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Liquid Chromatography/Electrospray Mass Spectrometry of Mustard-related Sulfonium Ions

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The detection and identification of degradation products of the chemical blister agent bis(2-chloroethyl) sulfide, commonly known as mustard or HD, is important for the future destruction of the mustard stockpile for safety and environmental reasons. Because stored mustard tends to degrade via intramolecular substitution forming stable sulfonium ions, conventional gas chromatographic/mass spectrometric analysis does not give a true picture of sample composition. In this study, liquid chromatography/electrospray ionization mass spectrometry (LC/MS) was applied for the detection and identification of six synthesized mustard related cyclic and open-chain sulfonium ions to demonstrate the applicability of the technique. The technique was then applied to confirm the identification of the major component of unknown solid heels isolated from mustard ton containers as the six-membered ring cyclic 1-(2-chloroethyl)-1,4-dithianium ion by comparison of spectra obtained by LC/MS and LC/MS/MS for a mustard heel and a reference standard. A mechanism to account for this sulfonium ion formation under long-term ton container storage conditions is provided. The technique was also applied to monitor open-chain sulfonium ion formation during the hydrolysis of 0.01 M mustard in 0.05, 2.3 and 10 vol.% thiodiglycol–water at 20 °C. © **1997 John Wiley & Sons, Ltd**

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INTRODUCTION

Safety and environmental impact concerns resulting from the impending destruction of the chemical agent stockpile have led to increased interest in the detection and identification of agent impurities and degradation products.[1] One agent slated for future destruction is the chemical blister agent bis(2-chloroethyl) sulfide, commonly known as sulfur mustard or HD. The characterization of both neat material stored in ton containers and hydrolyzed mustard is complicated by the presence of stable sulfonium ions which have low volatility and thermally degrade to neutral species and are therefore not detected by conventional gas chromatographic/mass spectrometric (GC/MS) analysis. Although the formation of sulfonium ions as intermediates during the thermal degradation of mustard was postulated over 40 years ago,[2] it is only recently that direct evidence for the presence of these ions in HD and 2-chloroethyl ethyl sulfide (CEES) hydrolysis products by NMR and in CEES hydrolysis products by static secondary ion mass spectrometry (SIMS) has been reported.[3–5] Some of these sulfonium ions are believed to be toxic[6] and react reversibly to reform mustard and other toxic compounds. The detection of these ions is especially critical because their presence in the environment may account for the observed persistent toxicity of HD. Although HD has been reported to hydrolyze to non-toxic thiodiglycol (TDG) with a half-life of 5 min at 25 °C,[7] toxicity has been observed in environmental samples exposed to HD as long ago as World War I.

The first objective is to demonstrate the applicability of electrospray ionization mass spectrometry (ESI-MS) for detection of sulfonium ions. Results are provided for the detection of six synthesized mustard related sulfonium ion standards. Although these ions can be identified by NMR spectroscopy, ESI provides an important complementary tool for structural confirmation with enhanced sensitivity.

The second objective is to apply electrospray ionization to the identification of an unknown solid material isolated from mustard stored in ton containers. This solid material, often referred to as a heel, settles in the bottom of the container and can pose a major problem for container decontamination. Recent sampling of two containers from the HD stockpile that had been in storage over 50 years revealed that the amount of heel present can be as high as an estimated 20–30% of the total volume of the liquid HD.[8] Characterization of this solid is critical for effective destruction of the HD stockpile and decontamination of the ton containers. Recent characterization by ^{13}C NMR revealed the presence of two major compounds, HD and a cyclic sulfonium salt identified as 1-(2-chloroethyl)-1,4-dithianium chloride.[9] In this study, comparison of electrospray ionization spectra obtained in both MS and MS/MS modes for a heel sample and a reference standard provides confirmation of the identification of the sulfonium ion.

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The third objective is to apply electrospray ionization for the monitoring of sulfonium ion formation during mustard hydrolysis. Neutralization through hydrolysis is currently being considered as a viable technique for destruction of the HD chemical stockpile.[10] In this study, the formation and degradation of three sulfonium ions during hydrolysis of 0.01 M HD in 0.5, 2.3 and 10 vol.% thiodiglycol–water at 20 °C are monitored by ESI.

