you-try-it-10answers.xlsx

ver. 9/20/2018

For use with:

Brian M. Tissue, *Basics of Analytical Chemistry and Chemical Equilibria,* (John Wiley: New York, 2013).

http://www.achem.org

Worksheets in this file

notes	This page with background information.
10.A TLC	Calculating R_F to identify components in a mixture.
10.B quantitation	Determining unknown concentrations with an internal standard.
10.C resolution	Resolution of chromatographic peaks.
10.D mass-spec	Fingerprint analysis of mass spectra.

Background

Refer to Chapter 10 in the text for equations and explanations.

Each worksheet has instructions in the blue shaded box.

For step-by-step help see you-try-it-10guide.pdf.

You-Try-It 10.A **TLC**

Table 10.A.1 lists the spot positions for a standard solution and Table 10.A.2 lists results for an unknown mixture.

1. Calculate R_F for each spot in the reference mixture.
2. Calculate R_F for each spot in the unknown mixture and identify analytes.

Table 10.A.1 Reference

spot	analyte	migration distance (cm)	R_F
1	unknown	0.2	0.03
2	unknown	0.4	0.06
3	tyrosine	0.6	0.10
4	lysine	1.1	0.18
5	glycine	2.0	0.32
6	alanine/tryptophan	2.6	0.42
7	methionine	3.0	0.48
8	phenylanlanine	3.5	0.56
9	valine	3.8	0.61
10	isoleucine	4.3	0.69
11	leucine	4.5	0.73
12	proline	5.3	0.85
	solvent front	6.2	--

Table 10.A.2 Unknown mixture

spot	migration distance (cm)	R_F	analyte
1	0.4	0.06	unknown
2	1.1	0.15	unknown
3	3.4	0.48	methionine
4	4.9	0.69	isoleucine
5	5.2	0.73	leucine
6	6.0	0.85	unknown
	7.1		solvent front

data adapted from: Poole, C. F.; Poole, S. K. *Analytical Chemistry* , **1989**, *61* , 12571.

You-Try-It 10.B Quantitation with Internal Standards

Table 10.B.1 lists the retention times and integrated areas for a standard mixture.
Table 10.B.2 and 10.B.3 lists the retention times and integrated areas for two unknown mixtures.

1. Use the peak area data in Table 10.B.1 to determine sensitivity factors for each analyte. The standard mixture contained 2000 ppm of each analyte in 40:60 ethanol-water solvent.
2. Use the sensitivity factors and the data in Tables 10.B.2 and 10.B.3 to determine the concentration of each analyte. The internal standard is spiked in at 2000 ppm.

Table 10.B.1

Peak	Retention Time (min)	identity	Concentration (ppm)	Peak Area (µV * s)	Relative Sensitivity
1	1.51	methanol	2000	2370	0.653
2	1.79	ethanol	40%	949746	---
3	1.97	2-propanol	2000	3853	1.062
4	2.46	1-propanol	2000	4073	1.123
5	3.15	ethyl acetate	2000	1424	0.393
6	3.34	2-methyl-1-propanol	2000	4291	1.183
7	3.98	1-butanol (IS)	2000	3628	1.000
8	5.45	3-methyl-1-butanol	2000	4428	1.221
9	5.51	2-methyl-1-butanol	2000	3812	1.051

Table 10.B.2 Gas chromatographic data for a bourbon whiskey spiked with 2000-ppm IS.

Peak	Retention Time (min)	identity	Peak Area (µV * s)	Relative Sensitivity	Concentration (ppm)
1	1.54	methanol	83.5	0.653	46.7
2	1.85	ethanol	1943410	---	---
3		2-propanol	ND	1.062	ND
4	2.49	1-propanol	131.9	1.123	43.0
5		ethyl acetate	213.8	0.393	199.2
6		2-methyl-1-propanol	832.5	1.183	257.4
7		1-butanol (IS)	5469.7	1.000	2000.0
8		3-methyl-1-butanol	2724.7	1.221	816.3
9	1.97	2-methyl-1-butanol	1122.2	1.051	390.5

Table 10.B.3 Gas chromatographic data for an Irish whiskey spiked with 2000-ppm IS.

Peak	Retention Time (min)	identity	Peak Area (µV * s)	Relative Sensitivity	Concentration (ppm)
1	1.56	methanol	83.5	0.65	46.7
2	1.85	ethanol	1680398	---	---
3	---	2-propanol	ND	1.06	ND
4	2.5	1-propanol	231.8	1.12	75.5
5	3.17	ethyl acetate	47.1	0.39	43.9
6	3.37	2-methyl-1-propanol	103.6	1.18	32.0
7	3.99	1-butanol (IS)	5267.7	1.00	2000.0
8	5.46	3-methyl-1-butanol	323	1.22	96.8
9	5.52	2-methyl-1-butanol	119.9	1.05	41.7

