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DETERMINATION OF EXPLOSIVES IN WATER USING DISPOSABLE PIPETTE EXTRACTION AND HIGH PERFORMANCE LIQUID CHROMATOGRAPHY

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The use of disposable pipette extraction was examined for the simple and rapid determination of seven high explosives (cyclotrimethyl-enetrinitramine, cyclotetramethyl-enetetranitramine, 2,4,6-trinitrophenyl-methylnitramine, 2,4,6-trinitrotoluene, 2,4-dinitrotoluene, nitroglycerin, and pentaerythritol tetranitrate) in water. The current study involved the determination of slightly polar and nonpolar explosives in water with a reversed phase sorbent followed by high performance liquid chromatography. The method was based on a styrene divinylbenzene sorbent loosely placed inside a 5-mL pipette tip. Water samples were drawn into the tip and mixed with the sorbent. Air bubbles were also drawn through the tip following sample solution to enhance mixing. Because disposable pipette extraction uses small amounts of sorbent, minimal solvent is required to elute analytes and solvent evaporation is not necessary. The method provided rapid sample preparation, and required less than five minutes to extract 1.0 mL of water sample in the current study. Matrix-matched calibration was performed, and the limits of detection (LOD) were determined to be below 0.1 μg mL^{-1} for all targeted explosives in water with an enrichment factor of two. Coefficients of determination (r^2) were greater than 0.9990 for all studied explosives, and the recoveries ranged from 69.76% to 87.51%, 83.77% to 91.25%, and 83.62% to 98.99% for samples spiked at 0.25 μg mL^{-1}, 1.0 μg mL^{-1}, and 5.0 μg mL^{-1}, respectively. The relative standard deviations of recoveries at all spiked levels were below 8.97%. These results indicate that the disposable pipette extraction method provided good accuracy and precision for the determination of explosives in water.

Keywords: Disposable pipette extraction (DPX); Explosives; High performance liquid chromatography (HPLC); Solid phase extraction (SPE); Water

INTRODUCTION

The destruction resulting from the illegal use of high explosive materials has led to countless tragedies; often neglected, however, is the environmental harm caused by explosive residues. The hydrophilic nature and high mobility of most explosives

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indicate their potential for contamination of surface and ground water, and improper manufacturing, handling, storage, and disposal of explosives have been reported to contribute to contamination of water and surrounding areas (Crescenzi et al. 2007; Gaurav and Rai 2009; Jenkins et al. 1996; Talmage et al. 1999). As most explosives are toxic and carcinogenic (Yinon 1990; Robidoux et al. 2000; Robidoux et al. 2002), regulatory and public concern over explosives in water have increased due to potential health risks to humans and indigenous species. For example, 2,4,6-trinitrotoluene experiences photodegradation upon exposure, producing di- and trinitrobenzene derivatives, all of which are potentially dangerous to human. Hexahydro-1,3,5-trinitro-1,3,5-triazine and octahydro-1,3,5,7-tetranitro-1,3,5,7-tetrazocine are toxic to numerous terrestrial and aquatic systems, whereas methyl-2,4,6-trinitrophenylnitramine is mutagenic (Yinon 1990). Routine and comprehensive testing of explosives in water is important for regulatory agencies to ensure that concentrations of explosives are below toxic levels. It is crucial to develop simple and reliable methods for determination of explosives in water.

Sample preparation is usually the first step in the analysis. Achieving accurate and precise results for the determination of explosives in water depends largely on the extraction and cleanup methods that are employed. The number of relevant extraction methods for organic explosive residues present in aqueous mediums is virtually endless although continual advancements provide faster, simpler, or more reliable routes to achieve enrichment. Perhaps the two most widely employed methods are salting out and solid phase extraction (SPE) (Crescenzi et al. 2007; Leggett, Jenkins, and Miyares 1990; Jenkins et al. 1994; U.S. Environmental Protection Agency 1998; Feltes et al. 1990; Smith, Collins, and Wang 2003; Ahmad et al. 2008; Chow et al. 2004). SPE techniques have gained increasing interest because of their selectivity and because large volumes of organic solvents are not necessary. Almost all adsorbent types can be packed into the SPE column format, and the use of molecularly imprinted polymers expands the range of binding mechanisms (Fontanals, Marcé, and Borrull 2007). SPE cartridges have been widely employed for extraction and concentration of explosives from a broad range of sample matrices prior to chromatographic analysis. Few reports regarding application of solid phase microextraction (SPME) for extraction of explosives in water has been reported (Gaurav and Rai 2008; Monteil-Rivera, Beaulieu, and Hawari 2005). However, the SPME method can be somewhat limited due to the effect of environmental conditions on analyte adsorption on fiber surface, such as temperature, presence of salt, and pH variation of the natural water samples.

