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OPERATIONAL GUIDELINE: TEFLON DRY SURFACE WIPE COLLECTION AND IN-INJECTION PORT THERMAL DESORPTION ANALYSIS OF TRACE LEVELS OF ORGANIC EXPLOSIVES

DECEMBER 1999

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Opening Remarks: This document contains operational guidelines for an organic explosives sampling and analysis method developed in the Chemical and Analytical Sciences Division at Oak Ridge National Laboratory under funding provided by the National Institute of Justice. This document is written in a format resembling that of a standard operating procedure (SOP); however, it is intended only as an operational guideline for those wishing to implement the method. We have chosen to prepare the document as an operational guideline rather than a rigorous SOP in keeping with current practices of explosives analysis in the forensics community.

1. *Scope and Application*

1.1. This procedure describes a method of preparing dry Teflon surface wipes for sampling, from solid surfaces, trace levels of organic explosives and organics related to the production and decomposition of organic explosives.

1.2. This procedure also describes a gas chromatographic method utilizing thermal desorption (TD) of dry Teflon surface wipes for explosives analysis. Thermal desorption is performed within a split/splitless injection port, followed by gas chromatographic analysis using either electron capture (GC/ECD) and/or negative ion chemical ionization (GC/NICI) mass spectrometric detection.

1.3. The method has been applied to post explosion debris and is suitable for sampling explosive residues from other surfaces including, luggage, clothing, hands, etc.

1.4. This method has been validated for the organic explosives and organics related to the manufacture or decomposition of organic explosives listed in Table 1.

1.5. Refer to Reference Section 13.1 for sources of supply for explosive and related standards.

2. *Summary of Method*

2.1. Trace explosive residues are collected by wiping post-explosion debris with a surface-abraded Teflon tubing.

2.2. The explosive residues are thermally desorbed from the Teflon tubing within a split/splitless injection port of a GC and focused onto the head of a capillary column.

2.3. Quantification of desorbed explosives are carried out by gas chromatographic analysis with either electron capture detection or negative ion chemical ionization mass spectrometry.

3. *Accuracy, precision, linearity and limit of detection*

3.1. The accuracy of the method ranges from 85 to 113% for nine analytes and 87% for internal standard (see list in Table 1).

3.2. The precision (defined as percent relative standard deviation, %RSD) of the method ranges from 3 to 71% for nine analytes determined by GC/ECD with loading range of 0.1-51.9 ng. The precision ranges from 1 to 38% for nine explosives analyzed by GC/NICI with loading range of 0.7-138.2 ng.

3.3. The method limit of detection (LOD, defined in Section 12.2) ranges from 29 to 340 pg for nine analytes determined by GC/ECD, and 0.1-5.6 ng by GC/NICI.

3.4. The method limit of quantitation (LOQ, defined in Section 12.3) ranges from 88 to 1030 pg for nine explosives analyzed by GC/ECD, and 300 to 17000 pg analyzed by GC/NICI.

4. *Hazards*

4.1. Benzene is a carcinogen. Wear nitrile gloves when handling it.

4.2. Methanol and acetonitrile are flammable. Wear nitrile gloves when handling them and aviod ignition sources.

4.3. Properly dispose of spent solvents according to appropriate waste handling procedures.

4.4. Insulated (thermal) gloves should be worn when handling liquid nitrogen.

4.5. Desorptions are performed at high temperatures (ca. 170 °C). Avoid contact with the thermal desorption injector while a desorption is in progress.

4.6. The gases used in the analytical operation are contained in cylinders under high pressure. Be sure all cylinders are securely strapped. Safety glasses should always be worn when changing cylinders.

4.7. Guidelines for handling explosives in the specific site should be followed (see Section 13.5, or explosives handling guidelines at your specific site).

5. *Interferences*

5.1. Since a short column with a thin film thickness is used in this method, excessive quantities of sample will eventually diminish the column performance.

5.2. Non-target components (such as phthalates and halogenated compounds), that respond to ECD and/or NICI detection, may interfere with the analysis of explosives.

6. *Personnel Qualifications*

6.1. Personnel with adequate instrumentation analysis background can be trained to perform this method.

6.2. It is recommended that personnel demonstrate proficiency before attempting to perform this method without supervision.

