SHIMADZU

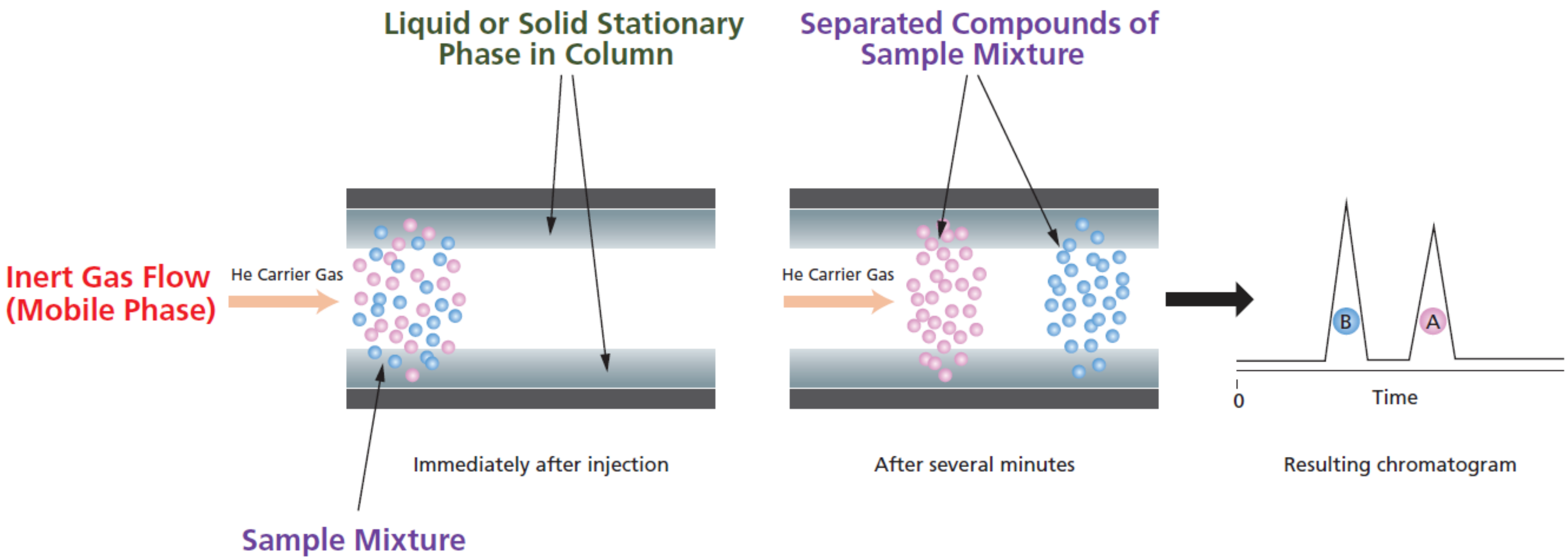
Fundamentals Of

SHIMADZU GCMS-QP2020 series

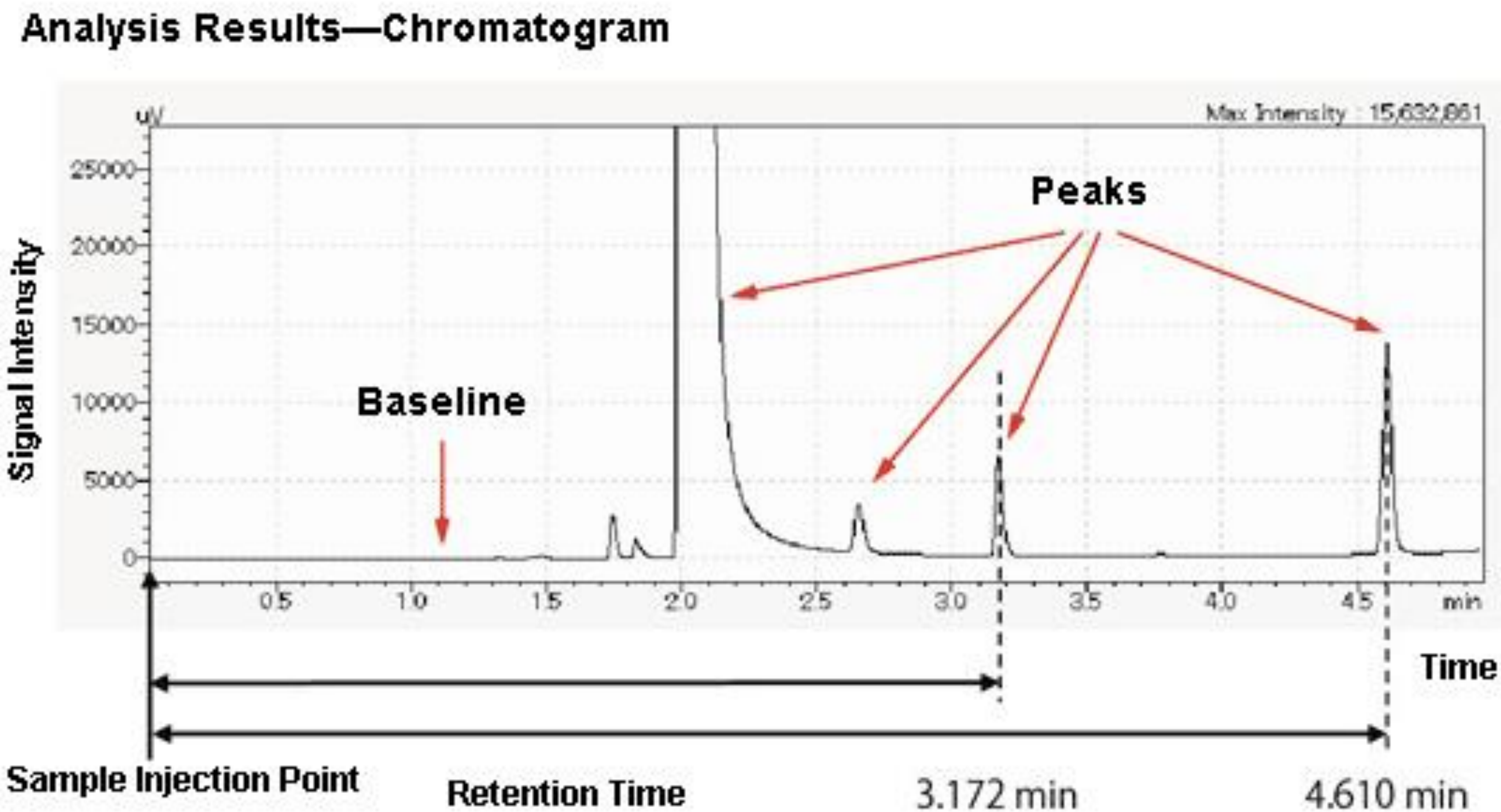
GCMS-QP2020series.

- **GCMS-QP2020 is Shimadzu gas chromatograph-mass spectrometer (GC-MS).**
- **GC-MS is a combination of two different analytical techniques, Gas Chromatography and Mass Spectrometry.**

