6 from the MH^+ ion, were observed for diisopropyl methylphosphonate (Figure 2c). Similar results were obtained for triisopropyl phosphate with product ions (and their acetonitrile adducts) being observed at m/z 183, m/z 141 and m/z 99.

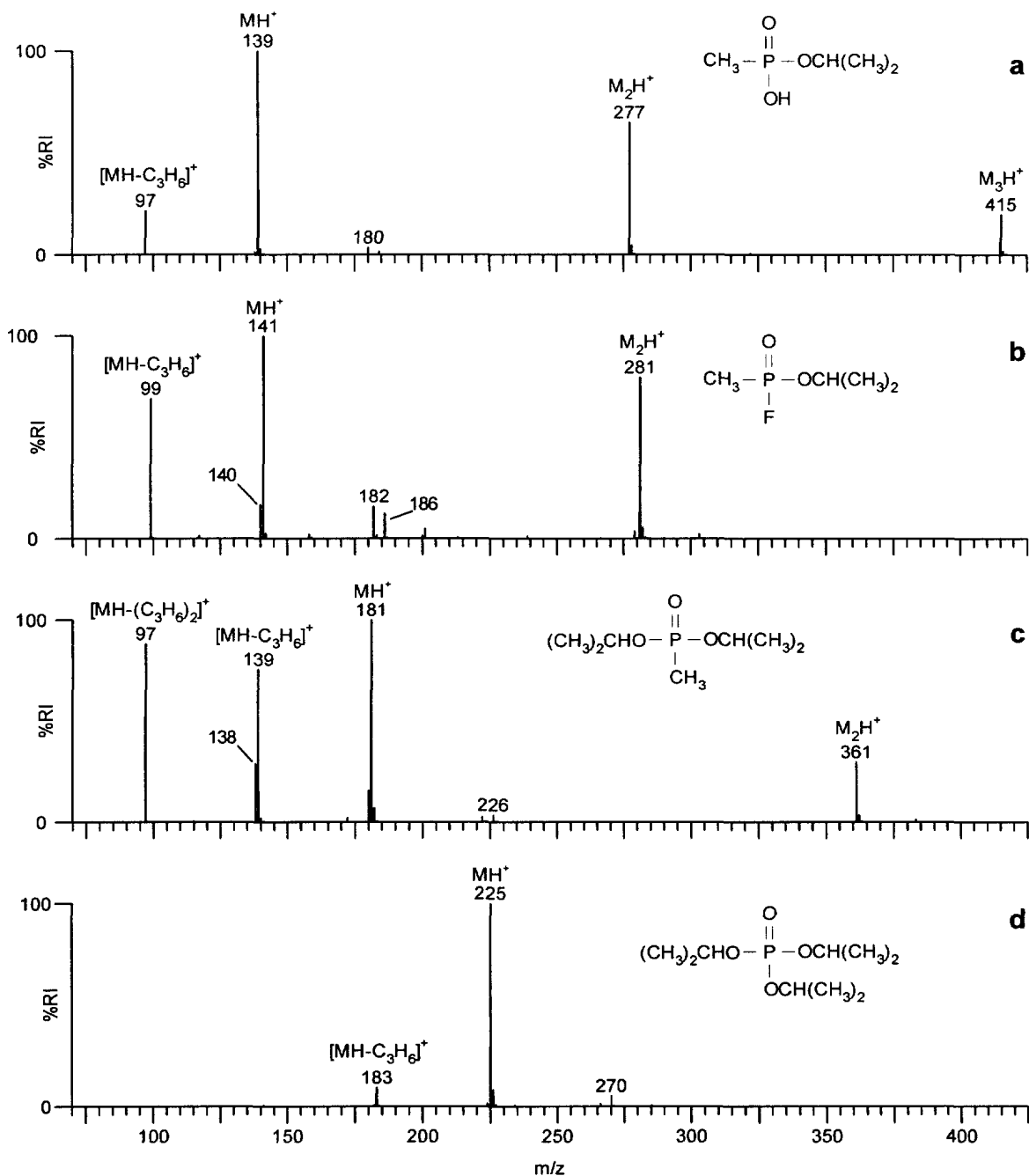


Figure 2. ESI-MS data acquired for a) isopropyl methylphosphonic acid (sampling cone voltage: 20 volts), b) sarin (sampling cone voltage: 20 volts), c) diisopropyl methylphosphonate (sampling cone voltage: 30 volts) and d) triisopropyl phosphate (sampling cone voltage: 20 volts) during LC-ESI-MS of the melted snow sample diluted 1:10 with distilled water

