Detection of nitroaromatic and cyclic nitramine compounds by cyclodextrin assisted capillary electrophoresis quadrupole ion trap mass spectrometry

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Abstract

An Agilent [3D]CE capillary electrophoresis system using sulfobutylether-β-cyclodextrin (SB-β-CD)–ammonium acetate separation buffer pH 6.9 was coupled to a Bruker Esquire 3000+ quadrupole ion trap mass detector via a commercially available electrospray ionization interface with acetonitrile sheath flow. The CE–MS system was applied in negative ionization mode for the resolution and detection of nitroaromatic and polar cyclic or caged nitramine energetic materials including TNT [2,4,6-trinitrotoluene, formula mass (FW) 227.13], TNB (1,3,5-trinitrobenzene, FW 213.12), RDX (hexahydro-1,3,5-trinitro-1,3,5-triazine, FW 222.26) HMX (octahydro-1,3,5,7-tetranitro-1,3,5,7-tetrazocine, FW 296.16), and CL-20 (2,4,6,8,10,12-hexanitro-2,4,6,8,10,12-hexaazaisowurtzitane, FW 438.19). The CE–MS system conformed to the high-performance liquid chromatography with ultraviolet absorbance detection (HPLC–UV) and HPLC–MS reference methods for the identification of energetic contaminants and their degradation products in soil and marine sediment samples.
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1. Introduction

The overproduction of military explosives and propellants has resulted in their inadvertent release to soil and aquatic environments [1], and the potential toxicity of these materials and their degradation products has prompted governments to initiate programs for the identification and characterization of contaminated sites [2,3]. The most frequently occurring soil contaminants (TNT: 2,4,6-trinitrotoluene, HMX: octahydro-1,3,5,7-tetranitro-1,3,5,7-tetrazocine, and RDX: hexahydro-1,3,5-trinitro-1,3,5-triazine) are analyzed by C_{18} reverse phase high-performance liquid chromatography with ultraviolet absorbance detection (HPLC–UV) [US Environmental Protection Agency (EPA) Method EPA 8330, Nitroaromatics and explosives in soil][4]. This method uses equipment readily available to analytical laboratories, and offers μg/l detec-

tion limits, but requires known reference analytical standards for unequivocal peak identification. The method is therefore somewhat problematic for the discovery of novel degradation products. The use of atmospheric electrospray ionization-mass spectrometry (ESI-MS) [5–10] or atmospheric pressure chemical ionization mass spectrometry (APCI-MS) [11–14] with HPLC has greatly assisted in the identification of new intermediates and the definition of environmental degradation pathways for energetic materials [15–20]. Frequently, however, the C_{18} reversed-phase HPLC system does not resolve polar metabolites, and analyte identification is heavily dependent on mass spectra. Alternative separation methods, including gas chromatography [21–23], CN- or C_8-reversed-phase HPLC [24], ion exclusion HPLC [25,26], and micellar electrokinetic chromatography [27] are consequently of interest for use with mass spectrometry for the improved resolution and identification of energetic contaminants. Cyclic nitramine explosives of moderate polarity are resolved using sulfobutylether-β-cyclodextrin (SB-β-CD) assisted capillary

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electrophoresis [28] and here we report on the coupling of this separation method to a quadrupole ion trap mass spectrometer for the identification of frequently encountered explosives and their degradation products in environmental samples.

2. Experimental

2.1. Materials

CL-20 (2,4,6,8,10,12-hexanitro-2,4,6,8,10,12-hexaazaisowurtzitane, purity 99.3% as determined by HPLC, 95% epsilon form as determined by IR) was provided by Thiokol Propulsion (Brigham City, UT, USA). HMX, RDX, and TNX (hexahydro-1,3,5-trinitroso-1,3,5-triazine, purity of all chemicals >99%) were provided by Defense Research and Development Canada (DRDC), Valcartier, Canada. MNX (hexahydro-1-nitroso-3,5-dinitro-1,3,5-triazine, purity 98%) was obtained from SRI International (Menlo Park, CA, USA). TNT and TNB (1,3,5-trinitrobenzene) analytical standards were purchased from Supelco (Oakville, Canada). Contaminated soil from an explosive manufacturing site (Produits Chimiques Expro, Valleyfield, Canada) was obtained from the BRI applied ecotoxicology group (Montreal, Canada). Contaminated marine sediment core samples (Ordinance range UXO, Oahu Island, HW, USA) were provided by the US Naval Research Laboratory (Arlington, VA, USA). Advasep 4 (sulfobutylether-β-cyclodextrin, sodium salt) [29], was purchased from Cydex (Overland Park, KS, USA). CE grade ammonium acetate was purchased from Agilent Technologies (New Castle, DE, USA). Formic acid was purchased from Fluka (Munich, Germany). HPLC grade acetone and 2-propanol were purchased from Fisher Scientific (Montreal, Canada). Acetonitrile and methanol were obtained from Mallinckrodt Baker (Phillipsburg, NJ, USA). Ethyl acetate was provided by BDH (Poole, UK). Aqueous solutions were prepared using deionized Milli-Q plus (Millipore) water. All other chemicals (HCHO, HCl, NaOH, H_3PO_4) used were obtained from Aldrich (Oakville, Canada). Standard analytical solutions (1000 mg/l) were prepared by adding crystals of analyte to acetonitrile with verification of concentration by HPLC as described earlier [16]. Samples were prepared by diluting analytical standards or soil extracts in 10 mM ammonium acetate buffer pH 6.9.