6.3. It is mandatory to have explosive handling guidelines implemented before using this method.

7. ***Material and Instrumentation***

7.1. Procedure for Manufacturing and Conditioning Teflon Tubes.

7.1.1. Teflon tubing, 1/8" OD and 1/16" ID (Upchurch Scientific, Oak Harbor, WA), is used as the surface wipe/thermal desorption material.

7.1.2. The tubing is cut to 55 mm in length and held in a standard drill chuck attached to a laboratory stirrer motor.

7.1.3. The tubing is lightly sanded with a 240-grit emory cloth at a rate of several hundred revolutions per minute. The abraded tubing provides a better surface contact when wiping debris (Figure 1).

7.1.4. The abraded tubes are heated at 220 ^{0}C under a flow of argon at 360 ml/min for at least 5 hours (Figure 2).

7.1.5. The resulting tubes should have little or no detectable background signal on the ECD or NICI when thermally desorbed by the procedure described below.

7.1.6. When analyzing explosive residue, a freshly conditioned Teflon tube should be used for wiping debris surface. It is recommended that the Teflon tube should make a good contact with the debris surface (Figure 3) during wiping. A known quantity of internal standard should be spiked onto the Teflon tube after sampling the debris and before analysis.

7.2. Preparation of calibration standards.

7.2.1. The stock solutions for each of the ten compounds (as listed in Table 1) can be prepared in either methanol or acetonitrile at a nominal concentration of 1 mg/ml. The stock solutions can be stored in a flamable-storage approved refrigerator for at least two months.

7.2.2. The individual calibration standard or mixture of standards with known concentrations is prepared in benzene from the individual stock solutions at levels given in Table 3.

7.3. GC column sppecifications.

7.3.1. A HT-5 column (5% phenyl polycarborane siloxane, Scientific Glass Engineering, Inc., Austin, TX) or an equivalent column (such as DB-5 or Rtx-5) can be used in this method.

7.3.2. A short (12 m), narrow bore (0.22 mm ID) column, coated with thin film (0.1 µm) is recommended for the optimal analytical separation and recovery of explosives.

7.3.3. A helium (or nitrogen for ECD) carrier flow rate of greater than 2 ml/min is recommended for GC/ECD analysis. With vacuum compensation, a flow rate of greater than 1 ml/min is recommended for GC/NICI analysis. Slower flow rates or longer column length can lead to degradation of PETN on the column.

7.4. Instrumental conditions for GC/ECD analysis.

7.4.1. A Hewlett-Packard 5890 Series II Gas Chromatograph, equipped with an electron capture detector or an equivalent gas chromatograph can be used for this method.

7.4.2. An on/off valve should be attached to the split-vent on the front of the gas chromatograph (Figure 4a), and a single gooseneck splitless liner (4.0 mm ID, 6.5 mm OD x 78.5 mm) should be installed in the injection port (Figure 4b).

7.4.3. The auxiliary flow should be set at an optimal flow rate (ca 100 ml/min) to minimize ECD background signal and maximize ECD sensitivity.

7.4.4. The ECD detector temperature should be set between 250 to 300 °C for optimal detection.

7.5. Instrumentatal conditions for GC/NICI analysis.

7.5.1. Hewlett-Packard 5989B Gas Chromatograph/Mass Spectrometer with dual ion source or an equivalent instrument can be used for this method.

7.5.2. The analysis should be performed in electron capture negative ion chemical ionization mode.

7.5.3. With methane as reagent gas, the source pressure (0.8 to 1.2 Torr) should be optimized for maximum sensitivity and mass accuracy (see Quality Control section for details).

7.5.4. Source temperature should be set at 150 ^{0}C, source temperature at 100 ^{0}C.

7.5.5. With the electron energy set at 230 eV, and emission current at 300µA, full scan (50-550 amu) spectral data should be acquired at a rate as predetermined by the manufacture's software.

7.6. Calibration and internal standard spiking.

7.6.1. Over a pre-determined concentration range, a five-point calibration curve with replicate measurements at each point should be constructed for each target analyte.

