

Class on 02-03-20

1

Looking at the data with a 2D view

Formula

Ionization type

Charge

m/z tolerance m/z or ppm

Type of plot

Raw data files

Scans MS level: 1

m/z -

2

Set the run time for GenGlcA

Scan number -

Base Filtering Integer

Retention time - min.

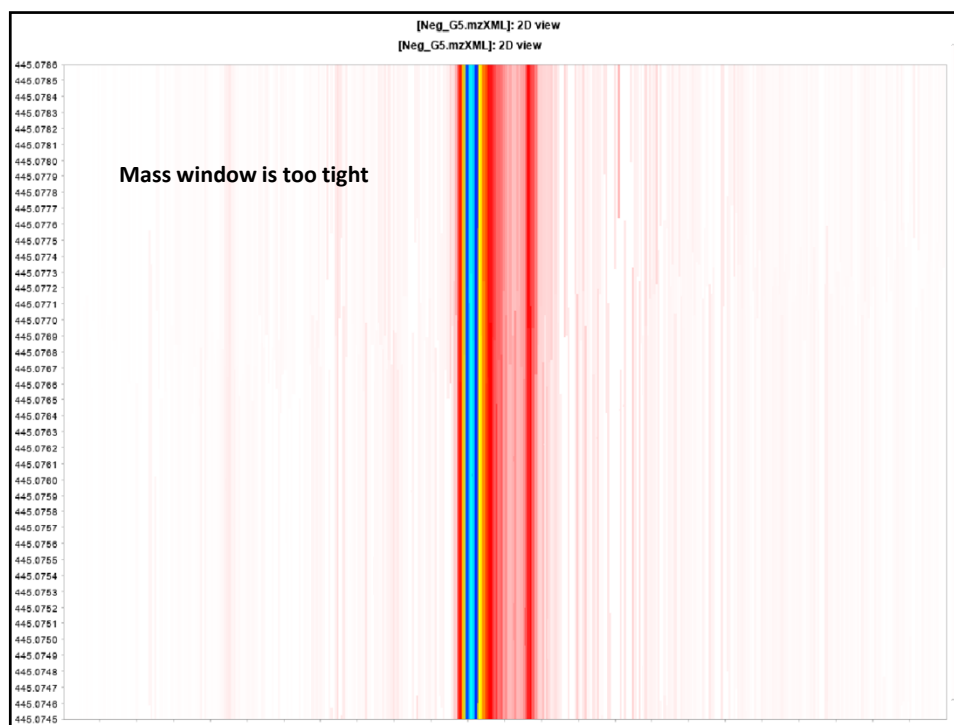
MS level

Scan definition

Polarity

Spectrum type

3



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Reset the retention time and mass ranges