2.2. CE system

CE separations were carried out using an Agilent ^{3D}CE instrument (Agilent, Waldbronn, Germany). Fused silica capillaries (total length (L) 90 cm, effective length to UV detection window (l) 21.5 cm, internal diameter 50 μm) were purchased from Agilent. Unless otherwise indicated, separations were carried out at 30 kV and 25 °C. Sample injections were performed hydrodynamically (50 mbar, 1–10 s). The separation buffer was 10 mM sulfobutylether-β-cyclodextrin,

10 mM ammonium acetate pH 6.9. New capillaries were conditioned by flushing with water (5 min), 1 M HCl (5 min), water (5 min), 1 M NaOH (5 min) and separation buffer (15 min). Following this the capillaries were conditioned electrophoretically by applying 30 kV for 15 min with subsequent replenishment of anodic and cathodic buffer vials. In between runs the capillaries were rinsed for 30 s in 10 mM ammonium acetate, and then flushed with separation buffer for 3 min.

2.3. MS system

A Bruker Esquire 3000 plus ion trap spectrometer (Bruker-Daltonics, Boston, MA, USA) equipped with an Agilent atmospheric electrospray ionization source (Agilent, Waldbronn, Germany) was employed for mass detection. The electrospray was operated in negative ion mode to produce mainly deprotonated molecular mass ions $[M-H]^{-1}$. Nitrogen was used as drying gas at 150 °C, with a pressure of 816 mbar (12 psi) and at a flow rate of 7 l/min. Sheath liquid was pumped to the atmospheric pressure interface using an Agilent model 1100 isocratic pump with a 100:1 flow splitter to deliver 6 μl/min. The capillary voltage was set at 6000 V with an end plate offset of −500 V. Observed electrospray currents ranged from 5 to 17 nA. The scanning mass to charge range was 40 to 600 *m/z* with a scanning speed of 13000 *m/z* per second. The resulting baseline peak width resolution was 0.6 U. The maximum accumulation time was 50 ms.

2.4. Soil and sediment analysis

Soil and sediment samples were extracted by sonication in acetonitrile in accordance with EPA Method 8330 [4]. The HPLC–UV analyses were performed as described by Fournier et al. [30]. Sediment samples were also analyzed as follows using a Hewlett-Packard 6890 gas chromatograph equipped with electron-capture detection (GC-ECD). Acetonitrile extracts (2 μl) were injected onto a Restek (Bellefonte, PA, USA) Rtx-TNT capillary column (6 m × 0.530 mm, 1.50 μm) with helium as carrier gas at a flow rate of 60 ml/min and 680 mbar (10 psi). The column was initially held at 100 °C for 2 min, then raised to 200 °C at a rate of 10 °C/min. The temperature was then ramped at a rate of 20 °C/min to 230 °C and maintained there for 13.5 min. The injector and detector temperatures were 250 and 300 °C, respectively. A Restek 8095 calibration mix A was employed for reference calibration. The instrumental quantification limits (μg/l) for this method were as follows: TNT, 0.6; RDX, 8.9; HMX, 10.9; 1,3,5-TNB, 1.9; 2,6-DNT, 0.3; 2,4-DNT, 1.6.

3. Results and discussion

3.1. Mass spectra of analytes in the presence of cyclodextrin

Beta cyclodextrins are widely known to be cyclic oligosaccharides containing seven glucopyranose units

linked exclusively through alpha (1–4) saccharide bonds to form a single ring. The cyclic orientation provides a truncated cone structure that is hydrophilic on the exterior and lipophilic at the interior cavity. Native cyclodextrins are of limited aqueous solubility or inclusion capacity, so the parent macrocycles are frequently modified by electrophilic persubstitution at the available hydroxyl groups of carbons 2, 3, or 6 of each glucopyranose subunit to produce charged or alkylated derivatives [31–33]. Selective modification at the specified hydroxyl groups is difficult, and a range of isomers is usually obtained using the above methods. It is theoretically possible to achieve a degree of substitution of 21 in the case of beta cyclodextrin, with numerous possibilities for the synthesis of positional or regioisomers. Commercially available modified cyclodextrins are therefore obtained as a mixture of isomers with varied degrees of substitution, and this has an effect on both pseudostationary phase electrophoretic mobilities, and on the observed mass spectrum.

The cyclodextrin applied in this study was Advasep 4 (Cydex, Overland Park, KS, USA) [29], a mixture of sulfobutyl ether derivatives of beta cyclodextrin (SB-β-CD) that vary in the degree of substitution from one to nine with a claimed overall effective average degree of substitution (d.s.) of 4. The low pK_a of the acid groups at the end of the carbon butyl chains causes the side chains to be charged at all pH values >2 ($pK_a < 1$). The presence of such diverse highly charged species in the CE separation buffer results in complex, but well defined CE–MS background spectra. The spectrum observed for an infusion of 10 mM ammonium acetate, 10 mM SB-β-CD (pH 6.9) separation buffer and acetonitrile sheath fluid is presented in Fig. 1A. The tentative identities of the observed molecular mass ions are listed in Table 1. Mass-to-charge values (m/z) were found for cyclodextrin derivatives with degrees of substitution of 4, 5, 6, and 7 (m/z values 418.7, 362.1, 324.4 and 297.4, respectively). Spectral lines are also observed for the monosodium adducts of cyclodextrin derivatives with degrees of substitution 5–7 (m/z values

458.3, 393.4 and 350.6, respectively). Similar mass spectra for preparations of SB-β-CD were presented by Grard et al. [34].

