Qualitative Organic Analysis – CH 351

Mass Spectrometry

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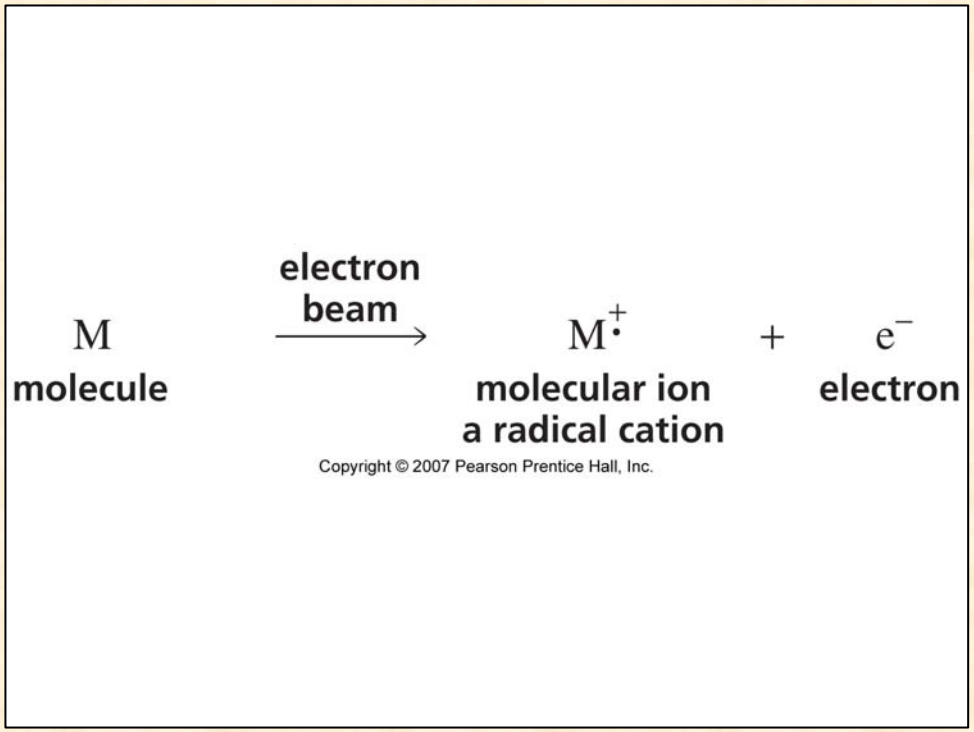
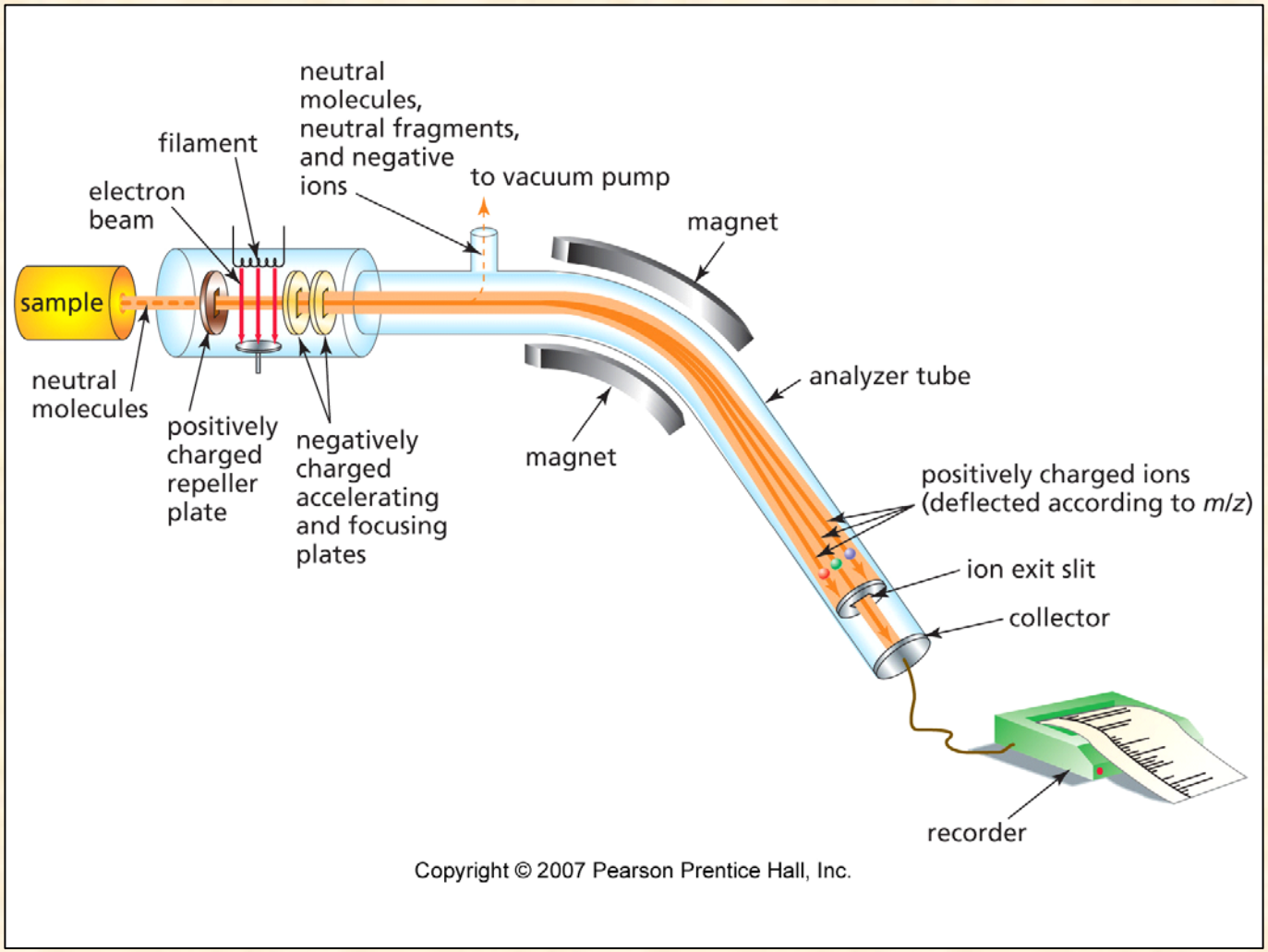
General Aspects

Theoretical basis of mass spectrometry



General Aspects

Basic Instrumentation



General Aspects - Ionization Methods

Gas Phase Ionization Methods

Electron Impact Ionization (EI)

electron beam

Chemical Ionization (CI)

ionized molecules (CH_3^+, or $(CH_3)_3C^+$

General Aspects - Ionization Methods

Gas Phase Ionization Methods

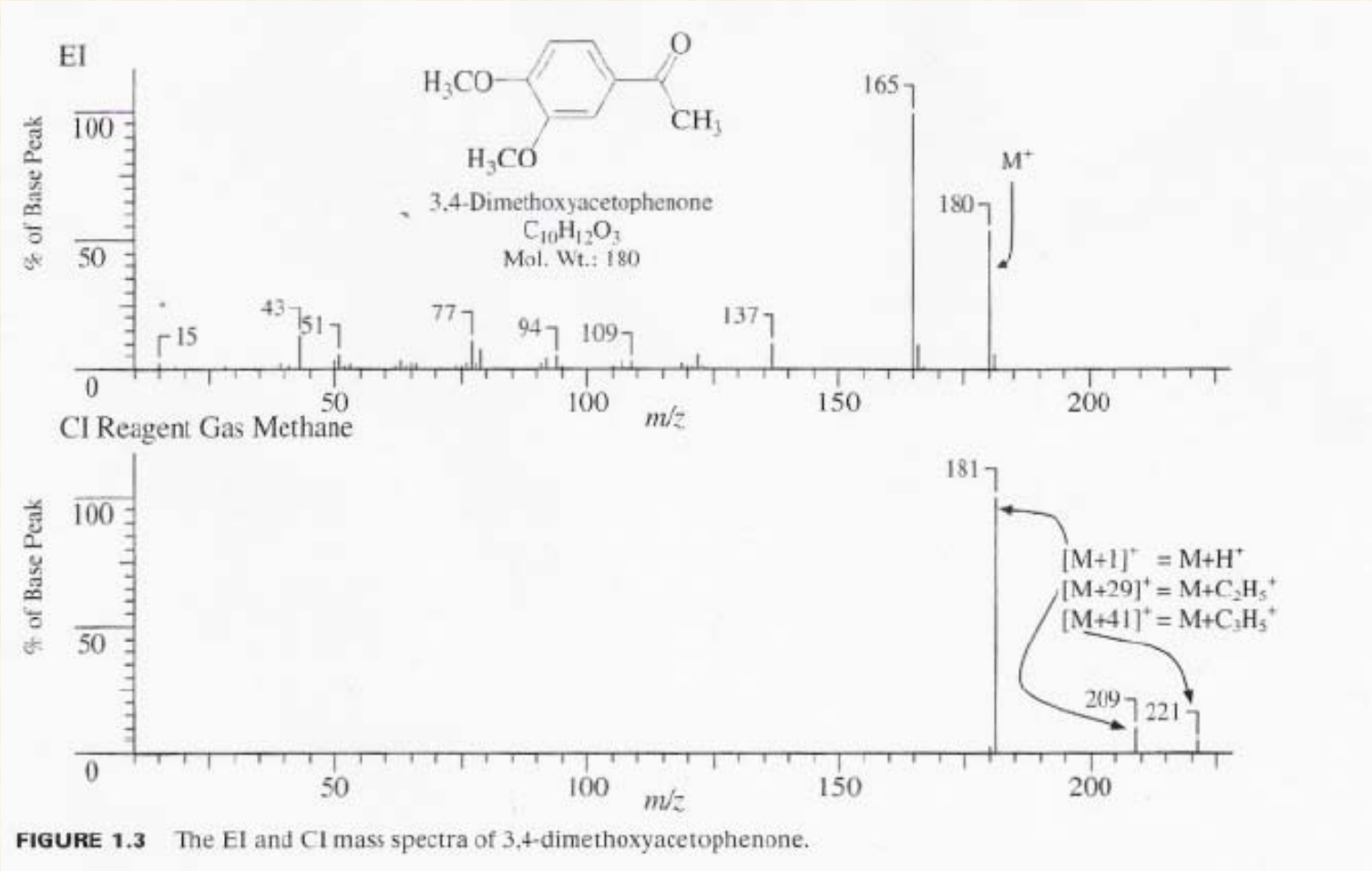


FIGURE 1.3 The EI and CI mass spectra of 3,4-dimethoxyacetophenone.

General Aspects - Ionization Methods

Desorption Ionization Methods

Field Desorption Ionization (FD)

emits ions fro surface

Fast Atom Bombardment (FAB)

high energy Xe or Ar atoms (liquid sample)

Liquid Secondary Ionization Mass Spec. – LSIMS

Cs^+ ions

Plasma Desorption Ionization

fission product of ^{252}Cf

Laser Desorption Ionization

laser beam

General Aspects - Ionization Methods

[Desorption Ionization Methods]

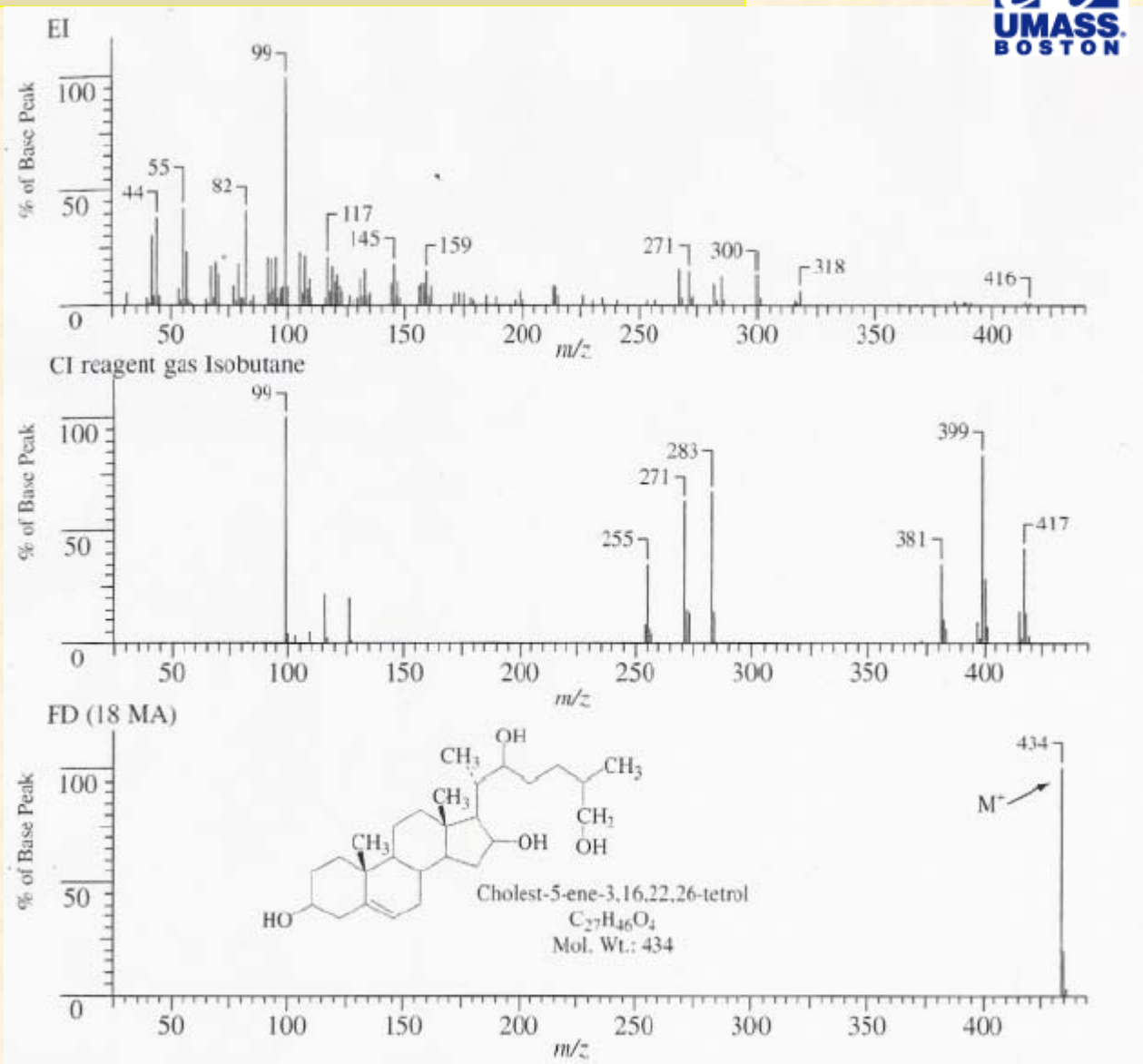


FIGURE 1.4 The electron impact (EI), chemical ionization (CI), and field desorption (FD) mass spectra of cholest-5-ene-3, 16, 22, 26-tetrol.

General Aspects - Ionization Methods

Evaporative Ionization Methods

Thermospray MS (TSMS)

heated capillary tube (outdated)

Electrospray MS (ESI-MS)

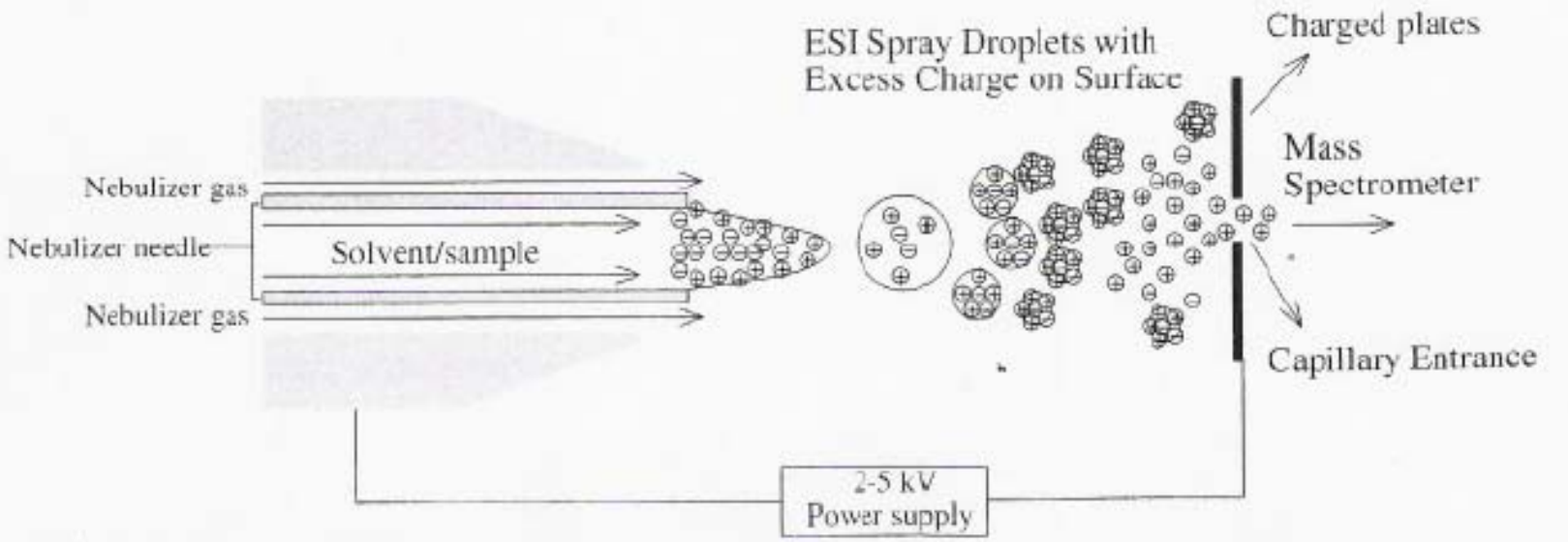
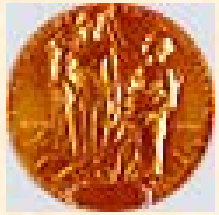
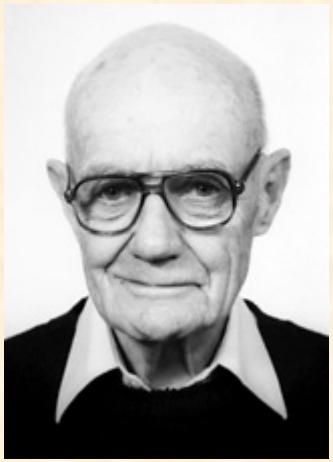


FIGURE 1.5 A diagram showing the evaporation of solvent leading to individual ions in an electrospray instrument.

General Aspects - Ionization Methods

The Nobel Prize in Chemistry 2002





John B. Fenn **Koichi Tanaka** **Kurt Wüthrich**

General Aspects - Ionization Methods

The Nobel Prize in Chemistry 2002

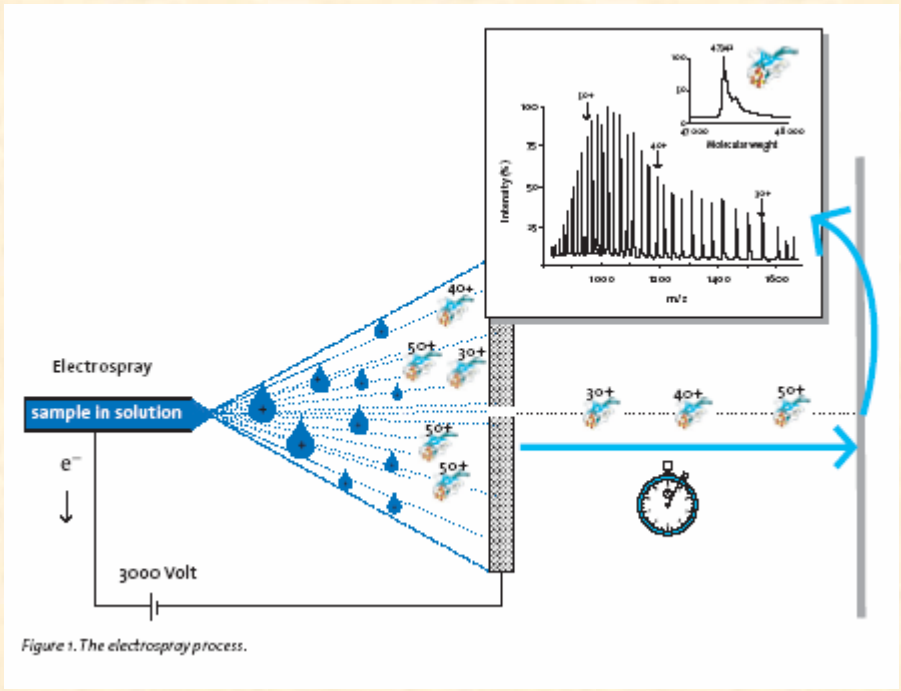


Figure 1. The electrospray process.

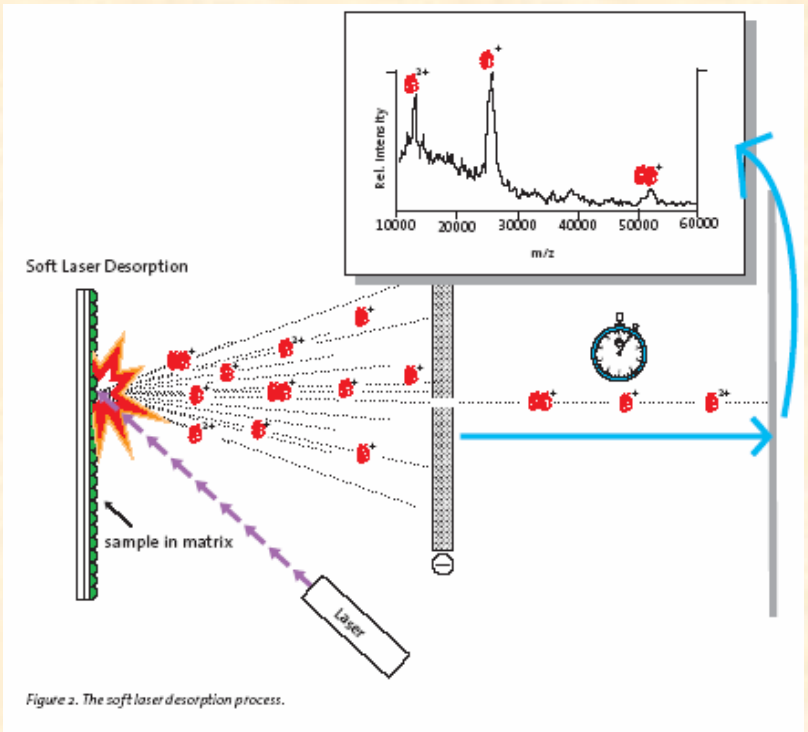


Figure 2. The soft laser desorption process.

