

The Agilent 5975 inert MSD: New Tools for the Forensic Analyst

Chemical Analysis Group

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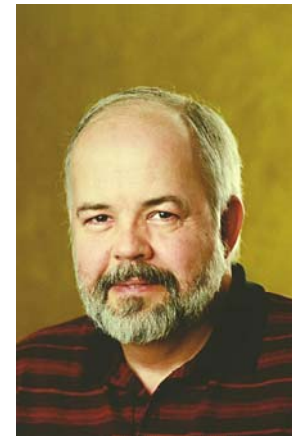


The Speaker

Bruce Quimby is a Senior Applications Chemist in the Chemical Analysis Solutions unit of Agilent Technologies in Wilmington, Delaware. He received a Ph.D. in analytical chemistry from the University of Massachusetts (Amherst) in 1980 and a batchelors degree in chemistry from Mansfield State College (PA) in 1974 . He has been at Agilent Technologies (formerly Hewlett-Packard) since 1979, working the first 10 years in research and development. He has authored or co-authored 18 journal articles and 10 patents in the field of gas chromatography. He is currently working in Forensic and Homeland Security applications.



R&D ID Photo



Marketing ID Photo

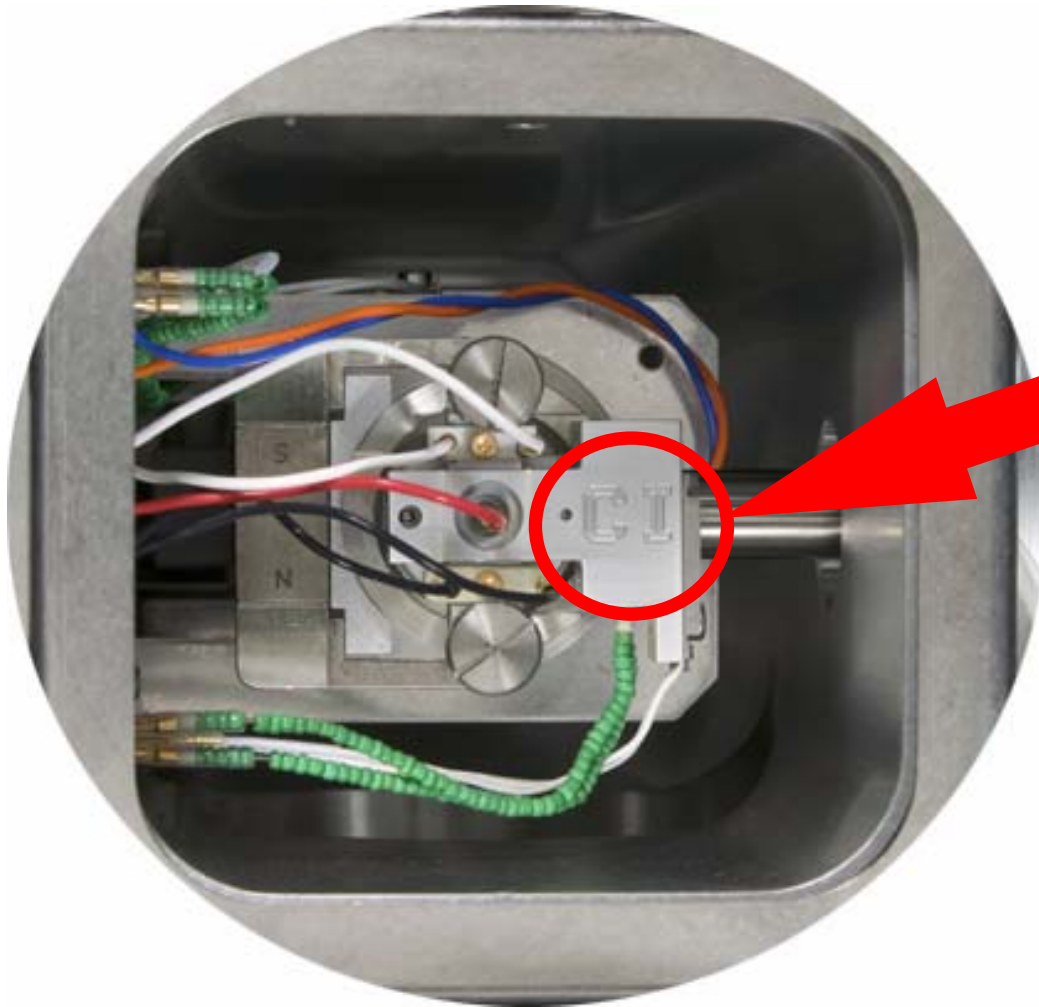
Introducing the 5975 inert MSD



What's Different?

- Does everything a 5973 does, plus...
- Synchronous SIM/Scan
- Extended mass range
- AutoCI and EI with CI source
- 6850 GC control
- QuickSwap MSD interface
- Permanent Effluent Splitters
- Deconvolution Reporting Software

Functional Front Window – Ease of Use



- ***Wire connections***
- ***Filament operation***
- ***Column insertion***
- ***CI Source identification***

Quick Access – Ease of Use



Easy analyzer access –
hinged side cover

Extended Mass Range – Performance

1050 m/z for 5975 inert MSD

- Quadrupole modified
- New electronics
- 800 m/z for all 5973 MSDs
(Software upgrade does not enable 1050 m/z)

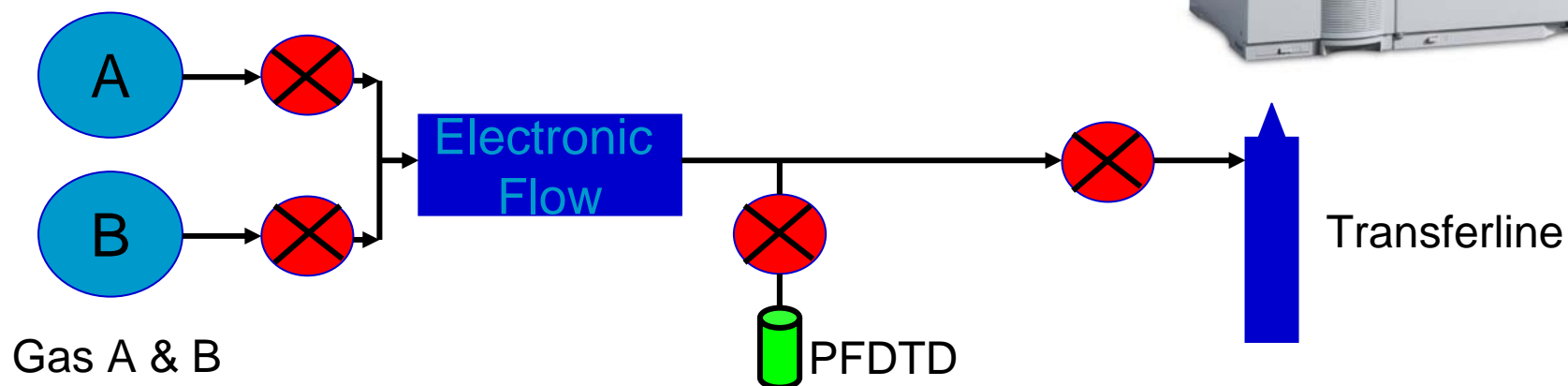


AutoCI – Performance

- No external manual control
- Fully automated setup/tuning
- Flexible Configuration
- EI capability with CI Source
- AutoCI – as easy as EI

AutoCI Electronic Flow Control – Ease of Use

- Reagent gas automatically adjusted by tune
 - Ammonia compatibility
- Ensures easy reproduction of application



Ion Source – Improved Response

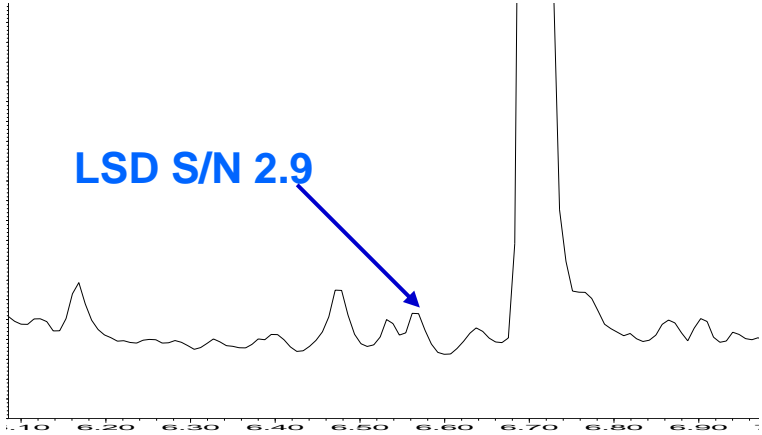
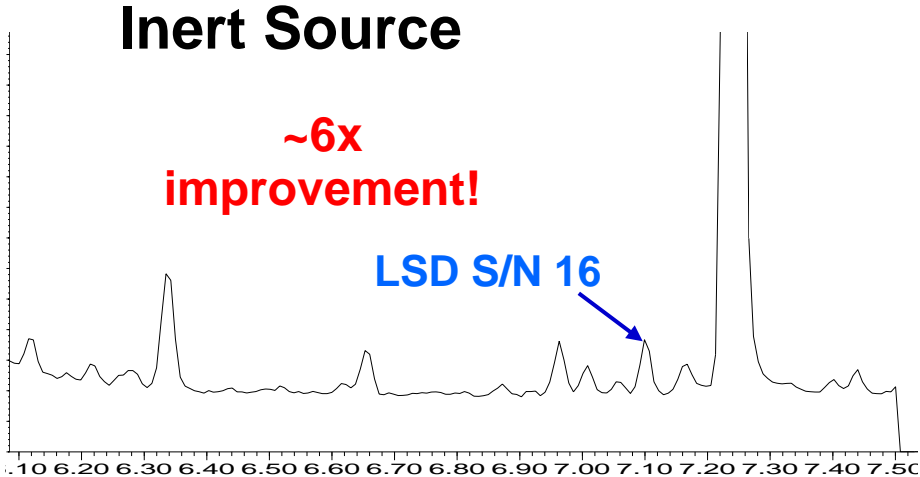
Data from a production forensic drug lab...