Although traditional SPE has been widely employed and shown positive results for extraction of explosives from water, the standard procedure involving conditioning, loading, washing, and elution can be lengthy and time-consuming. Disposable pipette extraction (DPX) is a novel solid phase extraction technique used for rapid extraction of analytes. A schematic diagram of DPX method is shown in Figure 1. Unlike traditional SPE that has sorbent tightly pack in cartridges; DPX has sorbent loosely placed in a pipette tip. DPX also provides flexibility for sample introduction. The sample solution may be either loaded from the top of the pipette tip as in conventional SPE or drawn in from the bottom of the pipette tip, although in most applications, sample solutions are drawn in from the bottom of the tip and mix with the sorbent in a dispersive manner. Rapid equilibration, partitioning, and enhanced

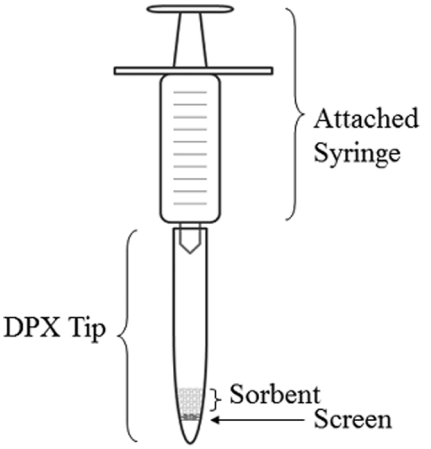


Figure 1. Schematic diagram of the DPX method. The DPX tip contains a loose sorbent confined inside the tip by a screen/filter. The syringe attachment allows for the dispensing or aspiration of solutions.

contact between analytes and solid phase sorbent are also enhanced by allowing air bubbles to be drawn through the tip following sample solution. DPX has been successfully employed in extraction of analytes in various matrices, such as drugs of abuse (Samanidou et al. 2013; Kovatsi et al. 2011; Ellison, Brewer, and Morgan 2009) and food contaminants (Guan, Brewer, and Morgan 2009; Guan, Brewer, Garris, Morgan, et al. 2010; Guan, Brewer, Garris, Craft et al. 2010) in biological samples. However, there has been no report for applicability of DPX on the determination of explosives in water.

The focus of the present research was to develop and validate a simple and reliable sample preparation method for the determination of high explosives in untreated water source using a DPX method followed by high-performance liquid chromatography (HPLC). The method may be applicable to finished drinking water, ground water, well water, and other types of surface water. In this study, a DPX method was used for extraction of explosives in river water, which is based on loosely contained styrene divinylbenzene sorbent placed inside a 5-mL pipette tip. The eluent was analyzed by HPLC with ultraviolet-visible detection. The DPX method was shown to be rapid, requiring just a few minutes to perform without any solvent evaporation. Seven explosives compounds were targeted in the study, representing three nitro-functional groups: nitroaromatics, nitramines, and nitrate esters (U.S. Environmental Protection Agency 2006). The method was optimized and fully validated. The accuracy, precision, limits of detection, limit of quantification, and linearity were determined. The applicability of the method to natural samples was evaluated with untreated river water from Beardstown, Illinois.

EXPERIMENTAL

Chemicals and Reagents

HPLC-grade acetonitrile, methanol, and water were purchased from Fisher Scientific (Pittsburgh, PA). Individual standard solutions for all explosive compounds

(1000 μg mL^{-1}), hexahydro-1,3,5-trinitro-1,3,5-triazine, octahydro-1,3,5,7-tetranitro-1,3,5,7-tetrazocine, methyl-2,4,6-trinitrophenylnitramine, 2,4,6-trinitrotoluene, 2,4-dinitrotoluene, trinitroglycerin, and pentaerythritol tetranitrate were purchased from Cerilliant (Round Rock, Texas). The properties of the targeted explosive compounds are shown in Table 1. 3,4-Dinitrotoluene (250 μg mL^{-1}) was purchased from Ultra Scientific (Kingstown, RI) and used as internal standard. All explosive standards and the internal standard were originally present in acetonitrile. Working solutions of standards and internal standard were prepared by dissolving original stock solutions in acetonitrile and diluting to 100 μg mL^{-1}. Disposable pipette extraction (DPX) tips (5 mL) containing styrene divinylbenzene sorbent (60 mg) were obtained from DPX Labs, LLC (Columbia, SC, USA). River water samples were collected from Illinois River located in Beardstown, Illinois.

Instrumentation

HPLC analysis was carried out on a Shimadzu Prominence LC-20AT LC, using a Shimadzu Prominence DGU-20A5 degasser, a Shimadzu Prominence SIL-20A auto-sampler, and a Shimadzu Prominence SPD-20A UV/VIS detector. The column employed was a Grace Brava ODS (150 mm × 4.6 mm with 5 μm particle size). The injection volume was 20 μL, and the detection wavelength was set at 220 nm. An isocratic mobile phase was used, consisting of a mixture of methanol and water (52/48, *v*/*v*), with a flow rate of 0.75 mL min^{-1}.

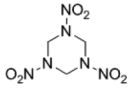
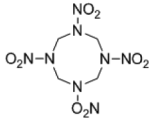
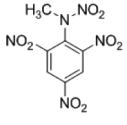
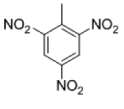
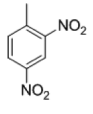
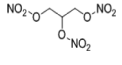
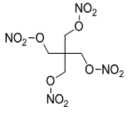
Sample Extraction

The DPX tips were first conditioned with 500 μL of acetonitrile. The water sample was then aspirated into the tip from the bottom followed by drawing in air bubbles to ensure the analytes were fully mixed with the sorbent for fast and efficient equilibration. After 30 to 60 seconds, the sample solution was dispensed to waste. One-half milliliter of HPLC-grade water was added to the top of the tip and dispensed to remove any interfering polar and water soluble matrix interferences. Lastly, the explosive analytes were eluted by adding 0.5 mL of acetonitrile and dispensing the organic solvent through the DPX tip into a HPLC autosampler vial. Twenty-five microliter of 3,4-dinitrotoluene (100 μg mL^{-1}) were added as internal standard to all final eluents before injection. A schematic diagram of DPX extraction of explosives in water is shown in Figure 2.