General Aspects - Ionization Methods

Evaporative Ionization Methods

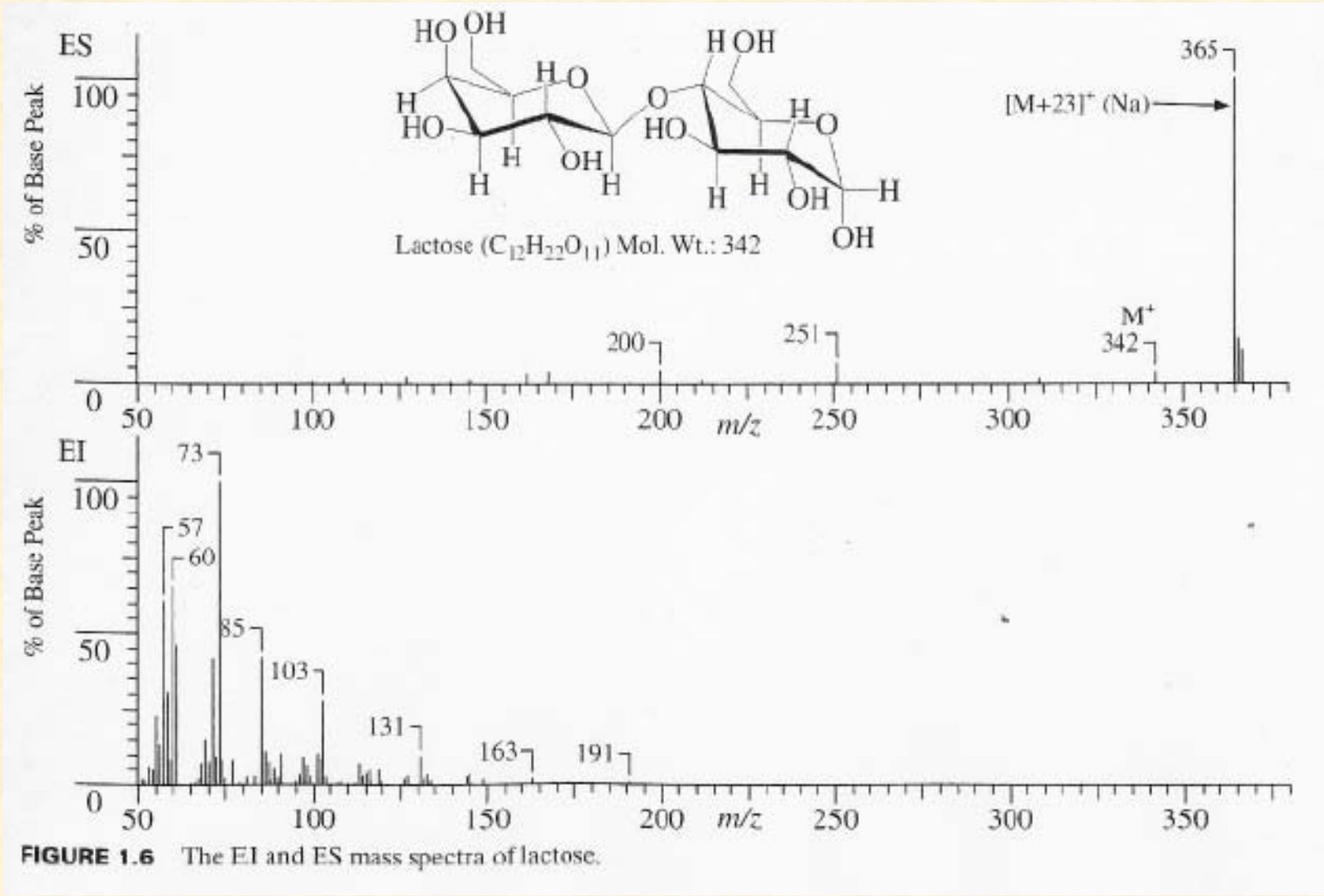


FIGURE 1.6 The EI and ES mass spectra of lactose.

General Aspects - Ionization Methods

TABLE 1.1 Summary of Ionization Methods.

Ionization Method	Ions Formed	Sensitivity	Advantage	Disadvantage
Electron impact	M^+	ng–pg	Data base searchable Structural information	M^+ occasionally absent
Chemical ionization	M + 1, M + 18, etc	ng–pg	M+ usually present	Little structural information
Field desorption	M^+	μg–ng	Non volatile compounds	Specialized equipment
Fast atom bombardment	M + 1, M + cation M + matrix	μg–ng	Non volatile compounds Sequencing information	Matrix interference Difficult to interpret
Plasma desorption	M+	μg–ng	Non volatile compounds	Matrix interference
Laser desorption	M + 1, M + matrix	μg–ng	Non volatile compounds Burst of ions	Matrix interference
Thermospray	M^+	μg–ng	Non volatile compounds	Outdated
Electrospray	M^+, M^{++}, M^{+++}, etc.	ng–pg	Non volatile compounds interfaces w/ LC Forms multiply charged ions	Limited classes of compounds Little structural information

General Aspects – Mass Analyzers

Magnetic Sector Mass Spectrometers

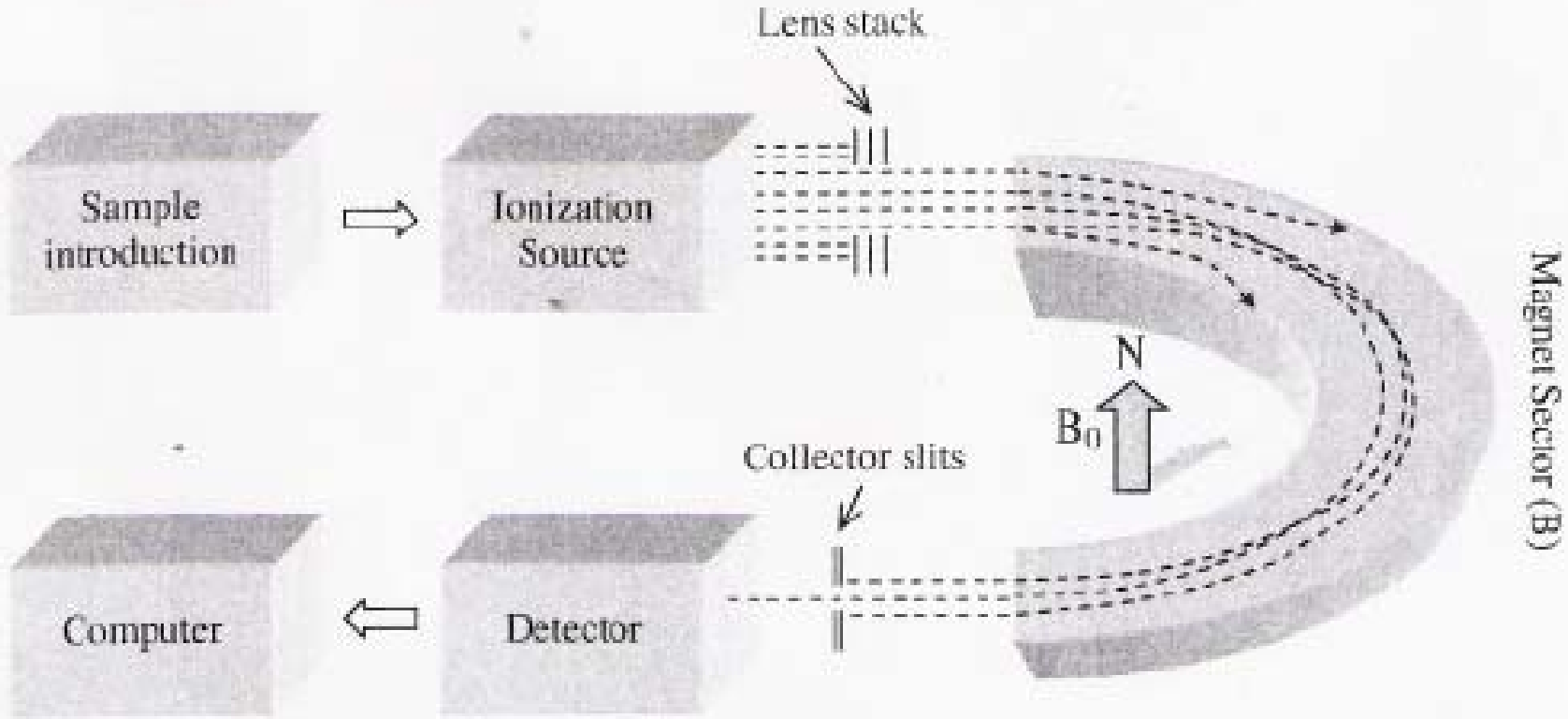


FIGURE 1.8 Schematic diagram of a single focusing, 180° sector mass analyzer. The magnetic field is perpendicular to the page. The radius of curvature varies from one instrument to another.

General Aspects – Mass Analyzers

Magnetic Sector Mass Spectrometers

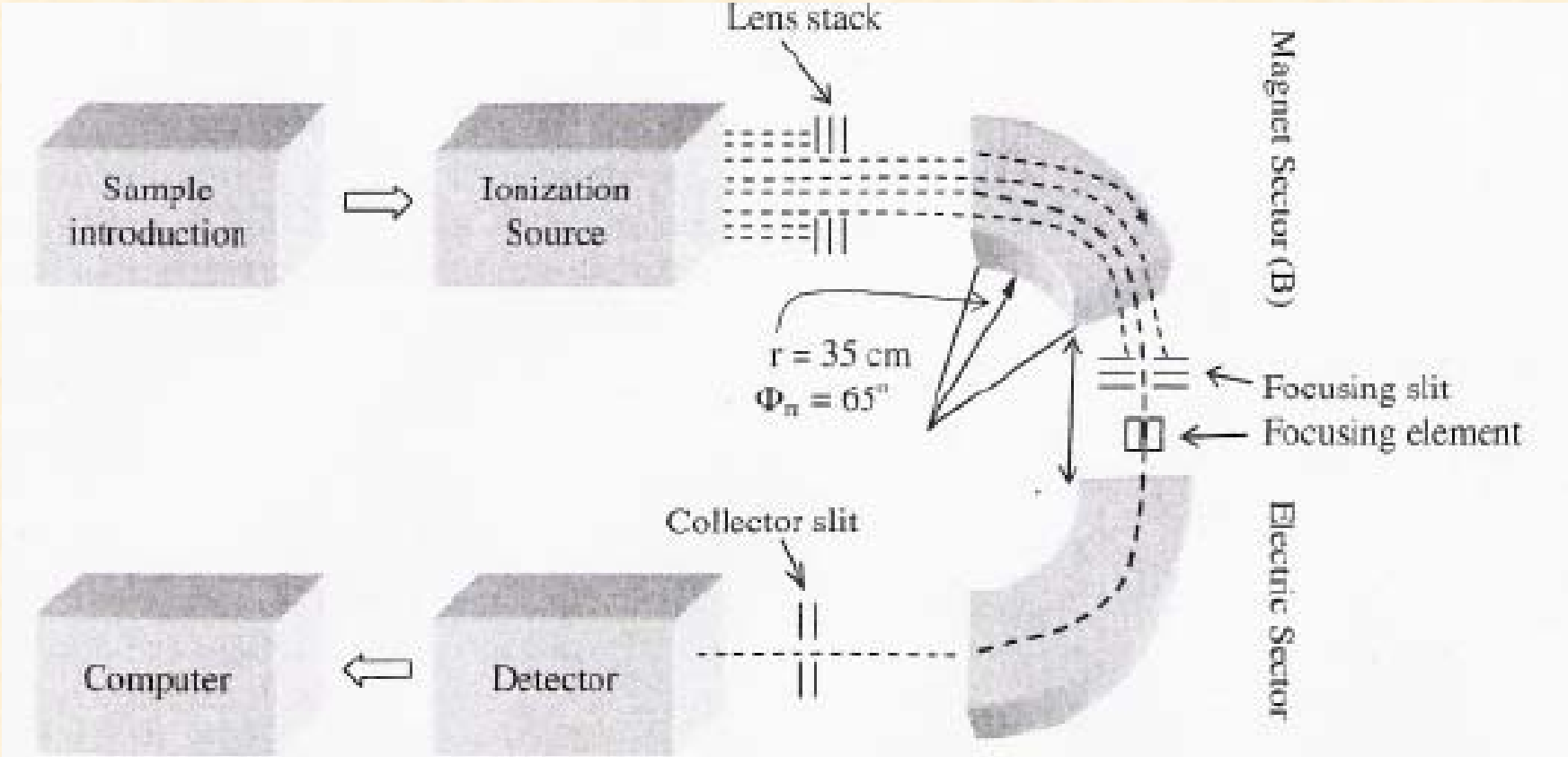


FIGURE 1.9 Schematic of double-focusing mass spectrometer.

General Aspects – Mass Analyzers

Quadrupole Mass Spectrometers

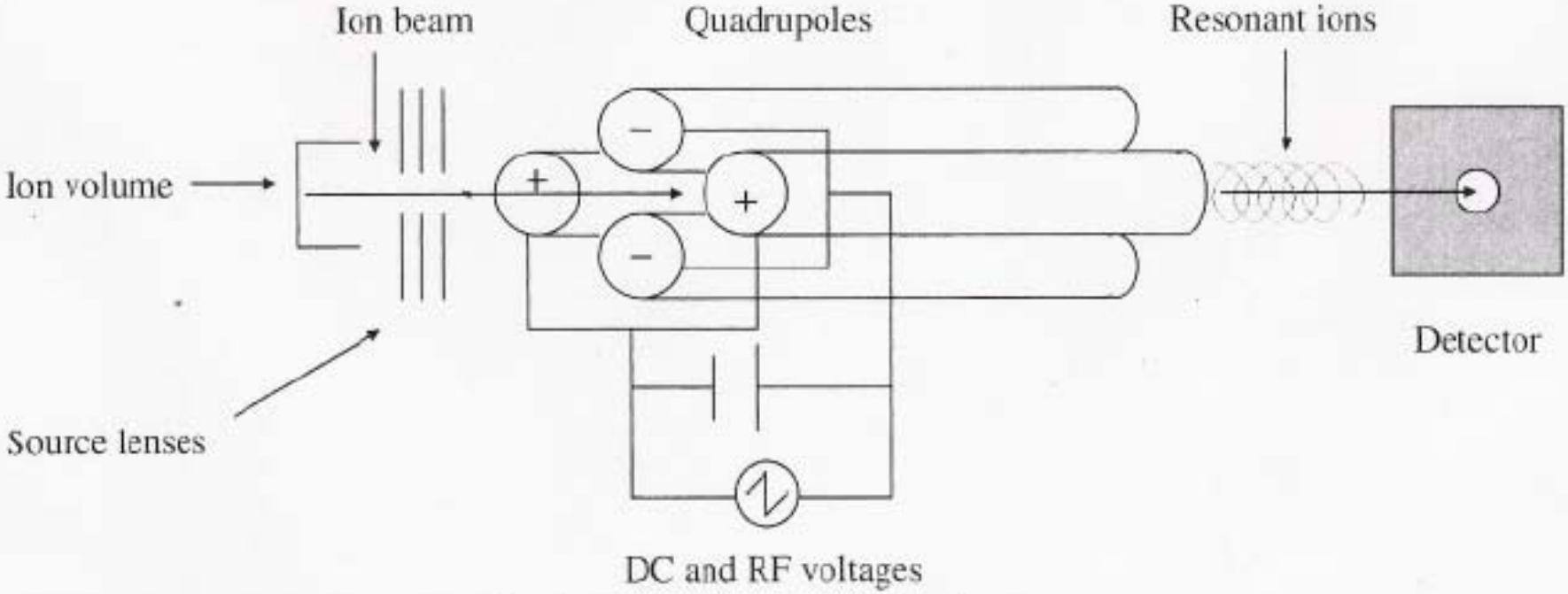


FIGURE 1.10 Schematic representation of a quadrupole "mass filter" or ion separator.

General Aspects – Mass Analyzers

Ion Trap Mass Spectrometers

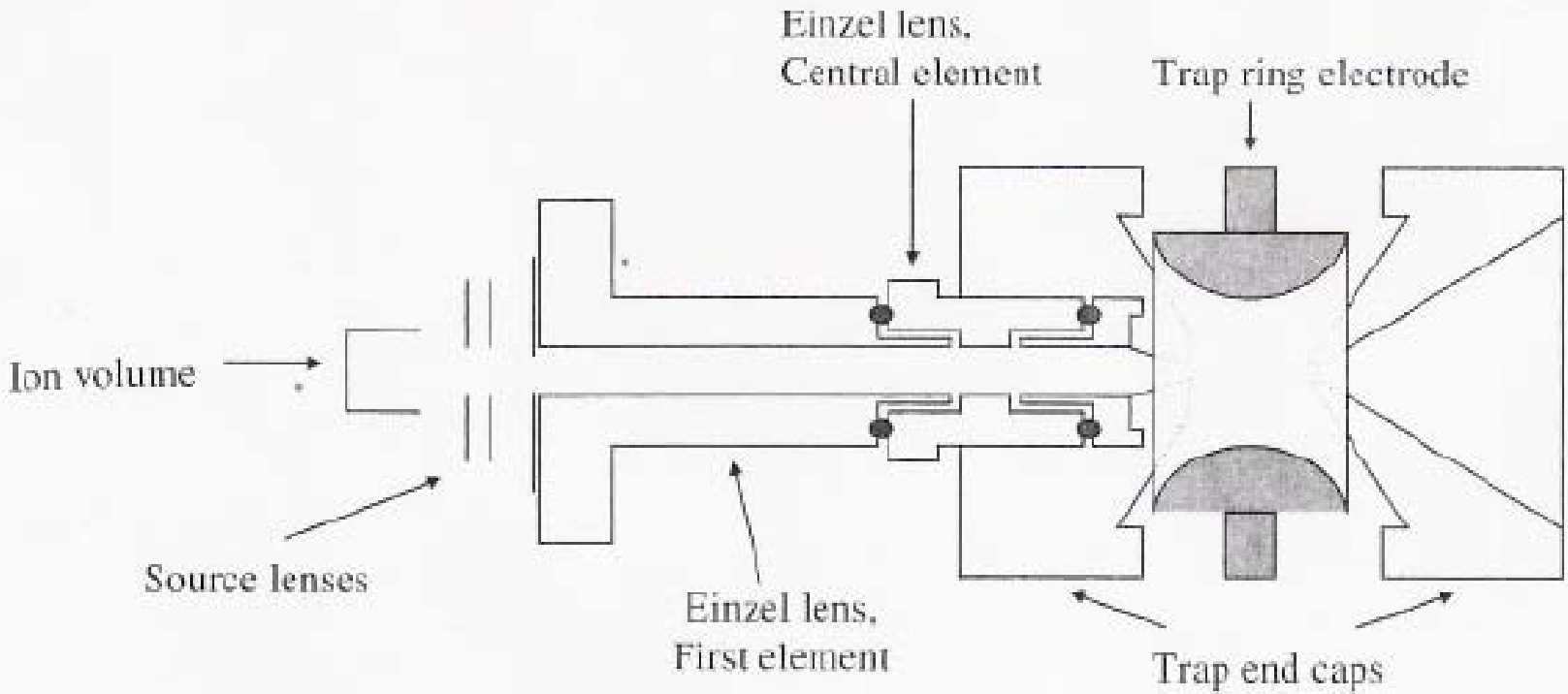
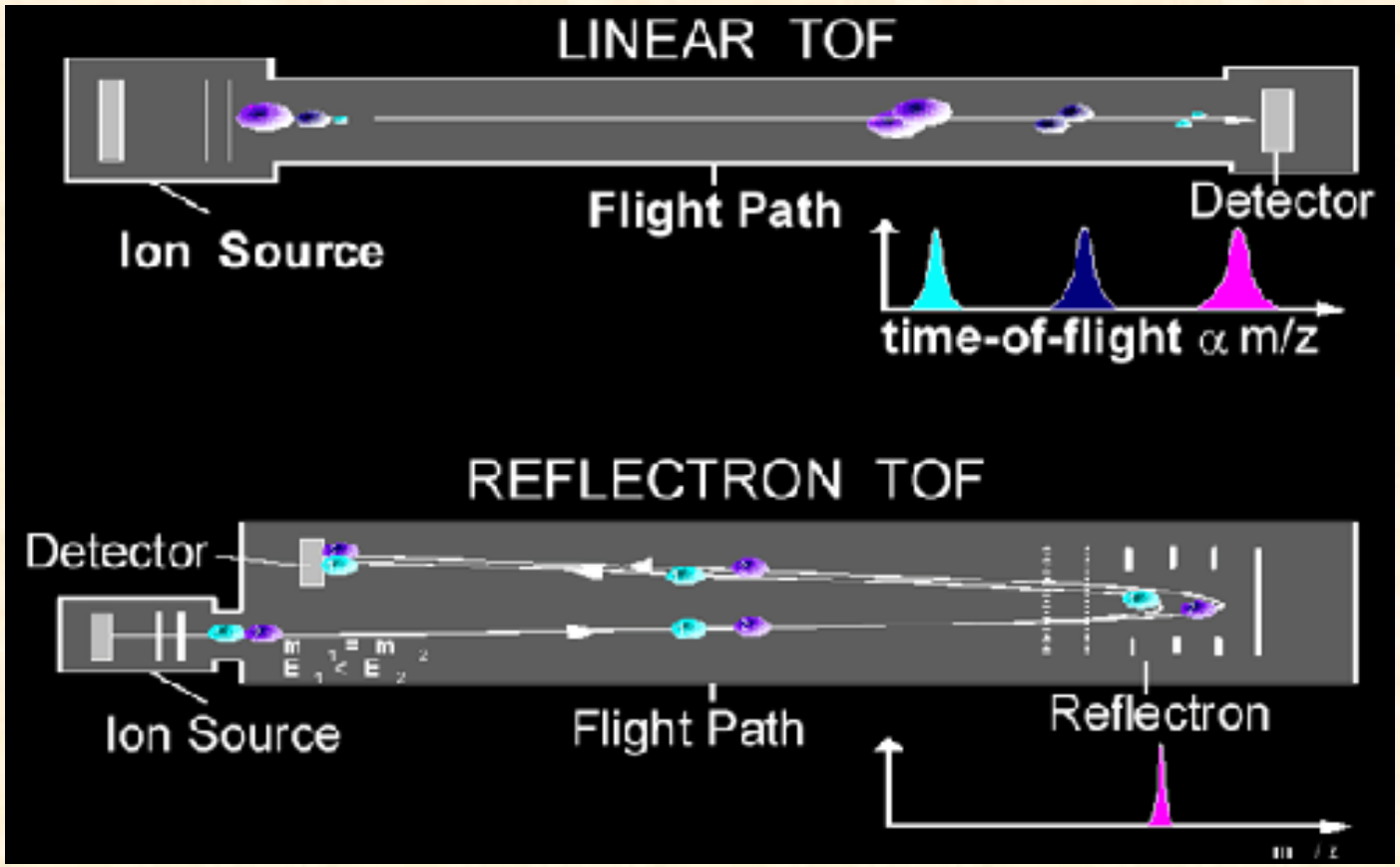


FIGURE 1.11 Cross sectional view of an ion trap.