Inert Ion source – Improved response

50pg LSD

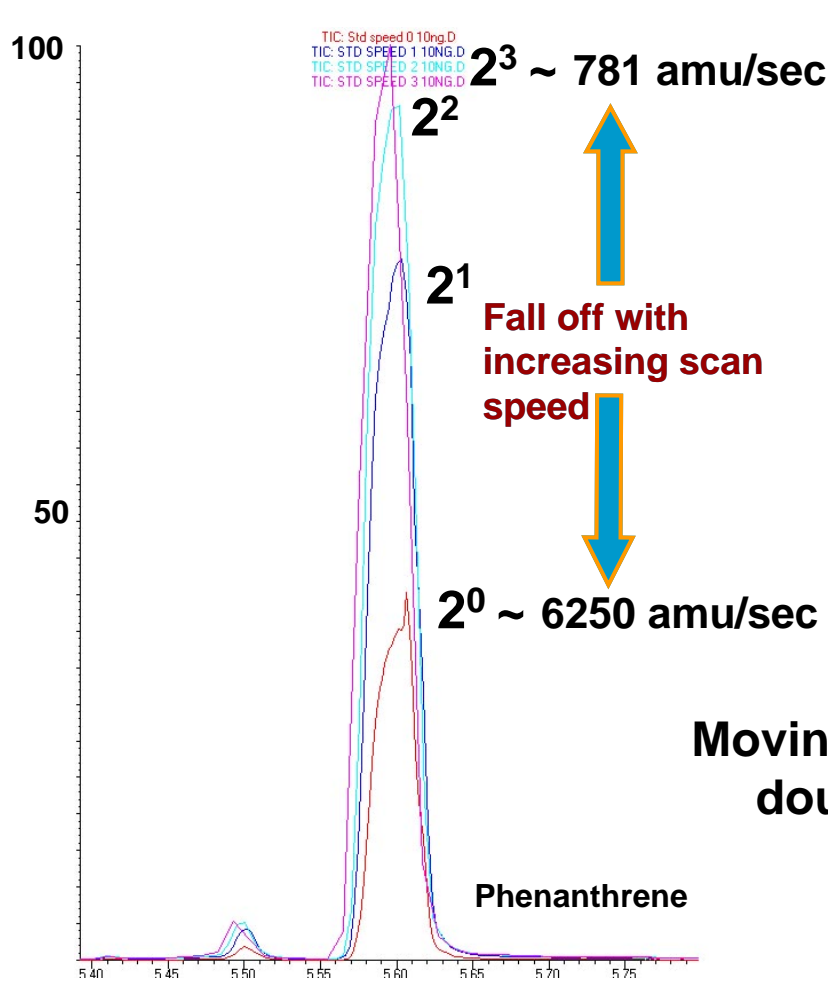
SS Source

Extracted Ion 253 m/z

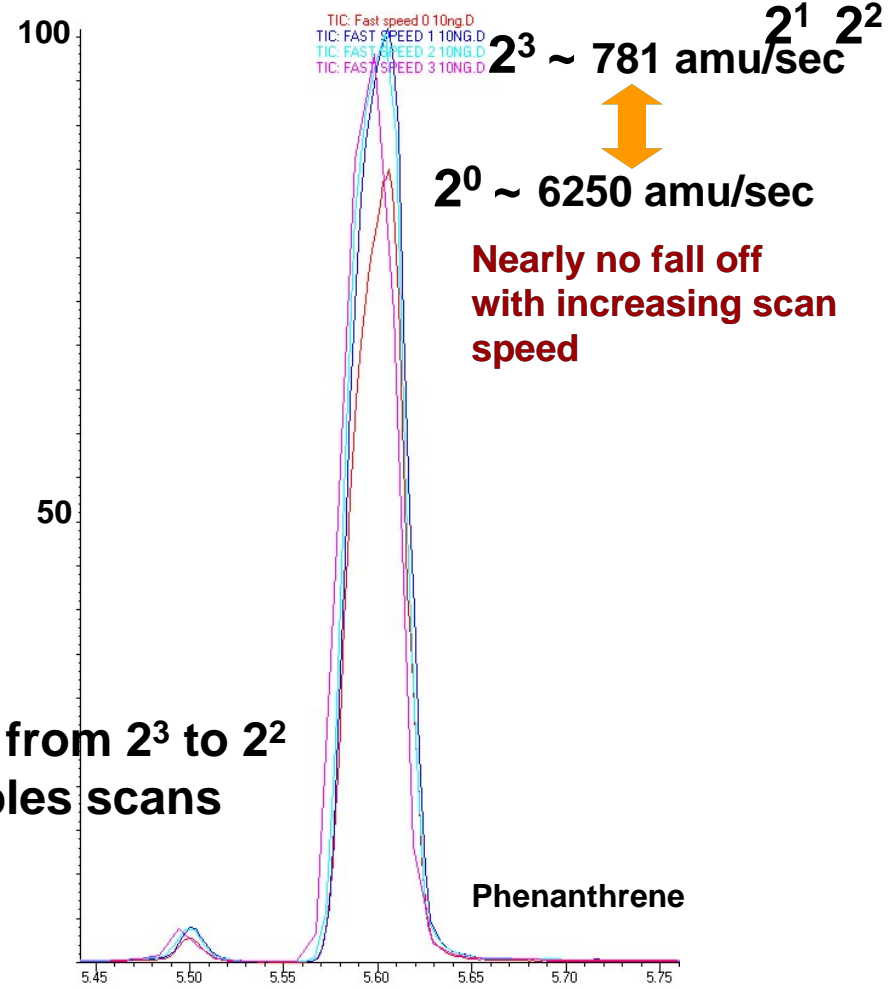


Faster Scan Speeds and 5 msec SIM Dwell

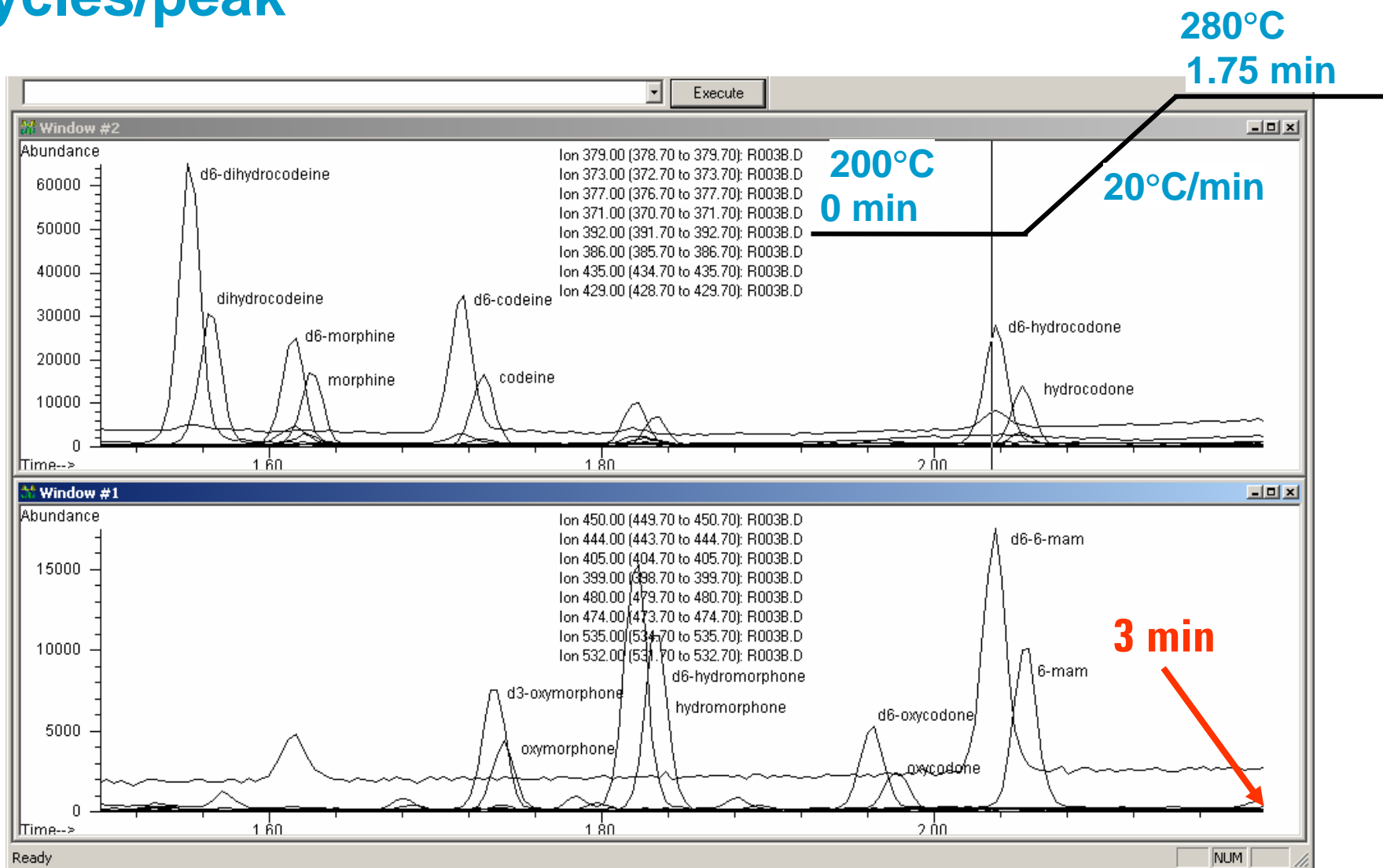
Standard Electronics



Performance Electronics



Opiates, 38 ions (5 msec dwell) in 1 group, 10 SIM cycles/peak



What is Synchronous SIM/Scan?

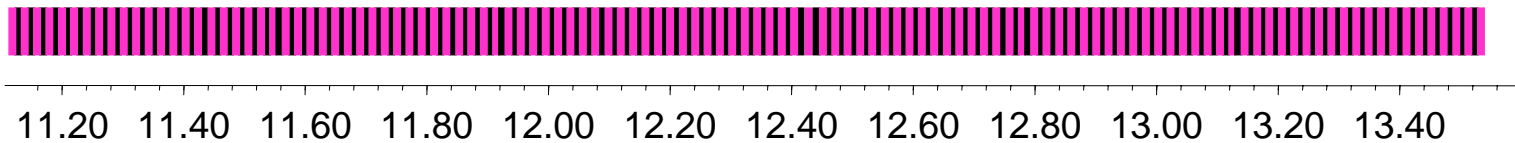
Agilent's Synchronous SIM/Scan allows for the continuous collection of both SIM and Scan data in the SAME acquisition. More data without sacrificing sensitivity or library searching capability

Great for target analysis (SIM) and identifying unknowns (Scan)

- SIM data for quantitation of targets
- Scan data for confirmation of unknowns

Agilent Synchronous SIM/Scan

Alternating SIM (black) and scan (magenta) data collection



SIM + scan in one cycle, alternating rapidly

**High sensitivity + high information throughout
the whole chromatogram**

Agilent Instrumentation Used

Gas Chromatograph

- 6890N with Autosampler (tray & injector) and split/splitless inlet
- Capillary NPD with extended jet
- G3180B - Microfluidic Splitter with Makeup Gas

Mass Spectrometer

- 5975 inert MSD with EI source

Software

- GC/MSD Chemstation G1701 DA version D.02.00 or higher (includes RTL and Screener)

Column

- DB-17MS 15m x 250 μ m x 0.25 μ m part # 122-4712

Method Parameters

GC **Agilent 6890N**
Oven
Ramp **'C/min** **'C** **Hold min**
Initial **80** **0.00**
Ramp 1 **20** **320** **5.00**
Runtime **17 min**

Inlet **Split/Splitless**
Temp **280 'C**
Mode **Splitless**
Pressure **23.16 psi**
Purge Flow **50 mL/min**
Purge time **2.00 min**
Total Flow **56.3 mL/min**

Column **DB-17MS part # 122-4712**
Mode **Constant pressure**
Pressure **23.16 psi**
Initial Flow **3.8 mL/min**
Outlet **Splitter**
Outlet Pressure **3.8 psig**

MSD **Agilent 5975inert**
Solvent Delay **2.5 min**
Scan Range **42 to 450**
Threshold **0**
Sampling **2² for scan, 2¹ for SIMSCAN**
Quad Temp **150 'C**
Source Temp **280 'C**
Transfer Line **280 'C**

Injector
Sample Washes **0**
Sample Pumps **6**
Injection volume **2 uL**
Solv A washes **4**
Solv B washes **4**
Viscosity Delay **3**
Plunger Speed **Fast**

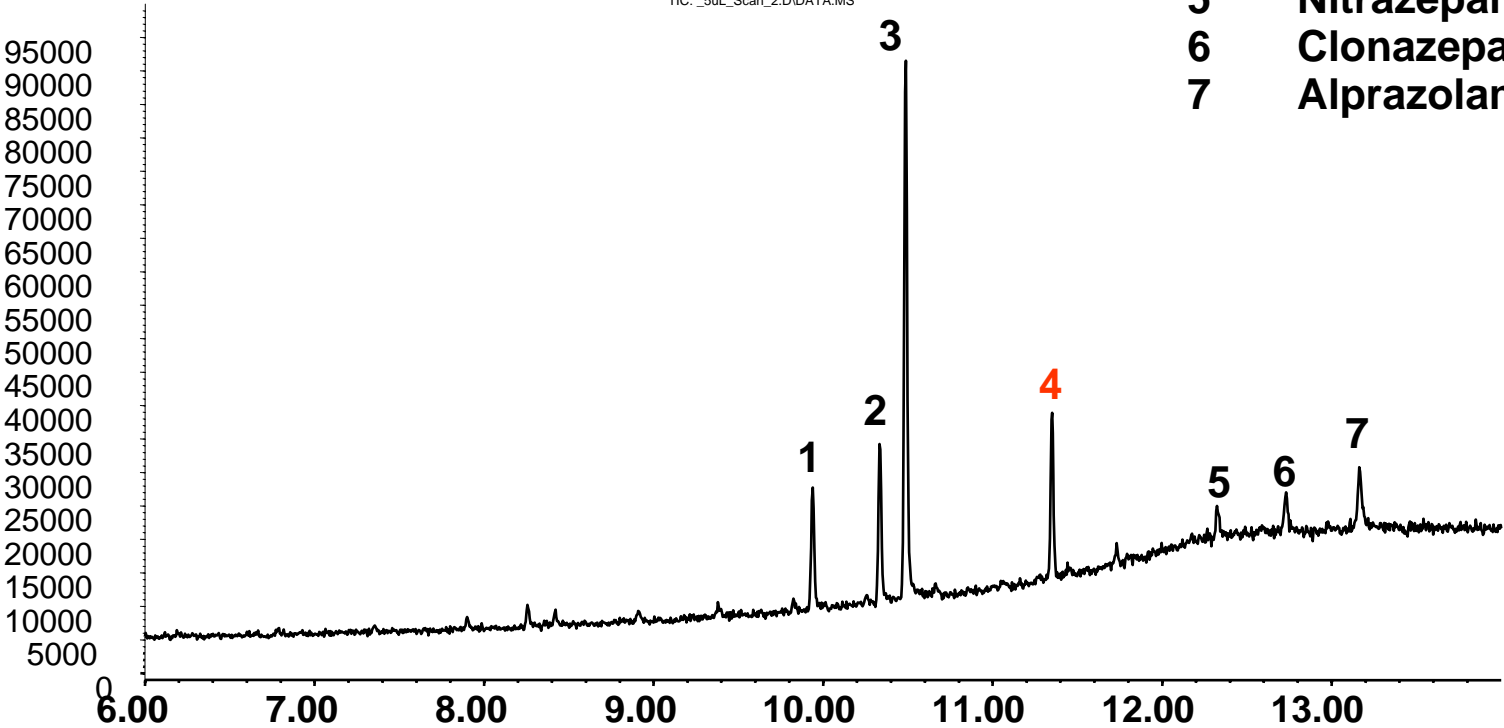
Benzodiazepines

0.63 ng each to MSD

- 1 Oxazepam
- 2 Lorazepam
- 3 Diazepam
- 4 Flunitazepam
- 5 Nitrazepam
- 6 Clonazepam
- 7 Alprazolam