Please set ranges for axes ✕

Retention time

Auto range

Minimum

Maximum

Auto tick size

Tick size

m/z

Auto range

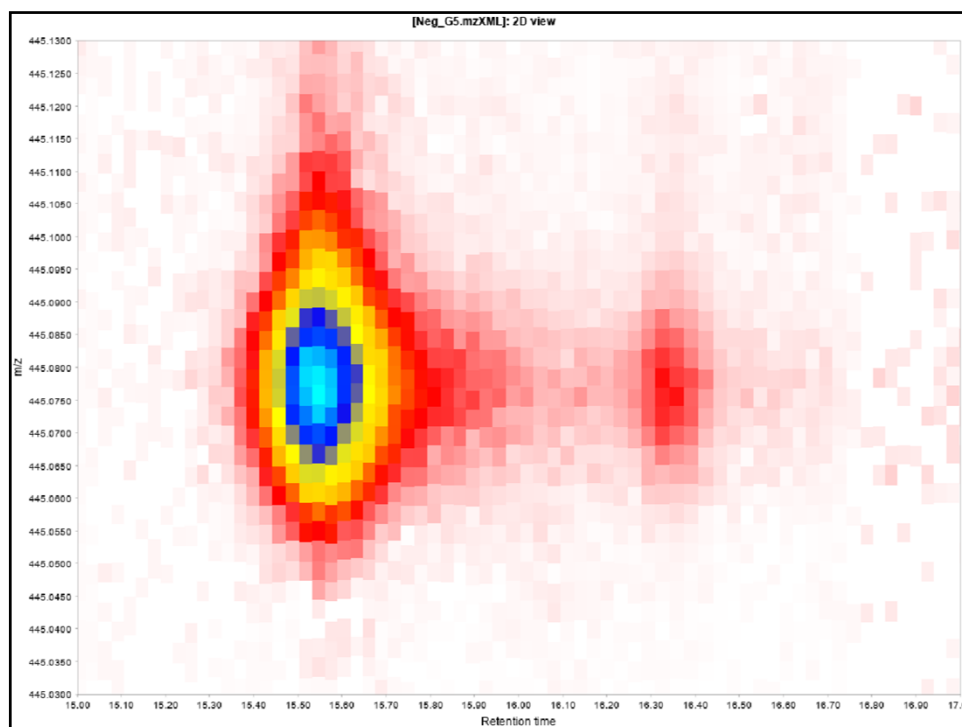
Minimum

Maximum

Auto tick size

Tick size

5



6

Select // and masses

The main dialog box contains the following fields and controls:

- Scan number: [] - []
- Base Filtering Integer: []
- Retention time: 10 - 22 min. [Auto range]
- MS level: 1
- Scan definition: []
- Polarity: -
- Spectrum type: Any
- Buttons: OK, Cancel
- Raw data files: 6 selected As selected in main window
- Scans: MS level: 1 [Set filters] [Clear filters]
- Mass detector: Centroid
- Mass list name: masses
- Output netCDF filename (optional): [] [last] []
- Buttons: OK, Cancel, Help

The secondary dialog box contains the following fields and controls:

- Noise level: 1e2
- Scale level: 5
- Wavelet window size (%): 30 %
- [] Show preview
- Buttons: OK, Cancel, Help

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The initial raw files now have masses

The screenshot shows a software interface with a menu bar (Project, Raw data methods, Feature list methods, Visualization, Tools, Windows, Help) and a list of raw data files. The list is organized into a tree view with expandable folders. A blue arrow points to the 'masses' column header in the list.

Key entries in the list include:

- #1654 @12.28 MS1 p -
- #1655 @12.30 MS2 (96.9631) p -
- #1656 @12.30 MS2 (303.0190) p -
- #1657 @12.31 MS2 (409.1716) p -
- #1658 @12.31 MS1 p -
- #1659 @12.33 MS2 (303.0193) p -
- #1660 @12.33 MS2 (391.1377) p -
- #1661 @12.33 MS2 (409.1723) p -
- #1662 @12.34 MS2 (443.1743) p -
- #1663 @12.34 MS2 (505.1346) p -
- #1664 @12.34 MS1 p -
- masses
- #1665 @12.36 MS2 (112.9883) p -
- #1666 @12.36 MS2 (174.9578) p -
- #1667 @12.37 MS2 (391.1388) p -
- #1668 @12.37 MS2 (505.1359) p -
- #1669 @12.38 MS1 p -
- #1670 @12.39 MS2 (112.9884) p -
- #1671 @12.40 MS2 (419.1493) p -
- #1672 @12.41 MS1 p -
- #1673 @12.42 MS2 (174.9579) p -
- #1674 @12.43 MS2 (218.0701) p -
- #1675 @12.43 MS2 (235.9279) p -
- #1676 @12.44 MS1 p -
- #1677 @12.45 MS2 (218.0699) p -
- #1678 @12.45 MS2 (235.9274) p -
- #1679 @12.45 MS2 (356.1831) p -
- #1680 @12.46 MS2 (423.2248) p -
- #1681 @12.46 MS2 (434.1595) c -

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Go to // and select ADAP chromatogram builder

First set the retention time range

Scan number -

Base Filtering Integer

Retention time - min.

MS level

Scan definition

Polarity

Spectrum type

9

Setting the parameters

Raw data files 6 selected As selected in main window

Scans -22.00 min.

