Advanced Mass Spectrometric Techniques for DOD analytes of interest

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SEVERN TRENT STL

Non-standard MS techniques

- HPLC/MS/MS
- IC/MS/MS
- GC/MS/MS
- Electrospray ionisation
- Atmospheric Pressure Chemical Ionization
- Chemical Ionization

Electron Impact GC/MS

- Advantages
  - Powerful separation
  - Structural information from fragmentation
  - Affordable instrumentation
  - Universal detector (if the analyte gets to the MS)
- Disadvantages
  - Most organic compounds will not go through a gas chromatograph
  - Some compounds fragment too much
  - No selectivity

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Desirable method characteristics

- Linearity - predictable instrument response
- Sensitivity - low concentration reliably detected
- Precision - reproducibility of results
- Accuracy - proximity of results to true value
- Selectivity - ability to differentiate compound of interest from interferences
- Ruggedness - ability of method to work properly in a variety of types of samples

Energetic compounds

- Standard method is 8330 HPLC/UV
  - Insufficient
    - Sensitivity
    - Selectivity
    - Ruggedness
- Solution
  - LC/MS
  - Extraction – similar to 8330 – 2g sonicated in acetonitrile for soil, SPE of 1L water eluted with acetonitrile to 5 mL final volume.

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- Analysis
  - LC- 250 mm C18 column, mobile phase 0.01M ammonium acetate in water and methanol mixture
  - MS- APCI negative ion polarity – single stage MS detection of characteristic mass
  - 3 isotopic labeled internal standards and one surrogate used for QC compounds
  - Calibration – 10 to 300 ug/L instrument concentration

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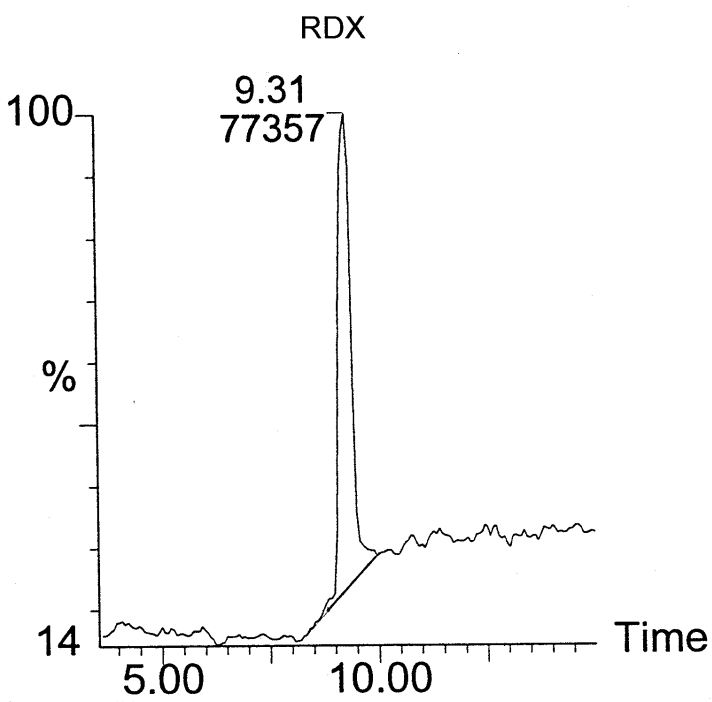
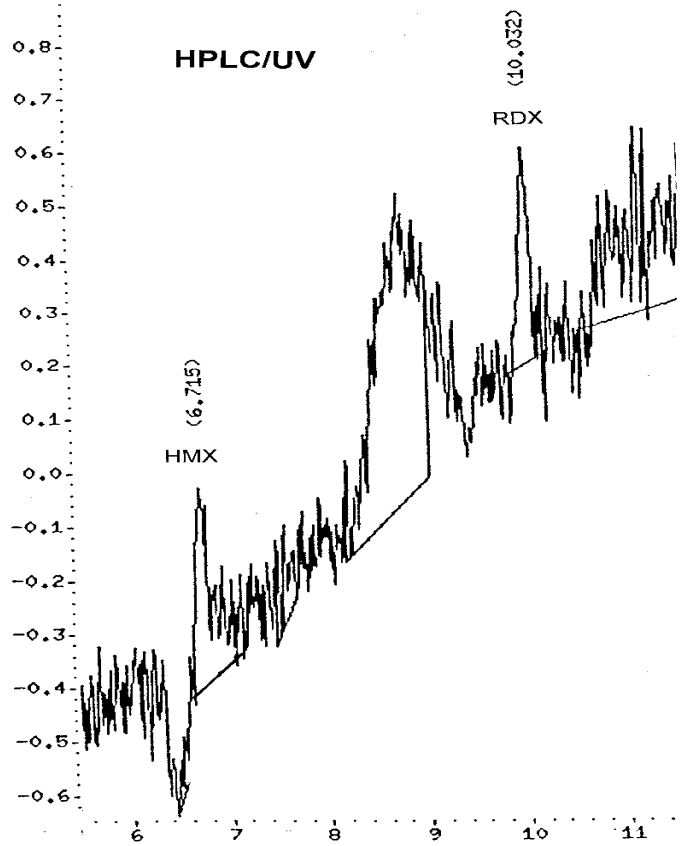
Detection limits, LC/MS vs. LC/UV

Analyte	LC/UV MDL	LC/MS MDL	Factor
1,3,5-Trinitrobenzene	0.037	0.015	2
1,3-Dinitrobenzene	0.065	0.008	8
2,4,6-Trinitrotoluene	0.047	0.015	3
2,4-Dinitrotoluene	0.068	0.013	5
2,6-Dinitrotoluene	0.075	0.013	6
2-Amino-4,6-dinitrotoluene	0.058	0.012	5
2-Nitrotoluene	0.065	0.022	3
3-Nitrotoluene	0.034	0.016	2
4-Amino-2,6-dinitrotoluene	0.028	0.015	2
4-Nitrotoluene	0.042	0.014	3
HMX	0.068	0.015	4
Nitrobenzene	0.096	0.020	5
Nitroglycerin	0.374	0.039	10
PETN	0.529	0.016	33
RDX	0.098	0.006	18
Tetryl	0.084	0.010	9