Recovery Study

Standard addition method was employed for recovery study of the DPX method using the river water from Beardstown, Illinois. Water samples before detonation was centrifuged for five minutes at 12,000 rpm to remove larger solid particles (i.e., dirt, organic plant matter, etc.) prior to use. Recovery studies were conducted using 1.0 mL river water samples spiked with a mixture of seven explosives at three different concentrations (0.25 μg mL^{-1}, 1.0 μg mL^{-1}, and 5.0 μg mL^{-1}). Water samples were then extracted as previously described. To reduce or eliminate matrix interferences, a matrix-matched sample was obtained by spiking the same

Table 1. Explosives and their properties

Name and (Abbrv.)	Cyclotrimethyl-enetrinitramine	Cyclotetramethyl-enetetranitramine	2,4,6-trinitrophenyl-methylnitramine	2,4,6-trinitrotoluene	2,4-dinitrotoluene	Nitroglycerin	Pentaerythritol tetranitrate
Emp. Formula[a]	$C_3H_6N_6O_6$	$C_4H_8N_8O_8$	$C_7H_5N_5O_8$	$C_7H_5N_3O_6$	$C_7H_6N_2O_4$	$C_3H_5N_3O_9$	$C_5H_8N_4O_{12}$
MW[b]	222	296	287	227	182	227	316
Structure							
Class[c]	N-AM	N-AM	N-AM/N-ARO	N-ARO	N-ARO	N-ES	N-ES
Solubility in water[d]	42	6.6	80	130	270	1500	2.1
Log K_{ow}[e,f]	0.87	0.06	1.65	1.86	1.98	1.62	3.71
Vapor Pressure[g]	4.1×10^{-9}	3.3×10^{-14}	5.7×10^{-9}	1.99×10^{-4}	1.4×10^{-4}	1.8×10^{-3}	5.4×10^{-9}

[a]Empirical Formula.
[b]MW: Molecular Weight, units of g mol^{-1}.
[c]Class: Nitramine: N-AM; Nitroaromatic: N-ARO; Nitrate Ester: N-ES.
[d]Clausen et al. 2006.
[e]Log K_{ow}, the Octanol/Water Partition Coefficient. 2010.
[f]U.S. Department of Health and Human Services, Public Health Service, Agency for Toxic Substances and Disease Registry (ATSDR), Division of Toxicology/Toxicology Information Branch. 1995, 1997, 1998.
[g]Vapor Pressure at 20°C (unless otherwise stated), in terms of mm Hg (Torr).

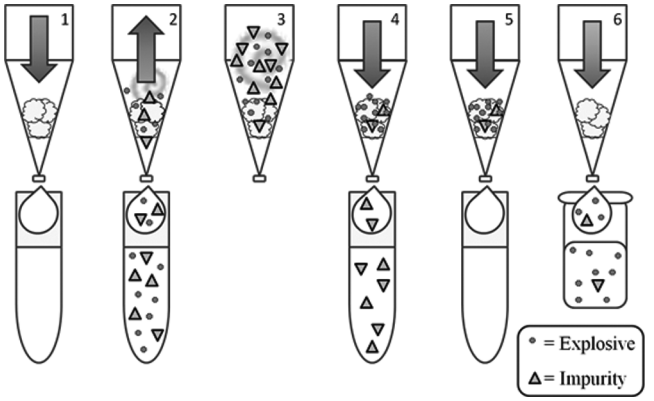


Figure 2. Schematic diagram of DPX extraction of explosives in water. Stage # 1: DPX tip is conditioned with acetonitrile; stage # 2: sample solution is aspirated into DPX tip from bottom; stage # 3: equilibrate for 30–60 s; stage # 4: dispense sample solution to waste; stage # 5: wash sorbent with 0.5 mL of water; stage # 6: elute explosives analytes using 0.5 mL acetonitrile.

concentration of explosives to a blank river water eluent following the same DPX procedures. Five replicates of spiked samples and duplicate of matrix matched samples were employed for all recovery studies. An internal standard was used to compensate for variations in the final volume, and 25 μL of 3,4-dinitrotoluene (100 μg mL^{-1}) was added to all final eluents before injection. Recoveries of explosives extracted from river water using the DPX method and HPLC analysis were calculated by the following equation:

$$\% \text{ recovery} = \frac{\begin{array}{c}\text{peak area of explosive in sample/}\\ \text{peak area of internal standard in sample}\end{array}}{\begin{array}{c}\text{peak area of explosive in matrix}-\text{matched sample/}\\ \text{internal standard in matrix}-\text{matched sample}\end{array}} \times 100\%$$

