From Trend to Discovery: Temporal Analysis of Contaminants in Watersheds Using High Resolution Mass Spectrometry

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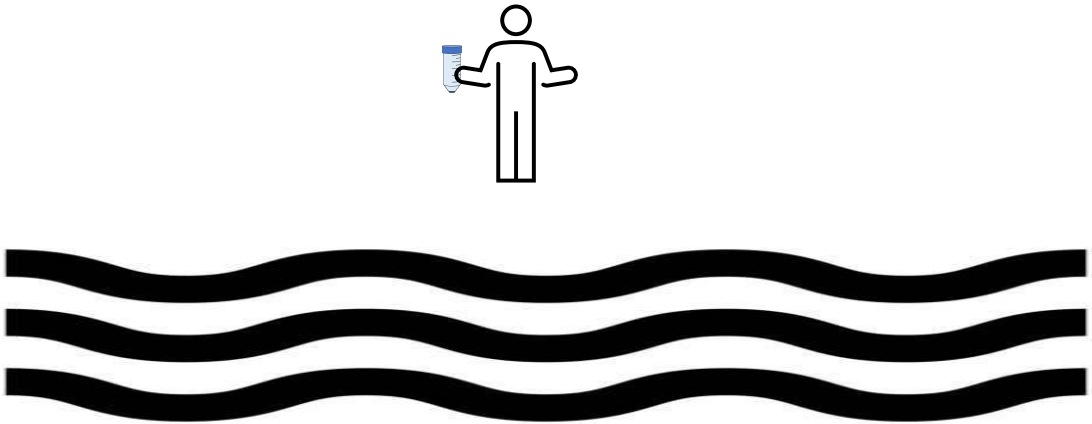
SCIEX
The Power of Precision

Overview

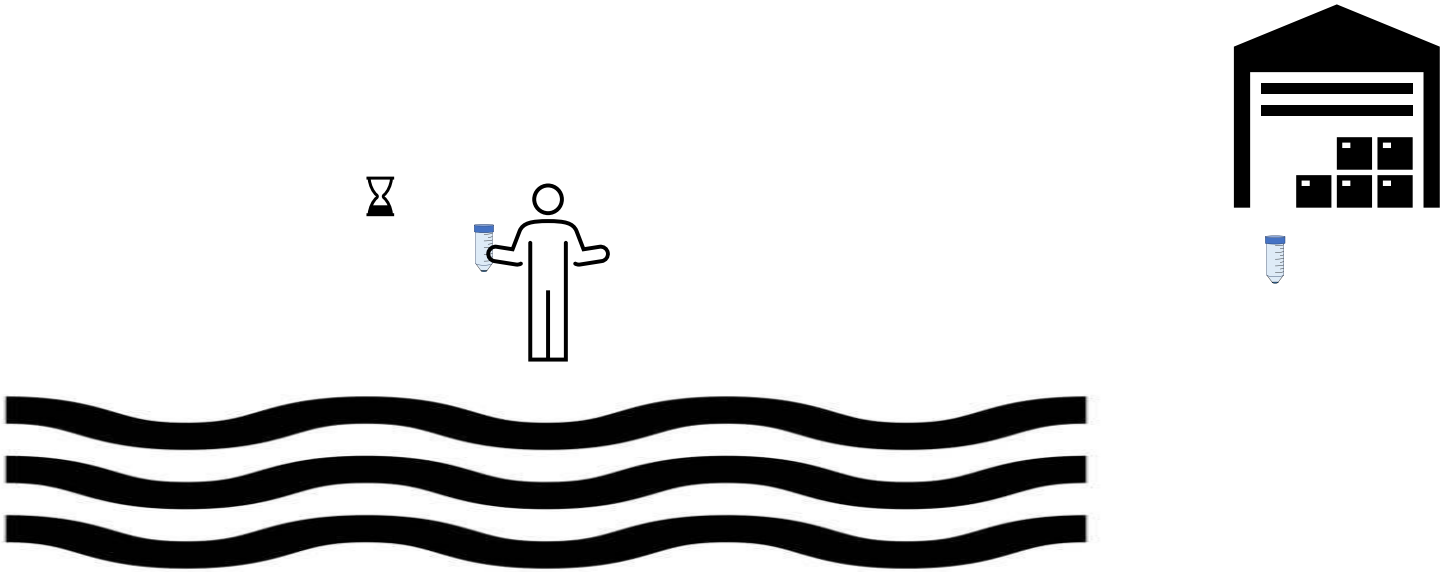
- Temporal monitoring
  - What is it?
  - How has it been used previously?
  - Rhine River research
- High resolution mass spectrometry for non-targeted analysis
- Our own monitoring study
  - Watershed
  - Sample collection
  - HRMS Analysis
  - Data Analysis
    - enviMass
    - ChemSpider
  - Trends and compounds discovered

Spatio-Temporal Monitoring

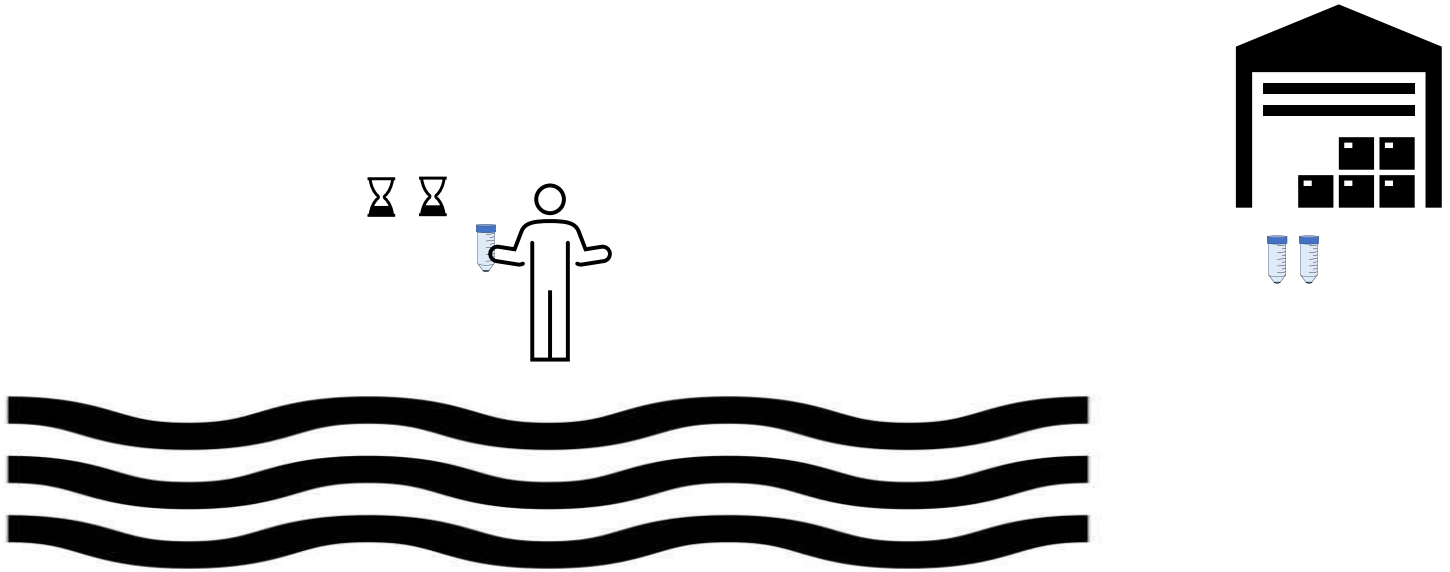
Spatio-Temporal Monitoring



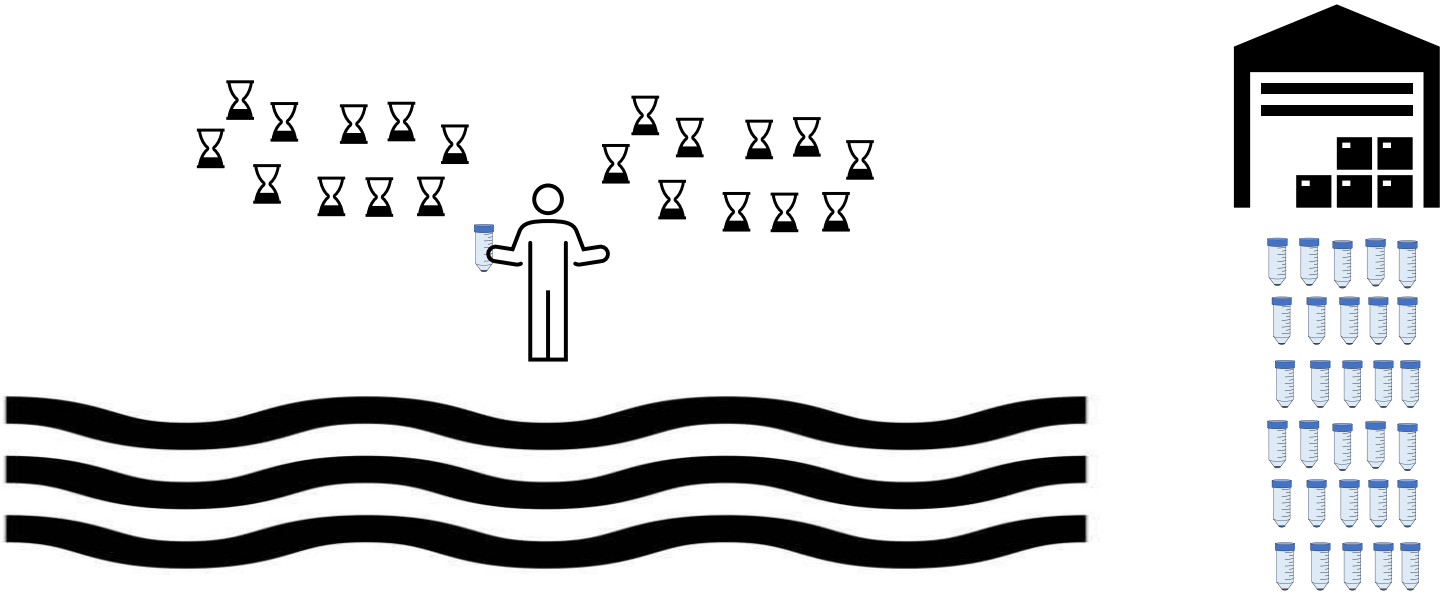
Spatio-Temporal Monitoring



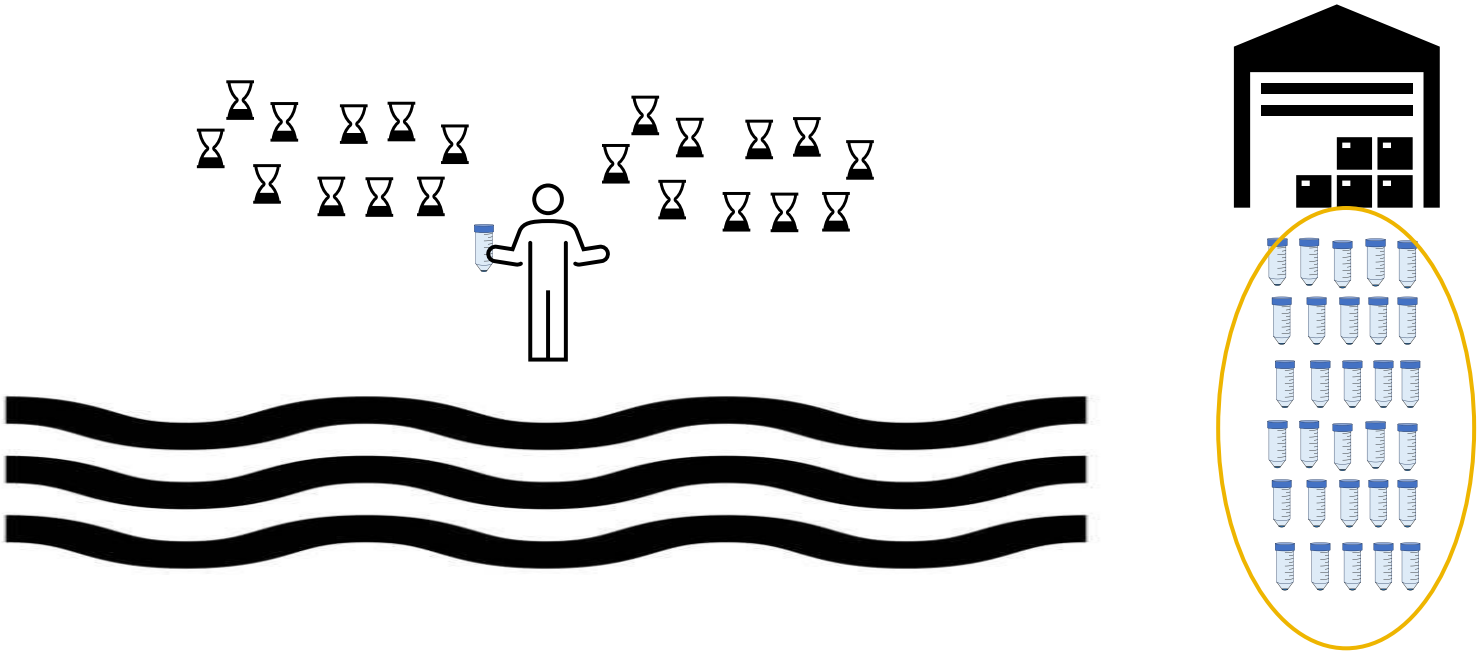
Spatio-Temporal Monitoring



Spatio-Temporal Monitoring



Spatio-Temporal Monitoring



Prepared for analysis
and data processing

Rhine River

- Water sampled across various locations along this river used for drinking water across Europe
- LC-HRMS performed for data analysis, spiking with IS and analyzing a mixture of standards so quantitation and identification of unknowns can be performed
- enviMass used for prioritization and trend discovery
- Locations of identified compounds could be traced to industrial sources in those areas

Water Research 87 (2015) 145–154

Contents lists available at ScienceDirect

Water Research

journal homepage: www.elsevier.com/locate/watres

Quantitative target and systematic non-target analysis of polar organic micro-pollutants along the river Rhine using high-resolution mass-spectrometry – Identification of unknown sources and compounds

Matthias Ruff, Miriam S. Mueller [1], Martin Loos, Heinz P. Singer*

Eawag, Swiss Federal Institute of Aquatic Science and Technology, 8600 Dubendorf, Switzerland

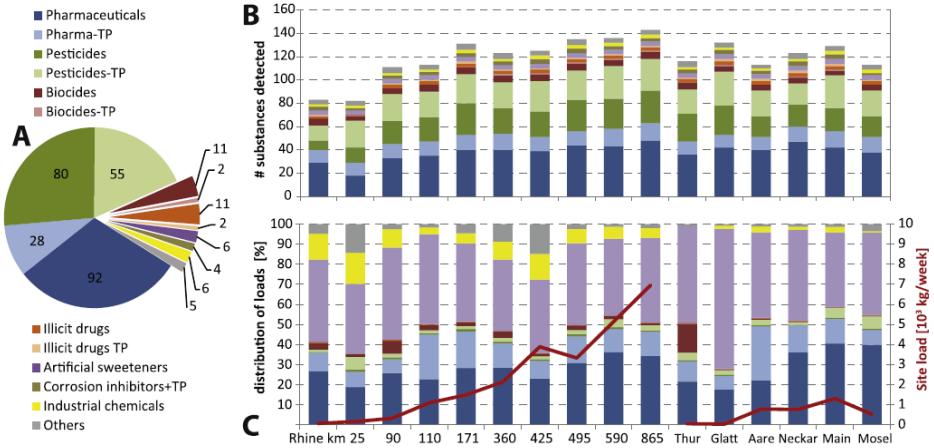


Fig. 2. A – Distribution of the investigated compounds grouped in substance classes; B – Number of substances detected at the sampling sites; C – Load distribution of the substance classes at each sampling site (left y-axis) and cumulative load of all quantifiable substances at each site (red curve on right y-axis). The color code of all graphs corresponds to legend 2A.