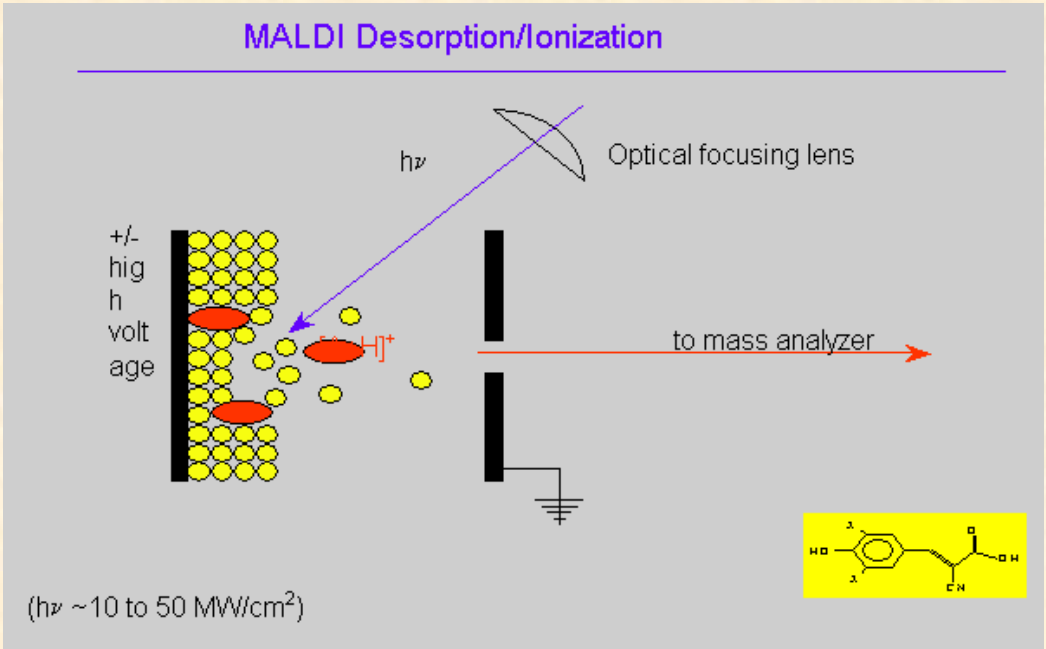
General Aspects – Mass Analyzers

Time-of-Flight Mass Spectrometers



General Aspects – Mass Analyzers

MALDI TOF Mass Spectrometers

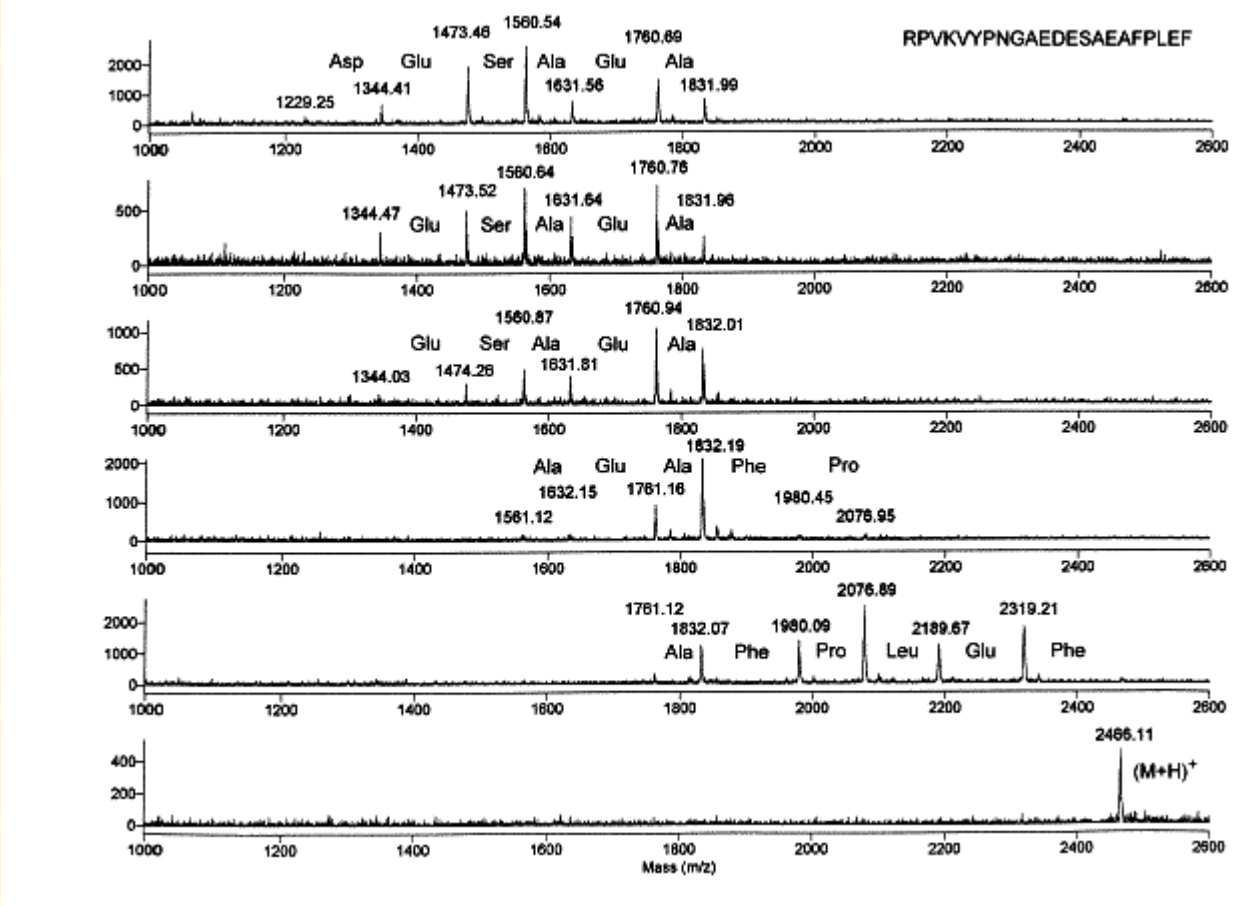


Some common MALDI matrices at 337 nm.

Matrix	*Application*
2,5-Dihydroxybenzoic acid (DHB)	Peptides, proteins, lipids, and oligosaccarides
3,5-Dimethoxy-4-hydroxycinnamic acid (sinapinic acid)	Peptides, proteins, and glycoproteins
α-Cyano-4-hydroxycinnamic acid (CHCA)	Peptides, proteins, lipids, and oligonucleotides

General Aspects – Mass Analyzers

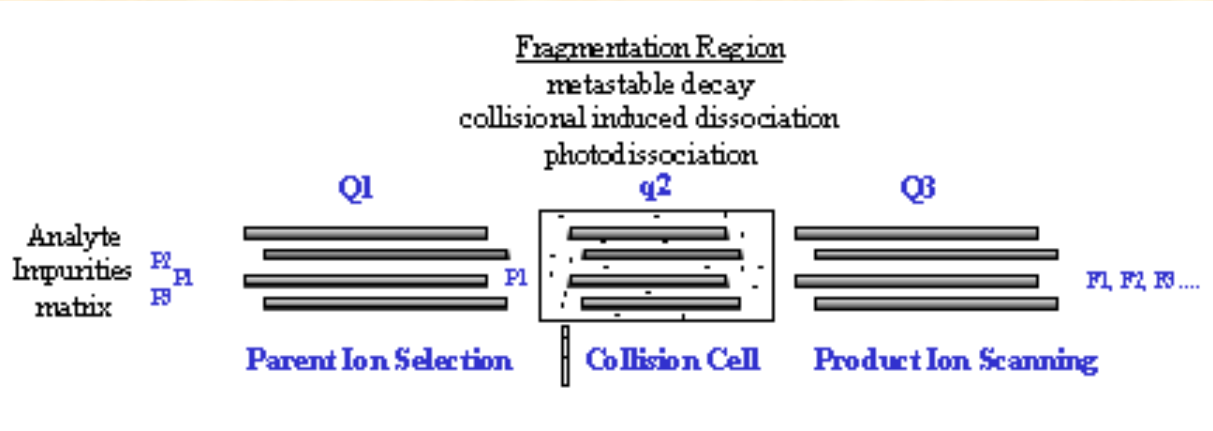
MALDI TOF Mass Spectrometers

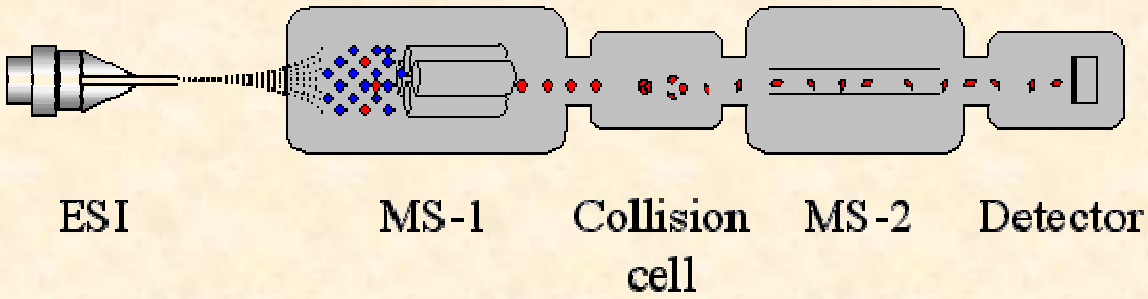


Ladder sequencing of the adrenocorticotropic hormone (ACTH) fragment 18-39 utilizing the CPY dilution technique and MALDI linear DE-TOF-MS

General Aspects – Mass Analyzers

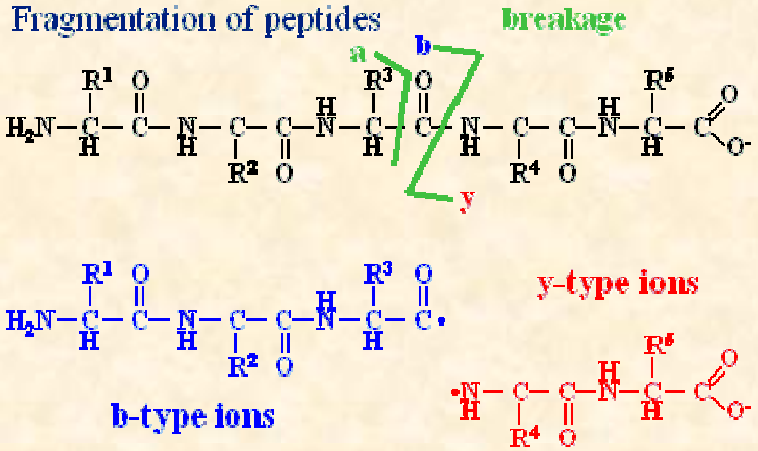
Tandem Mass Spectrometry

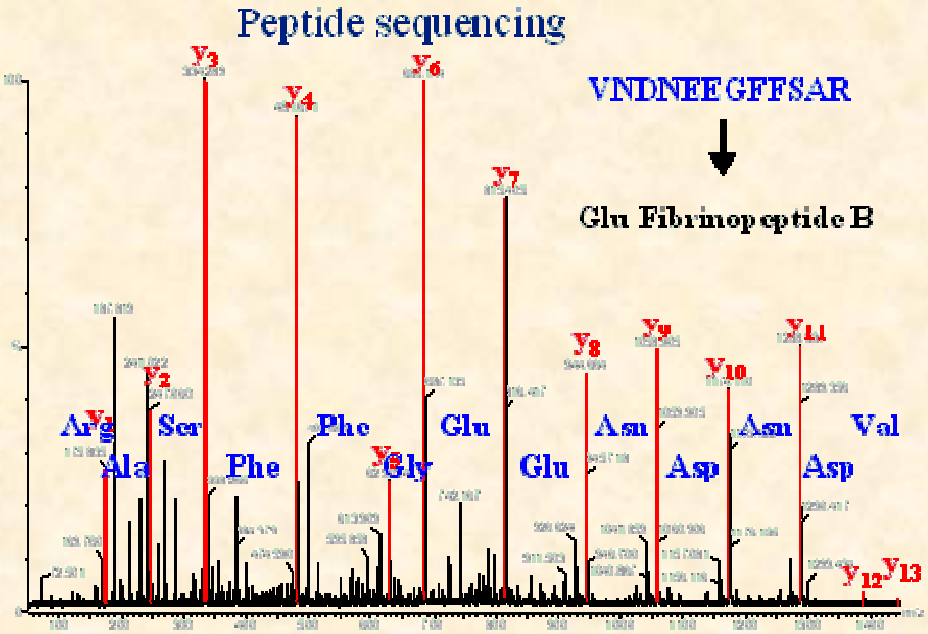




General Aspects – Mass Analyzers

Tandem Mass Spectrometry





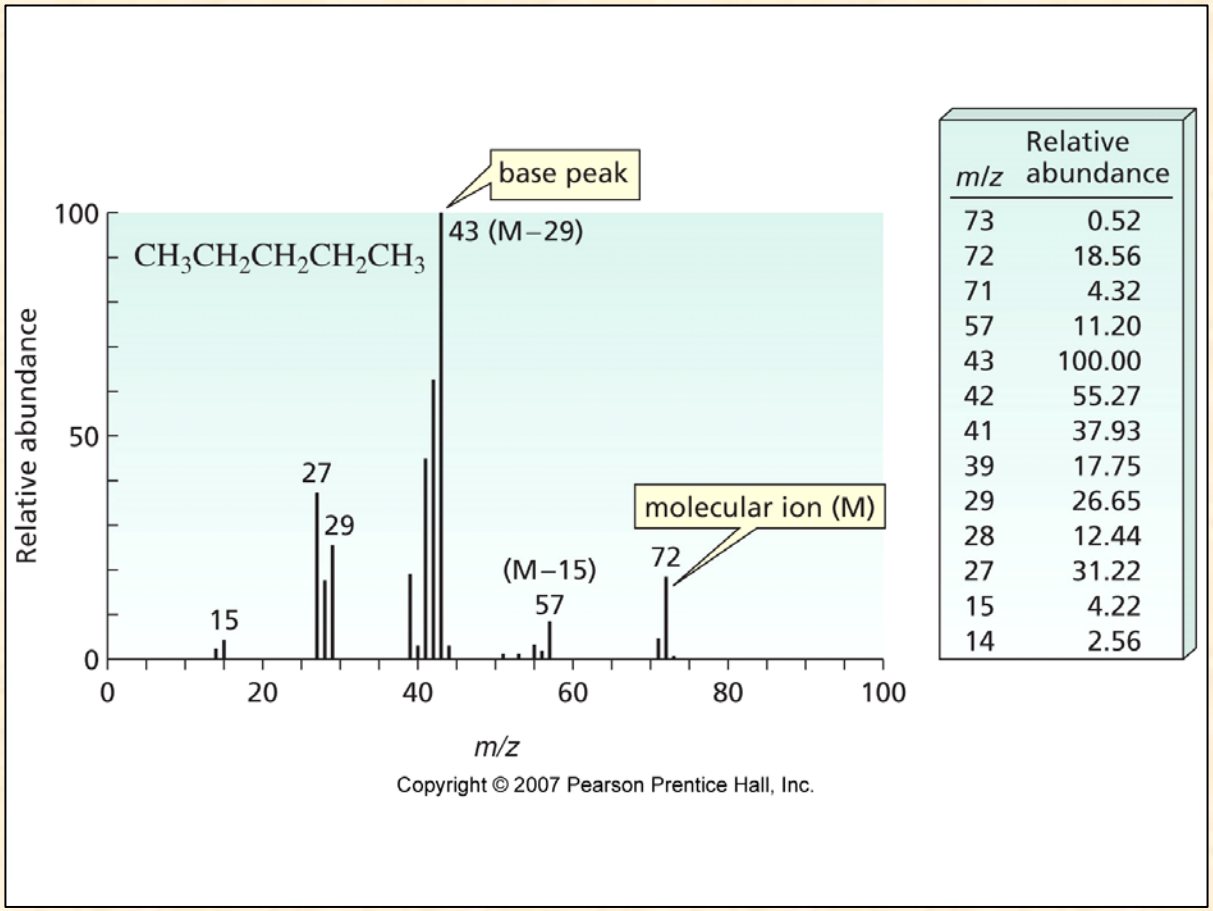
General Aspects – Mass Analyzers

TABLE 1.2 Summary of Mass Analyzers.

Mass Analyzer	Mass Range	Resolution	Sensitivity	Advantage	Disadvantage
Magnetic Sector	1–15,000 *m/z*	0.0001	Low	High res.	Low sensitivity Very expensive High technical expertise
Quadrupole	1–5000 *m/z*	unit	High	Easy to use Inexpensive High sensitivity	Low res. Low mass range
Ion trap	1–5000 *m/z*	unit	High	Easy to use Inexpensive High sensitivity Tandem MS (MS^n)	Low res. Low mass range
Time of flight	Unlimited	0.0001	High	High mass range Simple design	Very high res.
Fourier transform	up to 70 kDa	0.0001	High	Very High res. and mass range	Very expensive High technical expertise

General Aspects – Interpretation of Mass Spectra

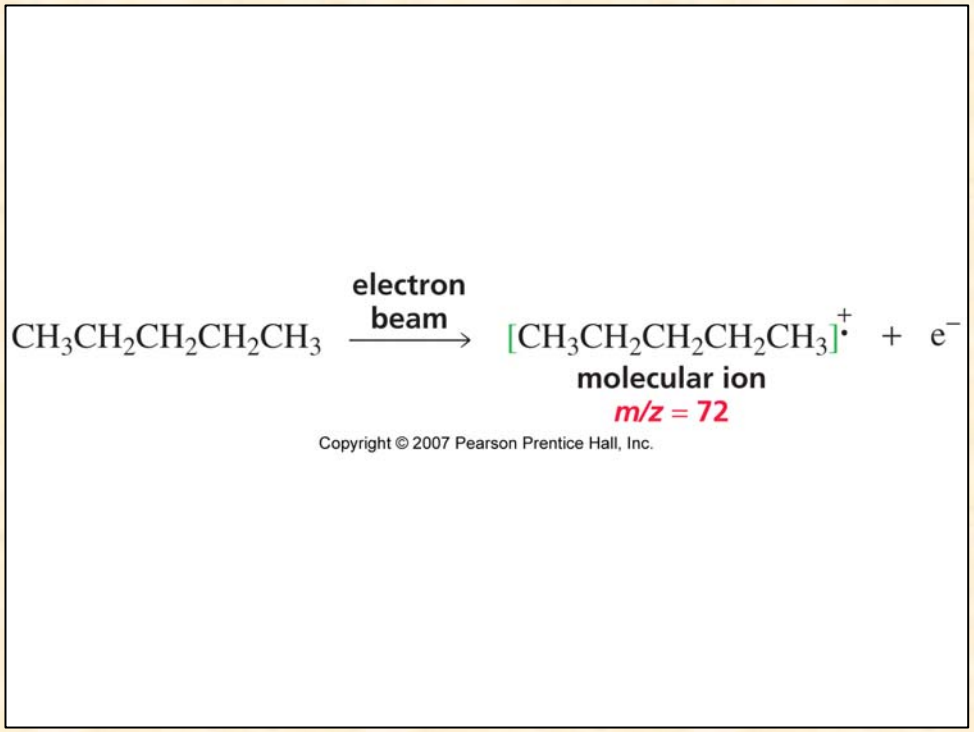
The mass spectrum



m/z	Relative abundance
73	0.52
72	18.56
71	4.32
57	11.20
43	100.00
42	55.27
41	37.93
39	17.75
29	26.65
28	12.44
27	31.22
15	4.22
14	2.56

General Aspects – Interpretation of Mass Spectra

The ionization



General Aspects – Interpretation of Mass Spectra

High Resolution Mass Spectra

Table 12.3 The Exact Masses of Some Common Isotopes

Isotope	Mass	Isotope	Mass
^{1}H	1.007825 amu	^{32}S	31.9721 amu
^{12}C	12.00000 amu	^{35}Cl	34.9689 amu
^{14}N	14.0031 amu	^{79}Br	78.9183 amu
^{16}O	15.9949 amu		

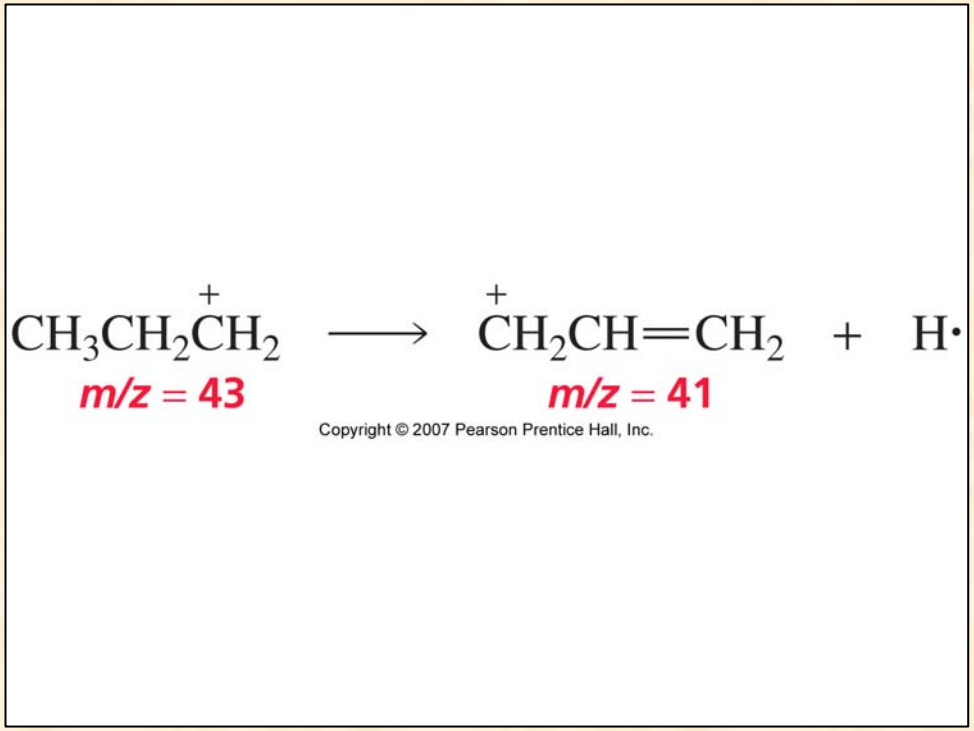
TABLE 1.4 Exact Masses of Isotopes.

Element	Atomic Weight	Nuclide	Mass
Hydrogen	1.00794	^{1}H	1.00783
		D(^{2}H)	2.01410
Carbon	12.01115	^{12}C	12.00000 (std)
		^{13}C	13.00336
Nitrogen	14.0067	^{14}N	14.0031
		^{15}N	15.0001
Oxygen	15.9994	^{16}O	15.9949
		^{17}O	16.9991
		^{18}O	17.9992
Fluorine	18.9984	^{19}F	18.9984
Silicon	28.0855	^{28}Si	27.9769
		^{29}Si	28.9765
		^{30}Si	29.9738
Phosphorus	30.9738	^{31}P	30.9738
Sulfur	32.0660	^{32}S	31.9721
		^{33}S	32.9715
		^{34}S	33.9679
Chlorine	35.4527	^{35}Cl	34.9689
		^{37}Cl	36.9659
Bromine	79.9094	^{79}Br	78.9183
		^{81}Br	80.9163
Iodine	126.9045	^{127}I	126.9045

High resolution MS - molecular formula

General Aspects – Interpretation of Mass Spectra

M-2 peaks



Peaks are commonly observed at *m/z* values two units below the *m/z* values of a carbocation, because the carbocation can lose two hydrogens

General Aspects – Interpretation of Mass Spectra

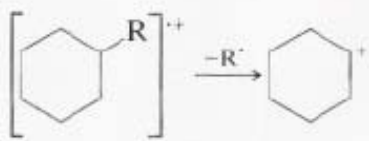
Fragmentation (EI only!!!)

1. The relative height of the molecular ion peak is greatest for the straight-chain compound and decreases as the degree of branching increases (see rule 3).
2. The relative height of the molecular ion peak usually decreases with increasing molecular weight in a homologous series. Fatty esters appear to be an exception.
3. Cleavage is favored at alkyl-substituted carbon atoms: the more substituted, the more likely is cleavage. This is a consequence of the increased stability of a tertiary carbocation over a secondary, which in turn is more stable than a primary.

Cation stability order:

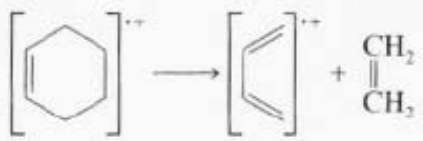
$$CH_3^+ < R_2CH_2^+ < R_3CH^+ < R_3C^+$$

4. Double bonds, cyclic structures, and especially aromatic (or heteroaromatic) rings stabilize the molecular ion and thus increase the probability of its appearance.
5. Double bonds favor allylic cleavage and give the resonance-stabilized allylic carbocation. This rule does not hold for simple alkenes because of the ready migration of the double bond, but it does hold for cycloalkenes.
6. Saturated rings tend to lose alkyl side chains at the α bond. This is merely a special case of branching (rule 3). The positive charge tends to stay with the ring fragment. See Scheme 1.4.



(Sch 1.4)

Unsaturated rings can undergo a *retro*-Diels-Alder reaction Scheme 1.5:

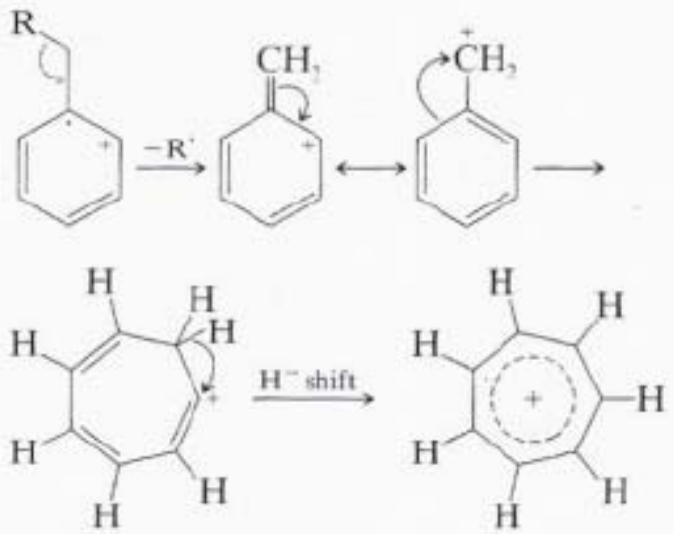


(Sch 1.5)

General Aspects – Interpretation of Mass Spectra

Fragmentation (EI only!!!!)

7. In alkyl-substituted aromatic compounds, cleavage is very probable at the bond β to the ring, giving the resonance-stabilized benzyl ion or, more likely, the tropylium ion (see Scheme 1.6).