7.6.2. In a typical calibration (or sample) analysis, a 2 µl sample of calibration solution (or internal standard solution) should be deposited on a Teflon tube by a micro-syringe (Figure 5) and the solvent should be allowed to dry (approximately 5 minutes).

7.7. Procedure for thermal desorption and gas chromatographic analysis.

7.7.1. The injection port temperature should be reduced to less than 50 ^{0}C to allow the Teflon tube to be inserted into the injection port. This step can be facilitated by blowing with a stream of house air or cold nitrogen drawn from a liquid nitrogen dewar (see Hazard section for proper handling of liquid nitrogen).

7.7.2. When analyzing explosive residue, a freshly conditioned Teflon tube should be used for wiping debris surface. It is recommended that the Teflon tube should make a good contact with the debris surface (Figure 3) during wiping. A known quantity of internal standard should be spiked onto the Teflon tube after sampling the debris and before analysis.

7.7.3. Prior to inserting the Teflon tube inside the injection port, the split purge valve should be turned off and the split vent should be blocked by closing the on/off valve.

7.7.4. After inserting the Teflon tube inside the injection port (Figure 6), the port should be closed securely and heated to 170 °C. The carrier flow during desorption process should be increased to at least 6.5 ml/min for GC/ECD, and 10 ml/min (with vacuum compensation) for GC/NICI analysis. Failure of the carrier flow to reach the pre-determined rate may indicate a leak in the system.

7.7.5. Thermal desorption should be allowed to proceed for 7 minutes, including 3.5 - 4 minutes injector heating time.

7.7.6. The GC oven should be held at ambient temperature during theraml desorption to allow the desorbed target analytes to be condensed and focused at the head of the column.

7.7.7. A two-tiered oven program is recommended for optimal separation of the analytes. The initial oven temperature should be held for 3 min at 70 °C, and then heated to 185 °C at a rate of 8 °C/min in the first tier of the program. The oven temperature should then be heated to 280 °C at a rate of 30 °C/min and held at 280 °C for 10 minutes.

7.8. Data reduction.

7.8.1. Calibration curves can be constructed by plotting the concentration ratios of target analytes relative to the internal standard versus their corresponding area ratios.

7.8.2. The integrated area for each analyte should be obtained either from the integrated area of a peak as anlyzed by TD/GC/ECD or from an integrated area of the most abundant ion as analyzed by TD/GC/NICI.

7.8.3. The concentration and area ratios can be used to perform linear regression with, or without, intercept to obtain the Response Factor (slope) and correlation coefficient (r) for each target analyte. The Response Factor is defined as the slope of a plot of Area Ratio (ordinate) versus the Concentration Ratio (abscissa). A standard t-test should be used to determine whether the intercept from each calibration curve is significantly different from zero at the 95% confidence level.

7.8.4. The quantity of explosives collected on the Teflon tube can be calculated using one of the following equations:

Quantity (ng) = (Area ratio /Response factor) * (ng of IS spiked)
Quantity (ng) = [(Area ratio – Intercept)/Response factor]*(ng of IS spiked)

8. *Quality Control and Quality Assurance*

8.1. Reproducibility of Teflon tube preparation.

8.1.1. If more than one batch of abraded tubes are prepared, reproducibility of tube preparation should be ensured so that the method accuracy and precision are independent of the surface-wipe preparation.

8.1.2. One tube randomly selected from each batch of prepared tubes should be spiked with known quantities of explosive standards and analyzed for their recovery either by TD/GC/ECD or by TD/GC/NICI.

8.1.3. The spiking and analysis should be repeated in triplicate for each tube, and the average analyte recovery based on an established calibration should be determined. The resulting average recovery should be used to calculate the percent relative standard deviation (%RSD) for each tube.

8.1.4. In general, the %RSD should be less than 10% for every analyte as listed in Section 2.5 except for PETN. The thermal instability of PETN will result in a higher %RSD (ca. 15%) for this analyte.

8.2. Column Performance.

8.2.1. By using a short capillary column coated with thin film thickness of moderately polar liquid phase (as recommended in Section 7.3.1), baseline separation should be achieved for all the explosives and related compounds including internal standards.

8.2.2. The order of gas chromatographic elution should be as the following (Figure 7): beyond 7 min, DEGN; 2,6-DNT; 2,5-DNT; 2,4-DNT; 2,3-DNT; 3,4-DNT; TNT; PETN; RDX; and DNN (IS).