Various forms of ion spray mass spectrometry have been applied to the analysis of explosives by many workers. Using electrospray ionization triple-quadrupole MS on acetonitrile solutions of a wide range of explosives, Casetta and Garofolo [6] noted that TNT and other resonance stabilized nitroaromatic compounds formed deprotonated anionic $[M - H]^{-1}$ molecular mass ions, while explosives lacking in acidic protons tended to form acetate $[M + 59]^{-1}$ or formate $[M + 46]^{-1}$ mass ion adducts. The presence of acetate or formate was attributed to impurity of the acetonitrile infusion solvent. Fig. 1B provides the mass spectrum for an infusion of separation buffer containing TNT (FW 227.13), TNB (FW 213.12), RDX (FW 222.26), HMX (FW 296.16), and CL-20 (FW 438.19) at 10 mg/l concentration. The dominant mass ion signals observed in the spectrum are those of the substituted cyclodextrin derivatives (i.e. m/z 362.1, etc.). The aromatic explosives TNT and TNB produced signals at 226 and 212 m/z corresponding to their deprotonated parent ions $[TNT - H]^{-1}$ and $[TNB - H]^{-1}$, and signals at m/z 244 and 230 representing their deprotonated ion water adducts $[TNT - H + H_2O]^{-1}$ and $[TNB - H + H_2O]^{-1}$, respectively. Acetate adducts for RDX and HMX were indicated by the observation of signals at m/z values of 281 ($[RDX + CH_3CO_2]^{-1}$) and 355 ($[HMX + CH_3CO_2]^{-1}$), respectively. A weak signal observed for CL-20 at m/z 500 was tentatively assigned to the composition $[CL-20 + NO_3]^{-1}$, as the signal for NO_3^- (m/z 62) is observed in the mass spectra for Cl-20 and other analytes (Figs. 1B and 2). CL-20 was reported to undergo denitration in aqueous or water–acetonitrile solutions at neutral [35] or alkaline [18] pH, to yield nitrite anion (NO_2^-, m/z 46), but not nitrate [18,35–37], and this reaction is accelerated in the presence of dimethyl-β-cyclodextrin [18,35]. The SB-β-CD CE separation buffer at pH 6.9 is therefore not well suited to the detection of CL-20. It is interesting to note that nitrate for-

Table 1
Identities of potential and observed ion fragments using negative mode electrospray ionization with 10 mM sulfobutylether-β-cyclodextrin (SB-β-CD) in 10 mM ammonium acetate pH 6.9 as capillary electrophoretic buffer and 100% acetonitrile as sheath fluid

| Tentative mass ion	Degree of substitution (d.s.)	Charge (z)	Molecular mass (m)	m/z calculated	m/z observed
$[CH_3CO_2]^{-1}$	–	1	59	59	59
$[2CH_3CO_2 + Na]^{-1}$	–	1	141	141	141
$[SB_1 - H]^{-1}$	1	1	1269	1269	
$[SB_2 - 2H]^{-2}$	2	2	1404	702	
$[SB_3 - 3H]^{-3}$	3	3	1539	513	513
$[SB_4 - 4H]^{-4}$	4	4	1674	418.5	419.3
$[SB_5 - 5H]^{-5}$	5	5	1809	361.8	362.1
$[SB_6 - 6H]^{-6}$	6	6	1944	324	324.4
$[SB_7 - 7H]^{-7}$	7	7	2079	297	297.4
$[SB_8 - 8H]^{-8}$	8	8	2214	276.75	277
$[SB_9 - 9H]^{-9}$	9	9	2349	261	
$[SB_5 - 5H + Na]^{-4}$	5	4	1832	458	458.5
$[SB_6 - 6H + Na]^{-5}$	6	5	1961	392.5	393.1
$[SB_7 - 7H + Na]^{-6}$	7	6	2102	350.3	350.6
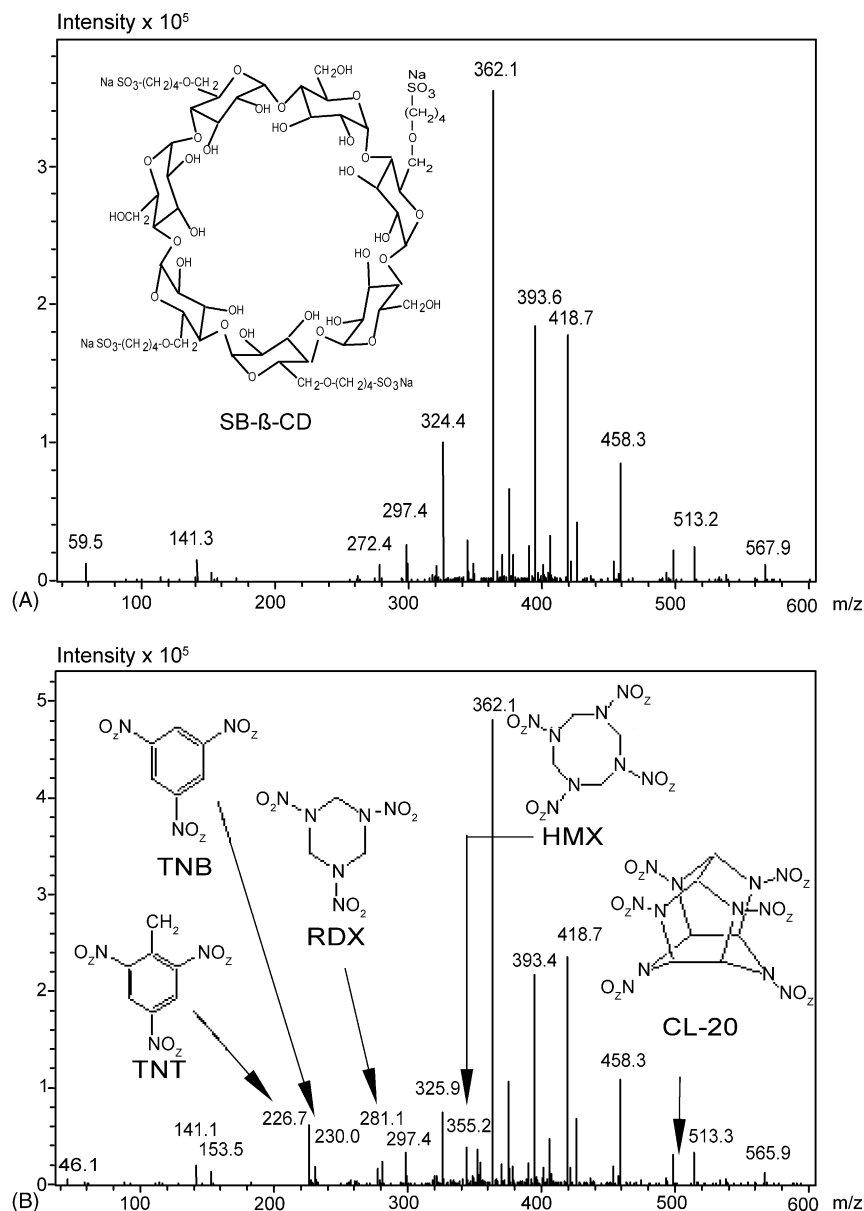
Fig. 1. (A) Mass spectrum for a 10 mM infusion of Advasep 4 sulfobutylether-β-cyclodextrin SB-β-CD (structure shown as inset, d.s. 4) in 10 mM ammonium acetate pH 6.9, with acetonitrile sheath fluid. (B) Mass spectrum for the above infusion containing 10 mg/l TNT, TNB, RDX, HMX and CL-20 (structures shown as inset) with mass ion intensities indicated respectively for *m/z* values of 226, 230, 281, 355, and 500.