(Sch 1.6)

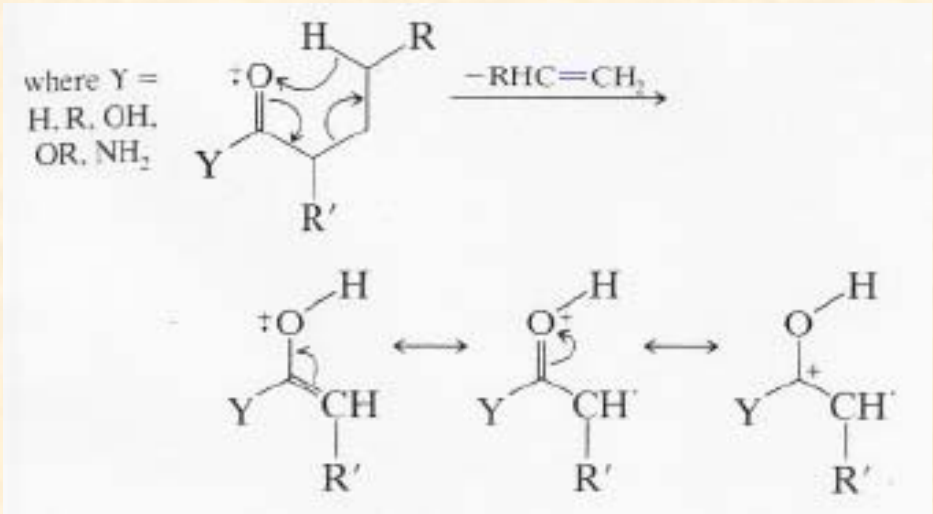
8. The C—C bonds next to a heteroatom are frequently cleaved, leaving the charge on the fragment containing the heteroatom whose nonbonding electrons provide resonance stabilization.

9. Cleavage is often associated with elimination of small, stable, neutral molecules, such as carbon monoxide, olefins, water, ammonia, hydrogen sulfide, hydrogen cyanide, mercaptans, ketene, or alcohols, often with rearrangement (Section 1.5.5).

General Aspects – Interpretation of Mass Spectra

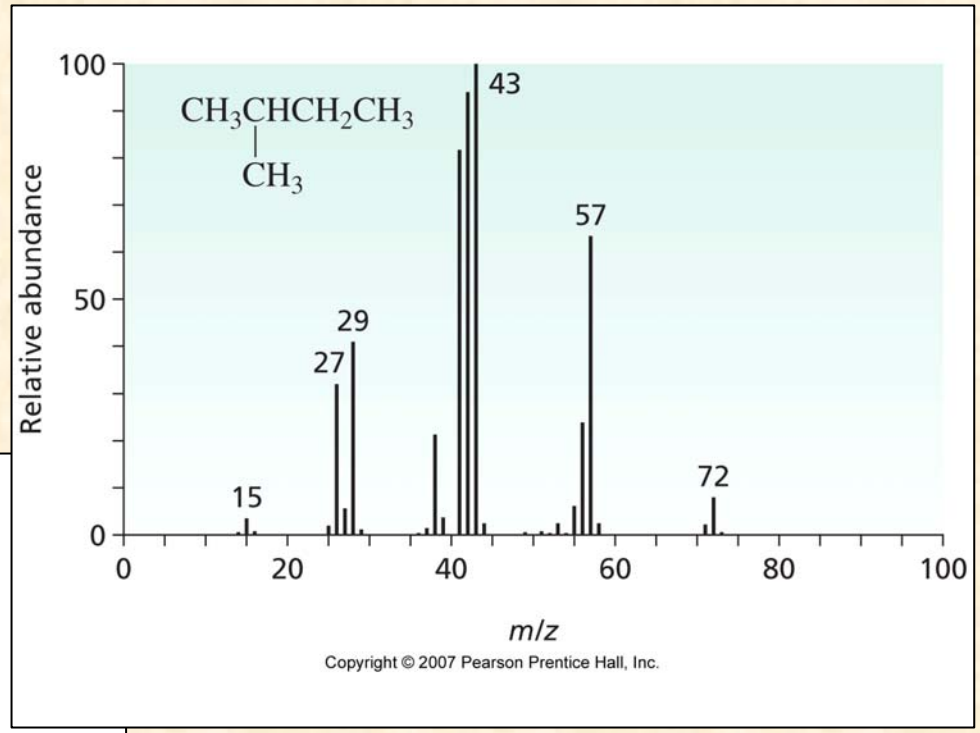
Rearrangements

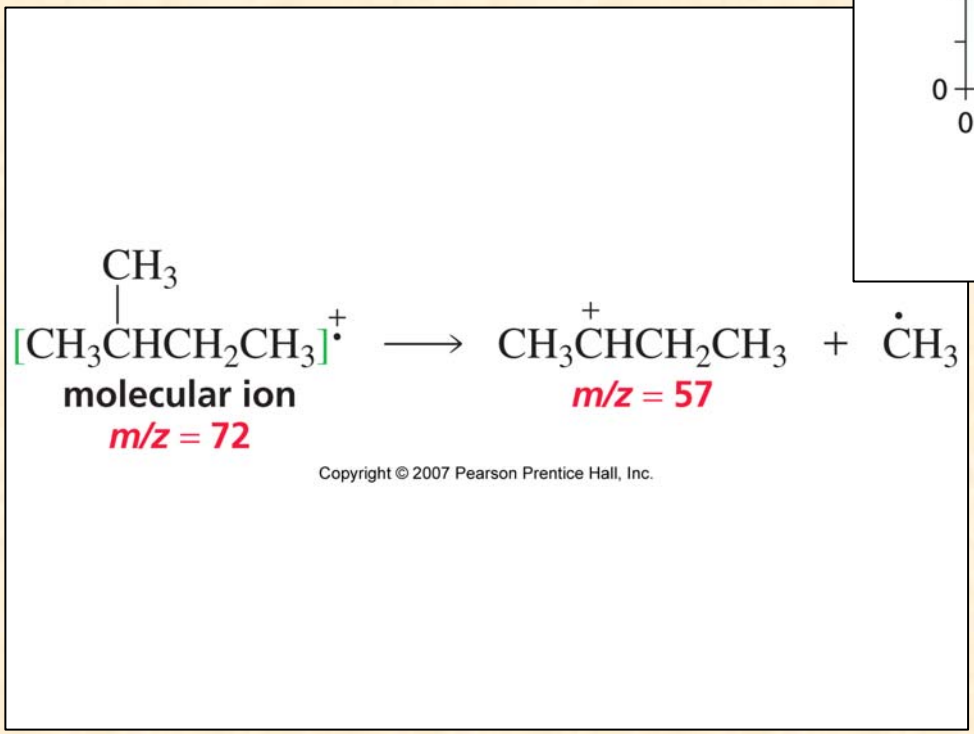
McLafferty rearrangement



General Aspects – Interpretation of Mass Spectra

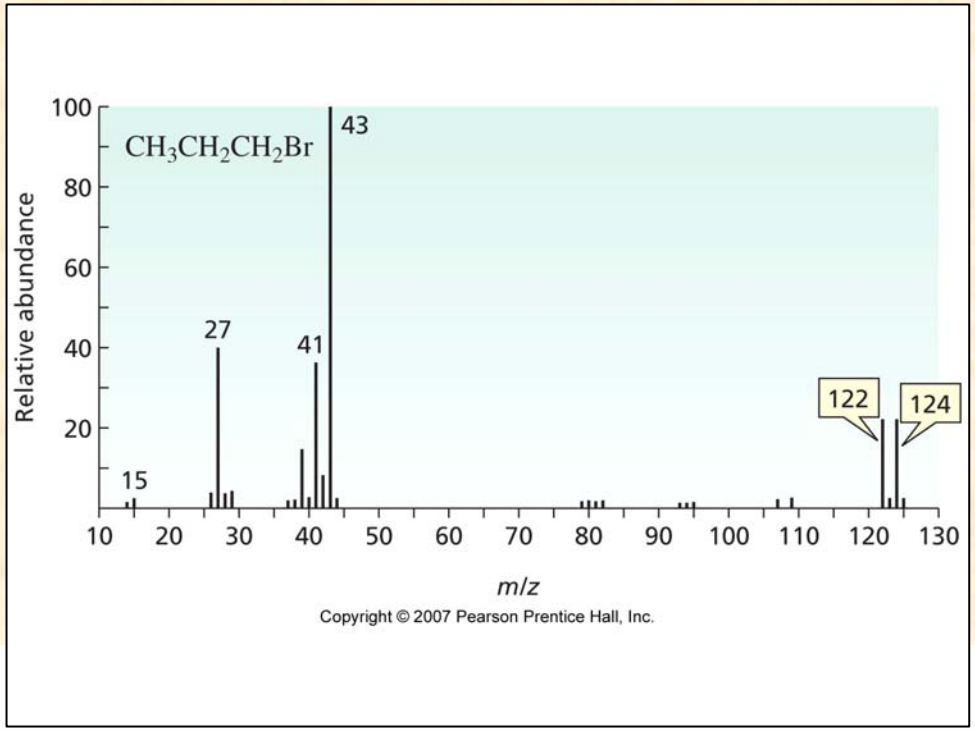
Mass Spectrum – 2-Me-butane





General Aspects – Interpretation of Mass Spectra

Isotope Effects



$CH_3CH_2CH_2{}^{79}\ddot{B}r: + CH_3CH_2CH_2{}^{81}\ddot{B}r: \xrightarrow{-e^-} CH_3CH_2CH_2{}^{79}\ddot{B}r:^{+} + CH_3CH_2CH_2{}^{81}\ddot{B}r:^{+} \longrightarrow CH_3CH_2\overset{+}{C}H_2 + {}^{79}\dot{B}r: + {}^{81}\dot{B}r:$

1-bromopropane

m/z = 122

m/z = 124

m/z = 43

General Aspects – Interpretation of Mass Spectra

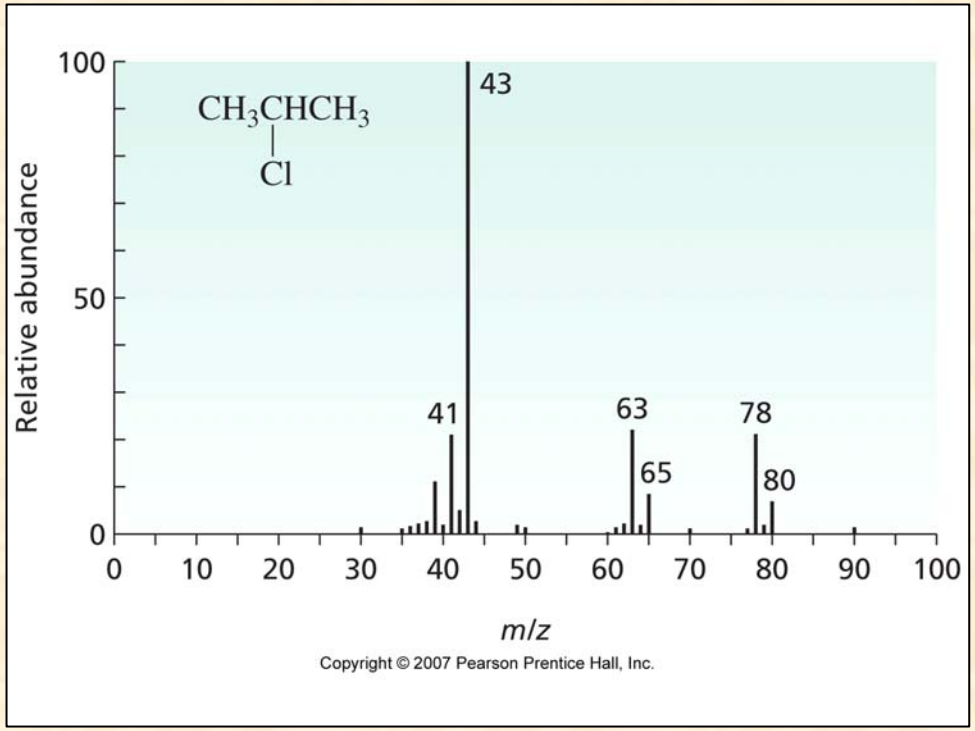
Isotope Effects

Table 12.2 The Natural Abundance of Isotopes Commonly Found in Organic Compounds

Element	Natural abundance			
Carbon	^{12}C 98.89%	^{13}C 1.11%		
Hydrogen	^{1}H 99.99%	^{2}H 0.01%		
Nitrogen	^{14}N 99.64%	^{15}N 0.36%		
Oxygen	^{16}O 99.76%	^{17}O 0.04%	^{18}O 0.20%	
Sulfur	^{32}S 95.0%	^{33}S 0.76%	^{34}S 4.22%	^{36}S 0.02%
Fluorine	^{19}F 100%			
Chlorine	^{35}Cl 75.77%		^{37}Cl 24.23%	
Bromine	^{79}Br 50.69%		^{81}Br 49.31%	
Iodine	^{127}I 100%			

General Aspects – Interpretation of Mass Spectra

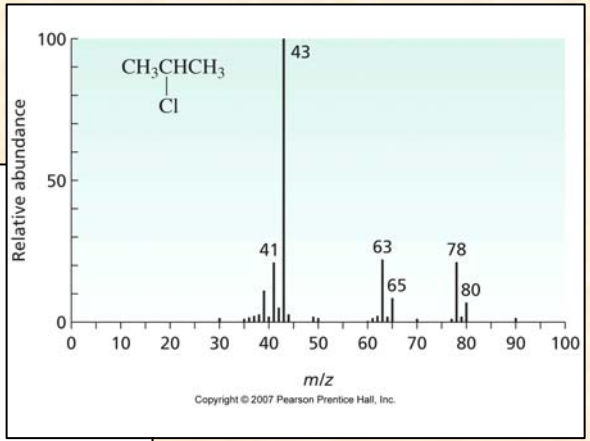
Mass Spectrum – 2-Cl-propane

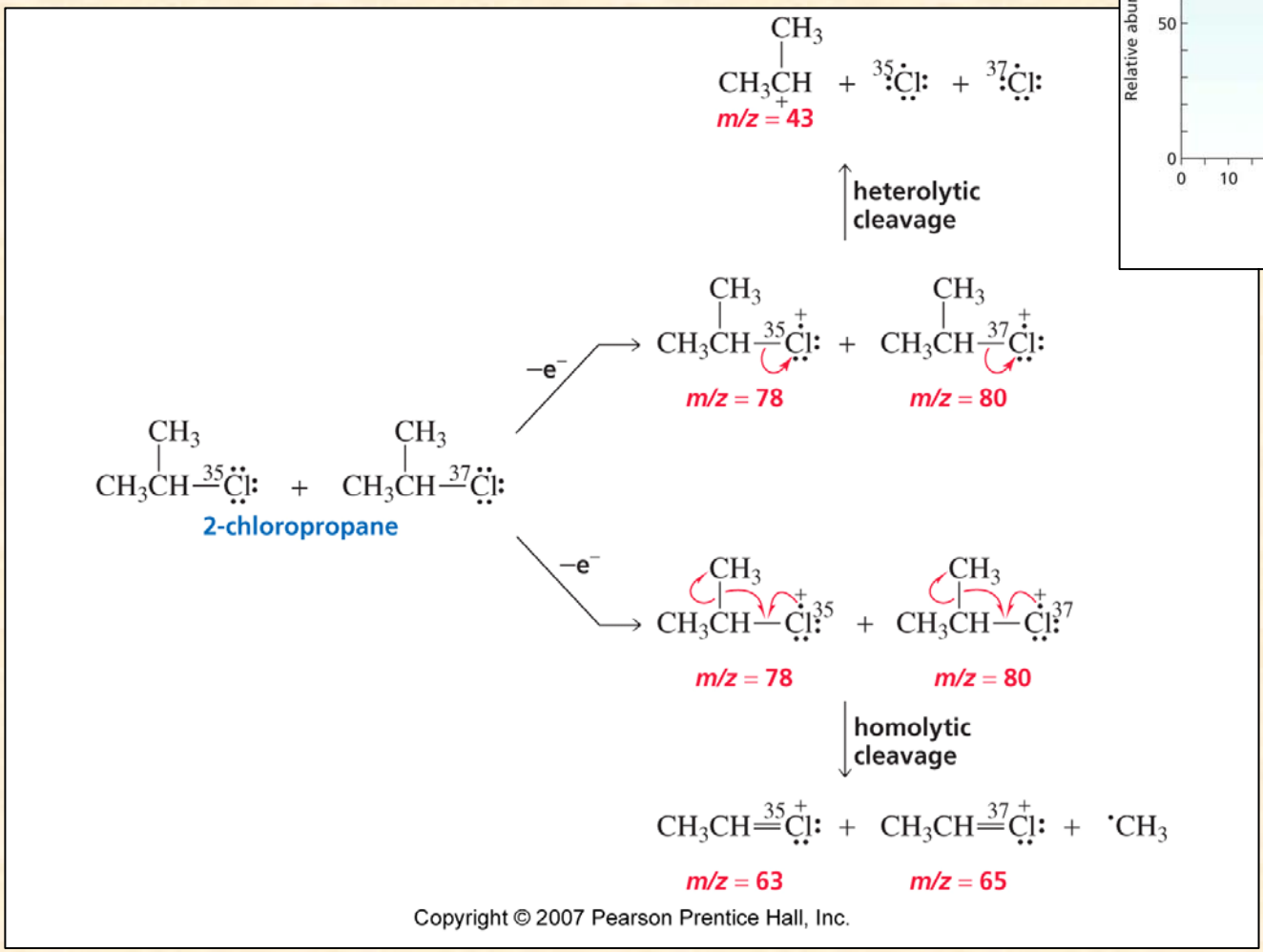


$$CH_3CH{=}\ddot{\underset{\cdot\cdot}{\overset{+}{C}}}l: \longleftrightarrow CH_3\overset{+}{C}H{-}\ddot{C}l$$

General Aspects – Interpretation of Mass Spectra

Fragmentation – 2-Cl-propane





Mass Spectra of the Common Classes of Compounds

Hydrocarbons – Alkanes

- Large compounds show fairly similar spectra (M^+ !)

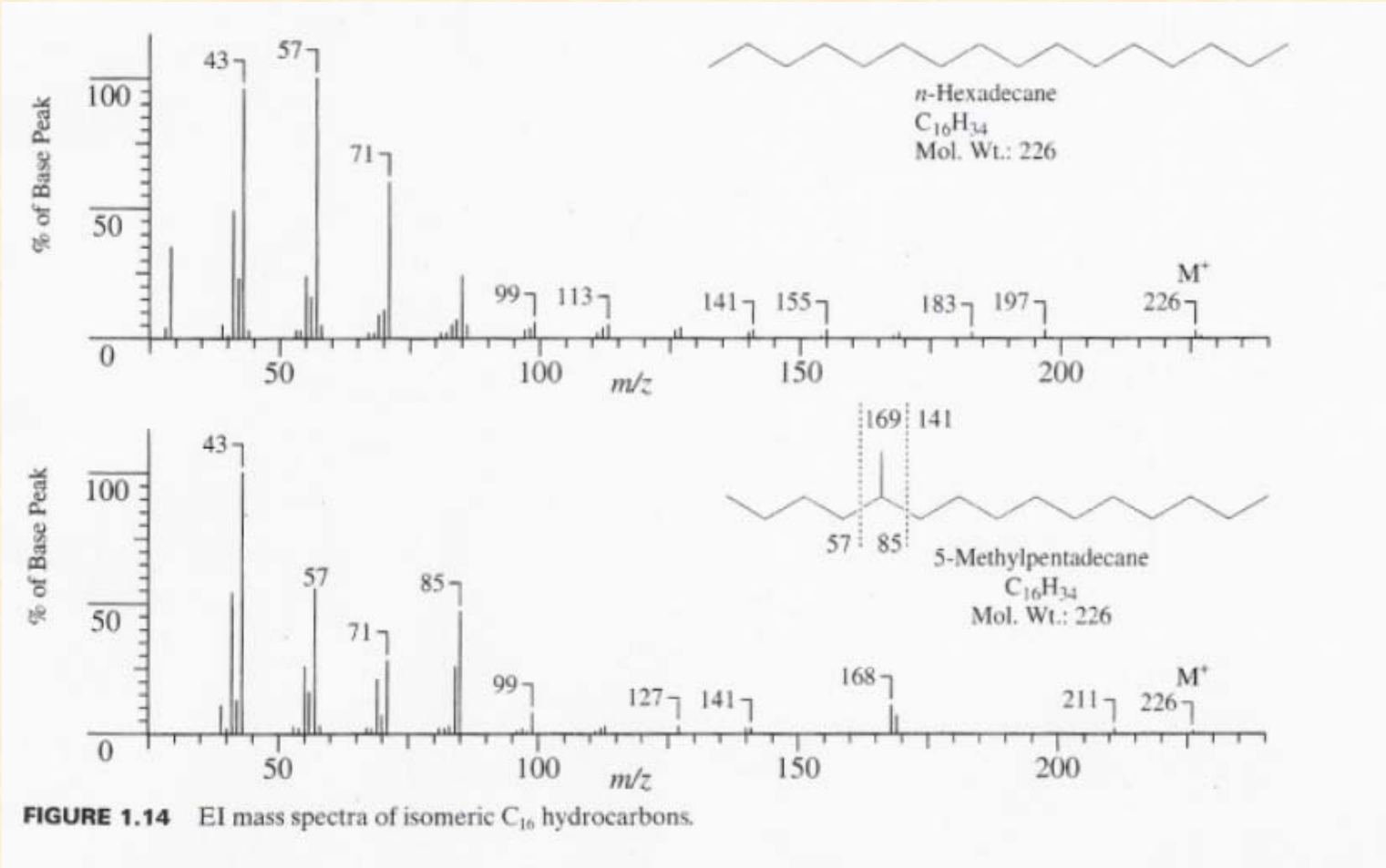


FIGURE 1.14 EI mass spectra of isomeric C_{16} hydrocarbons.

Mass Spectra of the Common Classes of Compounds

Hydrocarbons – Cycloalkanes

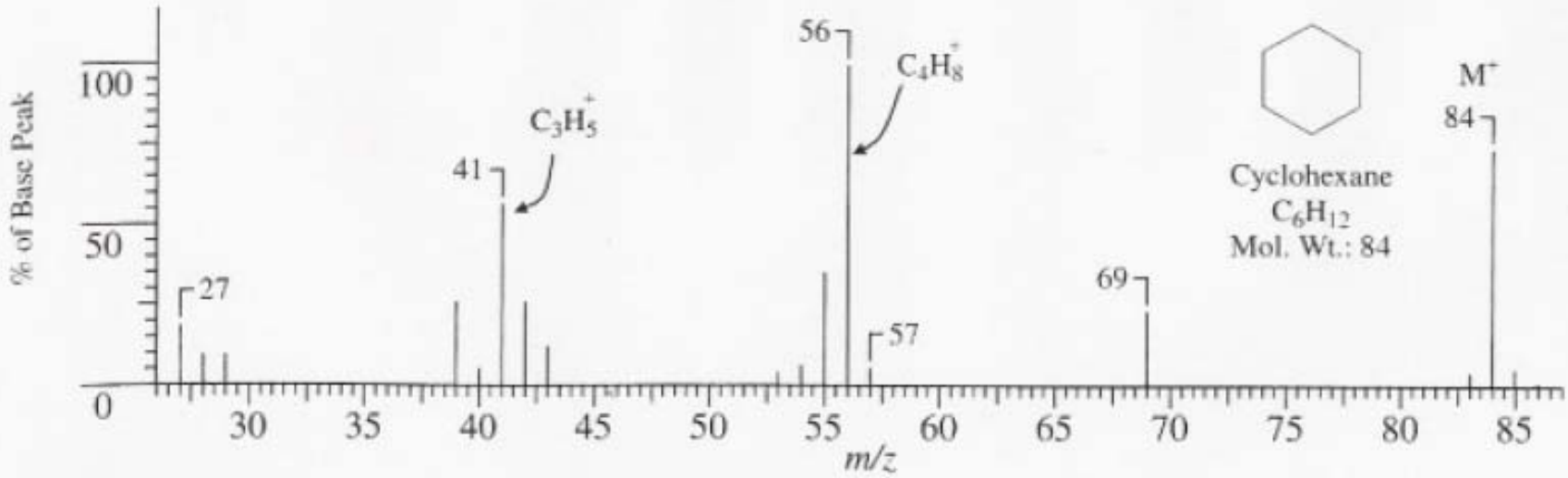


FIGURE 1.15 EI mass spectrum of cyclohexane.

