

## Detection of chemical signatures of 2,4,6-Trinitrotoluene (TNT) in water samples using automated Solid-Phase Micro-extraction (SPME) coupled to FT-Raman spectroscopy

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**ABSTRACT:** 2,4,6-Trinitrotoluene, a high military explosive extensively used in bursting charge of shells, mines and bomb blasts by terrorists. It has become potential environmental pollutant near military installations and sites where bulk explosives are disposed through incineration and detonation. We investigated the water samples collected from near military training centre and for enhanced recovery from water samples; C8-SPME (Solid phase micro extraction) technique was used. The recovered extract was analyzed using Fourier Transform Raman spectroscopy. The spectrum of samples was compared to that of the standard spectrum. The study revealed that Raman spectroscopy provides fingerprint identification of TNT for water samples.

**Keywords:** 2,4,6-trinitrotoluene, hazardous substance, FT-Raman spectroscopy

### Introduction

2,4,6-Trinitrotoluene (TNT), a high military explosive extensively used in military and commercial activities including underwater blasting, mining, dyestuffs and as a chemical intermediate in the manufacture of photographic chemicals. It is released in environment and contaminates air, water and soil. It is a persistent contaminant which poses threat to ecosystem including humans. TNT is released in water through disposal of solid waste, detonation of explosives during military mock drill and commercial blasting activities. Waste water containing TNT in industrial production has caused many serious environmental problems worldwide. Numerous studies confirm that TNT exposure results in anemia, abnormal liver function, enlargement of spleen cataract, allergic reaction of skin etc. The occurrence of urinary tract tumors correlated with intensity of contact with nitro aromatic explosives. The harmless concentration of TNT in soil and water is less than 30mg/kg and 0.14 mg/l respectively.

Characterization of TNT in such water samples containing traces is challenge to forensic chemists. SPME utilizes a coated fiber housed inside a syringe needle as an adsorbent. The needle is inserted directly into the aqueous or organic extract of the sample and the fiber then exposed for collection. Once collected, the sample can be desorbed

either thermally or by solvent extraction. A number of articles on this technique have been reported, and SPME appears to have considerable potential for the analysis of explosives, combining speed and simplicity [1]. One study used cotton swabs to remove potential organic explosive residues, followed by aqueous extraction and collection on the SPME fiber [2]. They report that water extracted the explosive as effectively as acetone, and did not co-extract interfering materials. Thus Solid phase micro extraction has the potential to recover explosives even at very low concentrations. Numerous research studies on recovery of TNT using C8 sorbent based micro extraction cartridges have shown hundred percent recoveries. In recent years, explosive examination schemes or protocols are becoming much more formalized. This is dictated not only by technical considerations, but also the recognition that the procedures must meet the requirements of the legal system. The European forensic community discussed the question of which and how many techniques are required for confirmation of an explosive identification [1, 2, 3, 4].

Raman spectroscopy is a rapid, nonintrusive, and nondestructive technique (in most cases) that can be used for the analysis of many classes of hazardous and potentially explosive compounds. With developments in fiber optics filters, diode lasers, electromagnetic wave detectors and data analysis software, Raman spectroscopy has been able to move from the

laboratory environment to the field [5,6,7,8]. Raman spectroscopy provides the ability to analyze bulk materials such as milligram (or kilogram) quantities of drugs present in bags or vials, as well as liquids consisting of solvent mixtures present in a variety of containers [9]. Raman spectroscopy is sensitive to slight differences in chemical structure and has been used to detect contaminants present within a medicinal tablet or gel cap. Raman spectroscopy can also easily and rapidly distinguish between the nitramine high explosives RDX (cyclo-1, 3, 5-trimethylene-2, 4, 6-trinitramine) and HMX (cyclotetramethylene tetranitramine). Thus Raman spectroscopy is regarded as chemical fingerprinting techniques in organic compounds characterization. In Raman spectra of a compound peak number, Raman shift, coupling factor is parameter to determine molecule structure. In this study Raman spectroscopy is used to characterize explosives because of nondestructive method and its unique spectrum. Trinitrotoluene (TNT), its impurity 2, 4-dinitrotoluene (2, 4-DNT) and its degradation products, 1, 3-dinitrobenzene and 4-amino-2, 6-dinitrotoluene, have been detected by SERS in the vapor phase [10]. There has been lot of study related to other explosives such as RDX or TNT by other methods but hardly any study related to TNT by SPME coupled to FT-Raman, therefore in these study efficiency of solid phase micro-extraction followed by detection by FT-Raman spectroscopy has been focused for TNT.