10 ug/L RDX

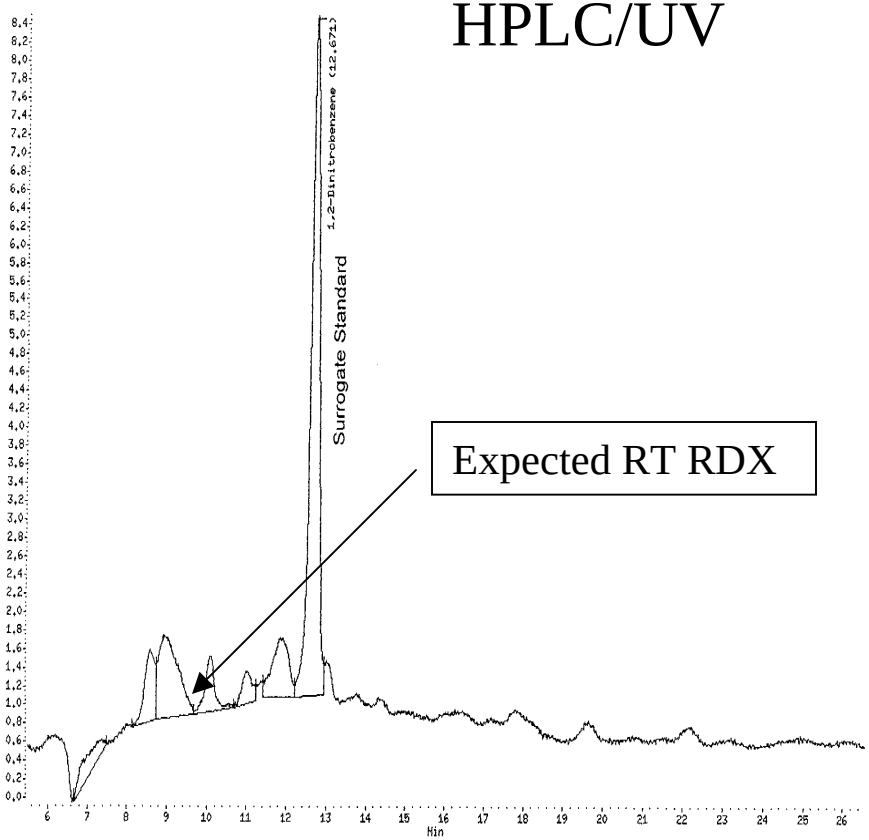
LC/MS





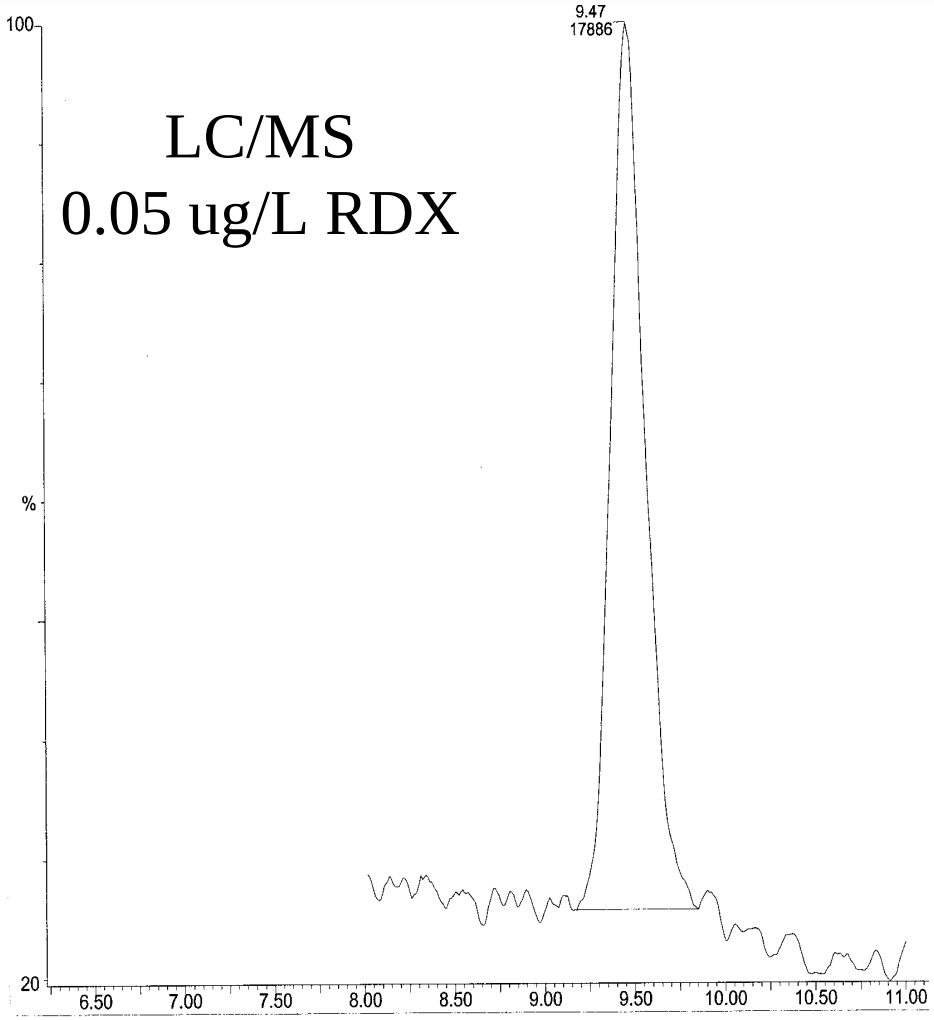
Groundwater containing 10 mg/L JP4

HPLC/UV



Expected RT RDX

LC/MS
0.05 ug/L RDX



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Explosives low std Compounds 1-8

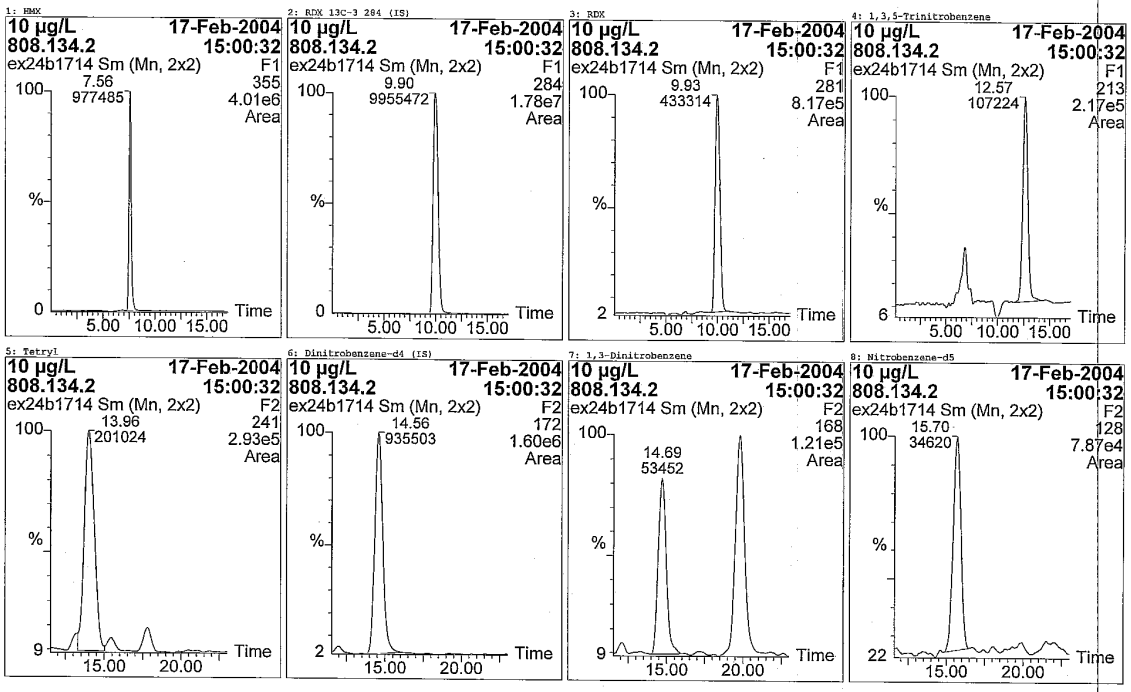
Quantify Sample Report
Explosives Analysis

Page 4

Sample List: C:\Masslynx\Explosives.PRO\SampleDB\ex24b17(2)
Last modified: Wed Feb 18 08:09:15 2004
Method: C:\Masslynx\Explosives.PRO\MethDB\ex24b17
Last modified: Wed Feb 18 07:54:11 2004
Job Code:

Printed: Wed Feb 18 08:10:42 2004

Name: ex24b1714
Text: 10 µg/L



Analyst: Steve Cowling

Explosives low std Compounds 9-16

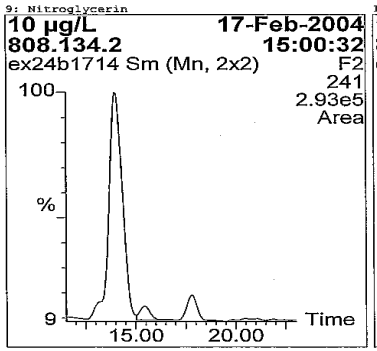
Quantify Sample Report
Explosives Analysis

Sample List: C:\Masslynx\Explosives.PRO\SampleDB\ex24b17(2)
Last modified: Wed Feb 18 09:09:15 2004
Method: C:\Masslynx\Explosives.PRO\MethDB\ex24b17
Last modified: Wed Feb 18 07:54:11 2004
Job Code:

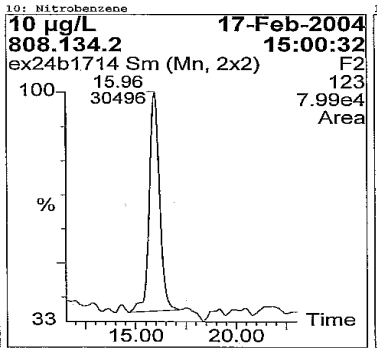
Printed: Wed Feb 18 08:10:42 2004

Name: ex24b1714
Text: 10 µg/L

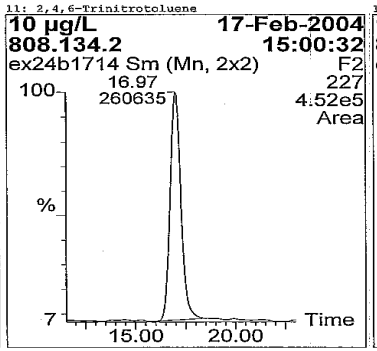
9: Nitroglycerin



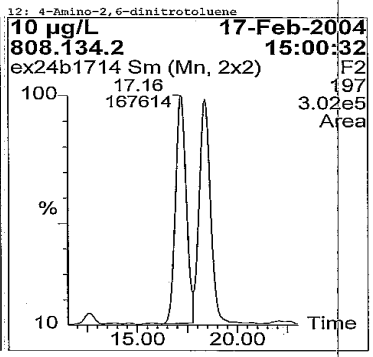
10: Nitrobenzene



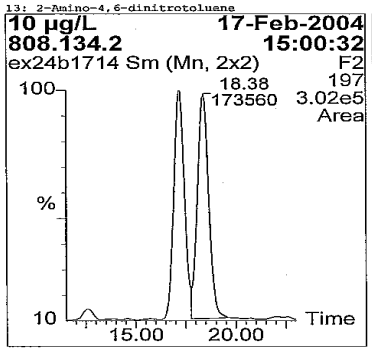
11: 2,4,6-Trinitrotoluene



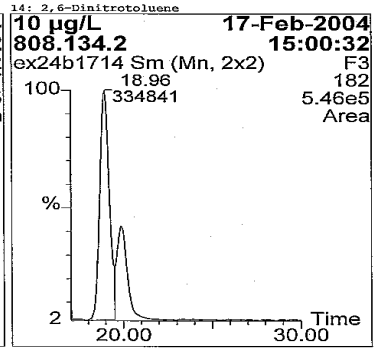
12: 4-Amino-2,6-dinitrotoluene



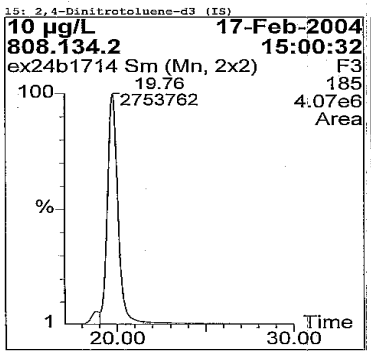
13: 2-Amino-4,6-dinitrotoluene



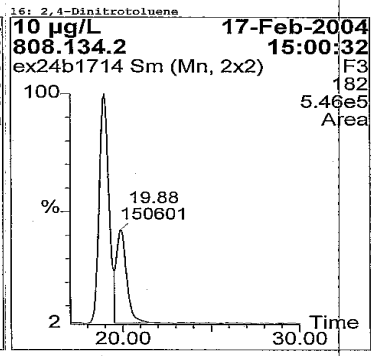
14: 2,6-Dinitrotoluene



15: 2,4-Dinitrotoluene-d3 (IS)