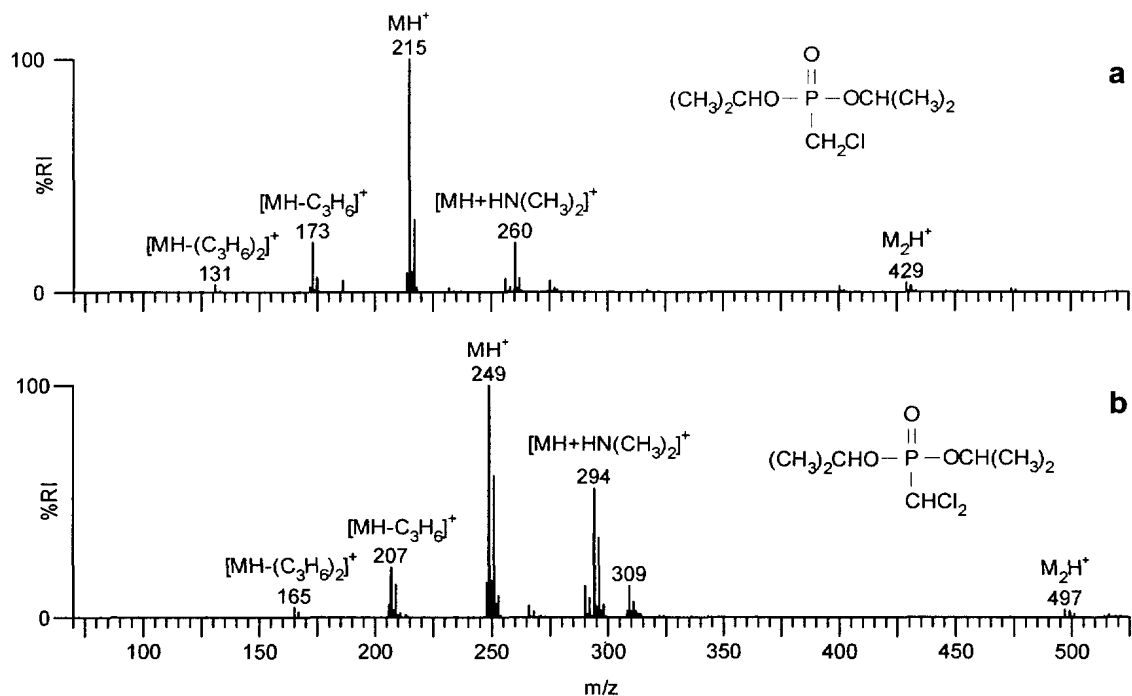


Figure 3. ESI-MS data acquired for a) diisopropyl chloromethylphosphonate (sampling cone voltage: 20 volts) and b) diisopropyl dichloromethylphosphonate (sampling cone voltage: 20 volts) during LC-ESI-MS of the melted snow sample diluted 1:10 with distilled water.

Two novel chlorinated compounds, exhibiting isotopic clusters consistent with the presence of one and two chlorines, respectively, were identified as diisopropyl chloromethylphosphonate and diisopropyl dichloromethylphosphonate based on the interpretation of acquired ESI-MS data (Figure 3). Chlorination was thought to be associated with the methyl substituent (possibly during the chlorination step in the sarin synthetic process), since both compounds exhibited sequential loss of C_3H_6 from the MH^+ ion, consistent with the presence of two isopropoxy substituents. Product ions were observed at m/z 173 and m/z 131 for diisopropyl chloromethylphosphonate and at m/z 207 and m/z 165 for diisopropyl dichloromethylphosphonate with a sampling cone voltage of 20 volts. An increase in the relative intensity of the product ions was observed with a sampling cone voltage of 30 volts, but the acquired data were complicated by a corresponding increase in acetonitrile adduct formation. The identification of both these compounds was strengthened by the high resolution data acquired for the MH^+ and $[MH-C_3H_6]^+$ ions for both compounds (Table 1). For example, the observed mass of the $[MH-C_3H_6]^+$ ion for diisopropyl chloromethylphosphonate was mass deficient, as predicted theoretically.

