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Identification of Fenton's Reagent-generated Atrazine Degradation Products by High-performance Liquid Chromatography and Megaflow Electrospray Ionization Tandem Mass Spectrometry†

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High-performance liquid chromatography megaflow electrospray tandem mass spectrometry (HPLC/ES-MS/MS) with an on-line radioisotope detector was used to identify [2,4,6-^{14}C]atrazine degradation products generated by treatment with Fenton's reagent (Fe^{2+} and H_2O_2). Fenton's reagent produced dealkylated and/or partially oxidized [2,4,6-^{14}C]atrazine products in preference to dechlorinated products. Seven major products were identified by collision-induced dissociation spectra: 4-acetamido-2-chloro-6-(isopropylamino)-*s*-triazine, 4-amino-2-chloro-6-(isopropylamino)-*s*-triazine, 4-acetamido-2-chloro-6-(ethylamino)-*s*-triazine, 6-amino-2-chloro-4-(ethylamino)-*s*-triazine, 4-acetamido-6-amino-2-chloro-*s*-triazine, 2-chloro-4,6-diamino-*s*-triazine and the first report of 4-acetamido-2-hydroxy-6-(isopropylamino)-*s*-triazine. HPLC/ES-MS/MS provided a rapid method for identifying a wide range of atrazine transformation products in aqueous samples and obviated the need for fraction collection, extraction, and chemical derivatization of the more polar atrazine products. Furthermore, because analyte retention times in HPLC/UV and HPLC/ES-MS/MS were similar, compound identities determined using the latter could be directly assigned to peaks in UV chromatograms.

INTRODUCTION

Atrazine (2-chloro-4-(ethylamino)-6-(isopropylamino)-*s*-triazine) is a widely used herbicide and common groundwater contaminant in the USA.[1] The highest levels of atrazine in groundwater occur where atrazine has been improperly disposed of or spilled in overlying soils.[2] Since atrazine is a possible human carcinogen,[3] remediation of soils and groundwaters contaminated by this chemical is a high priority for state and federal regulatory agencies.

A number of chemically and/or biologically based processes have been examined as technologies for treating atrazine-contaminated wastes. Chemical oxidation systems that generate hydroxyl radical (OH•) such as UV radiation combined with ozone, H_2O_2, Fe^{3+} or TiO_2[4–9] are faster than biological systems in initiating atraxine dealkylation and/or dechlorination. However, chemical oxidation approaches typically do not effect atrazine mineralization (i.e. total conversion to CO_2, NH_3 and Cl^-), but rather result in the accumulation of one or more degradation products. Since some atrazine derivatives may have toxicological characteristics similar to the parent compound,[3,5,7,8–10] product identification is essential for evaluating treatment efficacy.

Many investigators have used gas chromatography/mass spectrometry (GC/MS) to study *s*-triazine products generated by chemical oxidation systems.[5,8,9,11–13] However, since analysis by GC methods requires compounds with high vapor pressures, the less volatile, polar products need to be derivatized for identification by this approach.[13–15] Furthermore, mixture separations achieved by GC may not be directly comparable to those produced by high-performance liquid chromatography (HPLC). Since routine analysis of atrazine oxidation products is most conveniently done by HPLC, unambiguous assignment of the identity of an HPLC peak necessitates its isolation (e.g. fraction collection and extraction) prior to GC/MS analysis; identification of the more polar oxidation products by this approach may be particularly problematic.

Thermospray and particle beam LC/MS techniques have been used for the analysis of triazines.[15–17] However, confirmation of compounds can be difficult in thermospray because of insufficient fragmentation, and

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particle beam techniques often cannot ionize non-volatile compounds.[14] Development of atmospheric pressure chemical ionization and the electrospray (ES) interfaces[14,18] have made HPLC/ES-MS a practical alternative for identifying non-volatile, polar and temperature-labile compounds.[19–23] The advantages of ES interfaces are that the LC inlet is operated at atmospheric pressure and is decoupled from the mass spectrometer operating at 10^{-6} Torr, allowing optimized operating conditions for both instruments.[14] Solvents eluting from the chromatograph remain in the atmospheric pressure chamber and do not reach the mass spectrometer. Desolvation of liquid droplets is more efficient at atmospheric pressure, and the integrity of labile compounds is maintained because of free jet expansion as they enter the vacuum region. Structural information is generated by collision-induced dissociation (CID) in the collision cell.

We are studying the use of hydroxyl radicals generated by Fenton's reagent[24] (Fe^{2+} and H_2O_2) to initiate atrazine dealkylation in sequential combination with bacterial cultures that degrade residual atrazine products.[25] Product identification is the key to optimizing each treatment phase and maximizing the overall process efficacy. This paper describes the identification of seven [2,4,6-^{14}C]atrazine products generated by Fenton's reagent treatment using HPLC/ES-MS/MS with on-line radioisotope detection to improve parent ion scanning. Atrazine products were isolated for confirmatory analysis by HPLC/ES-MS/MS, high-resolution electron impact mass spectrometry (HR-EI-MS) and GC/MS.