Mass Spectra of the Common Classes of Compounds

Hydrocarbons – Alkenes (olefins)

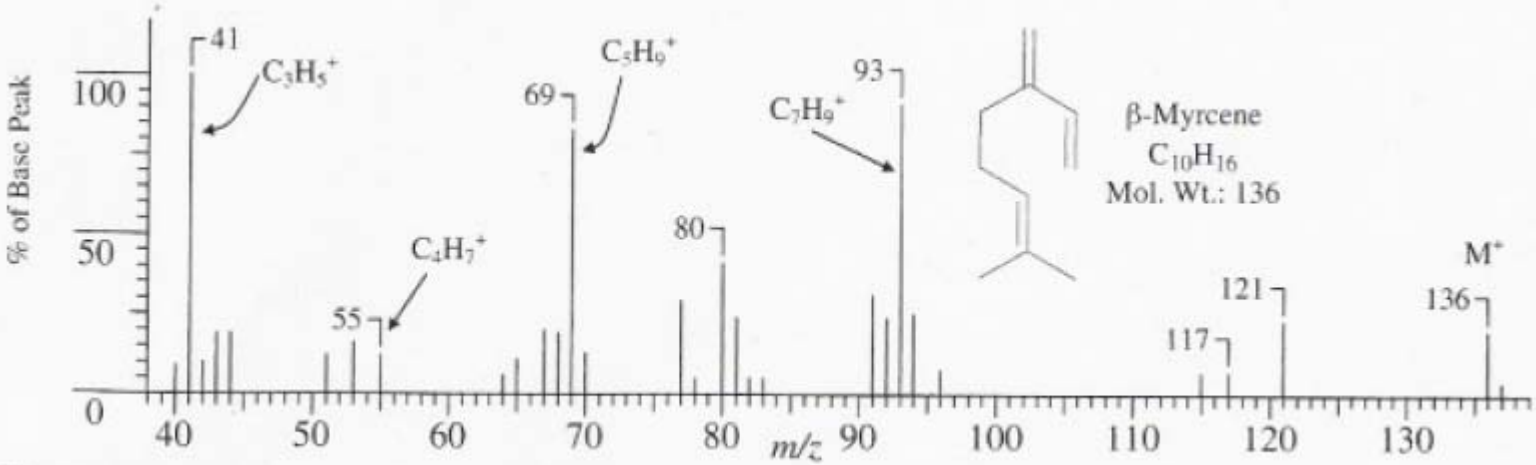
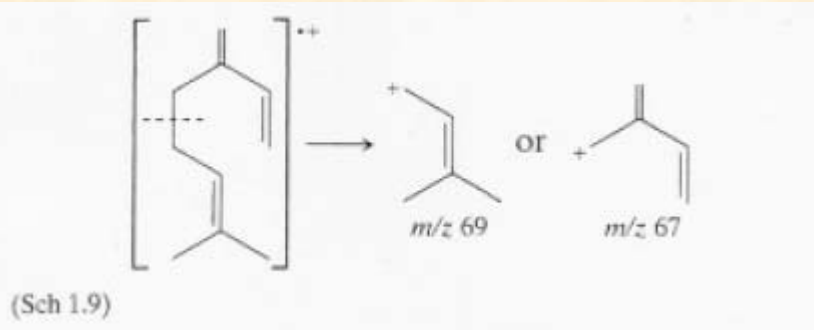
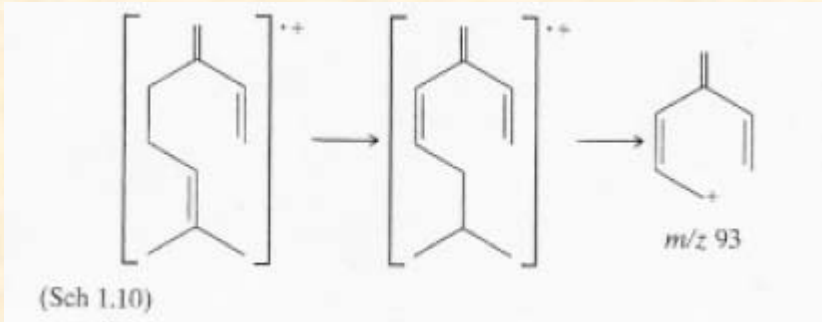


FIGURE 1.16 EI mass spectrum of β-myrcene.





Mass Spectra of the Common Classes of Compounds

Hydrocarbons – Aromatics and alkylaromatics

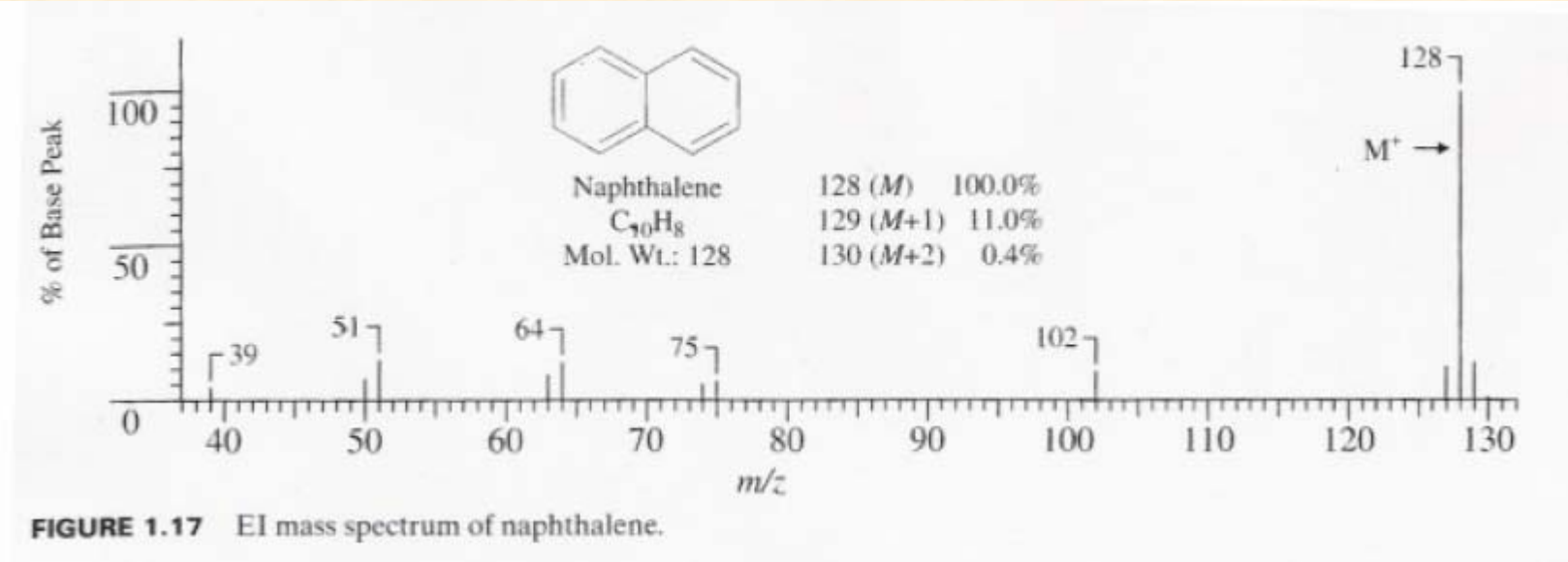
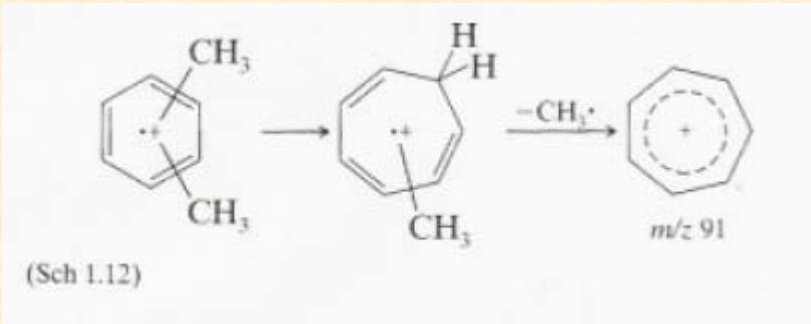
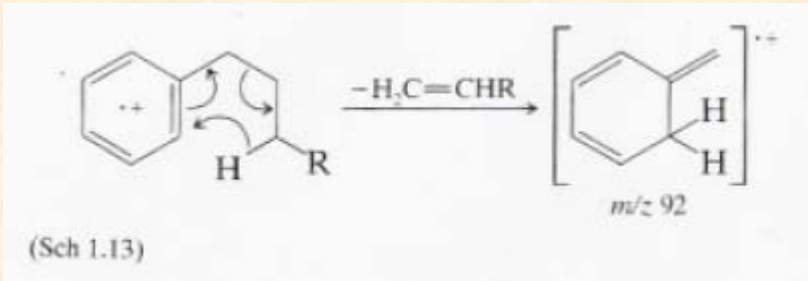


FIGURE 1.17 EI mass spectrum of naphthalene.



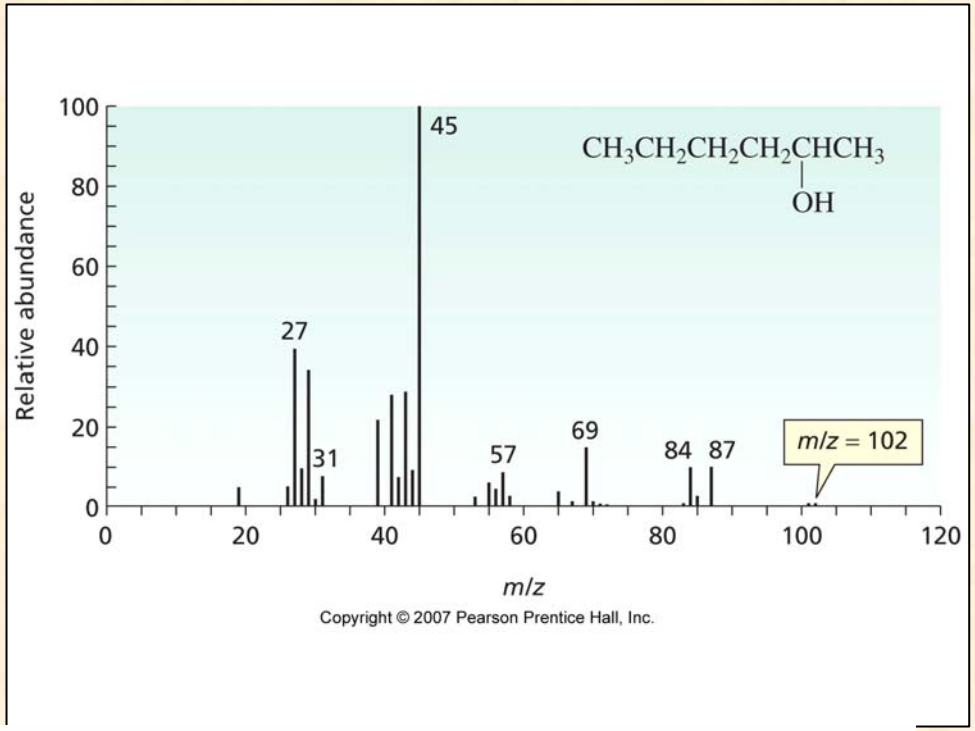
(Sch 1.12)

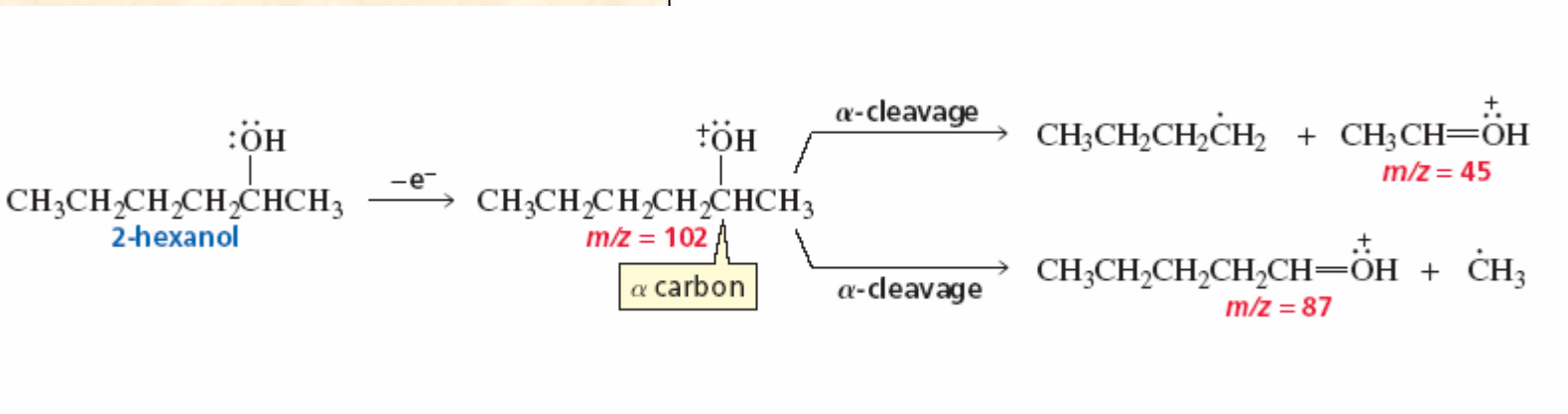


(Sch 1.13)

Mass Spectra of the Common Classes of Compounds

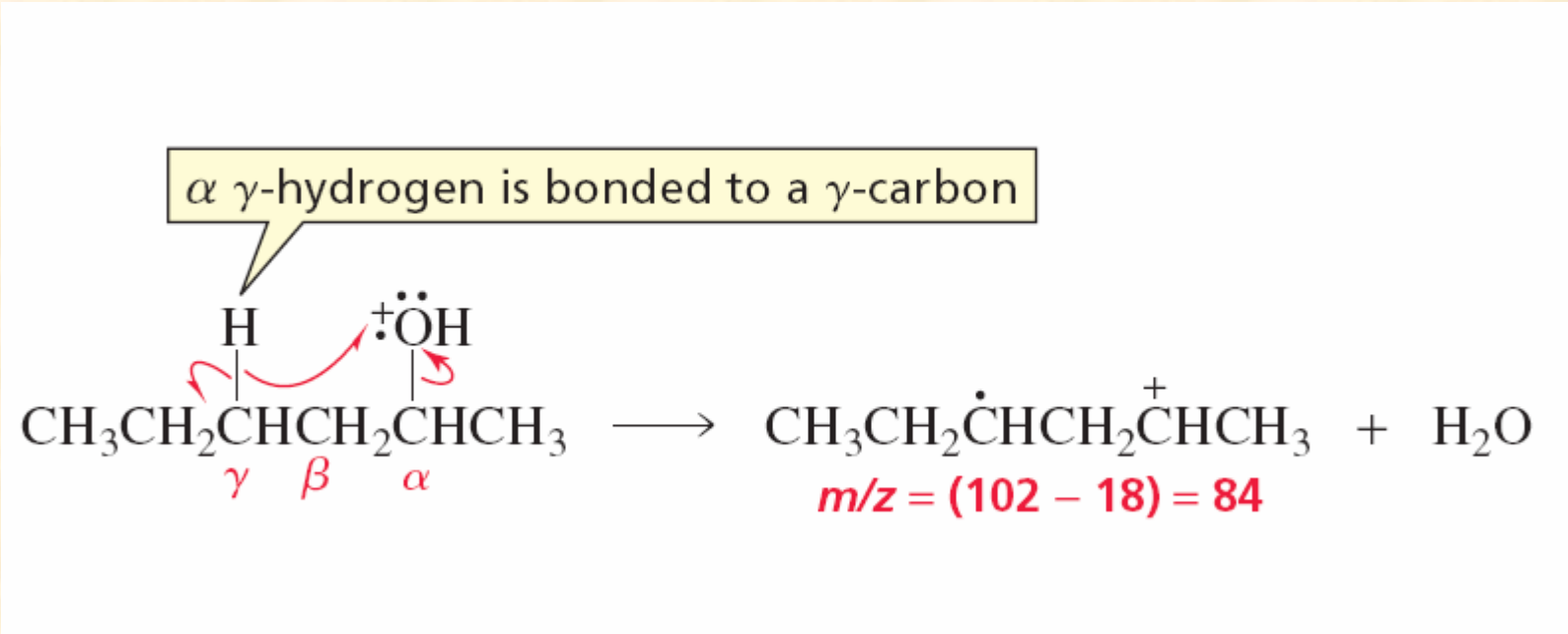
Alcohols





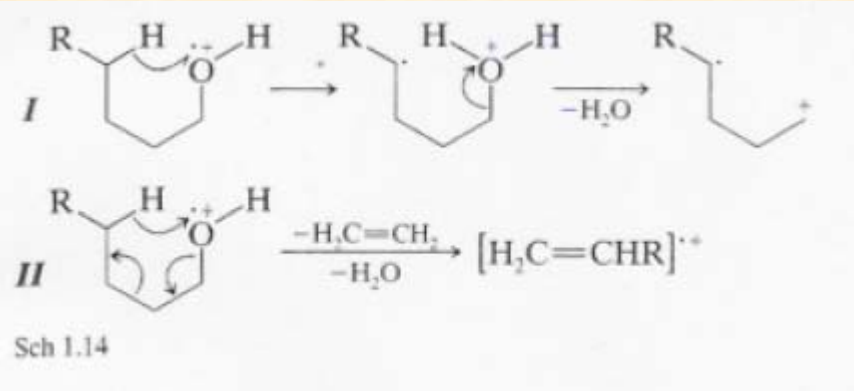
Mass Spectra of the Common Classes of Compounds

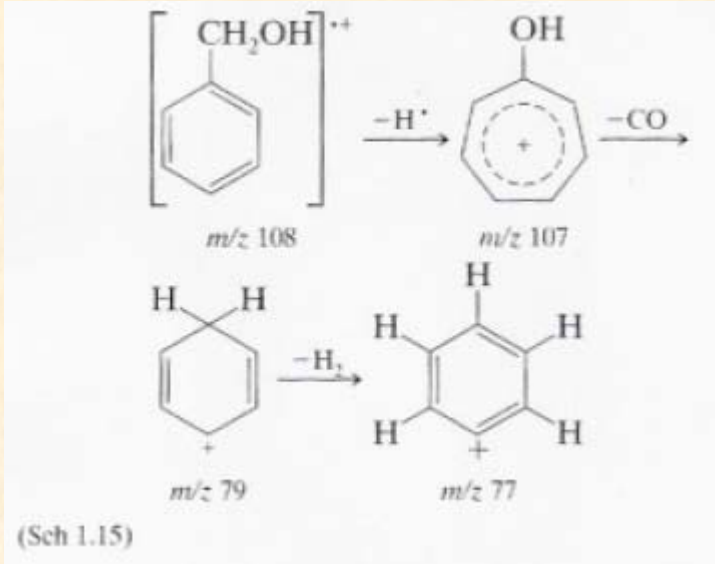
Alcohols

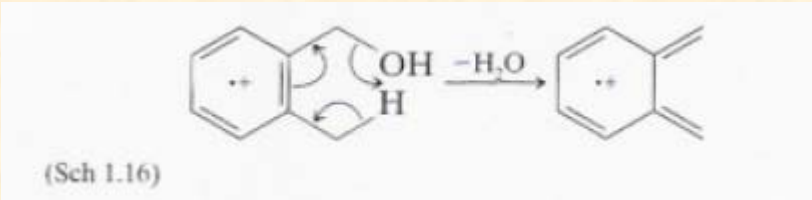


Mass Spectra of the Common Classes of Compounds

Alcohols







Mass Spectra of the Common Classes of Compounds

Alcohols

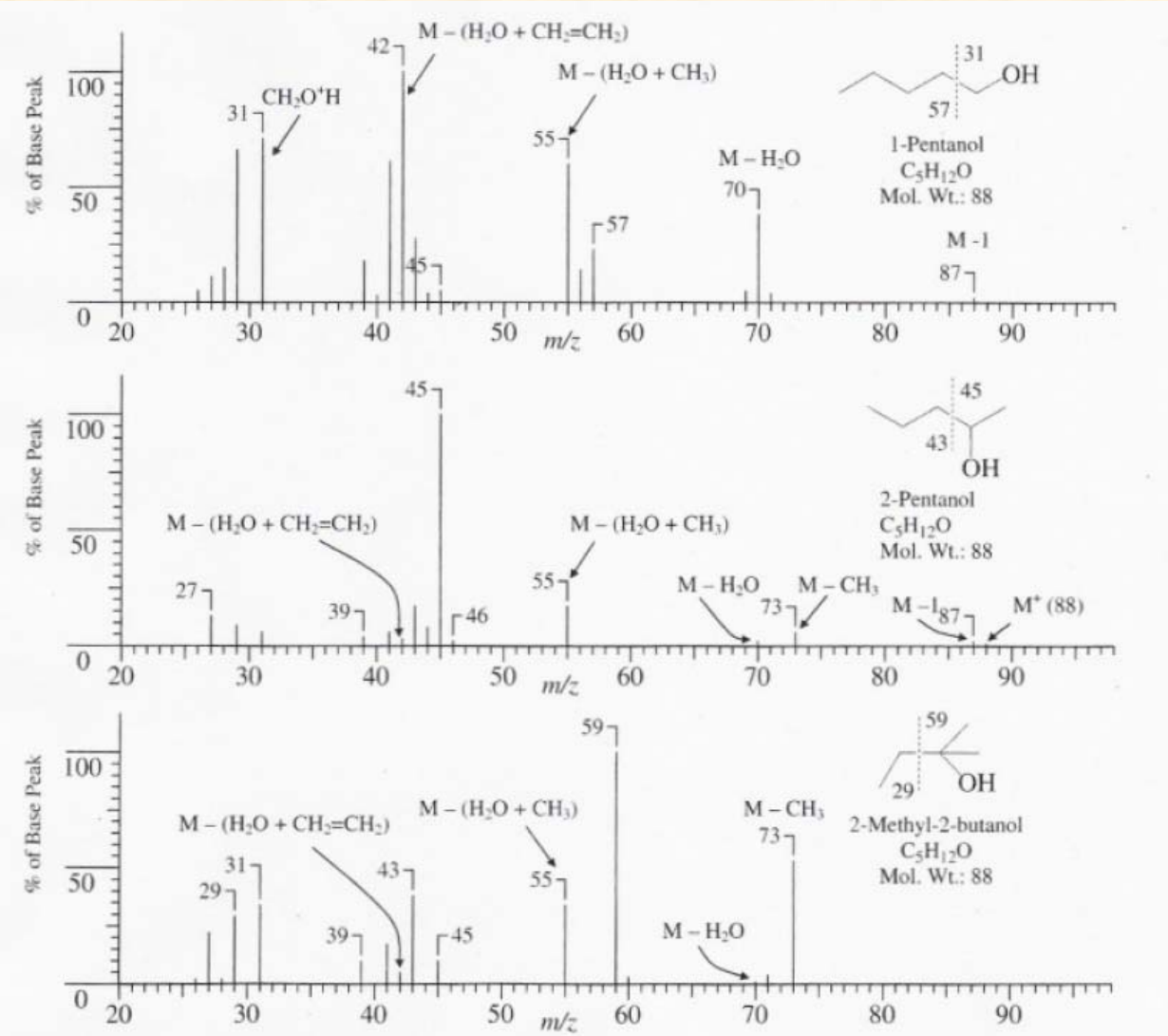


FIGURE 1.18 EI mass spectra of isomeric pentanols.

