Chemistry 4631

Instrumental Analysis

Lecture 30

Mass Spectrometry (MS)

From molecular to elemental analysis there are three major techniques used for elemental analysis:

- Optical spectrometry
- <u>Mass spectrometry</u>
- X-ray spectrometry

Mass Spectrometry (MS)

Process of Mass Spectrometry

- Atomization
- Conversion of atoms to ions (usually positive)
- Separation of ions by mass-to-charge ratio (m/z)
- Measurement of ion current by transducer

Mass Spectrometry (MS)

Atomic and molecular weights are measured in atomic mass units (amu)

The atomic weights are determined relative to carbon 12.

Carbon 12 is set to exactly 12 amu.

1 mol of carbon 12 weighs 12.0000 g.

Mass Spectrometry (MS)

For MS must know the exact mass, m.

$^{12}C^{1}H_4$

m = (12.000 x 1) + (1.007825 x 4) = 16.031 amu

$^{13}C^{1}H_4$

m = (13.00335 x 1) + (1.007825 x 4) = 17.035 amu

Mass Spectrometry (MS)

Mass – to - Charge ratio

Divide the atomic or molecular mass of an ion, m, by the charge, z, on the ion.

$^{12}C^{1}H_4^{+}$ m/z = 16.031/1 = 16.031

$^{13}C^{1}H_4^{2+}$ m/z = 17.035/2 = 8.518

Majority of ions in MS have a single charge.

Mass Spectrometry (MS)

Instrumentation

The MS separates rapidly moving ions on the basis of their mass-to-charge ratio, m/z.

The most popular are:

- Quadrapole
- Time-of-flight
- Double-focusing
- Ion Trap

Mass Spectrometry (MS)

Instrumentation

Principle components:

- Inlet
- Ion source
- Mass analyzer
- Ion transducer
- Pumps
- Signal processor

Mass Spectrometry (MS)

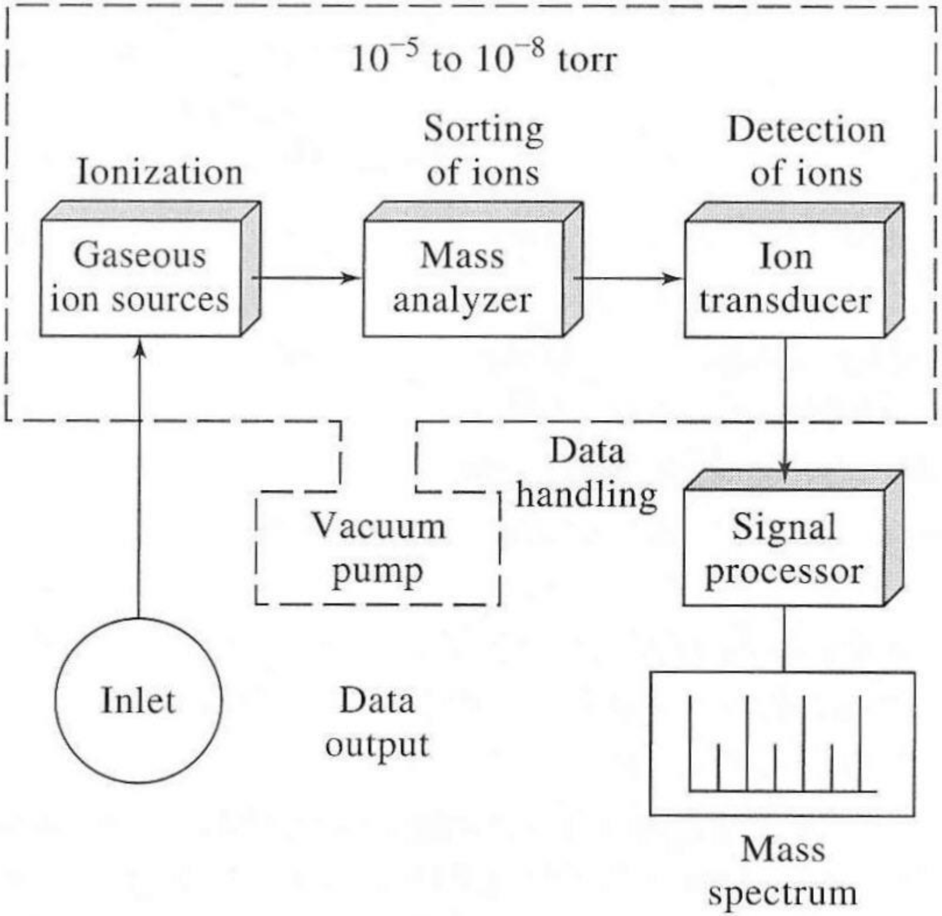


Figure 11-1 Components of a mass spectrometer.

Mass Spectrometry (MS)

Instrumentation

Principle components:

- Inlet
- Ion source
- Mass analyzer
- Ion transducer
- Pumps
- Signal processor

Mass Spectrometry (MS)

Instrumentation

Inlet Systems

Allow introduction of representative sample into ion source with minimal losses of vacuum.

Sample is volatilized externally and allowed to leak into evacuated ionization region.

For gases and liquids with boiling points up to about 500°C.

Mass Spectrometry (MS)

Inlet Systems

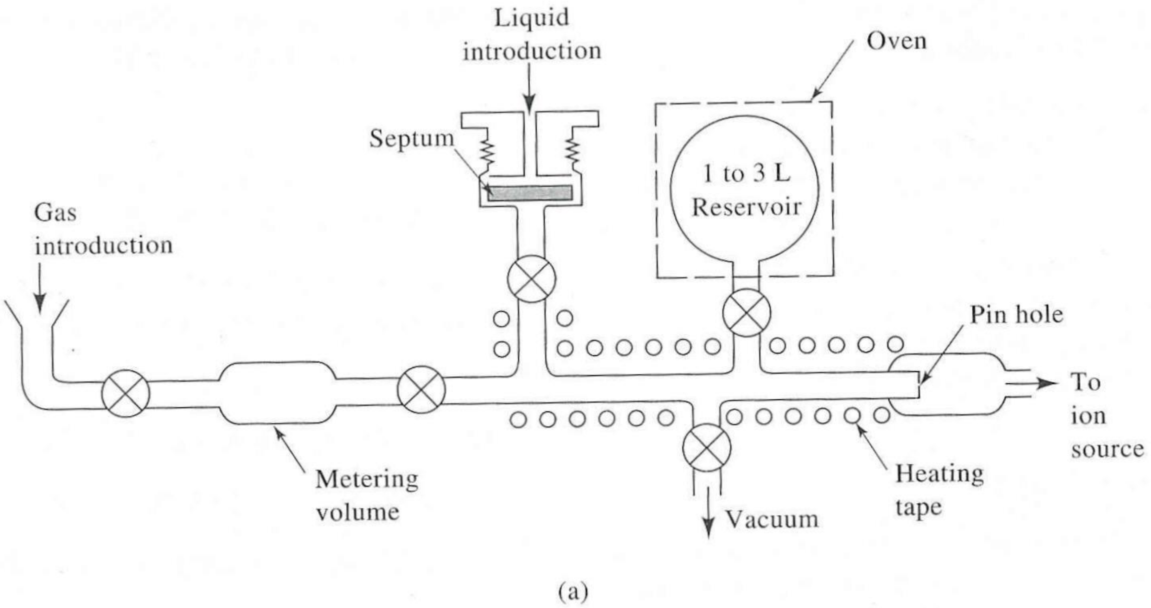


Figure 20-11 Schematic of (a) an external sample introduction system—note that the various parts are not to scale—and (b) a sample probe for inserting a sample directly into the ion source. *(From G. A. Eadon, in* Treatise on Analytical Chemistry, *2nd ed., J. D. Winefordner, M. M. Bursey, and I. M. Kolthoff, Eds., Part I. Vol. 11, p. 9. New York: Wiley, 1989. Reprinted by permission of John Wiley & Sons, Inc.)*