EXPERIMENTAL

Materials

Acetonitrile (Baxter, McGaw Park, IL, USA) and ammonium acetate (Fisher Scientific, Itasca, IL, USA) were of HPLC grade or better and filtered and degassed before use. Iron (II) sulfate was purchased from Fisher Scientific and H_2O_2 (30%) from Mallinckrodt (Paris, KY, USA). Non-labeled atrazine was certified 99% pure and purchased from Chem Service (West Chester, PA, USA). [2,4,6-^{14}C]Atrazine (19.4 μCi mg^{-1}) and standards (common name and chemical purity in parentheses) were gifts from Ciba-Geigy (Greensboro, NC, USA): 4-amino-2-chloro-6-(isopropylamino)-*s*-triazine (deethylatrazine, 99%), 6-amino-2-chloro-4-(ethylamino)-*s*-triazine (deisopropylatrazine, 98%), 2-chloro-4,6-diamino-*s*-triazine (diaminoatrazine, 90%), 4-(ethylamino)-2-hydroxy-6-(isopropylamino)-*s*-triazine (hydroxyatrazine, 98%), 6-amino-4-(ethylamino)-2-hydroxy-*s*-triazine (deisopropylhydroxyatrazine, 95%) and 4-amino-2-hydroxy-6-(isopropylamino)-*s*-triazine (deethylhydroxyatrazine, 97%).

Fenton's reagent treatment

A solution of $FeSO_4$ (0.73 mM) and atrazine (140 μM in 100 ml of distilled, deionized water) spiked with 0.38 μCi of [2,4,6-^{14}C]atrazine were mixed in a 150 ml Erlenmeyer flask. Hydroxyl radical production was initiated by adding H_2O_2 to a final concentration of 0.73 mM. The flasks were wrapped with aluminum foil and incubated for 24 h on a rotary shaker at 200 rpm (25 ± 1 °C). Iron precipitates produced by the reaction were removed by centrifuging the samples (12 500 rpm for 30 min) in Teflon tubes.

Identification of atrazine products

HPLC/UV. HPLC/UV analysis and fraction collection were carried out with a Hewlett-Packard (Palo Alto, CA, USA) Model 1050 system equipped with a variable-wavelength UV detector (λ = 220 nm). Analytes were separated using a Hewlett-Packard ODS Hypersil C-18 reversed-phase column (200 mm × 4.6 mm i.d.; 5 μm mean particle diameter) at a flow-rate of 1 ml min^{-1}. The mobile phase, ammonium acetate (5 mM, pH 4.6)–acetonitrile, was run in a gradient from 5 to 89% acetonitrile in 18 min. The column was re-equilibrated at the starting conditions in a 10 min post-run.

HPLC/ES-MS/MS. A 20–50 μl volume of sample was injected through a Rheodyne (Cotati, CA, USA) valve on to a Hewlett-Packard ODS Hypersil column (200 × 2.1 mm i.d., 5 μm mean particle diameter). A Hewlett-Packard Model 1090 HPLC system was used to generate a mobile phase gradient that consisted of ammonium acetate (5 mM, pH 4.6) – acetonitrile, and was run from 5 to 90% acetonitrile in 19 min at a flow-rate of 280 μl min^{-1}. Starting conditions were re-established 25 min after injection. The column eluate flowed in series to a Romona 92 solid cell (330 μl) radioisotope detector (Raytest, Pittsburgh, PA, USA) and then to the Megaflow ES interface (Fig. 1). The offset time between the radioisotope detector and the MS configuration was approximately 1.4 min. The Megaflow ES interface was maintained at an operating temperature of 120 °C. A low flow-rate of 280 μl min^{-1}

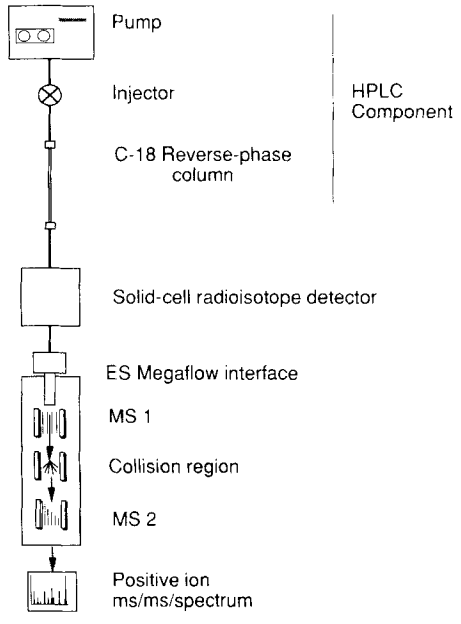


Figure 1. Schematic diagram of the HPLC/ES-MS/MS system.

allowed a longer sample residence time in the splitless on-line radioisotope detector. Solvent was pumped through the high-voltage needle (3.15 kV) forming a supersonic jet. Ions were formed at atmospheric pressure and then entered the mass spectrometer under high vacuum. The instrument was tuned to exclude fragmentation in the ES interface. The ES spectra were acquired for the protonated analyte and its products on a VG Quattro triple quadrupole mass spectrometer (VG Instruments, Beverly, MA, USA), which was controlled through a MassLynx Multitasking operating system. The spectra were acquired in the scan mode, using a scan rate of 4 s decade^{-1} over the m/z range 50–600. Parent ions were focused using Q_1 at 50% main beam attenuation and passed to the second quadrupole, where they were dissociated by collision with ultra-pure argon gas. The third quadrupole was scanned from m/z 40 to 250 in 2 s. Collision energies of 30–50 eV were used to obtain CID spectra in the multi-channel analysis mode.