EXPERIMENTAL

Materials

The sulfonium salts 1-methyl-1,4-dithianium iodide, 1-(2-chloroethyl)-1,4-dithianium tetrafluoroborate, 1-(3-chloropropyl)-1,4-dithianium tetrafluoroborate, 1-(2-chloroethyl)-1,4-oxothionium tetrafluoroborate, *S*-(2-chloroethyl)pentamethylene sulfonium tetrafluoroborate and methyl 2-hydroxyethyl 2-(methylthio)ethyl sulfonium chloride reference standards were synthesized in-house.[11]

Thiodiglycol, lot 69F-0720, was purchased from Sigma and was used as received. HD was obtained in-house and contained (GC/MS peak area%) 97.7% HD, 1.1% 1,4-dithiane, 0.6% 1,2-dichloroethane, 0.4% $ClCH_2CH_2SCH_2CH_2SCH_2CH_2Cl$ (Q) and 0.2% propyl and butyl analogs of HD.

Mustard hydrolysis reaction

A 0.01 M solution of HD in 2.3 vol.% thiodiglycol–water was prepared by adding 2.93 ml of deionized, distilled water to a mixture of 0.004 86 g of HD and 0.07 ml of thiodiglycol. Thiodiglycol, the major hydrolysis product of HD, was added to the mixture to enhance HD solubility. The mixture was shaken until the cloudiness disappeared. The sample was then stored at room temperature (20 °C) and samples were withdrawn at intervals for electrospray analysis. Solutions of 0.01 M HD were prepared in a similar fashion in 0.05 and 10 vol.% thiodiglycol–water.

Instrumental analysis

Electrospray mass spectra were obtained using a Finnigan TSQ-7000 LC/MS/MS system equipped with a Waters Model 616 LC pump. Methanol was used as the mobile phase with a flow rate of 0.1 ml min^{-1}. The nitrogen sheath gas pressure was 70 psi, the spray voltage was 4.5 kV and the heated capillary temperature was 200 °C. Better sensitivity was obtained with the auxiliary gas off. A mass range of 100–500 Da was scanned at 1 scan s^{-1}. MS/MS fragment ion spectra were obtained using argon as the collision gas with a collision-induced dissociation (CID) pressure of 1.4 mT, a collision energy of 20 V and a scan range of 50–200 Da (1 scan s^{-1}). Reference standards and heel samples were dissolved in methanol to give an approximately 0.01 M solution prior to analysis. A 1 μl volume of each sample was introduced into the source by direct flow injection.

GC/MS analysis of neat HD used for the hydrolysis reactions was obtained on a Finnigan TSQ-7000 GC/MS/MS system equipped with a 30 m × 0.25 mm i.d. DB-5ms column, film thickness 0.25 μm (J&W Scientific, Folsom, CA, USA). The carrier gas was helium with a flow rate of 1 ml min^{-1} and the oven temperature was ramped from 60 to 270 °C at 15 °C min^{-1}. The injection port temperature was 220 °C, the GC/MS interface temperature was 250 °C and the source temperature was 150 °C. The instrument was operated in the chemical ionization (CI) mode using methane as reagent gas with a source pressure of 3500 mT, an emission current of 300 μA and an electron energy of 200 eV. The mass range scanned was from 60 to 450 Da at 0.7 s per scan.

GC/MS analysis of the HD heel sample was obtained on a Finnigan 5100 GC/MS system equipped with a 15 m × 0.25 mm i.d. Rtx-5 column, film thickness 0.25 μm (Restek, Bellefonte, PA, USA). The oven temperature was programmed from 60 to 270 °C at 10 °C min^{-1} with an interface temperature of 230 °C and an injection port temperature of 210 °C. Samples were analyzed as dilute solutions in ethanol in both electron ionization (EI) and methane CI modes with an emission current of 500 μA, an electron energy of 70 eV and a methane source pressure of 0.5 Torr. The mass range scanned was from 60 to 450 Da (CI) or from 45 to 450 Da (EI) at 1 scan s^{-1}.