Mass Spectrometry (MS)

Instrumentation

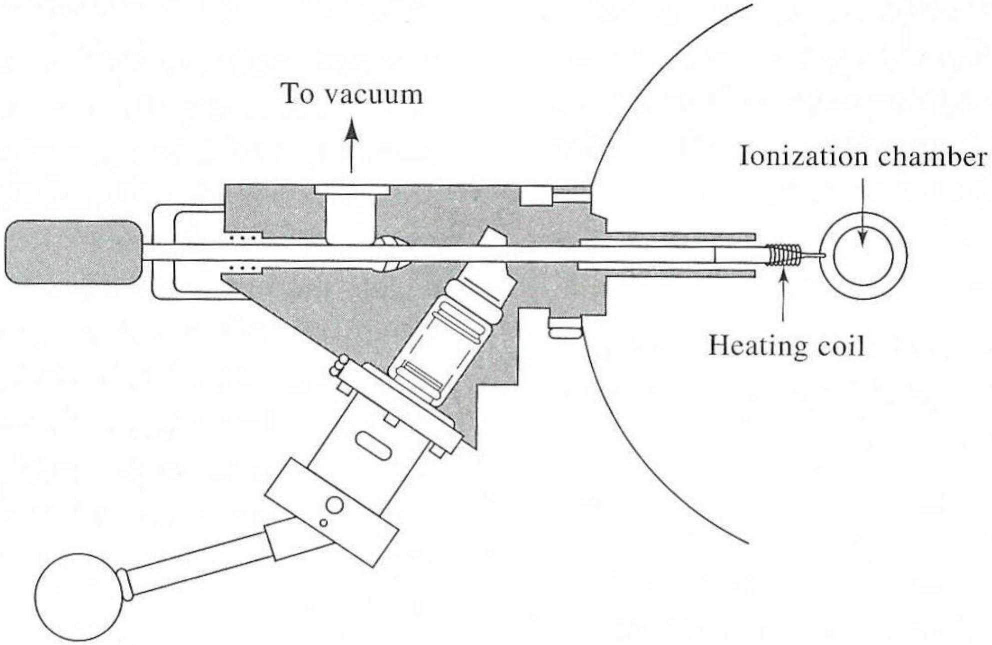
Inlet Systems

Direct probe Inlet

Sample or nonvolatile liquid introduced on a probe into ionization chamber.

Probe can be heated or cooled.

Mass Spectrometry (MS)



(b)

Figure 20-11 Schematic of (a) an external sample introduction system—note that the various parts are not to scale—and (b) a sample probe for inserting a sample directly into the ion source. *(From G. A. Eadon, in* Treatise on Analytical Chemistry, *2nd ed., J. D. Winefordner, M. M. Bursey, and I. M. Kolthoff, Eds., Part I. Vol. 11, p. 9. New York: Wiley, 1989. Reprinted by permission of John Wiley & Sons, Inc.)*

Mass Spectrometry (MS)

Instrumentation

Principle components:

- Inlet
- **Ion source**
- Mass analyzer
- Ion transducer
- Pumps
- Signal processor

Mass Spectroscopy (MS)

Instrumentation

Ion source

- ionizes and fragments molecules
- form the ions into a focused beam
- introduce the beam into the mass analyzer

Mass Spectrometry (MS)

Instrumentation

Ion Sources

Two major categories:

- Gas phase sources - sample is vaporized and then ionized.

  Restricted to thermally stable compounds with boiling points less than 500°C.

  MW limited to 10^3 daltons.

- Desorption sources - sample converted to gaseous ions from solid or liquid state residing on a surface.

  For MWs up to 10^5 daltons.

Mass Spectrometry (MS)