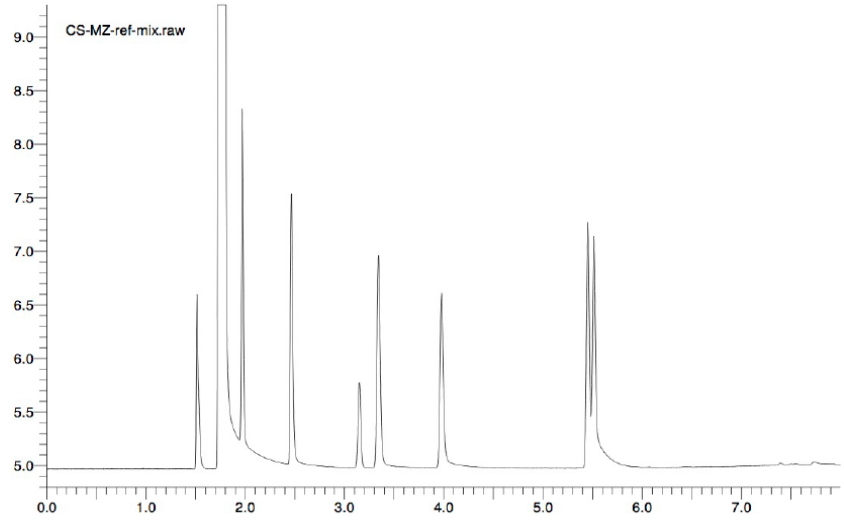


Figure 10.B.1. Chromatogram of 2000-ppm standard solution.

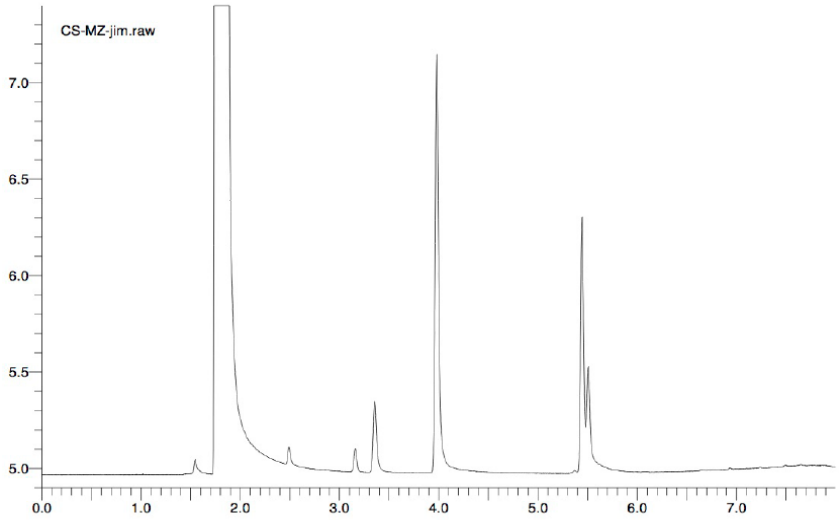


Figure 10.B.2. Chromatogram of a whiskey spiked with 2000-ppm internal standard.

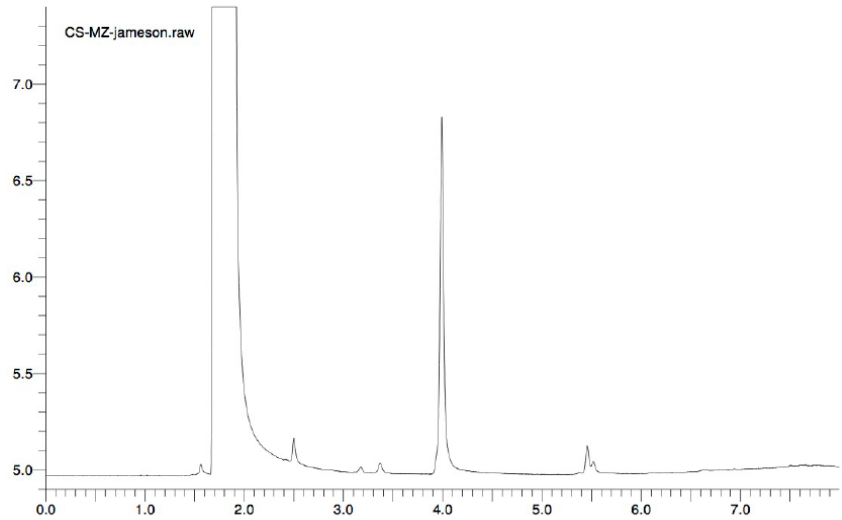


Figure 10.B.2. Chromatogram of a whiskey spiked with 2000-ppm internal standard.

You-Try-It 10.C **Chromatographic Resolution**

Tables 10.C.1 and 10.C.2 list retention times and peak widths for the chromatograms below each table.

1. Calculate the resolution of the close pairs in each chromatogram.

Table 10.C.1 GC peak parameters for a column with *N* = 1500.