Lek River Study

- The Lek River is a natural drinking water source and a branch of the Rhine River in The Netherlands
- Sampling of riverbank filtrate from a 60-year travel time
- Non-targeted MS analysis revealed trends over time maximized in the 1990s with increases in human activity, and wastewater treatment plant upgrades
- enviMass used for prioritization and ChemSpider utilized for structural ID

Environmental Science & Technology

Article

Cite This: Environ. Sci. Technol. 2019, 53, 7584−7594

pubs.acs.org/est

Nontarget Screening Reveals Time Trends of Polar Micropollutants in a Riverbank Filtration System

Vittorio Albergamo,*[,†] Jennifer E. Schollée,[‡] Emma L. Schymanski,[‡,§] Rick Helmus,[†] Harrie Timmer,[∥] Juliane Hollender,[‡,⊥] and Pim de Voogt[†,#]

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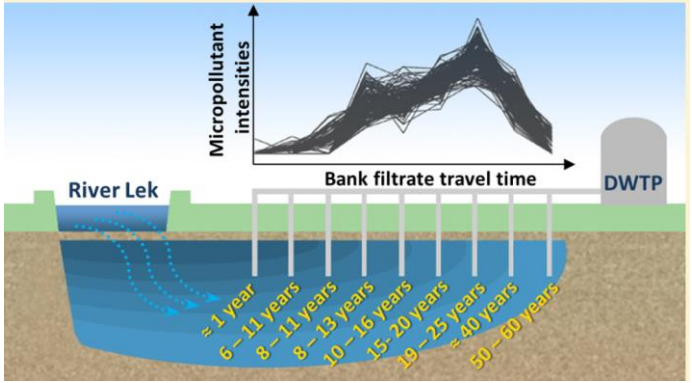
[§]Luxembourg Centre for Systems Biomedicine, University of Luxembourg, House of Biomedicine II 6, avenue du Swing, L-4367 Belvaux, Luxembourg

[∥]Oasen, Nieuwe Gouwe O.Z 3, 2801 SB Gouda, The Netherlands

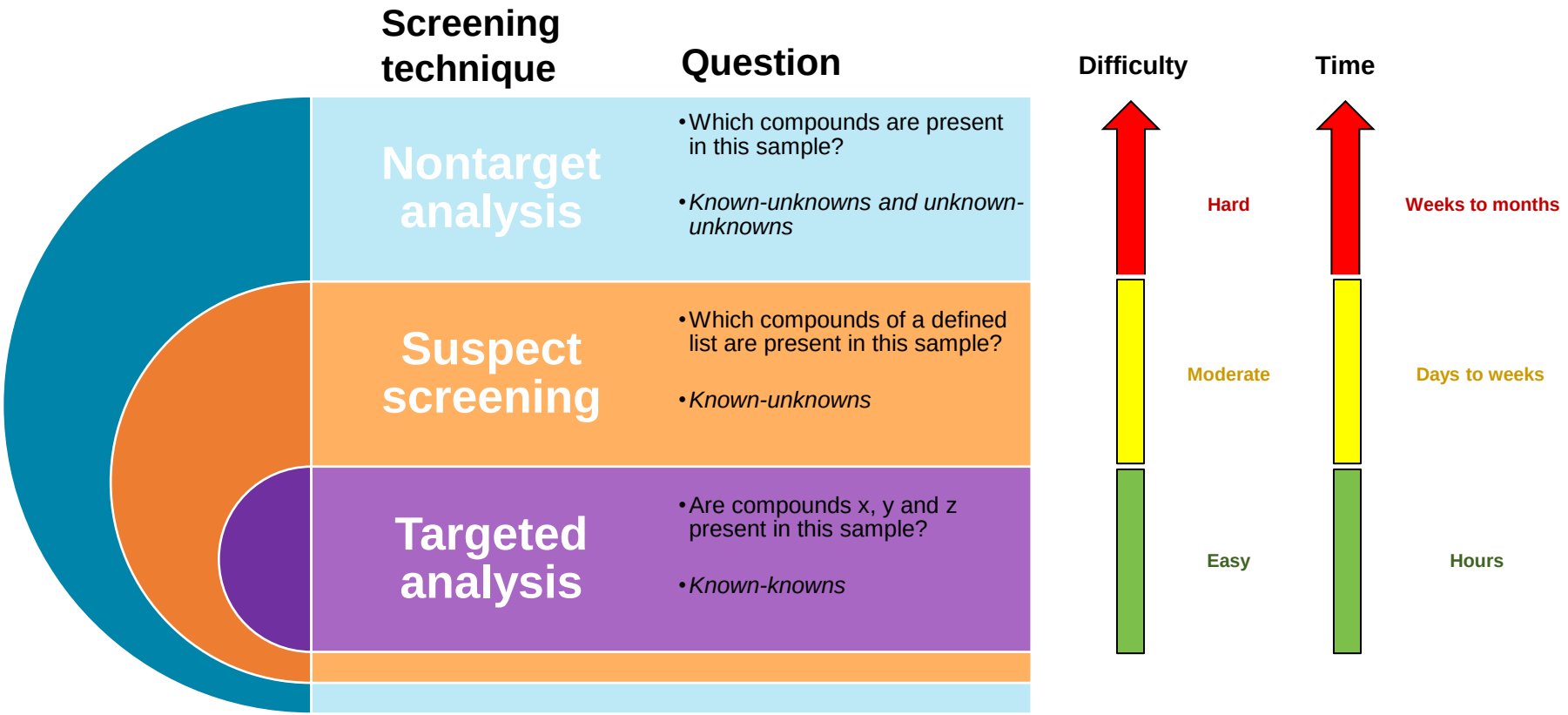
[⊥]Institute of Biogeochemistry and Pollutant Dynamics, ETH Zürich, Universitätstrasse 16, 8092 Zürich, Switzerland

[#]KWR Watercycle Research Institute, Groningenhaven 7, 3430 BB, Nieuwegein, The Netherlands



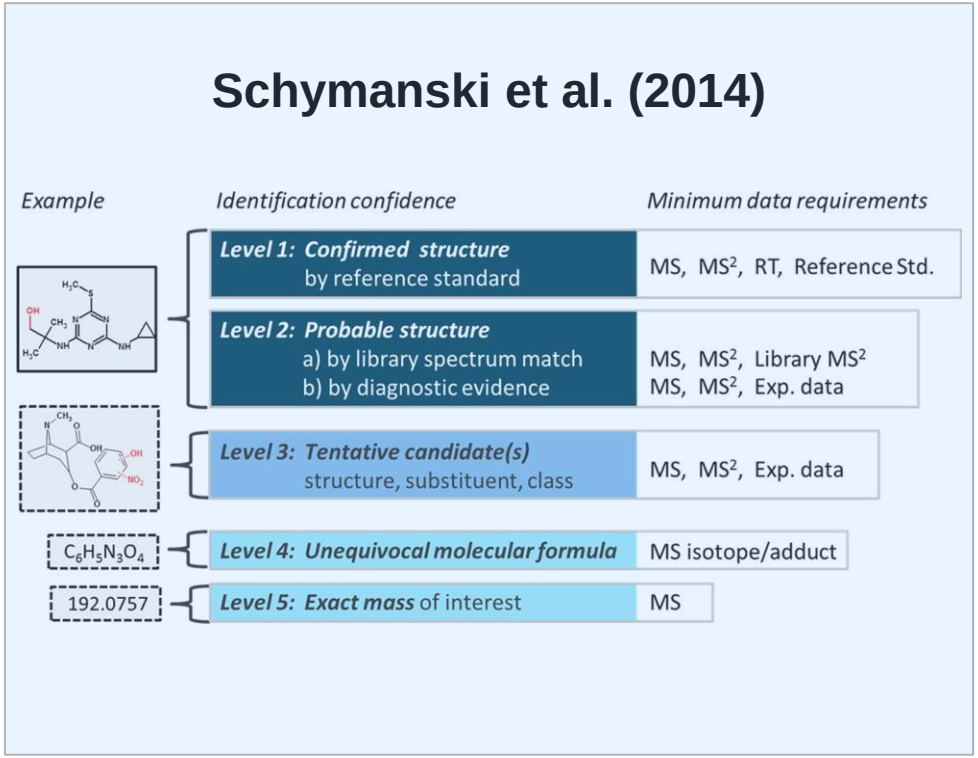
HRMS for NTA

LC-HRMS workflows



Communicating confidence

The most-used ranking system in environmental analysis:



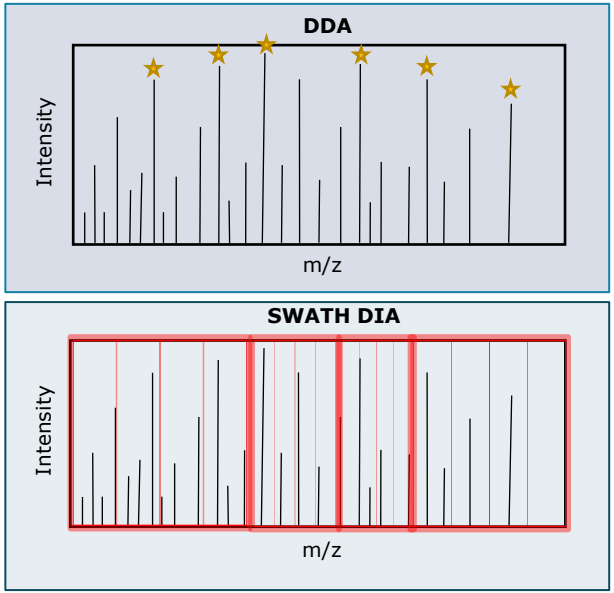
HRMS data acquisition and workflows

Workflow	QTOF system
Traditional high selectivity quantitation	MRM^{HR}
Suspect screening	MRM^{HR}, SWATH DIA, DDA
Unknown screening	SWATH DIA, DDA

Which compounds are present in this sample?

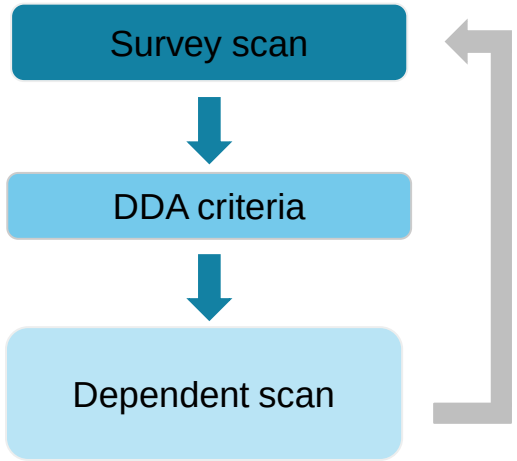
MS/MS data acquisition

Scan type	Meaning	Data dependent/ independent
DDA	Data dependent acquisition	Dependent
SWATH DIA	SWATH data independent acquisition	Independent



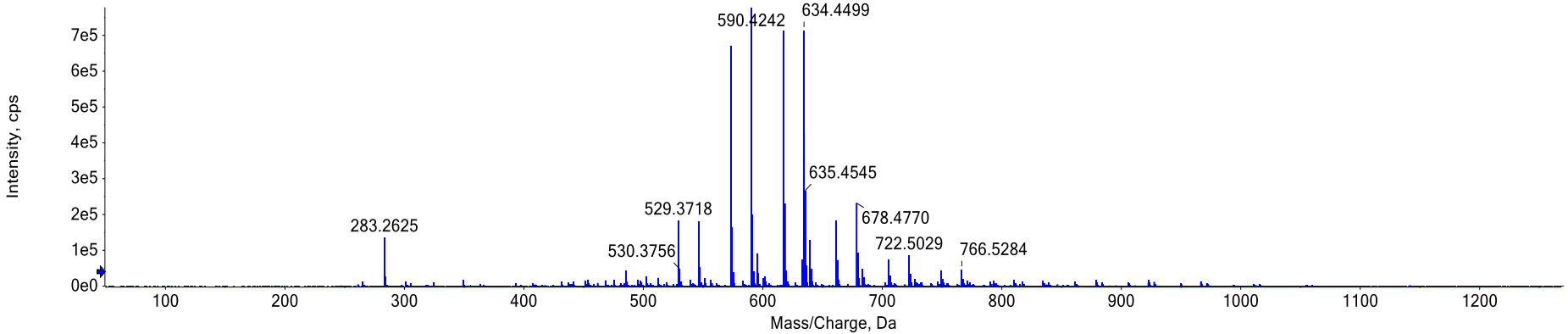
Data dependent acquisition (DDA)

- DDA is an acquisition strategy that automatically selects candidate ions for MS/MS study
- First, a survey scan is performed (TOF MS)
- If any candidate ions acquired in the survey scan meet the DDA criteria, a dependent MS/MS experiment is initiated for qualifying candidates
- After all dependent scans are completed, the process repeats



DDA overview

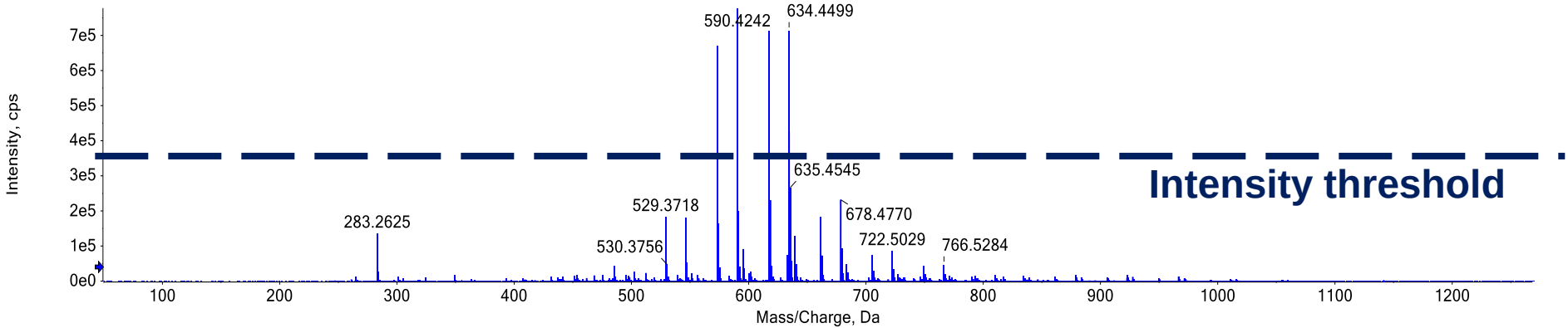
Survey scan: TOF MS



DDA criteria: select the top 10–12 most intense candidates

DDA overview

Survey scan: TOF MS



DDA criteria: select the top 10–12 most intense candidates

DDA overview

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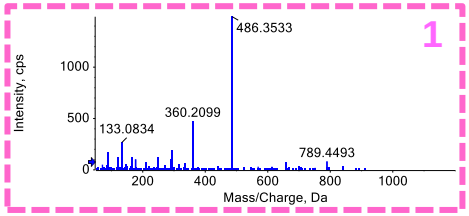
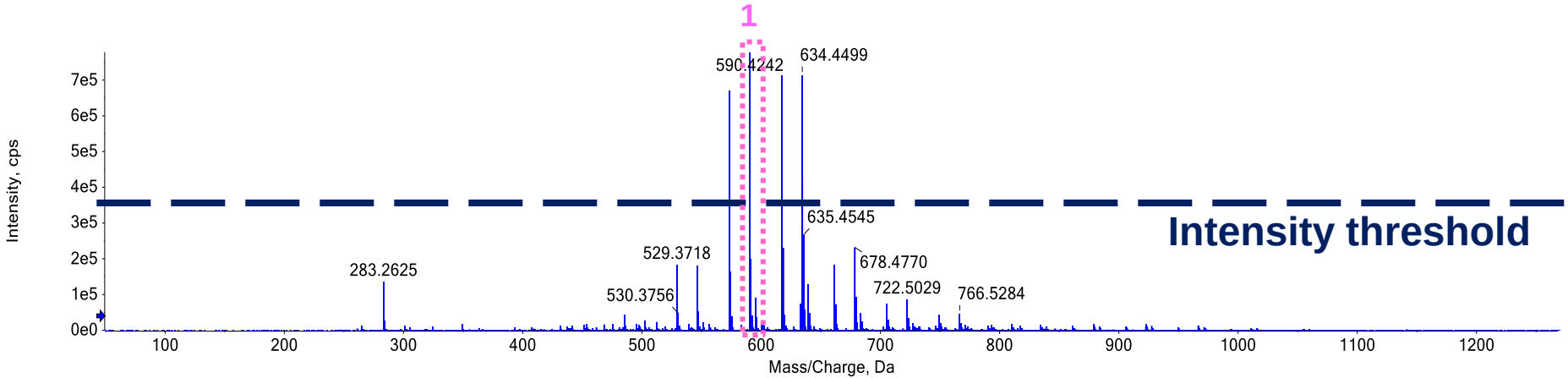
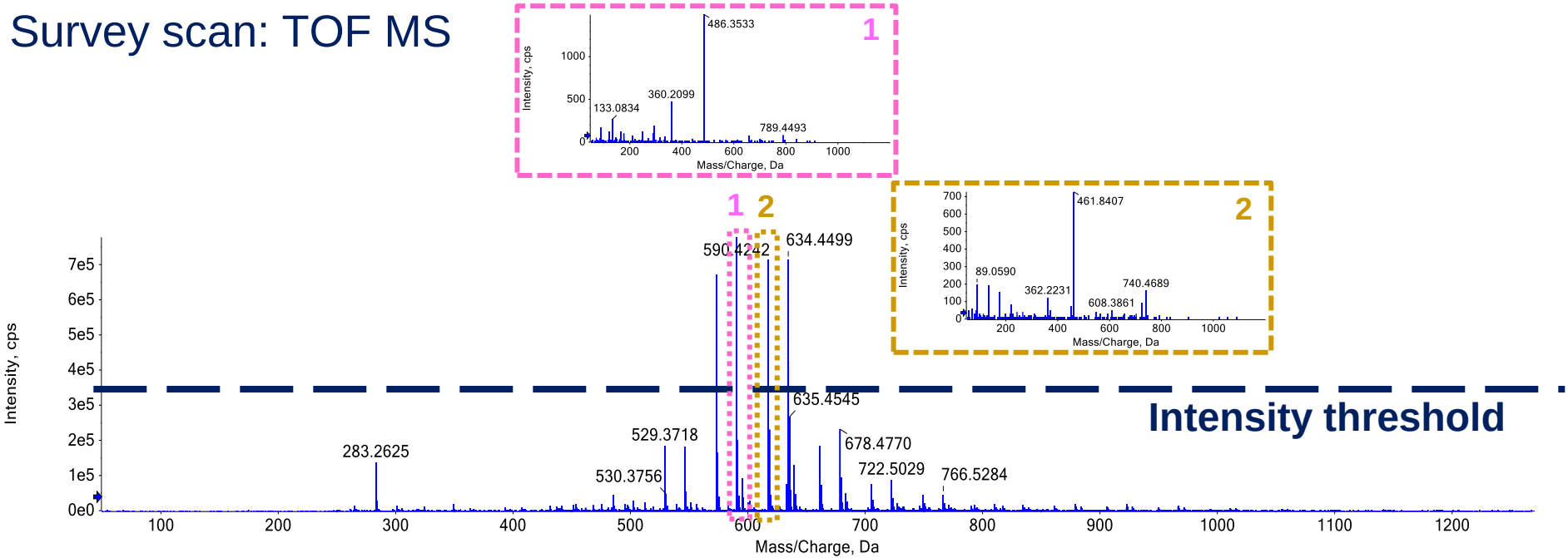