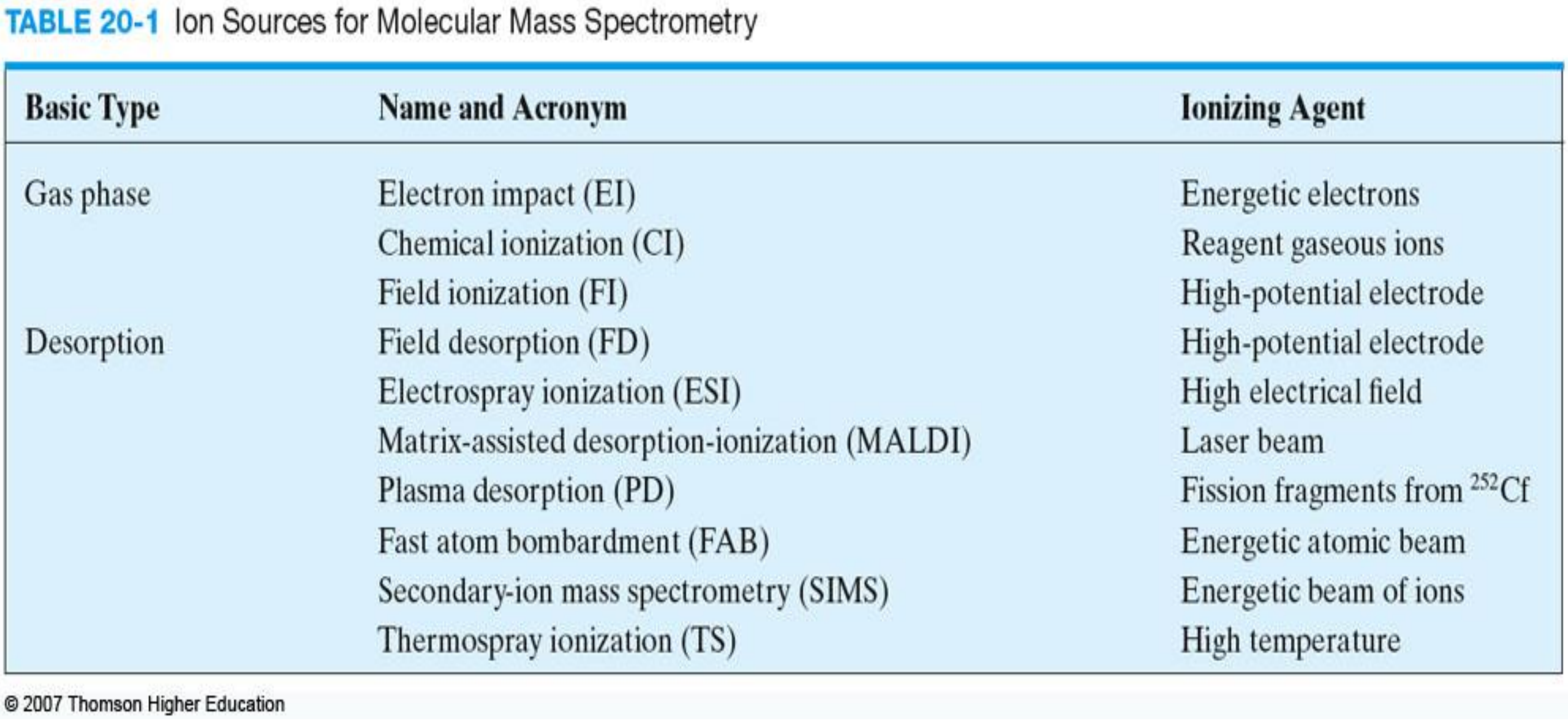


TABLE 20-1 Ion Sources for Molecular Mass Spectrometry

Basic Type	Name and Acronym	Ionizing Agent
Gas phase	Electron impact (EI)	Energetic electrons
	Chemical ionization (CI)	Reagent gaseous ions
	Field ionization (FI)	High-potential electrode
Desorption	Field desorption (FD)	High-potential electrode
	Electrospray ionization (ESI)	High electrical field
	Matrix-assisted desorption-ionization (MALDI)	Laser beam
	Plasma desorption (PD)	Fission fragments from ^{252}Cf
	Fast atom bombardment (FAB)	Energetic atomic beam
	Secondary-ion mass spectrometry (SIMS)	Energetic beam of ions
	Thermospray ionization (TS)	High temperature

Mass Spectrometry (MS)

Instrumentation

Ion Sources

Ion sources are classified as:

- **Hard sources**

  Impart sufficient energy to analyte molecules to promote to a highly excited energetic state.

  Relaxation involves rupture of bonds, producing fragment ions with m/z ratios less than the molecular ion.

- **Soft sources**

  Causes little fragmentation, producing few ions.

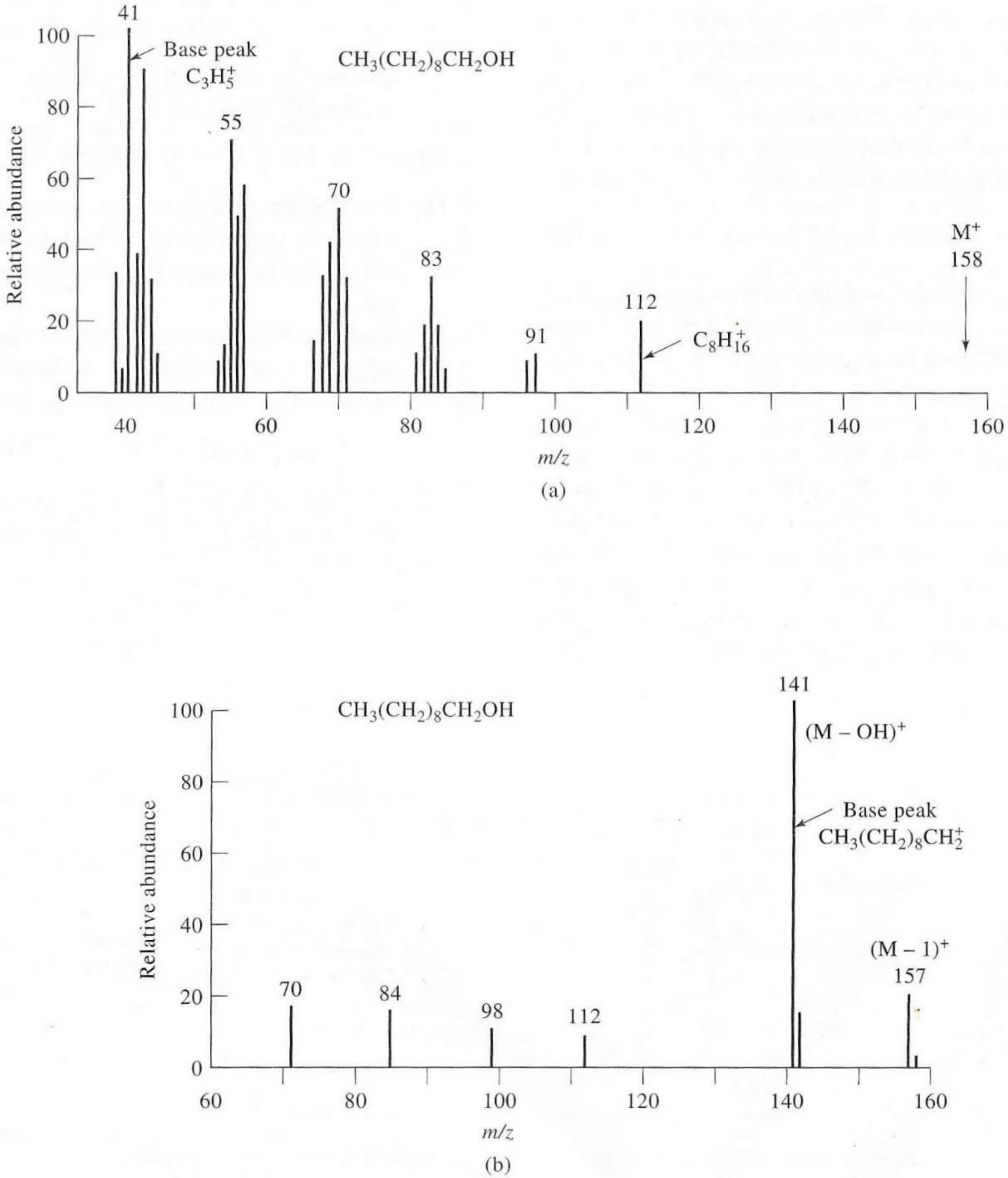


Figure 20-2 Mass spectrum of 1-decanol from (a) a hard source and (b) a soft source.

Mass Spectrometry (MS)

Instrumentation

Ion Sources

Electron-impact source (gas phase source)

Ions produced by electron impact.

Sample brought to a temperature high enough to produce a molecular vapor, which is then ionized by bombardment.

Electrons are emitted from a heated W or Rh filament and accelerated by ~ 70 V towards the anode.

Molecules are passed through this stream of electrons and primarily form singly charge positive ions.

Ions produced are accelerated through electrostatic plates which then goes to the mass analyzer.