mation from nitrite was reported for the photodegradation of RDX [17] and that a dominant mass signal of *m/z* 500 is observed in the LC–MS spectra reported for the photodegradation of CL-20 [38].

The presence of highly charged cyclodextrins in the electrospray aerosol may affect the identification of target explosive analytes in at least two respects: (1) The presence of high molecular weight, non-volatile ions in high concentration will inhibit the passage of target analytes to the vapour phase, and (2) If the analyte in question forms a highly stable inclusion complex with the cyclodextrin, then the observed mass signal will be that of the non-covalent inclusion complex and not the parent molecular ion. Bakhtiar and Bulusu [39] examined

the complexation of HMX, RDX, and 1,3,3-trinitroazetidine (TNAZ) with neutral cyclodextrins using positive ionization electrospray triple quadrupole MS and observed complexes for the cationic (Na$^+$, NH$_4^+$, K$^+$) adducts of RDX and neutral β- and γ-cyclodextrins in infusions containing 250 ppm RDX and cyclodextrin. The tentative β-CD complexes and mass to charge ratios were [2β-CD + RDX + 2NH$_4$ + K]$^{2+}$ (*m/z* 1283), [β-CD + RDX + NH$_4$]$^+$ (*m/z* 1375), [β-CD + RDX + Na]$^+$ (*m/z* 1380), and [β-CD + RDX + K]$^+$ (*m/z* 1396). Similar phenomena were reported by Srinivasan and Bartlett [40] for the CE–MS analysis of neutral β-cyclodextrins complexed with barbiturate ion.

Table 2
Average peak height intensities of extracted ions (cps) observed for 10 mg/l injections (10 s 50 mbar) of selected energetic compounds using different sheath fluids (standard deviation provided in brackets, $n = 5$)

Sheath fluid	Isopropanol	Ethanol	Methanol	Ethyl acetate	Acetone	Acetonitrile	Water
Compound							
CL-20 (m/z 500)	N.D.	N.D.	N.D.	N.D.	87 (79)	370 (85)	N.D.
TNT (m/z 226)	1880 (228)	342 (112)	623 (287)	3912 (351)	17165 (1030)	36903 (559)	N.D.
TNB (m/z 230)	1670 (170)	187 (320)	498 (172)	23439 (1523)	22473 (1143)	42178 (1898)	N.D.
HMX (m/z 355)	8387 (528)	237 (281)	10169 (543)	137 (162)	24724 (506)	23690 (1092)	N.D.
RDX (m/z 281)	9664 (473)	871 (138)	12853 (753)	518 (751)	18872 (430)	21635 (1073)	N.D.
MNX (m/z 265)	9444 (483)	181 (243)	5069 (101)	4178 (376)	28592 (202)	35178 (372)	N.D.
TNX (m/z 233)	1543 (338)	9664 (453)	18887 (491)	851 (383)	61764 (982)	54970 (1923)	N.D.

A previous CE–UV study [28] indicated that nitroaromatic and cyclic nitramine explosives associated weakly with SB-β-CD, and it is unlikely that analyte–cyclodextrin complexes would be stable in the presence of acetonitrile sheath fluid. Nevertheless, the scan ranges were extended with limits of 40–1000 m/z to detect possible noncovalent complexes with the distribution of substituted cyclodextrins observed in the separation buffer mass spectrum. In this case the expected m/z values for inclusion complexes [i.e. m/z values of 492.5 and 474 for HMX and RDX complexes respectively, with SB-β-CD (d.s. = 4)] were not observed. Attempts to observe the negative ions for TNT ([M]$^{-1}$; m/z 227), RDX ([M + NO$_2$ − H]$^{-1}$; m/z 267) and HMX ([M + NO$_2$ − H]$^{-1}$; m/z 341) as reported by Yinon et al. [7] were also unsuccessful.