DDA criteria: select the top 10–12 most intense candidates

DDA overview

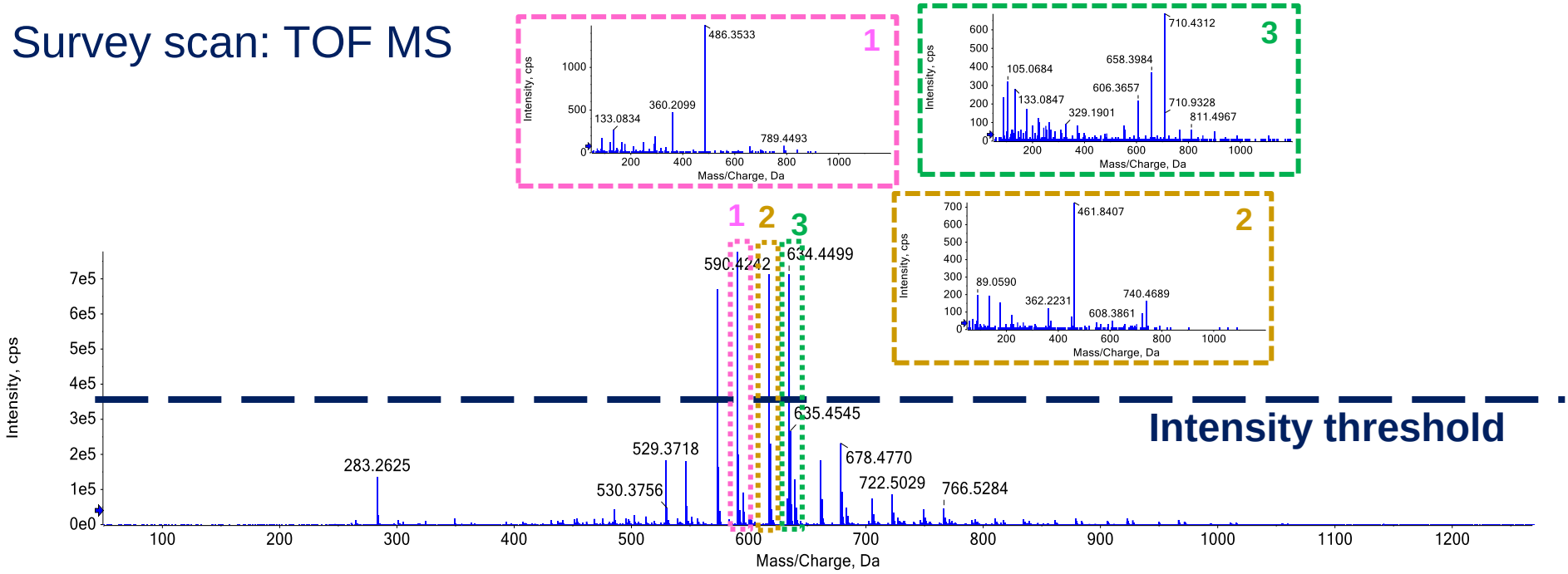
Survey scan: TOF MS



DDA criteria: select the top 10–12 most intense candidates

DDA overview

Survey scan: TOF MS



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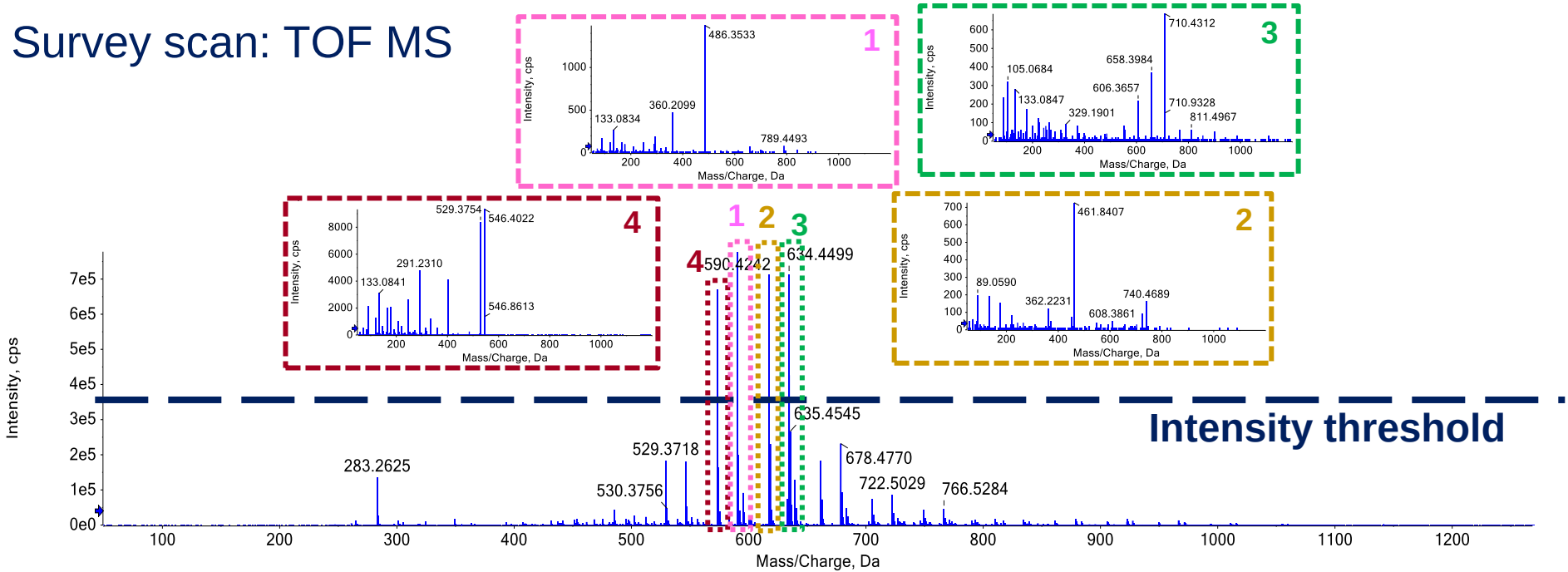
DDA overview

Survey scan: TOF MS



DDA criteria: select the top 10–12 most intense candidates

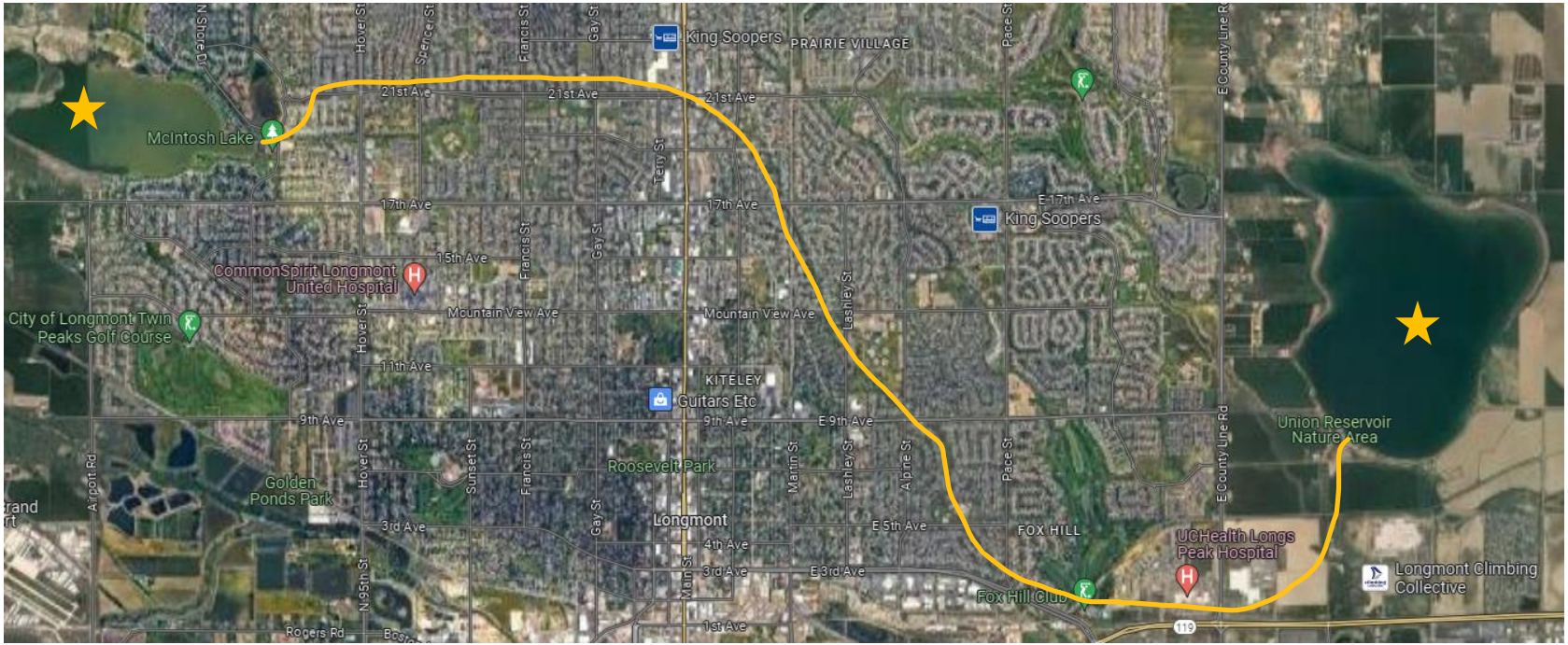
Our Water Study

Sample Information

- Water source near Karl's house in Longmont, CO
  - Oligarchy Ditch runs from McIntosh Lake to Union Reservoir

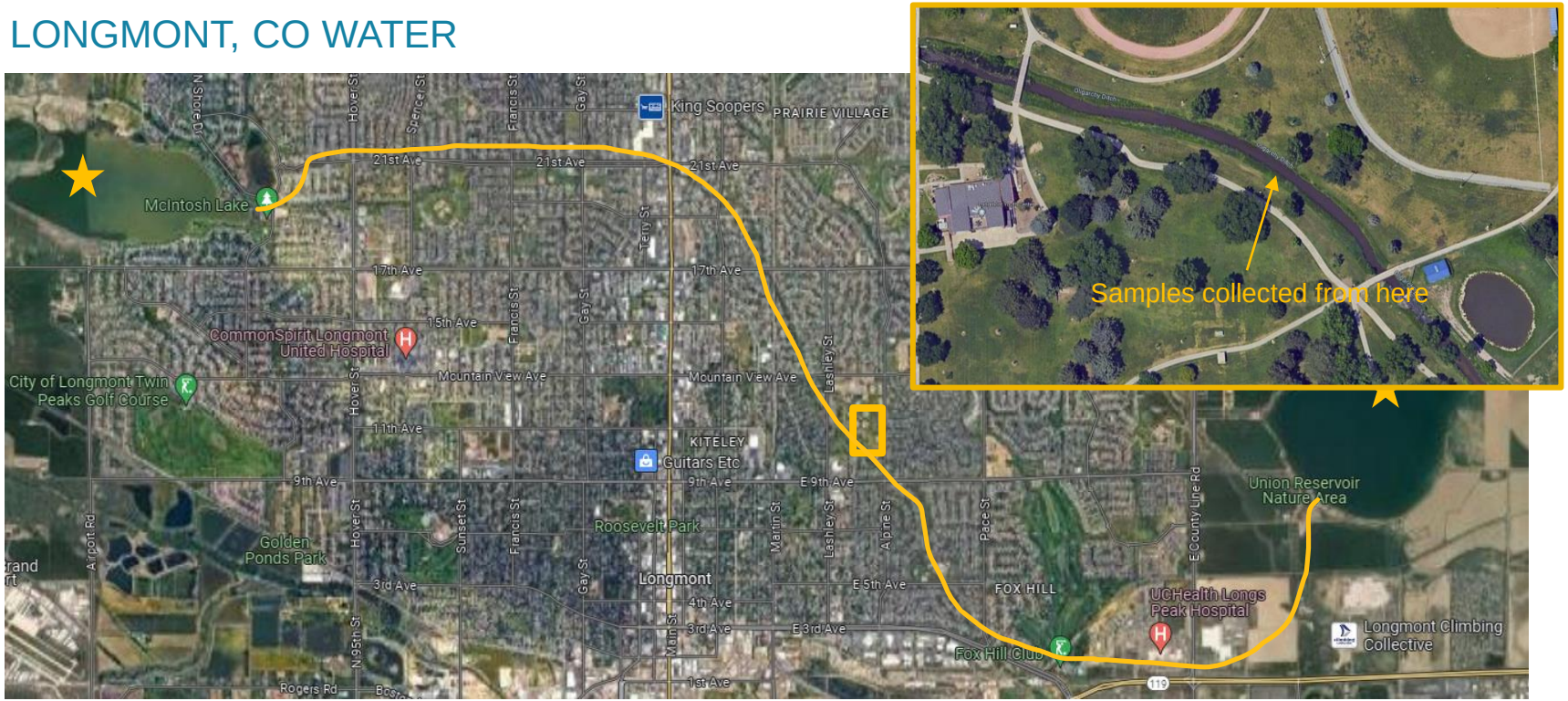
Sample Information

LONGMONT, CO



Sample Information

LONGMONT, CO WATER

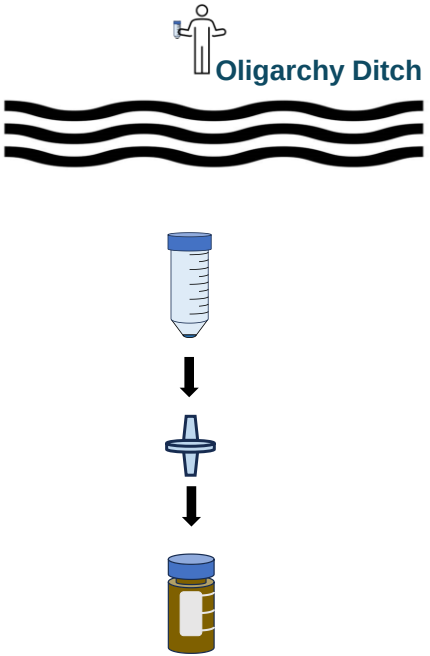


Sample Information

- Water source near Karl's house
  - Oligarchy Ditch runs from McIntosh Lake to Union Reservoir
- Collected samples on a daily-ish basis across 1.5 months in the spring

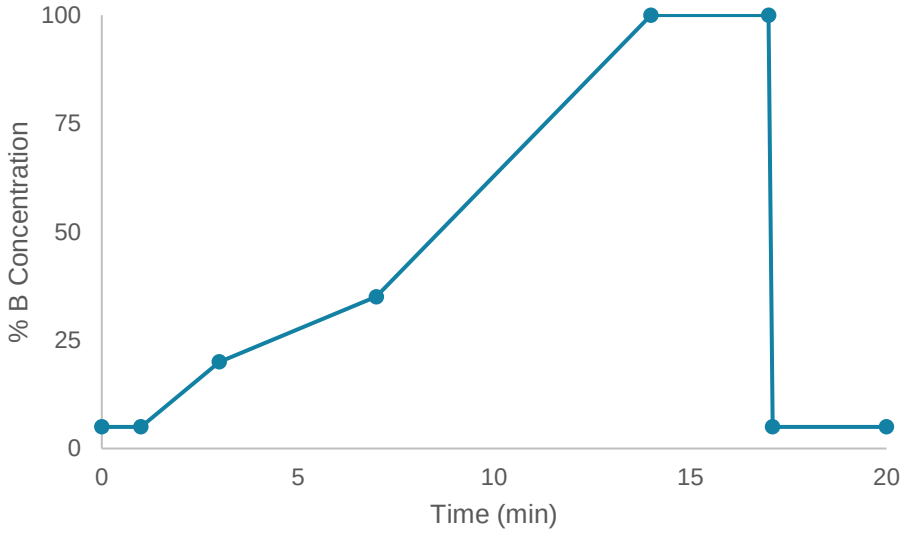
Simple Sample Preparation

- Water samples were filtered using a 0.2 µm nylon syringe filter directly to LC autosampler vial
- In this study we are only performing screening and identification, not quantification



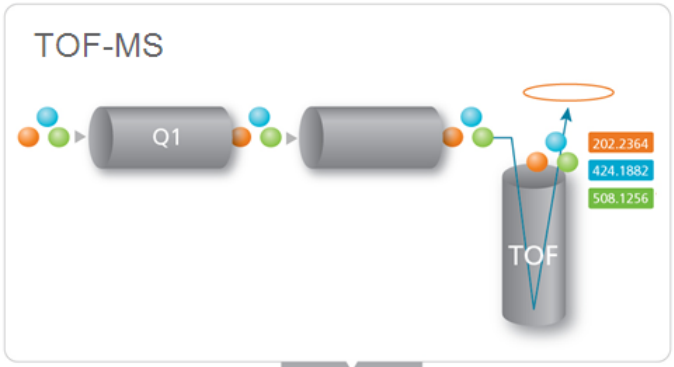
Chromatographic Separation

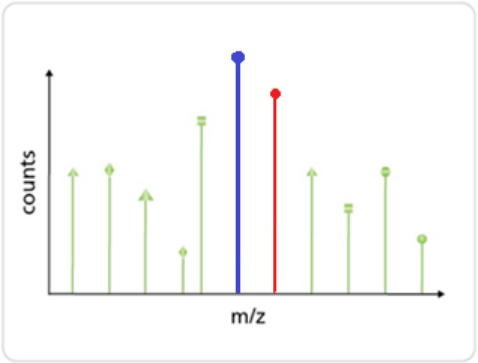
- Column: Phenomenex Luna Omega Polar C18 150 x 3 mm
- Flow rate: 0.7 mL/min
- Mobile phases
  - A: Water with 10mM ammonium formate and 0.1% formic acid
  - B: Acetonitrile with 10mM ammonium formate and 0.1% formic acid