Mass Spectrometry (MS)

Ion Sources

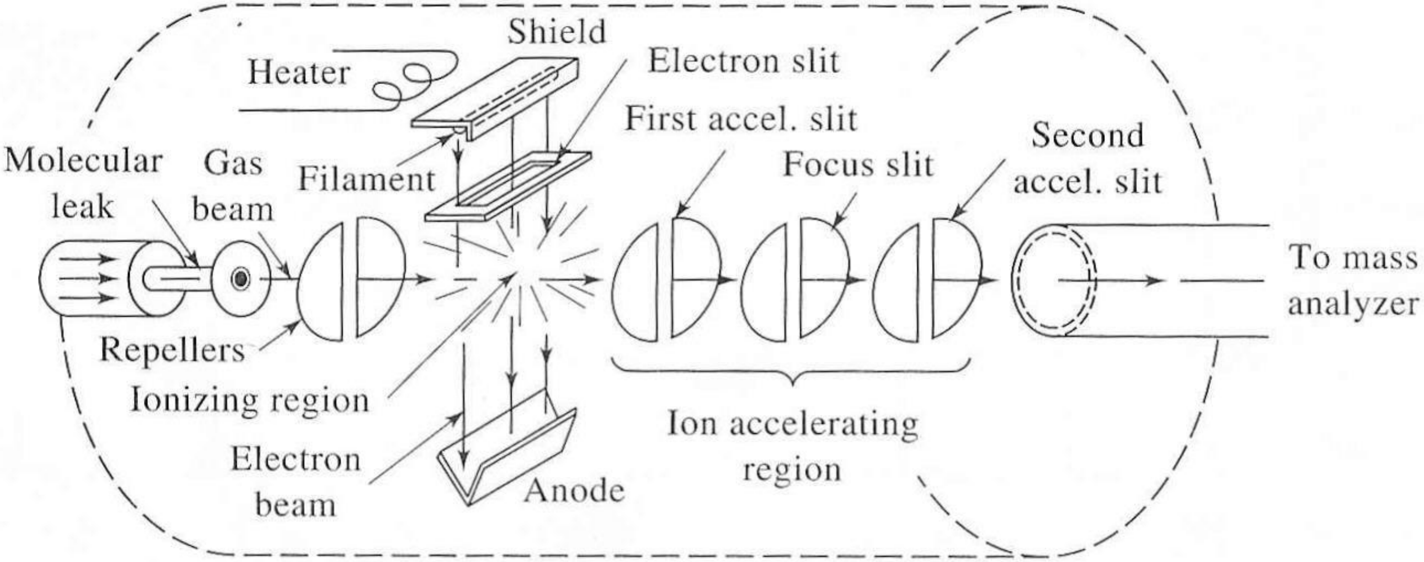


Figure 20-3 An electron-impact ion source. *(From R. M. Silverstein, G. C. Bassler, and T. C. Morrill,* Spectrometric Identification of Organic Compounds, *5th ed., p. 4. New York: Wiley, 1991. Reprinted by permission of John Wiley & Sons, Inc.)*

Mass Spectrometry (MS)

Instrumentation

Ion Sources

Electron-impact source (gas phase source)

Electron-impact spectra

Produce a complicated fragmentation spectra, that can be useful for compound identification.

Collision product peaks:

Sometimes peaks are produced at higher mass numbers than that of the molecular ion.

Usually occurs where collision transfers a H atom to the ion to give a $(M + 1)^+$ peak.

Mass Spectrometry (MS)

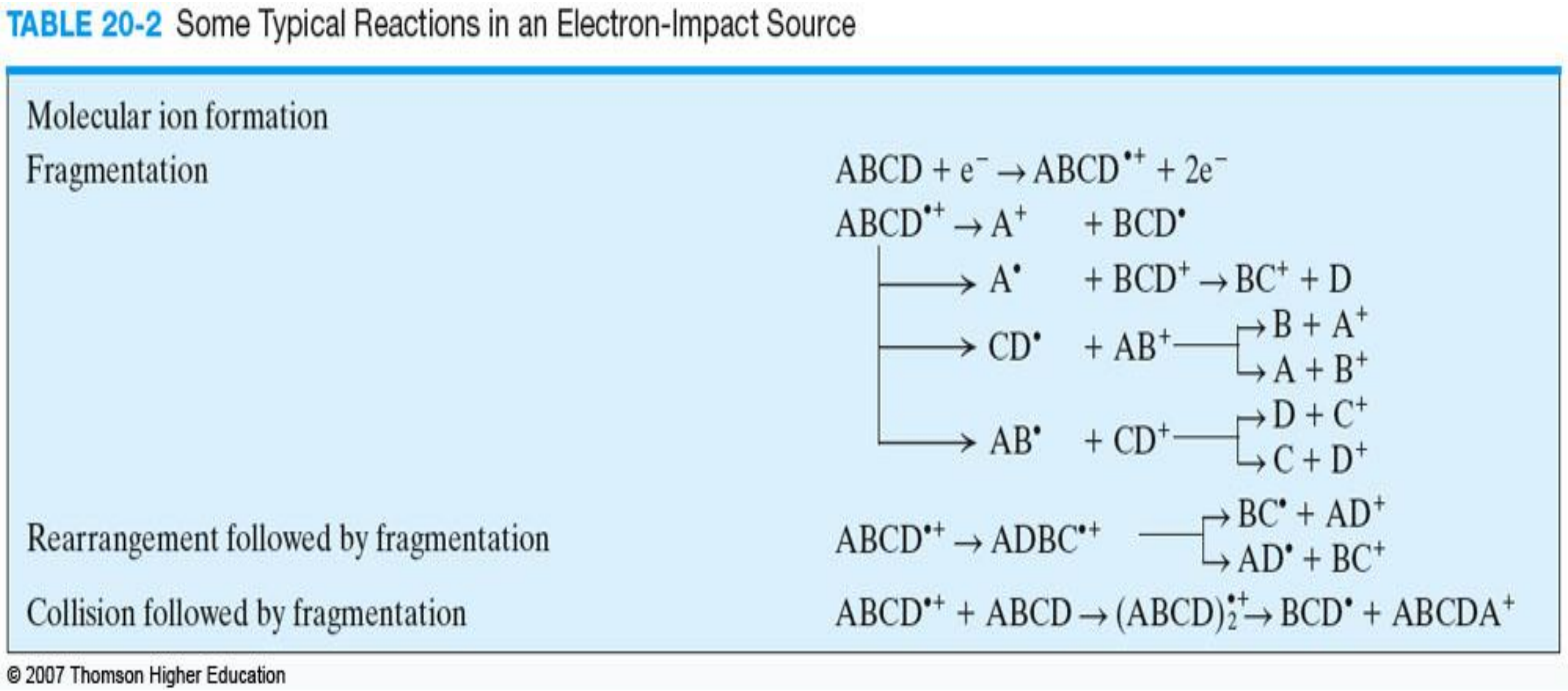


TABLE 20-2 Some Typical Reactions in an Electron-Impact Source

Molecular ion formation	
Fragmentation	$ABCD + e^- \rightarrow ABCD^{\bullet+} + 2e^-$ $ABCD^{\bullet+} \rightarrow A^+ + BCD^\bullet$ $\rightarrow A^\bullet + BCD^+ \rightarrow BC^+ + D$ $\rightarrow CD^\bullet + AB^+ \rightarrow B + A^+$ $\qquad\qquad\qquad\quad \rightarrow A + B^+$ $\rightarrow AB^\bullet + CD^+ \rightarrow D + C^+$ $\qquad\qquad\qquad\quad \rightarrow C + D^+$
Rearrangement followed by fragmentation	$ABCD^{\bullet+} \rightarrow ADBC^{\bullet+} \rightarrow BC^\bullet + AD^+$ $\qquad\qquad\qquad\qquad\quad \rightarrow AD^\bullet + BC^+$
Collision followed by fragmentation	$ABCD^{\bullet+} + ABCD \rightarrow (ABCD)_2^{\bullet+} \rightarrow BCD^\bullet + ABCDA^+$

Mass Spectrometry (MS)

Instrumentation

Ion Sources

Electron-impact source (gas phase source)

Advantages:

- Convenient
- Sensitive - Produce high ion currents
- Fragmentation

Disadvantages:

- Fragmentation
- Only for volatile samples

Mass Spectrometry (MS)

Instrumentation

Ion Sources

Chemical Ionization (gas phase source)

Chemical ionization (CI) takes place in an ion source similar to the EI source.