Mass list masses

Min group size in # of scans

Group intensity threshold

Min highest intensity

m/z tolerance m/z or ppm

Suffix

ADAP Module Disclaimer:
If you use the ADAP Chromatogram Builder Module, please cite the [MZmine2 paper](#) and the following article:
[Myers OD, Sumner SJ, Li S, Barnes S, Du X. One Step Forward for Reducing False Positive and False Negative Compound Identifications from Mass Spectrometry Metabolomics Data: New Algorithms for Constructing Extracted](#)

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Mass chromatogram file content

The screenshot displays a software interface with a menu bar (Project, Raw data methods, Feature list methods, Visualization, Tools, Windows, Help) and two main panels. The left panel, titled 'Raw data files', lists six files: Neg_C4.mzXML, Neg_C5.mzXML, Neg_C6.mzXML, Neg_G4.mzXML, Neg_G5.mzXML, and Neg_G6.mzXML. The right panel, titled 'Neg_G6.mzXML chromatograms', lists 28 peaks with their retention times and m/z values.

Peak ID	m/z	Retention Time (min)
#1	61.9921	20.28
#2	61.9976	19.10
#3	62.0035	17.57
#4	62.0098	16.89
#5	62.0164	19.00
#6	62.0220	19.00
#7	62.0291	17.51
#8	68.9987	18.48
#9	69.0057	16.89
#10	69.0137	19.02
#11	69.0302	18.97
#12	79.9639	17.51
#13	81.9581	17.49
#14	91.0053	18.76
#15	91.0111	17.70
#16	92.0543	13.21
#17	96.9620	17.75
#18	106.0454	17.51
#19	107.0517	15.17
#20	112.9875	20.31
#21	113.0012	17.54
#22	113.0073	17.83
#23	113.0130	20.17
#24	113.0238	16.83
#25	113.9893	17.78
#26	114.0586	18.70
#27	116.0374	16.53
#28	116.0532	17.81

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Selecting files for chromatogram deconvolution

The screenshot displays the same software interface as slide 11. The 'Raw data files' panel on the left remains the same. The 'Feature lists' panel on the right now lists the same six files as in slide 11, but they are all highlighted in blue, indicating they have been selected for deconvolution.

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Setting parameters for deconvolution

S/N threshold: 5

S/N estimator: Intensity window SN

min feature height: 100

coefficient/area threshold: 30

Peak duration range: 0.01 - .8

RT wavelet range: 0.01 - 0.30

Show preview

ADAP Module Disclaimer:
If you use the ADAP Chromatogram Deconvolution Module, please cite the [MZmine2 paper](#) and the following article:
[Myers OD, Sumner SJ, Li S, Barnes S, Du X: One Step Forward for Reducing False Positive and False Negative Compound Identifications from Mass Spectrometry Metabolomics Data: New Algorithms for Constructing Extracted Ion Chromatograms and Detecting Chromatographic Peaks. Anal Chem 2017, DOI: 10.1021/acs.analchem.7b00947](#)

OK Cancel Help

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Content of the deconvoluted files

Project Raw data methods Feature list methods Visualization Tools Windows Help

Raw data files

- Neg_C4.mzXML
- Neg_C5.mzXML
- Neg_C6.mzXML
- Neg_G4.mzXML
- Neg_G5.mzXML
- Neg_G6.mzXML

Feature lists

- Neg_C4.mzXML chromatograms
- Neg_C5.mzXML chromatograms
- Neg_C6.mzXML chromatograms
- Neg_G4.mzXML chromatograms
- Neg_G5.mzXML chromatograms
- Neg_G6.mzXML chromatograms
- Neg_C6.mzXML chromatograms deconvoluted
- Neg_C4.mzXML chromatograms deconvoluted
- Neg_G5.mzXML chromatograms deconvoluted
- Neg_G4.mzXML chromatograms deconvoluted
- Neg_G6.mzXML chromatograms deconvoluted
- Neg_C5.mzXML chromatograms deconvoluted