16: 2,4-Dinitrotoluene



Analyst: Steve Cowling

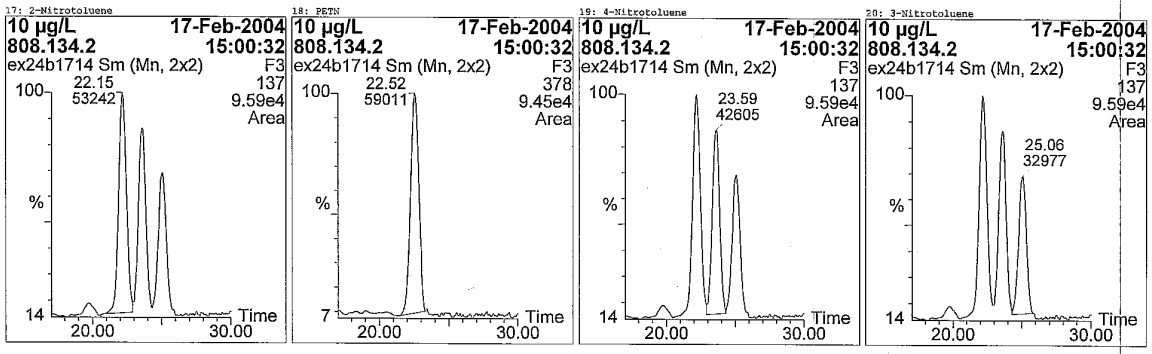
Explosives low std Compounds 17-20

Quantify Sample Report
Explosives Analysis

Sample List: C:\Masslynx\Explosives.PRO\SampleDB\ex24b17(2)
Last modified: Wed Feb 18 08:09:15 2004
Method: C:\Masslynx\Explosives.PRO\MethDB\ex24b17
Last modified: Wed Feb 18 07:54:11 2004
Job Code:

Printed: Wed Feb 18 08:10:42 2004

Name: ex24b1714
Text: 10 µg/L



#	Name	RT	Area	Response	Flags	Ion Ratio	Result µg/(L or kg)	%Rec	Mod.Date	Mod.Comment
1	HMX	7.56	977485	0.098	bb		9.5019	95.02		
2	RDX 13C-3 284 (IS)	9.90	9955472	9955472.000	bb		1.1071	110.71		
3	RDX	9.93	433314	0.044	bb		9.7358	97.36		
4	1,3,5-Trinitrobenzene	12.57	107224	0.115	bb		9.7866	97.87		
5	Tetryl	13.95	201024	0.215	dd		8.8059	88.06		
6	Dinitrobenzene-d4 (IS)	14.56	935503	935503.438	bb		0.9792	97.92		
7	1,3-Dinitrobenzene	14.69	53452	0.057	bb		10.4226	104.23		
8	Nitrobenzene-d5	15.69	34620	0.037	bb		9.7768	97.77		
9	Nitroglycerin	15.43	9846	0.011	ds		8.1835	81.83		
10	Nitrobenzene	15.96	30496	0.033	bb		9.9783	99.78		
11	2,4,6-Trinitrotoluene	16.97	260635	0.279	bb		10.2677	102.68		
12	4-Amino-2,6-dinitrotoluene	17.16	167614	0.061	bd		9.3638	93.64		
13	2-Amino-4,6-dinitrotoluene	18.39	173560	0.063	db		8.7447	87.45		
14	2,6-Dinitrotoluene	18.96	334841	0.122	bd		10.3072	103.07		
15	2,4-Dinitrotoluene-d3 (IS)	19.76	2753762	2753762.000	ds		0.9507	95.07		
16	2,4-Dinitrotoluene	19.88	150601	0.055	db		10.2509	102.51		
17	2-Nitrotoluene	22.15	53242	0.019	bd		9.4410	94.41		
18	PETN	22.52	59011	0.021	bb		9.5228	95.23		
19	4-Nitrotoluene	23.59	42605	0.015	dd		10.0234	100.23		
20	3-Nitrotoluene	25.06	32977	0.012	db		9.7548	97.55		

Analyst: Steve Cowling

Perfluorooctanoic acid, PFOA

- Used in the manufacture of fluoropolymers – non-stick cookware, water and stain resistant finishes, fire resistant finishes
- Persistent in the environment
- Related compounds, Perfluorooctyl sulfonate (PFOS) and perfluorooctanesulfonic acid (PFOSA) can be analyzed using the same method

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PFOA

- Extraction
  - Aqueous – SPE extraction using C18 cartridge
  - Solids - 10g sonicated with methanol
- LC – 250 mm C18 column, aqueous formic acid and methanol mobile phase
- MS – ESI negative ion MS/MS detection
- C13 labeled PFOA used as an internal standard and PFNA (closely related cmpd) used for a surrogate
- Calibration - 1 to 50 ug/L instrument concentration

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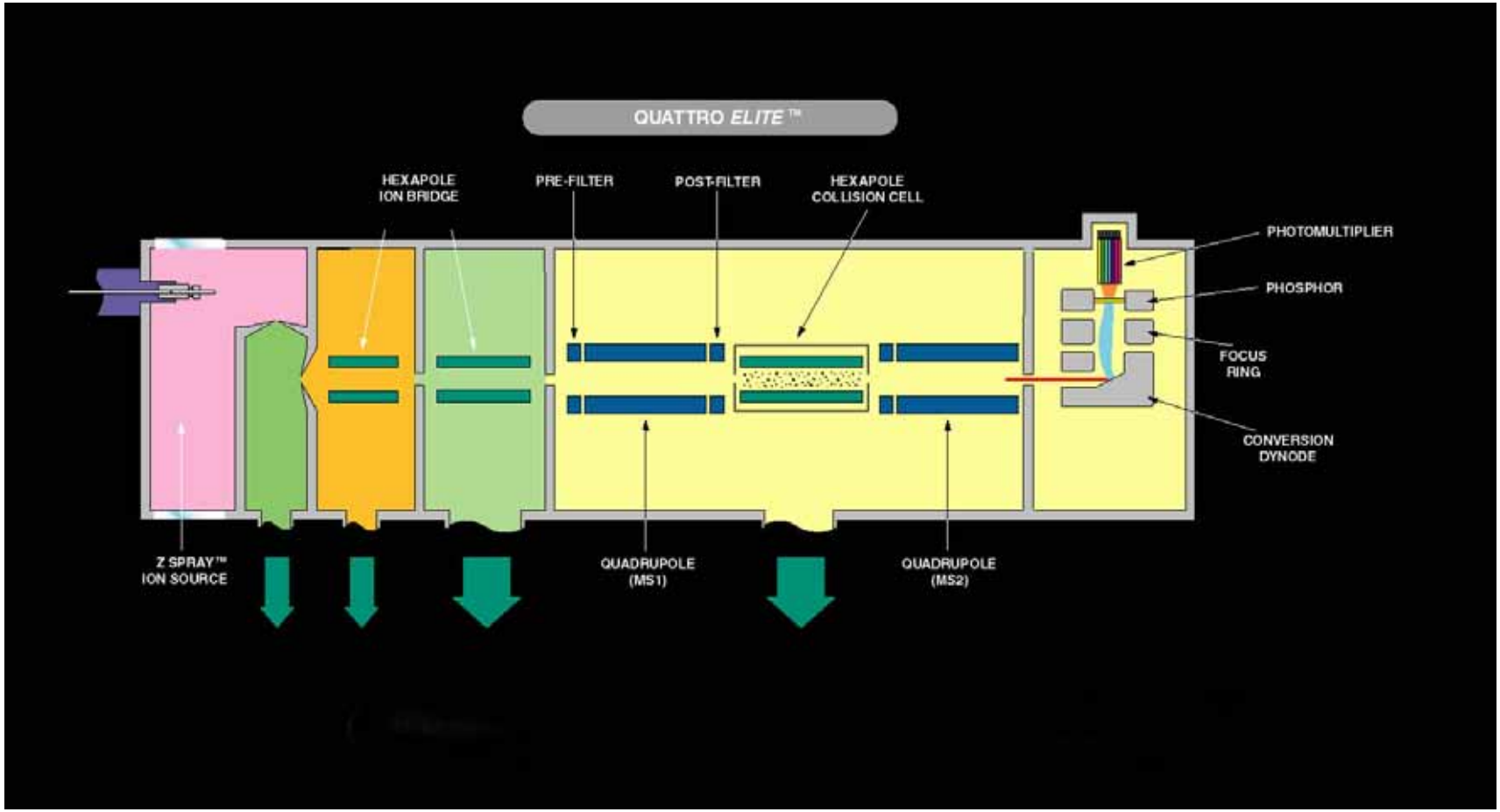
LC/MS/MS



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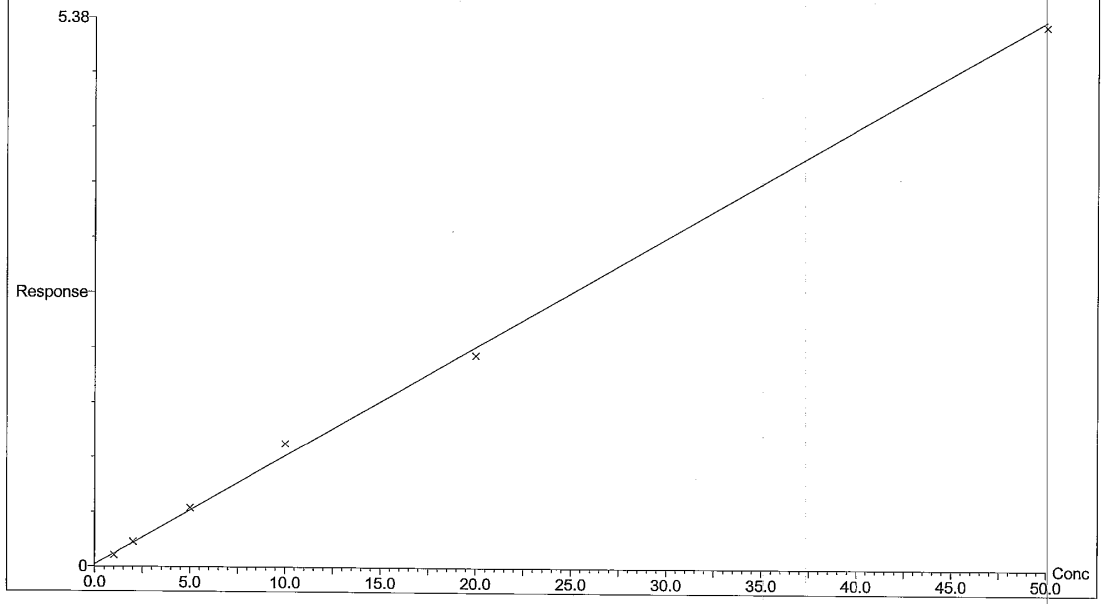
PFOA ICAL