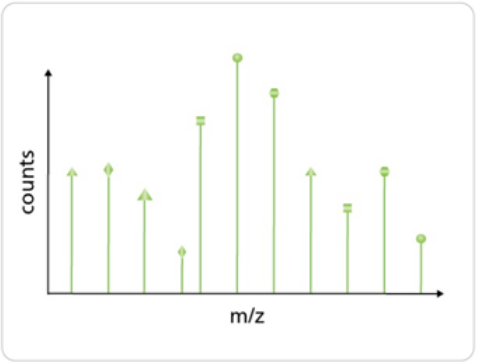


ZenoTOF 7600 Analysis

- Data Dependent Analysis
- Top 14 candidate precursor ions
- Collision energy: 20 v
- Collision energy spread: 10 v

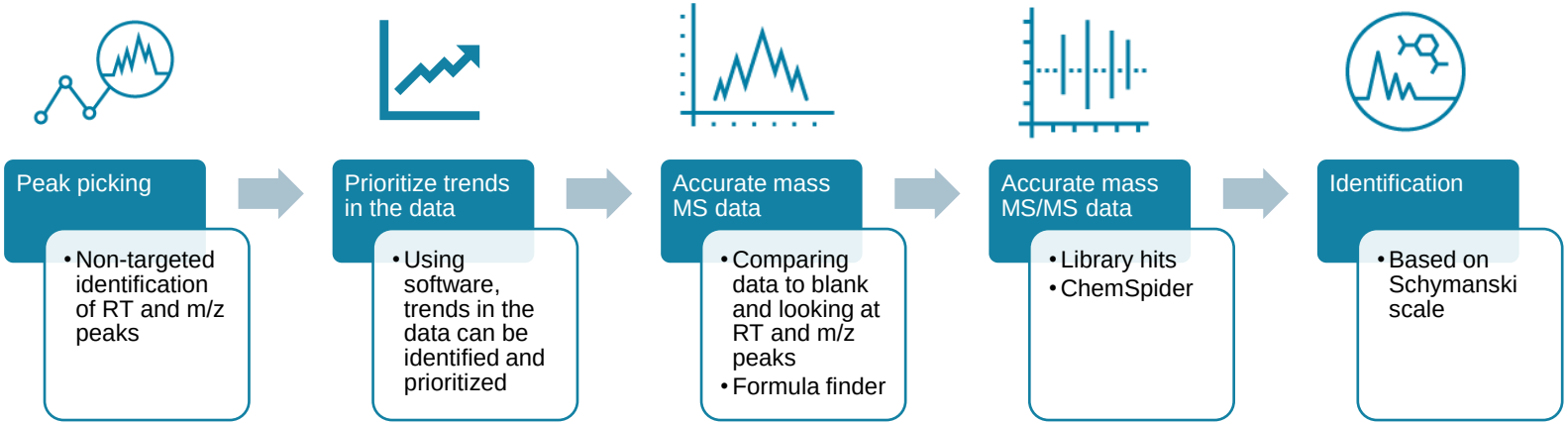






Monitoring Trends

Data processing workflow



Non-Targeted Peak Picking

Configure the parameters to use to find the non-targeted peaks to add to the extracted ion chromatogram list

Minimum retention time 1 min

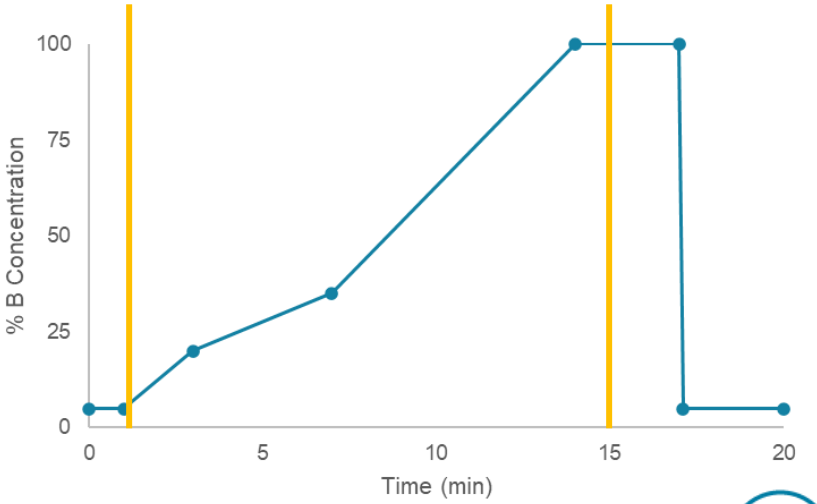
Maximum retention time 15 min

Peak detection sensitivity

Fast Exhaustive

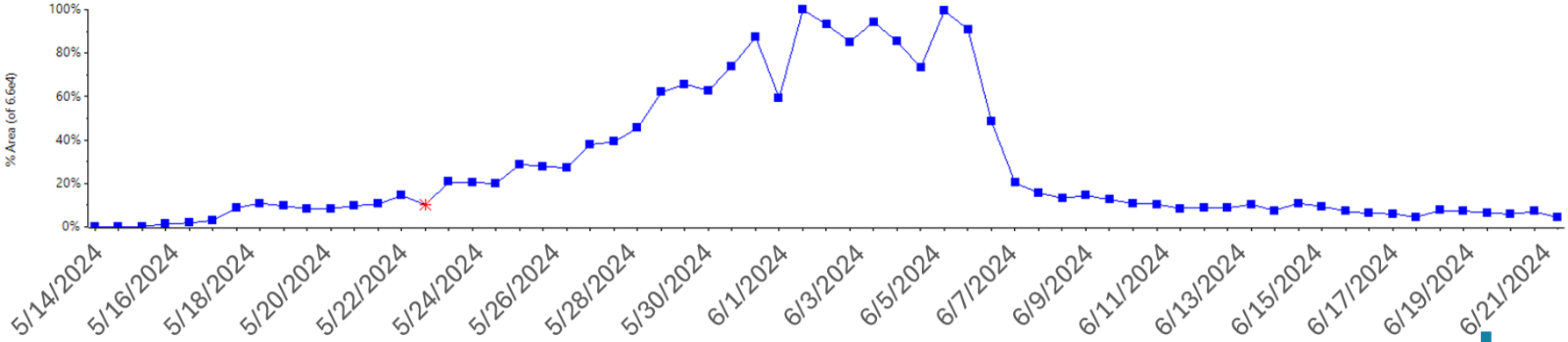
Area Ratio Threshold (Unknown/Control) 10

Group peaks by adduct or charge

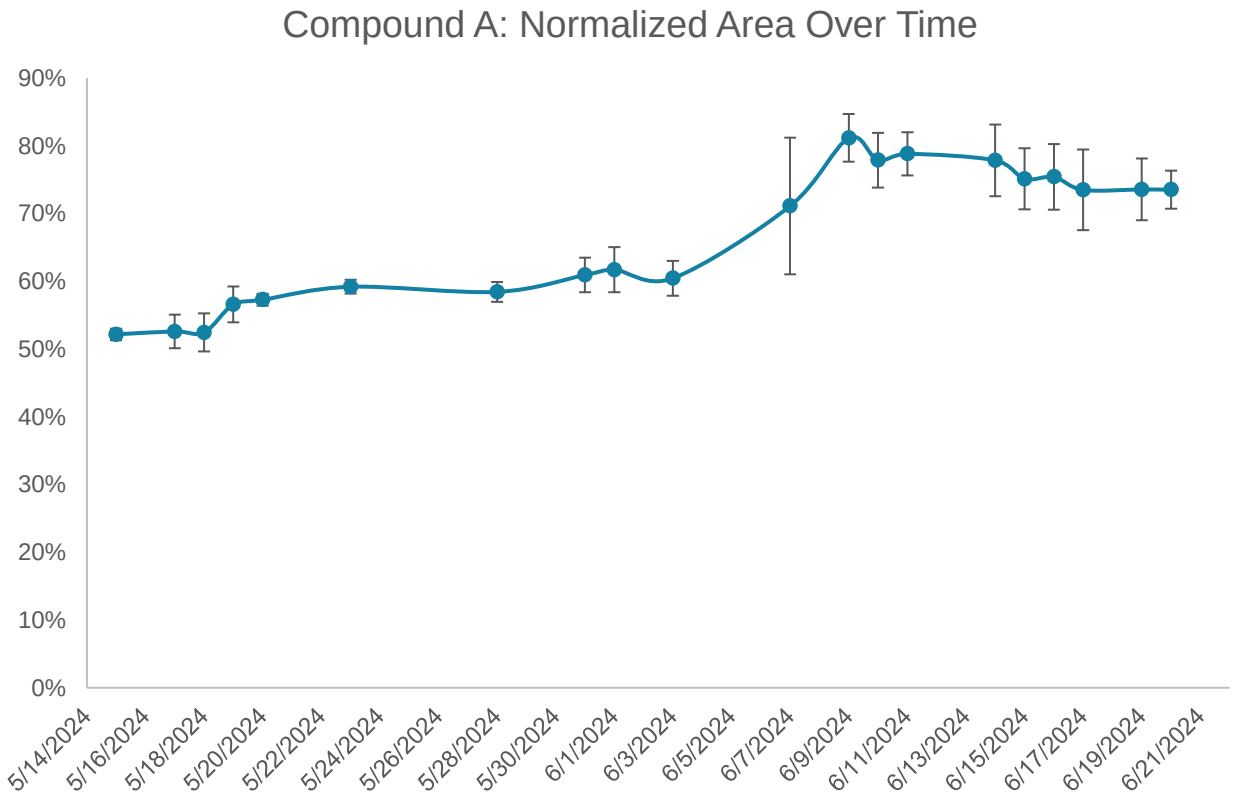


Data Analysis

- Using enviMass for prioritization of untargeted data
- Unknown spectral features with distinct temporal trends were identified based on intensity patterns
- Using libraries and ChemSpider integrated into SciexOS to tentatively ID our prioritized compounds based on MS/MS fragmentation patterns and *in silico* fragmentation based on chemical structure

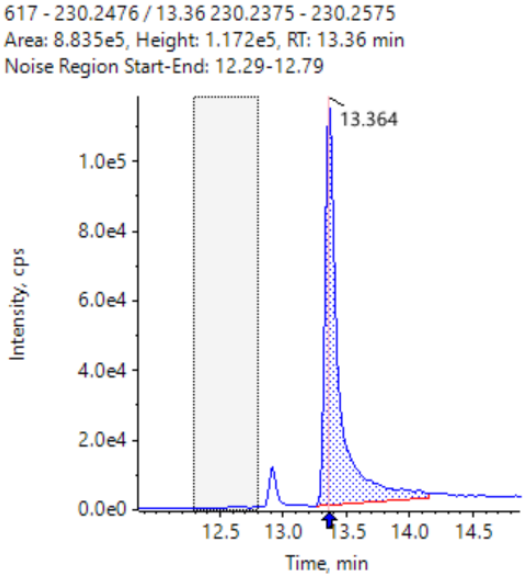


Compound A

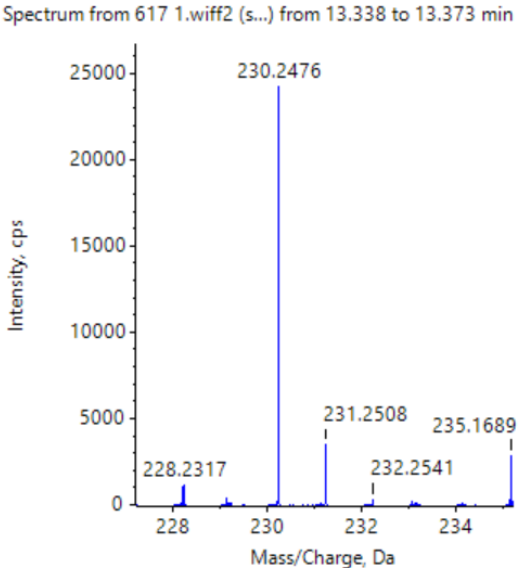


Compound A

RT: 13.3 min *m/z:* 230.2476

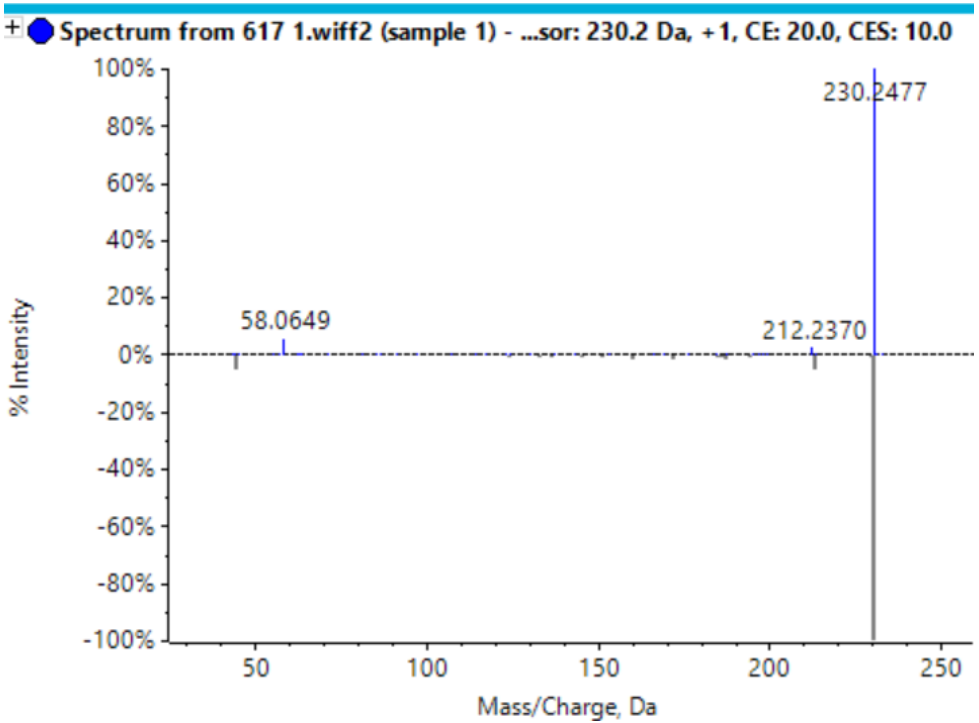


Extracted ion chromatogram



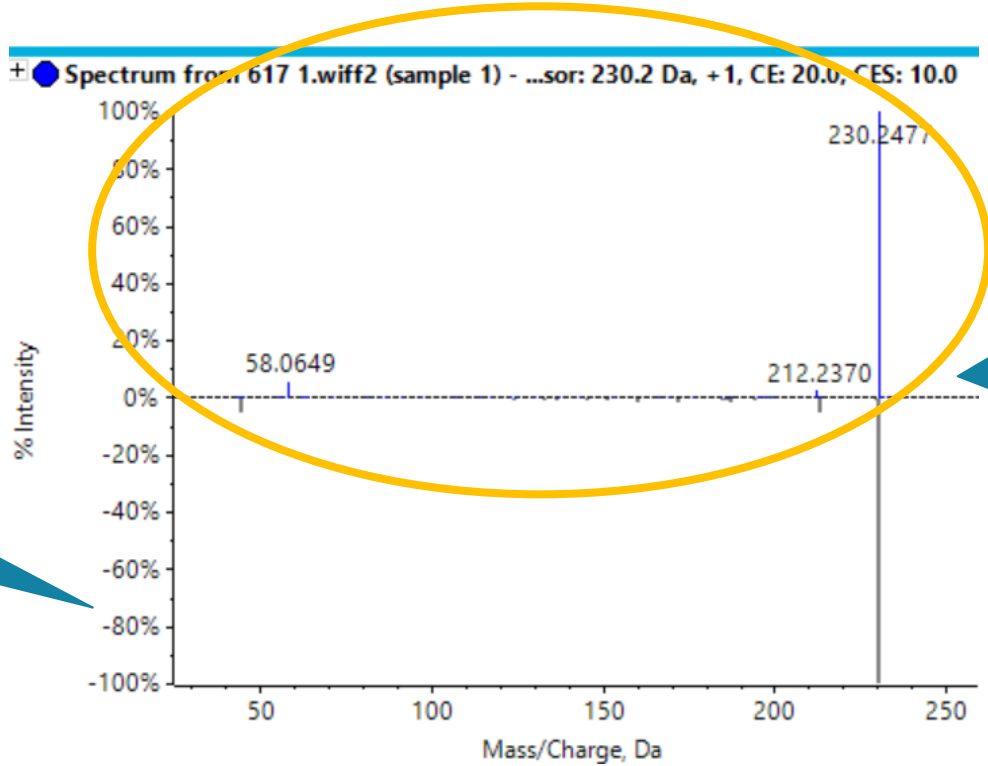
Precursor ion mass spectrum

Compound A



MS/MS Spectrum

Compound A



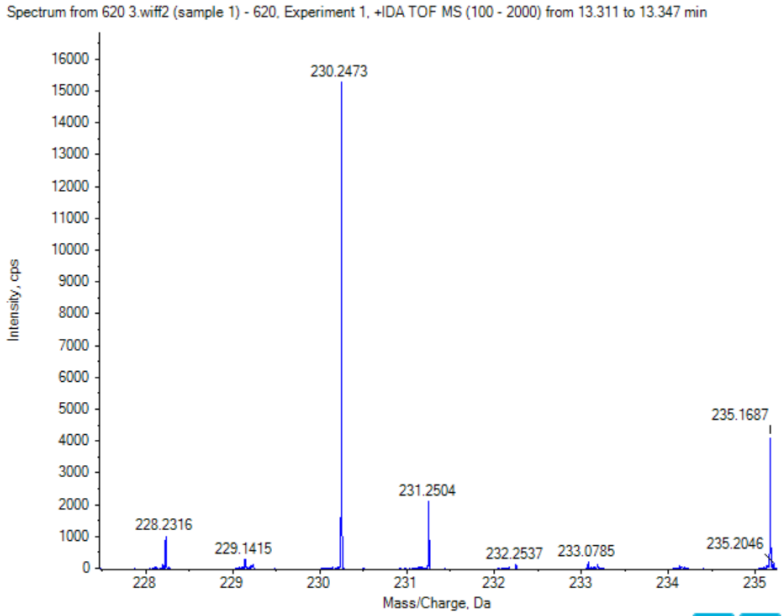
MS/MS spectrum lacking diagnostic fragmentation information