#1 59.0170 m/z @17.25

#2 61.8035 m/z @19.08

#3 61.8035 m/z @20.35

#4 61.8034 m/z @16.08

#5 61.8063 m/z @16.58

#6 61.8066 m/z @17.83

#7 62.0070 m/z @19.03

#8 62.0165 m/z @19.03

#9 62.0165 m/z @11.20

#10 62.0198 m/z @11.20

#11 62.0313 m/z @17.33

#12 62.0369 m/z @18.95

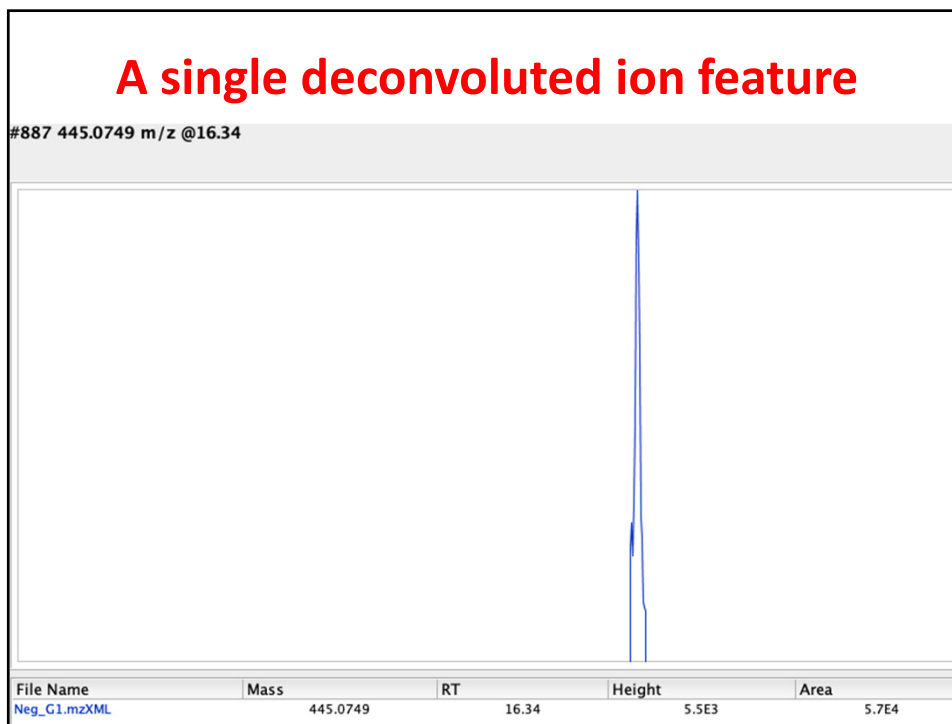
#13 62.0369 m/z @17.11

#14 62.0369 m/z @14.91

#15 62.0368 m/z @12.66

#16 62.0450 m/z @20.43

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Isotope grouping

Feature lists 6 selected As selected in main window

Name suffix deisotoped

m/z tolerance .005 m/z or 10 ppm

Retention time tolerance 0.5 absolute (min)

Monotonic shape

Maximum charge 1

Representative isotope Most intense

Remove original peaklist

OK Cancel Help

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De-isotoped files

Project Raw data methods Feature list methods Visualization Tools Windows Help

Raw data files

- Neg_C4.mzXML
- Neg_C5.mzXML
- Neg_C6.mzXML
- Neg_G4.mzXML
- Neg_G5.mzXML
- Neg_G6.mzXML

Feature lists

- Neg_C4.mzXML chromatograms
- Neg_C5.mzXML chromatograms
- Neg_C6.mzXML chromatograms
- Neg_G4.mzXML chromatograms
- Neg_G5.mzXML chromatograms
- Neg_G6.mzXML chromatograms
- Neg_C4.mzXML chromatograms deconvoluted
- Neg_C5.mzXML chromatograms deconvoluted
- Neg_C6.mzXML chromatograms deconvoluted
- Neg_G4.mzXML chromatograms deconvoluted
- Neg_G5.mzXML chromatograms deconvoluted
- Neg_G6.mzXML chromatograms deconvoluted
- Neg_C4.mzXML chromatograms deconvoluted deisotoped
- Neg_C5.mzXML chromatograms deconvoluted deisotoped
- Neg_C6.mzXML chromatograms deconvoluted deisotoped
- Neg_G4.mzXML chromatograms deconvoluted deisotoped
- Neg_G5.mzXML chromatograms deconvoluted deisotoped
- Neg_G6.mzXML chromatograms deconvoluted deisotoped