The main difference is a moderate pressure (~1 torr) of reagent gas is maintained in the source.

Gaseous atoms of sample are ionized by collision with ions (usually positive) produced by electron bombardment of an excess of a reagent gas.

Mass Spectrometry (MS)

Instrumentation

Ion Sources

Chemical Ionization (gas phase source)

Most common reagent is methane which gives CH_4^+, CH_3^+, and CH_2^+ under electron bombardment.

$$CH_4^+ + CH_4 \rightarrow CH_5^+ + CH_3$$

$$CH_3^+ + CH_4 \rightarrow C_2H_5^+ + H_2$$

Mass Spectrometry (MS)

Instrumentation

Ion Sources

Chemical Ionization (gas phase source)

The produced ions interact with the analyte molecules by four basic mechanisms:

$CH_4^+ + MH \rightarrow MH^+ + CH_4$ (Charge transfer)

$CH_5^+ + MH \rightarrow MH_2^+ + CH_4$ (Proton transfer)

$C_2H_5^+ + MH \rightarrow M^+(M-1) + C_2H_6$ (Hydride abstraction)

$C_2H_5^+ + MH \rightarrow C_2H_5{:}MH^+(M+29)$ (Addition)

Mass Spectrometry (MS)

Instrumentation

Ion Sources

Chemical Ionization (gas phase source)

CI is a soft ionization process and compared to EI produces fewer fragment ions.

Besides methane, isobutene and ammonia are common reagent CI gases.

Mass Spectrometry (MS)

Instrumentation

Ion Sources

Field-Ionization Sources (Gas phase source)

Ions are formed under large electric fields (10^8 V/cm).

The fields are produced by applying high voltages (10-20 kV) to emitters made up of fine tips with diameters less than 1 μm.

The emitter is often a W wire that is coated carbon dendrites.

Mass Spectrometry (MS)

Ion Sources

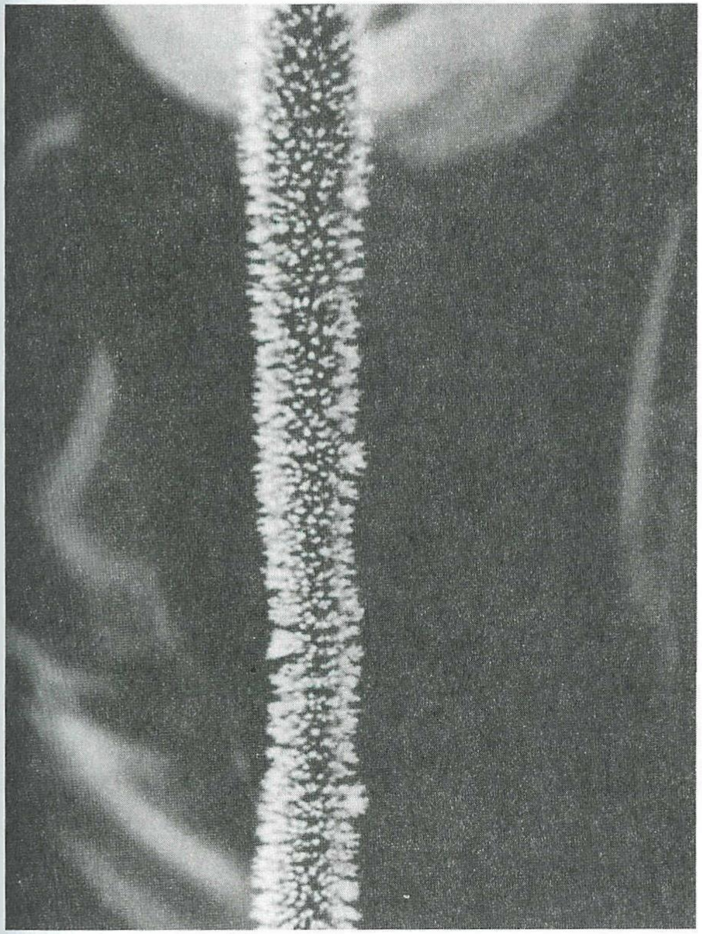


Figure 20-5 Photomicrograph of a carbon microneedle emitter. *(Courtesy of R. P. Lattimer, BF Goodrich Research and Development Center.)*

Mass Spectrometry (MS)

Instrumentation

Ion Sources

Field-Ionization Sources (Gas phase source)

The emitters are mounted 0.5-2 mm from the cathode, the sample diffuses above the microtips and are ionized by quantum mechanical tunneling mechanism.

The electrons of the analyte are extracted by the microtips.

Ionization occurs but very little fragmentation.