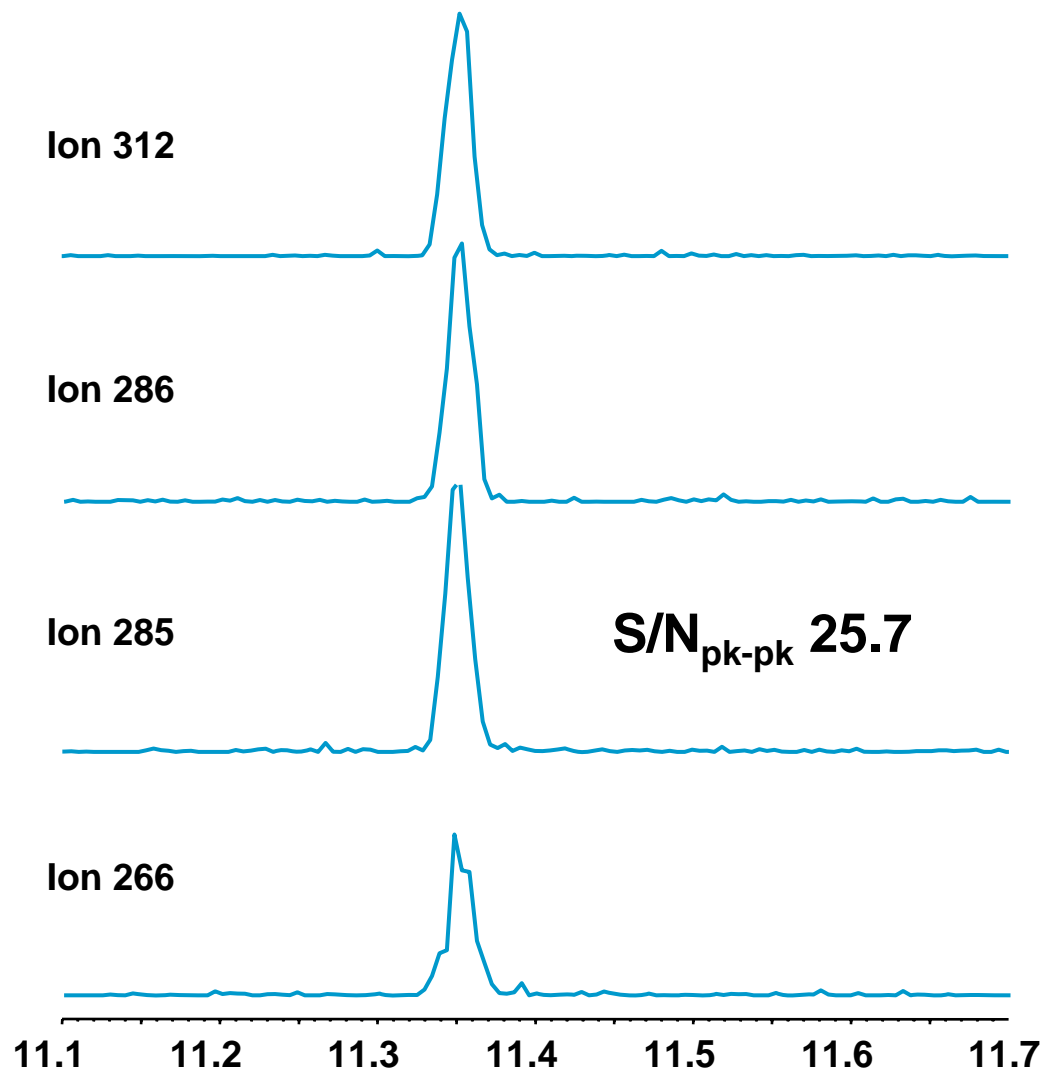
Abundance

TIC: _5uL_Scan_2.D\\DATA.MS



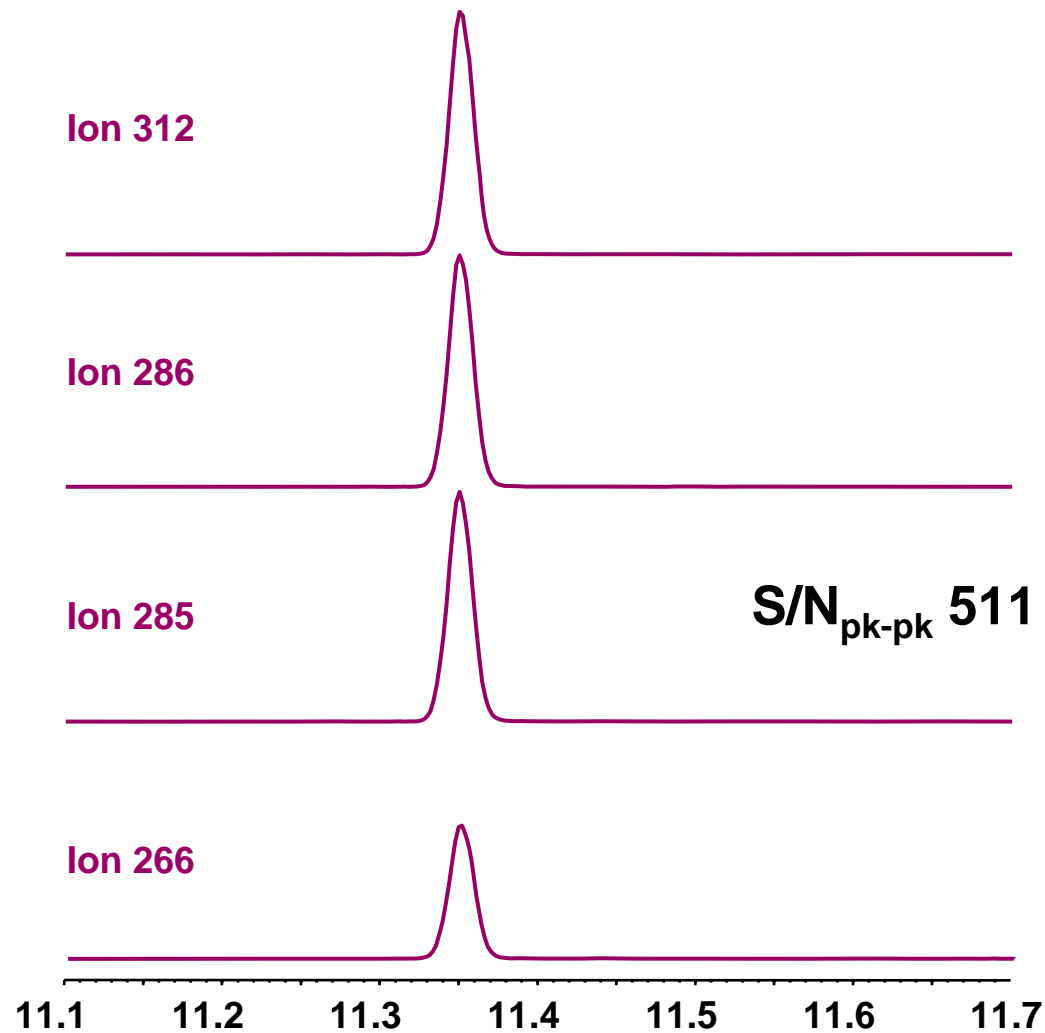
Flunitrazepam- Scan Only Mode

0.63 ng

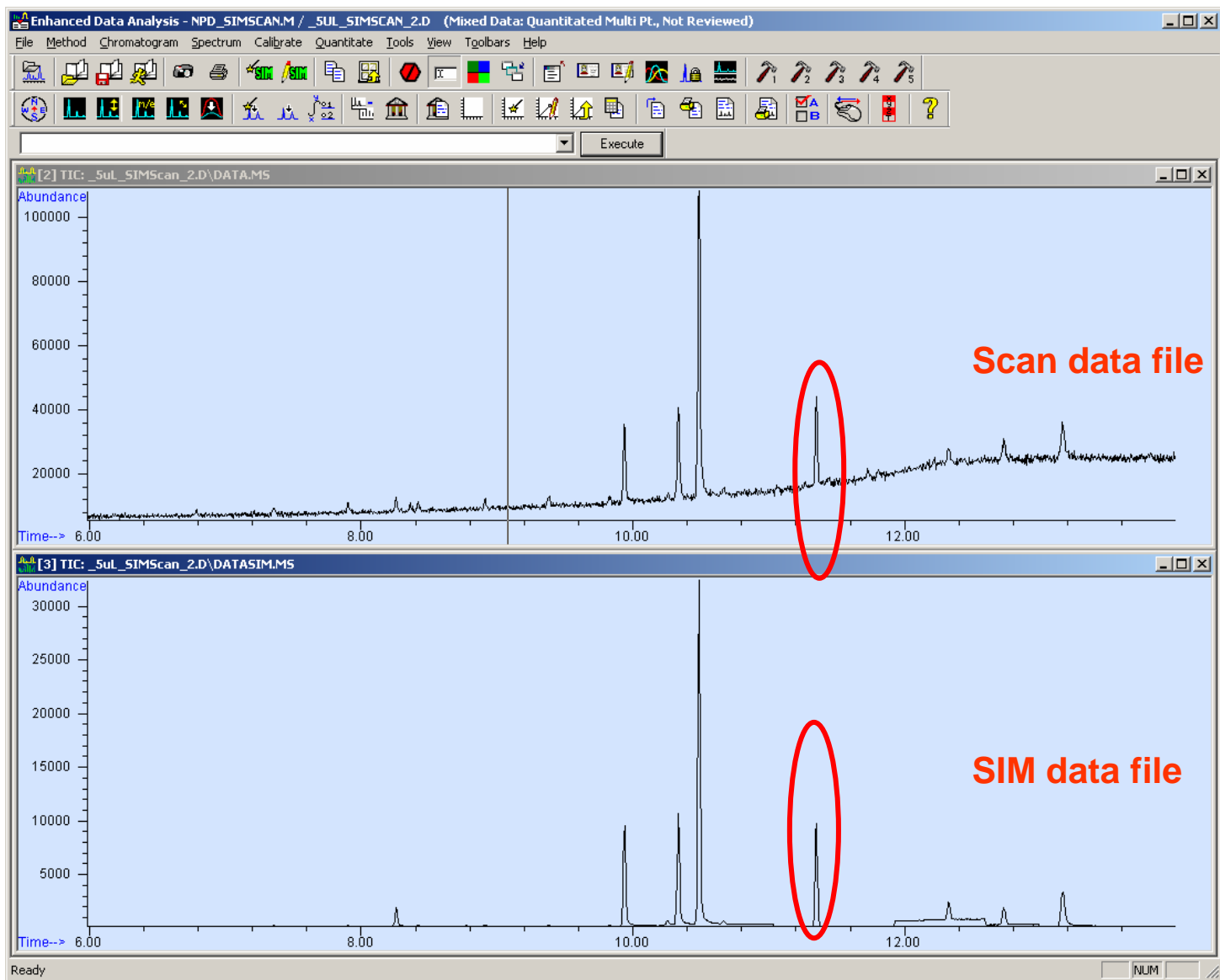


Flunitrazepam- SIM From SIM Only Run

0.63 ng

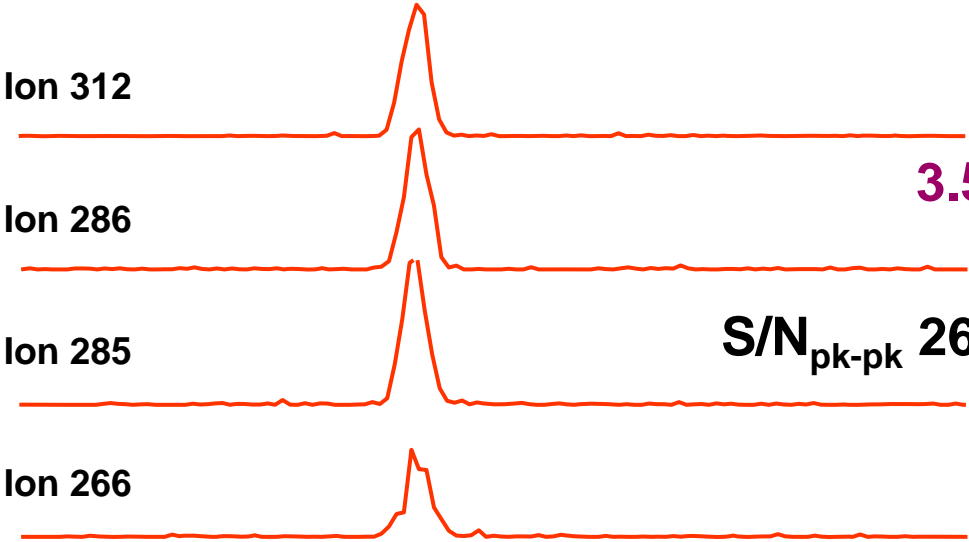


Synchronous Data

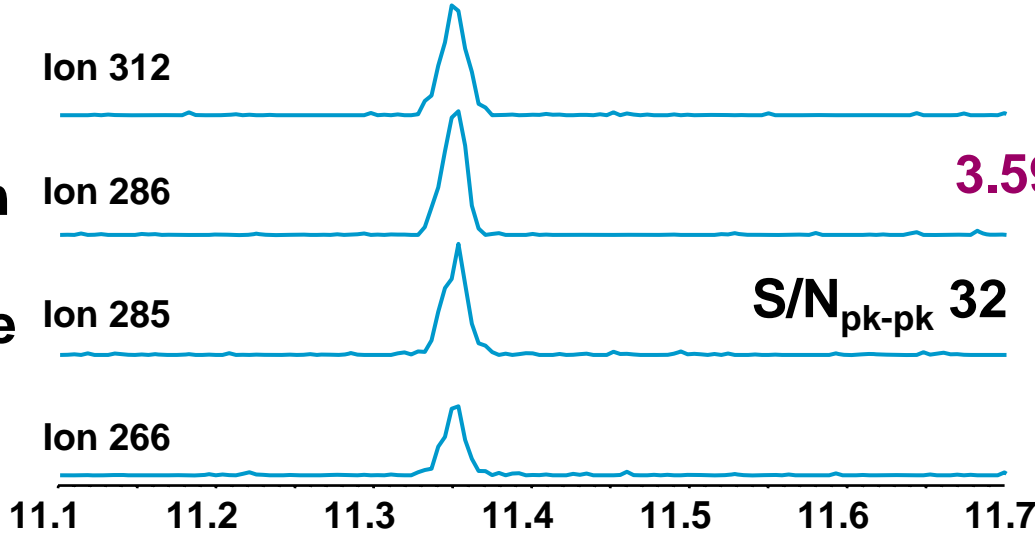


Comparison of Scan Only and Scan from SIM/Scan

Extracted Ions From Scan Mode

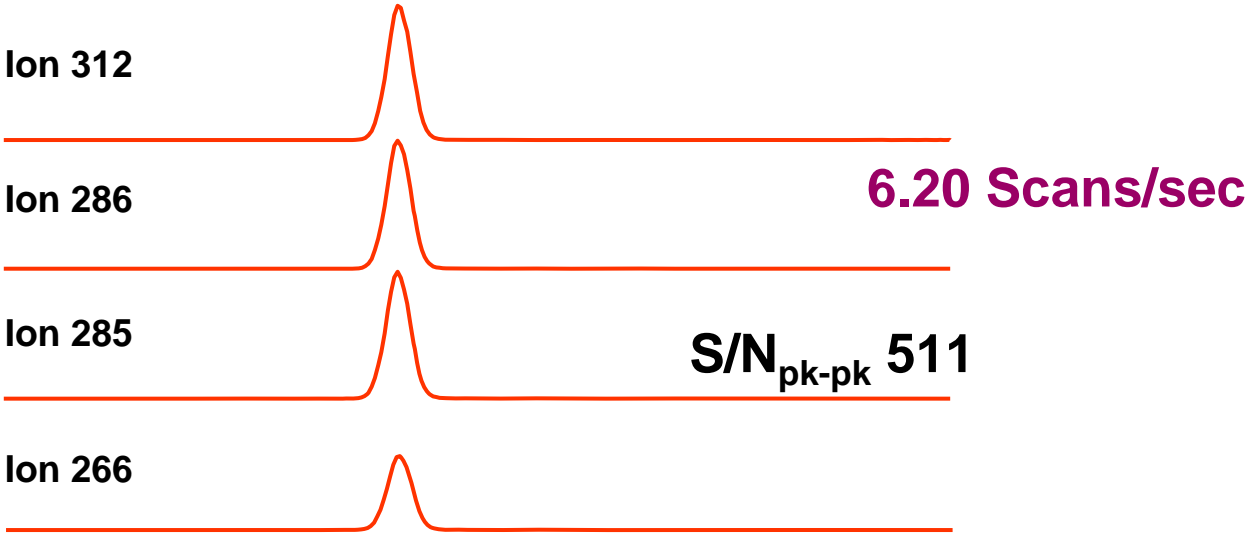


Extracted Scan Ions From SIM/Scan Mode