Peak	Retention Time (min)	Base Width (min)	*R*
1	1.4	0.24	
2	1.8	0.26	1.60
3	3	0.45	
4	3.4	0.48	0.86
5	4.5	0.52	
6	4.9	0.52	0.77

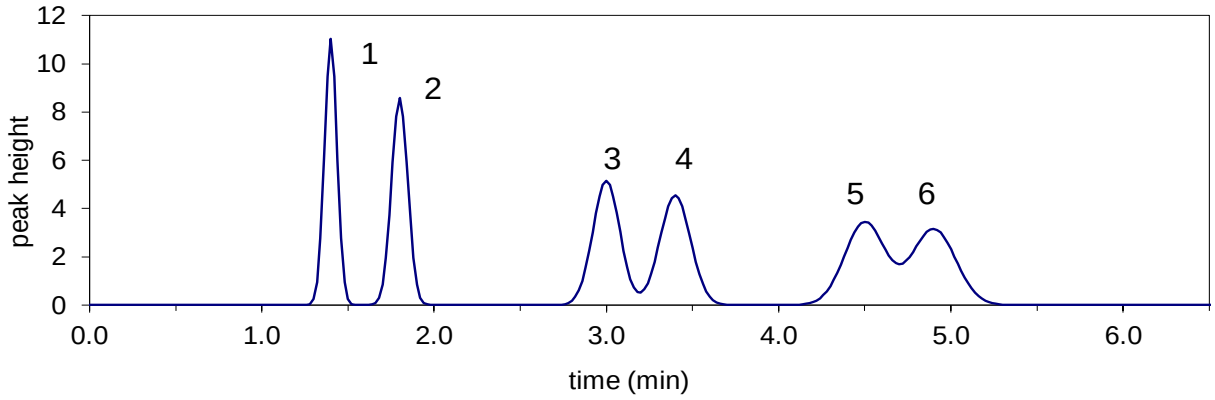
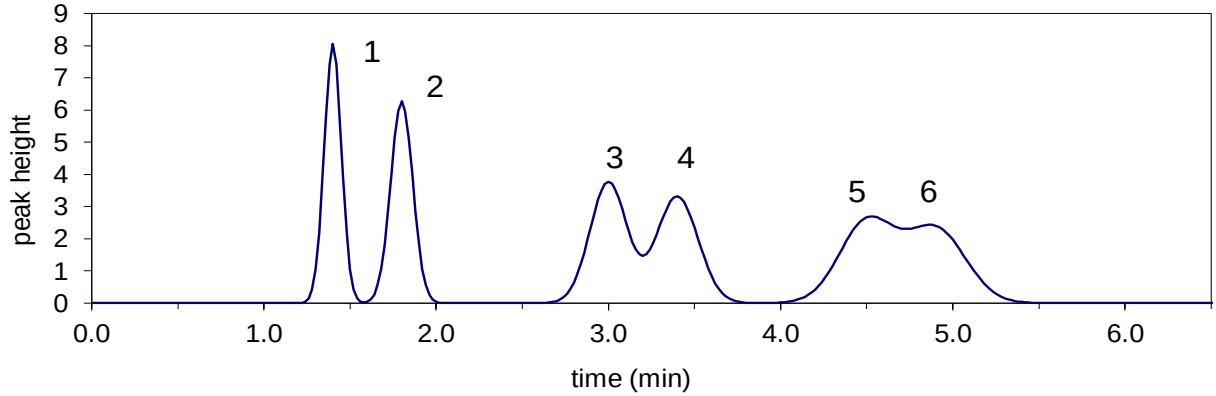


Table 10.C.2 GC peak parameters for a column with *N* = 800.

Peak	Retention Time (min)	Base Width (min)	*R*
1	1.4	0.26	
2	1.8	0.28	1.48
3	3	0.5	
4	3.4	0.51	0.79
5	4.5	0.7	
6	4.9	0.7	0.57

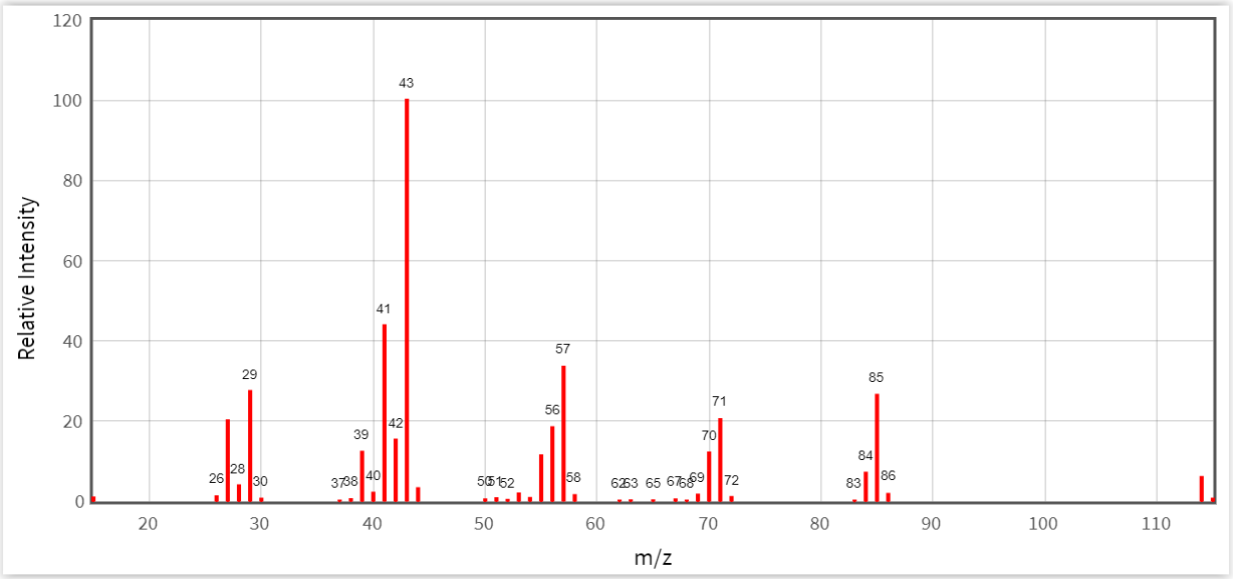


You-Try-It 10.D **Mass Spectrometry**

This worksheet contains mass spectra of several analytes.