RESULTS AND DISCUSSION

Electrospray mass spectra of synthesized sulfonium ion standards

Mass spectra obtained under electrospray conditions for the six synthesized cyclic and open-chain sulfonium ions listed in Table 1 are provided in Fig. 1. All six ions were readily detected at the 0.01 M level with little or no ion fragmentation observed. The fragment ions observed in the spectrum of the 1-(2-chloroethyl)-1,4-dithianium ion at *m/z* 147 and the 1-(2-chloroethyl)-1,4-oxothionium ion at *m/z* 131 may be impurities present in the samples resulting from HCl elimination. This assumption is supported by the absence of *m/z* 147 as a product ion from the corresponding collisionally activated precursor sulfonium ion (see Fig. 3). The *m/z* 131 ion also is not observed as a product ion from the oxothionium precursor ion. Because of the absence of significant fragmentation the technique can be used for the detection of mixtures of sulfonium ions without chromatographic separation.

Mustard heel analysis

A comparison of electrospray spectra obtained for a mustard heel sample isolated from a ton container and a synthesized reference standard of the 1-(2-chloroethyl)-1,4-dithianium ion (**1**) is shown in Fig. 2. Both spectra are dominated by the base peak at *m/z* 183 with the chlorine isotope at *m/z* 185. The presence of the *m/z*

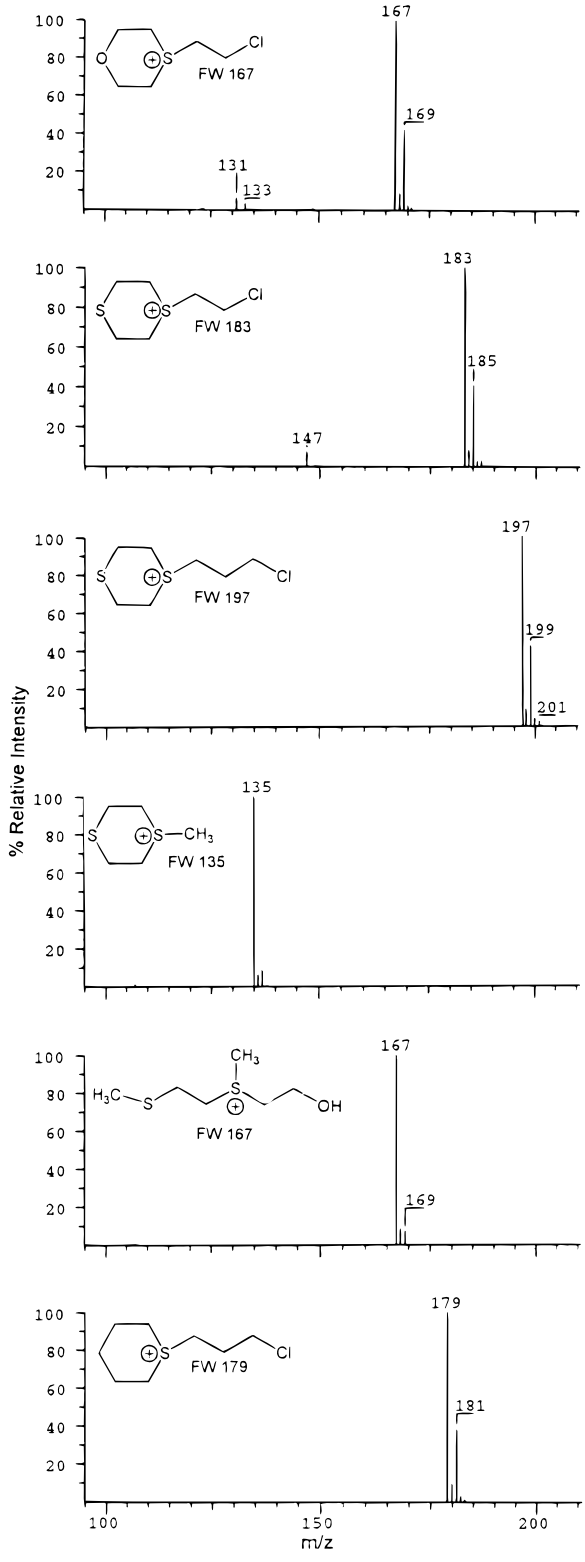


Figure 1. Electrospray mass spectra of synthesized sulfonium ions.