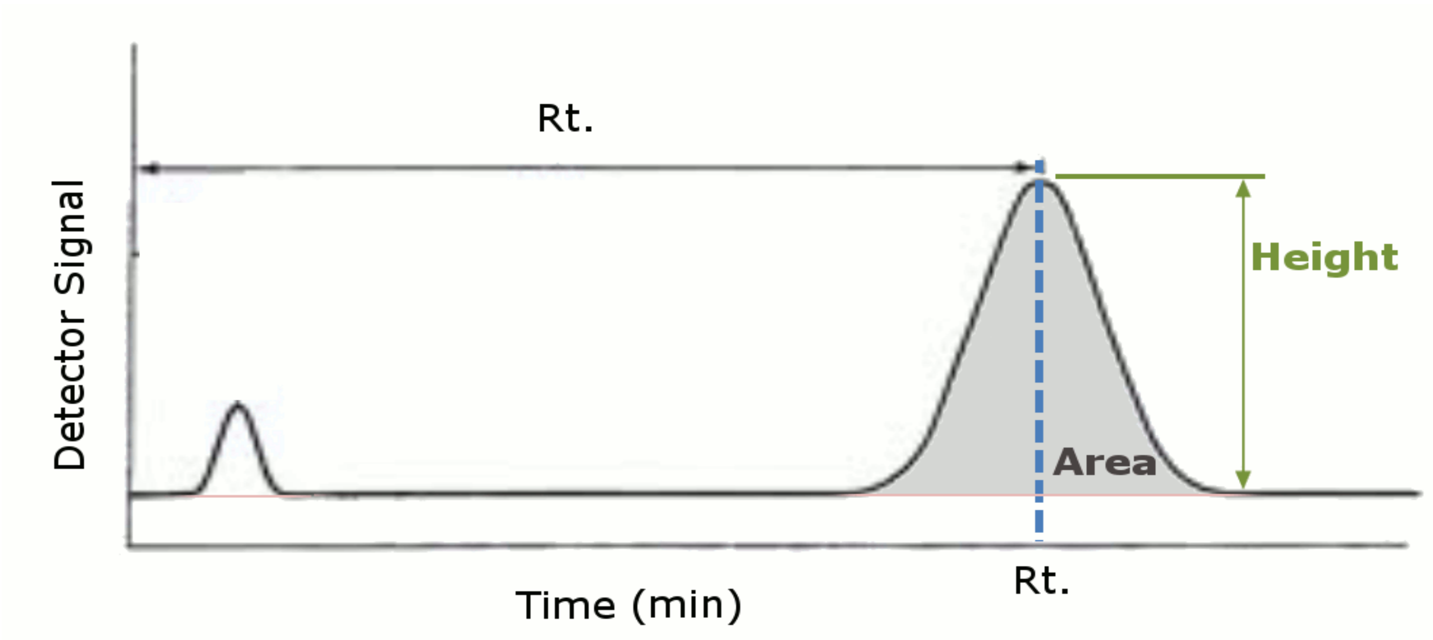
Overview of GC Analysis



How to read Chromatogram.

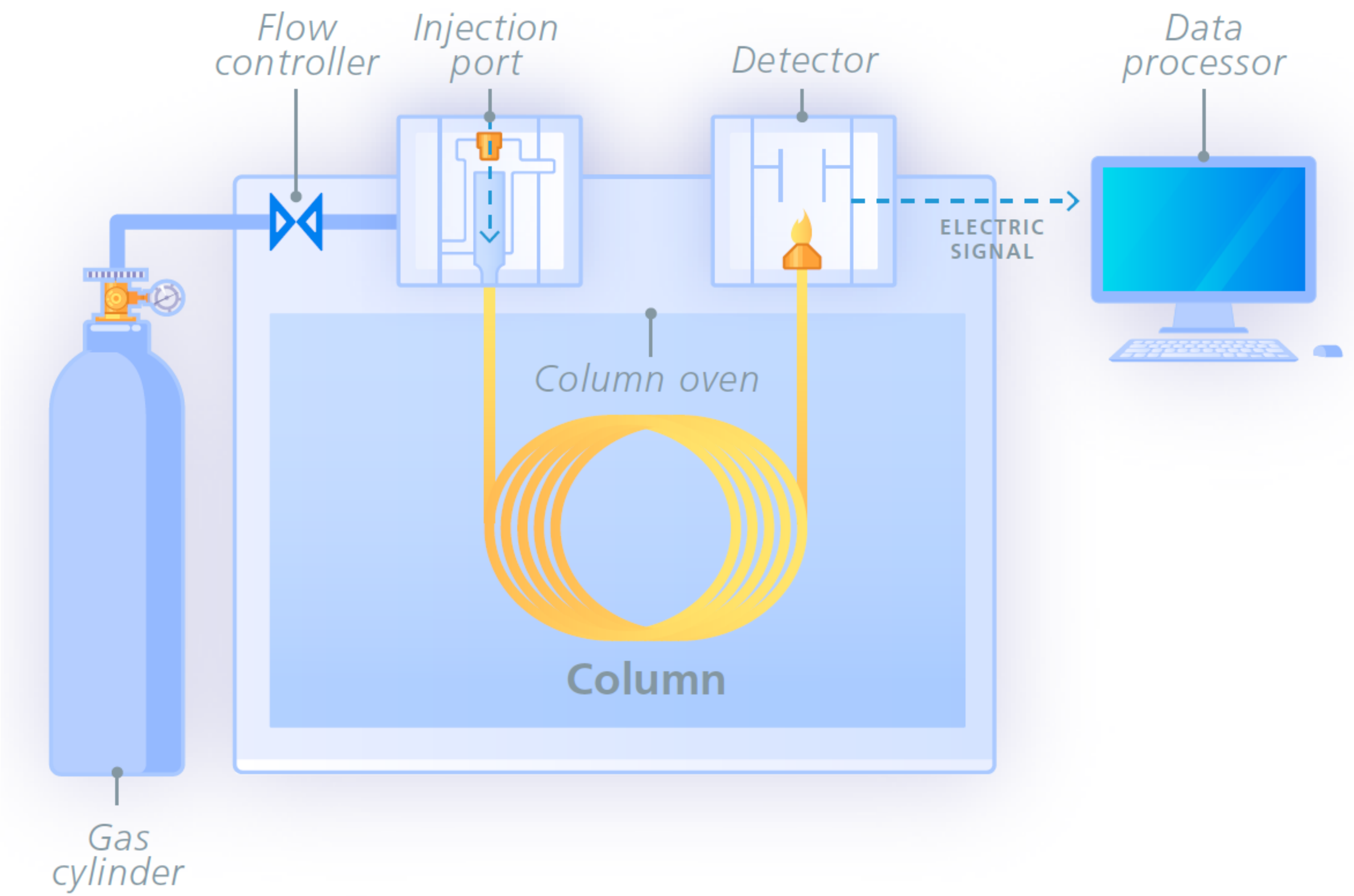


How to read Chromatogram.