Method Validation

Matrix-matched calibration was performed to account for potential matrix effects, and river water samples were used to validate the method by displaying its ability to extract explosive residues from natural surface water. Coefficients of determination (r^2), limits of detection (LOD), and limits of quantitation (LOQ) were obtained. The river water samples were centrifuged at 12,000 rpm for five minutes to remove larger solid particles (i.e., dirt, organic plant matter, etc.). Matrix-matched calibration was performed with river water after centrifugation and spiked with analytes at five levels ranging from 0.25 to 5.0 μg mL^{-1}. Calibration data were generated from 5 replicate samples at 0.25 μg mL^{-1}, 2 replicate samples at 0.5 μg mL^{-1}, 2 replicate samples at 1.0 μg mL^{-1}, 2 replicate samples at 2.0 μg mL^{-1}, and 5 replicate samples at 5.0 μg mL^{-1}. The LOD and LOQ were determined by Eq. 1 and Eq. 2.

$$\text{LOD} = \frac{3.3\, s_{bl}}{m} \tag{1}$$

$$\mathrm{LOQ} = \frac{10\, s_{bl}}{m} \tag{2}$$

where m is the slope of the calibration plot and the standard deviation of the blank (s_{bl}) was estimated by calculating the standard deviation of the replicate results at the lowest fortification level (0.25 μg mL^{-1}).

Screening of Nitroglycerin Residue in Post-Blast River Water

River water samples from Beardstown, Illinois were collected before and after detonation of D-Gel 1000 (Desensitized Extra Gelatin Nitroglycerin Dynamite) by an independent institution. The dynamite, composed mainly of nitroglycerin (NG) and ammonium nitrate, was used to destroy heavily compacted, oversized beaver dams that had blocked water movement. All samples were stored at 4°C until analysis.

Water sample collected before detonation was used as a control and compared with post-blast water samples. Both controlled water samples and post-blast water samples were collected at the same locations 10 feet from the blast site. All water samples were subjected to the centrifugation process at 12,000 rpm for five minutes prior to the disposable pipette extraction. Extraction of nitroglycerin residue using the DPX method in river water was similar to the procedure described in sample extraction section except 10 mL of water sample was used to increase detection sensitivity. Isocratic HPLC conditions were the same as previously described with a total analysis time of 20 minutes.

RESULTS AND DISCUSSION

Determination of Explosives

EPA Method 8330 recommends an octadecyl-bonded silica stationary phase and a water/methanol mixture for the mobile phase for separation of explosives (U.S. Environmental Protection Agency 2006). In the current study, a Grace Brava ODS column (150 mm × 4.6 mm) was employed, and different conditions were investigated involving mobile phase composition, flow rate, and detection wavelength. To obtain baseline separation of the seven explosives and the internal standard, different mixtures of methanol and water were investigated, and the most desirable conditions were determined to be an isocratic mobile phase composed of 52% methanol in water (*v*/*v*). The flow rate was also optimized by varying from 0.4 to 1.0 mL min^{-1}. A flow rate of 0.75 mL min^{-1} provided fast and baseline separation. Detection wavelengths of 220 nm and 254 nm were compared. Due to a lack of UV absorption at 254 nm, 2,4,6-trinitrophenyl-methylnitramine, nitroglycerin, and pentaerythritol tetranitrate were not detected at 10 μg mL^{-1} using this wavelength, whereas 220 nm was found to provide better sensitivity for all target explosives at 10 μg mL^{-1}. As shown in Figure 3, good separation was achieved for all explosives and the internal standard with total analysis time of less than 16 minutes.

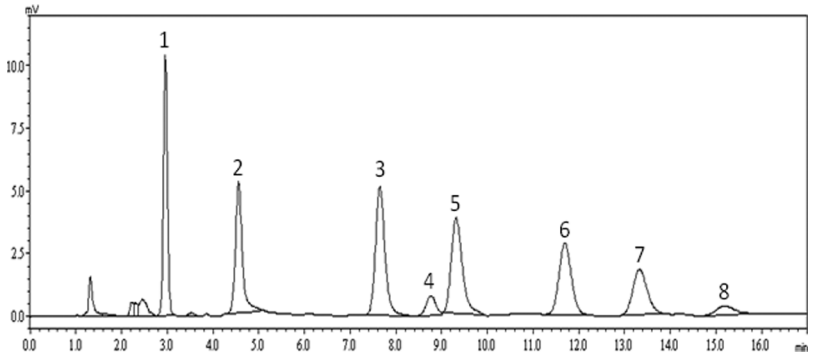


Figure 3. HPLC determination of explosives. Mobile phase: isocratic mobile phase composed of 52% methanol/water (*v*/*v*); column: Grace Brava ODS column (octadecylsilane, 150 mm × 4.6 mm); detection wavelength: 220 nm. Peak Identification: 1. cyclotetramethyl-enetetranitramine at 2.95 min; 2. cyclotrimethyl-enetrinitramine at 4.55 min; 3. 2,4,6-trinitrophenyl-methylnitramine at 7.65 min; 4. nitroglycerin at 8.76 min; 5. 2,4,6-trinitrotoluene at 9.31 min; 6. 3,4- dinitrotoluene (internal standard.) at 11.69 min; 7. 2,4-dinitrotoluene at 13.33 min; and 8. pentaerythritol tetranitrate at 15.19 min.