8.2.3. If the gas chromatograph is equipped with an electronic pressure control for maintaining a constant column flow throughout the entire GC program, then a baseline resolution should be achieved with the pair of 2,3- and 2,4-dinitrotoluene isomers. Otherwise, partial resolution of this pair of isomers is acceptable.

8.2.4. As part of QA/QC procedure, GC column performance (peak resolution, elution order, etc.) check should be carried out prior to calibration and/or sample analysis.

9. *Method Performance*

9.1. If the analysis is to be performed on a mass spectrometer, the following procedure is recommended for checking the NICI tune:

9.1.1. 300 pg of decachlorobiphenyl and 100 pg of hexachlorobenzene should be injected onto the column and analyzed with the following temperature program: 100 °C hold for 3 min, ramp to 280 °C at 20 °C/min.

9.1.2. The resulting spectra should be compared with reference spectra (Reference 13.6): for decachlorobiphenyl *m/z* 498 (100%), 464 (>27%), and 430 (>5%); for hexachlorobenzene *m/z* 284 (100%), 286 (>70%), 250 (>10%).

9.1.3. If the relative abundance is out of range, the instrument must be re-tuned. The GC peak height for both compounds acquired over the full scan (100-550 amu) must be at least three times greater than the noise.

9.1.4. The eight most abundant ions (m/z) and their relative intensities (% abundance) for the nine target analytes are listed in Table 2.

10. *Thermal Desorption Efficiency of the Methodology*

10.1. The thermal desorption (TD) efficiency (method accuracy) should be determined by comparing the ratios of instrumental response due to spiked analyte thermally desorbed from Teflon tube relative to the response from a liquid injection of the same quantity of analyte.

10.2. The analysis can be carried out either by TD/GC/ECD, or by TD/GC/NICI. It is recommended to obtain average recovery rate value from at least three successive sets of thermal desorption and liquid injections.

10.3. The resulting desorption efficiencies (%) for the analytes should be comparable to those listed in Table 3 (ranged from 85% for 2,5-DNT to 113% for RDX) with comparable loading quantities for each analyte.

11. Precision and Linearity of the Methodology

11.1. Teflon tubes should be spiked in triplicate with calibration standard mixtures that include nine target analytes and an internal standard at minimal of five concentrations ranging 0.5 to 80 ng/tube.

11.2. The spiked Teflon tubes should be analyzed either by TD/GC/ECD or by TD/GC/NICI. The spiking and analysis should take place in a random order over a period of a week.

11.3. The relative response and relative concentration for each analyte at each concentration should be calculated:

Relative response = Area of analyte/Area of IS
Relative concentration = Concentration of analyte/Concentration of IS

11.4. The relative responses obtained from triplicate analysis for each analyte at each concentration should be used to calculate the average response, which in turns should be used to calculated the percent relative standard deviation (%RSD).

11.5. The ranges for %RSD should be comparable to those listed in Table 3 for the specified loading range.

11.6. A linear regression curve should be fitted between the arrays of relative responses and relative concentrations to construct a five-point calibration.

11.7. The linearity (r^2) resulting from linear regression should be comparable to those listed in Table 4 for the specified loading range.

12 *Limit of Detection and Limit of Quantitation*

12.1 The data used for the precision and linearity should also be employed to determined the method limit of detection (LOD) and limit of quantitation (LOQ) for each analyte using both methods of detection.

12.2 The LOD should be determined as:

LOD (ng) = 3.3 (SD/S) * (ng of IS spiked)

Where SD is the standard deviation of the relative response near the detection limit and S is the slope of the calibration curve

12.3 The LOQ should be determined as :

LOQ = 10 (SD/S) * (ng of IS spiked)

12.4 The resulting LOD and LOQ for each analyte determined for TD/GC/ECD and TD/GC/NICI should be comparable to those listed in Table 4.

13 *References*

13.1 Michael E. Sigman, and Cheng Yu Ma, "In-Injection Port Thermal Desorption for Explosives Trace Evidence Analysis" Analytical Chemistry, *71*, pp 4119-4124, 1999.