243/245 ions in the heel spectrum suggests that a small amount of the polymeric sulfonium ion analog $S[CH_2CH_2]_2S^+CH_2CH_2SCH_2CH_2Cl$ may also be present. To provide further confirmation that the major ion detected in the heel is **1**, MS/MS fragment ion spectra were obtained for the *m/z* 183 parent ion for both the heel and the standard and are shown in Fig. 3.

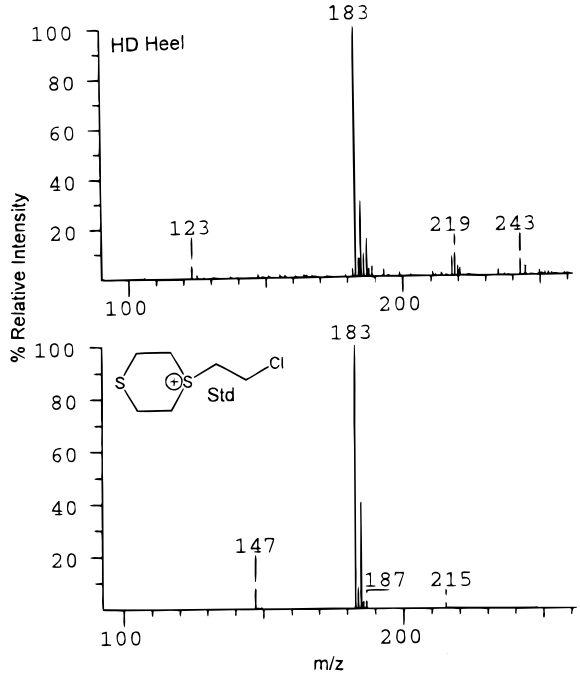


Figure 2. Electrospray mass spectra of HD heel and 1-(2-chloroethyl)-1,4-dithianium ion standard.

The heel sample was also analyzed under GC/MS conditions in methanol solution. As shown previously, sulfonium ions derived from HD-related compounds are not stable under GC/MS conditions and decompose to neutral species.[12,13] The results obtained are given in Table 2. As expected, no sulfonium ion was detected. Direct exposure probe analysis of the heel solid also resulted in thermal degradation of the sulfonium ion with the composite spectrum obtained having ions characteristic of the compounds in Table 2. Thermal degradation under GC conditions occurs via nucleophilic

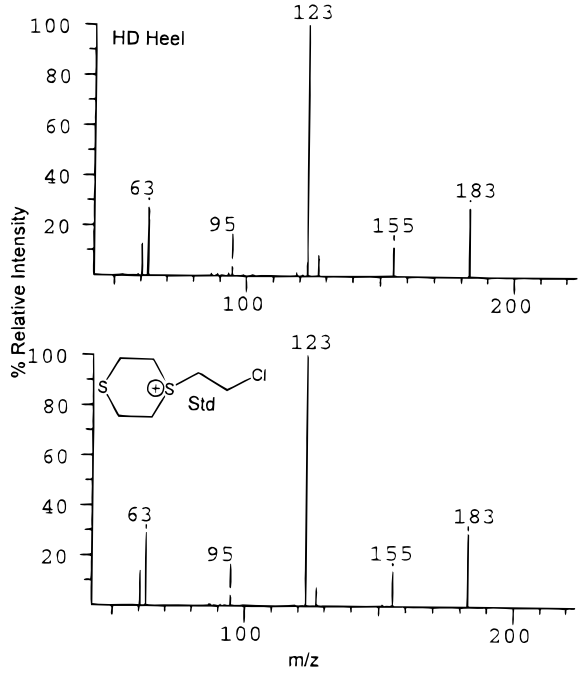
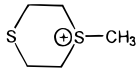
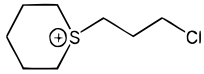
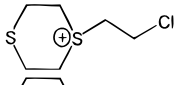
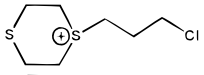
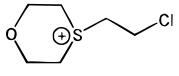
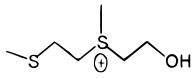
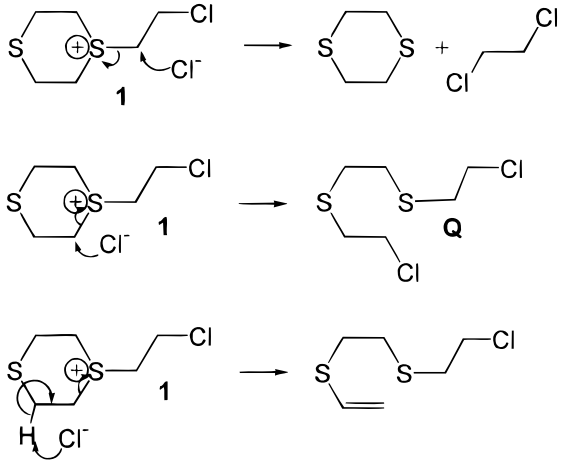