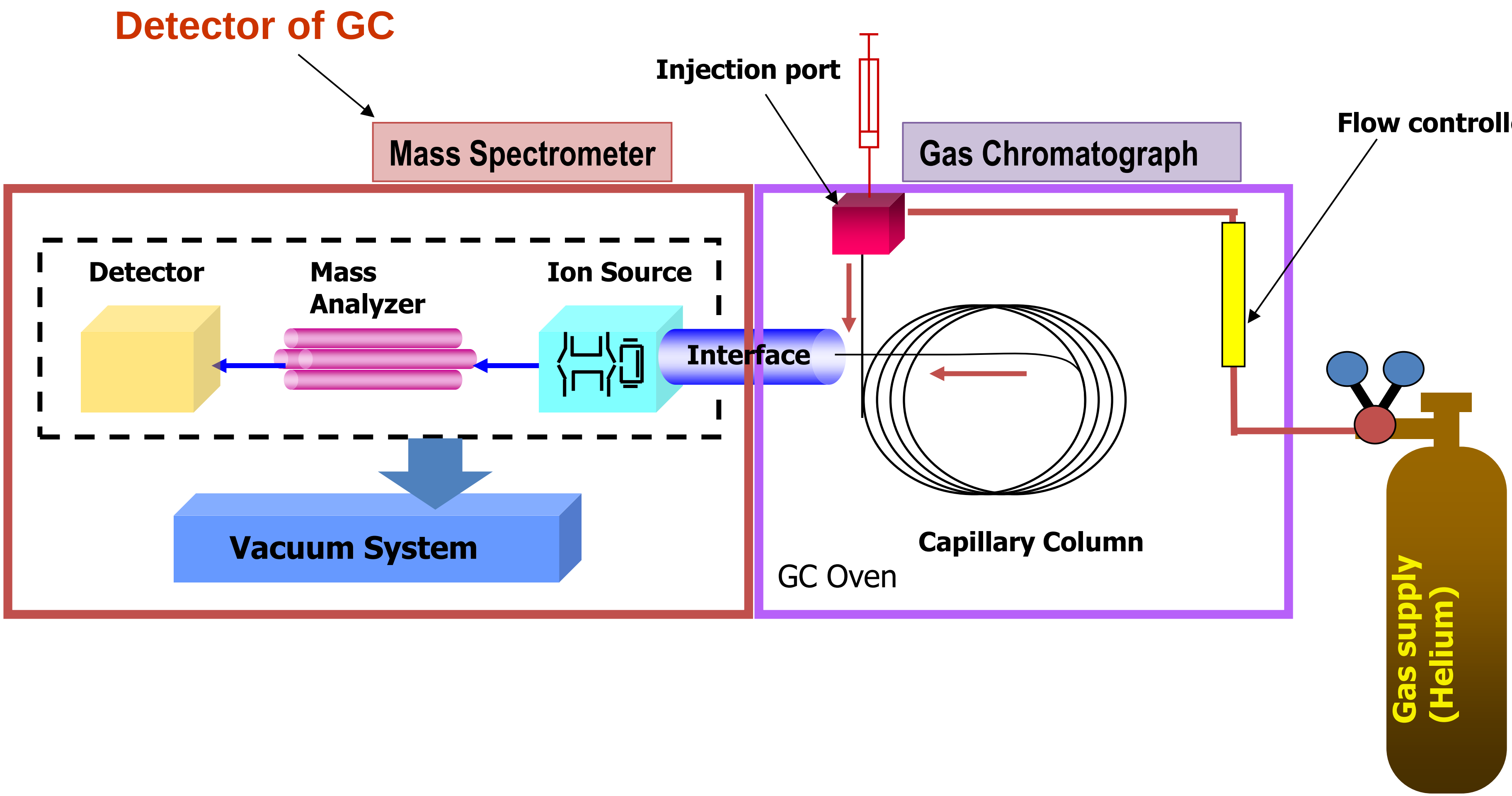


Retention time (Rt.) : Qualitative analysis.
Peak area, peak height : Quantitative analysis.

Overview of GC system configuration



Gas Chromatograph-Mass Spectrometer Components



Carrier gas

Carrier gas for GCMS

Carrier gas

- ชนิดของ carrier gas ที่นิยมใช้กับ GC detectorปกติ ได้แก่ H2, He และ N2 แต่ carrier gas ที่นิยมใช้กับเครื่อง GCMS ได้แก่แก็ส He
- ควรมีความบริสุทธิ์(Purity) สูงไม่น้อยกว่า 99.999%
- ควรติดตั้ง gas purifiers หรือ gas trap ไว้ที่ line gas ก่อนเข้าเครื่อง GC ด้วย

INJECTOR

Injector

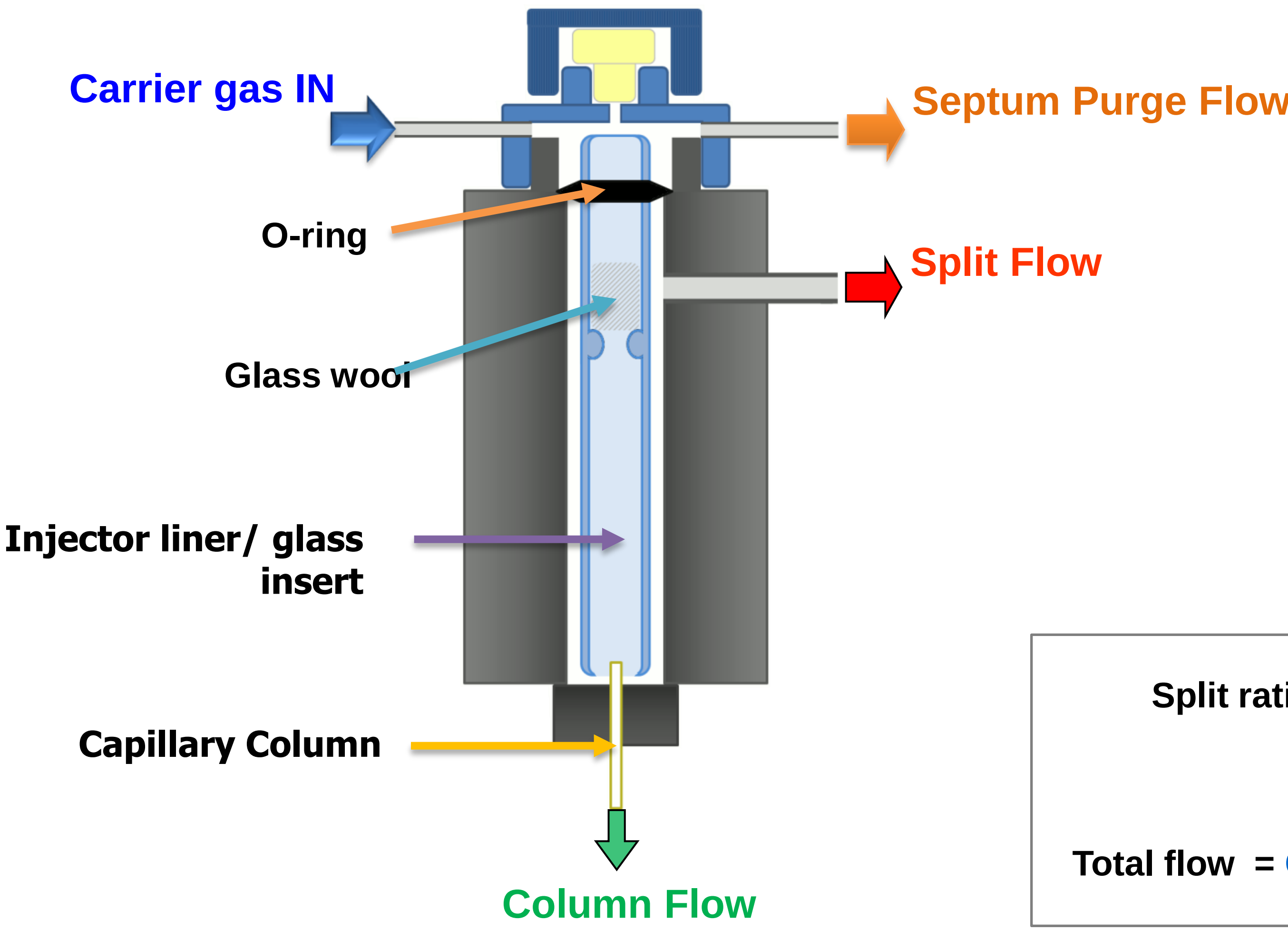
Injector

คือส่วนที่ทำหน้าที่เป็นทางเข้าของสารตัวอย่างและเปลี่ยนสถานะของสารตัวอย่างที่ฉีดเข้าไปให้กลายเป็นสถานะแก๊ส

Split/Splitless Injector

(SPL)

SPL Injector – Split mode



Total Flow :	75.4	mL/min
Column Flow :	1.42	mL/min
Linear Velocity :	37.6	cm/sec
Purge Flow :	3.0	mL/min
Split Ratio :	50.0	