1. Predict the mass of the molecular ion for each analyte.
   Comment on the intensity of this peak in the mass spectrum.
2. Predict the chemical formula for the three most abundant peaks in each mass spectrum.
3. Calculate the expected ratio for the following peaks.
   benzene 78 and 79
   chlorobenzene 112 and 114
   benzoic acid 122, 123, and 124

		molecular ion mass:		C	H	O
octane	C_8H_{18}	114	Da	8	18	



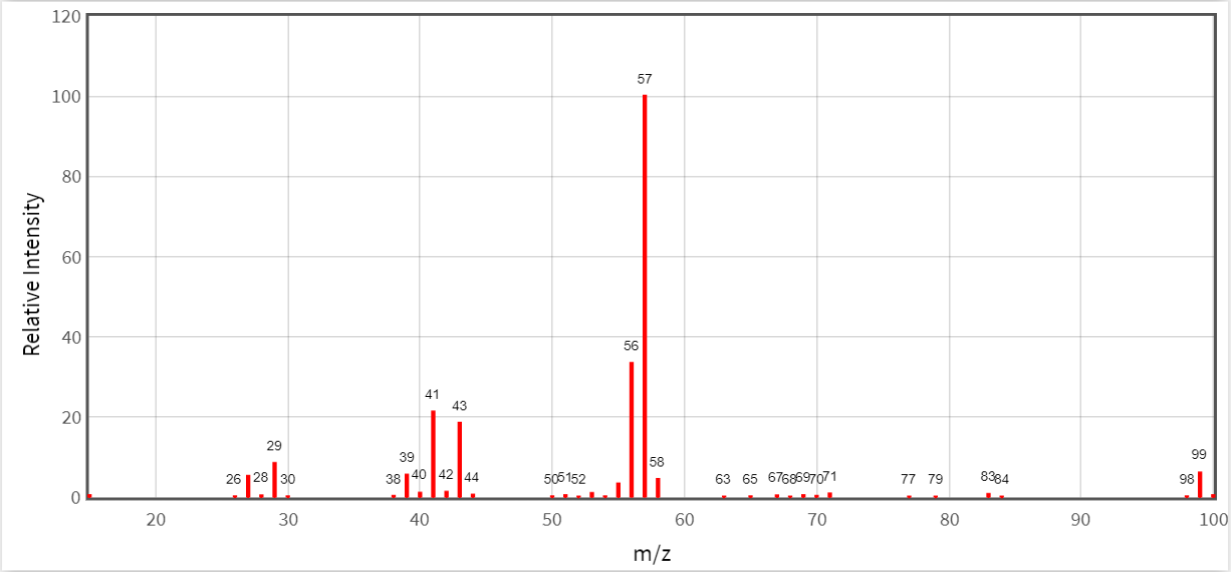
comments:
Mass 114 peak is small due to fragmentation.

Three most abundant masses:

1	43	$[C_3H_7]^+$
2	41	$[C_3H_5]^+$
3	57	$[C_4H_9]^+$

You will see a recurring pattern for alkanes.
For octane, note the sequence 29, 43, 57, 71, and 85.
These are alkyl ions that differ by a CH_2 fragment, i.e., 14 Da.

		mass:	C	H	O
isooctane	C_8H_{18}	114	8	18	
(2,2,4-trimethylpentane)					