Mass Spectra of the Common Classes of Compounds

Alcohols (Phenols)

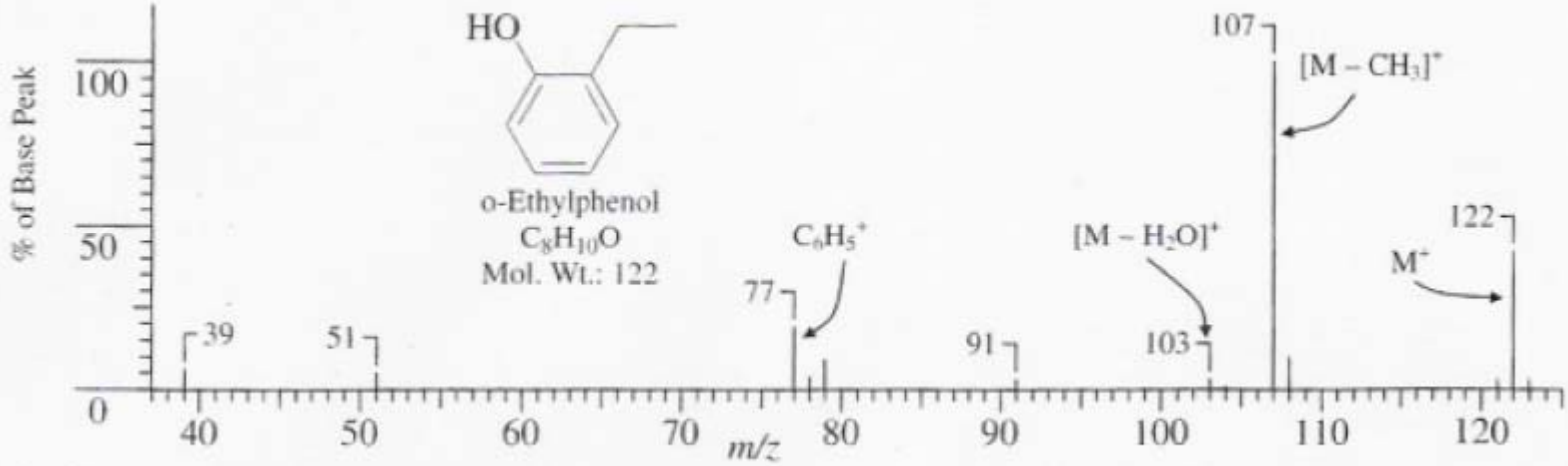
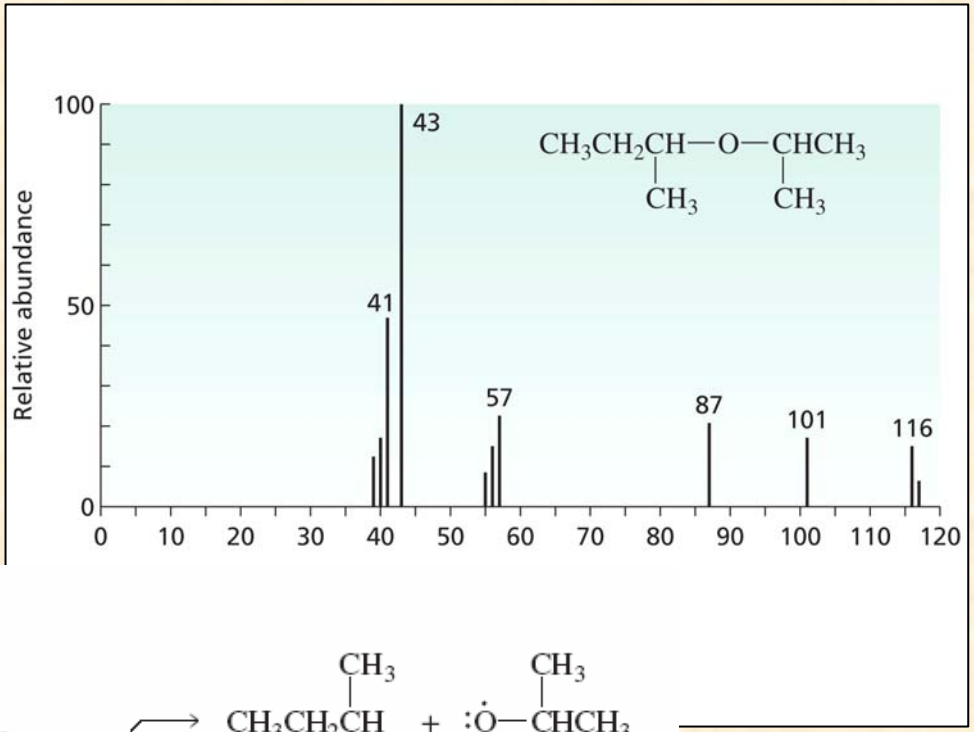
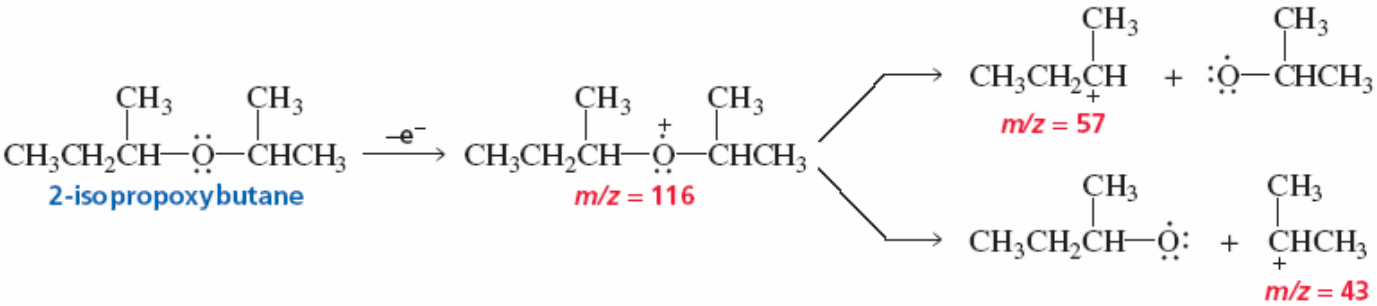


FIGURE 1.19 EI mass spectrum of *o*-ethylphenol.

Mass Spectra of the Common Classes of Compounds

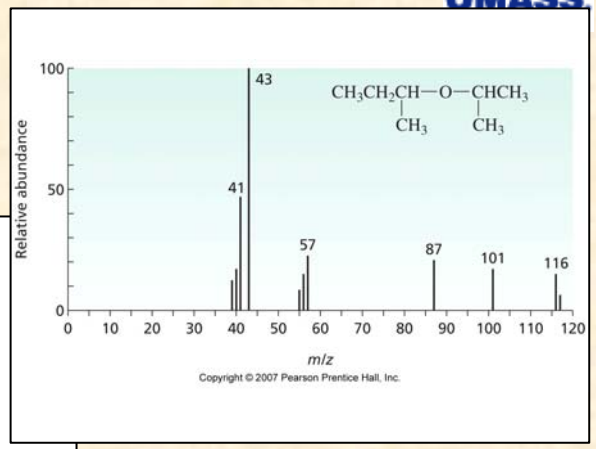
Ethers (aliphatic)

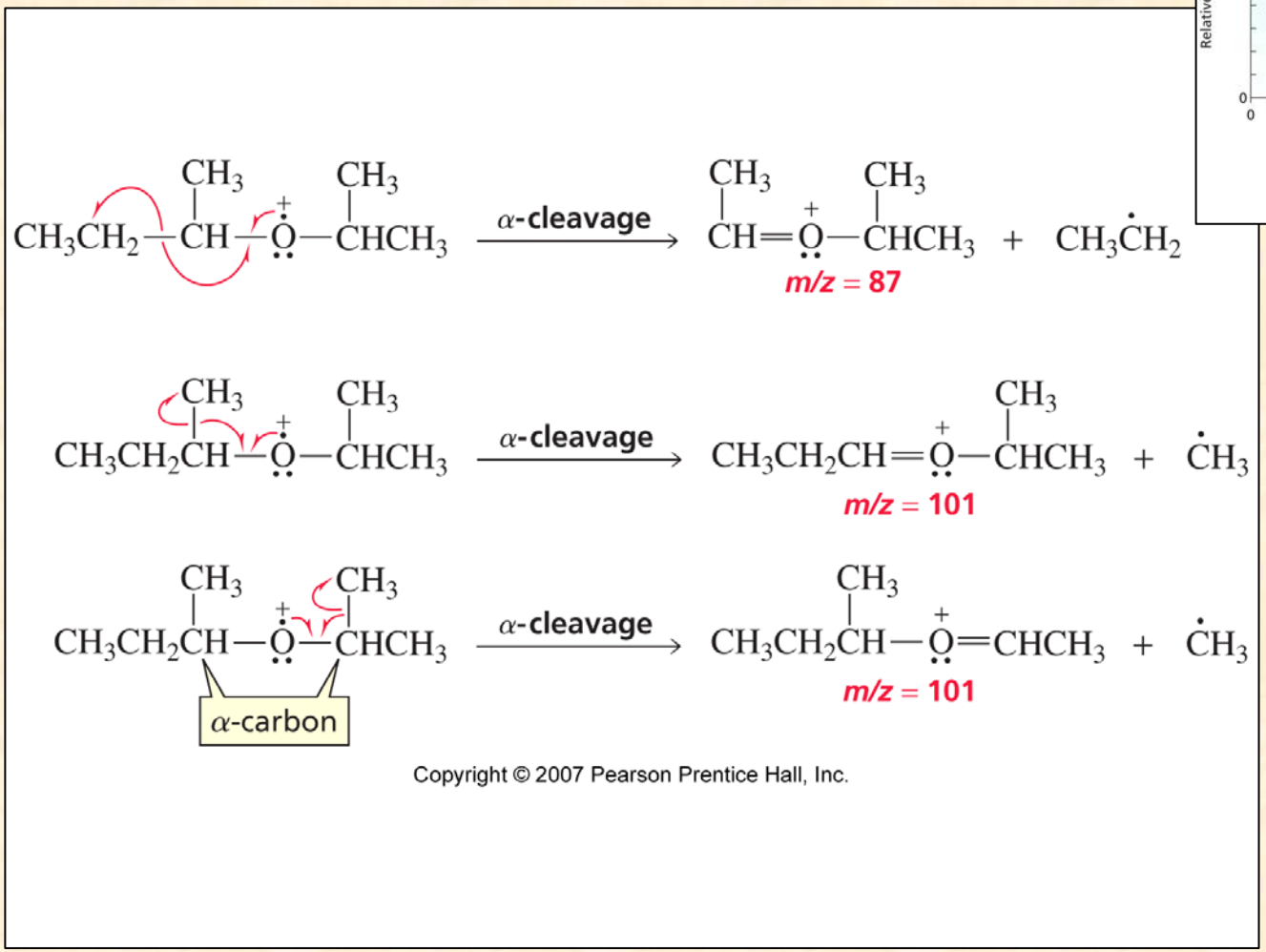




Mass Spectra of the Common Classes of Compounds

Ethers





Mass Spectra of the Common Classes of Compounds

Ethers

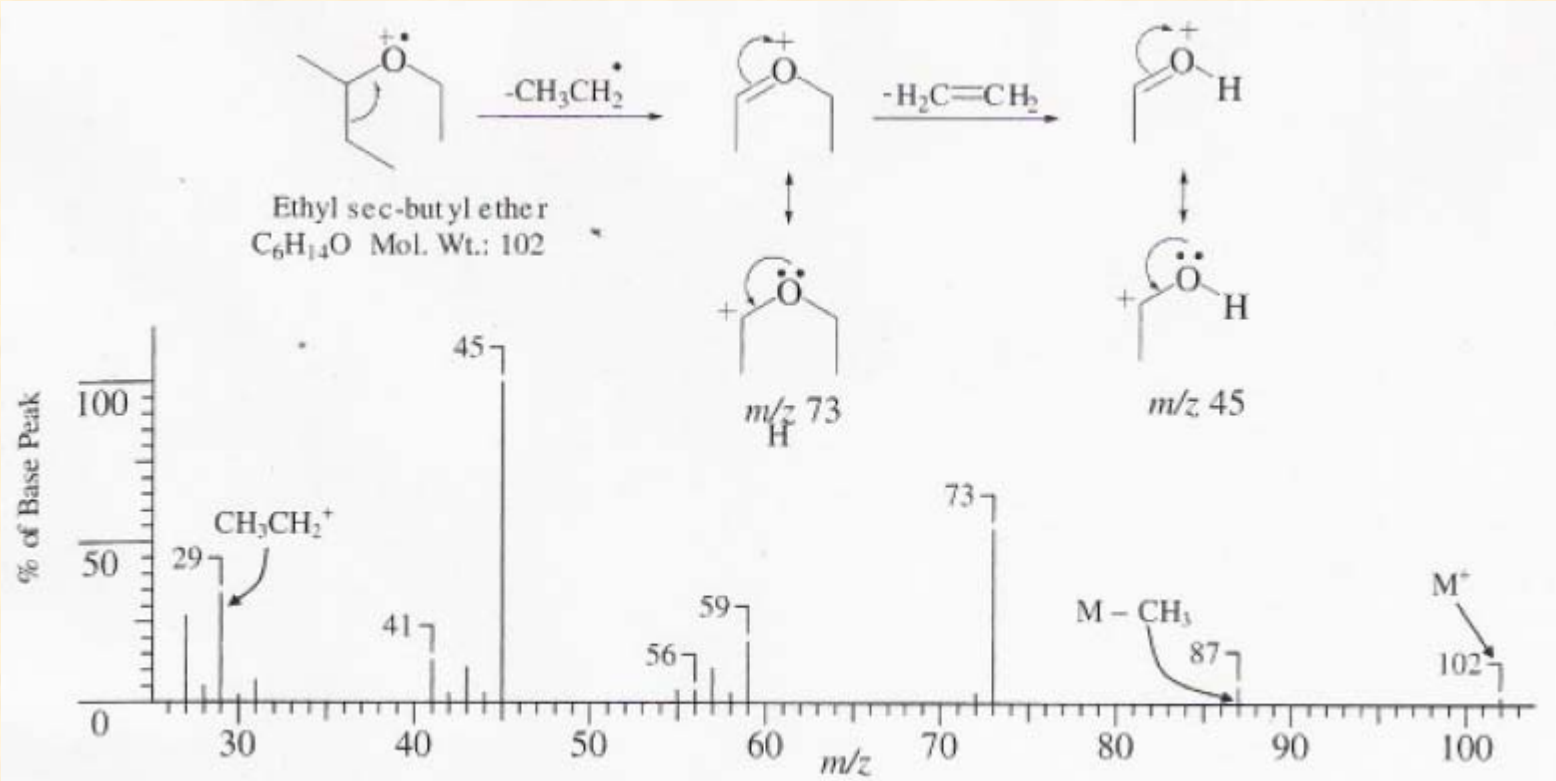
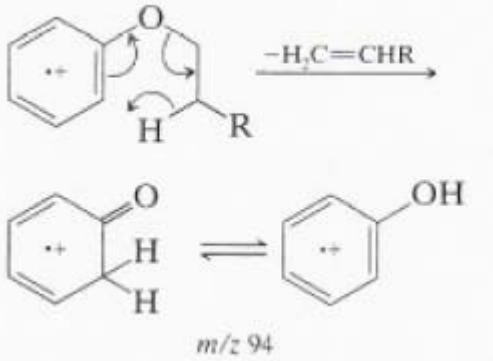


FIGURE 1.20 EI mass spectrum of ethyl *sec*-butyl ether.

Mass Spectra of the Common Classes of Compounds

Aromatic Ethers (phenolethers)



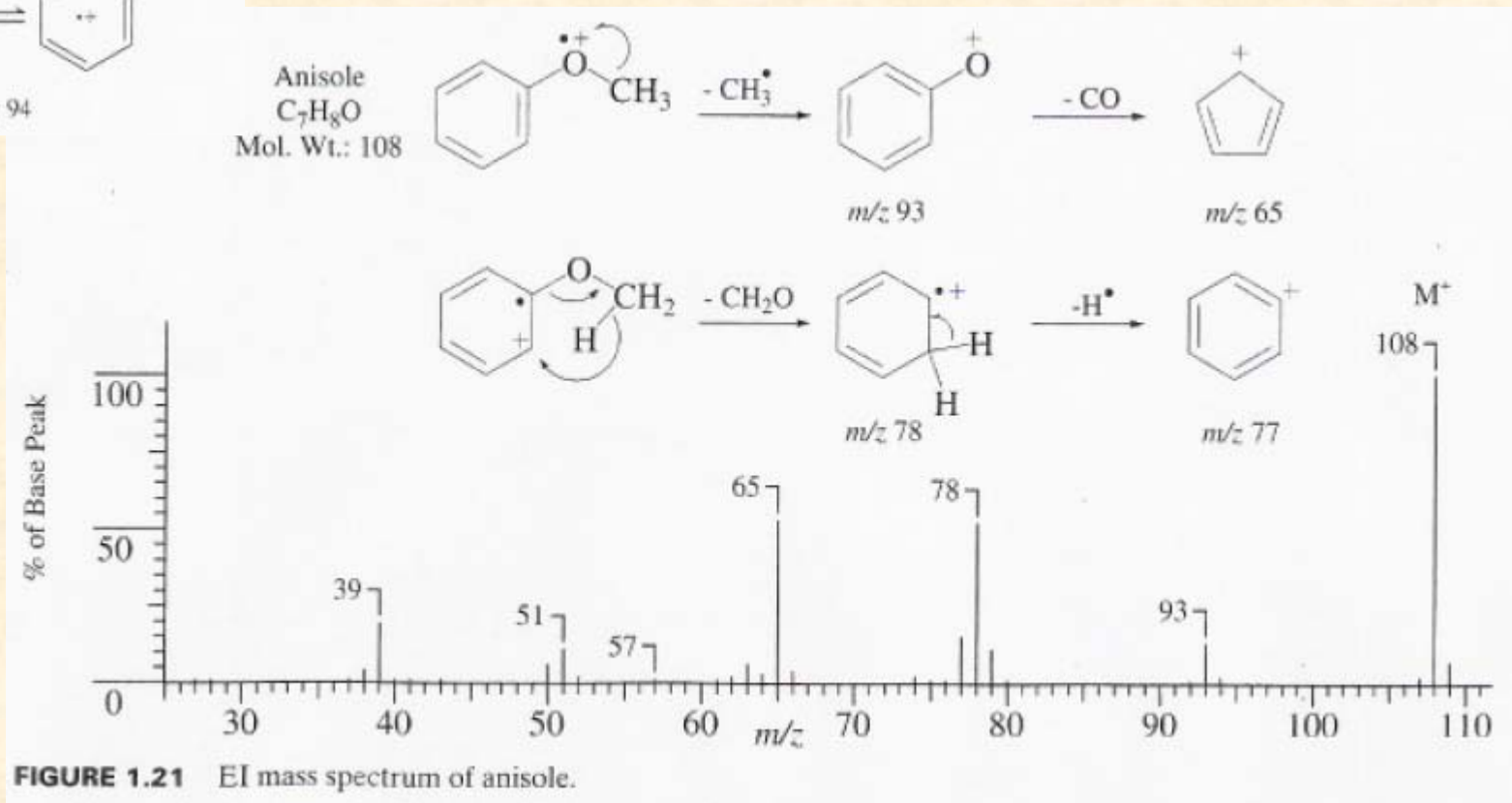
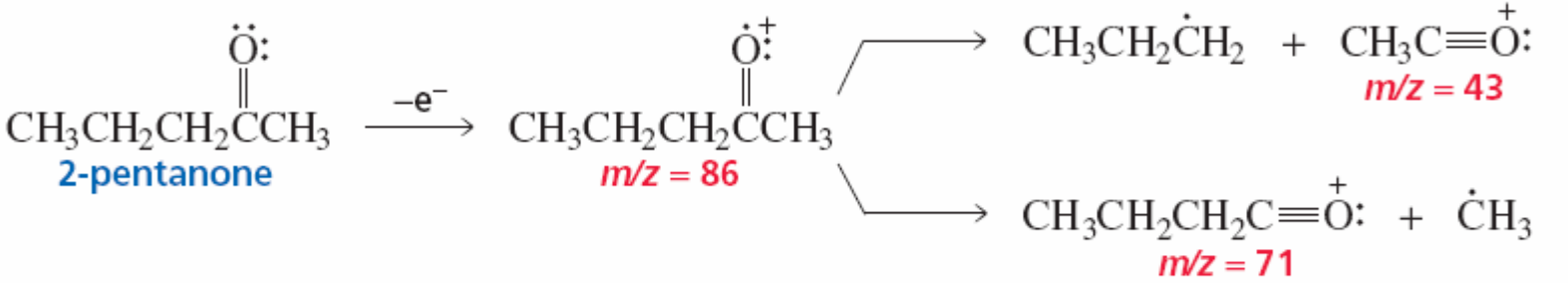
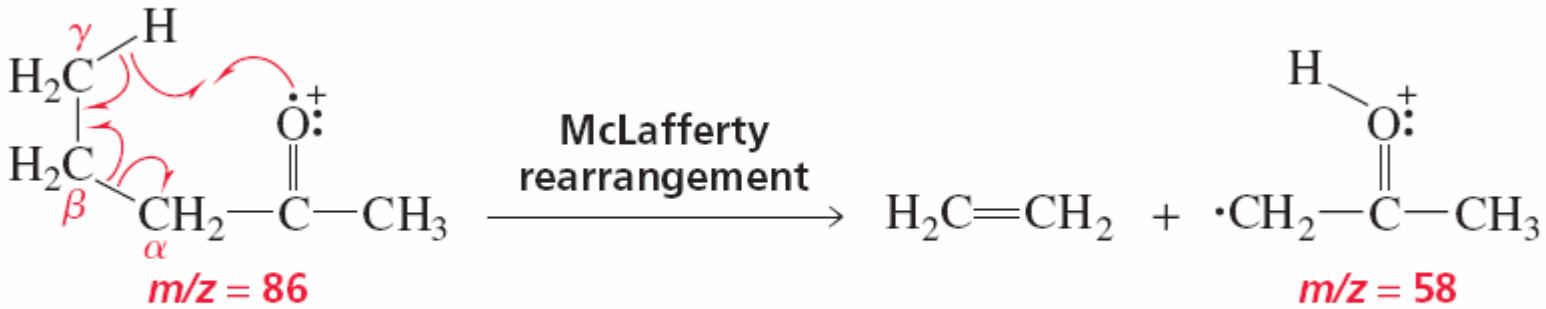


FIGURE 1.21 EI mass spectrum of anisole.