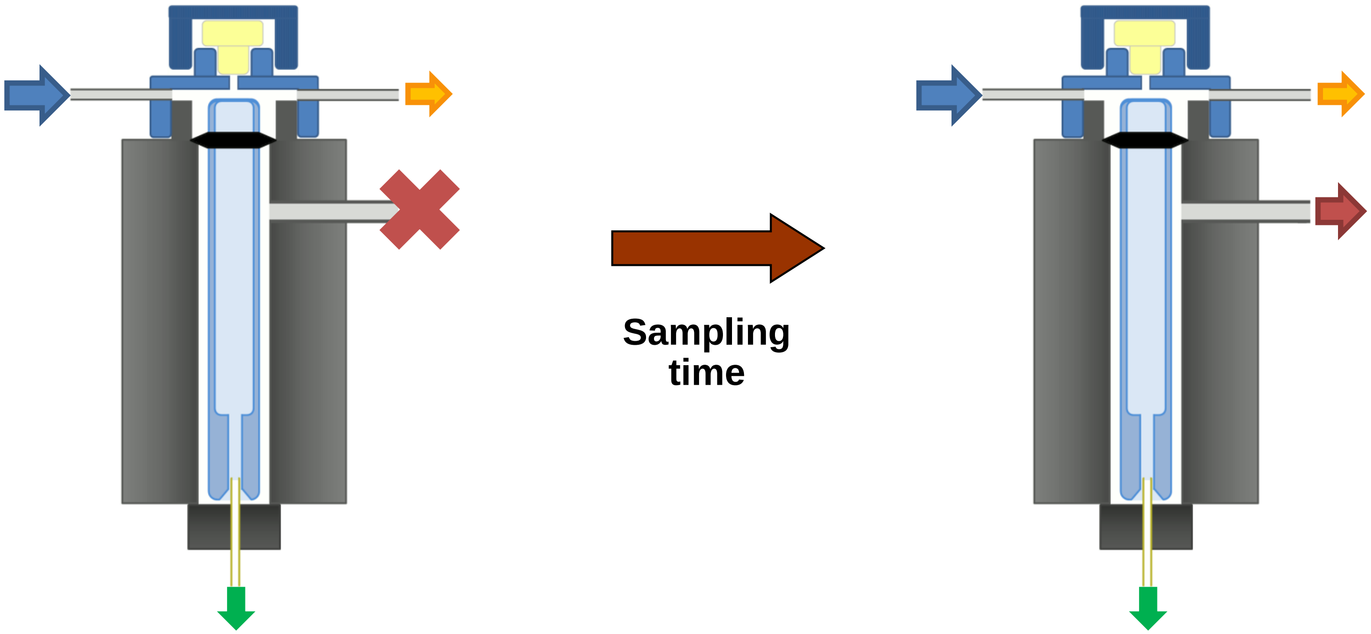
$$\text{Split ratio} = \frac{\text{Split Flow}}{\text{Column Flow}}$$

Total flow = Column flow + Split flow + Purge flow

SPL Injector – splitless mode

2.Splitless mode (ใช้เมื่อสารตัวอย่างมีความเข้มข้นต่ำ)

จะทำงานในโหมดDirectก่อนเป็นเวลาเท่ากับ Sampling time หลังจากนั้นจะเปลี่ยนไปทำงานในโหมด Split จนจบการวิเคราะห์



และเพื่อให้ได้ลักษณะPeakในผลการวิเคราะห์ที่ดี ควรตั้งอุณหภูมิเริ่มต้นของ Column Oven ให้ต่ำกว่า Boil point ของ Solvent อีกอย่างน้อย 10 องศา

SPL Injector

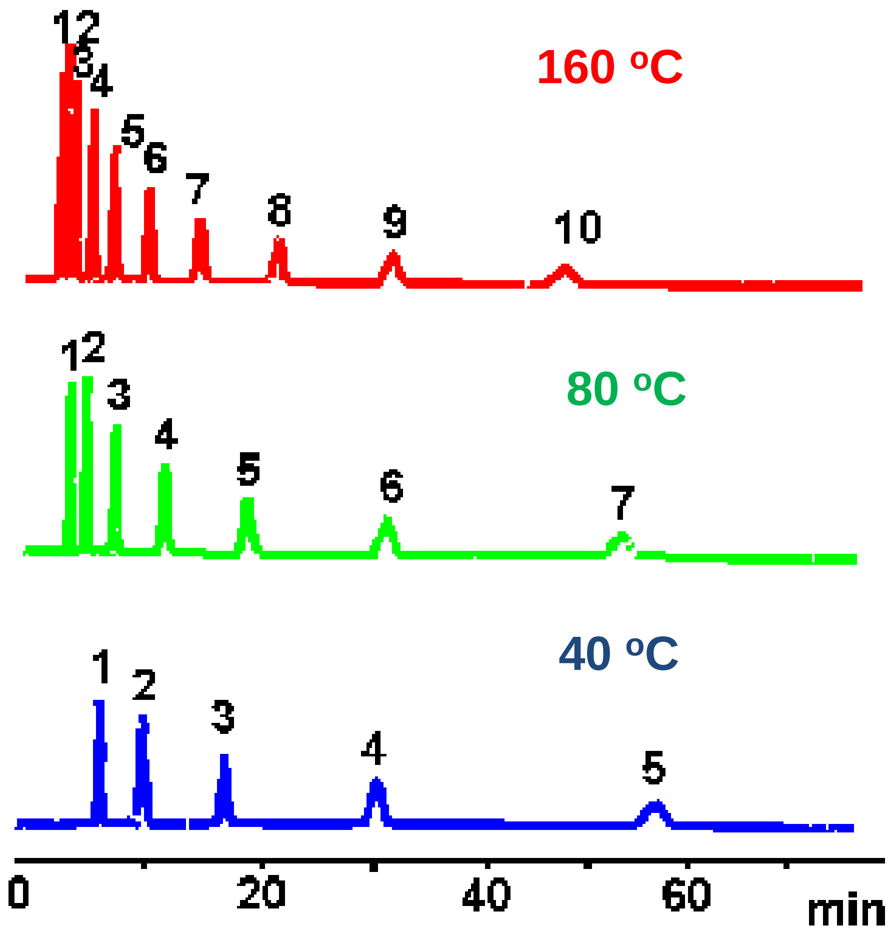
SPL Injection Mode

ดังนั้นจึงควรเลือก mode การทำงานของ SPL Injector ให้เหมาะสมกับความเข้มข้นของ sample ของเรา

- ***Split mode*** **: สารตัวอย่างมีความเข้มข้นปกติ-สูง**
- ***Splitless mode*** **: สารตัวอย่างมีความเข้มข้นต่ำ (Trace analysis)**