Quantify Calibration Report
APFO Analysis

Page 2

Calibration: C:\MASSLYNX\PFOA.PRO\CurveDB\pf14b13b
Last modified: Mon Feb 16 06:42:20 2004
Printed: Mon Feb 16 10:42:07 2004

Compound 2 name: APFO
Coefficient of Determination: 0.998740
Calibration curve: 0.107085 * x + 0.0231085
Response type: Internal Std (Ref 1), Area * (IS Conc. / IS Area)
Curve type: Linear, Origin: Exclude, Weighting: 1/x, Axis trans: None



Analyst: Steve Cowling

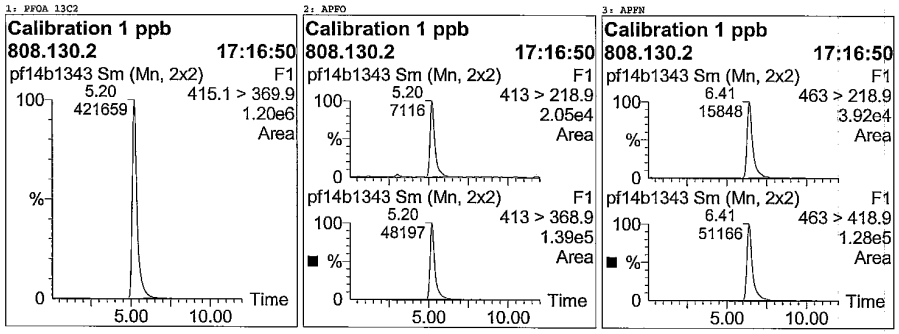
PFOA low std

Quantify Sample Report
APFO Analysis

Sample List: C:\MASSLYNX\PFOA.PRO\SampleDB\pf14b13(2)
Last modified: Mon Feb 16 10:41:14 2004
Method: C:\MASSLYNX\PFOA.PRO\MethDB\pf14b13
Last modified: Fri Feb 13 10:13:20 2004
Job Code:

Printed: Mon Feb 16 10:42:09 2004

Name: pf14b1343
Text: Calibration 1 ppb



#	Name	RT	Response	Flags	Result µg/(L or kg)	%Rec	Extract (L)	Sample (L or kg)	Dilution	Mod.Date	Mod.Comment
1	PFOA 13C2	5.20	421658.906	bb	1.0360	103.60	1.000	1.000	1.000		
2	APFO	5.20	0.114	bb	0.8516	85.16	1.000	1.000	1.000		
3	APFN	6.41	0.121	bb	1.0561	105.61	1.000	1.000	1.000		

Analyst: Steve Cowling

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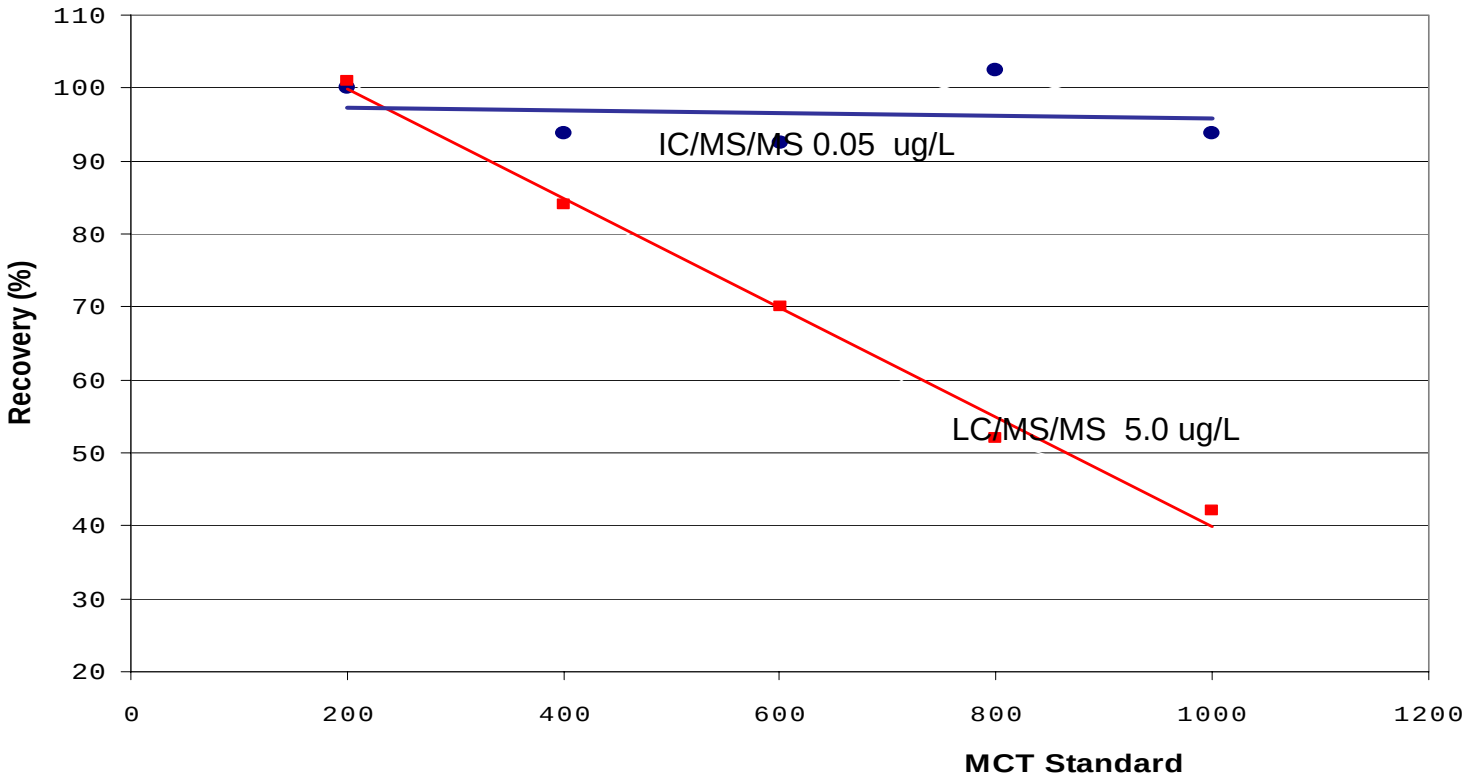
Perchlorate

- Extraction – soils tumbled with DI water, waters are analyzed directly
- IC Analysis
  - IC – AG16/AS16 column, using an eluant generator producing a potassium hydroxide mobile phase, a suppressor system and time actuated valves for sample diversion during the analysis
- MS – ESI negative ion MS/MS detection, heavy chlorine isotope monitored for confirmation
- O18 labeled perchlorate internal standard
- Calibration – 10 – 500 ng/L

ESI Ionization Suppression
(without IS correction)



Note: 1,000 MCT = 1,000 mg/L each chloride, sulfate, bicarbonate

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5,000 MCT Water Used for January 2004 Study

De-ionized water > 18 Megohm-cm

To which was added:

Chloride (NaCl)	= 5,000 mg/L
Sulfate (Na_2SO_4)	= 5,000 mg/L
Bicarbonate ($KHCO_3$)	= 5,000 mg/L
Total dissolved solids	= 22,600 mg/L

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Perchlorate Spikes Prepared for 2004 IC/MS/MS Study

Blank : 3 replicates of high TDS water

0.01 ug/L: 4 replicates in high TDS water

0.25 ug/L: 4 replicates in high TDS water

0.50 ug/L: 4 replicates in high TDS water

Notes:

- 1.0 ug/L O-18 perchlorate added to each
- Entire series prepared & analyzed on 3 days

IC/MS/MS Calibration

Each day of analysis:

Level	(ug/L)
1	0.005
2	0.01
3	0.02
4	0.05
5	0.10
6	0.20
7	0.50
8	1.0

$r = 0.9998_{\text{day 1}},\ 0.9999_{\text{day 2}},\ 0.9983_{\text{day 3}}$

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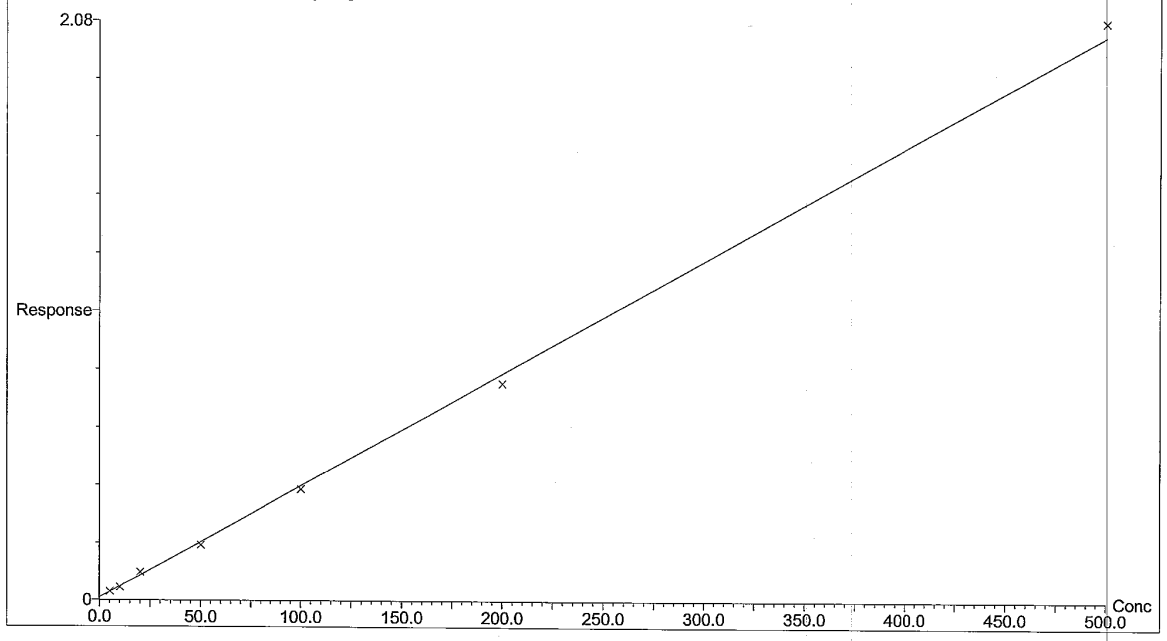
Perchlorate IC CAL

Quantify Calibration Report
Perchlorate Analysis

Page 1

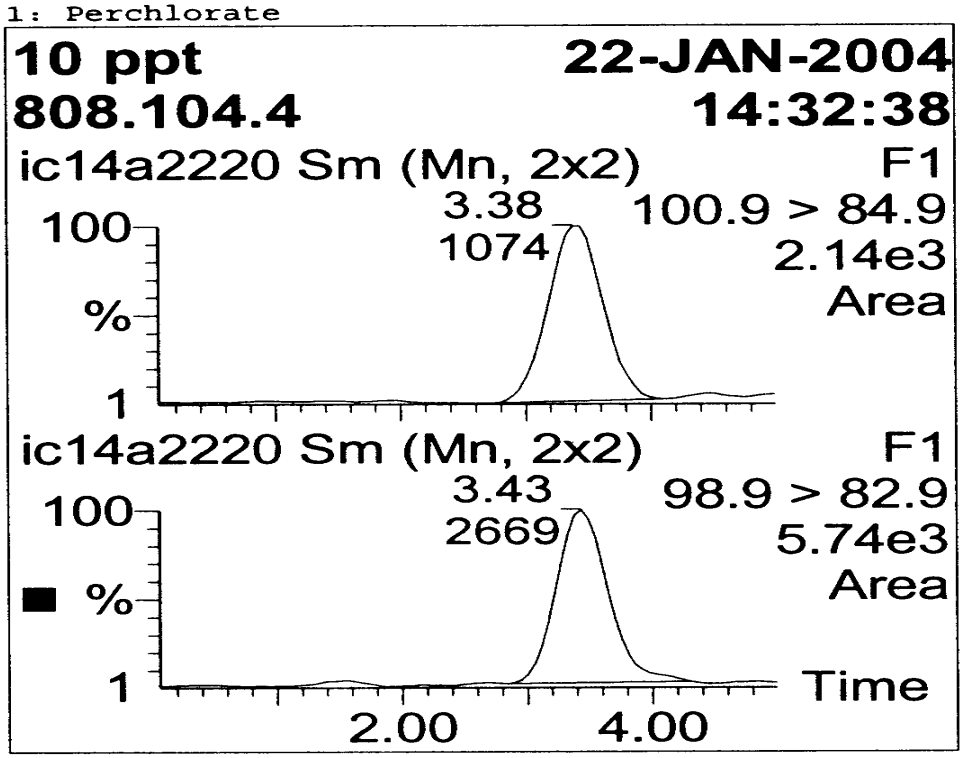
Calibration: C:\MASSLYNX\Perchlorate.PRO\CurveDB\ic14c10
Last modified: Wed Mar 10 14:19:48 2004
Printed: Thu Mar 11 06:56:39 2004

Compound 1 name: Perchlorate
Coefficient of Determination: 0.998726
Calibration curve: 0.00404910 * x + 0.0100597
Response type: Internal Std (Ref 2), Area * (IS Conc. / IS Area)
Curve type: Linear, Origin: Exclude, Weighting: 1/x, Axis trans: None



Analyst: Steve Cowling

0.01 ug/L Perchlorate Calibration Standard



← 85 ion

1,074 area counts

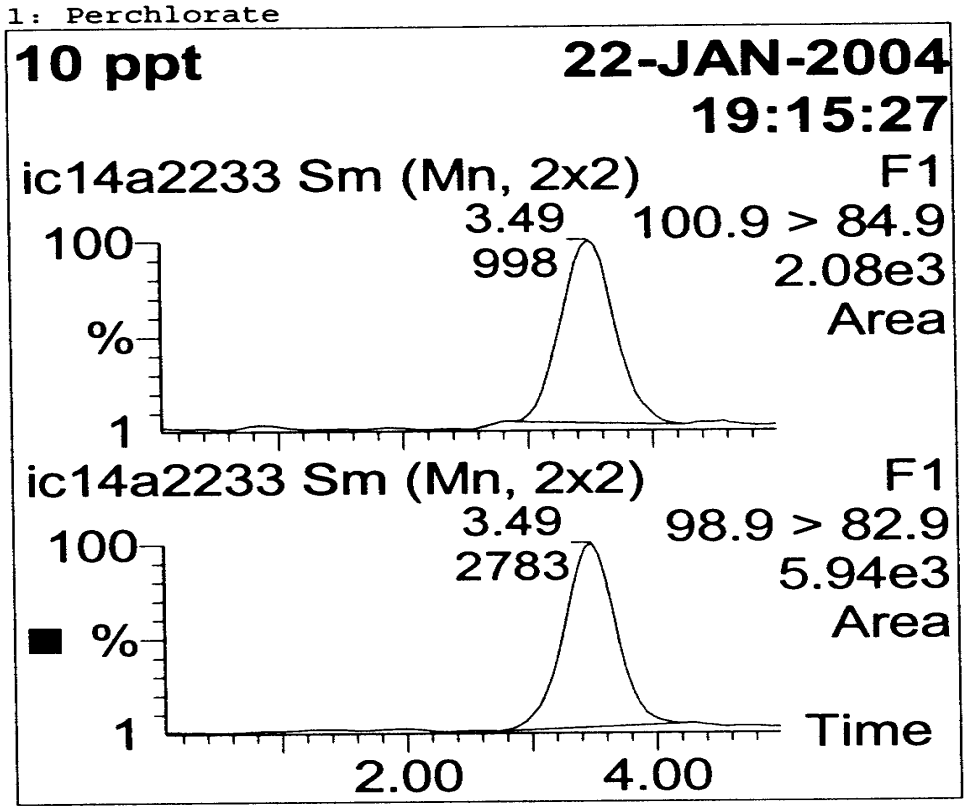
← 83 ion

2,669 area counts

True RT = 10.5+3.4 = 13.9 min.

0.01 ug/L Perchlorate In High TDS Water

After running high TDS samples for hours:



← 85 ion

998 area counts

← 83 ion

2,783 area counts

True RT = 10.5+3.5 = 14.0 min.

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Day 1 Data Could Not Be Used

- Perchlorate contamination in all samples at 0.06 - 0.08 ug/L
- Contamination control proved to be more challenging at ppt levels
- Previously confirmed perchlorate in some lab detergents at low mg/kg levels
- This time traced to vinyl lab gloves

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IC/MS/MS Precision / Accuracy Data in High TDS Water

Day2 &Day3:

Test No.*	True Value (ug/L)	Mean Recovery (%)	RSD (%)
1	0.01	116.8	14.9
2	0.25	99.2	2.71
3	0.5	93.6	2.84

- Spikes prepared in water with 22,600 mg/L TDS
- No pretreatment
- 8 replicates tested per concentration, 4 on each of 2 days
- O-18 labeled perchlorate used as internal standard

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N-Nitrosodimethylamine, NDMA

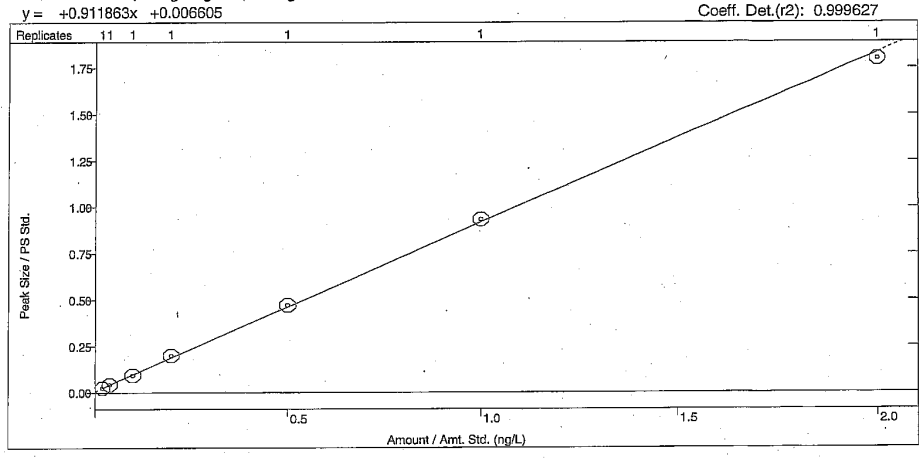
- GC/CI/MS/MS positive ion analysis
  - CI gas – ammonia
- Extraction – CLLE of 1L water with CH_2Cl_2, concentration to 1.0 ml final volume
- 624 type capillary column with helium carrier gas
- Cryogenic cool on-column injection
- NDMA-d6 used for an isotope dilution standard
- Concentration – 1.0 to 100 ug/L instrument concentration