A separation of CL-20, TNB, TNT, HMX, and RDX analytical standards (10 mg/l, hydrodynamic injection volume 96 nl, actual analyte mass amount 96 pg) in 10 mM ammonium acetate is shown as Fig. 2. The overall capillary length (L) was 90 cm and the effective length to the UV window (l) was 21.5 cm. The UV trace (230 nm) revealed the electroosmotic front (EOF) to arrive at the UV detection window after 2.5 min, and at 10.3 min a corresponding dip in capillary current from a steady state value of 23 μA signified the passage of the EOF to the electrospray ionization (ESI) interface. Peaks for the EOF and analytes arrived at the interface earlier than predicted using total length/effective length ratios. This slight increase in the bulk fluid velocity was due to the entrainment of all fluids exiting the electrospray needle by the nitrogen drying gas which flowed at 7 l/min (816 mbar and 150 °C). The capillary internal diameter was 50 μm and

the corresponding average flow rate from the capillary was 0.17 μl/min. The sheath fluid flow rate was set at 6 μl/min, which resulted in a 1/35 dilution of the CE flow in acetonitrile at the ESI interface. Under these conditions the total ion current decreased from 5×10^5 counts per second (cps) to a steady state value of 2.6×10^5 cps after 2 min. Extracted adduct or molecular ion traces with corresponding peak mass spectra (baseline mass spectra subtracted) for CL-20, TNB, TNT, HMX, and RDX are respectively indicated for m/z values of 500, 230, 226, 355 and 281.

3.2. Sheath flow liquid selection

The effect of sheath fluid choice on the negative electrospray ionization of cyclic nitramines resolved in 30 kV electrophoretic separations (10 mM SB-β-CD/10 mM ammonium acetate pH 6.9) was examined using the polar organic solvents acetone, acetonitrile, 2-propanol, ethyl acetate, ethanol, methanol, and water. The analytes in question (CL-20, TNT, TNB, HMX, RDX, MNX, TNX) were observed to ionize best in acetonitrile and acetone (Table 2) which possessed the highest volatilities and lowest viscosities of the selected solvents (Table 3). Ethyl acetate, which also has a high volatility and relatively low viscosity, apparently was not sufficiently polar to maintain analyte solubility. Generally, when applying negative ionization electrospray, ionization increases with sheath liquid volatility, and decreases with respect to solvent surface tension and viscosity. The dielectric constant (polarity) of the sheath liquid must also be sufficient to maintain the solubilization of analytes following droplet separation from the Taylor cone and desolvation

Table 3
Physical properties of selected sheath fluids (20 °C) [44]

| Sheath fluid	Vapour pressure (Torr)	Viscosity (cP)	Surface tension (dyn/cm)	Density (g/mL)	Polarity index	Dipole moment (Debye)	Dielectric constant
Isopropanol	33	2.3	18.3	0.785	3.9	1.66	19.9
Ethanol	43.9	1.2	23.75	0.9	5.2	1.69	24.55
Methanol	97.7	0.59	22.55	0.791	5.1	2.87	32.7
Ethyl acetate	73	0.45	23.75	0.9	4.4	1.88	6.02
Acetone	184.5	0.32	23.32	0.79	5.1	2.69	20.7[a]
Acetonitrile	88.8	0.37	19.1	0.782	5.8	3.44	37.5
Water	23.78	1	72.8	0.997	9	1.84	80.1

[a] 25 °C.
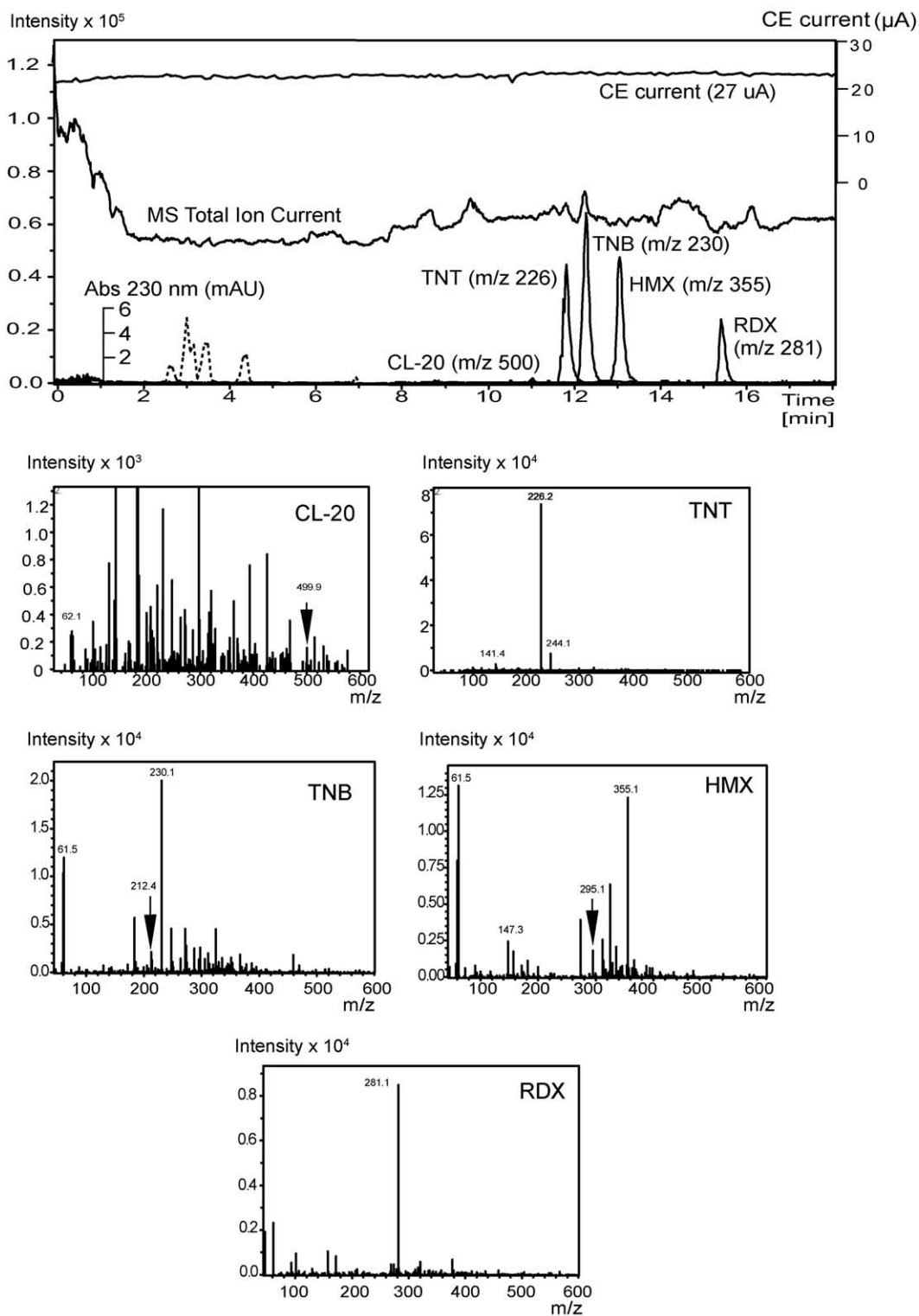
Fig. 2. CE–MS separation (30 kV, 10 mM acetate, 10 mM Advasep 4) with mass spectra for a 10 mg/l injection (10 s, 50 mbar, injection volume 56 nl) of TNT, TNB, RDX, HMX and CL-20 (actual mass amount: 96 pg) in 10 mM acetate pH 6.9. CE–UV trace (230 nm) as indicated by broken line. Extract ion traces (m/z values 226, 230, 281, 355, and 500, respectively) indicated by solid lines.