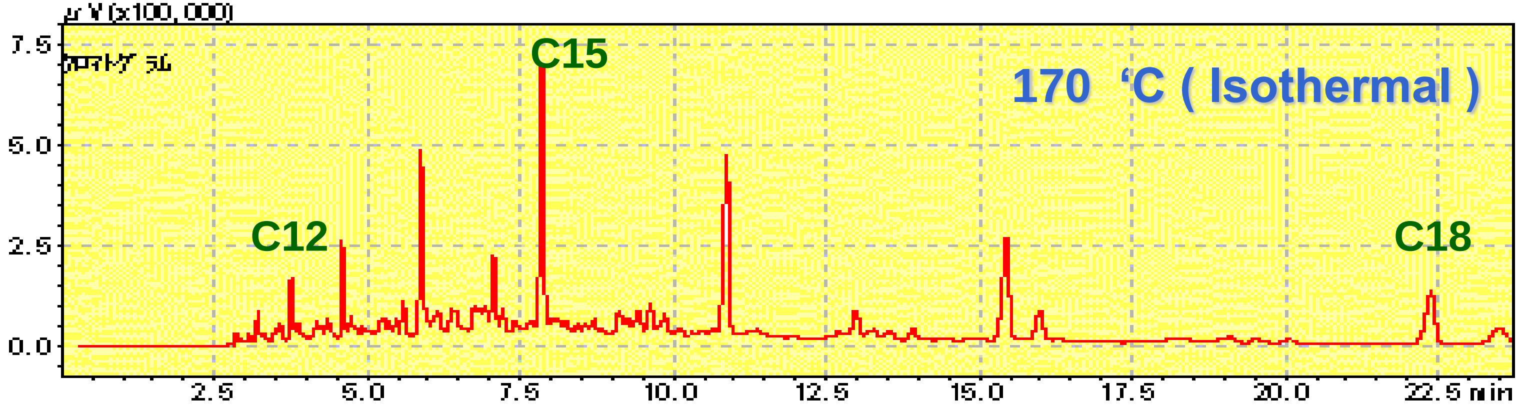
GC Column & Column Oven

Column & Column Oven

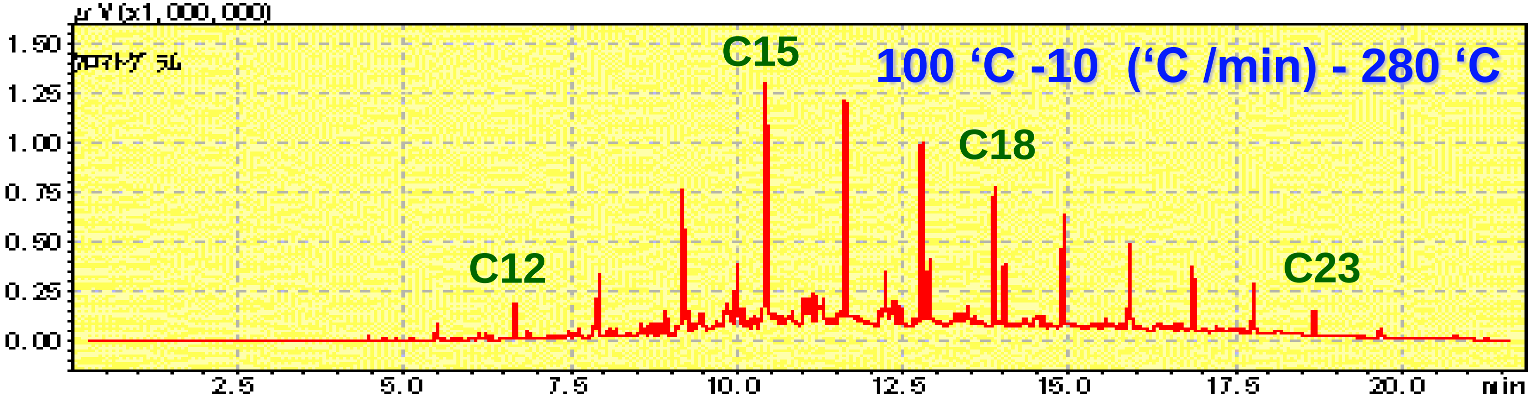


Column & Column Oven

Column Oven : Isothermal VS Temperature Program

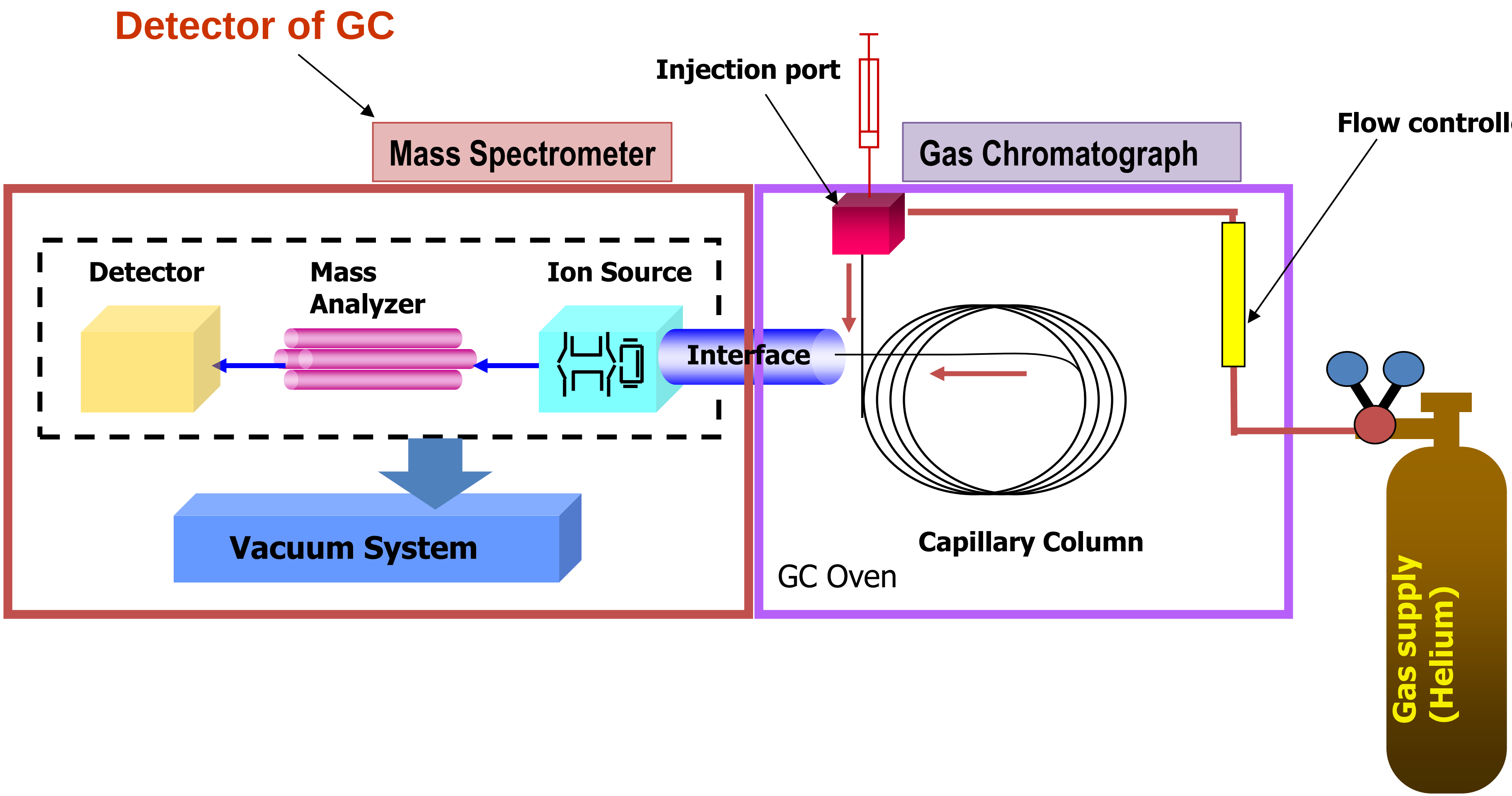


Isothermal