Poor library matching

MS/MS Spectrum

Compound A

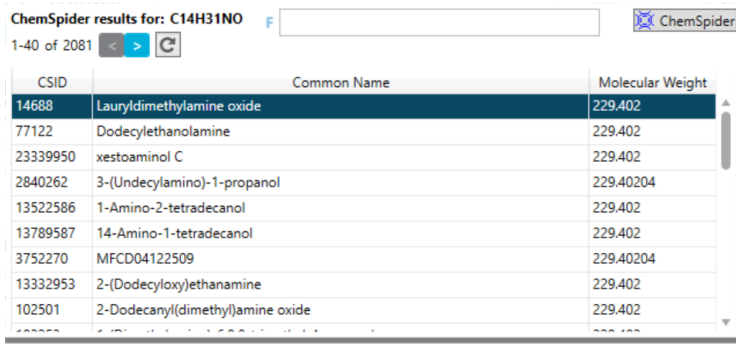
- Using exact mass to match potential chemical formulas
- Searching ChemSpider Database for entries which match potential formula
- $C_{14}H_{31}NO$ matches the precursor molecular weight with a mass error of 2.4 ppm
  - ChemSpider produces >2000 hits in the database matching this formula



Compound A

$C_{14}H_{31}NO$

Compounds matching formula



CSID	Common Name	Molecular Weight
14688	Lauryldimethylamine oxide	229.402
77122	Dodecylethanolamine	229.402
23339950	xestoaminol C	229.402
2840262	3-(Undecylamino)-1-propanol	229.40204
13522586	1-Amino-2-tetradecanol	229.402
13789587	14-Amino-1-tetradecanol	229.402
3752270	MFCD04122509	229.40204
13332953	2-(Dodecyloxy)ethanamine	229.402
102501	2-Dodecanyl(dimethyl)amine oxide	229.402

Compound A

FORMULA FINDER AND CHEMSPIDER

Compounds matching formula

ChemSpider results for: C14H31NO

1-40 of 2081

ChemSpider

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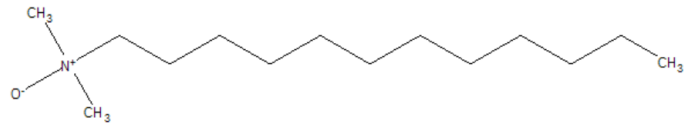
3752270	MFCD04122509	229.40204
13332953	2-(Dodecyloxy)ethanamine	229.402
102501	2-Dodecanyl(dimethyl)amine oxide	229.402

Display all Carbon Atoms

Options...

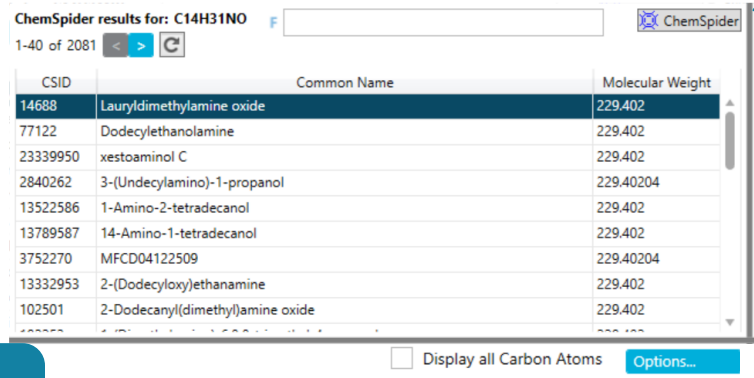
Structure of highlighted compound

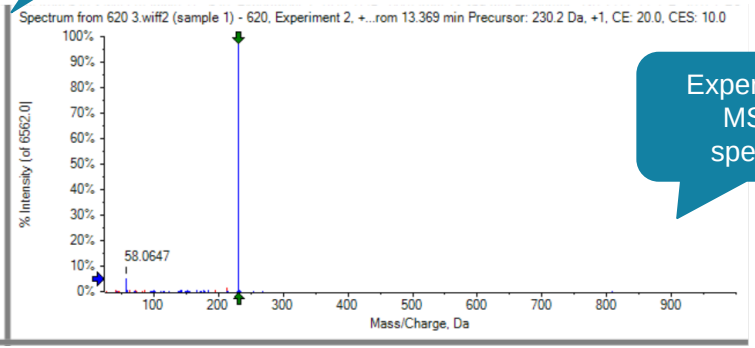


Compound A

FORMULA FINDER AND CHEMSPIDER

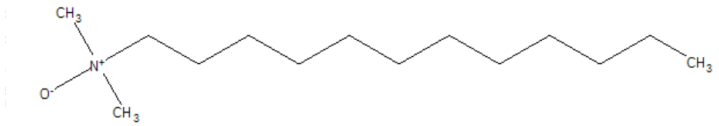
Compounds matching formula





Experimental MS/MS spectrum

Structure of highlighted compound



Compound A

FORMULA FINDER AND CHEMSPIDER

Compounds matching formula

ChemSpider results for: C14H31NO

1-40 of 2081

ChemSpider

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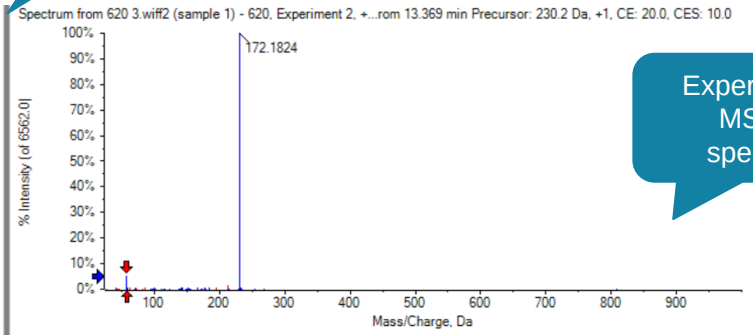
23339950	xestoaminol C	229.402
2840262	3-(Undecylamino)-1-propanol	229.40204
13522586	1-Amino-2-tetradecanol	229.402
13789587	14-Amino-1-tetradecanol	229.402
3752270	MFCD04122509	229.40204
13332953	2-(Dodecyloxy)ethanamine	229.402
102501	2-Dodecanyl(dimethyl)amine oxide	229.402



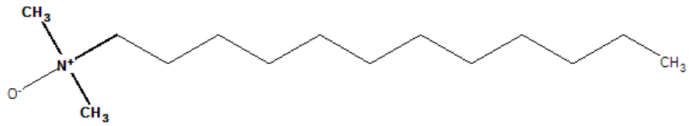
Experimental MS/MS spectrum

Structure of highlighted compound

Display all Carbon Atoms

Options...

cted composition: $C_3H_8N^+$ (58.0651 Da)



Options...

Fragments | Peaks

Mass/Charge	Intensity (%)	Assigned	Error (Da)
58.0647	5.39	☑	0.000
230.0807	0.61	☑	0.167
230.0888	0.32	☑	0.159
230.1965	0.32	☑	0.051
230.2475	100.00	☑	0.000
230.2761	0.52	☑	0.028
230.2943	0.56	☑	0.046
230.5088	0.32	☑	0.261
27.0219	0.30	☐	

Matches: 8 of 27 peaks, 92.2% of total intensity

Fragments corresponding to structure

Compound A

FORMULA FINDER AND CHEMSPIDER

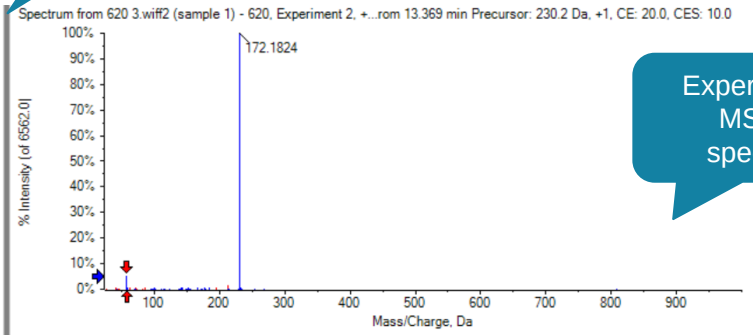
Compounds matching formula

ChemSpider results for: C14H31NO

1-40 of 2081

ChemSpider

CSID	Common Name	Molecular Weight
14688	Lauryldimethylamine oxide	229.402
77122	Dodecylethanolamine	229.402
23339950	xestoaminol C	229.402
2840262	3-(Undecylamino)-1-propanol	229.40204
13522586	1-Amino-2-tetradecanol	229.402
13789587	14-Amino-1-tetradecanol	229.402
3752270	MFCD04122509	229.40204
13332953	2-(Dodecyloxy)ethanamine	229.402
102501	2-Dodecanyl(dimethyl)amine oxide	229.402



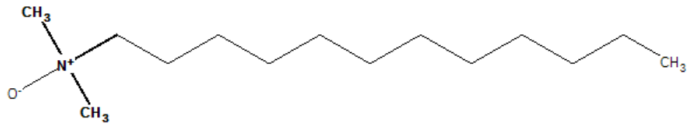
Experimental MS/MS spectrum

Structure of highlighted compound

Display all Carbon Atoms

Options...

cted composition: $C_3H_8N^+$ (58.0651 Da)



Options...

Fragments | Peaks

Mass/Charge	Intensity (%)	Assigned	Error (Da)
58.0647	5.39	☑	0.000
230.0807	0.61	☑	0.167
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230.1965	0.32	☑	0.051
230.2475	100.00	☑	0.000
230.2761	0.52	☑	0.028
230.2943	0.56	☑	0.046
230.5088	0.32	☑	0.261
27.0219		☐	

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Compounds matching formula

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1-40 of 2081

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3752270	MFCD04122509	229.40204
13332953	2-(Dodecyloxy)ethanamine	229.402
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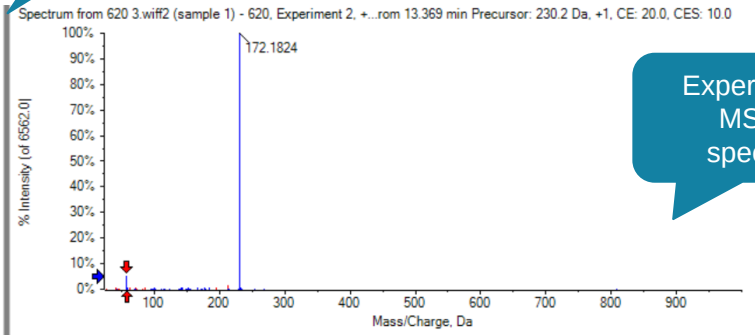

Experimental MS/MS spectrum

Structure of highlighted compound

Display all Carbon Atoms

Options...

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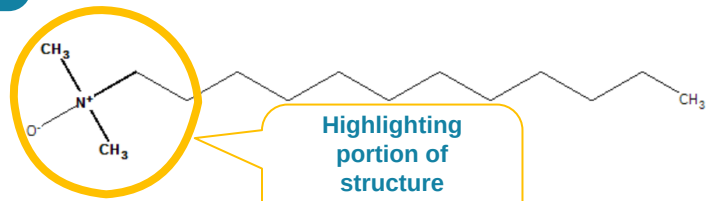

Highlighting portion of structure related to each fragment

Options...

Fragments | Peaks

Mass/Charge	Intensity (%)	Assigned	Error (Da)
58.0647	5.39	☑	0.000
230.0807	0.61	☑	0.167
230.0888	0.32	☑	0.159
230.1965	0.32	☑	0.051
230.2475	100.00	☑	0.000
230.2761	0.52	☑	0.028
230.2943	0.56	☑	0.046
230.5088	0.32	☑	0.261
27.0219	0.30	☐	

Matches: 8 of 27 peaks, 92.2% of total intensity

Fragments corresponding to structure

Compound A

FORMULA FINDER AND CHEMSPIDER

Compounds matching

Level 3 Identification with a tentative candidate structure

Experimental MS/MS spectrum

Structure of highlighted compound

Fragments corresponding to structure

Highlighting portion of structure related to each fragment

ChemSpider results for: C14H31NO

1-40 of 2081

CSID	Common Name
14688	Lauryldimethylamine oxide
77122	Dodecylethanolamine
23339950	xestoaminol C
2840262	3-(Undecylamino)-1-propanol
13522586	1-Amino-2-tetradecanol
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3752270	MFCD04122509
13332953	2-(Dodecyloxy)ethanamine
102501	2-Dodecanyl(dimethyl)amine oxide

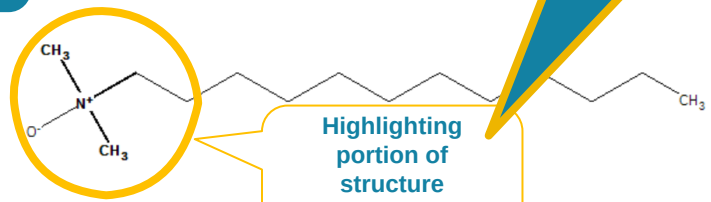
iment 2, +...rom 13.369 min Precursor: 230.2 Da, +1, CE: 20.0, CES: 10.0

400 500 600 700 800 900

Mass/Charge, Da

Display all Carbon Atom

cted composition: $C_3H_8N^+$ (58.0651 Da)



Options...

Fragments | Peaks

Mass/Charge	Intensity (%)	Assigned	Error (Da)
58.0647	5.39	☑	0.000
230.0807	0.61	☑	0.167
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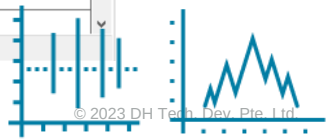
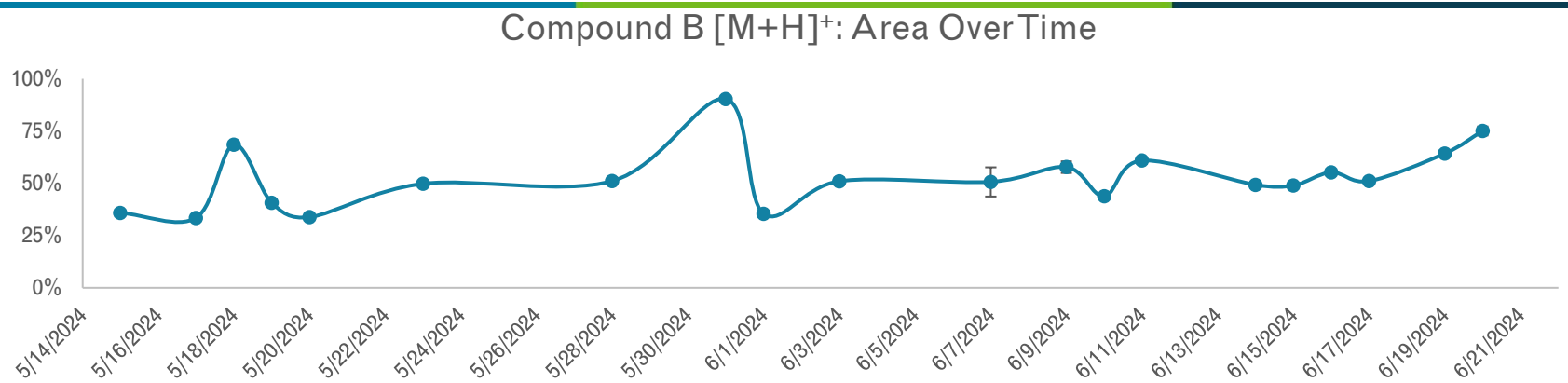
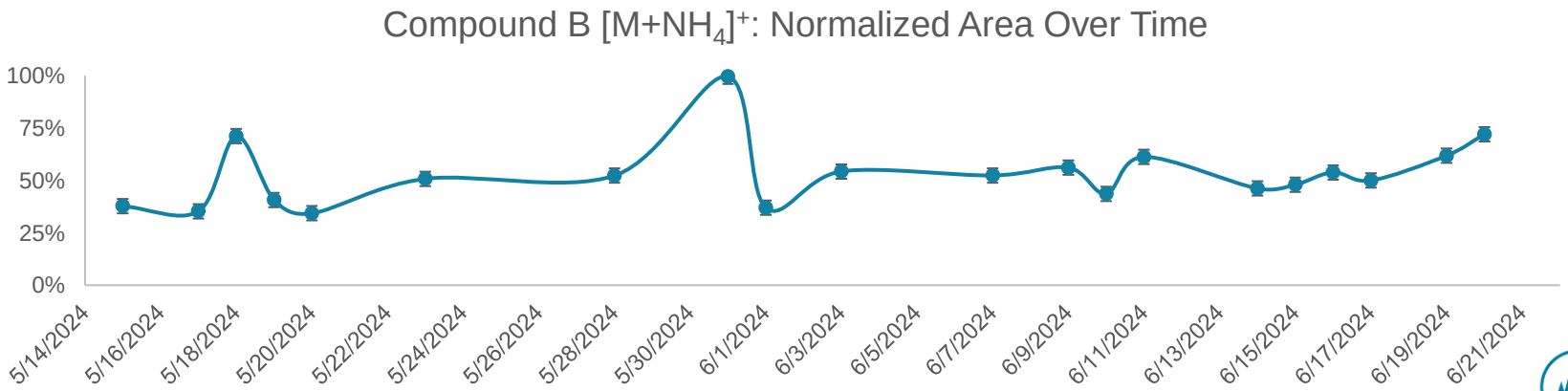
230.2475	100.00	☑	0.000
230.2761	0.52	☑	0.028
230.2943	0.56	☑	0.046
230.5088	0.32	☑	0.261
27.0219	0.30	☐	