Mass Spectra of the Common Classes of Compounds

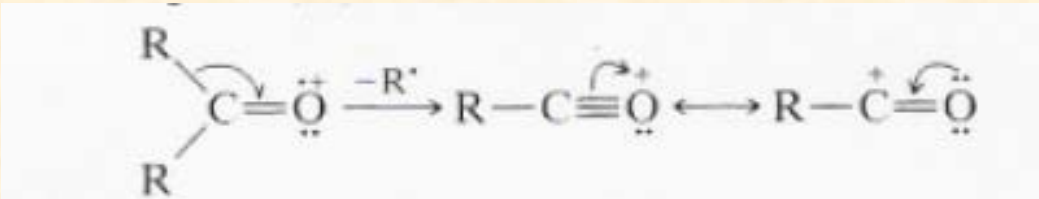
Ketones



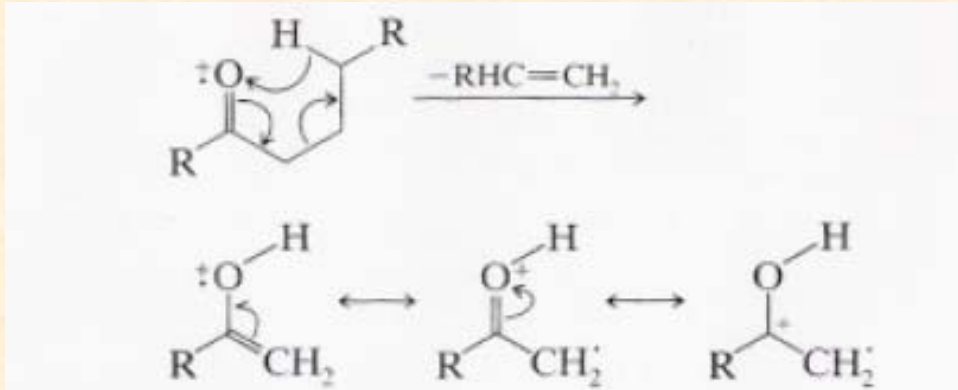


Mass Spectra of the Common Classes of Compounds

Ketones (aliphatic)



(Sch 1.18)



(Sch 1.19)

Mass Spectra of the Common Classes of Compounds

Ketones (cyclic)

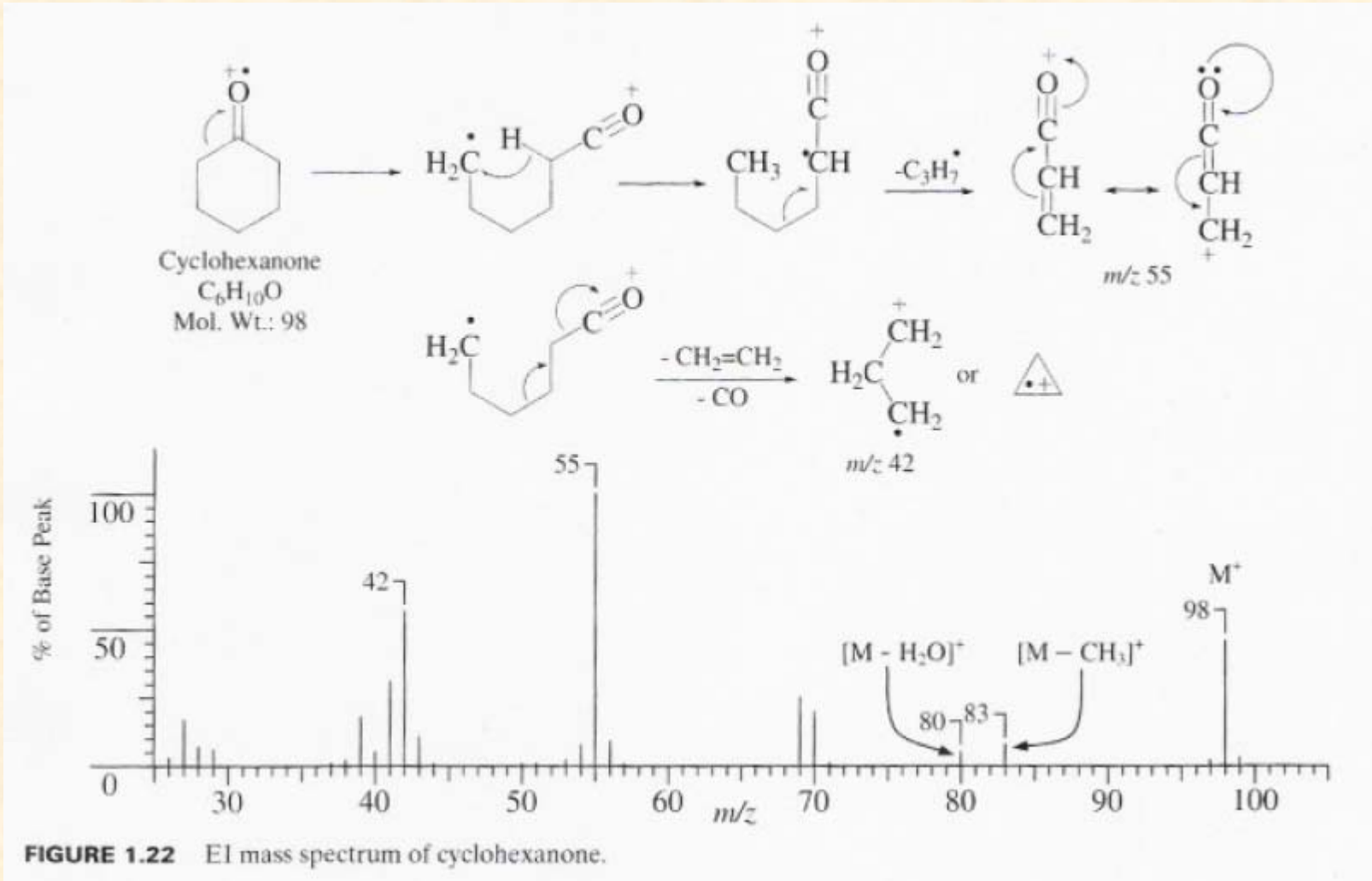


FIGURE 1.22 EI mass spectrum of cyclohexanone.

Mass Spectra of the Common Classes of Compounds

Ketones (aromatic)

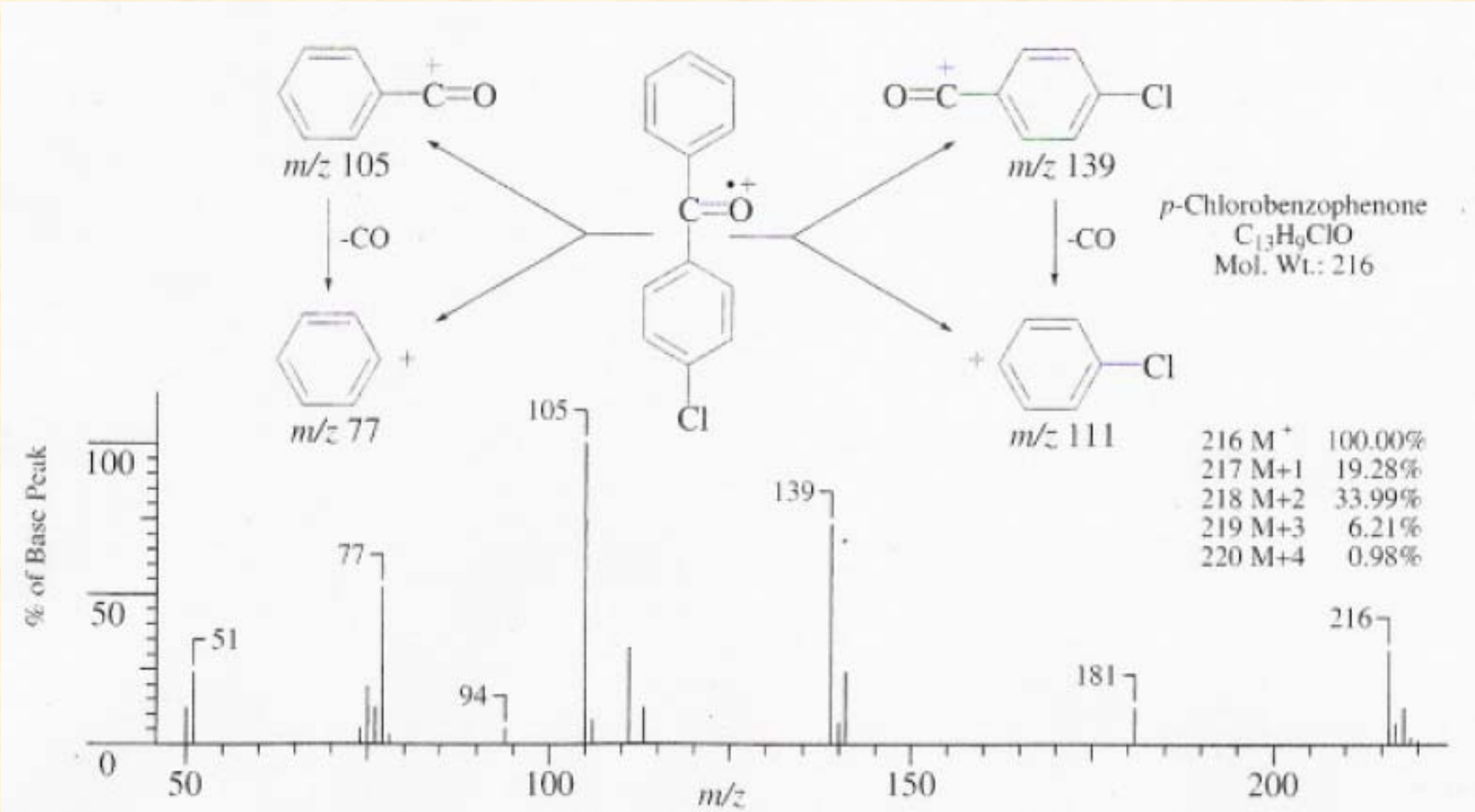
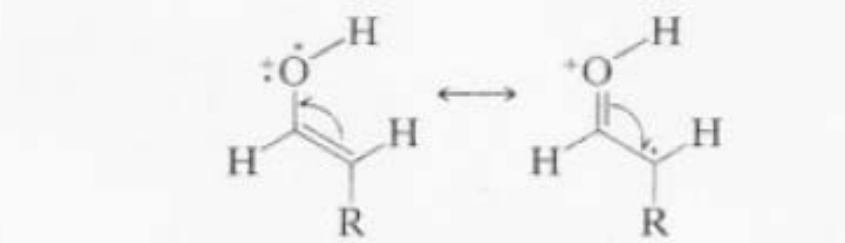


FIGURE 1.23 EI mass spectrum of *p*-chlorobenzophenone.

Mass Spectra of the Common Classes of Compounds

Aldehydes (aliphatic)



(Sch 1.20)

aromatic

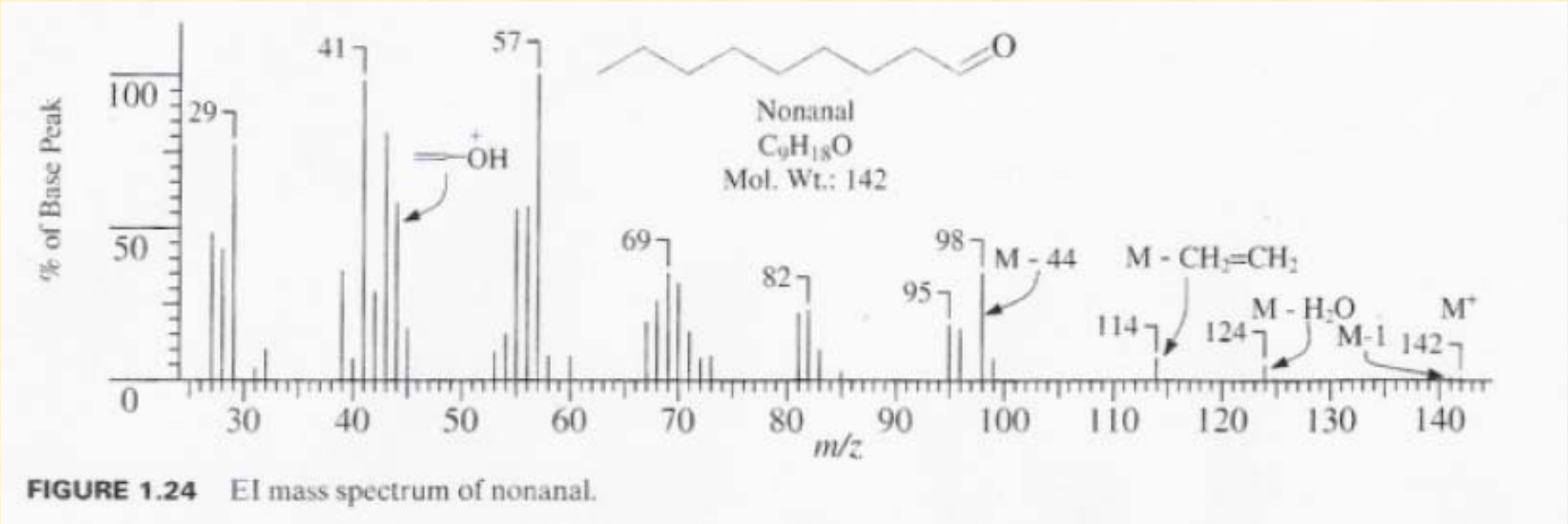
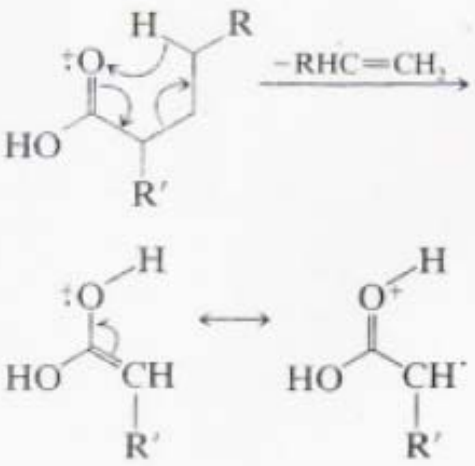


FIGURE 1.24 EI mass spectrum of nonanal.

Mass Spectra of the Common Classes of Compounds

Carboxylic Acids (aliphatic)



(Sch 1.21)

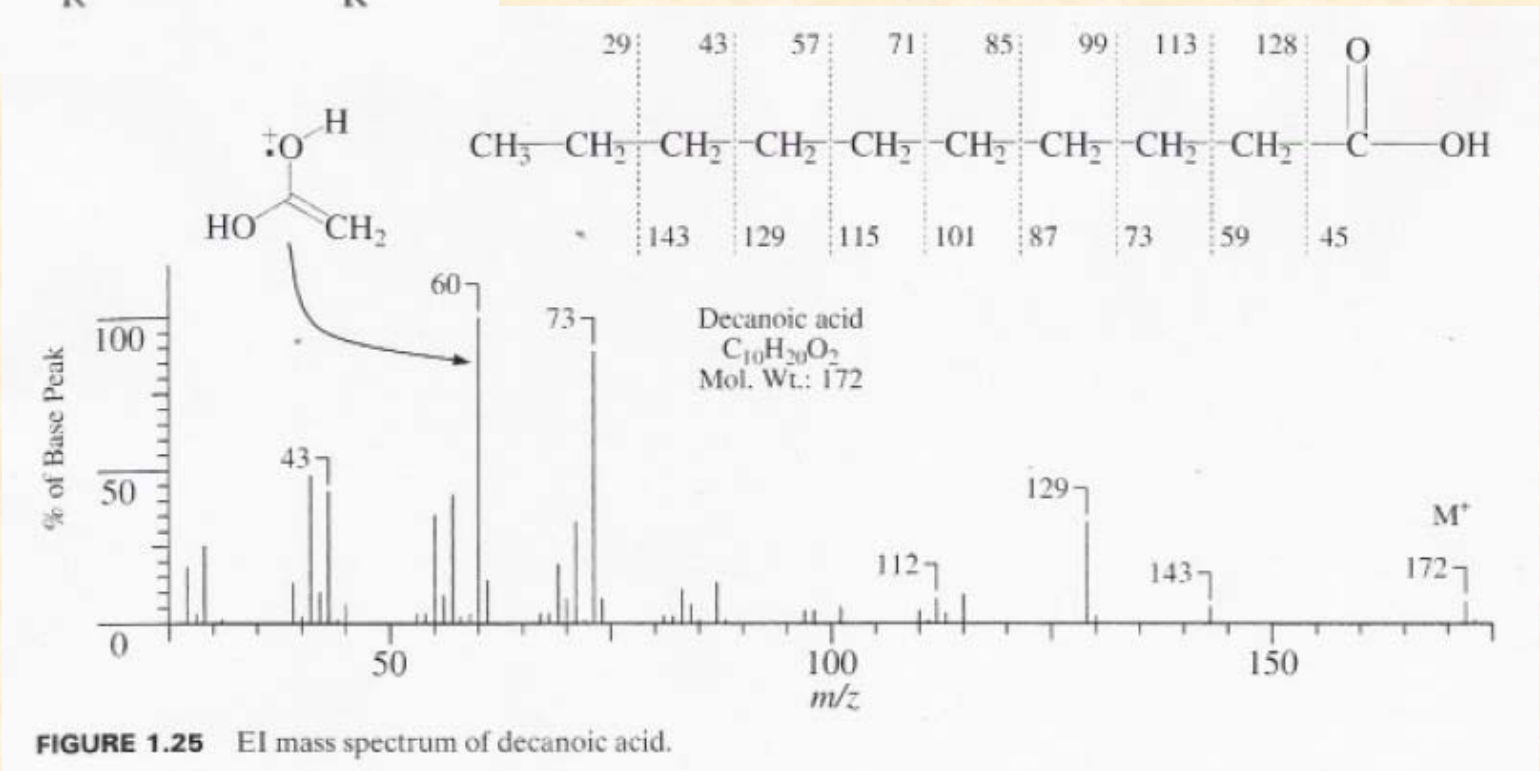
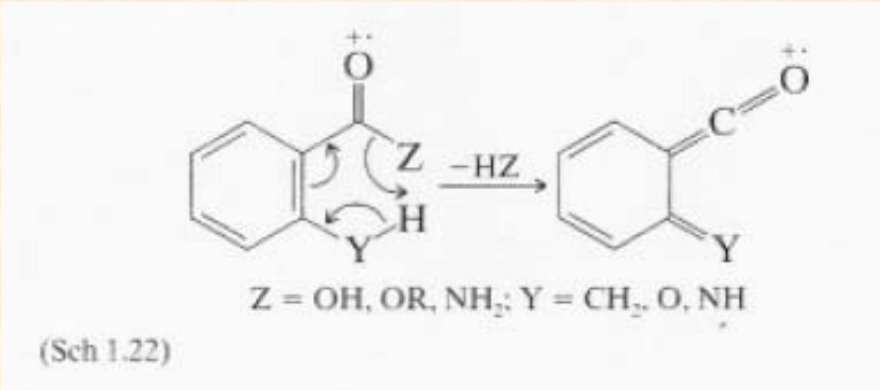


FIGURE 1.25 EI mass spectrum of decanoic acid.

Mass Spectra of the Common Classes of Compounds

Carboxylic Acids (aromatic)

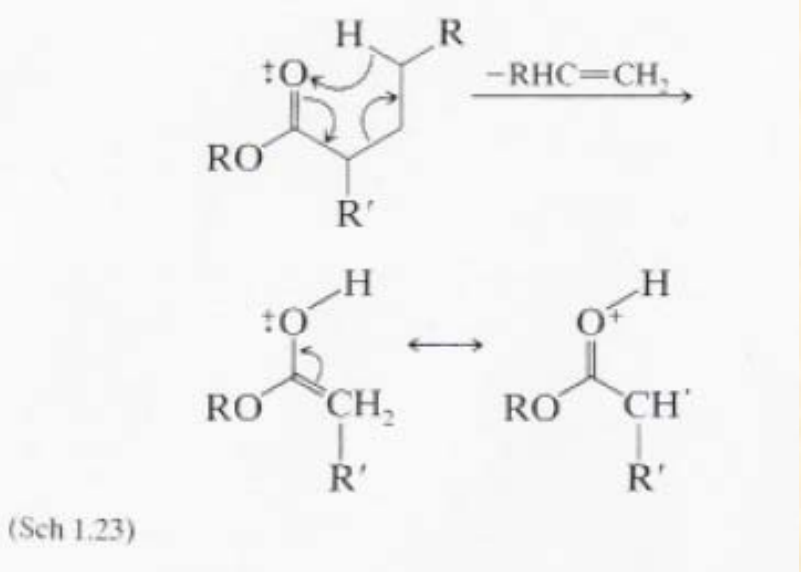


Z = OH, OR, NH_2; Y = CH_2, O, NH

(Sch 1.22)

Mass Spectra of the Common Classes of Compounds

Carboxylic Acid Esters (aliphatic)



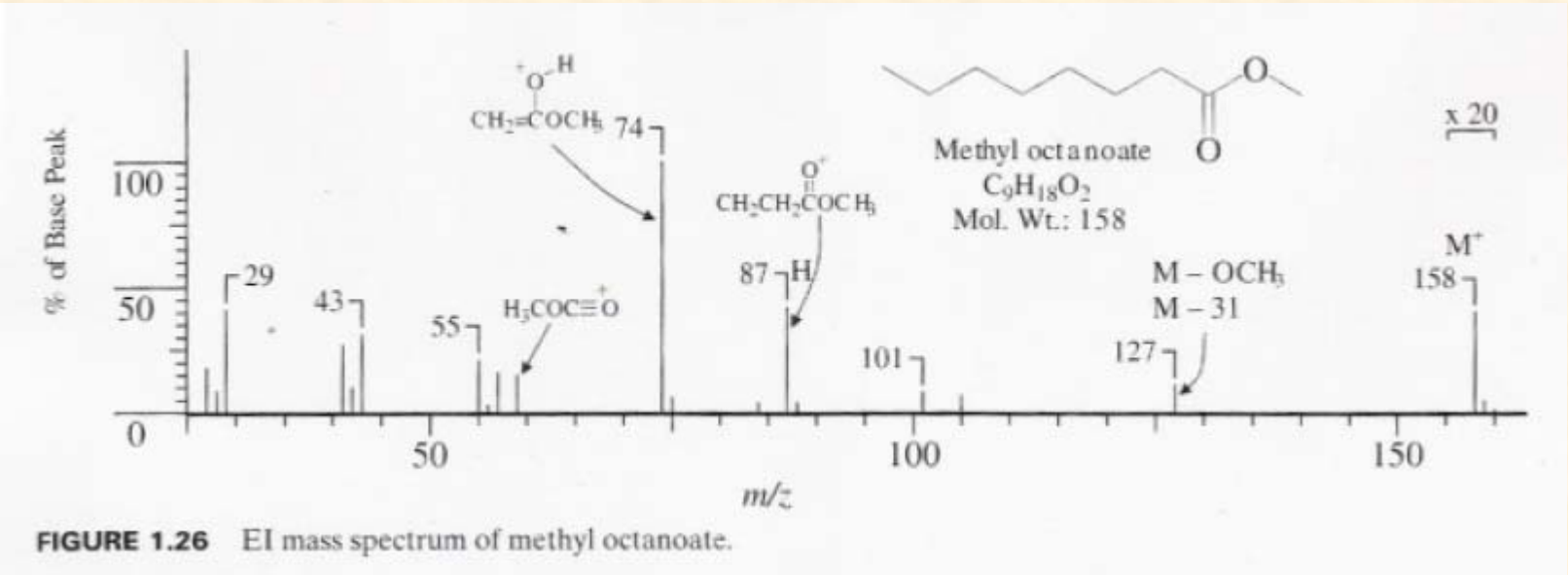
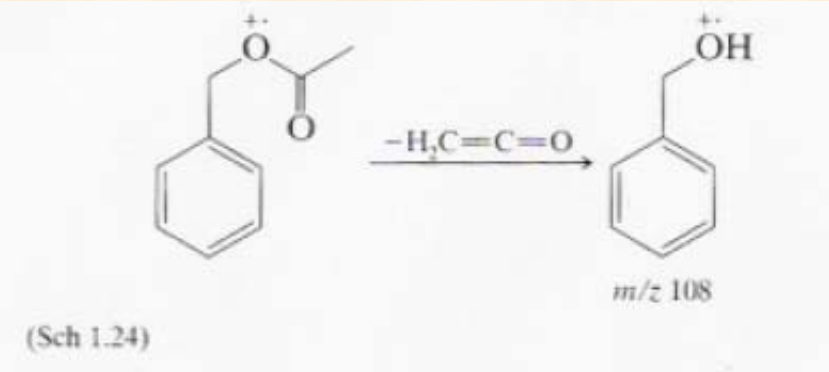


FIGURE 1.26 EI mass spectrum of methyl octanoate.