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Peak and chromatogram alignment

Feature lists 6 selected As selected in main window

Feature list name Aligned feature list

m/z tolerance .01 m/z or 10 ppm

RT tolerance 0.5 absolute (min)

RT tolerance after correction 0.3 absolute (min)

RANSAC iterations 0

Minimum number of points 90 %

Threshold value .5

Linear model

Require same charge state

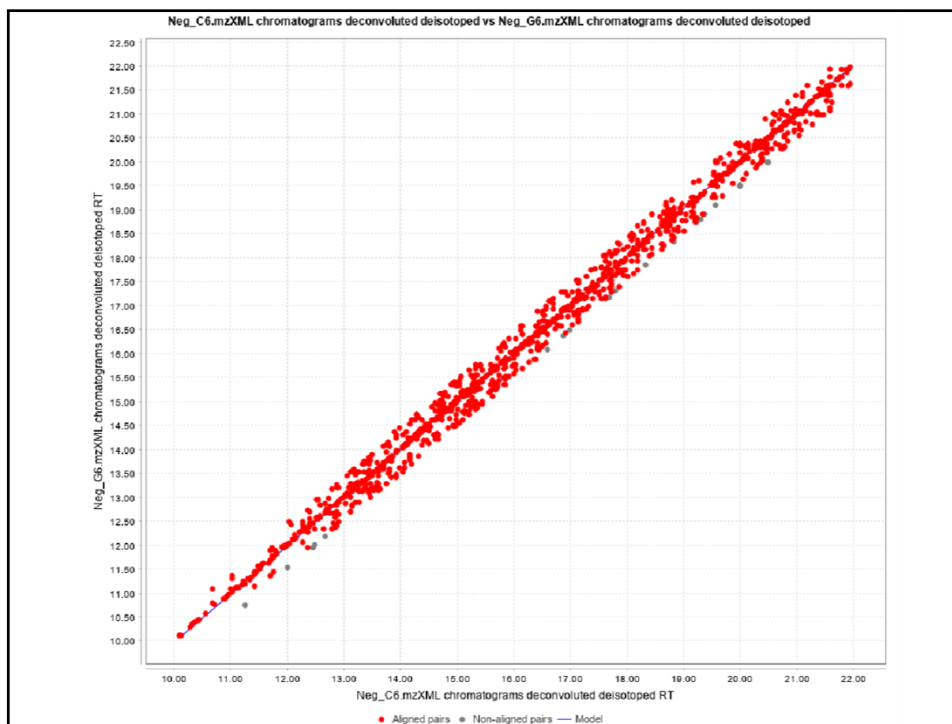
Show preview of RANSAC alignment

Neg_C6.mzXML chromatograms deconvoluted deisotoped

Neg_G6.mzXML chromatograms deconvoluted deisotoped

Preview alignment

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The aligned file appears

Project Raw data methods Feature list methods Visualization Tools Windows Help

Raw data files

- Neg_C4.mzXML
- Neg_C5.mzXML
- Neg_C6.mzXML
- Neg_G4.mzXML
- Neg_G5.mzXML
- Neg_G6.mzXML

Feature lists

- Neg_C4.mzXML chromatograms
- Neg_C5.mzXML chromatograms
- Neg_C6.mzXML chromatograms
- Neg_G4.mzXML chromatograms
- Neg_G5.mzXML chromatograms
- Neg_G6.mzXML chromatograms
- Neg_C4.mzXML chromatograms deconvoluted
- Neg_C5.mzXML chromatograms deconvoluted
- Neg_C6.mzXML chromatograms deconvoluted
- Neg_G4.mzXML chromatograms deconvoluted
- Neg_G5.mzXML chromatograms deconvoluted
- Neg_G6.mzXML chromatograms deconvoluted
- Neg_C4.mzXML chromatograms deconvoluted deisotoped
- Neg_C5.mzXML chromatograms deconvoluted deisotoped
- Neg_C6.mzXML chromatograms deconvoluted deisotoped
- Neg_G4.mzXML chromatograms deconvoluted deisotoped
- Neg_G5.mzXML chromatograms deconvoluted deisotoped
- Neg_G6.mzXML chromatograms deconvoluted deisotoped
- Aligned feature list