Adsorption of Explosives to DPX Sorbent

The logarithm of n-octanol/water partition coefficient (log K_{ow}) is commonly used to measure the hydrophobic/hydrophilic characteristics of compounds (Mitra 2003). Also, there is an apparent correlation between a compound's log K_{ow} value and its solubility in water. As shown in Table 1, the log K_{ow} values for the seven explosive compounds range from 0.06 to 3.71, indicating their wide range of polarities and different affinities toward aqueous phase and water solubility. Therefore, the development of a simple and reliable method for the determination of explosives in water is a challenging task. The mechanisms of retention depend on the hydrophobic nature of styrene divinyl benzene sorbent, which include Van der Waals forces and π-π bonding between the double bonds in explosives and the aromatic matrix of styrene divinyl benzene. The combination of the retention mechanisms provides selective enrichment of explosives and removal of polar matrix interferences in water.

Optimization of Elution Solvent for DPX Extraction

Methanol and acetonitrile were compared for elution of explosives by DPX with the styrene divinylbenzene sorbent. The explosives were spiked at 1.0 $\mu g\,mL^{-1}$ in river water before extraction. The relative recoveries achieved with the two solvents were evaluated by HPLC. Figure 4 shows recoveries obtained using methanol and acetonitrile. As can be seen, the recoveries ranged from 20% to 70% with most of recoveries below 60% when methanol was employed as elution solvent. However, when acetonitrile was investigated as elution solvent, the recoveries for all explosives were above 84%. A paired *t*-test using 95% confidence level was performed for comparison of the significance between methanol and acetonitrile, and the *p* value (0.0001) was below 0.05, indicating that acetonitrile provided significantly higher recoveries than methanol for the elution of explosives. The difference in the elution capabilities of the two solvents is speculated to be a result of their structural

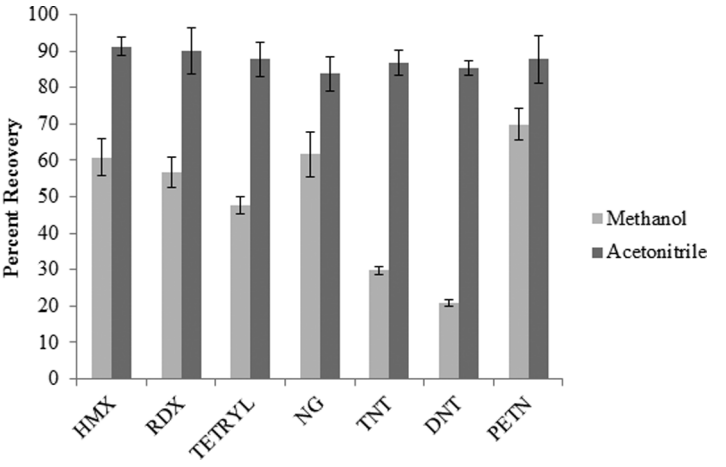


Figure 4. Comparison of the methanol and acetonitrile for elution of explosives from DPX with styrene divinylbenzene sorbent.

properties and the interactions offered to the analytes upon disrupting the adherence to the sorbent. Methanol and acetonitrile were both capable of participating in hydrophobic interactions with analytes during elution. However, only acetonitrile contains pi-bonds. The presence of pi-bonds presumably contributed to the ability of acetonitrile to elute the analytes from the sorbent due to the possible π-π interactions occurring in conjunction with the hydrophobic interactions between acetonitrile and styrene divinylbenzene sorbent. During the methanol elution study, poorly recovered analytes (2,4,6-trinitrophenyl-methylnitramine, 2,4,6-trinitrotoluene, and 2,4-dinitrotoluene) all contain aromatic rings, indicating strong π-π interactions between those compounds and styrene divinyl benzene sorbent. The poorly recovered nitroaromatic analytes also supported the assumption that methanol was incapable of disrupting the of the π-π interactions occurring between the analytes and sorbent.

Recoveries of Explosives at Different Fortification Levels

To evaluate the suitability of DPX for extraction of explosives at different levels, explosives were spiked at three different concentrations in river water: $0.25\,\mu g\,mL^{-1}$, $1.0\,\mu g\,mL^{-1}$, and $5.0\,\mu g\,mL^{-1}$. Table 2 shows recoveries of explosives

Table 2. Percent recoveries and RSD (in parentheses) based on 5 replicate experiments using DPX for the determination of explosives in water

Explosives	$0.25\,\mu g\,mL^{-1}$	$1.0\,\mu g\,mL^{-1}$	$5.0\,\mu g\,mL^{-1}$
Cyclotetramethyl-enetetranitramine	77.31 (3.31)	91.25 (2.90)	89.16 (2.17)
Cyclotrimethyl-enetrinitramine	80.80 (8.89)	89.95 (7.02)	87.92 (1.86)
2,4,6-trinitrophenyl-methylnitramine	69.76 (4.06)	87.71 (5.35)	87.51 (2.10)
Nitroglycerin	87.51 (3.45)	83.77 (5.56)	96.46 (2.35)
2,4,6-trinitrotoluene	73.69 (3.72)	86.72 (3.91)	90.53 (0.28)
2,4-dinitrotoluene	87.45 (4.72)	85.43 (2.38)	83.62 (0.64)
Pentaerythritol tetranitrate	71.84 (8.97)	87.72 (7.38)	98.99 (1.97)