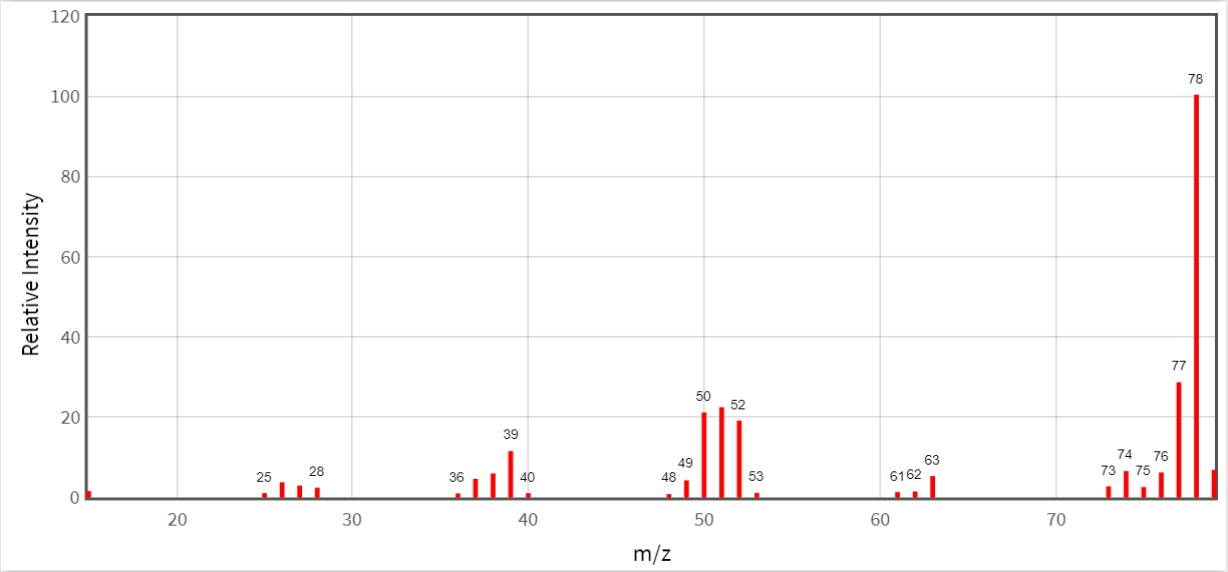


comments:
Mass 114 peak is not observed due to fragmentation.

Three most abundant masses:

1	57	$[C_4H_9]^+$
2	56	$[C_4H_8]^+$
3	41	$[C_3H_5]^+$

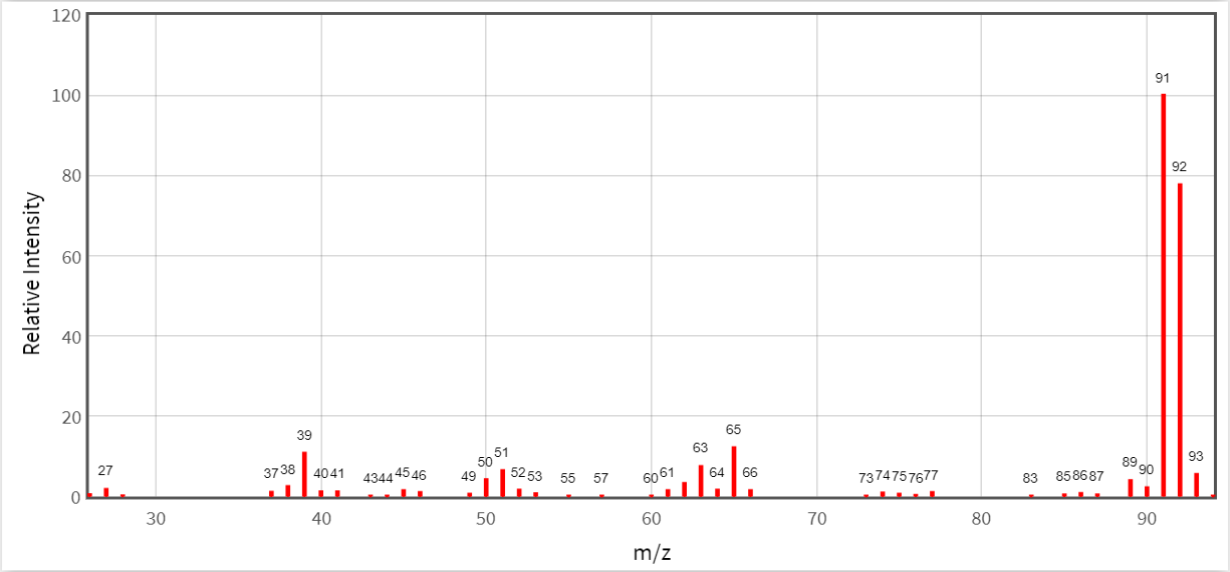
		mass:	C	H	O
benzene	C_6H_6	78	6	6	



comments:
Mass 78 peak is the most abundant peak in the spectrum.

Three most abundant masses:			SDBS-Mass peak data	
1	78	$[C_6H_6]^+$	77.0	15.9
2	77	$[C_6H_5]^+$	78.0	100.0
3	51	$[C_4H_3]^+$	79.0	6.7

		mass:	C	H	O
toluene	C_7H_8	92	7	8	



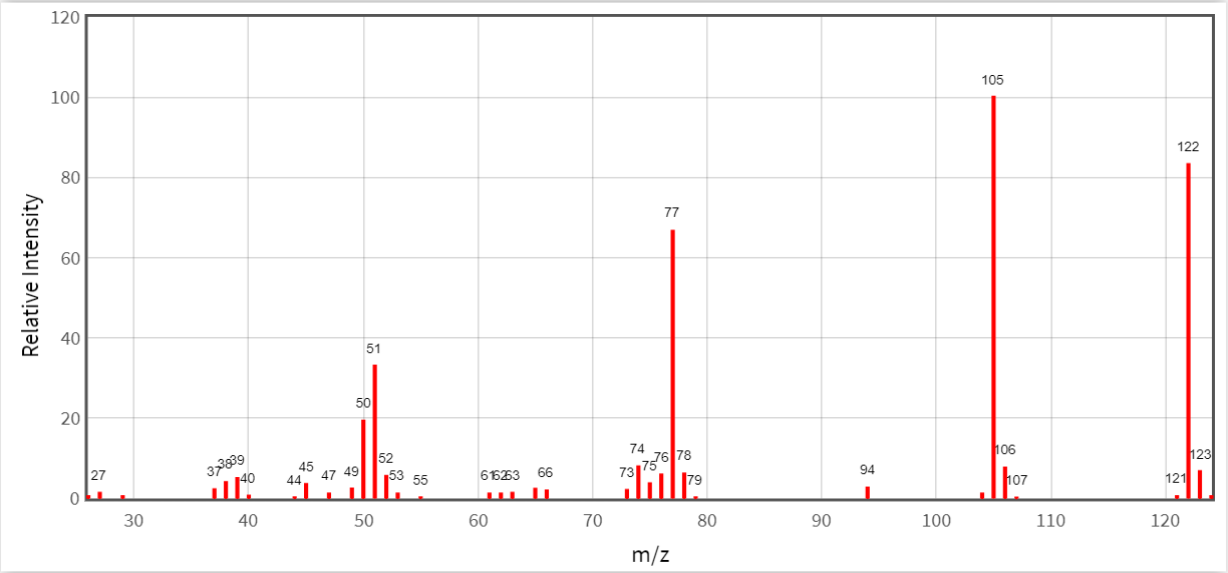
comments:
Mass 92 peak is the second most abundant peak in the spectrum.