13.2 Hewlett Packard Gas Chromatograph and Mass Spectrometer 5989B system Operating Manuals, including HP 5989B Hardware Manual (Copyright 1993, Part

05989-90071), MS Chemstation User's Guide (Copyright 1993, HP G 1034C, Part # HP G 1034-90043).

13.3 Hewlett-Packard 5890 Series II Gas Chromatograph Operating Manual (Copyright 1989, Part # 05890-90260).

13.4 Microsoft Excel 97 Spreadsheet Program (Microsoft, 1997, Part # 98722).

13.5 Lockheed Martin Energy Research Corporation ORNL Explosives Program, number ORNL-SH-P05, Rev. 1, dated 10/5/98.

13.6 E. A. Stemmler; R. A. Hites, *Electron Capture Negative Ion Mass Spectra of Environmental Contaminants and Related Compounds*, PP 39, and 253, VCH Publisher, New York, 1988.

Table 1. List of analytes (explosives and explosives related compounds), abbreviations and CAS numbers.

Analyte	Abbreviation	CAS Number
Diethyleneglycol dinitrate	(DEGN)	[693-21-0]
2,6-Dinitrotoluene	(2,6-DNT)	[606-20-2]
2,5-Dinitrotoluene	(2,5-DNT)	[619-15-8]
2,4-Dinitrotoluene	(2,4-DNT)	[121-14-2]
2,3-Dinitrotoluene	(2,3-DNT)	[602-01-7]
3,4-Dinitrotoluene	(3,4-DNT)	[610-39-9]
2,4,6-Trinitrotoluene	(TNT)	[118-96-7]
Pentaerythritol tetranitrate	(PETN)	[78-11-5]
Hexahydro-1,3,5-trinitro-1,3,5-triazine	(RDX)	[121-82-4]
1,3-Dinitronaphthalene (*internal standard, IS*)	(DNN)	[606-37-1]

Table 2 The eight most abundant NICI ions (m/z) and their relative intensities (% abundance) for the nine target analytes.

Analyte	m/z	% Abundance	Analyte	m/z	% Abundance
DEGN	62	100	**2,6-DNT**	182	100
	46	91		183	9
	242	71		152	8
	258	20		184	1
	90	10		166	1
	93	10		153	1
	103	9		150	1
	109	5		164	1
2,5-DNT	182	100	**2,4-DNT**	182	100
	183	9		152	12
	152	3		183	8
	166	2		166	2
	184	1		165	1
	153	1		150	1
				184	1
				164	1
2,3-DNT	182	100	**3,4-DNT**	182	100
	152	9		183	9
	166	1		152	6
	184	1		184	1
	153	1		166	1
				153	1
				134	1
TNT	227	100	**PETN**	62	100
	197	22		46	19
	210	16		101	16
	228	10		99	9
	198	3		84	8
	167	3		240	6
	195	2		256	5
	229	2		378	5
RDX	102	100			
	129	80			
	176	30			
	268	25			
	46	17			
	324	15			
	130	4			
	103	4			

Table 3 Thermal desorption efficiency and reproducibility determined by TD/GC/ECD and TD/GC/NICI.

		GC/ECD		GC/NICI	
Analyte	Accuracy (Desorption Efficiency)	Reproducibility (%RSD)	Loading Range (ng)	Reproducibility (%RSD)	Loading Range (ng)
DEGN	95	8-45	0.15-24.51	13-34	2.60-37.76
2,6-DNT	93	5-26	0.11-18.17	15-27	0.68-10.04
2,5-DNT	85	5-30	0.14-22.88	9-30	0.86-12.64
2,4-DNT	97	5-19	0.29-46.72	7-31	1.18-17.22
2,3-DNT	96	3-29	0.11-17.30	7-29	0.66-9.56
3,4-DNT	96	4-23	0.14-22.49	5-19	0.86-12.44
TNT	89	4-24	0.20-31.72	2-32	1.20-17.54
PETN	95	6-71	0.33-51.91	4-38	8.25-119.96
RDX	113	5-33	0.31-49.98	1-11	4.74-138.18

Table 4 Limit of detection and limit of quantitation determined by TD/GC/ECD and TD/GC/NICI.