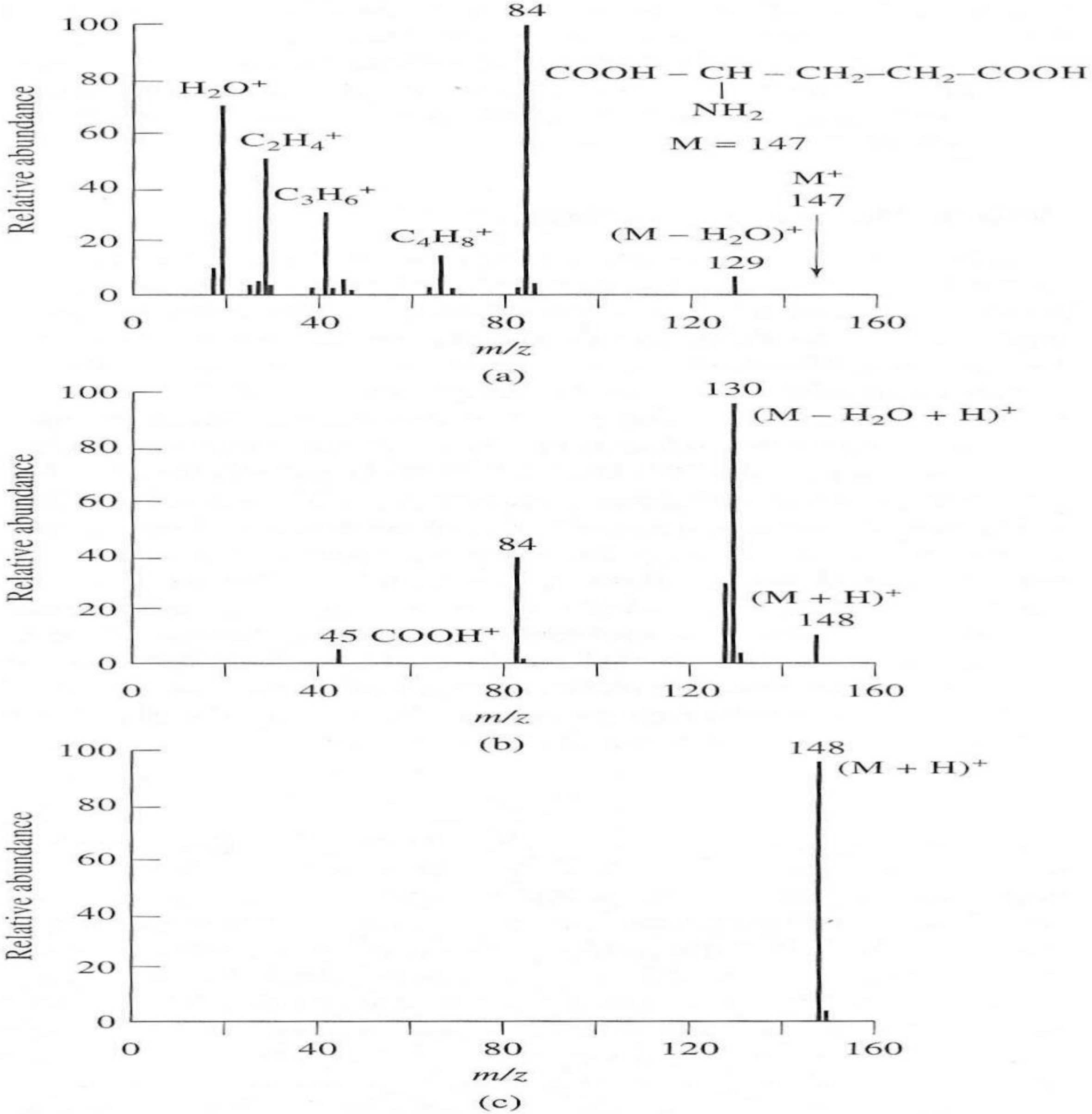


Figure 20-6 Mass spectra for glutamic acid: (a) electron-impact ionization, (b) field ionization, and (c) field desorption. *(From H. D. Beckey, A. Heindrich, and H. U. Winkler,* Int. J. Mass Spec. Ion Phys., ***1970****, 3, App. 11. With permission.)*

Mass Spectrometry (MS)

Instrumentation

Ion Sources

Matrix-Assisted Laser Desorption/Ionization (desorption source)

MALDI was developed in 1988.

An aqueous/alcohol solution of the sample is mixed with a large excess of a radiation absorbing matrix material.

Solution is evaporated on the surface of a metal inserted into MS and then hit with a pulse laser beam.

This sublimes the sample as ions into a time-of-flight spectrometer.

Mass Spectrometry (MS)

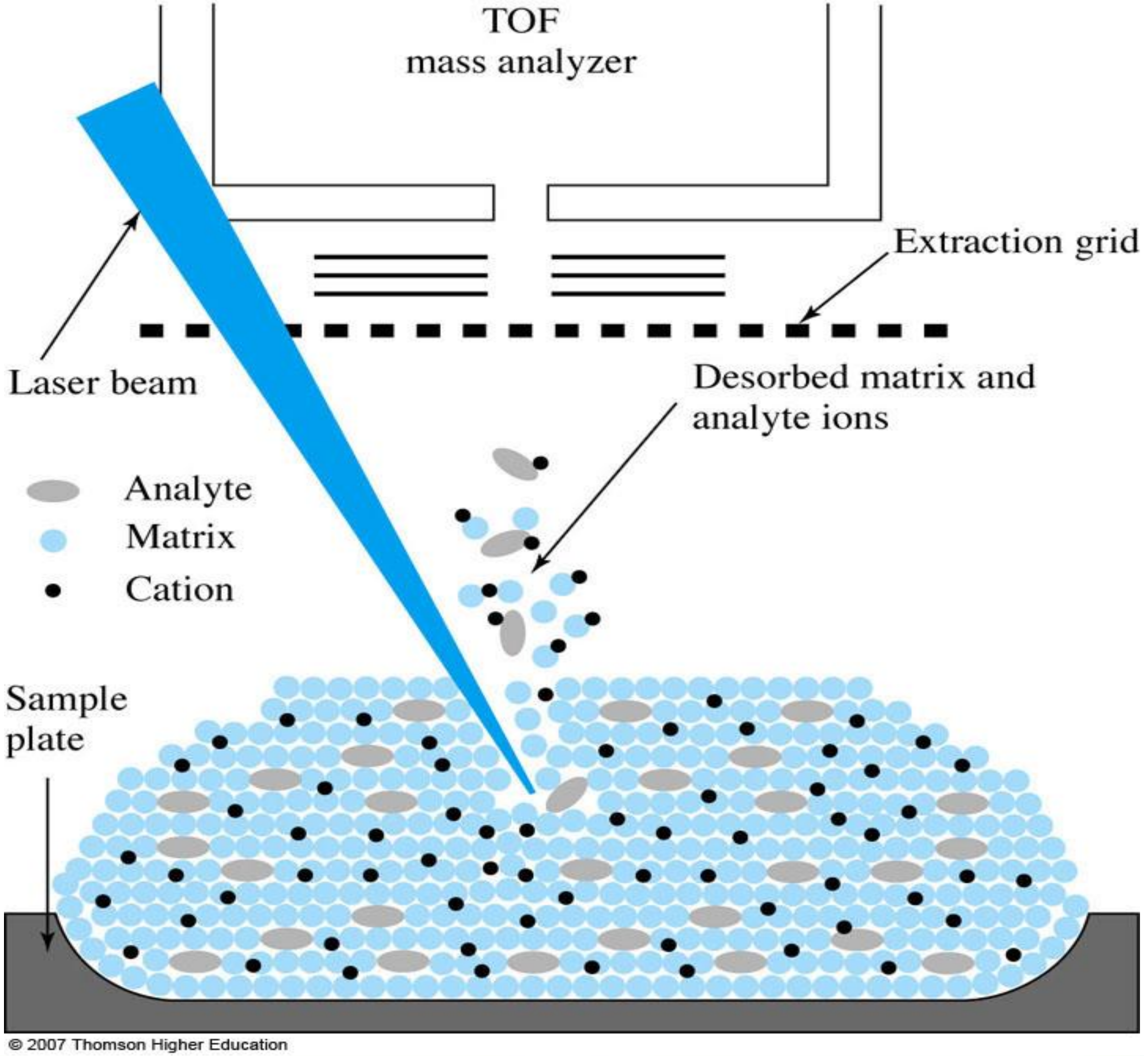
Instrumentation

Ion Sources - MALDI

A laser-based soft ionization method
-Important for protein analysis.
-Ionization occurs with bombarding the sample with laser light
-The sample is embedded in a chemical matrix (UV absorbant)
-The matrix makes the production of intact gas-phase ions from large, nonvolatile, and thermally decomposed compounds such as proteins, oligonucleotides, synthetic polymers easy
-The matrix absorbs the laser light energy thus small amount is vaporized

Mass Spectrometry (MS)

Matrix-Assisted Laser Desorption/Ionization



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Mass Spectrometry (MS)