GC/MS. Electron impact (EI) mass spectra were obtained with a Kratos MS-25 mass spectrometer interfaced to a Carlo Erba Fractovap 4162 gas chromatograph. Spectra were recorded at an electron energy of 70 eV and accelerating voltage of 4 kV. The source temperature was set at 250 °C. The magnet was scanned from 28 to 343 u at 1 s decade^{-1} and the resolution was set to $m/\delta m = 600$. Samples (1 µl) were injected splitless onto a 60 m DB 5 column. The oven temperature started at 40 °C and was increased to 280 °C at 15 °C min^{-1}.

HR-EI-MS. Electron impact (70 eV) mass spectra were obtained with a Kratos MS-80 mass spectrometer equipped with a direct exposure probe. Samples were dissolved in chloroform and placed directly on the probe tip. The resolution was set to $m/\delta m = 7500$ and the sample was scanned from 53 to 654 u at 10 s decade^{-1}. The source temperature was set at 250 °C.

Isolation of atrazine products. The Fenton's reagent product mixture (10 ml), prepared as described above, was extracted four times with 2 ml of ethyl acetate and then evaporated to dryness under a gentle stream of nitrogen. The samples were then dissolved in 500 µl of methanol–water (50:50). Fractions containing eight major peaks were collected from 20 injections (injection volume = 30 µl). This approach provided the 500 ng per analyte needed in a 20 µl sample volume for mass spectra analysis by HPLC/ES-MS/MS. Peak 3 was not recovered in ethyl acetate extracts; in this case, 5 ml of the Fenton's reagent product mixture were concentrated to 1 ml under nitrogen and the peak was isolated from multiple HPLC injections as described above.

RESULTS AND DISCUSSION

Atrazine degradation products were analyzed by conventional HPLC/UV and by monitoring parent ions, fragment ion products and radioactivity with HPLC/ES-MS/MS. The compounds were identified by comparison with authentic standards, published spectra and interpretation of fragment ions. The results are summarized in the following sections according to analysis method.

Comparisons of HPLC/UV, HPLC/RAD and total ion chromatogram

Fenton's reagent treatment generated seven major atrazine products detectable with HPLC/UV [Fig. 2(a)]. Direct comparisons of peaks between the HPLC/UV, HPLC/RAD (HPLC/radioisotope detection) and total ion chromatograms (TIC) are possible because the analytes' relative retention times were similiar in the HPLC/UV and HPLC/ES-MS/MS systems [Fig. 2(a), (b) and (c)]. Decreased peak resolution in the HPLC/RAD chromatogram resulted from using a large radioisotope detector cell. Atrazine (peak 8, retention time t_R = 16.9 min), deethylatrazine (peak 6, t_R = 11.1 min), deisopropylatrazine (peak 4, t_R = 8.8 min) and diaminoatrazine (peak 1, t_R = 4.7 min) were tentatively identified in UV chromatograms by co-elution with authentic standards. Peaks 2 (t_R = 6.0 min), 3 (t_R = 7.2 min), 5 (t_R = 10.4 min) and 7 (t_R = 12.7 min) did not co-elute with any of the seven standards used.

Mass spectra of atrazine products acquired by HPLC/ES-MS/MS

Initial experiments were carried out to develop and validate the HPLC/ES-MS/MS methodology using the eight major peaks detected by HPLC/UV on the Fenton's reagent product mixture. Examples of the CID spectra obtained for the isolated compounds are given for peaks 8, 7 and 3 (Figs 3–5). Fragmentation patterns obtained with HPLC/ES-MS/MS for all eight peaks are summarized in Table 1 and described further below.

Peak 8: atrazine. Atrazine has an abundant parent ion $[M + H]^+$ at m/z 216 with the corresponding ^{37}Cl at m/z 218 [Fig. 3(a) and (b)]. Cleavage of the isopropyl group yields deisopropylatrazine, m/z 174 (176). Loss of both side-chains gives diaminoatrazine, m/z 146 (148). The ions m/z 132 (134) are ring cleavage products present only when the ethylamino group is linked to the *s*-triazine ring. The ion at m/z 110 represents Cl loss from diaminoatrazine. Ions at m/z 104 (106), 96, 79 (81), 71 and 68 are ring cleavage fragments. The spectrum of the authentic standard was identical with that of the isolated compound.