	GC/ECD			GC/NICI		
Analyte	Calibration Linearity (r^2)	Method LOD (pg)	Method LOQ (pg)	Calibration Linearity (r^2)	Method LOD (ng)	Method LOQ (ng)
DEGN	0.991	229	694	0.995	1.8	5.4
2,6-DNT	0.998	29	88	0.988	0.5	1.5
2,5-DNT	0.967	78	236	0.990	0.5	1.5
2,4-DNT	0.997	174	527	0.993	0.7	2.1
2,3-DNT	0.979	39	118	0.994	0.5	1.5
3,4-DNT	0.999	59	179	0.995	0.1	0.3
TNT	0.998	340	1030	0.995	0.4	1.2
PETN	0.988	133	403	0.997	5.6	17.0
RDX	0.997	332	1006	0.983	1.8	5.6

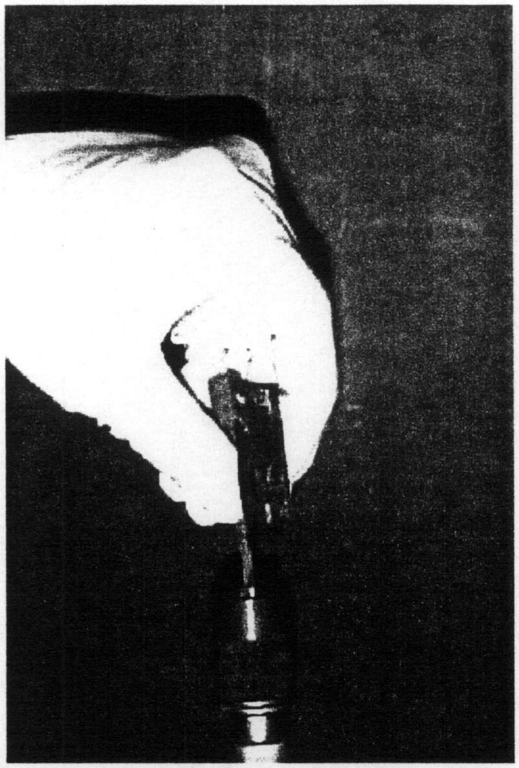

Figure 1: Abrasion of Teflon tube with 240-grit emory cloth.

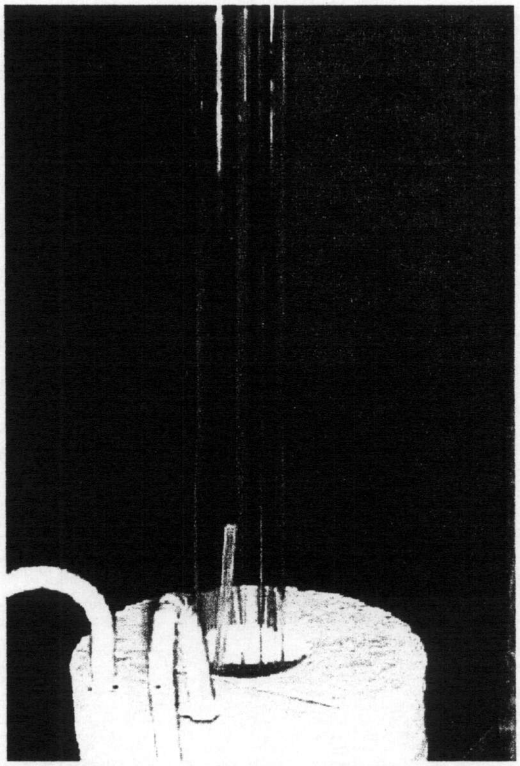

Figure 2: Heating of abraded Teflon tube in a tube furnace.