[41]. The aprotic nature of acetone and acetonitrile may act to enhance acetate adduct formation and ionization of the cyclic nitramines. The solubility of SB-β-CD in the sheath fluid appears to have little effect on analyte ionization, as evidenced by the poor performance of sheath fluids (methanol, ethanol, and water) in which SB-β-CD is known to be soluble [42]. The various sulfobutylether-β-cyclodextrin derivatives present in Advasep-4 are sufficiently soluble in acetonitrile to permit the passage of intact deprotonated parent ions $[SB_n\text{-}nH]^{-n}$ to the mass detector (Fig. 1, Table 1).

3.3. Effect of buffer components on signal intensity

Figs. 3 and 4 respectively represent the effects of ammonium acetate and cyclodextrin concentration in the separation buffer on analyte ionization. Fig. 3 indicated that increases in separation buffer acetate concentration decreased the ionization of nitroaromatic explosives, primarily due to competition with acetate ions at the electrospray droplet interface as indicated by the increased acetate extracted ion intensities (m/z 59) observed. Ion pairing of ammonium with $[M - H]^{-1}$ nitroaromatic parent anions may also decrease their abundance in the electrospray. The signal intensities for the nitramine explosives increased with acetate concentrations up to 20 mM, and indicated that increased acetate concentration served to enhance the formation of acetate adducts with these non-aromatic compounds at the electrospray interface. At acetate concentrations higher than 20 mM the ionization of cyclic nitramines gradually decreased in the same manner as for the aromatic analytes. This trend is an indication that once adduct ion formation is maximized, the passage of adduct ions to the gas phase will vary as a function of their mole fraction at the electrospray droplet surface.

As indicated in Fig. 4, three-fold increases in signal intensity were observed for the nitroaromatic compounds TNT
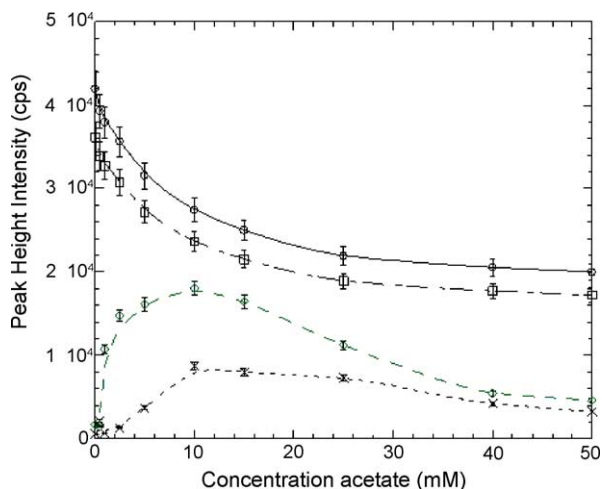

Fig. 3. Peak height intensities of selected analytes as a function of ammonium acetate concentration. All other conditions as in Fig. 2. Symbols: (○) TNB, (□) TNT, (◇) HMX, (×) RDX. (Error bar = 1 standard deviation, $n = 5$.)
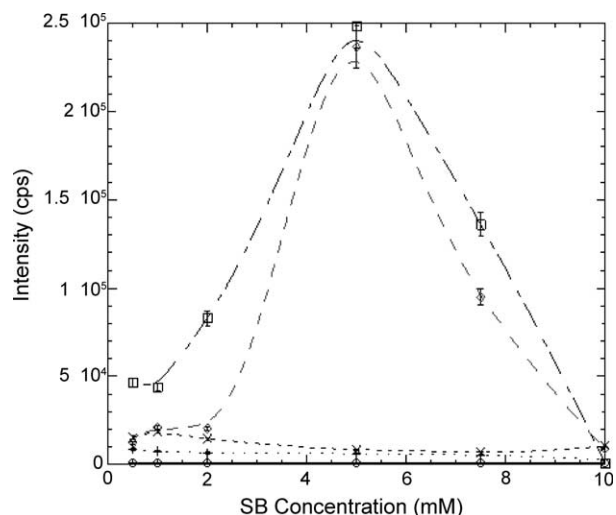

Fig. 4. Peak height intensities of selected analytes with variation in cyclodextrin concentration. All other conditions as in Fig. 2. Symbols: (○) EOF, (□) TNT, (◇) TNB, (×) HMX, (+) RDX. (Error bar = 1 standard deviation, $n = 5$.)

and TNB when the cyclodextrin concentration was increased from 2 to 5 mM. Signal intensities for HMX and RDX were relatively insensitive to increased cyclodextrin concentration in the range of 0.5–10 mM. The increase in signal observed for the aromatic compounds may be attributed to their 1000-fold greater volatility in comparison with HMX and RDX, as any improvement in the resolution of nitroaromatic compounds will be amplified by greater ionization efficiency which is a function of analyte volatility.