Matches: 8 of 27 peaks, 92.2% of total intensity

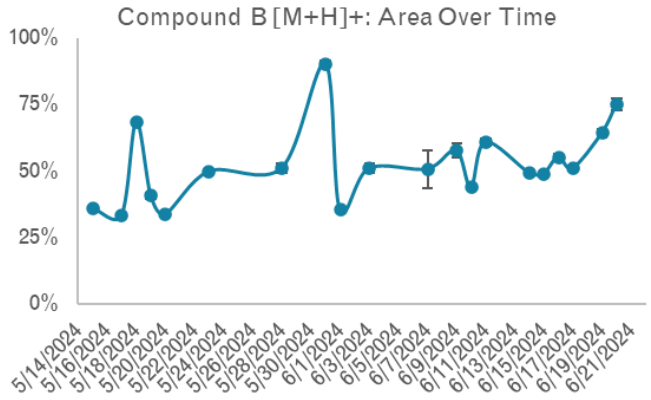


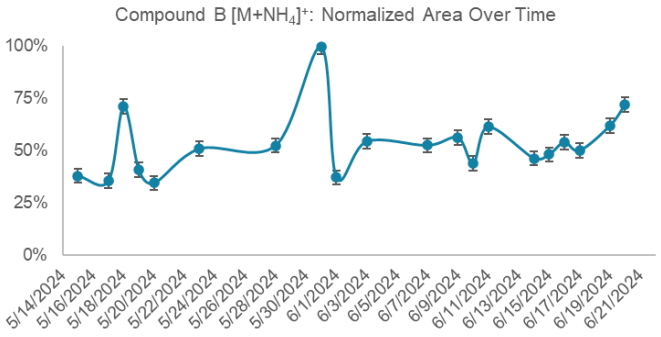
Compound B

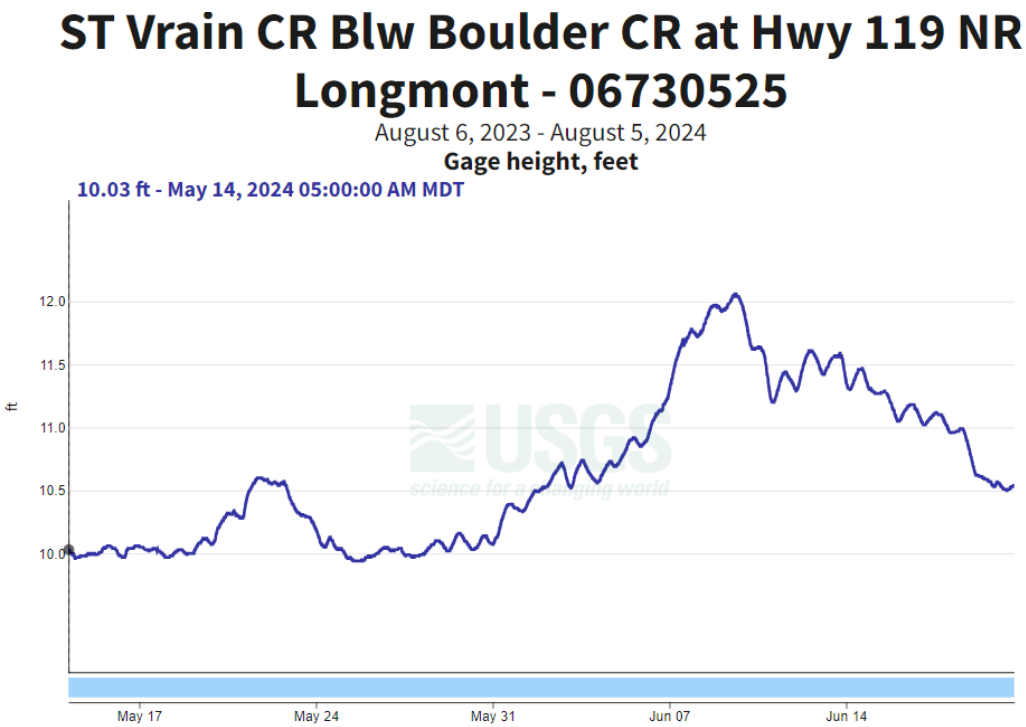




Gage height trends

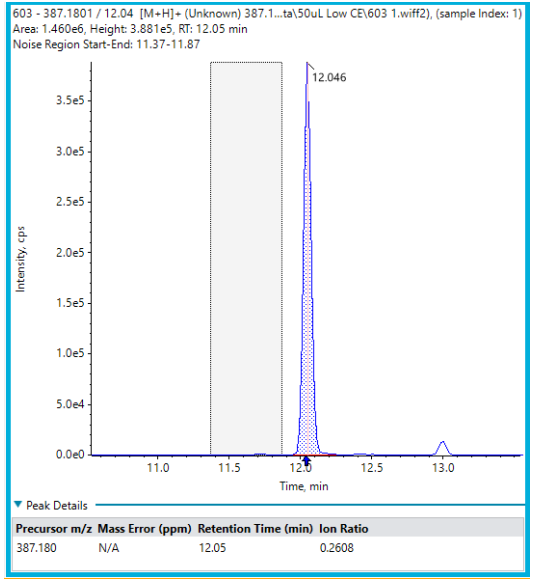




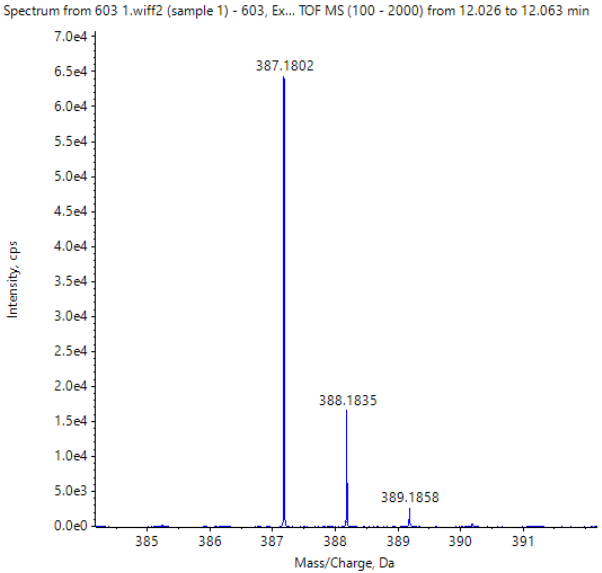


Compound B

RT: 12.05 *m/z:* 387.1802



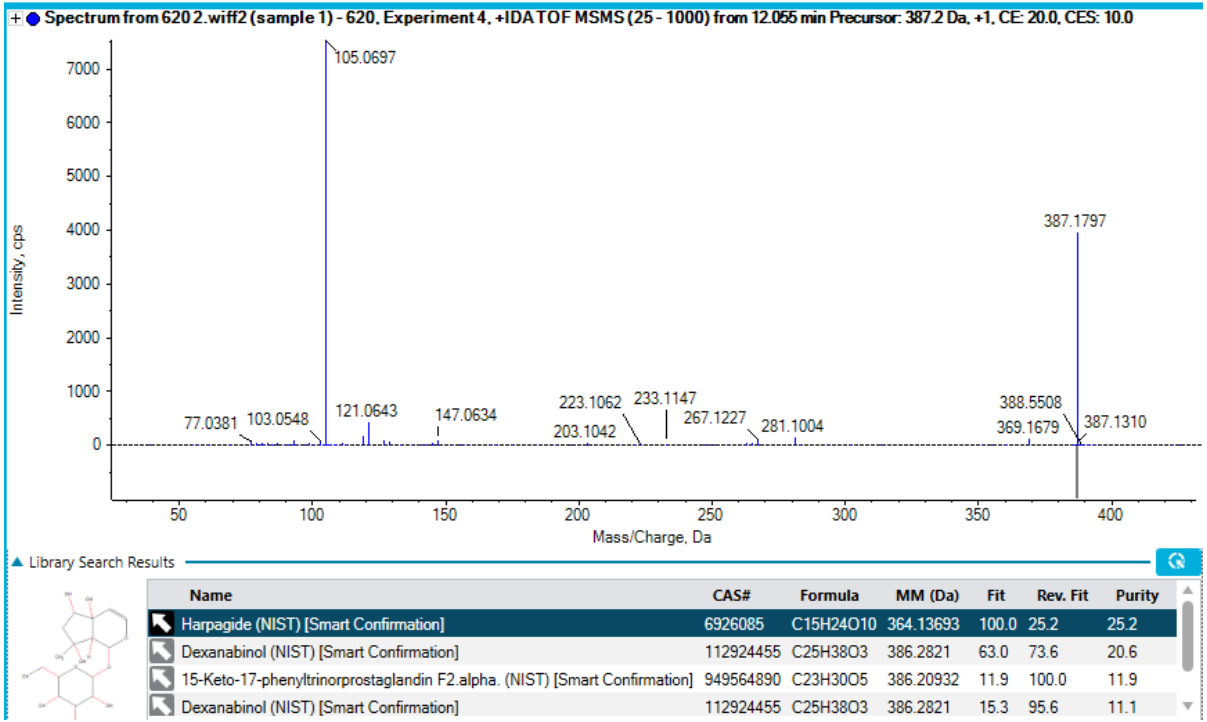
Extracted Ion Chromatogram Precursor Ion



Extracted Ion Chromatogram Precursor Ion

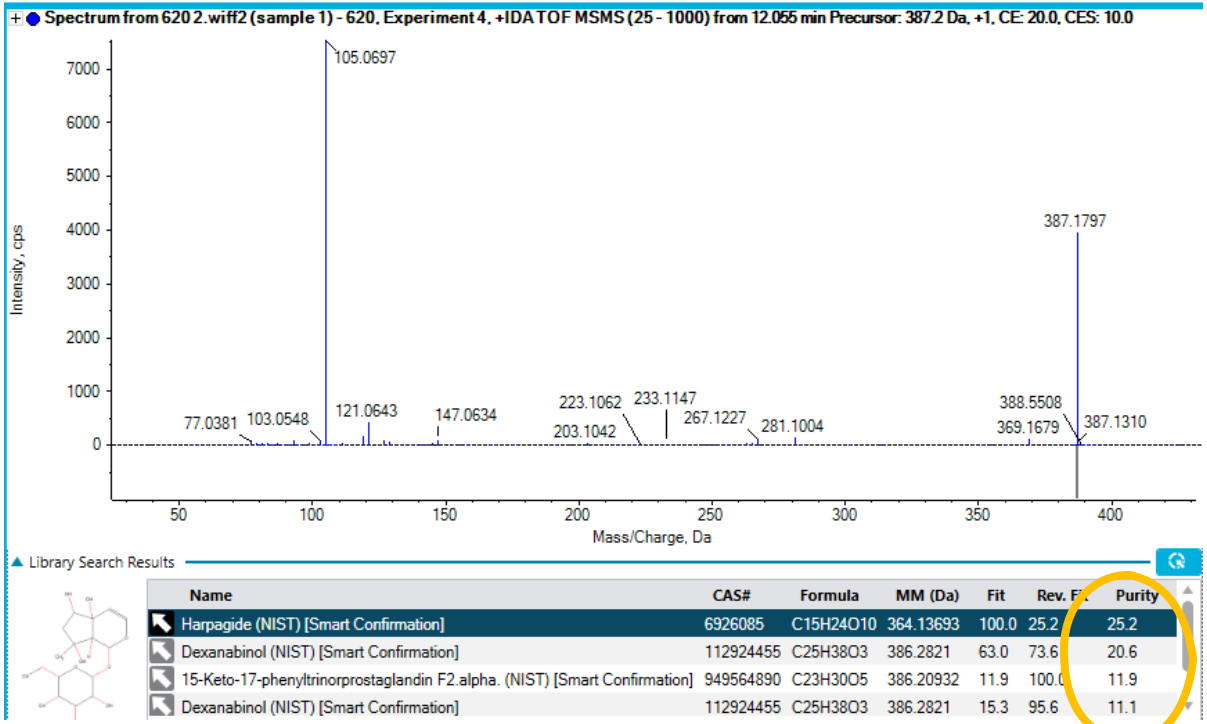
MS/MS Spectrum Compound B

LIBRARY MATCHES WITH LOW CONFIDENCE

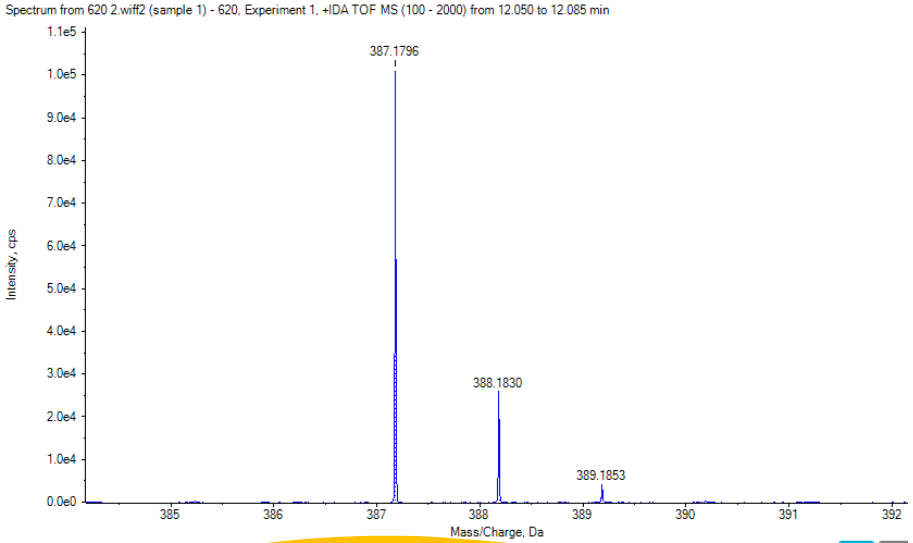


MS/MS Spectrum Compound B

LIBRARY MATCHES WITH LOW CONFIDENCE



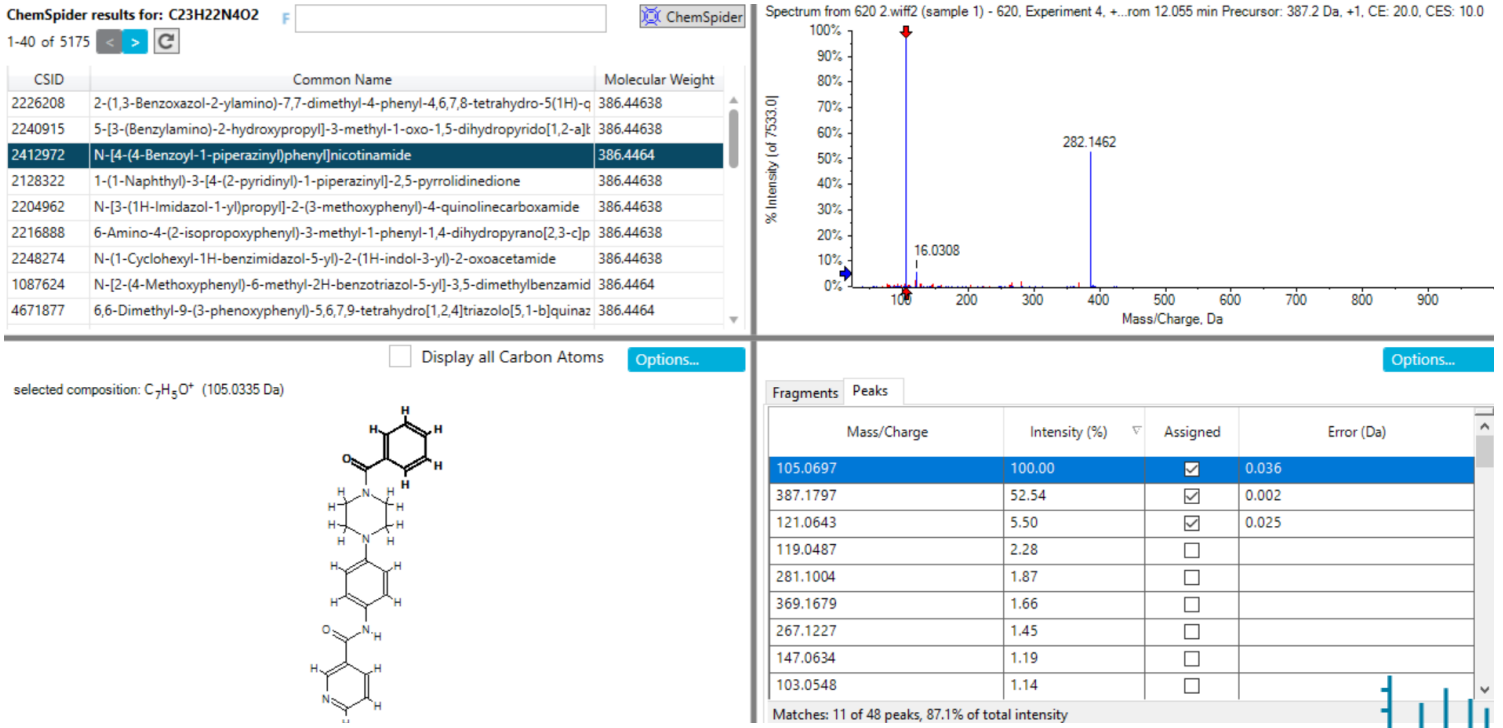
Formula Finder Results



Name	Formula	Score	m/z (Da)	Error (ppm)	Error MSMS (ppm)	Hit Count
	C20H25F3O4	57.4	387.17777	4.8	3.1	21
	C23H22N4O2	56.7	387.18155	4.9	3.1	5175
	C26H26OS	56.3	387.17771	5	3.2	2
	C15H31O9P	53.6	387.17785	4.6	4.6	1
	C18H22N6O4	52.4	387.17753	5.5	3.4	876
	C16H24F2N6OS	52.2	387.17731	6	2.3	0