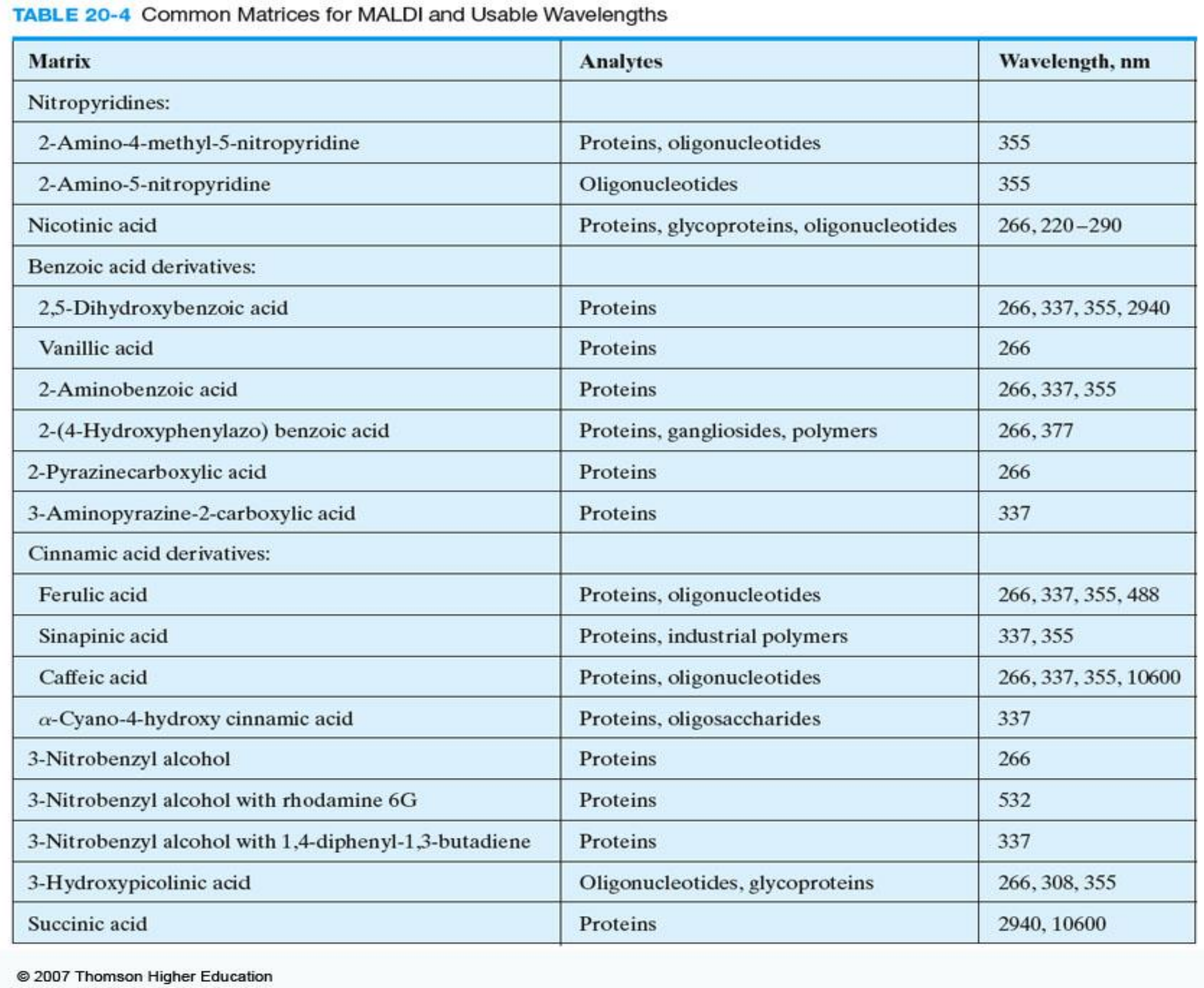


TABLE 20-4 Common Matrices for MALDI and Usable Wavelengths

Matrix	**Analytes**	**Wavelength, nm**
Nitropyridines:		
2-Amino-4-methyl-5-nitropyridine	Proteins, oligonucleotides	355
2-Amino-5-nitropyridine	Oligonucleotides	355
Nicotinic acid	Proteins, glycoproteins, oligonucleotides	266, 220–290
Benzoic acid derivatives:		
2,5-Dihydroxybenzoic acid	Proteins	266, 337, 355, 2940
Vanillic acid	Proteins	266
2-Aminobenzoic acid	Proteins	266, 337, 355
2-(4-Hydroxyphenylazo) benzoic acid	Proteins, gangliosides, polymers	266, 377
2-Pyrazinecarboxylic acid	Proteins	266
3-Aminopyrazine-2-carboxylic acid	Proteins	337
Cinnamic acid derivatives:		
Ferulic acid	Proteins, oligonucleotides	266, 337, 355, 488
Sinapinic acid	Proteins, industrial polymers	337, 355
Caffeic acid	Proteins, oligonucleotides	266, 337, 355, 10600
α-Cyano-4-hydroxy cinnamic acid	Proteins, oligosaccharides	337
3-Nitrobenzyl alcohol	Proteins	266
3-Nitrobenzyl alcohol with rhodamine 6G	Proteins	532
3-Nitrobenzyl alcohol with 1,4-diphenyl-1,3-butadiene	Proteins	337
3-Hydroxypicolinic acid	Oligonucleotides, glycoproteins	266, 308, 355
Succinic acid	Proteins	2940, 10600

Mass Spectrometry (MS)

Instrumentation

Ion Sources

Electrospray Ionization (Desorption source)

ESI/MS developed in 1984, now most important technique for analyzing biomolecules with MW > 100,000 daltons.

Can also be used to characterize inorganic species and synthetic polymers.

Mass Spectrometry (MS)

Instrumentation

Ion Sources

Electrospray Ionization (Desorption source)