3.4. Application to environmental samples

The practicality of the CE method for the analysis of real samples was examined using samples analyzed by other available methods (HPLC–UV, HPLC-MS, GC-ECD). Generally, the extracted molecular ion response curves for quadrupole ion trap spectrometers increase hyperbolically with ion concentration and saturate to a constant value at the upper analyte concentration limit. Consequently, the quantitative linear dynamic range for selected quantification standards using the CE–MS method was narrow; varying between 10 mg/l and the instrumental quantification limit (0.05 mg/l). The observed instrumental limits of detection and quantification for the CE–MS method and for the validated HPLC–UV reference method (EPA 8330) are compared in Table 4. Triple quadrupole and time of flight mass detectors are recognized to have greater dynamic ranges in comparison with ion traps, and to be more suitable for quantification, but the multiple MS (MS^n) capability of the latter instruments facilitates degradative product indentification [43]. However, the dominant factor in MS detector selection is often availability.

A CE–MS electropherogram for the extract (acetonitrile sonication) of explosive contaminated marine sediment is shown as Fig. 5. An extracted ion trace at m/z 181 revealed peaks observed at 15.2 and 16.3 min, and were respectively attributed to the deprotonated parent ions ($[M - H]^{-1}$) of 2,6-

Table 4
Instrumental detection limits (IDL)[a] and instrumental quantification limits (IQL)[b] for selected reference standards with cyclodextrin assisted CE–MS and with C_{18} HPLC–UV (Method 8330)

Method	CL-20	TNT	TNB	HMX	RDX	TNX	MNX
IDL (mg/l)							
CE–MS	0.50	0.025	0.025	0.025	0.025	0.025	0.025
HPLC–UV	0.005	0.003	0.002	0.011	0.008	0.005	0.005
IQL (mg/l)							
CE–MS	5	0.05	0.05	0.05	0.05	0.05	0.05
HPLC–UV	0.010	0.013	0.007	0.006	0.028	0.050	0.050

[a] The minimum concentration applied at which peak height was distinguished as greater than three times the baseline variation ($n > 5$).
[b] The minimum concentration applied at which the standard error of the mean was less than 5% ($n > 5$).

dinitrotoluene (2,6-DNT) and 2,4-dinitrotoluene (2,4-DNT). The C_{18} HPLC–UV analysis of this extract also indicated the presence of 2,6-DNT and 2,4-DNT, but with incomplete resolution of the two compounds, possibly due to matrix components. Analysis of the extract using GC-ECD identified 2,4-DNT and 2,6-DNT at concentrations of 0.716 and 0.095 mg/kg. The CE-MS method therefore conformed to the other available methods for the identification of explosives and their degradation products.

Fig. 6 shows the electropherogram for an extract of contaminated soil obtained from an ammunition manufacturing site. HPLC–UV analysis revealed that this extract contained 34 mg/l of TNX (hexahydro-1,3,5-trinitroso-1,3,5-triazine) as confirmed using a reference standard, and quantities of tetranitroso-HMX (4NO-HMX; octahydro-1,3,5,7-tetranitroso-1,3,5,7-tetrazacene), and trinitroso-HMX (3NO-HMX; octahydro-1-nitro-3,5,7-trinitroso-1,3,5,7-tetrazacene), compounds for which no
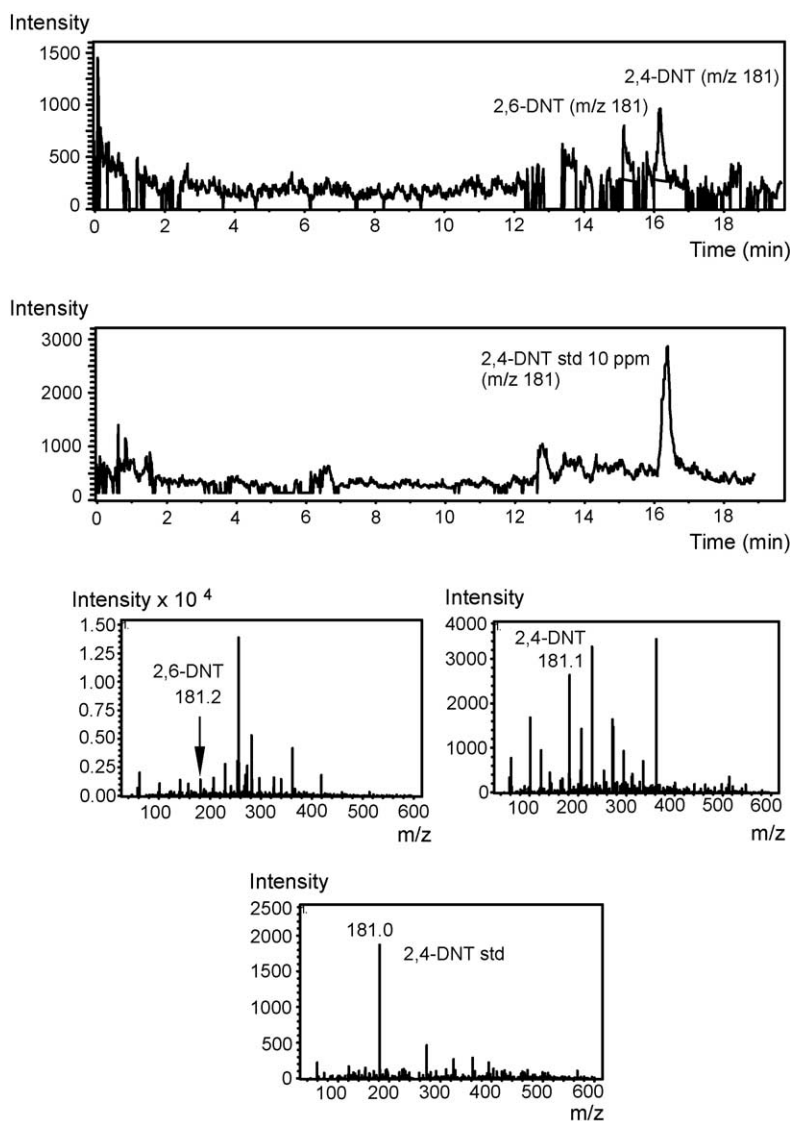