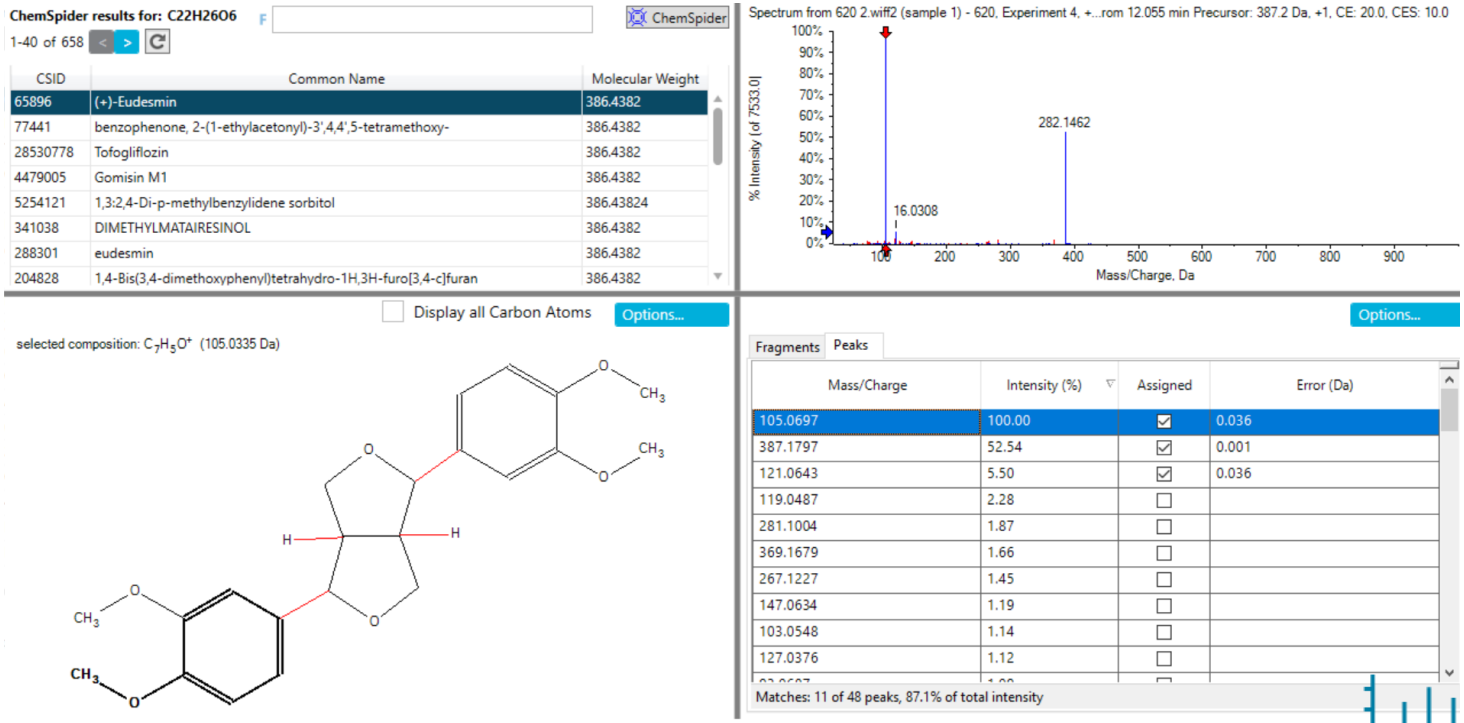
ChemSpider

$C_{23}H_{22}N_4O_2$



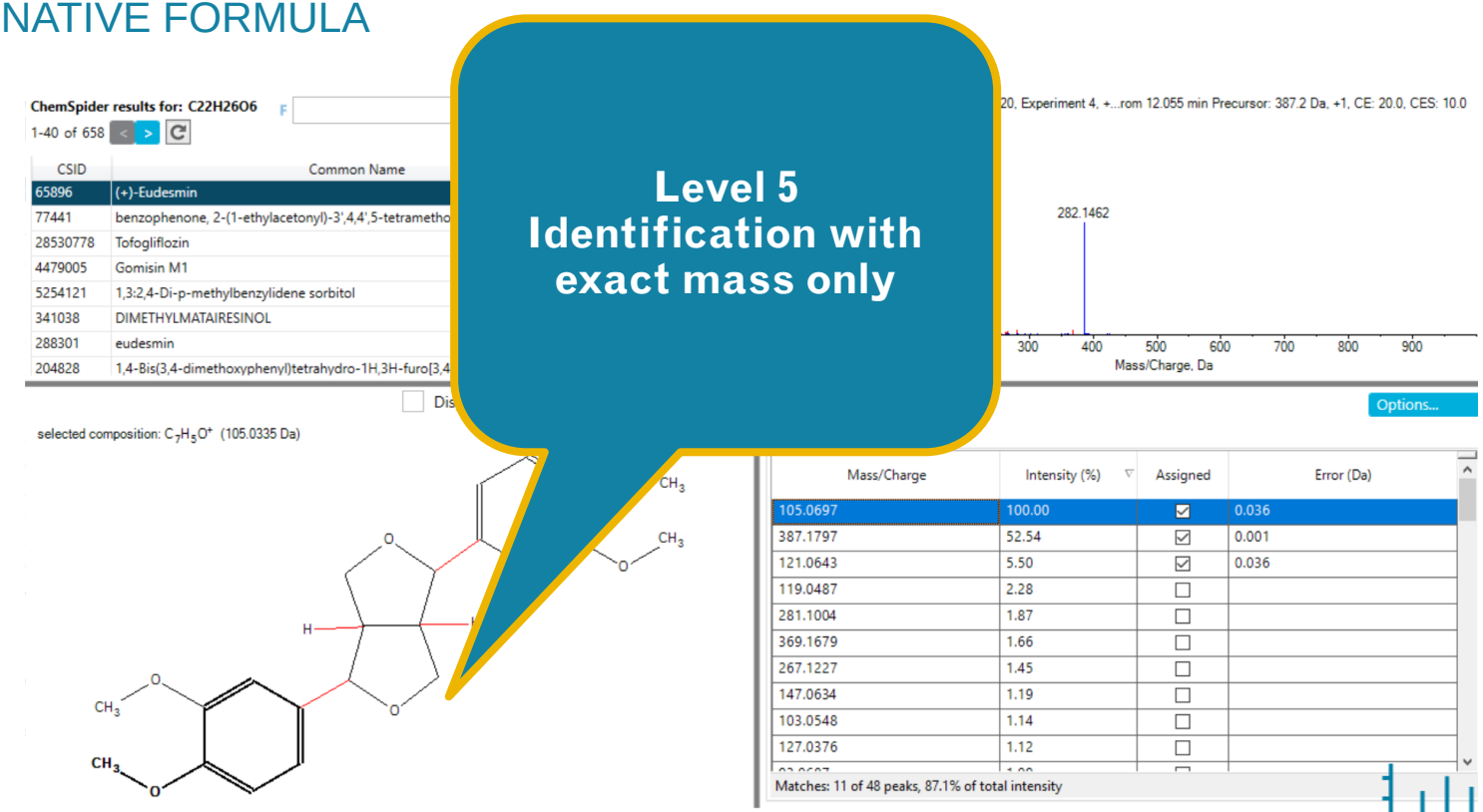
ChemSpider

$C_{22}H_{26}O_6$

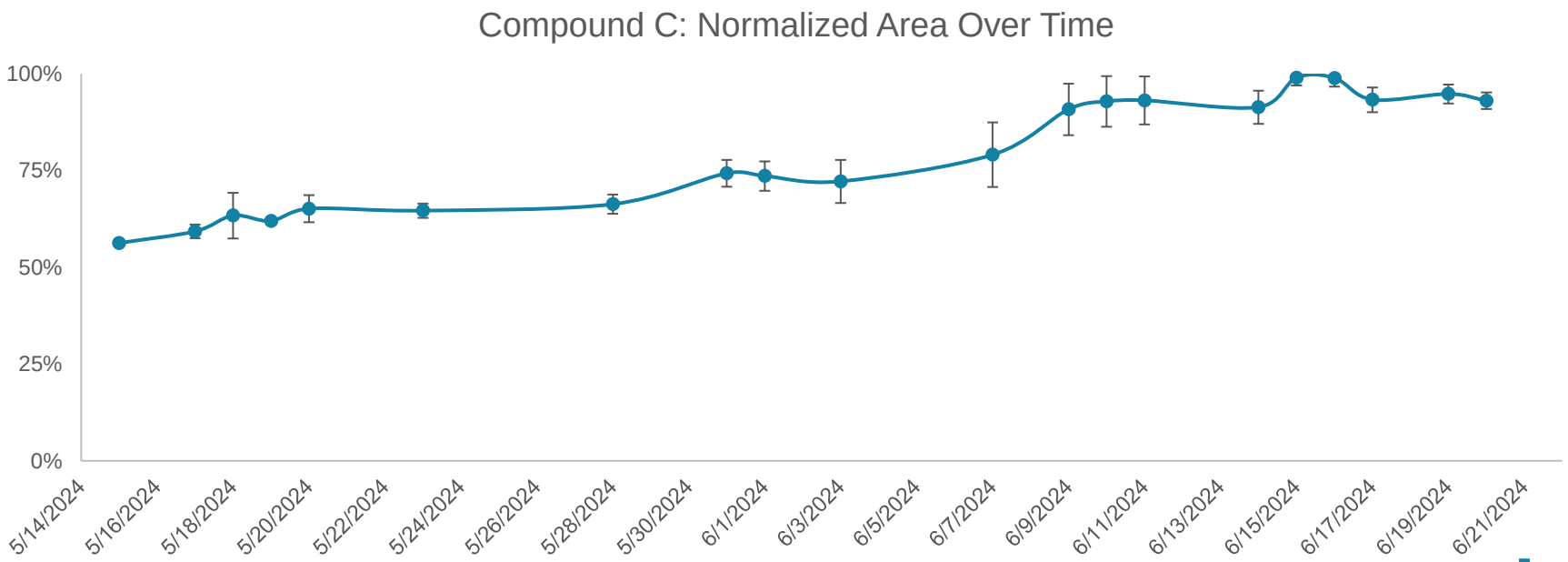


ChemSpider

ALTERNATIVE FORMULA

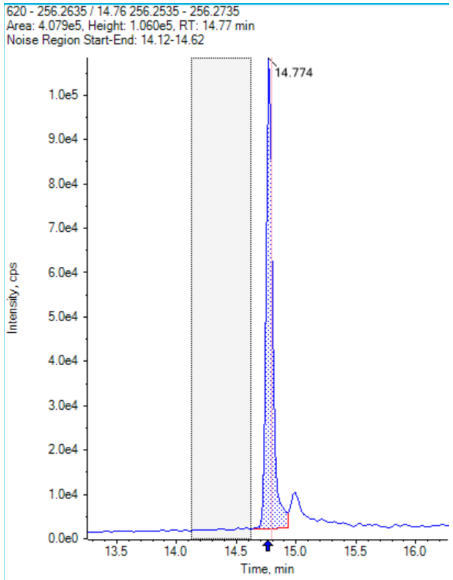


Compound C

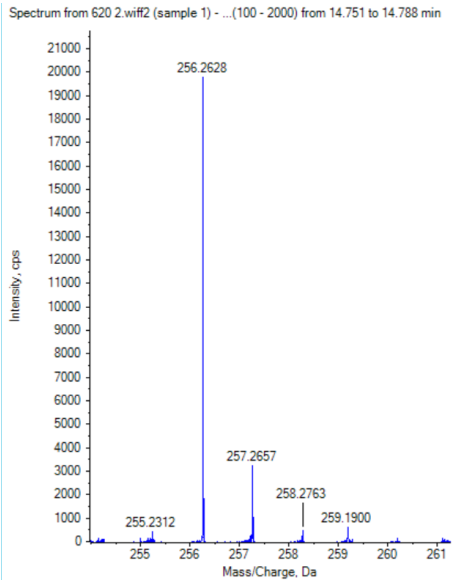


Compound C

RT: 14.77 *m/z* 256.2628



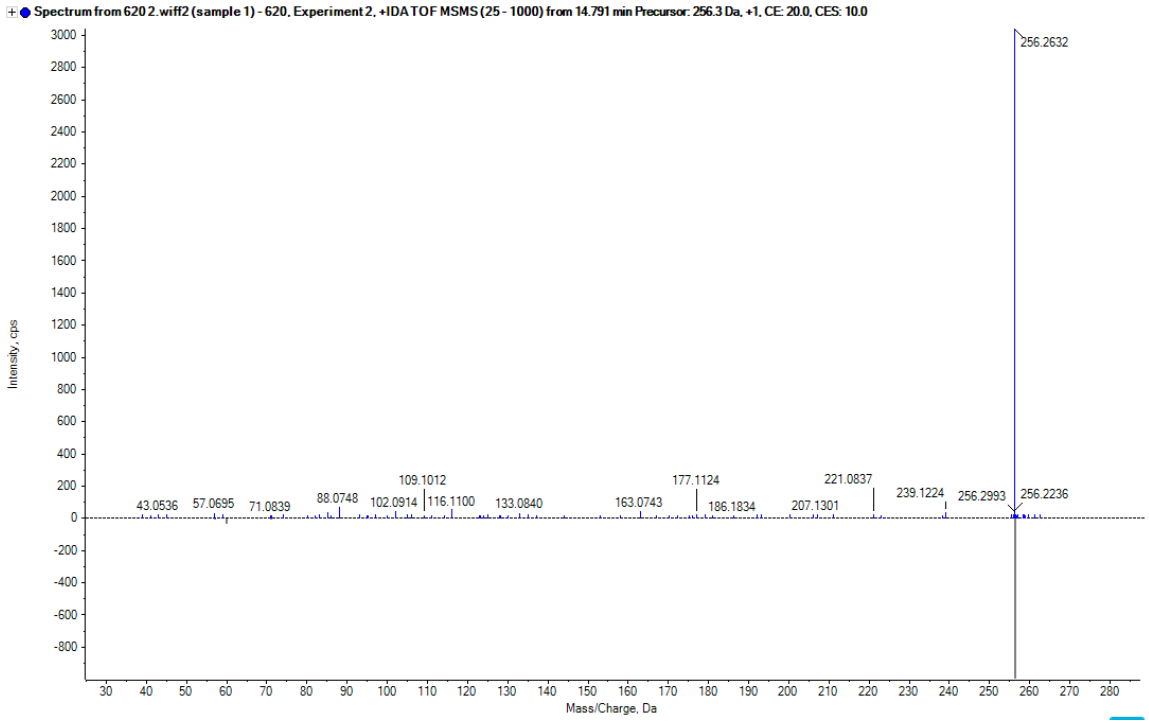
Extracted ion chromatogram



Precursor ion mass spectrum

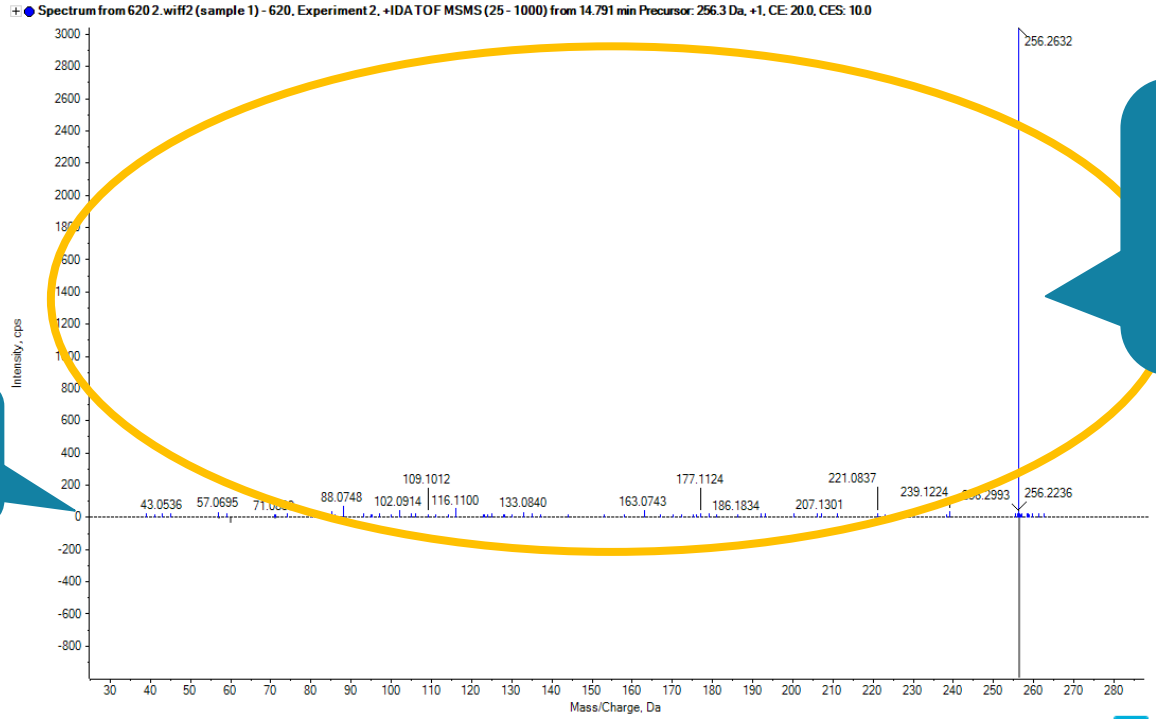
Compound C

Experimental MS/MS Data



MS/MS Spectrum

Compound C



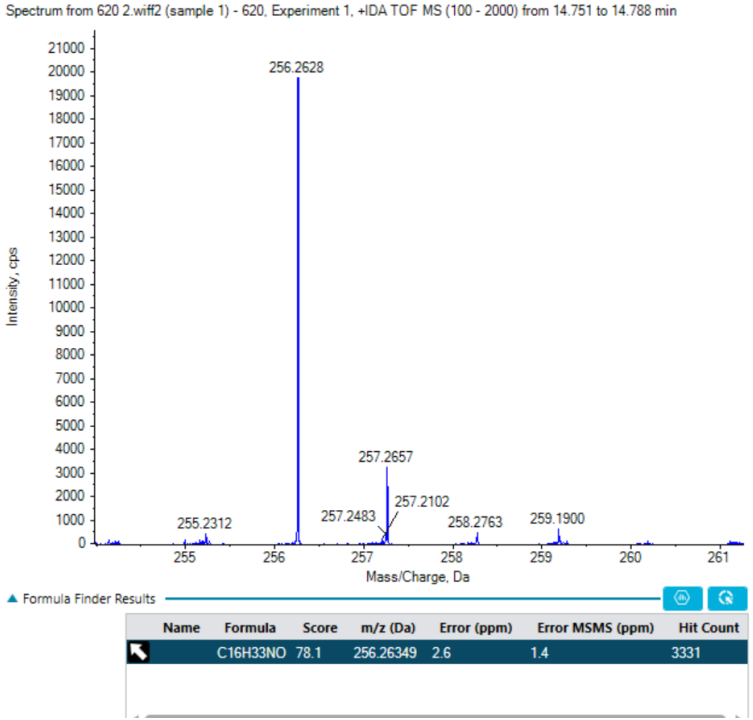
MS/MS spectrum lacking diagnostic fragmentation information

Poor library matching

MS/MS Spectrum

Compound C

FORMULA FINDER AND CHEMSPIDER



Compound C

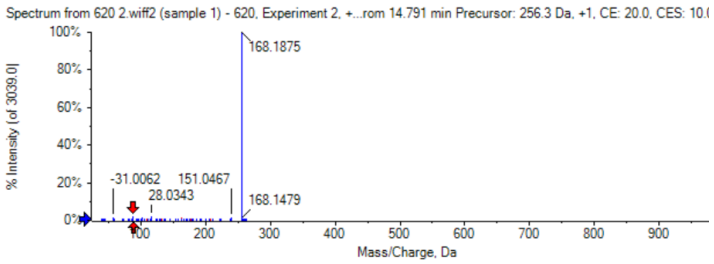
$C_{16}H_{33}NO$

ChemSpider results for: C16H33NO

1-40 of 3331

ChemSpider

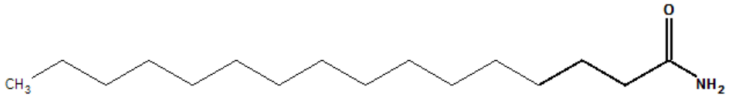
CSID	Common Name	Molecular Weight
62629	Palmitamide	255.4393
17736	N,N-Diethyldodecanamide	255.4393
66404	N-Laurylmorpholine	255.4393
68863	MFCD00026596	255.4393
84518	N,N-Dibutyloctanamide	255.4393
81853	N,N-Dipropyldecanamide	255.4393
3499401	MFCD00442362	255.43932



Display all Carbon Atoms

Options...

selected composition: $C_4H_{10}NO^+$ (88.0757 Da)



Options...

Fragments | Peaks

Mass/Charge	Intensity (%)	Assigned	Error (Da)
256.2632	100.00	☑	0.000
88.0748	2.30	☑	0.001
116.1100	1.74	☑	0.003
256.2236	1.61	☑	0.040
163.0743	1.48	☐	
102.0914	1.38	☑	0.000
256.2993	1.25	☑	0.036
239.1224	1.22	☑	0.115
57.0695	1.15	☑	0.000
85.1007	1.15	☑	0.001
133.0840	1.12	☐	

Matches: 20 of 26 peaks, 95.8% of total intensity

Compound C

CHEMSPIDER MATCHES