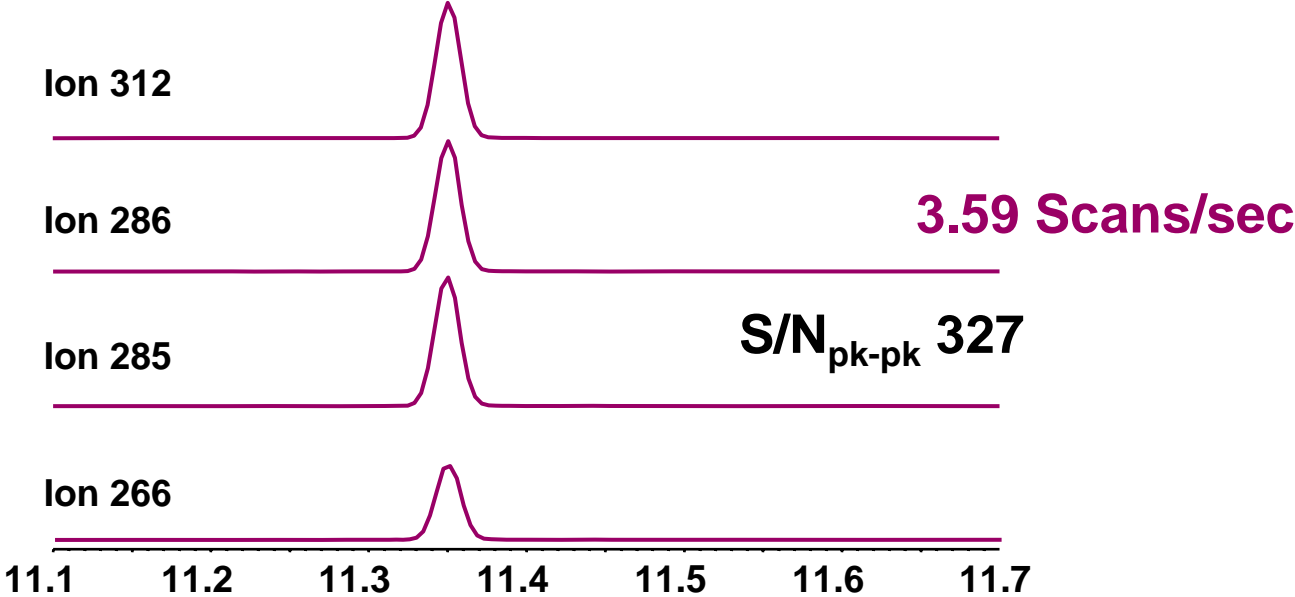


Comparison of SIM Only and SIM from SIM/Scan

Extracted SIM Ions From Scan Mode
25 msec dwell

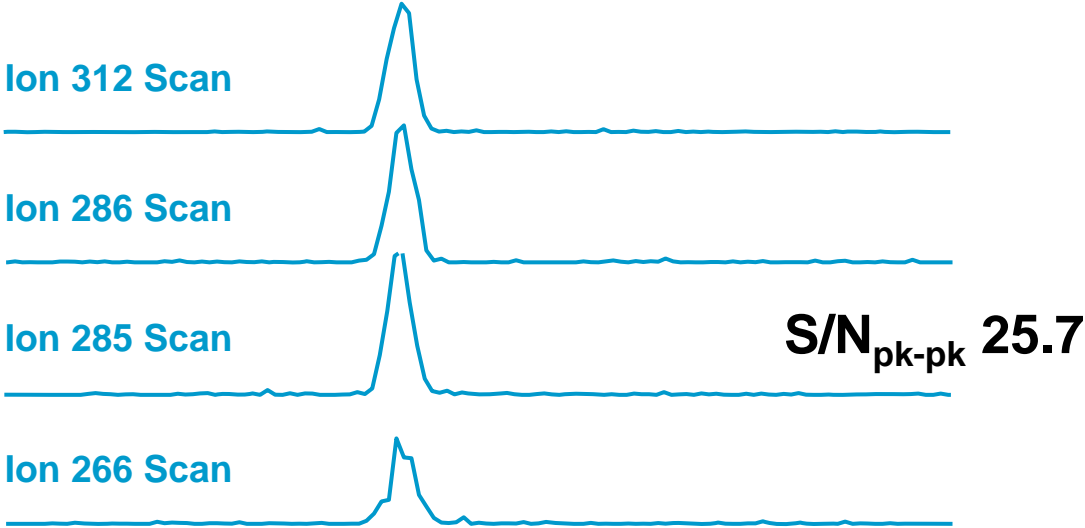


Extracted SIM Ions From SIM/Scan Mode
25 msec dwell

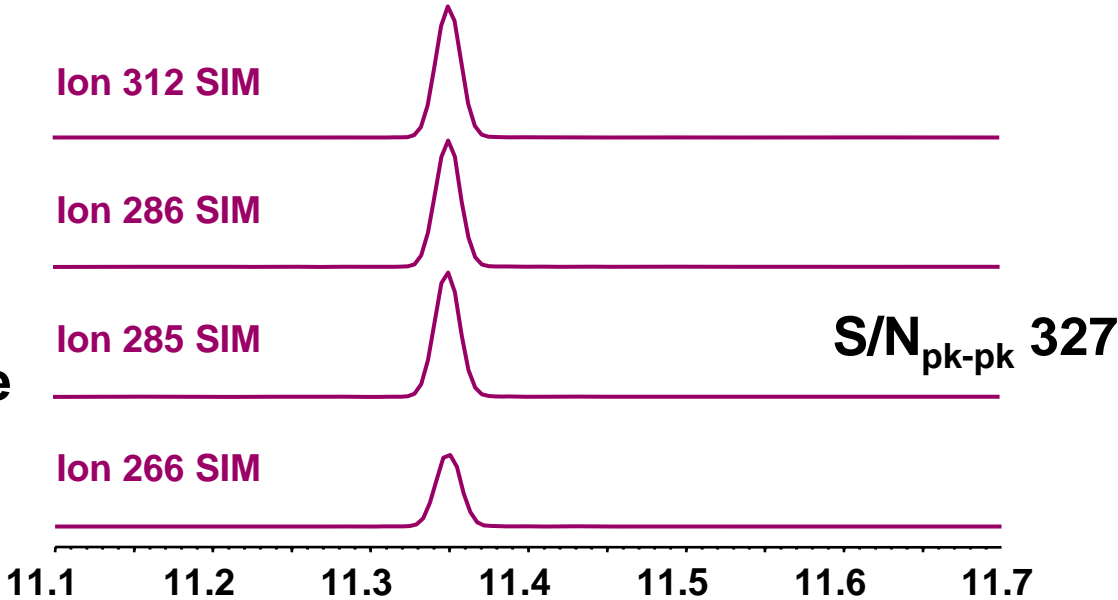


Flunitrazepam-Advantage for Quant with SIM/Scan 0.63 ng

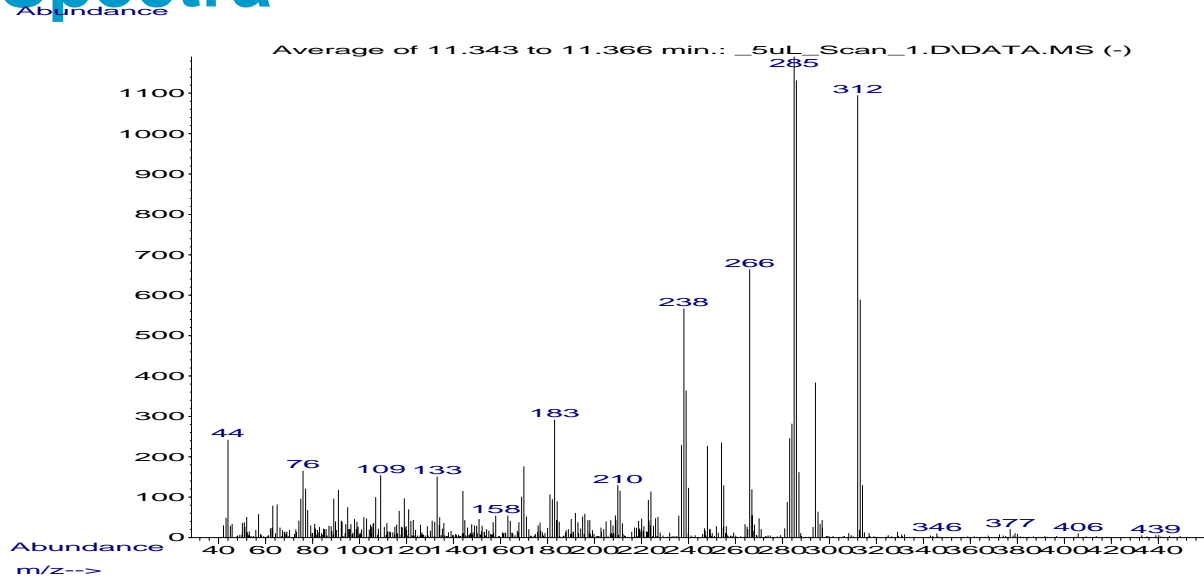
Extracted Ions
From Scan
Mode



Extracted SIM
Ions From
SIM/Scan Mode



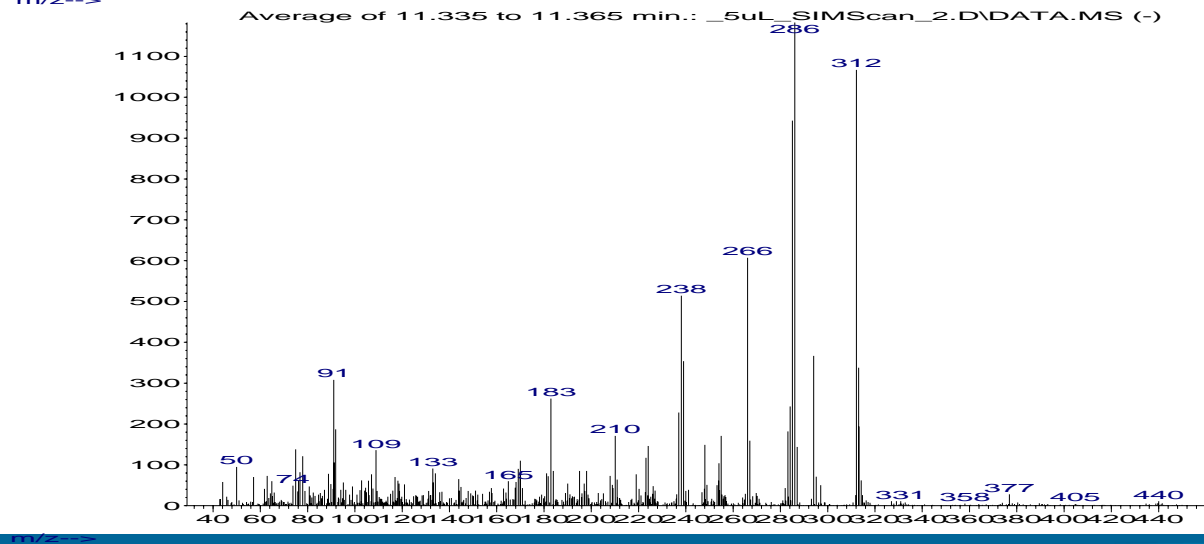
Synchronous SIM/Scan – Library Searchable Spectra