Mass Spectra of the Common Classes of Compounds

Carboxylic Acid Esters (benzyl, phenyl esters, aromatic acids)



$(ArCOOH)^+$ $(ArCOOH_2)^+$

Mass Spectra of the Common Classes of Compounds

Carboxylic Acid Esters (lactones)

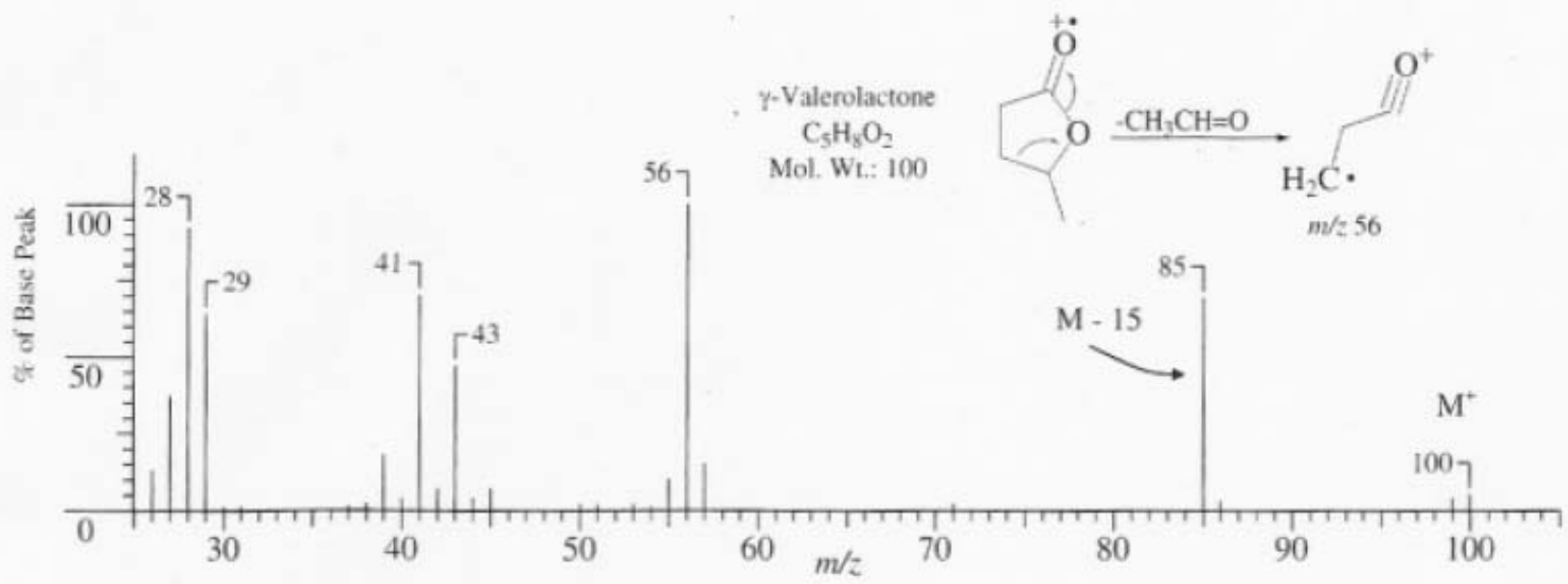
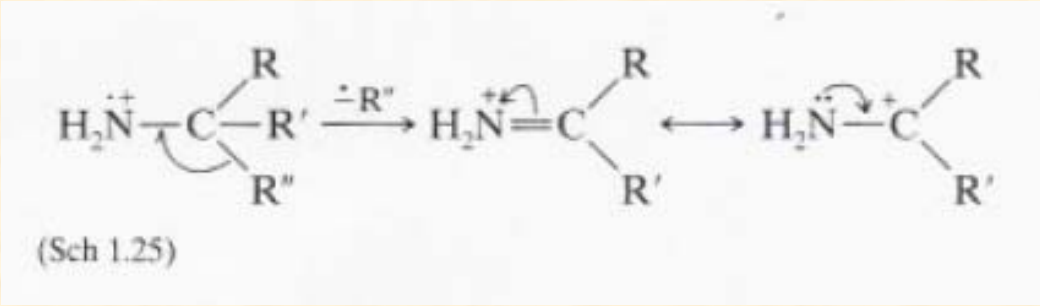


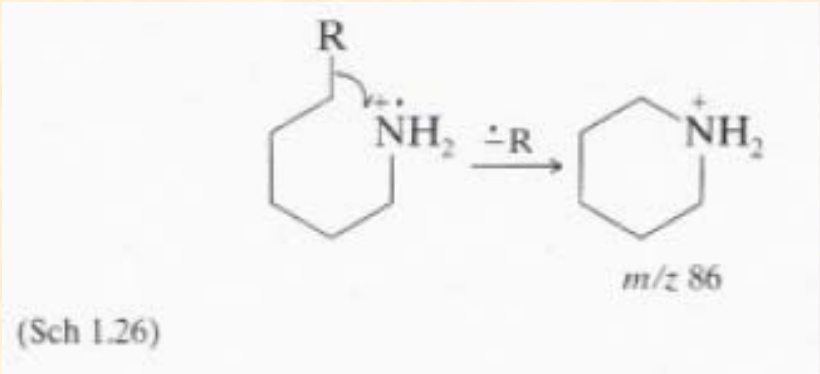
FIGURE 1.27 EI mass spectrum of γ-valerolactone.

Mass Spectra of the Common Classes of Compounds

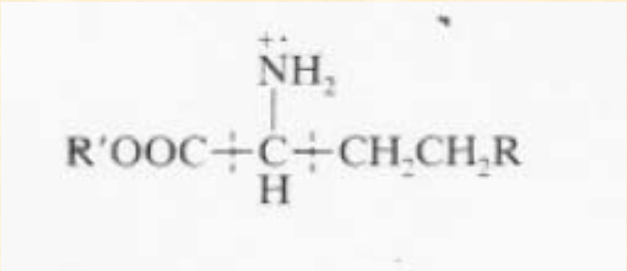
Amines (aliphatic)



(Sch 1.25)



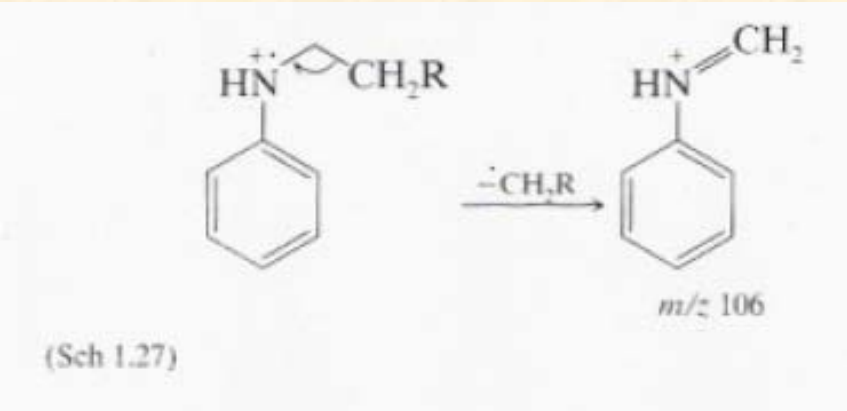
(Sch 1.26)



Mass Spectra of the Common Classes of Compounds

Amines (cyclic and aromatic)

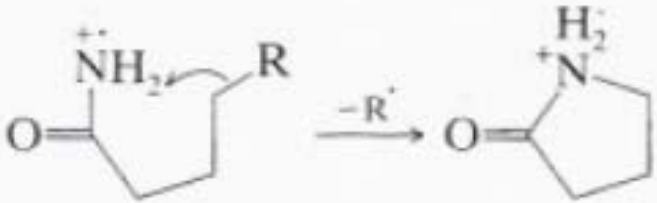
$\cdot CH_2—{}^{+}NH{=}CH_2$ (m/z 43



(Sch 1.27)

Mass Spectra of the Common Classes of Compounds

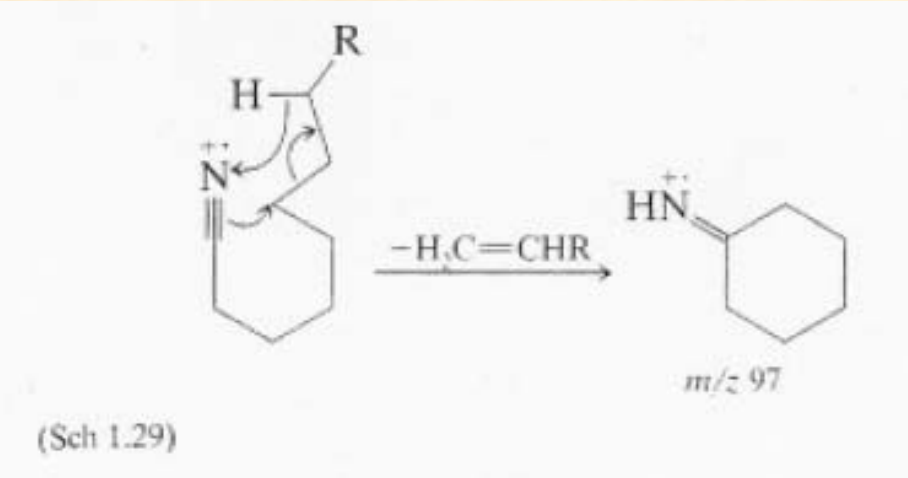
Amides



(Sch 1.28)

Mass Spectra of the Common Classes of Compounds

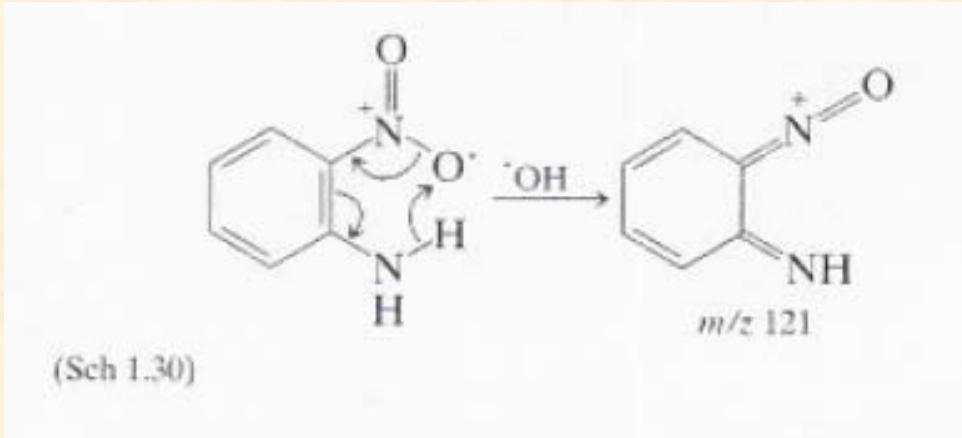
Nitriles



(Sch 1.29)

Mass Spectra of the Common Classes of Compounds

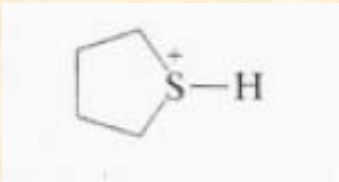
Nitro compounds

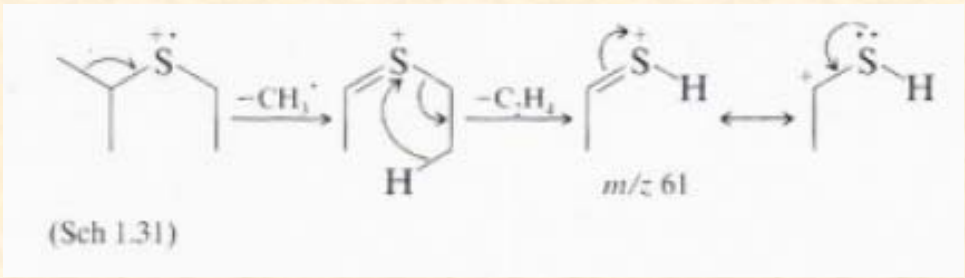


(Sch 1.30)

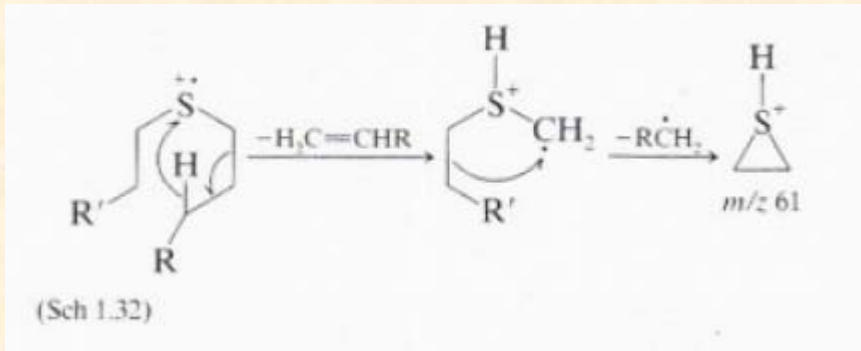
Mass Spectra of the Common Classes of Compounds

Sulfur compounds (thiols, sulfides)

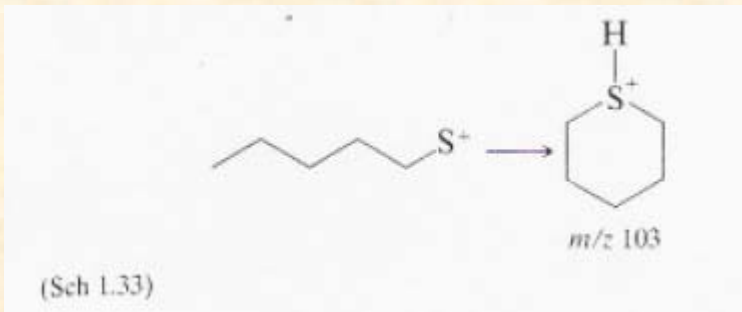




(Sch 1.31)



(Sch 1.32)



(Sch 1.33)

Mass Spectra of the Common Classes of Compounds

Sulfur compounds (thiols, sulfides)

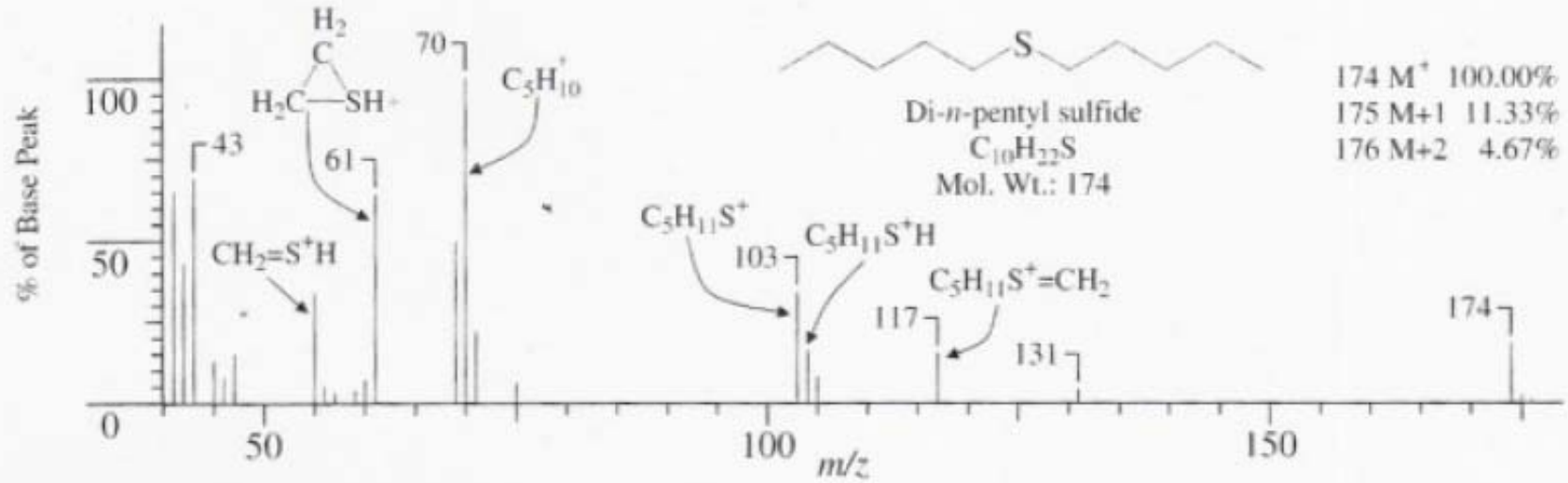


FIGURE 1.28 EI mass spectrum of di-*n*-pentyl sulfide.

Mass Spectra of the Common Classes of Compounds

Halogen compounds

TABLE 1.5 Intensities of Isotope Peaks (Relative to the Molecular Ion) for Combination of Chlorine and Bromine.

Halogen Present	% M+2	% M+4	% M+6	% M+8	% M+10	% M+12
Cl	32.6					
Cl_2	65.3	10.6				
Cl_3	97.8	31.9	3.5			
Cl_4	131.0	63.9	14.0	1.2		
Cl_5	163.0	106.0	34.7	5.7	0.4	
Cl_6	196.0	161.0	69.4	17.0	2.2	0.1
Br	97.9					
Br_2	195.0	95.5				
Br_3	293.0	286.0	93.4			
BrCl	130.0	31.9				
$BrCl_2$	163.0	74.4	10.4			
Br_2Cl	228.0	159.0	31.2			

Mass Spectra of the Common Classes of Compounds

Halogen compounds

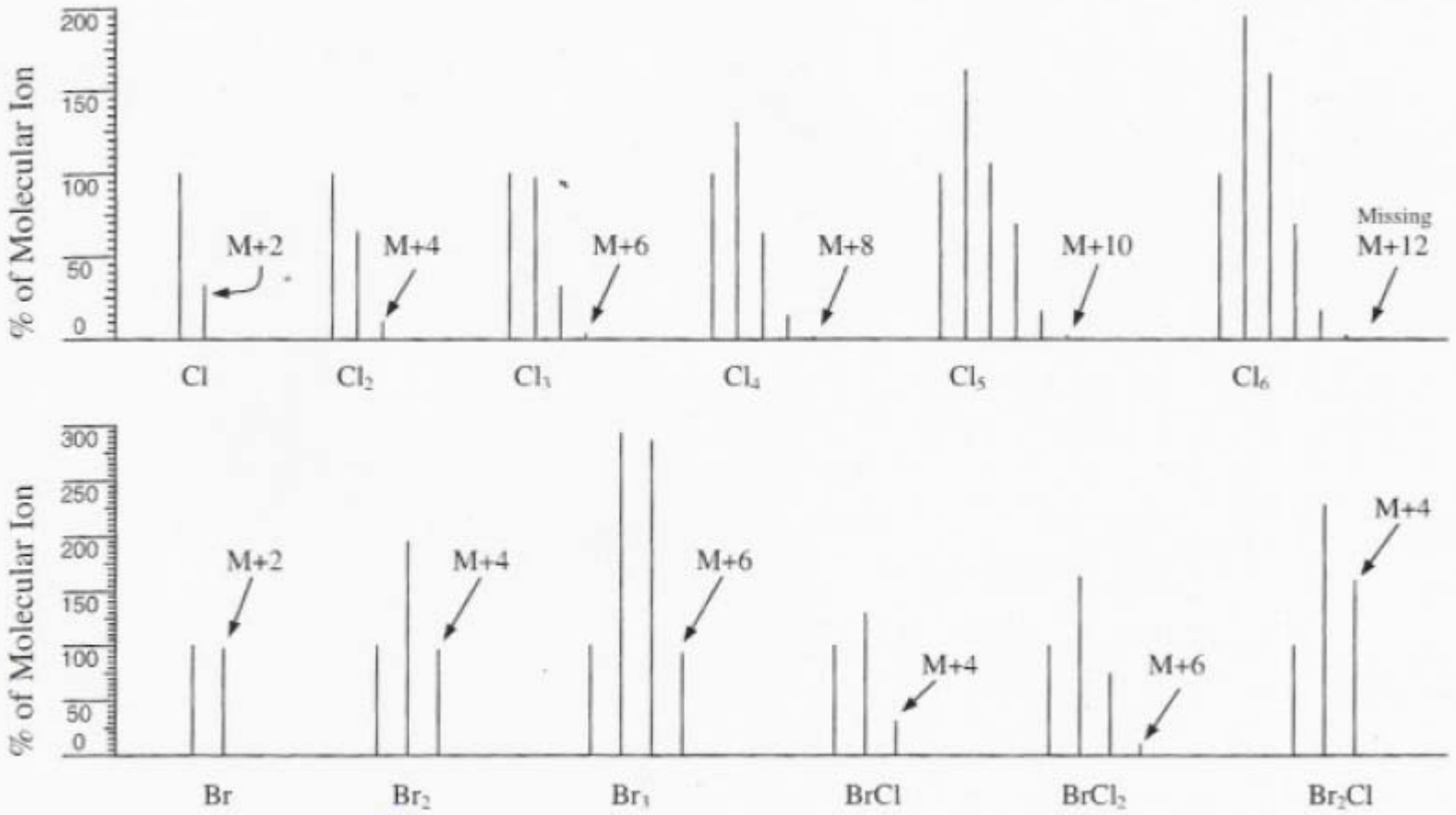
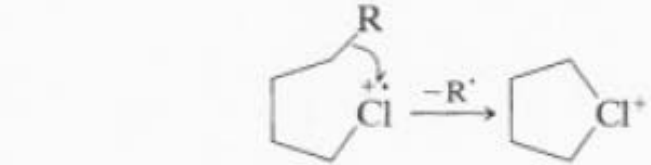


FIGURE 1.29 Predicted patterns of M, M + 2, M + 4, . . . for compounds with various combinations of chlorine and bromine.

Mass Spectra of the Common Classes of Compounds

Aliphatic Chlorides



(Sch 1.34)

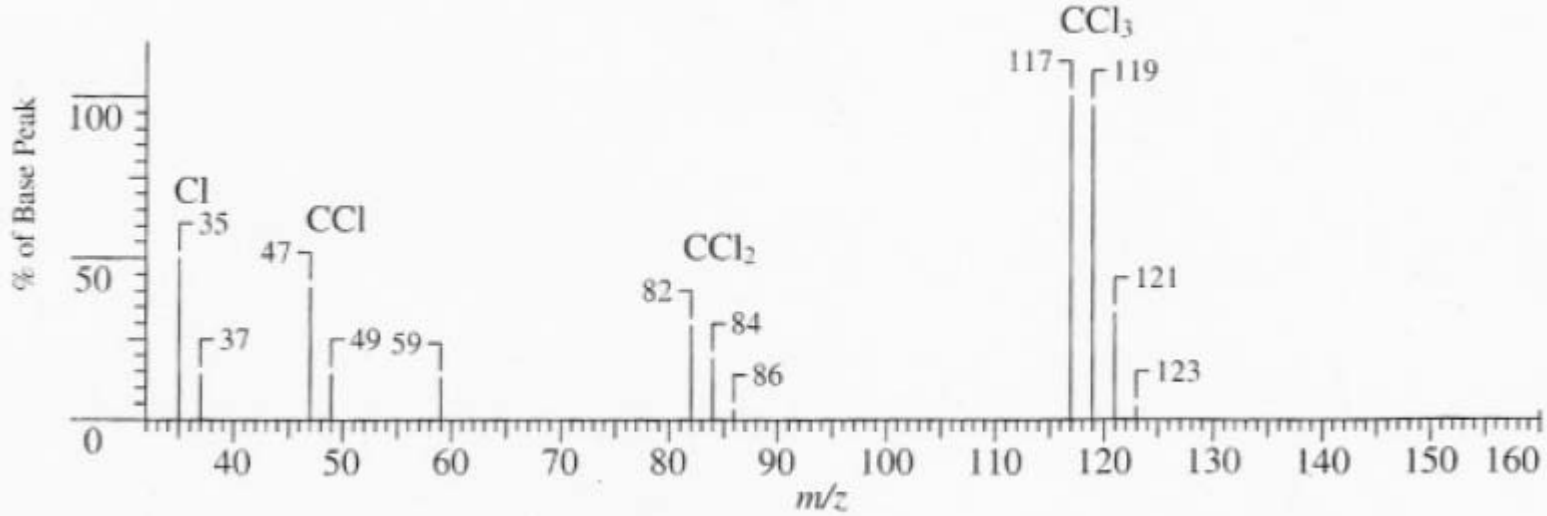
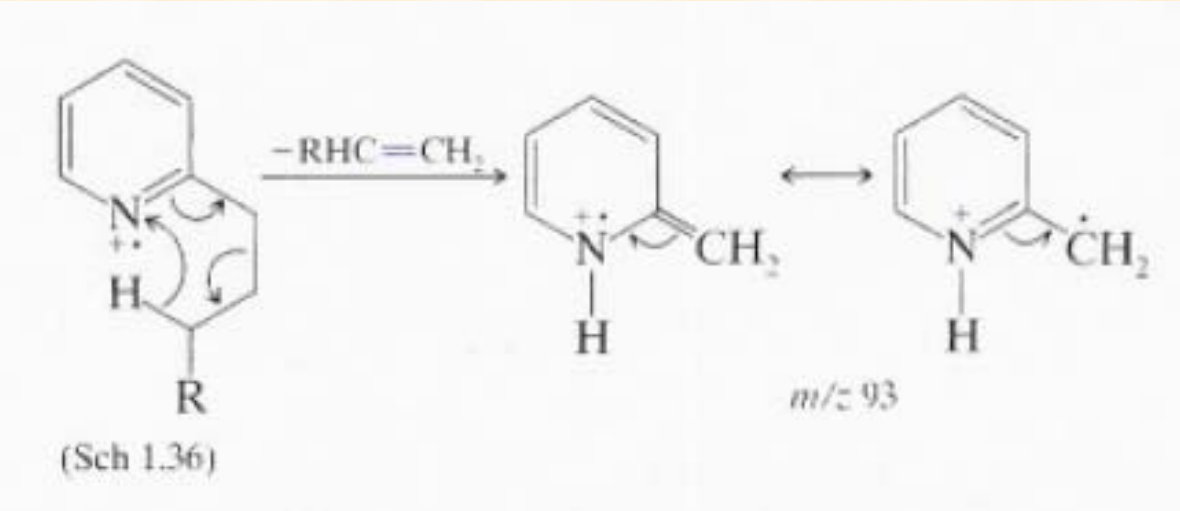


FIGURE 1.30 EI mass spectrum of carbon tetrachloride (CCl_4).

Mass Spectra of the Common Classes of Compounds

Heteroaromatic compounds



(Sch 1.36)

References

Silverstein, Webster, Kielme, Spectrometric Identification of Organic Compounds

Wiley, 2005

Bruice, Organic Chemistry, Prentice Hall, 2005