Peak 7: 4-acetamido-2-chloro-6-(isopropylamino)-*s*-triazine. Figure 4(a) and (b) show that peak 7 has an abundant parent ion $[M + H]^+$ of m/z 230 (232). The *N*-ethyl side-chain was oxidized to give C=O in the C(2) position (most stable). Other investigators have identified this product by comparison with the authentic compound using GC/MS[5,8,13] and by direct injection probe

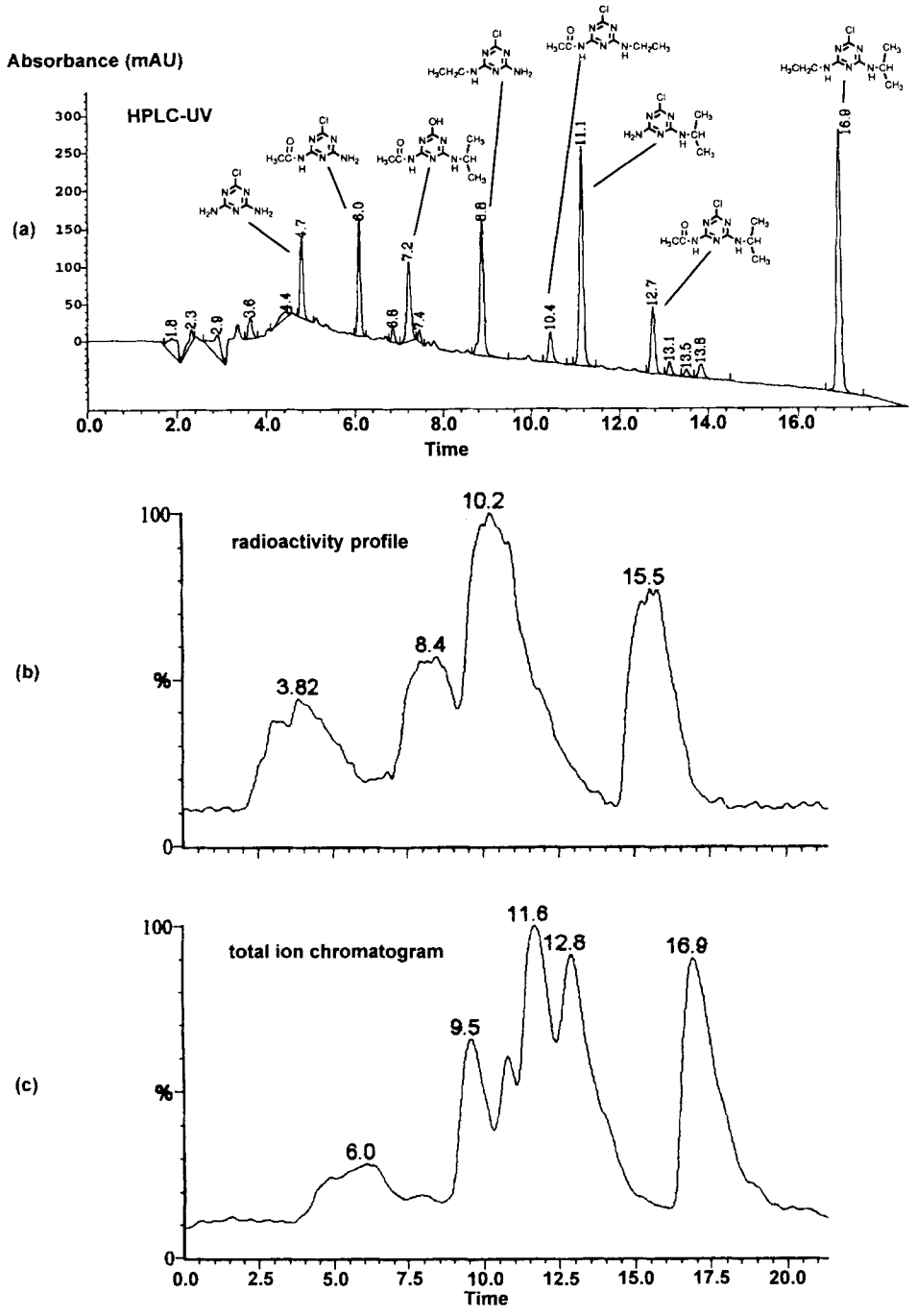


Figure 2. (a) HPLC/UV trace, (b) HPLC/ES-MS/MS radioactivity profile and (c) total ion chromatogram of an atrazine solution (140 μM) treated with 0.73 mM Fenton's reagent.

MS.[11] The ketene (*m/z* 42) is a better leaving group than the isopropyl group, and its loss gives ions at *m/z* 188 (190). Further fragmentation gives ions at *m/z* 146 (148) representing loss of both side-chains. The ions at *m/z* 110, 104 (106), 79 (81) and 68 are similar to that of atrazine.

Peak 6: deethylatrazine. Deethylatrazine has an abundant parent ion $[M + H]^+$ at *m/z* 188 (190) (Table 1). Loss of the isopropyl group yields the ions at *m/z* 146 (148). Ions at *m/z* 110, 104 (106), 79 (81) and 68 are similar to those for atrazine. The CID spectrum obtained for the authentic standard was identical with that of the isolated product.