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Content of the aligned file

The screenshot shows a software interface with a menu bar (Project, Raw data methods, Feature list methods, Visualization, Tools, Windows, Help) and two main panels. The left panel, titled 'Raw data files', lists six files: Neg_C4.mzXML, Neg_C5.mzXML, Neg_C6.mzXML, Neg_G4.mzXML, Neg_G5.mzXML, and Neg_G6.mzXML. The right panel, titled 'Aligned feature list', displays a list of 28 features, each with a blue arrow icon, a feature ID, m/z value, and retention time (RT).

Feature ID	m/z	RT
#1	427.1796	18.84
#2	283.0826	14.80
#3	411.1264	11.12
#4	144.0468	17.13
#5	341.1242	16.78
#6	187.0077	15.14
#7	567.1694	14.73
#8	275.0956	18.67
#9	361.2011	20.27
#10	291.0911	17.07
#11	441.1583	15.09
#12	823.2590	11.12
#13	443.1738	13.67
#14	186.1147	17.60
#15	389.1563	21.39
#16	319.1221	18.43
#17	178.0520	13.20
#18	404.1913	13.32
#19	291.0906	16.43
#20	216.1249	16.53
#21	349.0020	18.09
#22	407.1006	12.69
#23	537.2321	18.45
#24	198.1141	18.66
#25	275.0967	18.18
#26	587.3053	17.35
#27	273.0922	13.43
#28	377.1963	16.76

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Initial version of the aligned table

The screenshot shows a software interface with a 'Windows' menu and a table. The table has columns for ID, m/z, RT, Identity, Comment, Peak shape, and Neg_C4.mzXML. The Neg_C4.mzXML column is further divided into Status, Height, Area, and Status. The table contains 13 rows of data, corresponding to the first 13 features from the aligned feature list.

ID	Average		Identity	Comment	Peak shape	Neg_C4.mzXML			
	m/z	RT				Status	Height	Area	Status
1	427.1796	18.84				●	2.4E5	2.1E6	●
2	283.0826	14.80				●	1.6E5	1.2E6	●
3	411.1264	11.12				●	1.3E5	8.4E5	●
4	144.0468	17.13				●	1.3E5	1.2E6	●
5	341.1242	16.78				●	1.2E5	9.6E5	●
6	187.0077	15.14				●	1.1E5	1.2E6	●
7	567.1694	14.73				●	1.1E5	5.3E5	●
8	275.0956	18.67				●	8.4E4	6.7E5	●
9	361.2011	20.27				●	8.3E4	7.0E5	●
10	291.0911	17.07				●	7.5E4	6.6E5	●
11	441.1583	15.09				●	6.7E4	4.7E5	●
12	823.2590	11.12				●	6.5E4	3.0E5	●
13	443.1738	13.67				●	6.5E4	5.8E5	●

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Locating Gen GlcA in the table