Flunitrazepam

Scan Only Spectra

**NIST Forward
Match 837**



**Spectra from
SIM/Scan**

**NIST Forward
Match 826**

Signal to Noise Improvement with SIM/Scan

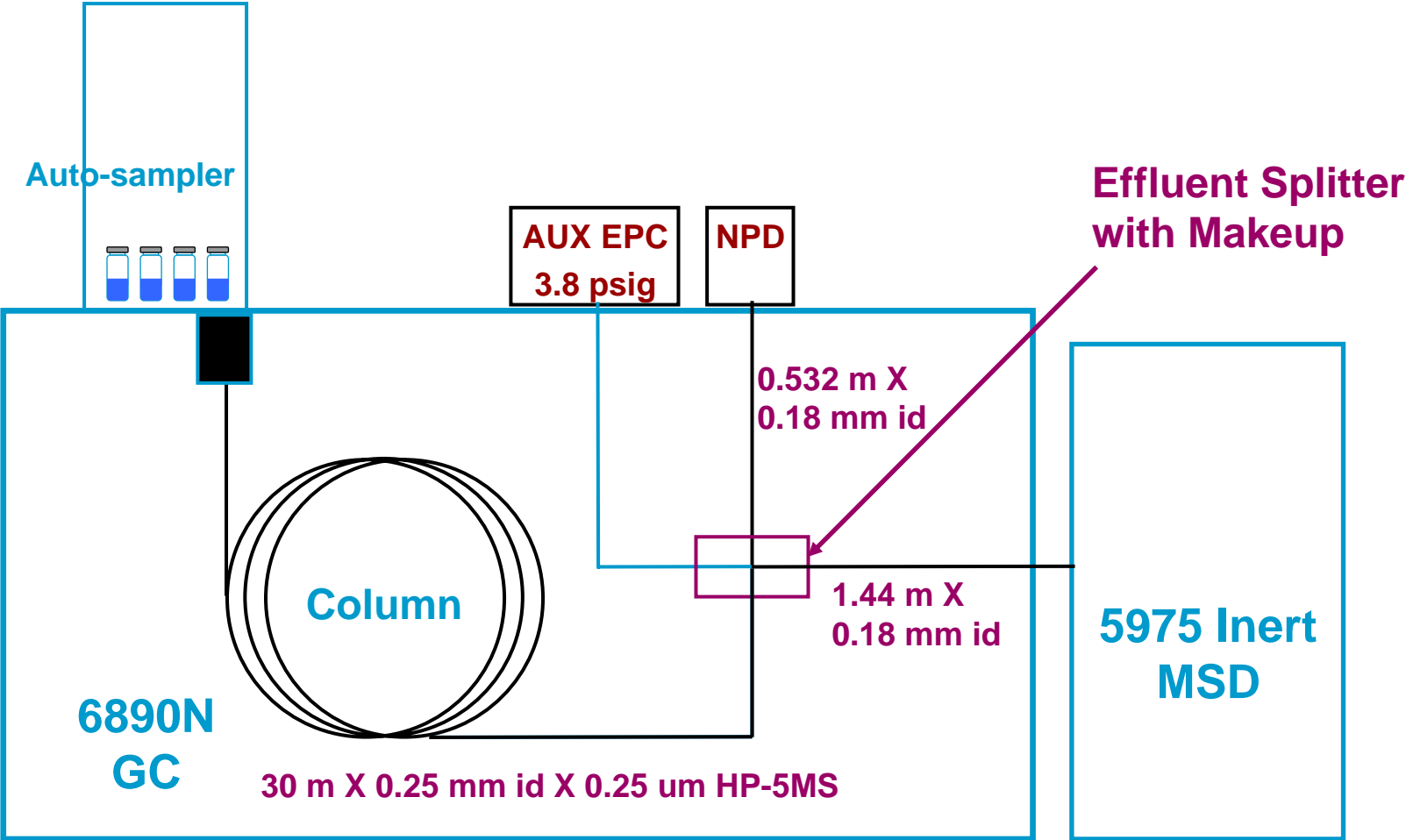
Ret Time	Compound	Tgt Ion	A	B	C	D	D/A	# ions in SIM
			Scan Only S/N _{pk-pk}	SIM/Scan Scan S/N _{pk-pk}	SIM only S/N _{pk-pk}	SIM/Scan SIM S/N _{pk-pk}	Improvement factor	
9.940	Oxazepam	267	43	49	687	676	16	4
10.338	Lorazepam	239	87	60	787	1098	13	7
10.490	Diazepam	256	242	388	2002	2638	11	7
11.353	Flunitazepam	285	50	42	461	597	12	4
12.327	Nitrazepam	280	6	12	115	116	18	8
12.733	Clonazepam	280	8	11	128	123	15	4
13.168	Alprazolam	308	18	24	173	192	11	4

avg 13.6

SIM and Scan performance of SIM/Scan is comparable to SIM only and Scan only modes

Permanent Splitter System GC/NPD/MSD

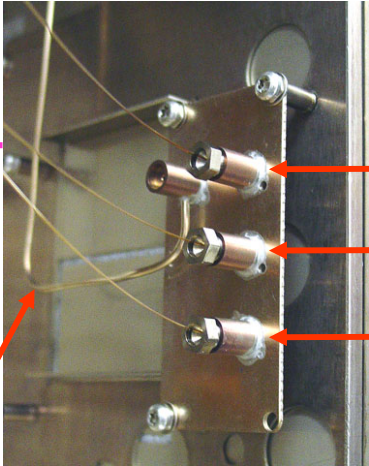
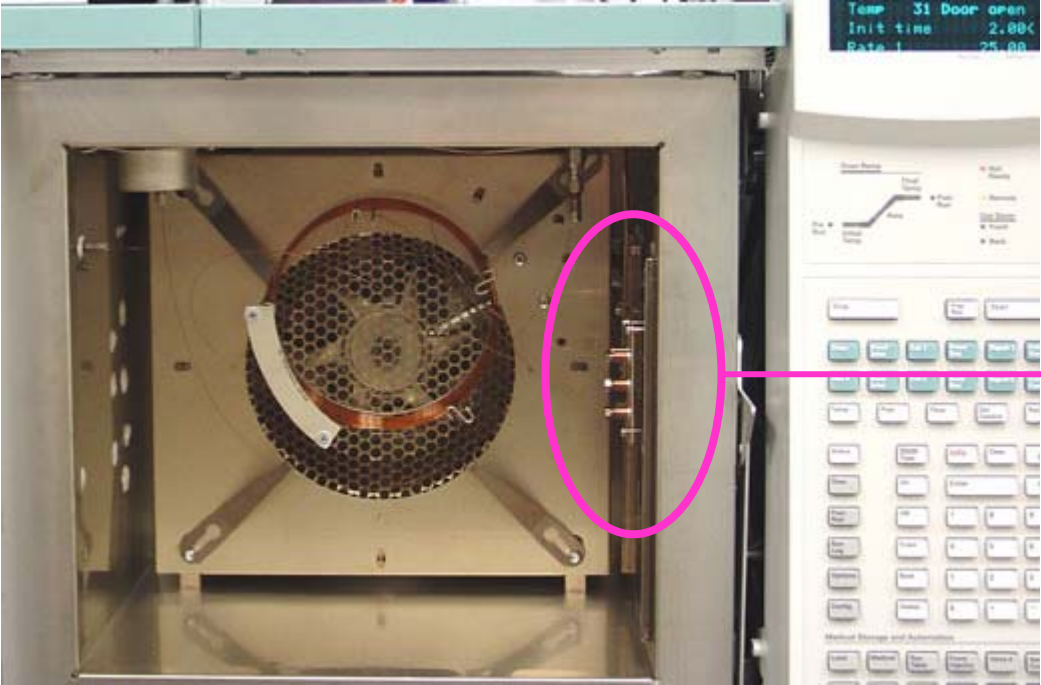
1:1 split NPD:MSD



uFluidic Splitter Hardware

Deactivated microfluidic splitter operates to 350 C.

Uses metal ferrules to eliminate leaks and retightening

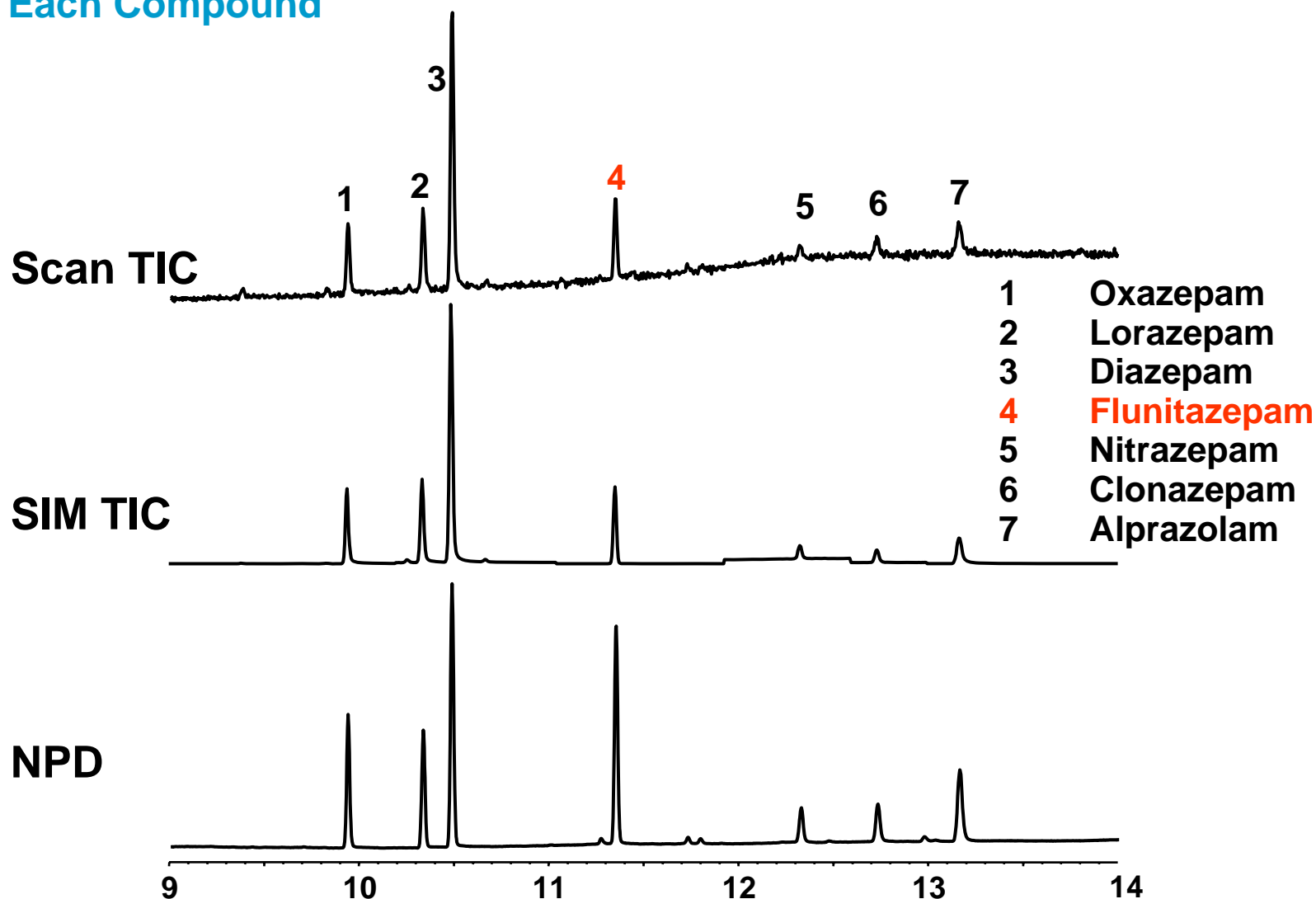


Column in
To NPD
To MSD

Makeup from EPC

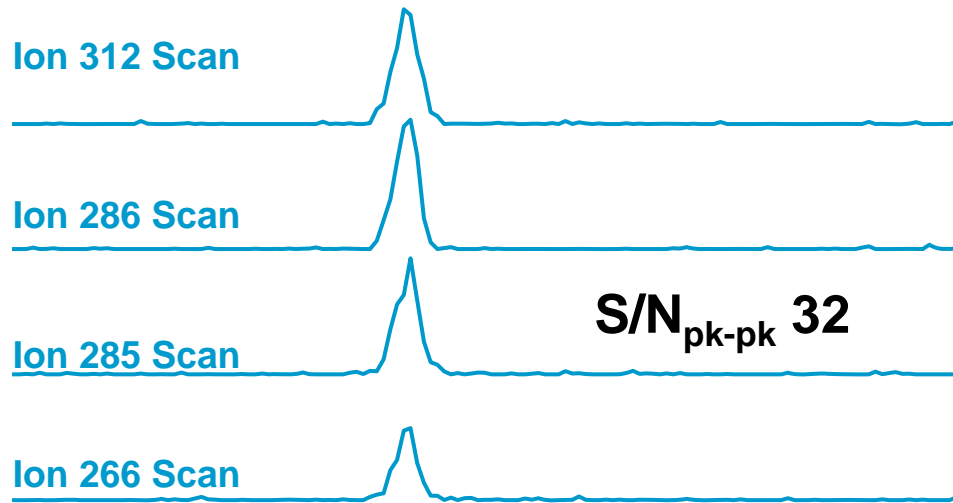
Three Signals from 1 Injection: Scan, SIM, and NPD