Peak 5: 4-acetamido-2-chloro-6-(ethylamino)-*s*-triazine. Peak 5 had the same parent ion as atrazine, *m/z* 216 (218) (Table 1), but different HPLC retention times and CID spectra. Peak 5 had an HPLC retention time of 10.4 min compared with 16.9 min for atrazine. CID of 4-acetamido-2-chloro-6-(ethylamino)-*s*-triazine yields ions at *m/z* 174 (176) resulting from ketene cleavage. This compound lacked the ions at *m/z* 146 (148) or *m/z* 110 that were evident for atrazine. A ring-cleavage product

Table 1. HPLC/ES-MS/MS fragmentation of atrazine degradation products (m/z)[a]

Ion or fragment	HPLC/UV peak No. 1[b]	2	3	4	5	6	7	8
$[M+H]^+$	146 (148)[c]	188 (190)	212	174 (176)	216 (218)	188 (190)	230 (232)	216 (218)
$[M+H]^+ - H_2C{=}CH_2$				146 (148)				
$[M+H]^+ - H_2C{=}C{=}O$		146 (148)	170		174 (176)	146 (148)	188 (190)	174 (176)
$[M+H]^+ - H_2C{=}C{=}O$ and $H_3CCH{=}CH_2$			128				146 (148)	
$[M+H]^+ - H_2C{=}CH_2$ and $H_3CCH{=}CH_2$								146 (148)
$[ClC{\equiv}NC(NHC_2H_5){=}NH]^+$				132 (134)	132 (134)			132 (134)
[Diaminoatrazine-Cl]$^+$	110			110		110	110	110
$[ClC{\equiv}NC(NH_2){=}NH]^+$	104 (106)			104 (106)	104 (106)	104 (106)	104 (106)	104 (106)
$[C_2H_5NHC{\equiv}NCN]^+$				96	96			96
$[HN{=}C(NH_2)N{\equiv}C{-}OH]^+$			86					
$[ClC(NH_2){=}NH_2]^+$	79 (81)			79 (81)	79 (81)	79 (81)	79 (81)	79 (81)
$[C_2H_5N(H)C{\equiv}NH]^+$				71	71			71
$[N{\equiv}CN{=}C{=}NH_2]^+$	68			68		68	68	68
$[ClC{\equiv}NH]^+$	62 (64)			62 (64)				
$[H_2NC{\equiv}NH]^+$		43	43	43	43	43	43	43

[a] Identified in collected fractions unless noted otherwise.
[b] Ion or fragment identified in mixed sample.
[c] ^{37}Cl isotope in parentheses.