ID	Average		Peak shape	Neg_C4.mzXML			Neg_C5.mzXML			Neg_C6.mzXML			Neg_G4.mzXML			Neg_G5.mzXML			Neg_G6.mzXML		
	m/z	RT		Status	Height	Area	Status	Height	Area	Status	Height	Area	Status	Height	Area	Status	Height	Area	Status	Height	Area
17013	445.0425	14.46		●			●			●			●			●	2.8E3	1.4E4	●		
21846	445.0456	15.94		●			●			●			●			●			●	1.1E2	7.2E2
3240	445.0581	13.58		●	1.6E2	1.1E3	●			●			●			●			●		
4815	445.0581	13.89		●	1.1E2	4.2E2	●			●			●			●			●		
19074	445.0758	20.38		●			●			●			●			●	1.3E2	5.4E2	●		
12253	445.0766	16.33		●			●			●			●	2.8E4	2.1E5	●	2.1E3	1.1E4	●	3.6E3	2.4E4
12247	445.0772	16.58		●			●			●			●	1.1E5	1.3E5	●			●	8.7E4	9.0E5
21281	445.0786	13.76		●			●			●			●			●			●	1.5E2	5.5E2
8000	445.1309	17.77		●			●	1.1E2	9.1E2	●			●			●			●		
6531	445.1328	10.91		●			●	2.4E2	7.8E2	●			●			●	9.1E2	5.6E3	●		
6667	445.1666	15.50		●			●	2.1E2	1.2E3	●			●			●			●		
7268	445.1668	18.26		●			●	1.5E2	7.3E2	●			●			●			●		
6685	445.1669	16.17		●			●	2.1E2	1.3E3	●			●			●			●		
20049	445.1705	14.51		●			●			●			●			●			●	1.7E3	9.6E3
20018	445.1884	13.57		●			●			●			●			●			●	4.0E3	2.1E4
20123	445.1884	16.09		●			●			●			●			●			●	9.1E2	7.2E3
21267	445.1884	18.32		●			●			●			●			●			●	1.5E2	6.1E2
20252	445.1884	17.32		●			●			●			●			●			●	5.5E2	4.8E3
17235	445.1916	14.57		●			●			●			●			●	7.5E2	3.6E3	●	1.3E3	8.4E3
17140	445.1948	14.61		●			●			●			●			●	7.5E2	3.6E3	●		
20335	445.2064	18.05		●			●			●			●			●			●	4.1E2	2.1E3

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Table ordered to ions found in Genistein-treated samples

ID	Average		Peak shape	Neg_C4.mzXML			Neg_C5.mzXML			Neg_C6.mzXML			Neg_G4.mzXML			Neg_G5.mzXML			Neg_G6.mzXML		
	m/z	RT		Status	Height	Area	Status	Height	Area	Status	Height	Area	Status	Height	Area	Status	Height	Area	Status	Height	Area
4	144.0468	17.13		●	1.3E5	1.2E6	●	6.6E4	7.0E5	●	8.3E4	8.8E5	●	4.9E4	4.9E5	●	8.7E4	7.9E5	●	7.5E4	7.4E5
5	341.1242	16.78		●	1.2E5	9.6E5	●	1.3E5	1.1E6	●	8.6E3	4.7E4	●	1.6E5	1.3E6	●	3.7E4	2.3E5	●	8.2E4	5.3E5
6	187.0077	15.14		●	1.1E5	1.2E6	●	3.6E4	3.4E5	●	8.0E4	8.7E5	●	3.6E4	3.5E5	●	8.7E4	9.1E5	●	5.1E4	4.9E5
9	361.0011	20.27		●	8.3E4	7.0E5	●	5.2E4	2.5E5	●	1.6E5	1.2E6	●	1.6E5	1.1E6	●	2.6E4	1.2E5	●	5.3E4	2.2E5
10	291.0911	17.07		●	7.5E4	6.6E5	●	3.2E4	2.9E5	●	2.6E4	2.1E5	●	9.8E4	7.2E5	●	1.8E4	1.1E5	●	2.9E4	2.7E5
14	186.1147	17.80		●	6.3E4	4.5E5	●	4.0E4	2.8E5	●	8.0E4	6.5E5	●	1.1E4	8.6E4	●	2.0E4	1.5E5	●	3.0E4	2.1E5
16	319.1221	18.43		●	5.8E4	3.6E5	●	5.4E4	6.4E5	●	2.4E4	1.6E5	●	6.3E4	7.0E5	●	1.1E4	1.5E5	●	2.6E4	3.1E5
17	178.0520	13.20		●	5.8E4	4.1E5	●	6.7E4	5.2E5	●	1.0E4	8.1E4	●	7.6E4	6.1E5	●	3.7E4	2.8E5	●	6.9E4	5.0E5
18	404.1913	13.32		●	5.7E4	3.4E5	●	2.7E4	1.6E5	●	6.6E4	2.7E5	●	2.7E4	1.3E5	●	1.1E4	6.6E4	●	2.1E4	1.4E5
19	291.0906	16.43		●	5.7E4	4.4E5	●	4.8E4	2.6E5	●	2.8E4	1.5E5	●	6.3E4	4.0E5	●	1.7E4	1.2E5	●	3.1E4	2.0E5
20	216.1249	16.53		●	5.6E4	4.0E5	●			●	9.9E4	8.7E5	●	2.2E4	1.5E5	●	2.8E4	2.1E5	●	2.2E4	1.6E5
23	537.2321	18.45		●	4.8E4	3.0E5	●	3.5E4	2.5E5	●	1.3E5	1.1E6	●	1.0E5	7.7E5	●	1.5E4	1.0E5	●	3.8E4	2.5E5
25	275.0967	18.18		●	4.3E4	3.4E5	●	6.1E4	4.5E5	●	1.0E5	9.6E5	●	4.7E4	3.6E5	●	3.0E4	1.5E5	●	1.4E4	8.9E4
30	242.1402	14.50		●	3.5E4	1.9E5	●	2.6E4	1.7E5	●	5.1E4	2.8E5	●	2.7E4	2.2E5	●	2.5E4	1.2E5	●	2.7E4	1.9E5
32	421.1353	13.04		●	3.4E4	1.9E5	●	3.1E4	2.2E5	●	2.6E4	1.8E5	●	3.9E4	2.7E5	●	2.9E4	2.1E5	●	3.2E4	2.1E5
34	172.0987	15.72		●	3.2E4	2.5E5	●	1.3E4	1.0E5	●	5.8E4	5.1E5	●	5.3E3	4.0E4	●	1.3E4	9.7E4	●	1.5E4	1.2E5
36	375.1304	13.03		●	3.1E4	2.0E5	●			●	2.4E4	1.7E5	●	3.6E4	2.9E5	●	3.1E4	2.0E5	●		
39	212.0033	13.36		●	3.0E4	3.1E5	●	2.1E4	1.9E5	●	1.6E4	1.7E5	●	1.9E4	1.6E5	●	2.8E4	2.6E5	●	2.9E4	2.6E5
43	204.0674	16.06		●	2.7E4	2.4E5	●	2.5E4	2.3E5	●	2.3E4	1.9E5	●	1.6E4	1.4E5	●	1.7E4	1.4E5	●	2.1E4	1.8E5
44	415.1969	19.14		●	2.7E4	1.1E5	●			●	3.9E4	2.4E5	●	1.1E4	6.1E4	●	6.1E3	2.1E4	●	1.1E4	7.4E4
46	317.1238	14.46		●	2.4E4	1.8E5	●	1.5E4	1.1E5	●	3.8E4	2.3E5	●	1.9E4	1.3E5	●	1.8E4	8.6E4	●	1.4E4	8.9E4