1.25 ng Each Compound

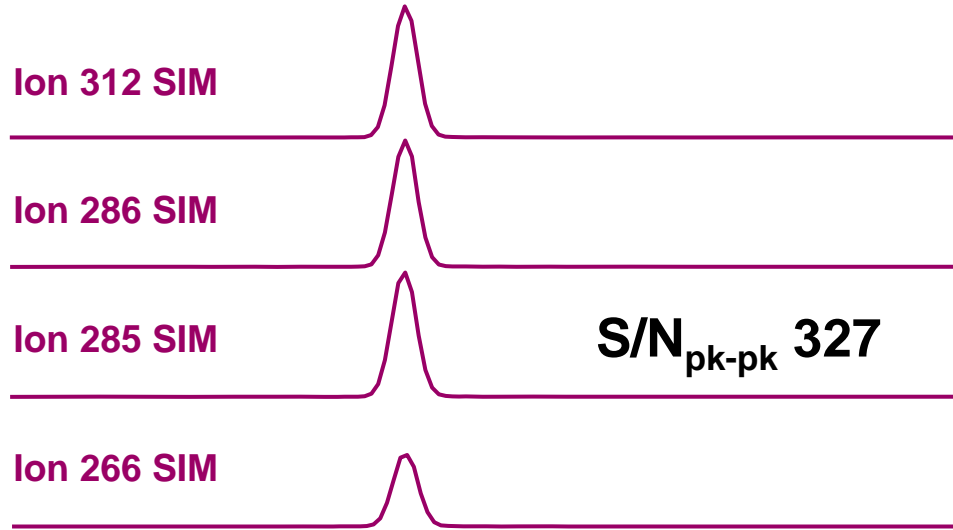


Flunitrazepam

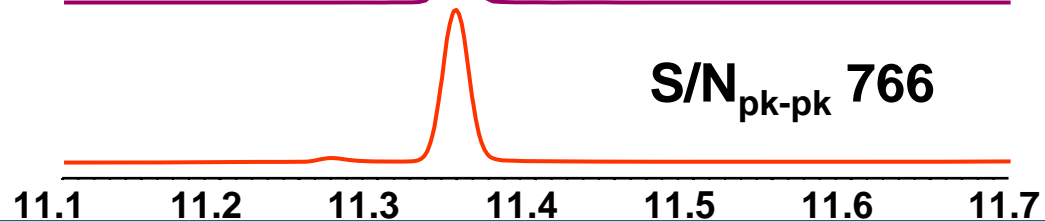
Extracted Ions
From Scan
Mode



Extracted SIM
Ions From
SIM/Scan Mode



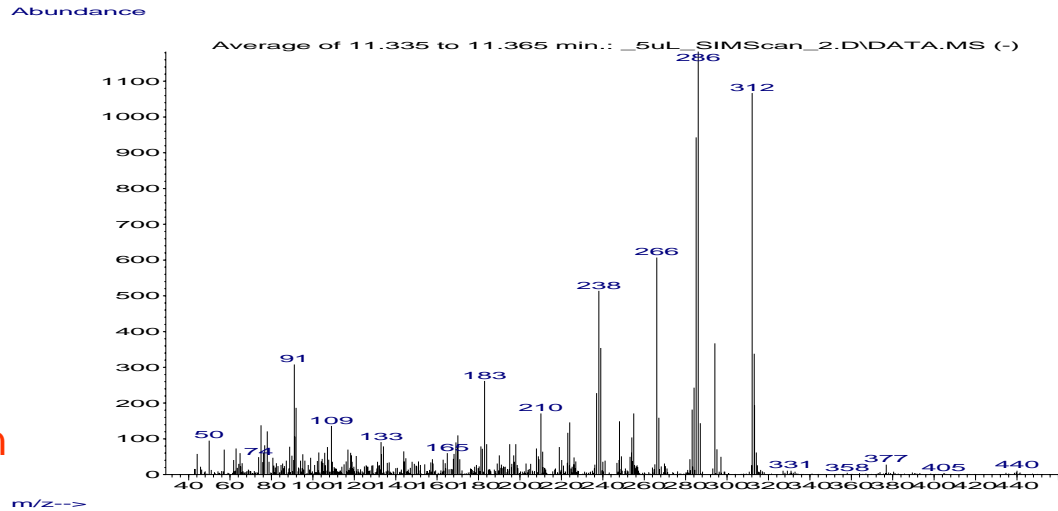
NPD



Flunitrazepam 1.25 ng

Scan from
SIM/Scan

Use for confirmation



Extracted SIM
Ions From
SIM/Scan Mode

Use for quant

Ion 312 SIM

Ion 286 SIM

Ion 285 SIM

Ion 266 SIM

S/N_{pk-pk} 327

S/N_{pk-pk} 766

NPD

Use for confirmation
and/or quant

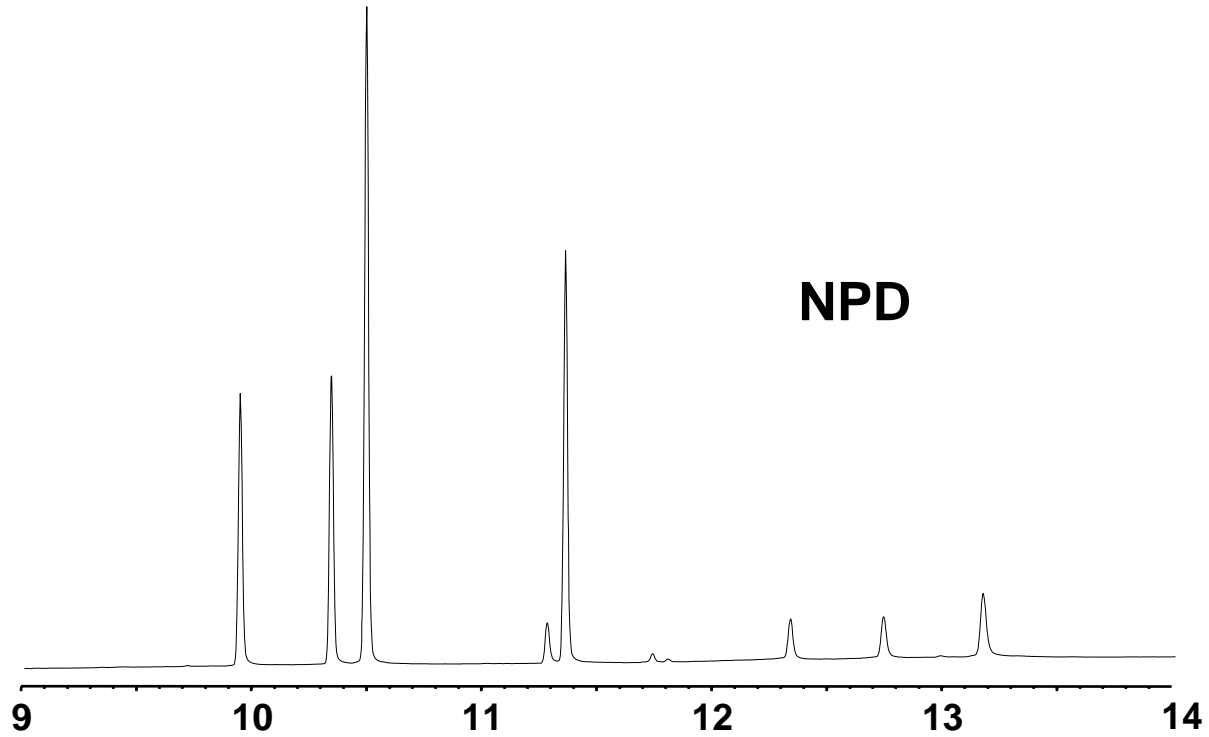
11.1 11.2 11.3 11.4 11.5 11.6 11.7



Agilent Technologies

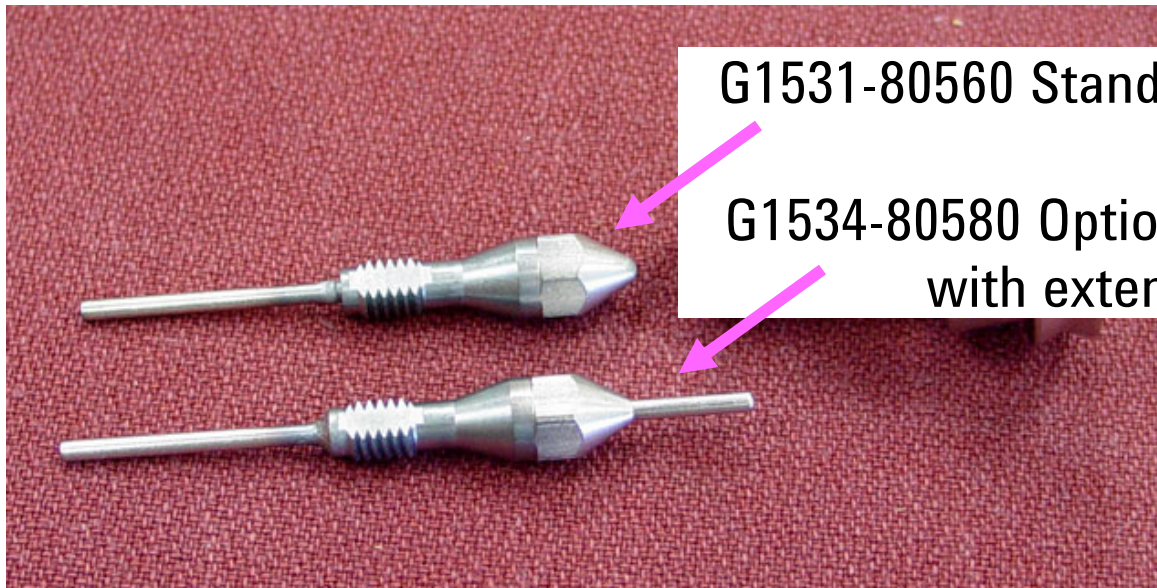
For Research Use Only. Not
for use in diagnostic
procedures.

Why Does My NPD Peakshape Look So Good?



NPD Jets, Standard vs Extended Tip

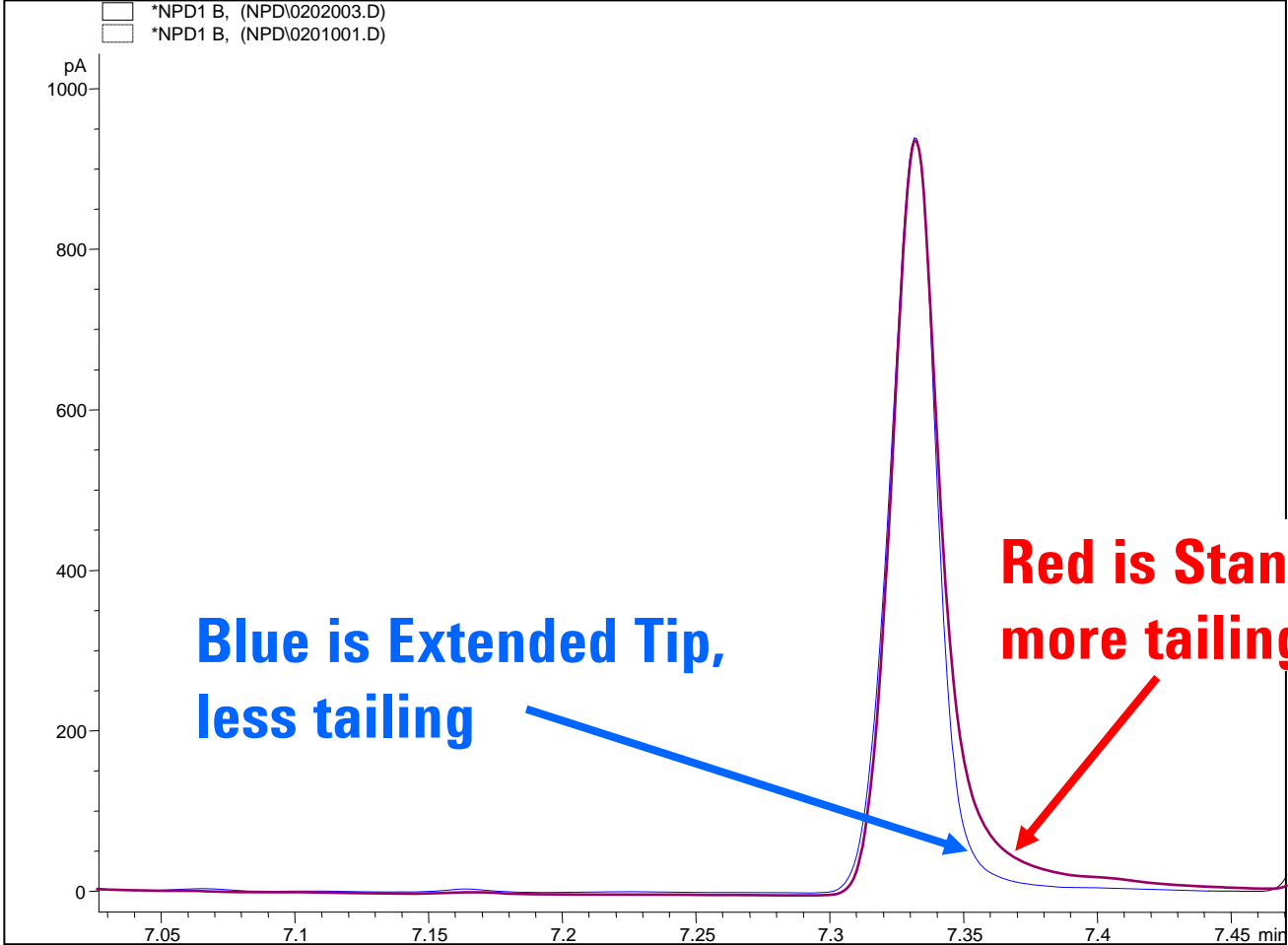
**Extended Tip extends ~ 9mm
Analytes are positioned closer to
the bead, minimizing tailing.**



G1531-80560 Standard Capillary Jet

G1534-80580 Optional Capillary Jet
with extended tip

Drug Standard by NPD using Different Jets



NPD Jet For Improved Nitrogen on 6890

There is a much better jet for nitrogen detection with the NPD:

- G1534-80580 NPD Jet Capillary Det
- G1534-80590 NPD Jet Universal Det
- G1530A-29 Service Note that describes best practices for optimal NPD operation
- [note: disregard suggested use of narrower collector]