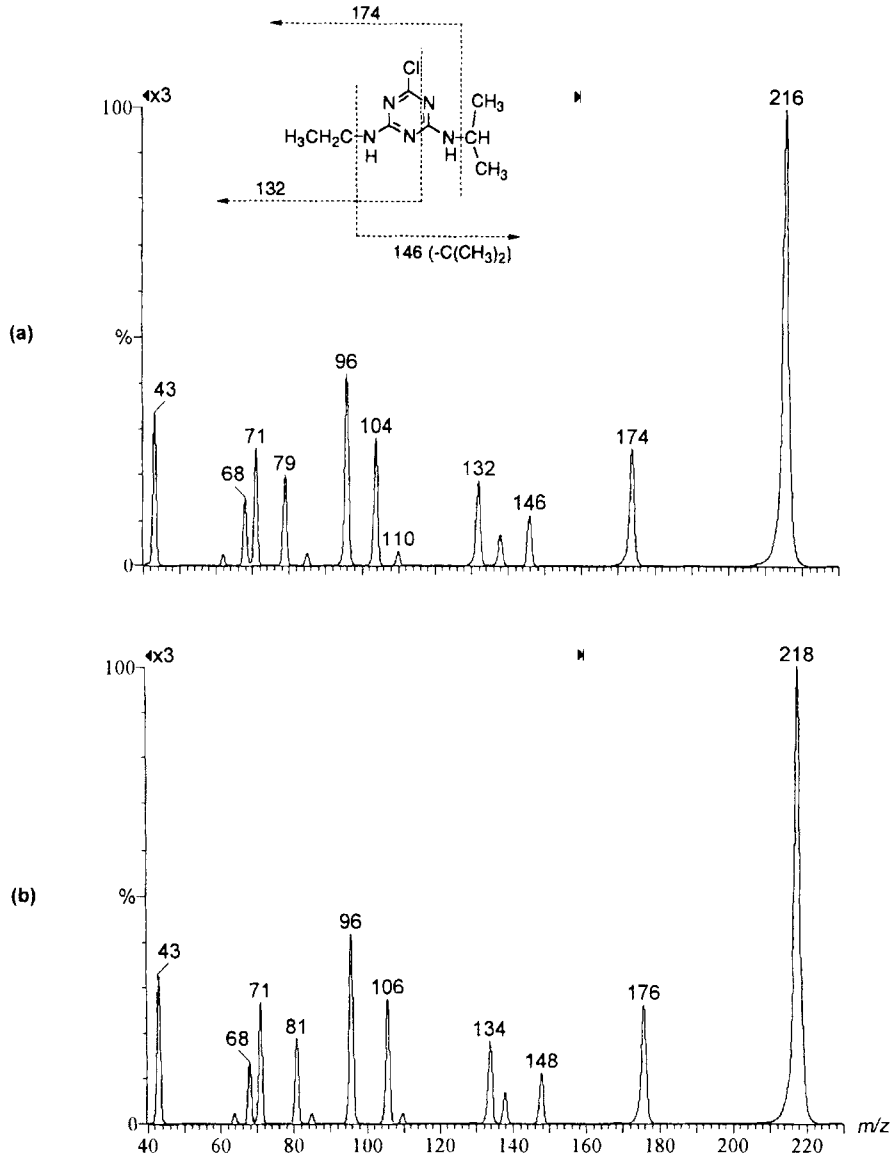


Figure 3. CID fragment ion spectra for atrazine (peak 8): (a) ions of m/z 216 and (b) ions of m/z 218 (^{37}Cl).

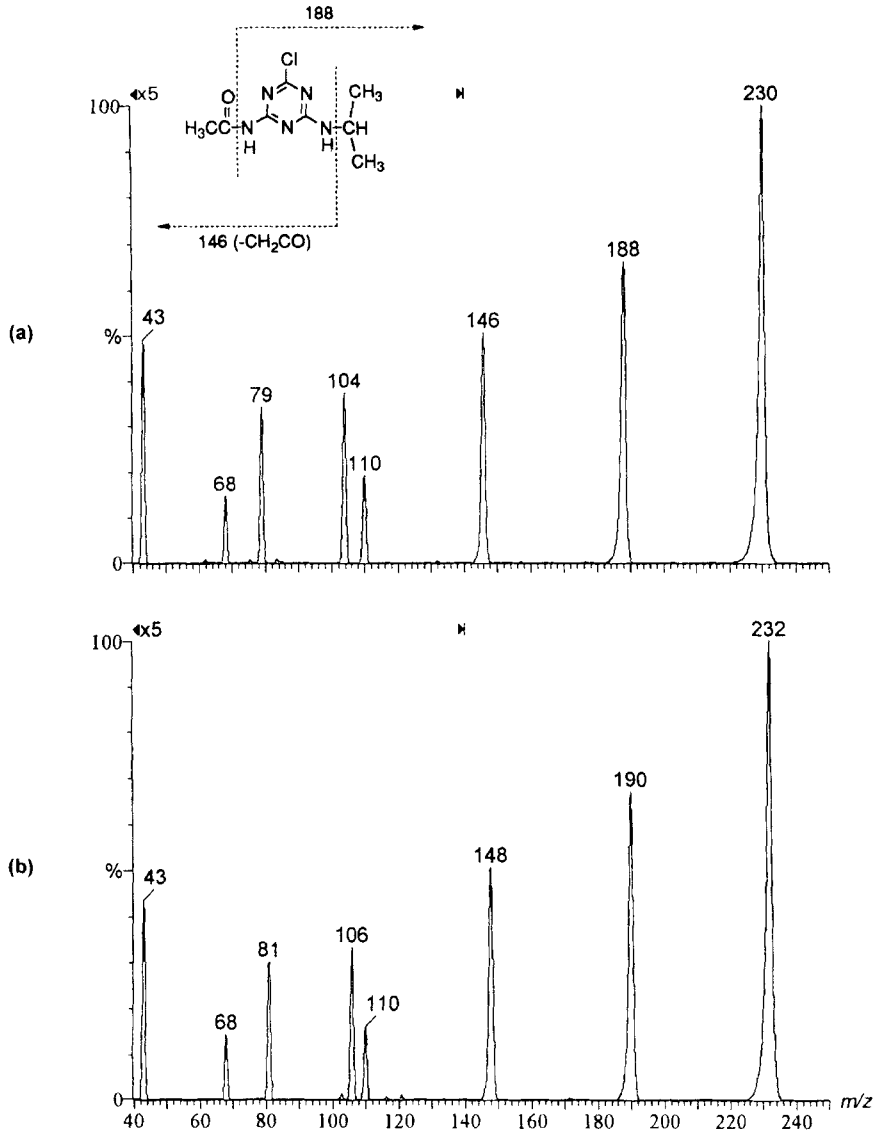


Figure 4. CID fragment ion spectra for 4-acetamido-2-chloro-6-(isopropylamino)-*s*-triazine (peak 7): (a) ions of *m/z* 230 and (b) ions of *m/z* 232 (^{37}Cl).

retaining the Cl atom and ethyl side-chain yields ions at *m/z* 132 (134). Ions at *m/z* 104 (106), 96, 79 (81) and 71 represent ring-cleavage fragments similar to those of atrazine. This product was identified elsewhere by comparison with the spectrum of a synthesized product using direct insertion probe MS.[11]

Peak 4: deisopropylatrazine. Table 1 lists the CID spectra of deisopropylatrazine. It has an abundant parent ion $[M + H]^+$ at *m/z* 174 (176). Loss of the ethyl group yields diaminoatrazine, *m/z* 146 (148). A chlorinated ring-cleavage product with the ethyl group yields the ions at *m/z* 132 (134). The ion at *m/z* 110 represents Cl loss from diaminoatrazine. Ions at *m/z* 104 (106), 96, 79 (81), 71, 68 and 62 (64) are ring-cleavage fragments. The spectrum for the authentic standard was identical with that of the isolated product.