Figure 4 illustrates the ESI-MS data acquired for three additional organophosphorus compounds identified following interpretation of the acquired ESI-MS data. Molecular mass for all three compounds were established by the presence of MH^+ , $[MH+ACN]^+$, $[MH+HN(CH_3)_2]^+$ ions and/or protonated dimers. Product ions due to loss of C_3H_6 were

observed at m/z 95 and m/z 111 for isopropyl dimethylphosphinate (Figure 4a) and isopropyl methyl methylphosphonate (Figure 4b), respectively, indicating the presence of isopropoxy substitution for both compounds. Sequential loss of C_3H_6 from the MH^+ ion was observed at m/z 143 and m/z 101 for diisopropyl phosphorofluoridate (Figure 4c), suggesting the presence of a compound with two isopropoxy substituents. Finally, the assignment of the structures illustrated in Figure 4 was consistent with the observed masses for the MH^+ and $[MH-C_3H_6]^+$ ions for all three organophosphorus compounds listed in Table 1.

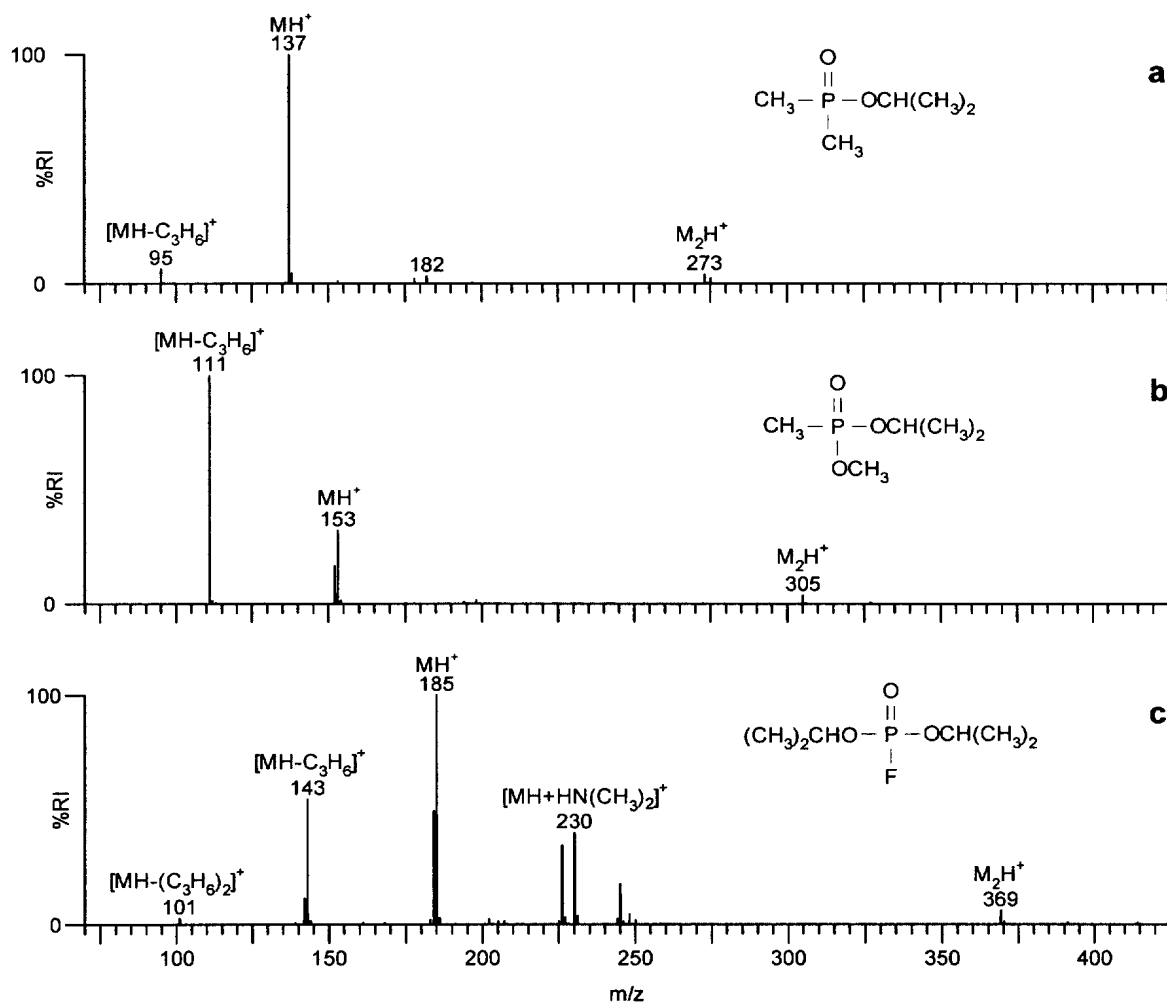


Figure 4. ESI-MS data acquired for a) isopropyl dimethylphosphinate (sampling cone voltage: 30 volts), b) isopropyl methyl methylphosphonate (sampling cone voltage: 20 volts) and c) diisopropyl phosphorofluoridate (sampling cone voltage: 20 volts) during LC-ESI-MS of the melted snow sample diluted 1:10 with distilled water.

A broad chromatographic component was observed just before fifteen minutes in the LC-ESI-MS chromatogram (Figure 1). Two components, containing an odd number of nitrogens, were detected with molecular masses of 200 Da and 186 Da based on the observed MH^+ ions (sampling cone voltages ranging from 20 to 70 volts). Acetonitrile adducts, generally observed for sarin, soman, tabun and related organophosphorus compounds (29,30) were not observed, suggesting the presence of two more basic sample components. The two compounds did not produce significant product ions until the sampling cone voltage was raised to 50 volts, with the most structurally informative mass spectra being acquired with a sampling cone voltage of 70 volts. Sarin related organophosphorus compounds fragment extensively in the ESI interface with a sampling cone voltage of 70 volts, giving rise to mass spectra containing only low mass product ions (e.g., m/z 97 or m/z 99) and their acetonitrile adducts. Product ions resulting from the loss of C_4H_8 suggested the two more basic compounds contained butyl substitution. Tributylamine, a stabilizer used in sarin munitions, and a possible impurity pentyl dibutylamine, were suspected based on this evidence. Comparison of the observed masses for the MH^+ and $[MH-C_3H_6]^+$ ions for both amines with theoretical values was consistent (Table 1), suggesting the presence of these two compounds in the sample.