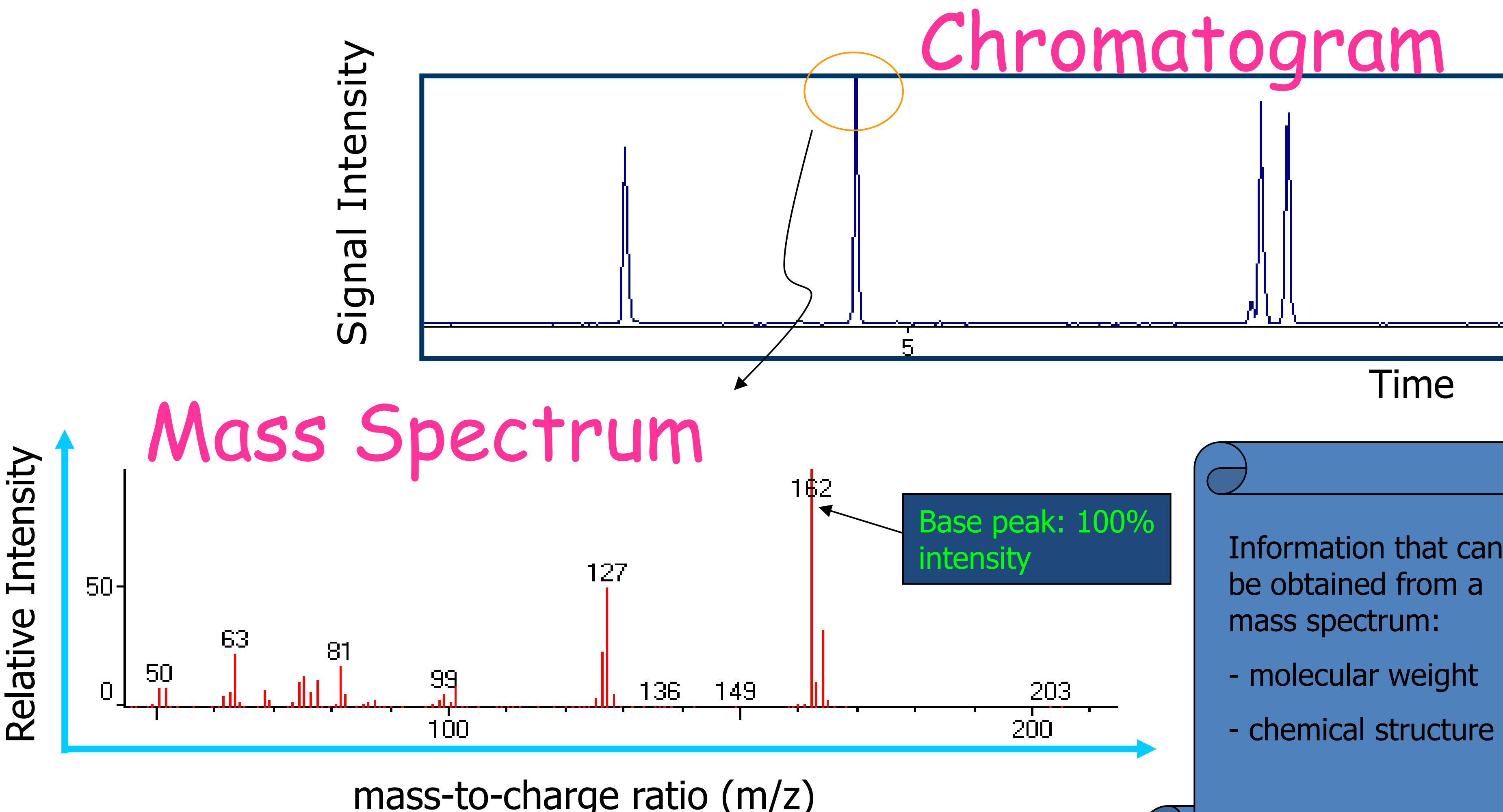


Temperature Program

Gas Chromatograph-Mass Spectrometer Components



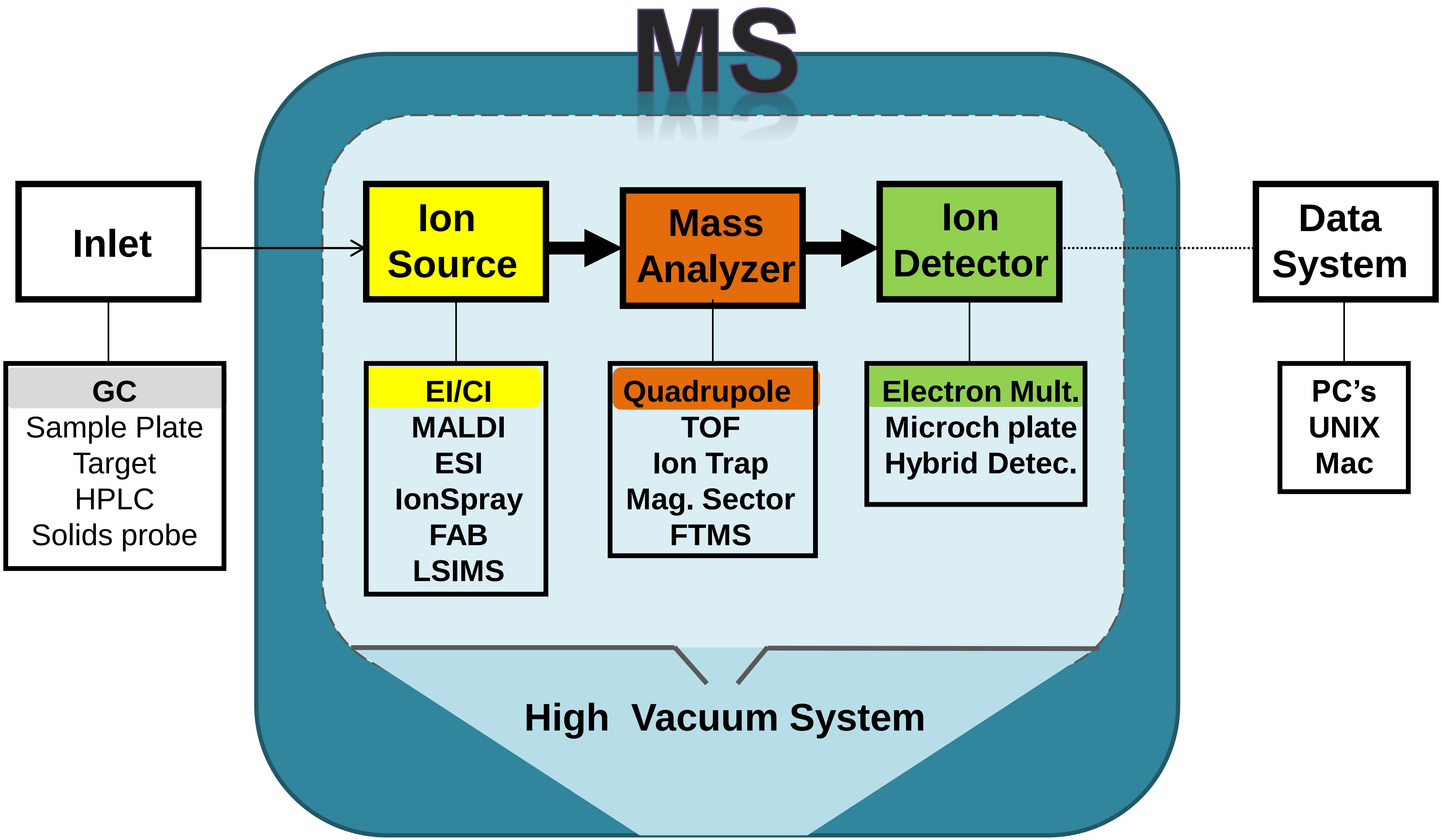
GCMS Data



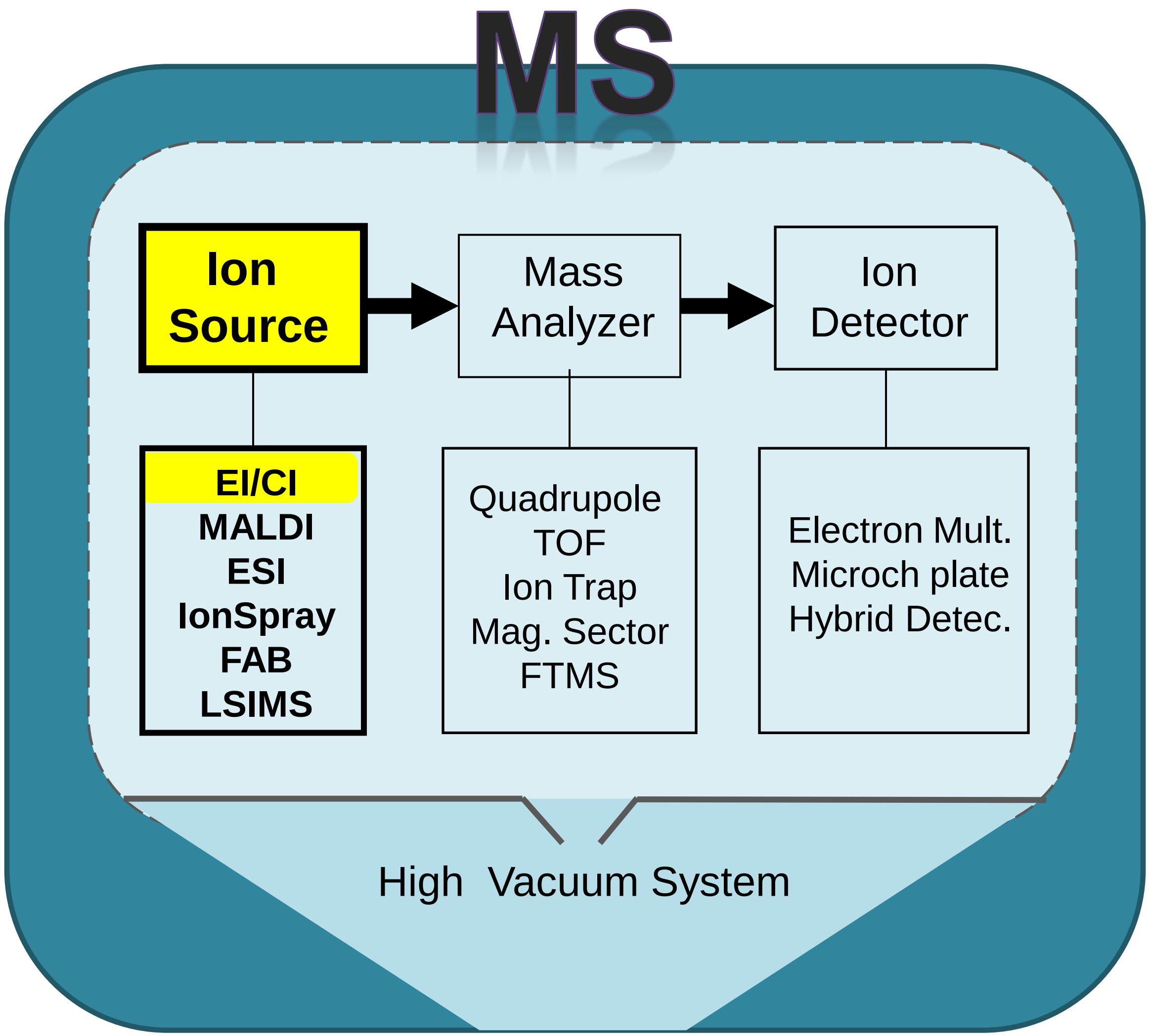
Information that can be obtained from a mass spectrum:

- molecular weight
- chemical structure

Mass Spectrometer Components



Ion Source


MS
Ion Source
Mass Analyzer
Ion Detector
EI/CI
MALDI
ESI
IonSpray
FAB
LSIMS
Quadrupole
TOF
Ion Trap
Mag. Sector
FTMS
Electron Mult.
Microch plate
Hybrid Detec.
High Vacuum System

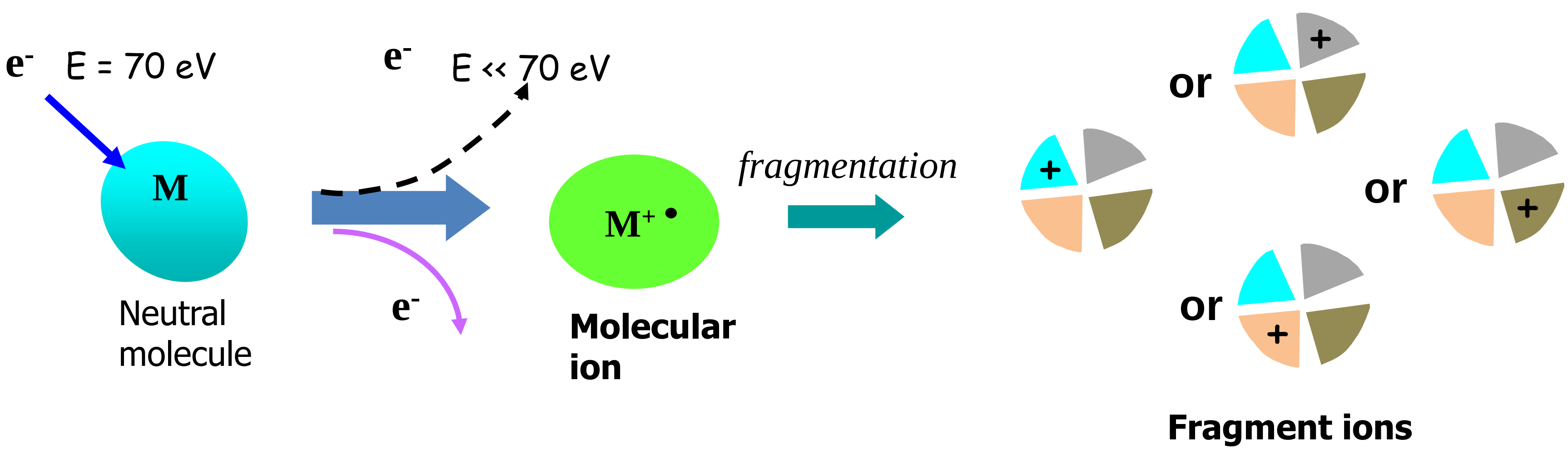
Ion Source

- **Sample molecules are ionized in the ion source after chromatographic separation by a GC. The structure and function of the ion source will be explained here.**

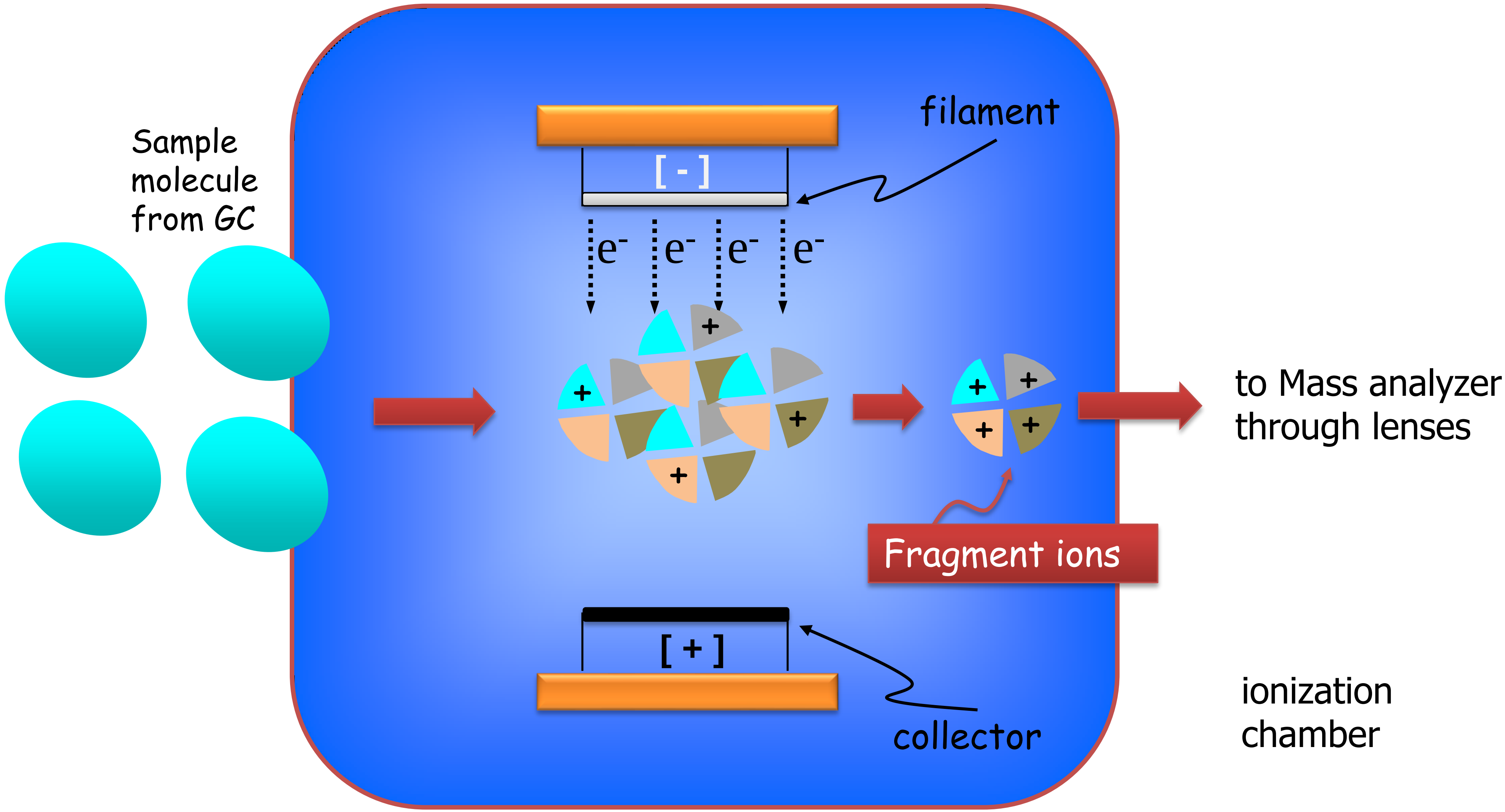
- **EI/CI is one of the important ionization methods for GC/MS.**

EI (Electron Impact Ionization)

- **EI Ionization usually produces singly charged ions (molecular Ion) containing one unpaired electron.**
- **Energy imparted by the electron impact and, more importantly, instability in a molecular ion can cause that ion to break into smaller pieces (fragments).**