Figure 3. Electrospray tandem mass spectra of *m/z* 183 fragment ion (1.4 mT argon, 20 V) of HD heel and 1-(2-chloroethyl)-1,4-dithianium ion standard.

Table 1. Synthesized sulfonium ions analyzed by ESI-MS

Compound	Ion structure	*m/z*
1-Methyl-1,4-dithianium		135
S-(3-Chloropropyl)pentamethylenesulfonium		179/181
1-(2-Chloroethyl)-1,4-dithianium		183/185
1-(3-Chloropropyl)-1,4-dithianium		197/199
1-(2-Chloroethyl)-1,4-oxothionium		167/169
Methyl-2-hydroxyethyl-2-(methylthio)ethylsulfonium		167

attack by the chloride anion on either of the carbons alpha to the charged sulfur and by simple elimination of the beta ring hydrogen as shown in Eqns (1)–(3). Occlusion of liquid HD in the heel as it precipitates (confirmed by NMR) results in the high level of HD observed.



(1)

(2)

(3)

A proposed mechanism to account for formation of the sulfonium ion **1** in mustard ton containers stored in Aberdeen Proving Ground (APG), Maryland, under nitrogen is shown in Fig. 4. We believe the degradation is initiated by the formation of the reactive ethylenesulfonium ion via neighboring sulfur assistance and polarization of the C—Cl bond (an S_N1 process). Nucleophilic attack by a second molecule of HD results in the more stable dimeric sulfonium ion,[3] which slowly decomposes to give Q and 1,2-dichloroethane. Q slowly cyclizes via intramolecular substitution to give **1**, which is not soluble in HD and therefore precipitates out of solution as a solid.

Table 2. GC/MS analysis of HD heel

Compound	Area%
$ClCH_2CH_2SCH_2CH_2Cl$ (HD)	54.2
1,4-Dithiane	16.8
1,4-Thioxane	0.2
1,2-Dichloroethane	4.6
$ClCH_2CH_2SCH_2CH_2SCH_2CH_2Cl$ (Q)	16.8
$CH_2{=}CHSCH_2CH_2SCH_2CH_2Cl$	7.4

Hydrolysis of HD/thiodiglycol solutions

A mechanism for the hydrolysis of HD to thiodiglycol ($HOCH_2CH_2SCH_2CH_2OH$, TDG) and the subsequent nucleophilic substitution reactions between HD, TDG and water to form sulfonium salts has been reported.[4,14] The structures for three sulfonium ions, H-TG, CH-TG and H-2TG, identified as major products of this reaction by ^1H and ^{13}C NMR, are shown in Fig. 5. Based on the above results for the analysis of sulfonium ion standards and HD heels, electrospray ionization should be a viable candidate for monitoring the formation and degradation of sulfonium ions during HD hydrolysis.