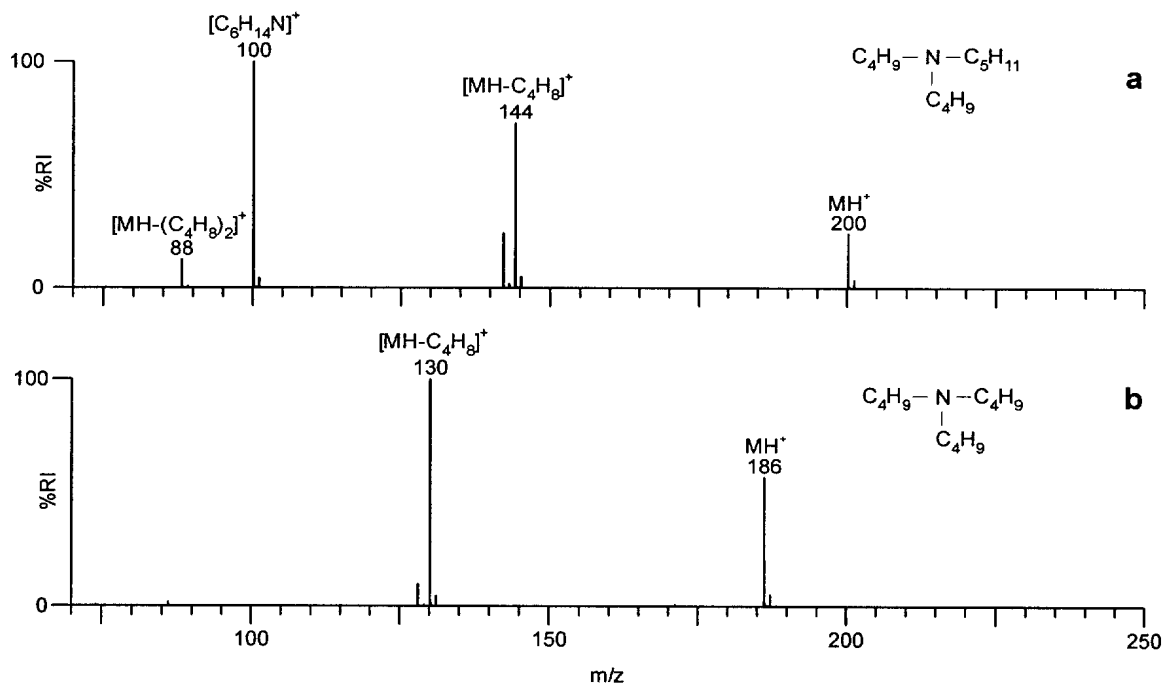


Figure 5. ESI-MS data acquired for a) pentyl dibutylamine (sampling cone voltage: 70 volts) and b) tributylamine (sampling cone voltage: 70 volts) during LC-ESI-MS of the melted snow sample diluted 1:10 with distilled water.

Conclusions

Packed capillary LC-ESI-MS was used for the analysis of organophosphorus chemical warfare agents and their hydrolysis products in a contaminated snow sample. Sarin, its hydrolysis products and a number of previously uncharacterized related compounds were identified on the basis of acquired LC-ESI-MS data. Full mass spectra were acquired for fourteen compounds detected during LC-ESI-MS analysis, with all compounds exhibiting MH^+ , $[MH+ACN]^+$ ions and/or protonated dimers that could be used to confirm molecular mass. Sampling cone voltages from 20 to 70 volts were utilized with the higher sampling voltages enhancing formation of structurally important product ions in the ESI interface. All data were acquired with a time-of-flight (TOF) mass spectrometer with a resolution of 5000 (50% valley definition), a resolution that aided in the assignment of elemental composition of the observed ions. All fourteen sample components, including two amines, were identified or provisionally identified on the basis of the ESI-MS data acquired during this investigation.

The application of LC-ESI-MS to snow analysis appears to be an attractive alternative to the GC-MS methods. A melted snow sample containing both chemical warfare agents and their hydrolysis products may be analysed directly during a single analysis, eliminating the need for the additional sample handling and derivatization steps associated with GC-MS analyses. Application of the acquired ESI-MS data is anticipated during future chemical warfare agent analyses, since presence of compounds in collected samples could help verify the prior presence of sarin, indicate the synthetic procedure used for munition production or suggest the origin of the munition.