NDMA Calibration

NDMA confirmation

Curve Fit: Linear, Origin: Ignore, Weight: 1/X
y = +0.911863x +0.006605

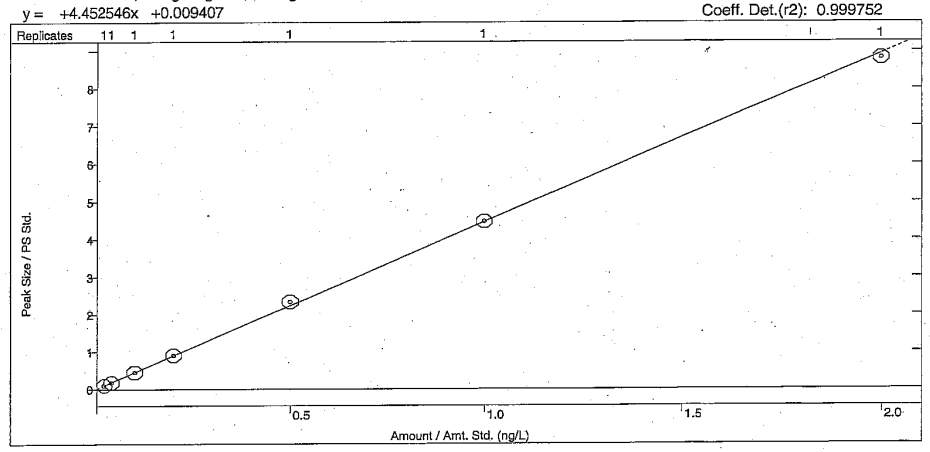
Resp. Fact. RSD: 10.77%
Coeff. Det.(r2): 0.999627



NDMA

Curve Fit: Linear, Origin: Ignore, Weight: 1/X
y = +4.452546x +0.009407

Resp. Fact. RSD: 4.714%
Coeff. Det.(r2): 0.999752



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NDMA low std

Print Date: 21 Mar 2004 12:57:13

Target Compound Report for #3 from nd34c20001.xms - Page 3

Sample ID:	1.0 ppb std	Operator:	md
Instrument ID:	Varian MS #1	Last Calibration:	3/21/2004 12:55 PM
Measurement Type:	Area	Calibration Type:	Internal Standard
Acquisition Date:	3/20/2004 3:43 PM	Data File:	...ch 04\nd34c20001.xms
Calculation Date:	3/21/2004 12:55 PM	Method:	...o1 initial split.mth
Sample Type:	Calibration		
Inj. Sample Notes:	None		

Compound Information

Peak Name: NDMA Adduct
Result Index: 3 Compound Number: 3 CAS Number: None Identified

Identification

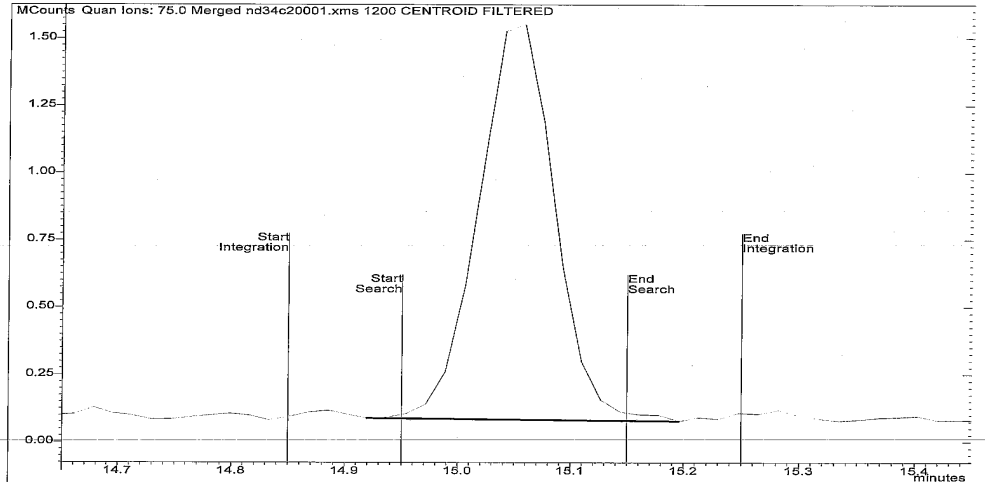
Parameter	Specification	Actual	Status
Search Type	Spectrum		
Retention Time	15.050 +/- 0.100	15.048 min.	Pass
Match Result	N-R >= 700	1000	Pass

Integration and Quantitation

Parameter	Specification	Actual	Status
Quan Ions	75.0		
IS Peak Name	NDMA-d6		
Calibration Equation	Linear, Include, 1/X	y = +3.7061x +0.0	
Area	>=100	6.920e+6	Pass
Height		1.456e+6	
Amount (RRF)		3.706	

0.074129

Match Types: N-R : Normal-Reverse



Seg 1, Time: 10.15-16.01, Channels: 3

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Conclusions

- Electron impact GC/MS works for many analytes, but not for everything
- LC/MS, LC/MS/MS , IC/MS/MS and CI-GC/MS/MS can provide definitive data
- MS/MS is very desirable when soft ionization techniques are used
- Ionization suppression is a concern in LC/MS, and isotopically labeled internal standards are the best solution
- When a lab claims a low detection limit, check the signal to noise

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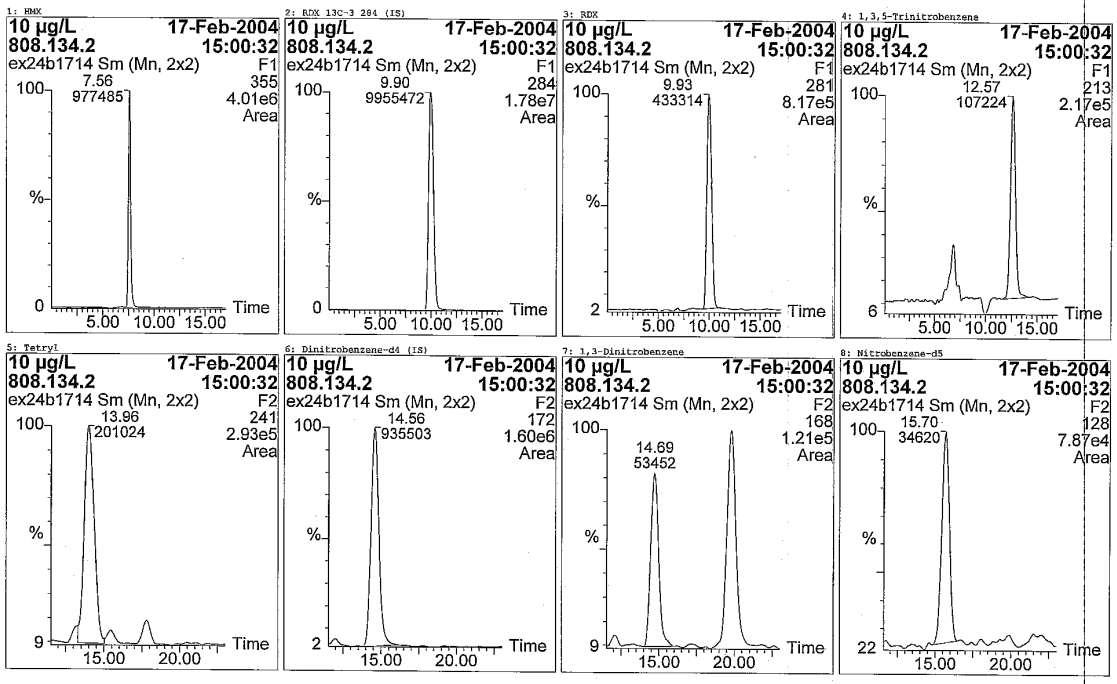
Good

Quantify Sample Report
Explosives Analysis

Sample List: C:\Masslynx\Explosives.PRO\SampleDB\ex24b17(2)
Last modified: Wed Feb 18 08:09:15 2004
Method: C:\Masslynx\Explosives.PRO\MethDB\ex24b17
Last modified: Wed Feb 18 07:54:11 2004
Job Code:

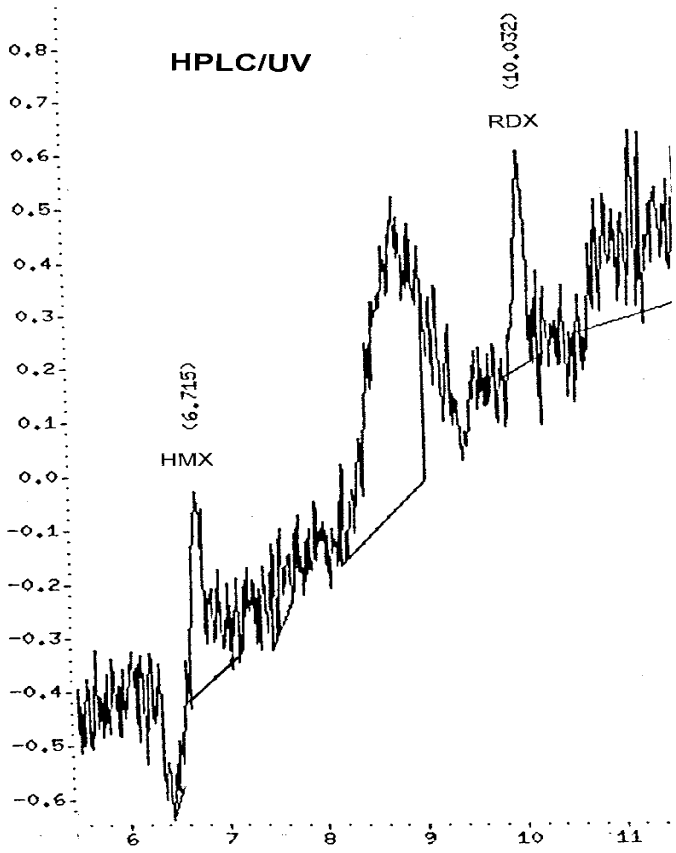
Printed: Wed Feb 18 08:10:42 2004

Name: ex24b1714
Text: 10 µg/L



Analyst: Steve Cowling

Not so good


HPLC/UV
(10.032)
RDX
(6.715)
HMX
0.8
0.7
0.6
0.5
0.4
0.3
0.2
0.1
0.0
-0.1
-0.2
-0.3
-0.4
-0.5
-0.6
6
7
8
9
10
11

Conclusions

- Electron impact GC/MS works for many analytes, but not for everything
- LC/MS, LC/MS/MS , IC/MS/MS and CI-GC/MS/MS can provide definitive data
- MS/MS is very desirable when soft ionization techniques are used
- Ionization suppression is a concern in LC/MS, and isotopically labeled internal standards are the best solution
- When a lab claims a low detection limit, check the signal to noise

Acknowledgments

Mark Dymerski Steve Cowling Larry Penfold

STL Denver

Further Information

Richard Burrows (303)736-0100 rburrows@stl-inc.com

Explosives CAL example RDX

Quantify Calibration Report
Explosives Analysis

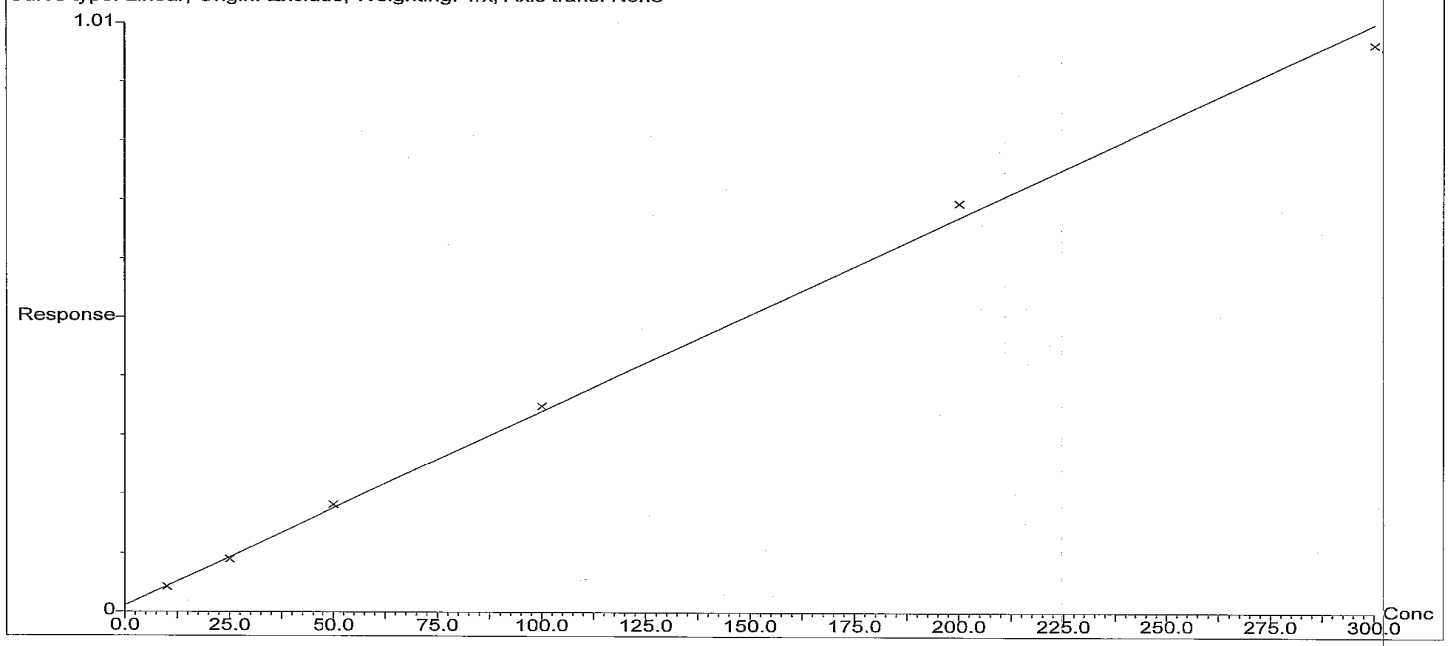
Page 3

Calibration: C:\Masslynx\Explosives.PRO\CurveDB\ex24b17
Last modified: Wed Feb 18 07:54:31 2004
Printed: Wed Feb 18 08:10:39 2004

Result (µg/L or kg) = Amt * DF * Vf / Vs

Amt = on-column concentration (µg/L)
DF = Dilutions after extraction (L/L)
Vf = Final volume at end of extraction (L)
Vs = Size of sample Extracted (L or kg)

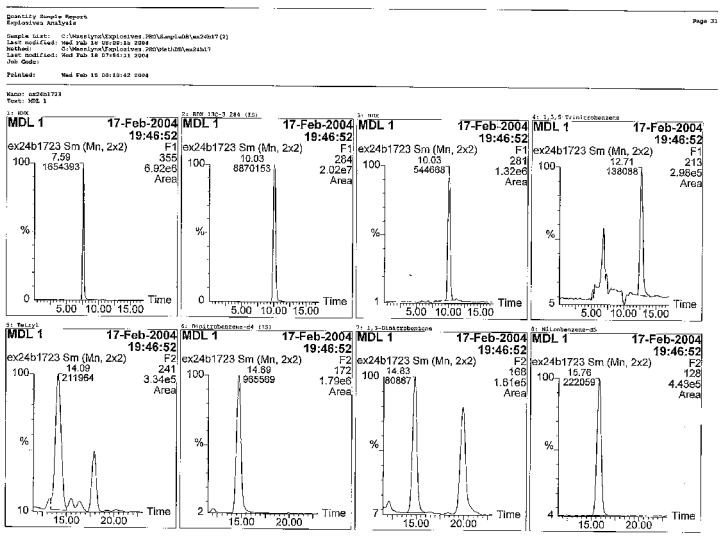
Compound 3 name: RDX Method File: ex24b17
Coefficient of Determination: 0.997169
Calibration curve: 0.00333146 * x + 0.0110907
Response type: Internal Std (Ref 2), Area * (IS Conc. / IS Area)
Curve type: Linear, Origin: Exclude, Weighting: 1/x, Axis trans: None



Analyst: Steve Cowling

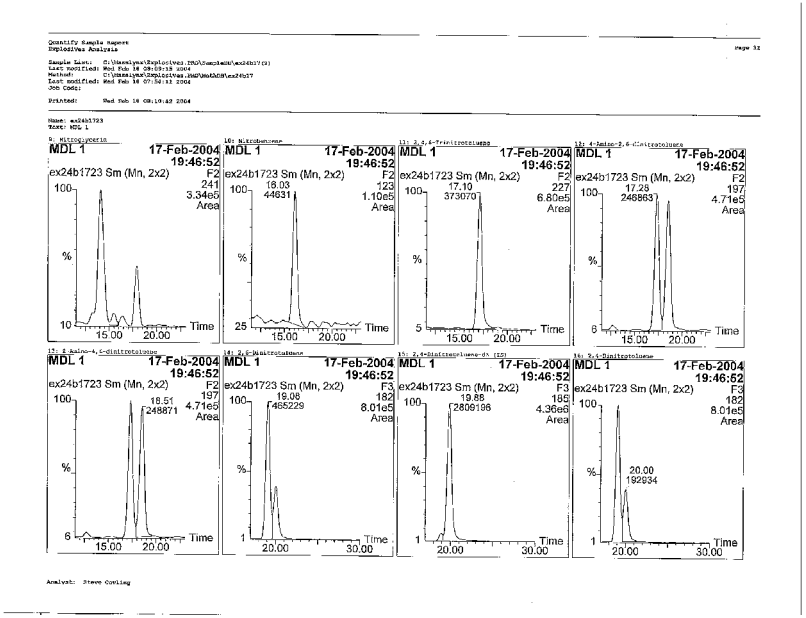
SEVERN TRENT STL

Exp MDL 1



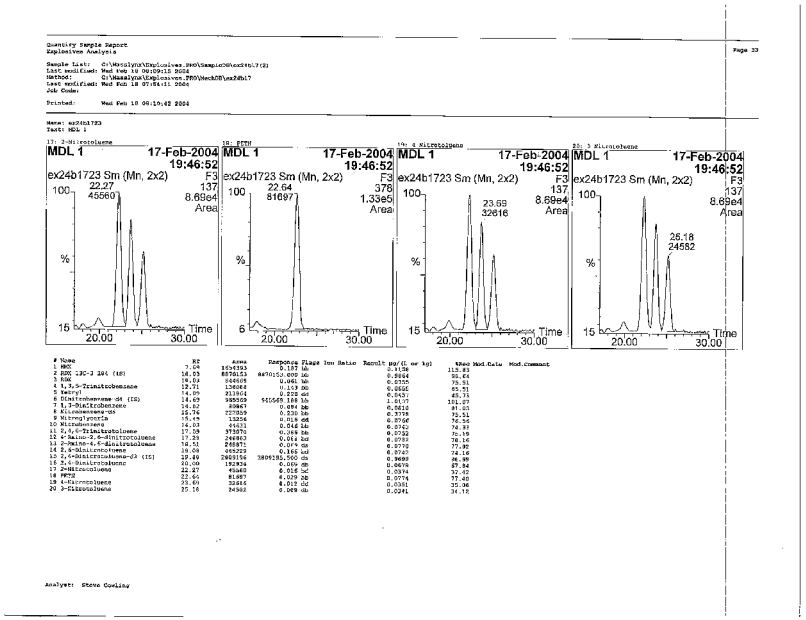
SEVERN TRENT STL

Exp mdl 2



SEVERN TRENT STL

Exp mdl 3



Quantity Sample Report
Explosives Analysis

Page 33

Sample List: C:\Mcolyn\Explosives.P80\Sample80\ex24b17(2)
Saved Time: Wed Feb 18 08:09:15 2004
Method: C:\MassLynx\Explosives.PM9\Meth58\ex24b17
Last modified: Wed Feb 18 07:54:11 2004
Job Code:

Printed: Wed Feb 18 08:10:42 2004

Name: ex24b1723
Test: MDL 1

17: 2-Nitrotoluene
MDL 1 17-Feb-2004 19:46:52
ex24b1723 Sm (Mn, 2x2) F3
137
8.69e4
Area
22.27
45560
100
%
15
20.00
30.00
Time

18: PETN
MDL 1 17-Feb-2004 19:46:52
ex24b1723 Sm (Mn, 2x2) F3
378
1.33e5
Area
22.64
81697
100
%
6
20.00
30.00
Time

19: 4 Nitrotoluene
MDL 1 17-Feb-2004 19:46:52
ex24b1723 Sm (Mn, 2x2) F3
137
8.69e4
Area
23.69
32616
100
%
15
20.00
30.00
Time

20: 3 Nitrotoluene
MDL 1 17-Feb-2004 19:46:52
ex24b1723 Sm (Mn, 2x2) F3
137
8.69e4
Area
26.18
24582
100
%
15
20.00
30.00
Time

#	Name	RT	Area	Response	Flags	Ion Ratio	Result	ug/L or kg	%Rec	Mod.Calc	Mod.Comment
1	MOC	7.04	1654353	0.157	Ub		2.1138		115.83		
2	RDX 13C-3 284 (IS)	16.03	8870153	8870153.000	Ub		0.8864		88.64		
3	RDX	16.03	348608	0.041	Ub		0.0355		79.31		
4	1,3,5-Trinitrobenzene	12.71	136084	0.153	Bb		0.0656		65.71		
5	HMX	14.09	211914	0.228	Bb		0.0431		43.73		
6	Dinitrobenzene d6 (IS)	14.09	9853647	9853647.000	Bb		1.0107		101.07		
7	1,3-Dinitrobenzene	14.62	355967	0.038	Bb		0.0818		81.83		
8	Nitrobenzene-d5	15.76	227859	0.230	Bb		0.0756		75.51		
9	Nitroglycerin	15.49	13254	0.015	dB		0.0746		74.59		
10	Nitrobenzene	16.03	41631	0.042	Bb		0.0242		24.23		
11	2,4,6-Trinitrotoluene	17.58	373870	0.268	Bb		0.0752		75.19		
12	4-Amino-2,6-Dinitrotoluene	17.23	246802	0.064	bd		0.0769		76.16		
13	2-Amino-4,6-Dinitrotoluene	16.51	266871	0.069	dd		0.0772		77.22		
14	2,4-Dinitrotoluene	18.08	465229	0.166	bd		0.0747		74.16		
15	2,6-Dinitrotoluene-d3 (IS)	18.49	2809156	2809156.500	db		0.8898		88.98		
16	1,3-Dinitrobenzene	20.08	152834	0.054	dB		0.0878		87.84		
17	2-Nitrotoluene	22.27	41468	0.016	bC		0.0374		37.42		
18	PETN	22.64	81697	4.079	dB		0.0774		77.40		
19	4-Nitrotoluene	23.69	32616	0.012	CC		0.0351		35.06		
20	3-Nitrotoluene	26.18	24582	0.009	db		0.0241		24.10		

Analyst: Steve Cowling

NDMA mdl example

Print Date: 21 Mar 2004 13:01:41

Target Compound Report for #3 from nd34c20011.xms - Page 3

Sample ID:	mdl #1	Operator:	md
Instrument ID:	Varian MS #1	Last Calibration:	3/21/2004 12:55 PM
Measurement Type:	Area	Calibration Type:	Internal Standard
Acquisition Date:	3/20/2004 7:36 PM	Data File:	...ch 04\nd34c20011.xms
Calculation Date:	3/21/2004 12:55 PM	Method:	...o1 initial split.mth
Sample Type:	Analysis		
Inj. Sample Notes:	None		

Compound Information

Peak Name: NDMA Adduct

Result Index: 3 Compound Number: 3 CAS Number: None Identified

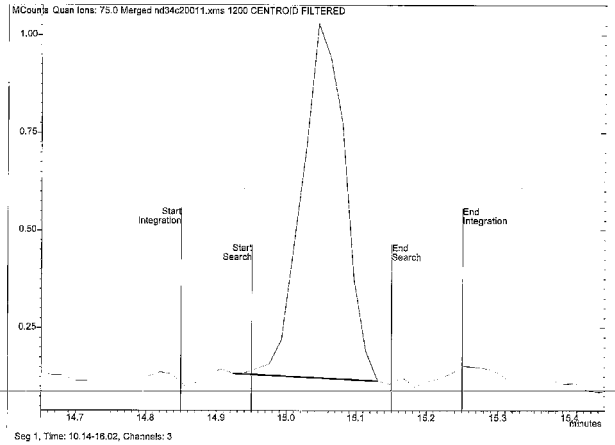
Identification

Parameter	Specification	Actual	Status
Search Type	Spectrum		
Retention Time	15.050 +/- 0.100	15.049 min.	Pass
Match Result	N-R >= 700	1000	Pass

Integration and Quantitation

Parameter	Specification	Actual	Status
Quan Ions	75.0		
IS Peak Name	NDMA-d6		
Calibration Equation	Linear, Ignore, 1/X	y = +3.3994x +0.0027	
Area	>=100	3.923e+6	Pass
Height		907862	
Amount	>= 0.000 ng/L	1.233 ng/L	Pass

Match Types: N-R : Normal-Reverse



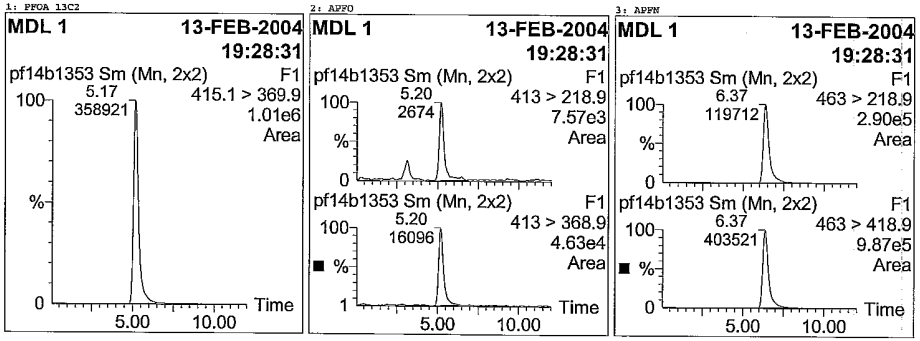
PFOA soil mdl example

Quantify Sample Report
APFO Analysis

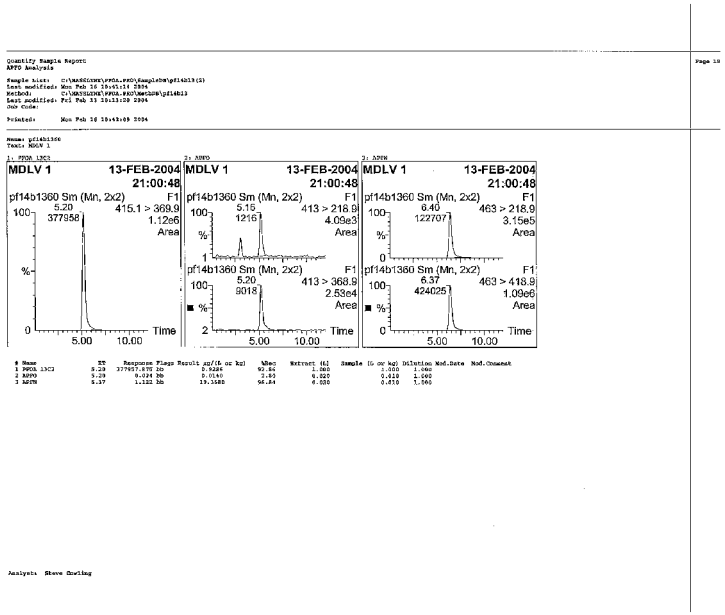
Sample List: C:\MASSLYNX\PFOA.PRO\SampleDB\pf14b13(2)
Last modified: Mon Feb 16 10:41:14 2004
Method: C:\MASSLYNX\PFOA.PRO\MethDB\pf14b13
Last modified: Fri Feb 13 10:13:20 2004
Job Code:

Printed: Mon Feb 16 10:42:09 2004

Name: pf14b1353
Text: MDL 1



#	Name	RT	Response	Flags	Result µg/(L or kg)	%Rec	Extract (L)	Sample (L or kg)	Dilution	Mod.Date	Mod.Comment
1	PFOA 13C2	5.17	358920.531	bb	0.8818	88.18	1.000	1.000	1.000		
2	APFO	5.20	0.045	bb	0.4060	40.60	0.020	0.010	1.000		
3	APFN	6.37	1.124	bb	19.4090	97.04	0.020	0.010	1.000		

Analyst: Steve Cowling

PFOA mdlv example