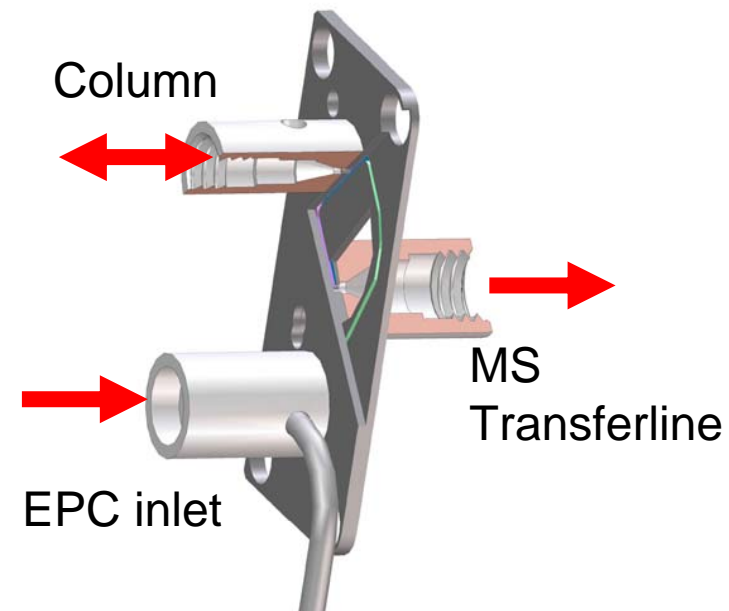
QuickSwap MSD Interface – Ease of Use

Remove column w/o venting

Backflush mode

- Inlet maintenance reverse flow
- Removes heavies from column

Maintain constant flow to MSD

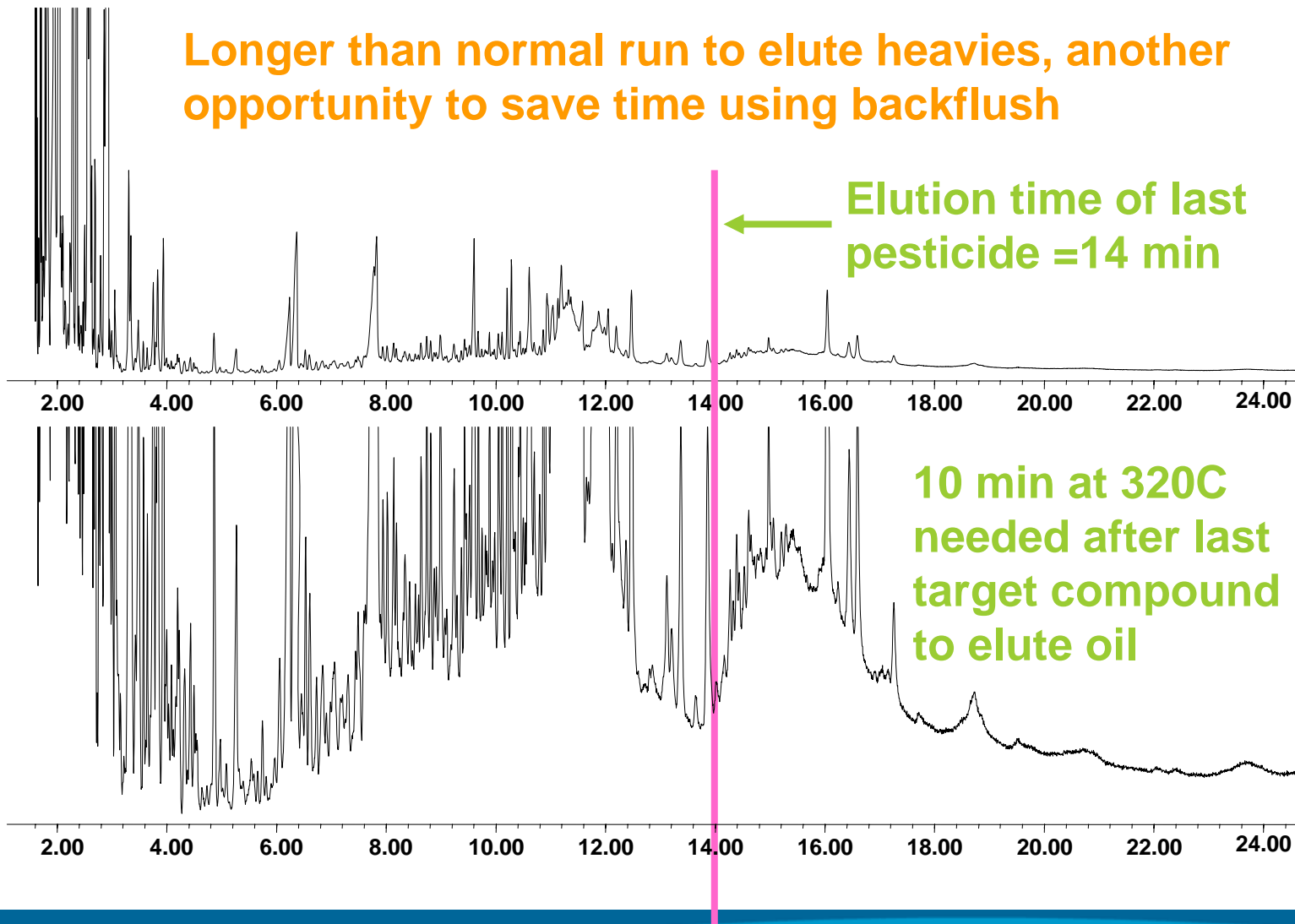


Microfluidic technology

Requires performance turbo for flow rates > 2 cc/min

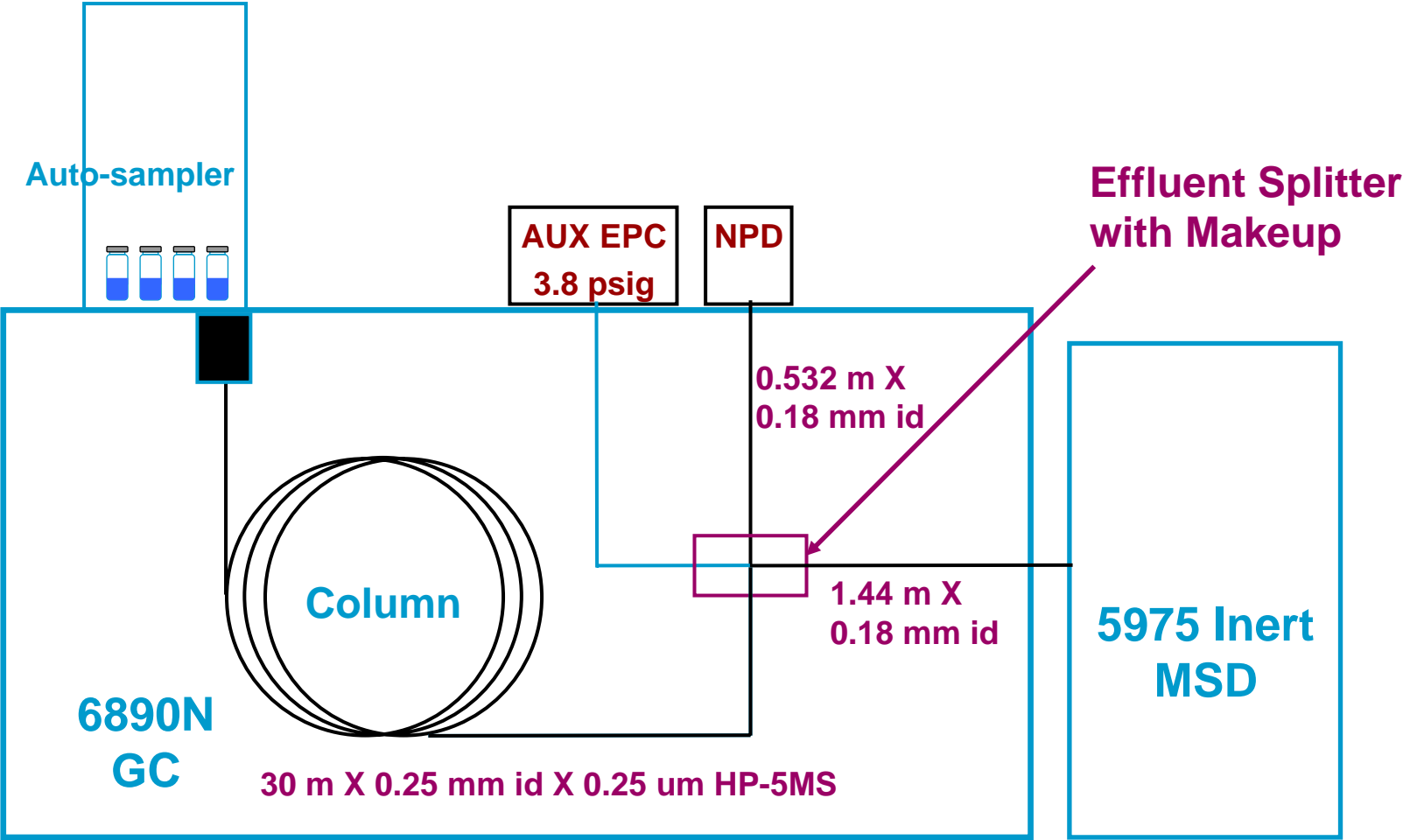
3x Speed, 15m, Lemon Oil TIC, no spike

Longer than normal run to elute heavies, another opportunity to save time using backflush



Permanent Splitter System GC/NPD/MSD

1:1 split NPD:MSD



How Do You Backflush ?

Program inlet pressure to ~1.0 psi (may have to fine tune)

Program make-up pressure to calculated value after last target peak elution time

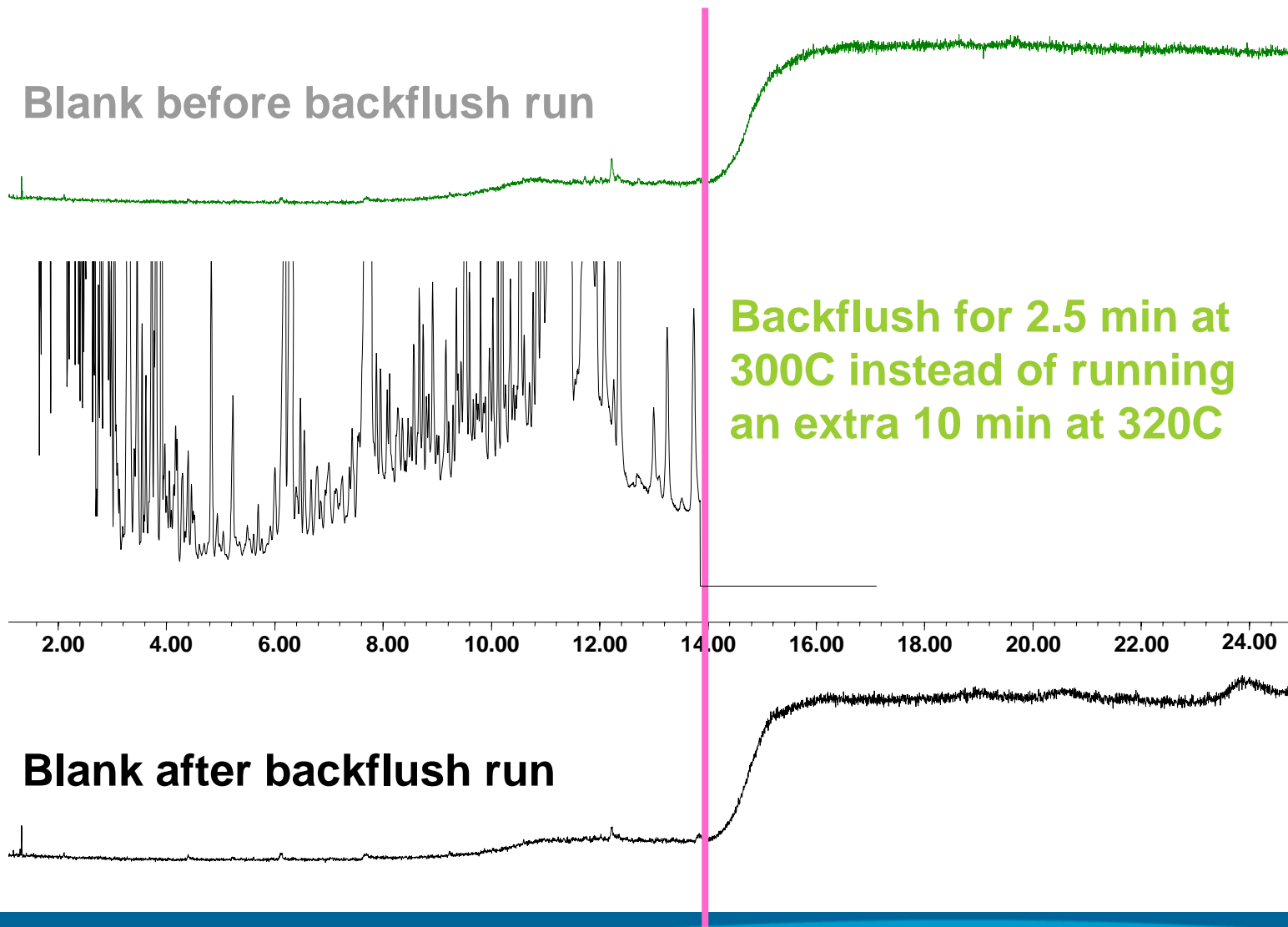
- Use flow calculator to get pressure for make-up to splitter (aux EPC or PCM). MS restrictor flow < 10 mL/min at backflush temp

Try a 3 void-times backflush, then run a blank for an extended time to make sure system is clean