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References

1. Kientz, Ch. E. (1998). Chromatography and mass spectrometry of chemical warfare agents, toxins and related compounds: state of the art and future prospects. *J. Chromatogr. A*, 814, 1-23.
2. Witkiewicz, Z., Mazurek, M. and Szulc, J. (1990). Chromatographic analysis of chemical warfare agents. *J. Chromatogr.*, 503, 293-357.
3. D'Agostino, P. A., Provost, L. R. and Looye, K. M. (1989). Identification of tabun impurities by combined capillary column gas chromatography-mass spectrometry. *J. Chromatogr.*, 465, 271-283.
4. D'Agostino, P. A., Provost, L. R. and Visintini, J. (1987). Analysis of O-ethyl S-[2-(diisopropylamino)ethyl] methylphosphonothiolate (VX) by capillary column gas chromatography-mass spectrometry. *J. Chromatogr.*, 402, 221-232.
5. Bonierbale, E., Debordes, L. and Coppet, L. (1997). Application of capillary gas chromatography to the study of hydrolysis of the nerve agent VX in rat plasma. *J. Chromatogr. B*, 688, 255-264.
6. Rohrbough, D.K. (1998). Characterization of equimolar VX-water reaction product by gas chromatography-mass spectrometry. *J. Chromatogr. A*, 809, 131-139.
7. Creasy, W. R., Stuff, J. R., Williams, B., Morrissey, K., Mays, J., Duevel, R. and Hurst, H. D. (1997). Identification of chemical-weapons-related compounds in decontamination solutions and other matrices by multiple chromatographic techniques. *J. Chromatogr. A*, 774, 253-263.
8. Sass, S. and Fisher, T. L. (1979). Chemical ionization and electron impact mass spectrometry of some organophosphonate compounds. *Org. Mass Spectrom.*, 14, 257-264.
9. D'Agostino, P. A. and Provost, L. R. (1986). Capillary column ammonia chemical ionization mass spectrometry of organophosphorus chemical warfare agents and simulants. *Biomed. Environ. Mass Spectrom.*, 13, 231-236.
10. Borrett, V. T., Mathews, R. J. and Mattsson, E. R. (1994). Verification of the chemical weapons convention: Mass spectrometry of alkyl methylphosphonofluoridates. *Aust. J. Chem.*, 47, 2065-2074.
11. Purdon, J. G., Pagotto, J. G. and Miller, R. K. (1989). Preparation, stability and quantitative analysis by gas chromatography and gas chromatography-electron impact mass spectrometry of tert-butyldimethylsilyl derivatives of some alkylphosphonic and alkyl methylphosphonic acids. *J. Chromatogr.*, 475, 261-272.

12. D'Agostino, P. A., and Provost, L. R. (1992). Determination of chemical warfare agents, their hydrolysis products and related compounds in soil. *J. Chromatogr.*, 589, 287-294.
13. Sega, G. A., Tomkins, B. A. and Griest, W. H. (1997). Analysis of methylphosphonic acid, ethyl methylphosphonic acid and isopropyl methylphosphonic acid at low microgram per litre levels in groundwater. *J. Chromatogr. A*, 790, 143-152.
14. Lakso, H-A. and Ng, W. F. (1997). Determination of chemical warfare agents in natural water samples by solid-phase microextraction. *Anal. Chem.*, 69, 1866-1872.
15. Kientz, C. E., Verweij, A., de Jong, G. J. and Brinkman, U. A. Th. (1992). Verification of nonproduction of chemical warfare agents: I. Determination of organophosphorus compounds by microcolumn liquid chromatography with flame photometric or thermionic detection. *J. Microcol. Sep.*, 4, 477-483.
16. Wils, E. R. J. and Hulst, A. G. (1988). Determination of organophosphorus acids by thermospray liquid chromatography-mass spectrometry. *J. Chromatogr.*, 454, 261-272.
17. Wils, E. R. J. and Hulst, A. G. (1990). Determination of O-ethyl S-2-diisopropylaminoethyl methylphosphonothiolate (VX) by thermospray liquid chromatography-mass spectrometry. *J. Chromatogr.*, 523, 151-161.
18. Wils, E. R. J. and Hulst, A. G. (1992). The use of thermospray-liquid chromatography/mass spectrometry for the verification of chemical warfare agents. *Fresenius J. Anal. Chem.*, 342, 749-758.
19. Tornes, J. A. (1996). Identification of some alkyl methylphosphonic acids by thermospray tandem mass spectrometry. *Rapid Commun. Mass Spectrom.*, 10, 878-882.
20. Black, R. M. and Read, R. W. (1997). Application of liquid chromatography-atmospheric pressure chemical ionization mass spectrometry, and tandem mass spectrometry, to the analysis and identification of degradation products of chemical warfare agents. *J. Chromatogr. A*, 759, 79-92.
21. Black, R. M. and Read, R. W. (1998). Analysis of degradation products of organophosphorus chemical warfare agents and related compounds by liquid chromatography-mass spectrometry using electrospray and atmospheric pressure chemical ionization. *J. Chromatogr. A*, 794, 233-244.
22. Read, R. W. and Black, R. M. (1999). Rapid screening procedures for the hydrolysis products of chemical warfare agents using positive and negative ion liquid chromatography-mass spectrometry and atmospheric pressure chemical ionization. *J. Chromatogr. A*, 862, 169-177.
23. Kostianen, R., Bruins, A. P. and Hakkinen, V. M. A. (1993). Identification of degradation products of some chemical warfare agents by capillary electrophoresis-ion spray mass spectrometry. *J. Chromatogr.*, 634, 113-118.