To test this theory and to study the effect of TDG concentration on sulfonium ion formation, hydrolysis of 0.01 M HD in 0.5, 2.3 and 10 vol.% TDG–H_2O at room temperature (20 °C) was monitored by ESI-MS. Three major ions at *m/z* 245/247, 227 and 166 were observed,

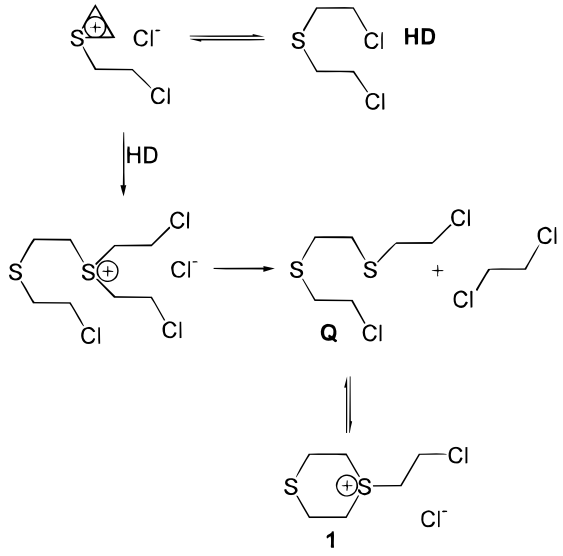


Figure 4. Proposed mechanism of HD degradation in APG ton containers.

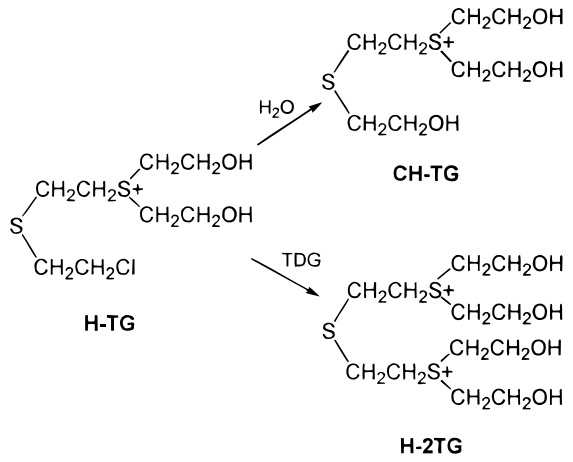


Figure 5. Sulfonium ions observed during hydrolysis of 0.01 M HD in TDG–H_2O.

corresponding to H-TG, CH-TG and H-2TG (doubly charged), respectively. Figure 6 is a plot of data obtained for this reaction using the relative abundances on the *m/z* 245/247, 227 and 166 ions observed over a 120 h period. As shown, the initial product is H-TG, which then decomposes to form CH-TG and H-2TG with a half-life of about 4.5 h. Equilibrium concentrations of CH-TG and H-2TG are reached at about 20 h with no further change in concentration observed up to 120 h.

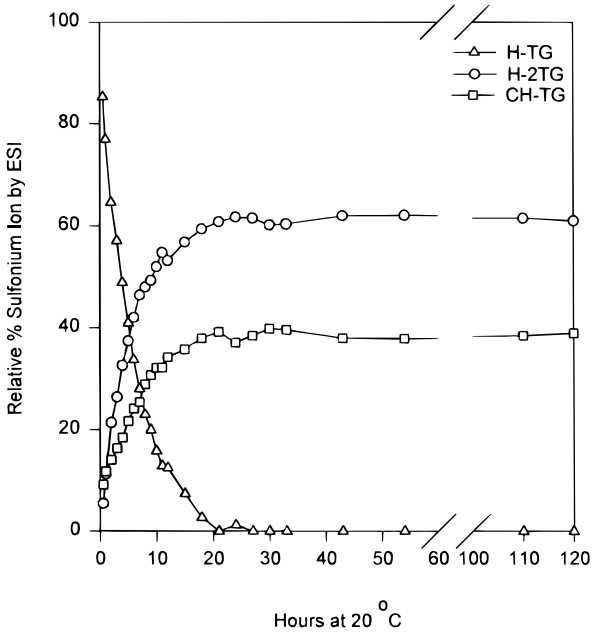


Figure 6. Sulfonium ion formation during hydrolysis of 0.01 M HD in 2.3 vol.% TDG–H_2O at 20 °C.

Similar results were obtained with TDG concentrations of 0.5 and 10 vol.%. The major difference observed is the final equilibrium distribution of products. As expected from the reactions shown in Fig. 5, increasing the TDG concentration results in increased formation of H-2TG relative to CH-TG. The rate of degradation of H-TG, and S_N1 process, decreases slightly as TDG concentration is increased (i.e. solvent polarity is decreased) with half-lives of ~4, 4.5 and 5.5 h observed at 0.5, 2.3 and 10 vol.% TDG, respectively.