Table 3. Percent recoveries and RSD (in parentheses) based on 5 replicate experiments performed by two individuals for reproducibility study

Explosives	Student 1 Recovery (RSD)	Student 2 Recovery (RSD)
Cyclotetramethyl-enetetranitramine	91.25 (2.90)	90.26 (1.86)
Cyclotrimethyl-enetrinitramine	89.95 (7.02)	88.68 (7.33)
2,4,6-trinitrophenyl-methylnitramine	87.71 (5.35)	86.27 (4.57)
Nitroglycerin	83.77 (5.56)	83.45 (6.37)
2,4,6-trinitrotoluene	86.72 (3.91)	85.37 (2.07)
2,4-dinitrotoluene	85.43 (2.38)	85.58 (2.70)
Pentaerythritol tetranitrate	87.72 (7.38)	87.20 (8.43)

in river water extracted using the DPX method developed. Of the studied explosives, recoveries ranged from 69.76% to 87.51%, 83.77% to 91.25%, and 83.62% to 98.99% for samples spiked at 0.25 $\mu g\,mL^{-1}$, 1.0 $\mu g\,mL^{-1}$, and 5.0 $\mu g\,mL^{-1}$, respectively. The relative standard deviations of recoveries at all spiked levels were below 8.97%. Good recoveries and standard deviations were obtained for the explosives at all levels in natural water samples, which indicate good accuracy and precision of the method developed in the study. In a separate study (Table 3), good reproducibility was obtained when a second person without any training of this particular project in our lab was able to repeat the recovery and relative standard deviation for samples spiked with 1.0 $\mu g\,mL^{-1}$ of explosives.

Calibration, LODs, and LOQs of Target Explosives

Table 4 summarizes the matrix-matched calibration results, along with LOD and LOQ values for the explosives. The calibration plots exhibit good linearity for explosives ranging from 0.25 to 5.0 $\mu g\,mL^{-1}$. Average coefficients of determination were greater than 0.9990. For all explosives studied using DPX – HPLC, the LODs and LOQs were less than 0.10 and 0.31 $\mu g\,mL^{-1}$. It should be noted that, the purpose of this study was to develop a DPX method for fast extraction of explosives in water

Table 4. Matrix-matched calibration: linearity, limit of detection (LOD), and limit of quantification (LOQ)

Explosives	r^{2a}	Calibration equation	LOD[b], $\mu g\,mL^{-1}$	LOQ[c], $\mu g\,mL^{-1}$
Cyclotetramethyl-enetetranitramine	0.9992	$y = 1.0407x + 0.0155$	0.03	0.09
Cyclotrimethyl-enetrinitramine	0.9989	$y = 0.7371x + 0.0895$	0.10	0.31
2,4,6-trinitrophenyl-methylnitramine	0.9993	$y = 1.1749x - 0.0205$	0.03	0.10
Nitroglycerin	0.9990	$y = 0.1488x + 0.003$	0.03	0.10
2,4,6-trinitrotoluene	0.9999	$y = 1.1377x - 0.025$	0.03	0.09
2,4-dinitrotoluene	0.9999	$y = 0.7491x + 0.0313$	0.05	0.15
Pentaerythritol tetranitrate	0.9992	$y = 0.1843x - 0.0139$	0.06	0.17

[a]Coefficient of determination.
[b]Limit of detection was calculated using the following equation: $LOD = \frac{3.3\,s_{bl}}{m}$.
[c]Limit of quantification was calculated using the following equation: $LOQ = \frac{10\,s_{bl}}{m}$.
s_{bl} = standard deviation of the response for the lowest concentration, and
m = the slope of the calibration plot.

that is compatible for different chromatography and mass spectrometry instruments. HPLC was employed to evaluate the DPX method developed in this study in terms of accuracy, precision, and linearity. There are several techniques to improve LOD and LOQ if sensitivity is a concern. Lower LOD and LOQ may be obtained by loading larger volumes of water sample for extraction or by evaporating the elution solvent. The LOD and LOQ can also be improved by doing large volume injection or by using chromatography coupled with tandem mass spectrometry. The focus of future research will be using liquid chromatography with tandem mass spectrometry for field and post-blast water samples for the desired sensitivity and confirmation.

Nitroglycerin Residue in Post-Blast River Water

River water from Beardstown, Illinois was tested for presence of nitroglycerin residue using the developed DPX method. Chromatograms of the water samples before and after detonation are shown in Figure 5 (a) and (b), respectively. As can be seen in Figure 5 (b), nitroglycerin had a higher concentration after detonation. The presence of nitroglycerin in the water sample before detonation, Figure 5 (a), may be attributed to the water or soil containing nitroglycerin residue from previous detonations. Since unknown samples identification is the main drawback of HPLC instrumentation using ultraviolet-visible detection, liquid chromatography with tandem mass spectrometry is desirable for intricate sample matrices in the future.