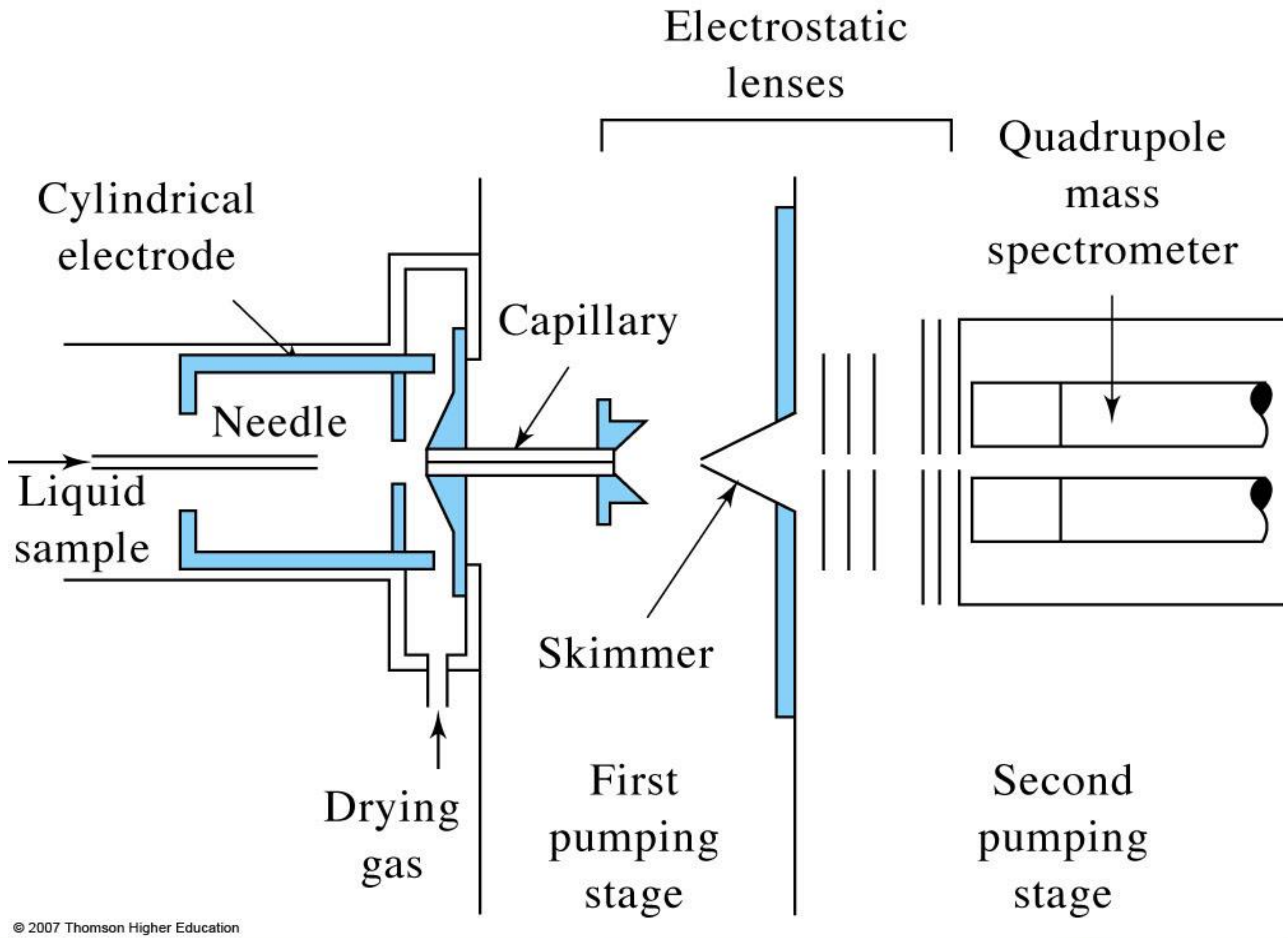
Solution of sample is pumped through a stainless steel capillary needle at few ml/min.

A few kV is applied between the needle and a surrounding cylindrical electrode.

The charged spray from the needle enters a desolvating capillary that evaporates the solvent and desorbs ions into ambient gas.

Mass Spectrometry (MS)

Ion Sources



Mass Spectrometry (MS)

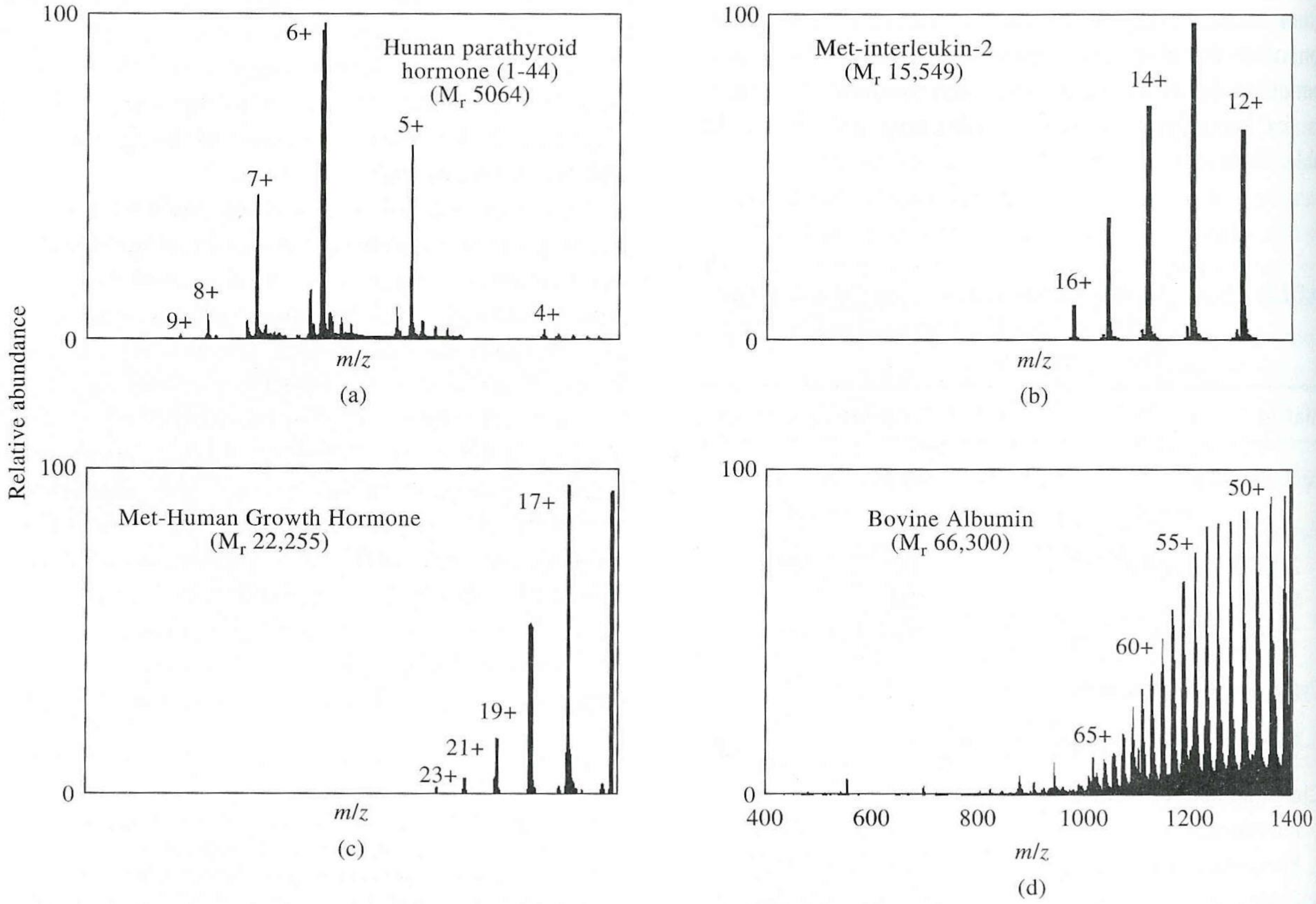


Figure 20-9 Typical electrospray mass spectra of proteins and peptides. The numbers above the peaks represent the molecular charge associated with each peak. *(From R. D. Smith et al.,* Anal. Chem., *1990,* 62, *887.)*

Mass Spectrometry (MS)

Instrumentation

Ion Sources

Fast Atom Bombardment Sources (Desorption source)

Samples condensed in glycerol and ionized by bombardment with energetic Xe or Ar atoms.

Desorption occurs when ions are sputtered from surface.

Mostly used for polar high MW species.

Assignment

- Read Chapter 11
- Read Chapter 20
- HW 17 Chapter 20: 1-5, 7-11, 17
- HW17 Due 4/29/24

- Test 4