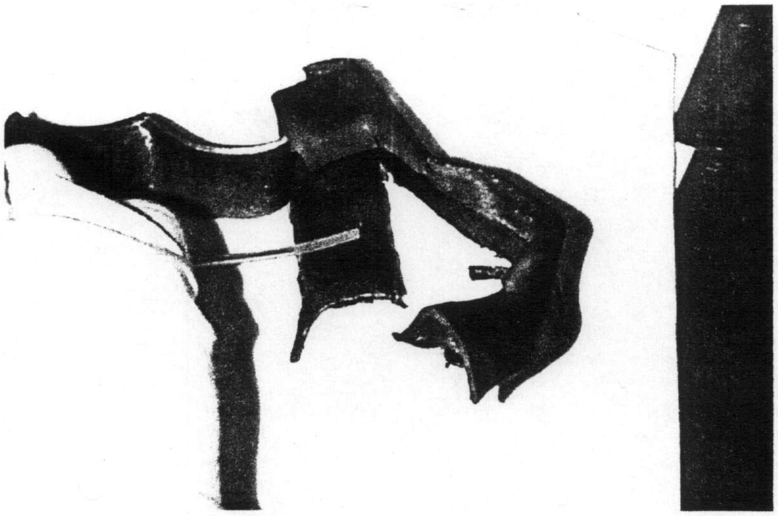

Figure 3: Example of debris sampling by surface wipe.

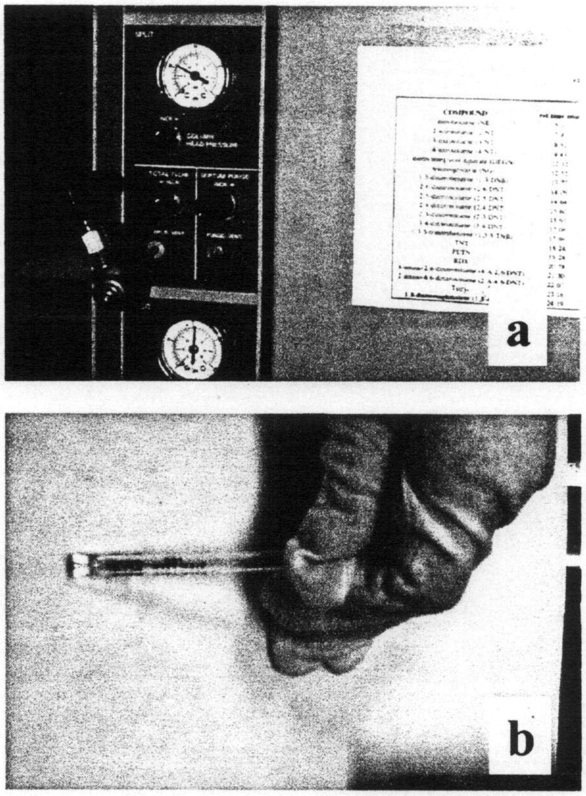

Figure 4: (a.) On/off valve attached to split-vent on the front of the gas chromatograph. (b.) Single gooseneck splitless liner.

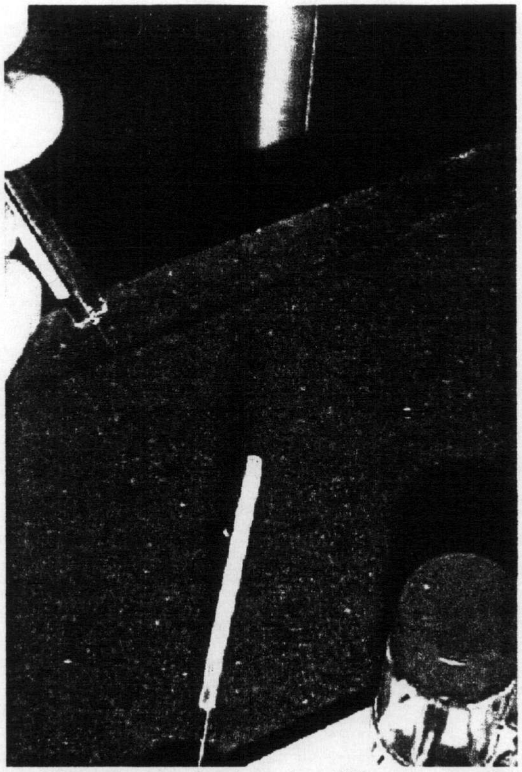

Figure 5: Deposition of calibration solution (or IS) on a Teflon tube by a microsyringe.