Advantages of DPX

Although the standard SPE procedure involving conditioning, loading, washing, and elution were still performed in the current DPX method, each step requires less time. The conditioning step only took a few seconds to add small amount of

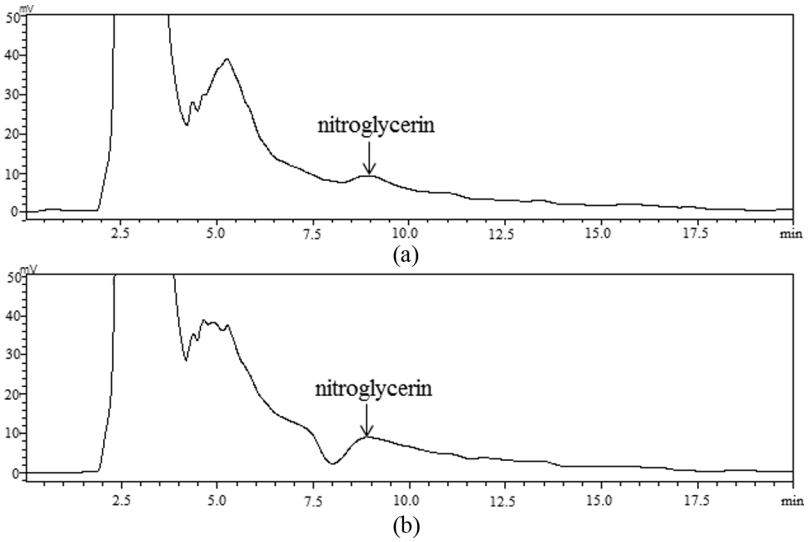


Figure 5. Screening of nitroglycerin residue in river water from Beardstown, Illinois using DPX extraction and HPLC: (a) river water before detonation; and (b) river water after detonation.

organic solvent (0.5 mL) to wet the sorbent. In traditional SPE, conditioning of the cartridge is a necessary step and usually requires multiple solvents to complete. DPX also provides the flexibility of sample loading from the top of the tip or aspirated from the bottom. Moreover, there is no vacuum system required for the DPX method since the sorbent is loosely contained in the tip and can be easily mixed with sample solution. Sample loading in DPX is usually done using a syringe as shown in Figure 1, and takes less than two minutes even for multiple loadings of large volume solution. The washing step is fast and usually takes a few seconds. The main advantages of DPX method include its speed and simplicity. The tips use small amount of sorbent and only require small amounts of solvent for elution; therefore, solvent evaporation is usually not necessary. The extraction of explosives (log $K_{ow} = 0.06$–3.71) in water has been demonstrated to be efficient due to the hydrophobic nature of the sorbent. In addition, good reproducibility was observed possibly because of the minimal matrix effects after extraction. Therefore, the DPX method developed in this study is suitable for the determination of explosives with wide range of polarities.

CONCLUSIONS

A simple and rapid sample preparation method for the extraction of explosives with a wide range of polarities in water has been demonstrated using DPX followed by HPLC. Good recoveries of over 70% were obtained with relative standard deviations of less than 10%, which indicates good accuracy and precision of the method. This method can be completely automated, providing a means for high-throughput determination of explosives. Future work will focus on the combination of DPX with liquid chromatography–tandem mass spectrometry both rapid and sensitive analysis of explosives.

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REFERENCES

Ahmad, U. K., S. Rajendran, L. W. Ling, and Y. C. Hooi. 2008. Forensic analysis of high explosives residues in post-blast water samples employing solid phase extraction for analyte pro-concentration. *Malaysian J. Anal. Sci.* 12(2): 367–374.

Chow, T. M., M. R. Wilcoxon, M. D. Piwoni, and N. R. Adrian. 2004. Trace level analysis of hexahydro-1,3,5-trinitro- 1,3,5- triazine (rdx) and its biodegradation intermediates in liquid media by solid-phase extraction and high-pressure liquid chromatography analysis. *J. Chromatogr. Sci.* 42(9): 470–473.

Clausen, J. L., N. Korte, M. Dodson, J. Robb, and S. Rieven. *Conceptual Model for the Transport of Energetic Residues from Surface Soil to Groundwater by Range Activities*. ERDC/CRREL TR-06–18. US Army Corps of Engineers: Engineer Research and Development Center/Cold Regions Research & Engineering Laboratory: Hanover, NH, 2006; pp 153–155.

Crescenzi, C., J. Albiñan, H. Carlsson, E. Holmgren, and R. Batlle. 2007. On-line strategies for determining trace levels of nitroaromatic explosives and related compounds in water. *J. Chromatogr. A.* 1153(1–2): 186–193.

Ellison, S. T., W. E. Brewer, and S. L. Morgan. 2009. Comprehensive analysis of drugs of abuse in urine using disposable pipette extraction. *J. Anal. Toxicol.* 33(7): 356–365.

Feltes, J., K. Levsen, D. Volmer, and M. Spiekermann. 1990. Gas chromatographic and mass spectrometric determination of nitroaromatics in water. *J. Chromatogr.* 518(1): 21–40.

Fontanals, N., R. M. Marcé, and F. Borrull. 2007. New materials in sorptive extraction techniques for polar compounds. *J. Chromatogr. A.* 1152(1–2): 14–31.