24. D'Agostino, P. A., Provost, L. R. and Hancock, J. R. (1994). Mass spectrometric identification of chemical warfare agent precursor and degradation products scheduled by the chemical weapons convention, In *Proceedings of the 42 nd Annual Conference on Mass Spectrometry and Allied Topics*, Chicago, Illinois, 275-276.
25. Borrett, V. T., Colton, R. and Traeger, J. C. (1995). The electrospray mass spectra of phosphonic acid, methyl phosphonic acid and its alkyl esters, and their complexes with alkali and alkali earth metal ions. *Eur. Mass Spectrom.*, 1, 131-140.
26. Borrett, V. T., Mathews, R. J., Colton, R. and Traeger, J. C. (1996). Verification of the United Nations chemical weapons convention: The application of electrospray mass spectrometry. *Rapid Commun. Mass Spectrom.*, 10, 114-118.
27. D'Agostino, P. A., Provost, L. R. and Hancock, J. R. (1998). Analysis of mustard hydrolysis products by packed capillary liquid chromatography-electrospray mass spectrometry. *J. Chromatogr. A*, 808, 177-184.
28. D'Agostino, P. A., Hancock, J. R. and Provost, L. R. (1999). Analysis of O-ethyl S-[2-(diisopropylamino)ethyl] methylphosphonothiolate (VX) and its degradation products by packed capillary liquid chromatography-electrospray mass spectrometry. *J. Chromatogr. A*, 837, 93-105.
29. D'Agostino, P. A., Hancock, J. R. and Provost, L. R. (1999). Packed capillary liquid chromatography-electrospray mass spectrometry analysis of organophosphorus chemical warfare agents. *J. Chromatogr. A*, 840, 289-294.
30. D'Agostino, P. A., Hancock, J. R. and Provost, L. R. (2001). Determination of sarin, soman and their hydrolysis products in soil by packed capillary liquid chromatography-electrospray mass spectrometry. *J. Chromatogr. A*, (in press).
31. Blanch, J.H., Odden, E. and Karlsen, P.J. (1982). Analysis of snow samples contaminated with chemical warfare agents. Norwegian Defence Research Establishment FFI/Rapport-82/6003, 30 pages.
32. Blanch, J.H., Johnsen, B.A. and Odden, E. (1983). Analysis of snow samples contaminated with chemical warfare agents - Part 2. Norwegian Defence Research Establishment FFI/Rapport-83/6003, 29 pages.
33. Blanch, J.H., Johnsen, B.A., Karlsen, P.J., Lyngaas, S. and Odden, E. (1984). Analysis of snow samples contaminated with chemical warfare agents - Part 3. Norwegian Defence Research Establishment FFI/Rapport-84/6003, 31 pages.
34. Tornes, J.A. and Opstad, A.M. (1996). Research report on verification of the Chemical Weapons Convention - Part XIII – Evaluation of recommended operating procedures for sampling and analysis. Norwegian Defence Research Establishment FFI/Rapport-96/05379, 28 pages.

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Packed capillary LC-ESI-MS was used for the analysis of a snow sample that was accidentally contaminated with an organophosphorus chemical warfare agent during the destruction of a chemical munition. Sarin, its hydrolysis products and a number of unique related compounds were identified on the basis of acquired LC-ESI-MS data. Full mass spectra were acquired for all fourteen compounds detected, with all compounds exhibiting MH^+ , $[MH+ACN]^+$ ions and/or protonated dimers that could be used to confirm molecular mass. Sampling cone voltages from 20 to 70 volts were utilized with the higher sampling voltages enhancing formation of structurally important product ions in the ESI interface. All data were acquired with a time-of-flight (TOF) mass spectrometer with a resolution of 5000 (50% valley definition), a resolution that aided in the assignment of elemental composition of the observed ions. The application of LC-ESI-MS to snow analysis appears to be an attractive alternative to the GC-MS methods, since both chemical warfare agents and their hydrolysis products may be analysed directly, eliminating the need for additional sample handling and derivatization steps.

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