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Exporting the aligned table

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Class exercise

- Load the C4-C6 and G4-G6 .mzxml files
- Locate the ions that have the ion (in negative) for p-ethylphenol glucuronide ($C_{14}H_{18}O_7$) and p-ethylphenol sulfate ($C_8H_{10}O_4S$) - what are their m/z values?
 - Get MSMS spectra of each one
- Identify all the *masses* in each file — from these generate chromatograms, and then deconvolute the chromatograms
- Output the data into a .csv file (choose row ID, m/z , retention time, peak height, peak area and FWHM)
- Sort the file by retention time – identify ions that are co-eluting and are isotopes.

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Export to .CSV file

<input checked="" type="checkbox"/> Export row ID	<input type="checkbox"/> Peak status
<input type="checkbox"/> Export row m/z	<input checked="" type="checkbox"/> Peak m/z
<input type="checkbox"/> Export row retention time	<input checked="" type="checkbox"/> Peak RT
<input type="checkbox"/> Export row identity (main ID)	<input type="checkbox"/> Peak RT start
<input type="checkbox"/> Export row identity (all IDs)	<input type="checkbox"/> Peak RT end
<input type="checkbox"/> Export row identity (main ID + details)	<input type="checkbox"/> Peak duration time
<input type="checkbox"/> Export row comment	<input checked="" type="checkbox"/> Peak height
	<input checked="" type="checkbox"/> Peak area
	<input type="checkbox"/> Peak charge
	<input type="checkbox"/> Peak # data points
	<input checked="" type="checkbox"/> Peak FWHM
	<input type="checkbox"/> Peak tailing factor
	<input type="checkbox"/> Peak asymmetry factor

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