Three most abundant masses:

1	91	$[C_7H_7]^+$
2	92	$[C_7H_8]^+$
3	65	$[C_5H_5]^+$

The mass 91 ion is an interesting case. It is the tropylium ion, which is the toluene structure rearranged in a 7-carbon ring.

		mass:	C	H	O
benzoic acid	$C_7H_6O_2$	122	7	6	2



comments:
Mass 122 peak is the second most abundant peak in the spectrum.

Three most abundant masses:

1	105	$[C_7H_5O]^+$
2	122	$[C_7H_6O_2]^+$
3	77	$[C_6H_5]^+$

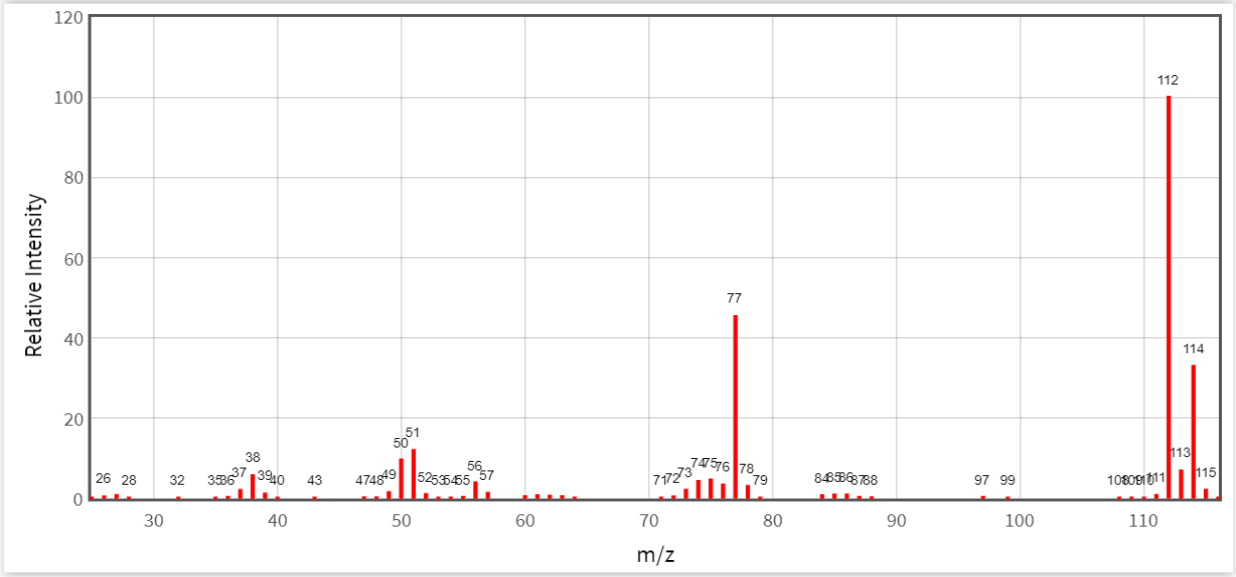
In many cases it is easier to identify the fragment that is lost
rather than predicting the fragment that is observed.
For the mass 105 and 77 peaks:

122 − 105 = 17

122 − 77 = 45

The mass 17 fragment could be the loss of OH, leaving C7H5O.
The mass 45 fragment could be the loss of COO, leaving C5H5.

		mass:	C	H	Cl
chlorobenzene	C_6H_5Cl	112	6	5	1



comments:
Mass 112 peak is the most abundant peak in the spectrum.