Guan, H., W. E. Brewer, S. T. Garris, C. Craft, and S. L. Morgan. 2010. Multiresidue analysis of pesticides in fruits and vegetables using disposable pipette extraction (DPX) and micro-luke method. *J. Agric. Food Chem.* 58: 5973–5981.

Guan, H., W. E. Brewer, S. T. Garris, and S. L. Morgan. 2010. Disposable pipette extraction for the analysis of pesticides in fruit and vegetables using gas chromatography/mass spectrometry. *J. Chromatogr. A.* 1217: 1867–1874.

Guan, H., W. E. Brewer, and S. L. Morgan. 2009. New approach to multiresidue pesticide determination in foods with high fat content using disposable pipette extraction (DPX) and gas chromatography-mass spectrometry (GC-MS). *J. Agric. Food Chem.* 57: 10531–10538.

Gaurav, A. K. M., and P. K. Rai. 2008. Solid phase microextraction-high performance liquid chromatographic determination of octahydro- 1,3,5,7-tetranitro-1,3,5,7-tetrazocine (HMX) and hexahydro-1,3,5-trinitro-1,3,5-triazine (RDX) in the presence of sodium dodecyl sulfate surfactant. *J. Sep. Sci.* 31: 2173–2181.

Gaurav, A. K. M., and P. K. Rai. 2009. Development of a new SPME-HPLC-UV method for the analysis of nitro explosives on reverse phase amide column and application to analysis of aqueous samples. *J. Hazard. Mater.* 172(2–3): 1652–1658.

Jenkins, T. F., P. H. Miyares, K. F. Myers, E. F. McCormick, and A. B. Strong. 1994. Comparison of solid-phase extraction with salting-out solvent extraction for preconcentration of nitroaromatic and nitra-mine explosives from water. *Anal. Chim. Acta.* 289: 69–78.

Jenkins, T. F., P. W. Schumacher, J. G. Mason, and P. T. Thorne. 1996. On-site analysis for high concentrations of explosives in soil: extraction kinetics and dilution procedures. CRREL Special Report 96–10. Hanover, NH: U. S. Army Cold Regions Research and Engineering Laboratory.

Kovatsi, L., K. Rentifis, D. Giannakis, S. Njau, and V. Samanidou. 2011. Disposable pipette extraction for gas chromatographic determination of codeine, morphine, and 6-monoacetylmorphine in vitreous humor. *J. Sep. Sci.* 34(14): 1716–1721.

Leggett, D. C., T. F. Jenkins, and P. H. Miyares. 1990. Salting-out solvent extraction for preconcentration of neutral polar organic solutes from water. *Anal. Chem.* 62(13): 1355–1356.

Mitra, S. 2003. *Sample preparation techniques in analytical chemistry*. New York, NY: John Wiley & Sons, Inc.

Monteil-Rivera, F., C. Beaulieu, and J. Hawari. 2005. Use of solid-phase microextraction/gas chromatography–electron capture detection for the determination of energetic chemicals in marine samples. *J. Chromatogr. A.* 1066: 177–187.

Robidoux, P. Y., J. Hawari, G. Bardai, L. Paquet, G. Ampleman, S. Thiboutot, and G. I. Sunahara. 2002. TNT, RDX, And HMX decrease earthworm (eisenia andrei) life-cycle responses in a spiked natural forest soil. *Arch. Environ. Contam. Toxicol.* 43(4): 379–388.

Robidoux, P. Y., C. Svendsen, J. Caumartin, J. Hawari, G. Ampleman, S. Thiboutot, J. M. Weeks, and G. I. Sunahara. 2000. Chronic toxicity of energetic compounds in soil determined using the earthworm (eisenia andrei) reproduction test. *Environ. Toxicol. Chem.* 19: 1764–1773.

Samanidou, V., C. Stathatos, S. Njau, and L. Kovatsi. 2013. Disposable pipette extraction for the simultaneous determination of biperiden and three antipsychotic drugs in human urine by GC-nitrogen phosphorus detection. *Bioanalysis.* 5(1): 21–29.

Smith, M., G. E. Collins, and J. Wang. 2003. Microscale solid-phase extraction system for explosives. *J Chromatogr A.* 991(2): 159–167.

Talmage, S. S., D. M. Opresko, C. J. Maxwell, C. J. Welsh, F. M. Cretella, P. H. Reno, and F. B. Daniel. 1999. Nitroaromatic munition compounds: environmental effects and screening values. *Rev. Environ. Contam. Toxicol.* 161: 1–156.

U.S. Department of Health and Human Services, Public Health Service, Agency for Toxic Substances and Disease Registry (ATSDR), Division of Toxicology/Toxicology Information Branch. 1995. T*oxicological Profile for Tetryl (2,4,6-Trinitrophenyl-N-methylnitramine),* 48–49 pp.

U.S. Environmental Protection Agency Method 3535A SW-846. 1998. *Solid-Phase Extraction (SPE)*. Washington, DC: Office of Solid Waste.

U.S. Environmental Protection Agency SW-846. 2006. Nitroaromatics and nitramines by high performance liquid chromatography. *Method 8330B*, Revision 2.

Yinon, J. 1990. *Toxicity and Metabolism of Explosive*. Boca Raton, FL: CRC Press.