Peak 3: 4-acetamido-2-hydroxy-6-(isopropylamino)-*s*-triazine. Peak 3 showed an abundant parent ion $[M + H]^+$ at *m/z* 212 (Table 1 and Fig. 5). The ^{37}Cl isotope was not present for this compound, indicating that Fenton's reagent treatment eliminated the Cl atom. It is postulated this compound is the dechlorinated analog of peak 7 because of the similar fragmentation patterns. CID spectra gave two losses of *m/z* 42 to yield ions at *m/z* 170 and 128. The first loss of *m/z* 42, representing the ketene, gives the ion at *m/z* 170. The second loss of *m/z* 42 (isopropyl group) yields ion at *m/z* 128. The ion at *m/z* 86 represents a ring-cleavage fragment. To the best of our knowledge, this compound has not been previously identified as an atrazine degradation product.

Peak 2: 4-acetamido-6-amino-2-chloro-*s*-triazine. The protonated parent ion for peak 2 shows a strong molecular ion of *m/z* 188 (190) (Table 1). This was the same molecular ion as deethylatrazine, but the HPLC retention times and mass spectra were different. Peak 2 had a retention time of 6.0 min whereas that of deethylatrazine was 8.8 min. CID spectra of 4-acetamido-6-amino-

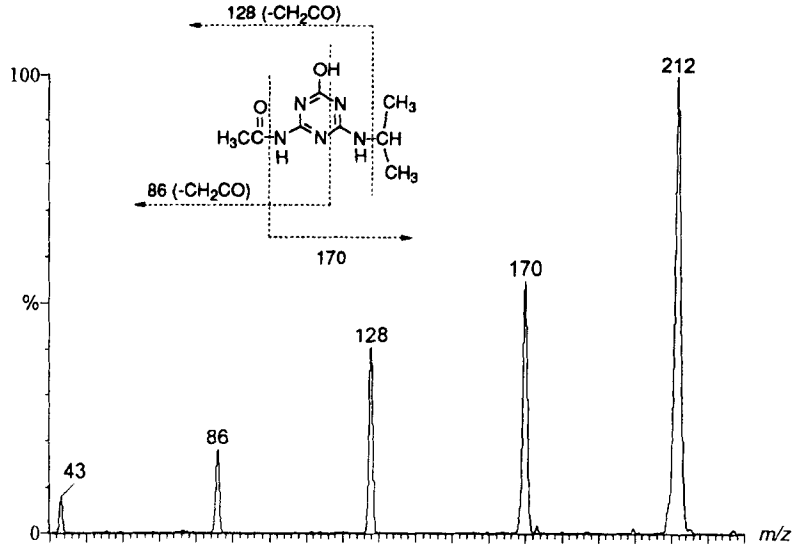


Figure 5. CID fragment ion spectra for 4-acetamido-2-hydroxy-6-(isopropylamino)-*s*-triazine (peak 3).

2-chloro-*s*-triazine showed the loss of *m/z* 42 to give ions at *m/z* 146 (148) resulting from cleavage of the *N*-alkyl side-chain. The *N*-alkyl side-chain consisted of an ethyl group with a C=O in the C(2) position. Other investigators have identified this compound by comparing GC/MS[5,13] or direct insertion probe MS[11] spectra with that of the authentic compound.

HR-EI-MS and GC/MS

HR-EI-MS was applied to peaks 5 and 7 to establish unambiguously the C(2) position of the C=O group in the alkyl side-chain. The HR-EI mass spectra confirmed the structural interpretation based on HPLC/ES-MS/MS and were identical with those obtained for 4-acetamido-2-chloro-6-(ethylamino)-*s*-triazine (peak 5) and 4-acetamido-2-chloro-6-(isopropylamino)-*s*-triazine (peak 7) by other investigators.[11] The HR-EI mass spectra of peak 5 showed abundant parent ions M^+ of *m/z* 215 (217). Cleavage of a methyl group (*m/z* 15) gave ions at *m/z* 200 (202). Loss of the ethyl (*m/z* 28) and ketene (*m/z* 42) moieties gave ions at *m/z* 187 (189) and 173 (175), respectively. Loss of the methyl and ketene yielded ions at *m/z* 158 (160). Further fragmentation gave ions at *m/z* 145 (147), representing loss of both side-chains. Peak 7 had abundant parent ions M^+ of *m/z* 229 (231). Methyl loss gave ions at *m/z* 214 (216). Loss of the ketene or the isopropyl group yielded ions at *m/z* 187 (189). Ions at *m/z* 172 (174) represented cleavage of the ketene and methyl groups. Cleavage of both side-chains yielded ions at *m/z* 145 (147).