CONCLUSION

ESI-MS has been successfully applied for the direct detection of sulfonium ions formed during HD storage and hydrolysis. Six synthesized mustard-related cyclic and open-chain sulfonium ions were all readily detected at a concentration of 0.01 M with little of no ion fragmentation observed.

ESI-MS was applied to successfully identify the major component of a mustard heel isolated from HD storage ton containers as the 1-(2-chloroethyl)-1,4-dithianium ion. Confirmation of the identification was provided by comparison of fragment ion tandem mass spectra obtained for the *m/z* 183 parent ion of the heel and a reference standard.

ESI-MS was also applied to successfully monitor the formation and reaction of sulfonium ions during 0.01 M mustard hydrolysis in 0.5, 2.3 and 10 vol.% thiodiglycol–water. Three major sulfonium ions were observed with equilibrium concentrations resulting from complete hydrolysis of the chlorinated H-TG sulfonium ion obtained after about 20 h at room temperature.

This is the first demonstration of the application of ESI-MS techniques for detecting and identifying sulfonium ions from mustard and mustard analogs. Because of the ubiquitous presence of sulfonium salts both in stored neat mustard and as products from mustard hydrolysis, and because of the difficulty in detecting these ions directly by most conventional techniques, ESI-MS may have important applications for monitoring HD degradation and environmental effects and for better understanding of the chemistry of future destruction processes of HD stockpiles.

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REFERENCES

1. Committee on Review and Evaluation of the Army Chemical Stockpile Disposal Program, National Research Council, *Recommendations for Disposal of Chemical Agents and Munitions*. National Academy Press, Washington, DC (1994).
2. R. C. Fuson, R. D. Lipscomb, B. C. McKusick and L. J. Reed, *J. Chem. Soc.* **11**, 513 (1946); M. A. Stahman, J. S. Fruton and M. Bergman, *J. Chem. Soc.* **11**, 704 (1946); and C. C. Price and A. Pohland, *J. Org. Chem.* **12**, 249 (1947).
3. Y.-C. Yang, L. L. Szafraniec, W. T. Beaudry and J. R. Ward, *J. Org. Chem.* **53**, 3293 (1988).
4. Y.-C. Yang, L. L. Szafraniec, W. T. Beaudry and J. R. Ward, *J. Org. Chem.* **51**, 1637 (1987).

5. G. S. Groenewold, J. C. Ingram, A. D. Appelhans, J. E. Delmore and D. A. Dahl, *Environ. Sci. Technol.* **29**, 2107 (1995).
6. W. P. Anslow, D. A. Karnofsky, B. Val Jager and H. W. Smith, *J. Pharmacol. Exp. Ther.* **93**, 1 (1948).
7. P. D. Bartlett and C. G. Swain, *J. Am. Chem. Soc.* **71**, 1406 (1949).
8. Dalton, D. W., in *Proceedings of the 1996 Scientific Conference on Chemical and Biological Defense Research, November 19–22, 1996, APG, MD*, in press.
9. Y.-C. Yang, L. L. Szafraniec, W. T. Beaudry and D. K. Rohrbaugh, in *Proceedings of the 1996 Scientific Conference on Chemical and Biological Defense Research, November 19–22, 1996, APG, MD*, in press.
10. Y.-C. Yang, *Chem. Ind.* (*London*) 334 (1995).
11. G. Hondrogiannis, Y.-C. Yang and F.-L. Hsu, in *Proceedings of the 1996 Scientific Conference on Chemical and Biological Defense Research, November 19–22, 1996, APG, MD*, in press.
12. D. K. Rohrbaugh, Y.-C. Yang and J. R. Ward, *Phosphorus Sulfur Silicon* **44**, 17 (1989).
13. D. K. Rohrbaugh, Y.-C. Yang and J. R. Ward, *J. Chromatogr.* **447**, 165 (1988).
14. Y.-C. Yang, J. A. Baker and J. R. Ward, *Chem. Rev.* **92**, 1729 (1992).