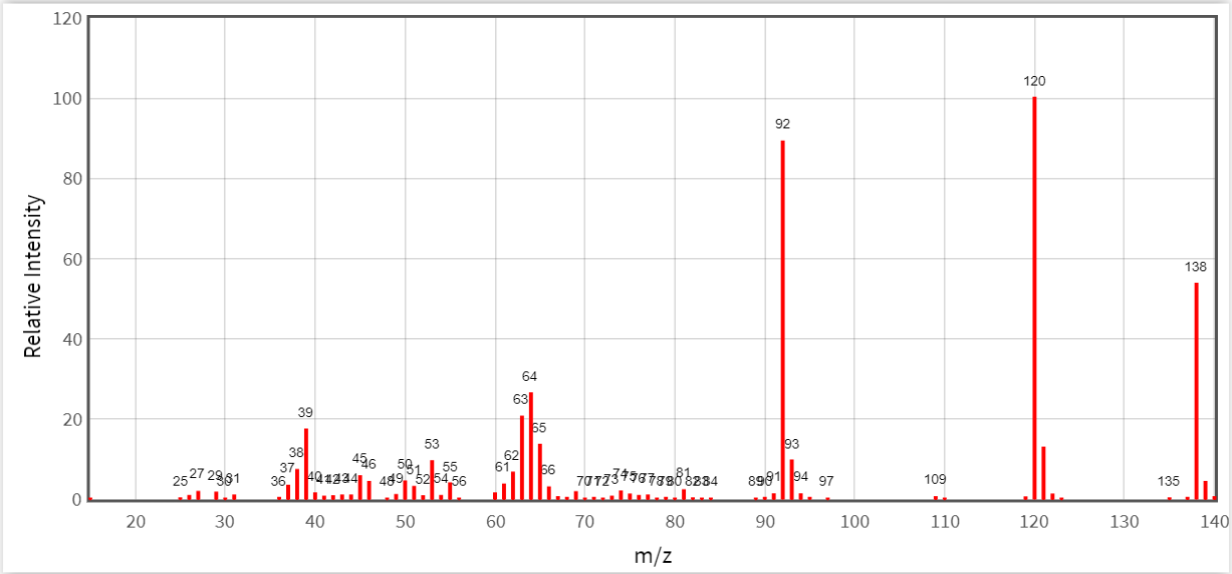
Three most abundant masses:

1	112	$[C_6H_5Cl]^+$	35Cl
2	77	$[C_6H_5]^+$	
3	114	$[C_6H_5Cl]^+$	37Cl

The mass 114 peak is approximately 1/3 the height of the mass 114 peak.
Anytime you see this ratio at a separation of 2 mass units,
you can suspect a Cl in the structure.
The natural abundance of Cl35 to Cl37 is 75.8:24.2, a ratio of:

3.13

		mass:	C	H	O
salicylic acid	$C_7H_6O_3$	138	7	6	3
(2-Hydroxybenzoic acid)					

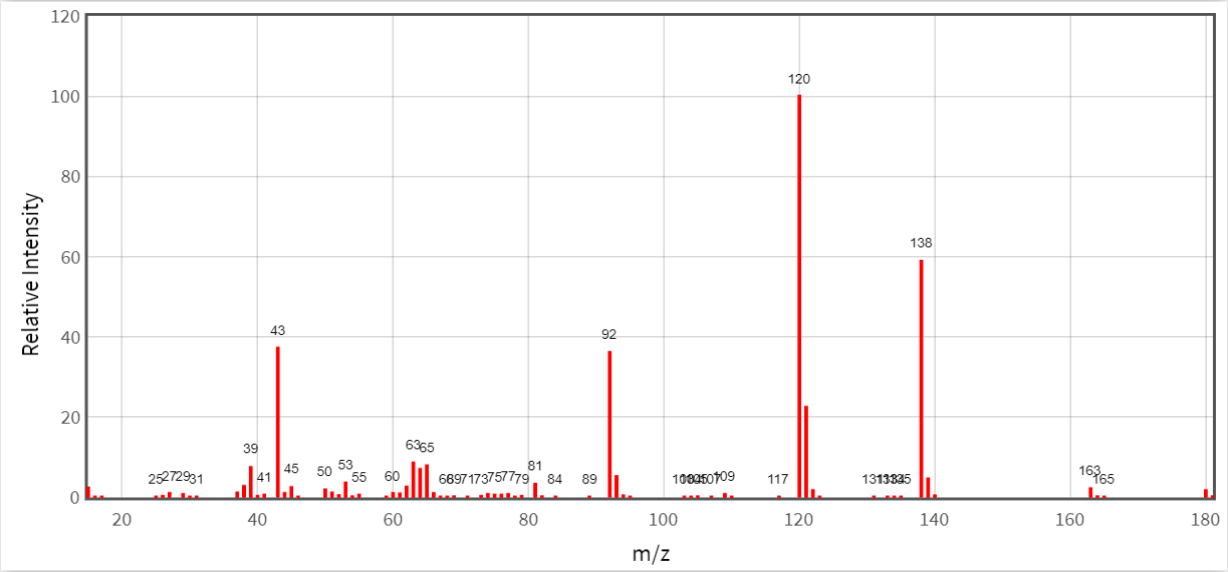


comments:
Mass 138 peak is the third most abundant peak in the spectrum.

Three most abundant masses:

1	120	$[C_7H_4O_2]^+$
2	92	$[C_6H_4O]^+$
3	138	$[C_7H_6O_3]^+$

acetylsalicylic acid		mass:	C	H	O
	$C_9H_8O_4$	180	9	8	4

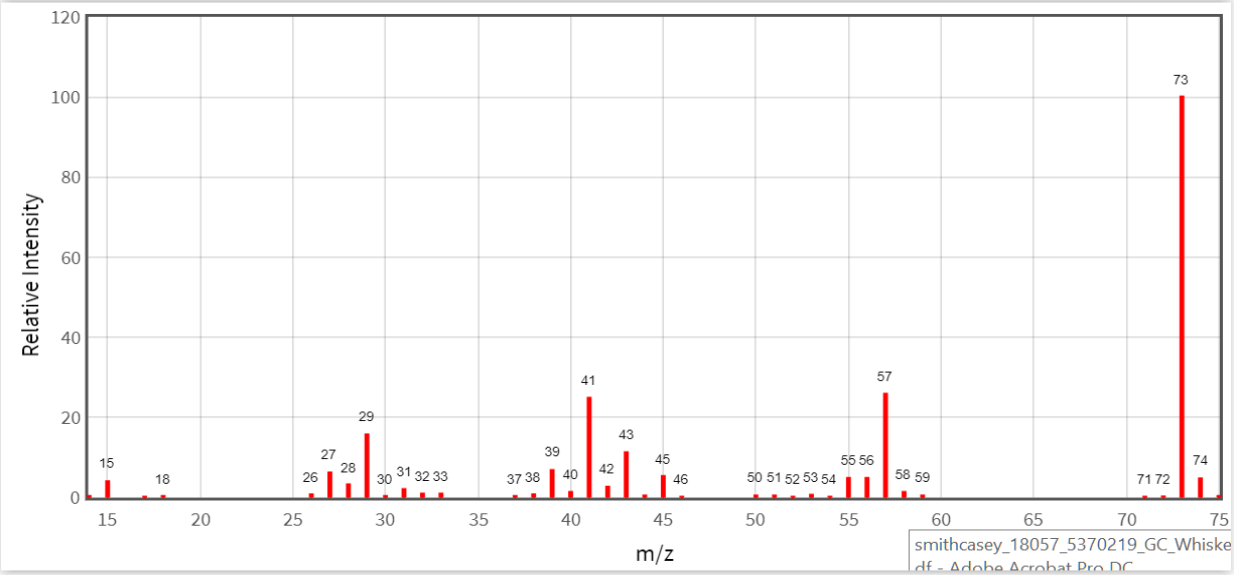


comments:
Mass 180 peak is small due to fragmentation.

Three most abundant masses:

1	120	$[C_7H_4O_2]^+$
2	138	$[C_7H_6O_3]^+$
3	43	$[C_3H_7]^+$
4	92	$[C_6H_4O]^+$

Methyl t-butyl ether (MTBE)	$C_5H_{12}O$	mass:	C	H	O
		88	5	12	1

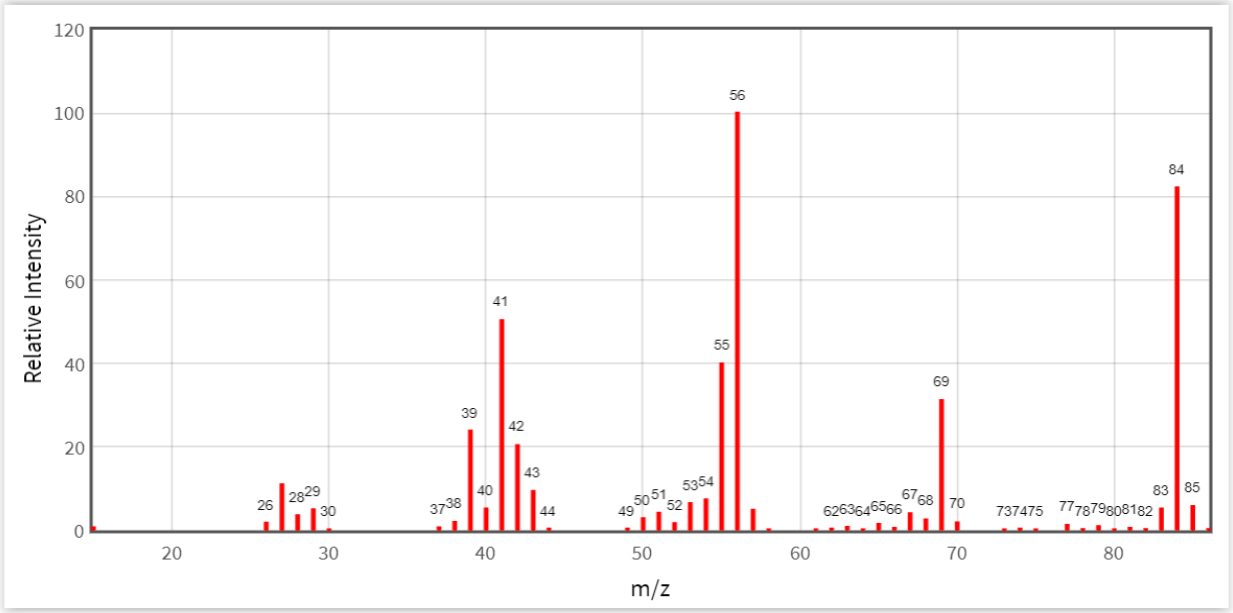


comments:
Mass 88 peak is not observed due to fragmentation.

Three most abundant masses:

1	73	$[C_4H_9O]^+$
2	57	$[C_4H_9]^+$
3	41	$[C_3H_5]^+$

		mass:	C	H	O
cyclohexane	C_6H_{12}	84	6	12	



comments:
Mass 84 peak is the second most abundant peak in the spectrum.

Three most abundant masses:

1	56	$[C_4H_8]^+$	$M - C_2H_4$
2	84	$[C_6H_{12}]^+$	
3	41	$[C_3H_5]^+$	$M - 43$