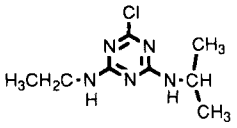
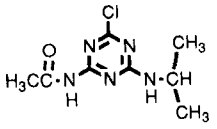
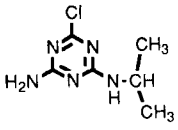
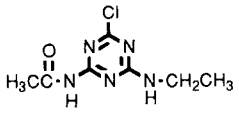
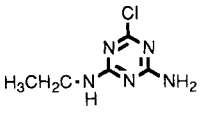
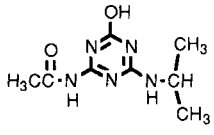
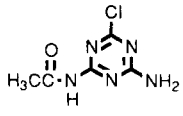
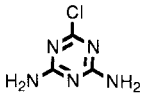
The peak 1 fraction was analyzed by GC/MS because chemical interferences introduced during product isolation rendered the HR-EI mass spectra and HPLC/ES tandem mass spectra uninterpretable. The GC mass spectra showed a strong parent ion M^+ of *m/z* 145 (147), and ions representing the dechlorinated parent compound (*m/z* 110) and ring-cleavage fragments (*m/z* 83 and 69) were also detected. The peak was thus identified as diaminoatrazine and confirmed by comparison with the spectrum of an authentic standard. Structural assignments for all eight peaks based on HPLC/ES-MS/MS, HR-EI-MS and GC/MS and/or co-elution with authentic standards are given in Table 2.

HPLC/ES-MS/MS analysis of atrazine products in mixed samples

Following validation with the eight isolated compounds, the HPLC/ES-MS/MS method was applied for direct analysis of the Fenton's reagent product mixture. Since *m/z* 43 is a common ion for all atrazine degradation products (Table 1), initial experiments were conducted in the parent ion scan mode using *m/z* 43. The results gave five compounds of interest: ions at *m/z* 212, 174 (176), 188 (190), 230 (232), 216 (218) for peaks 3, 4, 6, 7 and 8, respectively. The parent ion for 2,4-dihydroxy-6-amino-*s*-triazine (*m/z* 129) was also detected. In the next experiment, MS/MS was applied to on peaks 1–8 in the mixture. The fragmentation patterns for peaks 2–8 were similar to those obtained when these were analyzed as isolated components. In addition, peak 1 gave clear fragmentation in the mixed sample (Table 1).

Evaluating a technology's effectiveness for remediating atrazine-contaminated materials requires confirmation of parent compound depletion and identification of degradation intermediates. The latter is particularly important when chemical oxidation systems (e.g. Fenton's reagent) are used, since these can generate many degradation products. Chemical oxidation may be 100% effective in degrading atrazine, but still be incomplete in a remediation context if toxic degradation compounds persist at the treatments's end-point. Using the HPLC/UV and HPLC/ES-MS/MS combination we have shown that at Fenton's reagent end-point atrazine is completely transformed to a mixture of partially

Table 2. Atrazine degradation products produced by Fenton's reagent

HPLC peak No.[a]	Structure	Name	Identification method			
			HPLC/ES-MS/MS	HPLC[b]	HR-EI-MS	GC/MS
8		2-Chloro-4-(ethylamino)-6-(isopropylamino)-*s*-triazine(atrazine)	×[c]	×		
7		4-Acetamido-2-chloro-6-(isopropylamino)-*s*-triazine	×		×	
6		4-Amino-2-chloro-6-(isopropylamino)-*s*-triazine(deethylatrazine)	×[c]	×		
5		4-Acetamido-2-chloro-6-(ethylamino)-*s*-triazine	×		×	
4		6-Amino-2-chloro-4-(ethylamino)-*s*-triazine (deisopropylatrazine)	×[c]	×		
3		4-Acetamido-2-hydroxy-6-(isopropylamino)-*s*-triazine	×			
2		4-Acetamido-6-amino-2-chloro-*s*-triazine	×			
1		2-Chloro-4,6-diamino-*s*-triazine (diaminoatrazine)	×	×		×[c]

[a] See Fig. 2(a).
[b] Co-elution with authentic standard.
[c] Spectra matched authentic standard.

dealkylated products. However, because some of these daughter compounds are chlorinated, Fenton's reagent treatment may not effect total chemical detoxification. These results indicate that, for remediating atrazine-contaminated wastes, Fenton's reagent may be best used in combination with biological treatment: although the chlorinated atrazine products generated by Fenton's reagent may retain atrazine's mammalian toxicity potential, they are readily decomposed microbiologically.

CONCLUSIONS

This study has demonstrated the applicability of HPLC/ES-MS/MS for the identification of atrazine

products. Parent ions in mixed samples were easily identified and CID spectra of analytes in the product mixture were comparable to those obtained with the isolated component. The reliability of the HPLC/ES-MS/MS results was verified by comparison with traditional MS methods. Since atrazine products are typically analyzed by HPLC/UV, HPLC/ES-MS/MS should serve as the method of choice for the identification of a wide spectrum of atrazine derivatives. This method allows investigators to compare peak retention times and analyze more polar products, making fraction collection unnecessary. For remediation system development, the compatibility of the HPLC/UV and HPLC/ES-MS/MS techniques can be of particular value by allowing rapid evaluation of treatment endpoints.

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