Fig. 5. Extracted ion electropherograms and mass spectra for 2,6-dinitrotoluene and 2,4-dinitrotoluene obtained from acetonitrile extract injections (10 s, 50 mbar) of contaminated marine sediment.
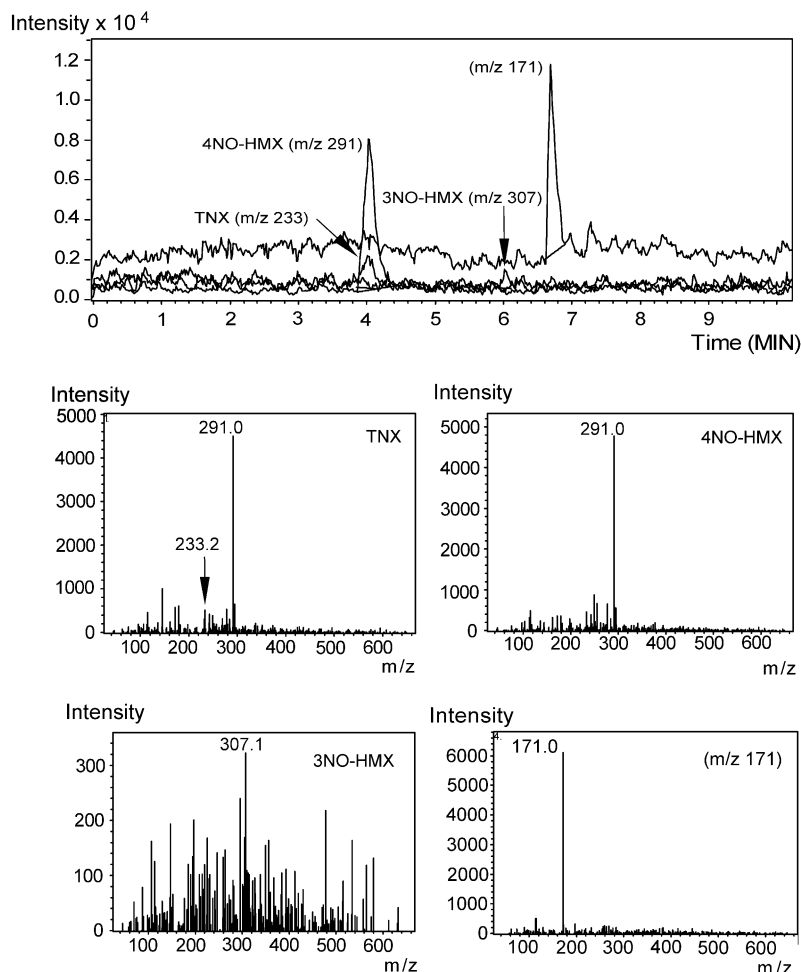
Fig. 6. Extracted ion electropherograms for hexahydro-1,3,5-trinitroso-1,3,5-triazine (TNX), octahydro-1,3,5-trinitroso-7-nitro-1,3,5,7-tetrazocine (3NO-HMX), octahydro-1,3,5,7-tetranitroso-1,3,5,7-tetrazocine (4NO-HMX) obtained from acetonitrile extracts of soil from an explosive manufacturing site.

commercial analytical standards were available. The electropherogram for this sample has peaks for extracted ions with mass to charge ratios of 233 (TNX, acetate adduct), 291 (4NO-HMX, acetate adduct), 307 (3NO-HMX, acetate adduct). A peak observed at 6.7 min (m/z 171) was initially hypothesized to be a diacetyl sodium adduct ion of formaldehyde $[HCHO + 2(CH_3CO_2) + Na]^{-1}$. Diacetyl sodium adduct ions $[2(CH_3CO_2) + Na]^{-1}$ (m/z 141.1) are uniformly observed in the electrospray background spectrum (Fig. 1, Table 1) and formaldehyde is a recognized degradation product of cyclic nitramines, including RDX and HMX [18]. However, no mass signals at m/z 171 were observed for injections of formaldehyde analytical standards. Further study using ^{15}N-labeled HMX, purified 4NO-HMX, and the quadrupole's MS-MS capability will better serve to define the identity of this degradation product.

4. Conclusion

A sulfobutylether-β-cyclodextrin assisted capillary electrophoresis system was applied in conjunction with a quadrupole ion-trap mass spectrometer for the identification of contaminant nitroaromatic and cyclic nitramine explosives and their degradation products. The work demonstrates the use of cyclodextrin dependent CE–MS for the analysis of explosives in environmental samples, and offers potential for the identification of highly polar or charged degradation products.

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