Palmitamide is used as an additive in tires and asphalt

ChemSpider results for: C16H33NO

1-40 of 3331

CSID	Common Name
62629	Palmitamide
17736	N,N-Diethyldodecanamide
66404	N-Laurylmorpholine
68863	MFCD00026596
84518	N,N-Dibutyloctanamide
81853	N,N-Dipropyldecanamide
3499401	MFCD00442362

4.791 min Precursor: 256.3 Da, +1, CE: 20.0, CES: 10.0

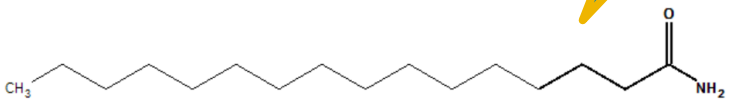
500 600 700 800 900

Mass/Charge, Da

Display all Carbon Atoms

Options...

selected composition: $C_4H_{10}NO^+$ (88.0757 Da)



Fragments | Peaks

Mass/Charge	Intensity (%)	Assigned	Error (Da)
256.2632	100.00	☑	0.000
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102.0914	1.38	☑	0.000
256.2993	1.25	☑	0.036
239.1224	1.22	☑	0.115
57.0695	1.15	☑	0.000
85.1007	1.15	☑	0.001
133.0840	1.12	☐	

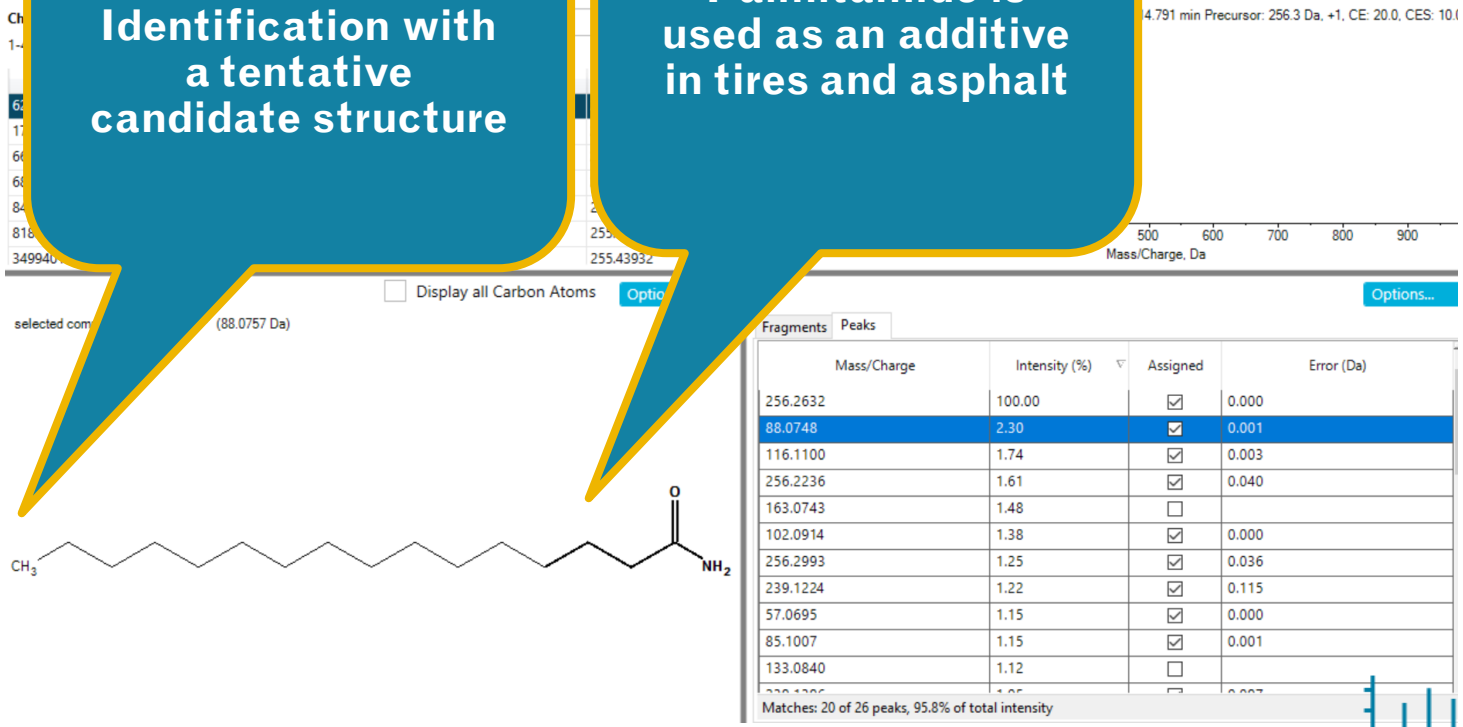
Matches: 20 of 26 peaks, 95.8% of total intensity

Compound C

CHEMSP

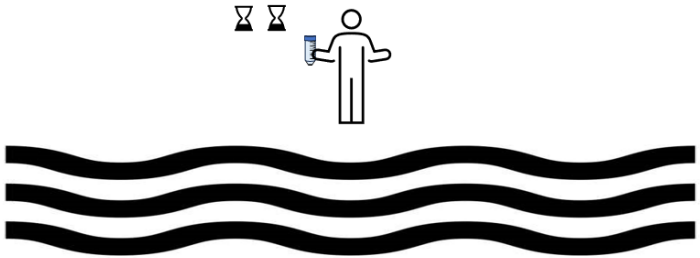
Level 3 Identification with a tentative candidate structure

Palmitamide is used as an additive in tires and asphalt



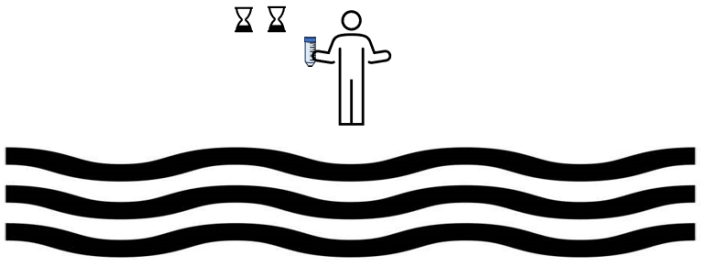
Summary

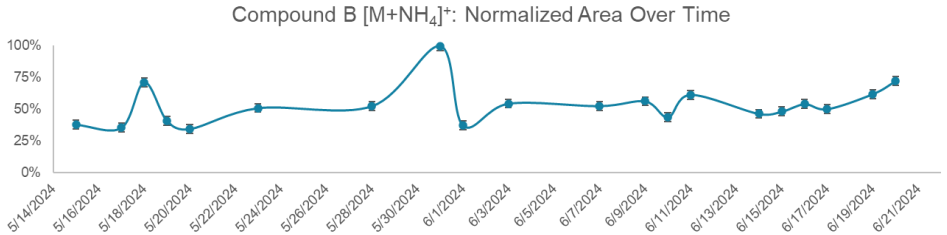
Spatio-temporal Monitoring and HRMS



Summary

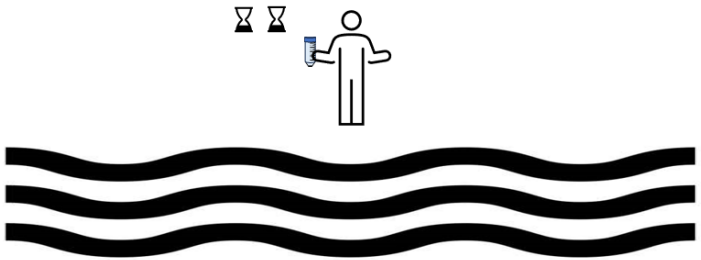
Spatio-temporal Monitoring And HRMS

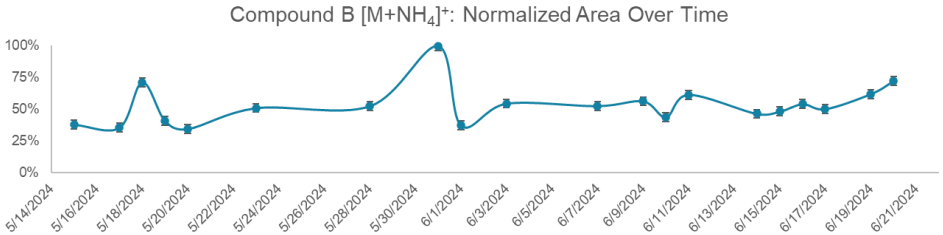


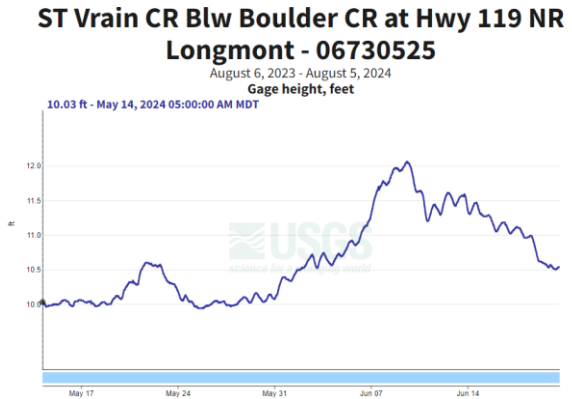


Summary

Spatio-temporal Monitoring And HRMS







Summary

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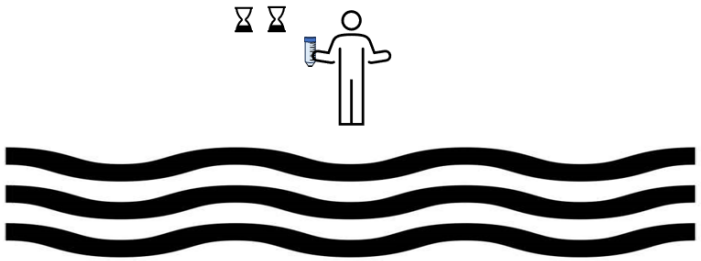

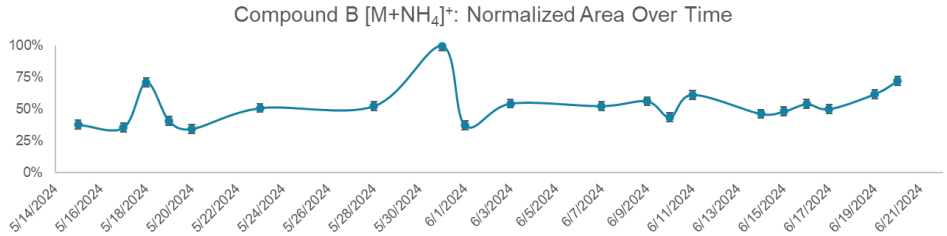

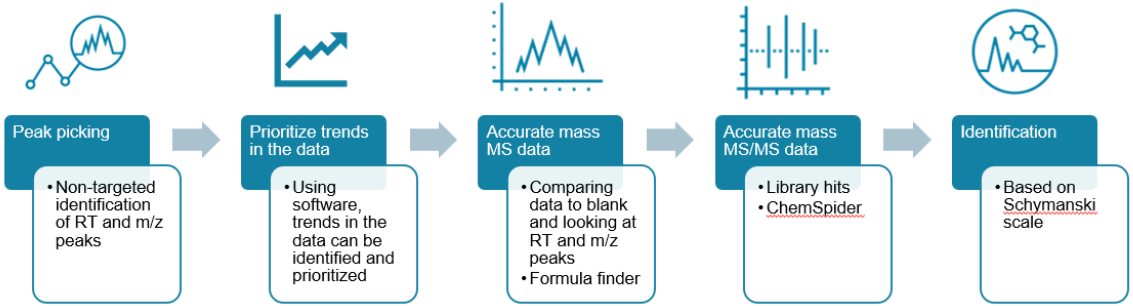

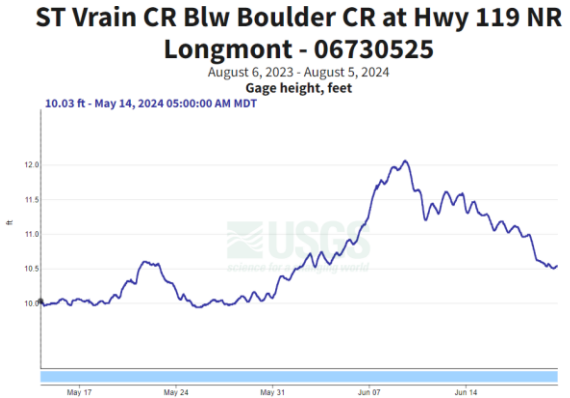

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