Shorten this time if blank is clean, in 0.5 min steps

Performance Turbo a must

3x Speed, 15m, Lemon Oil TIC, no spike

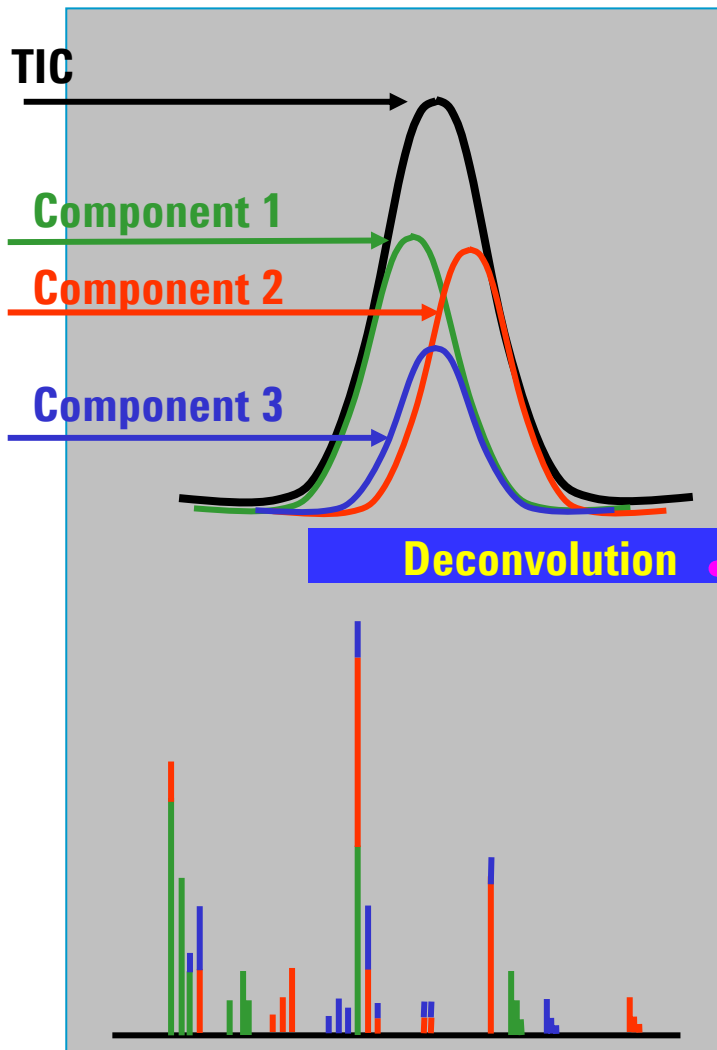


Spectral Deconvolution

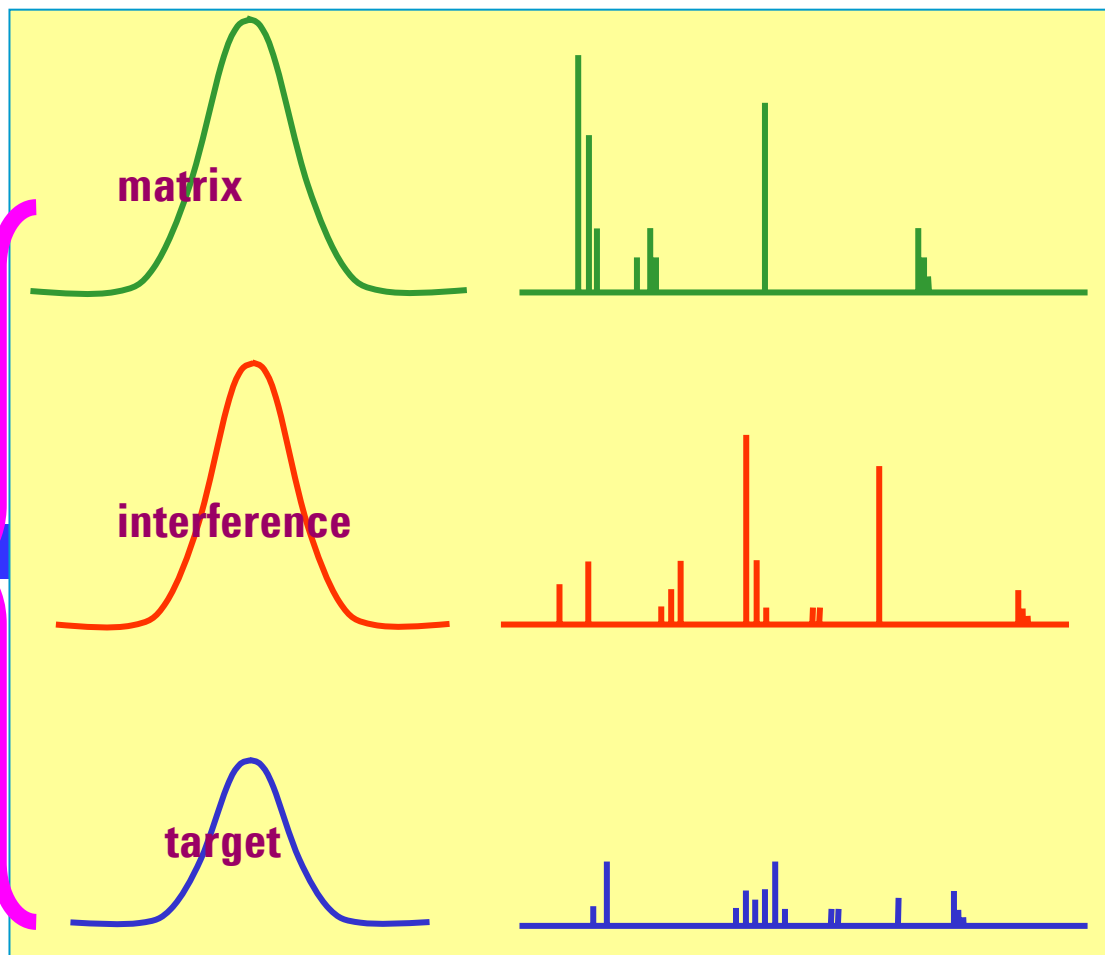
- Extracts cleaner mass spectra from overlap situations – easier, reliable identification
- Faster data review, a free second opinion from a mass spec expert
- When coupled with RTL and NPD data, gives higher quality identifications
- Can run searches of data not restricted by RT to find “spectral brethren” like designer drugs

AMDIS Deconvolution Pulls Out Individual Components and their Spectra

TIC & Spectrum

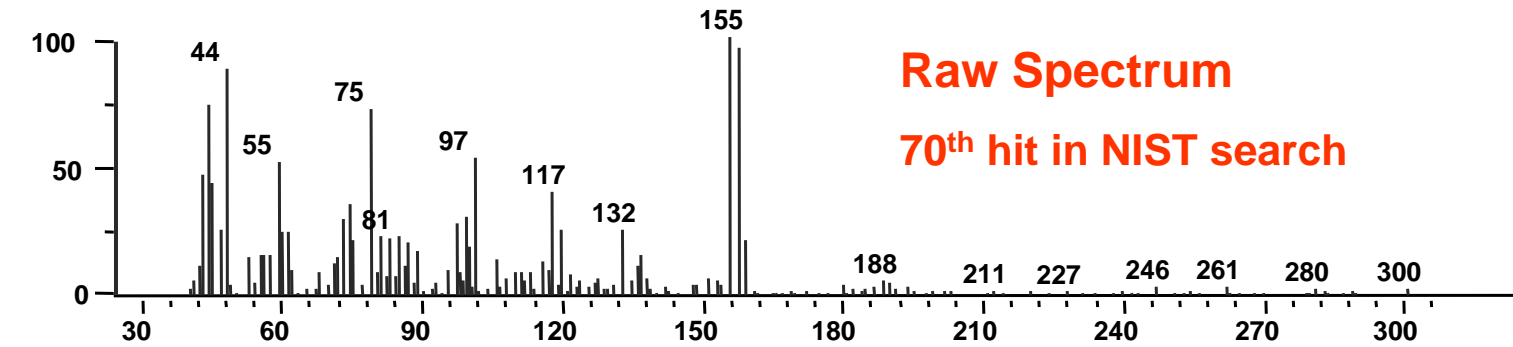


Deconvoluted peaks and spectra



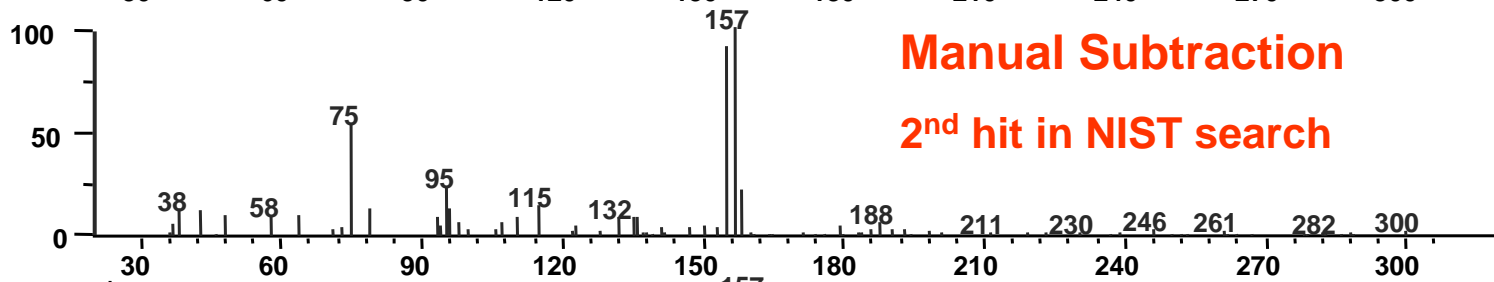
Deconvolution Removes Spectral Interferences

3-Chloro-1,2-dibromopropane overlapped with fuel components



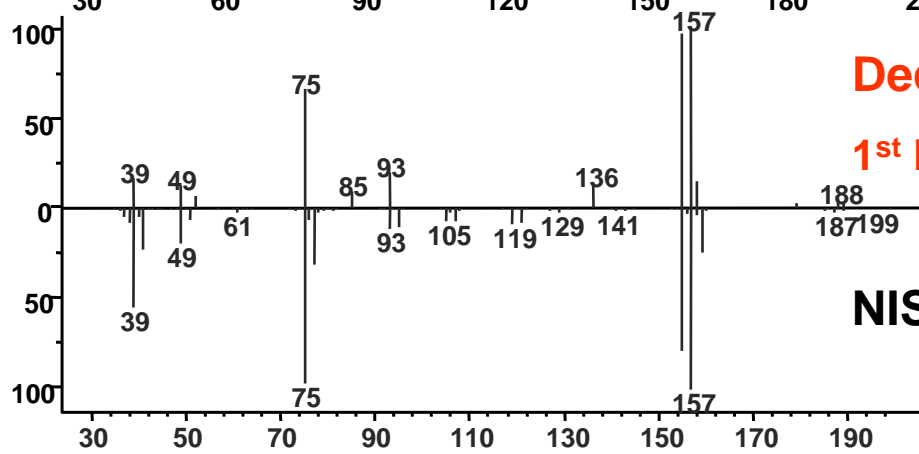
Raw Spectrum

70th hit in NIST search



Manual Subtraction

2nd hit in NIST search



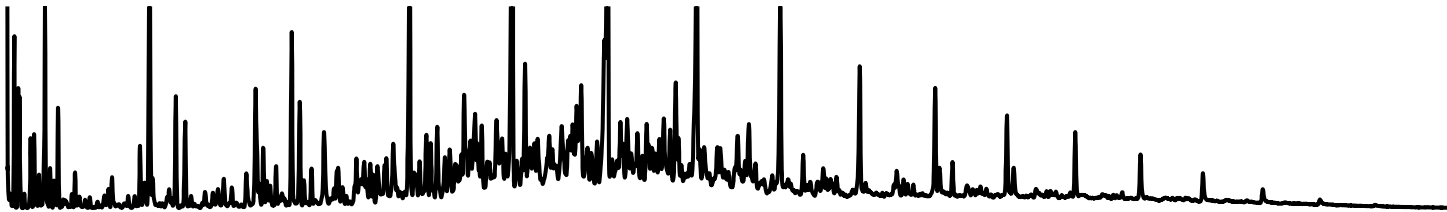
Deconvolved Spectrum

1st hit in NIST search

NIST 05 Library

DRS: 3 Integrated Processes

Total ion chromatogram



Targets are identified by locked R.T.s and 3 qualifier ions, then quantified using target ion area vs ISTD cal table

Quant Results

AMDIS 32 deconvolutes spectra and searches target MS database using locked RT as a qualifier

Confirmed AMDIS hits

Deconvoluted Target spectra confirmed by AMDIS, searched against NIST02 MS database

Confirmed NIST05 hits

Combined quantitative and qualitative HTML Summary report

DRS Report

MSD Deconvolution Report

Sample Name: 2 ppmVOA MIX 3 Only

Data File: C:\msdchem\1\DATA\042205AMHS_SimScan\2ppmMIX 3_Only_simscan.D

Date/Time: 05:13:03 PM Friday, Dec 9 2005

The NIST library was searched for the components that were found in the AMDIS target library.

R.T.	Cas #	Compound Name	Agilent	AMDIS		NIST	
			ChemStation Amount (ng)	Match	R.T. Diff sec.	Reverse Match	Hit Num.
1.497	107062	1,2-Dichloroethane	2.27	97	0.6	94	1
1.540	563586	1,1-Dichloropropylene	7.6	100	0.5	96	1
1.867	78875	1,2-Dichloropropane	4.92	95	0.7	90	1
1.871	79016	Trichloroethylene	7.58	99	0.6	91	1
2.330	10061015	cis-1,3-Dichloropropylene	4.39	98	1.0	92	2
2.677	10061026	trans-1,3-Dichloropropylene	3.3	97	1.5	94	1
2.758	79005	1,1,2-Trichloroethane	2.82	99	1.7	92	1
2.961	142289	1,3-Dichloropropane	3.39	98	1.5	92	1
3.273	106934	1,2-Dibromoethane	2.6	91	1.5	76	4
4.032	630206	1,1,1,2-Tetrachloroethane	5.15	100	1.9	94	1
5.187	79345	1,1,2,2-Tetrachloroethane	2.38	99	2.3	89	1
5.322	96184	1,2,3-Trichloropropane	1.89	98	2.5	94	1
6.323	76017	pentachloroethane	0.08	63	2.0	76	1
8.232	96128	3-Chloro-1,2-dibromopropane	1.62	93	1.6	87	1
10.439	87683	hexachlorobutadiene	16.46	94	0.9	95	1



Summary

The 5975 and associated new products bring several new tools to the forensic analyst:

- Auto-CI
- SIMScan
- Splitters
- DRS