### Material and Methods

2,4,6-Trinitrotoluene (1 $\mu$ g/ ml) standard and Solid TNT (99.9% purity, Crystalline) was purchased from Supelco inc. USA. Other organic solvents like Acetone, Ethyl alcohol, water were purchased from Merck, Poole, UK. C8 Solid phase Micro extraction Cartridges were purchased from Waters Ltd. USA. All glassware, spatula, forceps and sample bottles used for analysis were previously soaked in 10% nitric acid overnight before use. C8 SPME cartridge was pre washed with acetone 12 ml HPLC grade by creating vacuum with flow rate 5 ml/minute. It is allowed to air dried for 20-30 minutes. Cartridge rinsed with 3 ml HPLC acetone and collected drop by drop at flow rate 5 ml/minute. Water Samples were collected from the military base outside the shooting range. A series of 5ml samples were placed in glass vials. Samples were subjected to

cartridge and eluted using 3 ml deuterated chloroform. The eluant then collected and The Raman spectra of sample and control were taken using Bruker FT-Raman spectrometer. The Raman spectra were obtained in the 100-3800  $\text{cm}^{-1}$  range with 64 scans and a resolution of 2  $\text{cm}^{-1}$ . Some fraction of recovered explosive was crystallized and observed under SEM and results were compared with control

### Results and Discussion

Raman spectra of purified extracted explosive in Figure 1 show peaks at 2955, 1619.24, 1535.94, 1360.86, 1212.95, 825.23, 794.73  $\text{cm}^{-1}$ . Raman spectral peak observed at 2955  $\text{cm}^{-1}$  can be assigned to asymmetric C-H stretch vibration due to alkyl  $-\text{CH}_3$  group. The peak at 1619.24  $\text{cm}^{-1}$  represents C=C aromatic stretching vibration. Peak at 1535.94  $\text{cm}^{-1}$  may be due to C=C- $\text{NO}_2$  vibration. Peak at 1360.86 represent  $\text{NO}_2$  symmetric vibration. Peak at 1212.95  $\text{cm}^{-1}$  represents C6 H<sub>2</sub>-C vibration. Peak at 825.23 may be due to  $\text{NO}_2$  scissoring vibrations. Peak at 794.73  $\text{cm}^{-1}$  represent C-H out of plane vibrations. The spectral results were compared with standard (Figure 2) and peak matching confirmed presence of TNT [11,12].

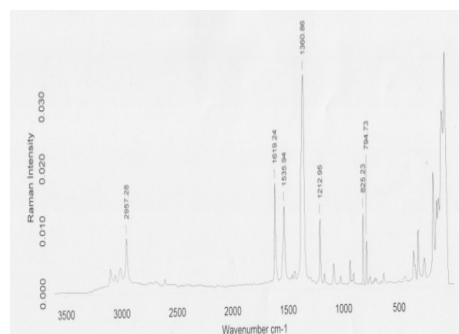


Figure 1: Water sample containing TNT

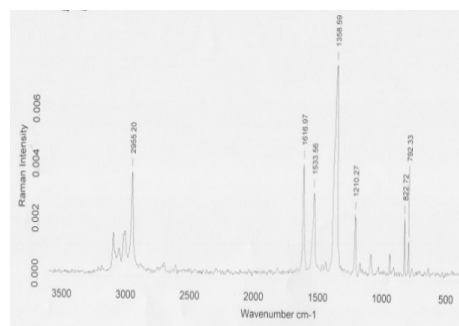


Figure 2: Standard TNT Sample

## Conclusion

In present study, Solid phase micro-extraction process has shown high recovery of TNT from aqueous samples. Addition of sodium chloride has improved extraction efficiency. Detection of TNT using FT- Raman spectroscopy provided fingerprint characterization even at very trace concentration. The present study could also be useful to forensic scientist whereby automated SPME coupled to FT-Raman spectroscopy could be used in forensic analysis of explosive residues recovered from explosion site.

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