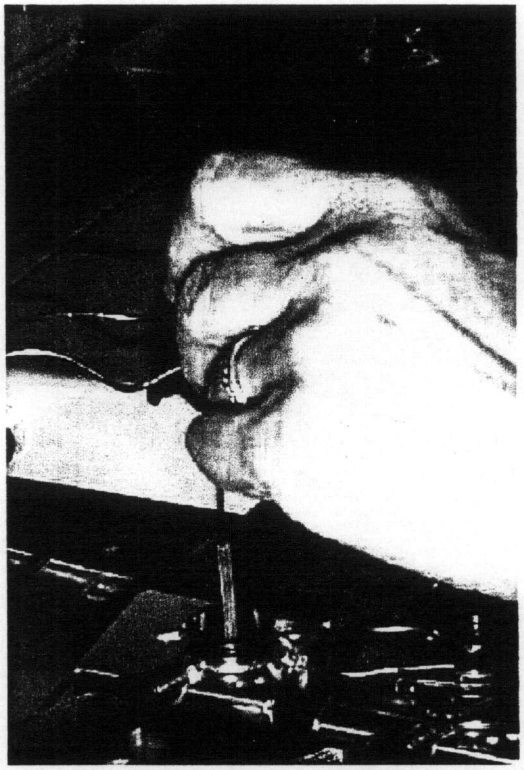

Figure 6: Inserting Teflon tube inside GC injection port.

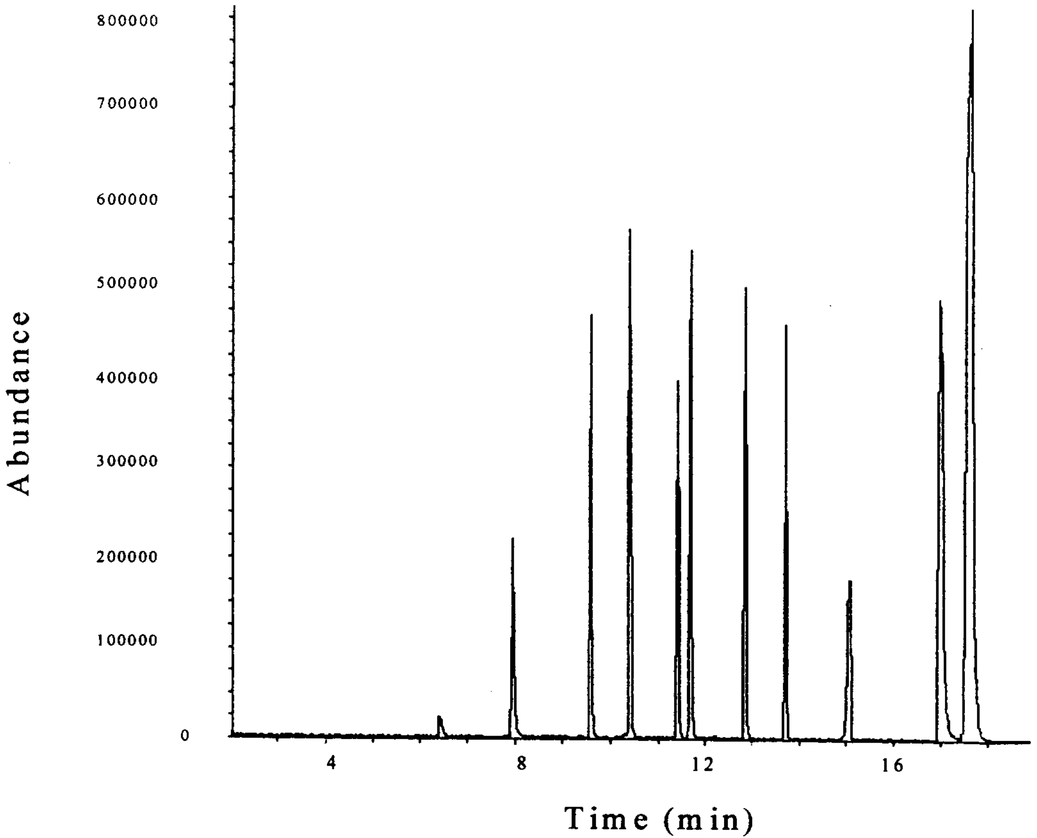

Figure 7: In-injection port thermal desorption GC/NICI chromatogram of a standard mix of explosives. The peak identities (starting at 8 min) are listed in Table 1.

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