EI Ionization in the MS



Electron Impact Ionization

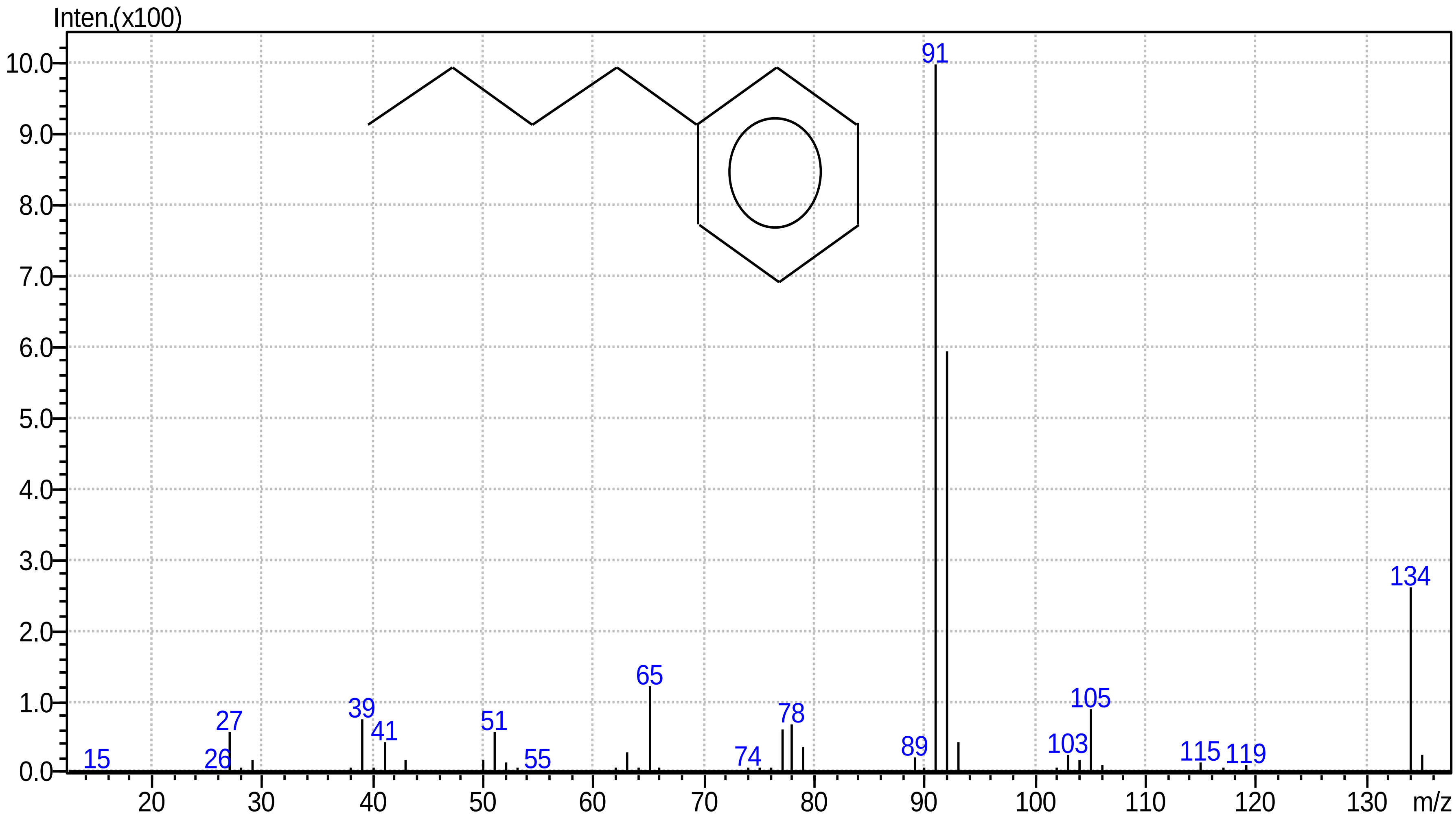
- Extensive fragmentation of sample molecules gives rise to “fingerprint” of compound, useful for identification purpose

- Every time a molecule of the same compound is ionized under the same conditions, it forms the same quantity and pattern of ions.

- m/z value of the molecular ion (M+) gives information about the molecular weight of the compound, also useful for identification purpose

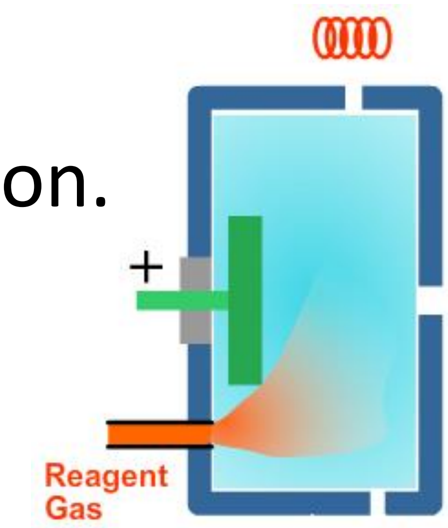
- In some cases, M+ ion does not survive fragmentation due to the high energy involved in the process. Two approaches can be taken:
  - Reduce ionization voltage (possible with GCMS-QP2020)
  - Change ionization mode to Positive Chemical Ionization (PCI) – possible with GCMS-QP2020 or GCMS-QP2020 models

EI Mass Spectrum of Butylbenzene



CI, Chemical Ionization

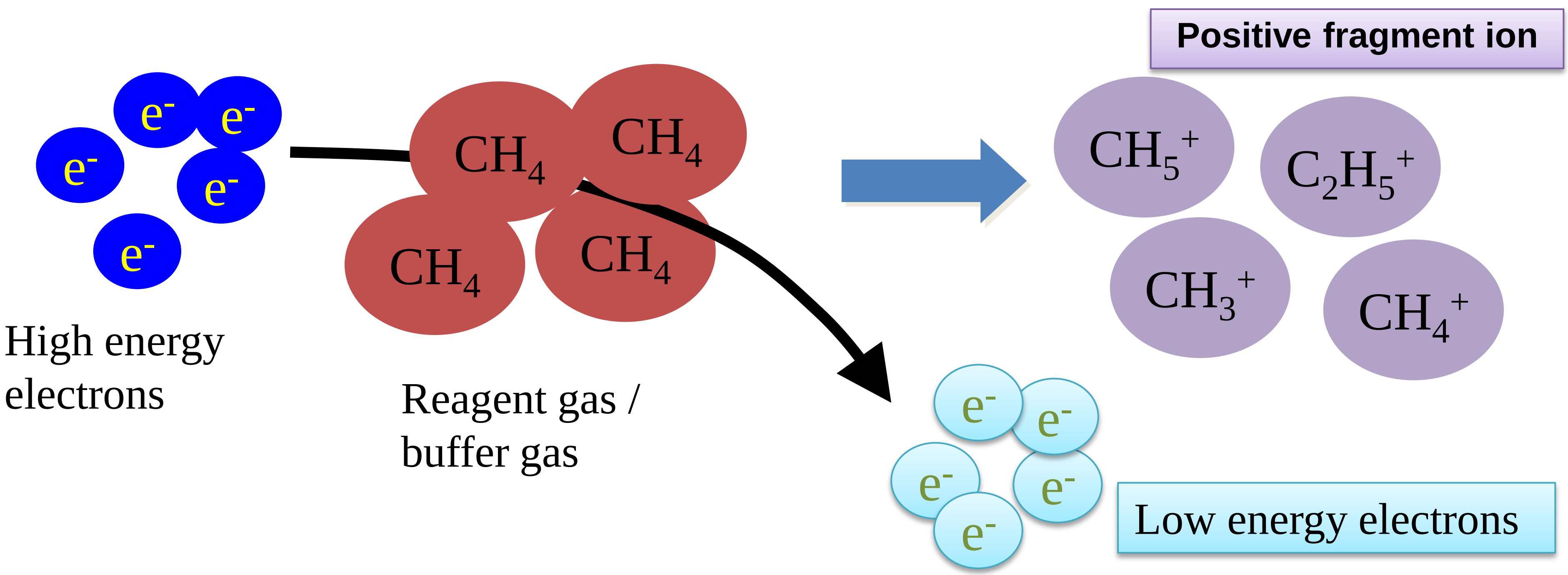
- CI is soft Ionization method causing less fragmentation.
- Reagent gas is used for this ionization. (methane, iso-butane or ammonia)



- 2 type of CI method: PCI and NCI.

CI – Reagent gas

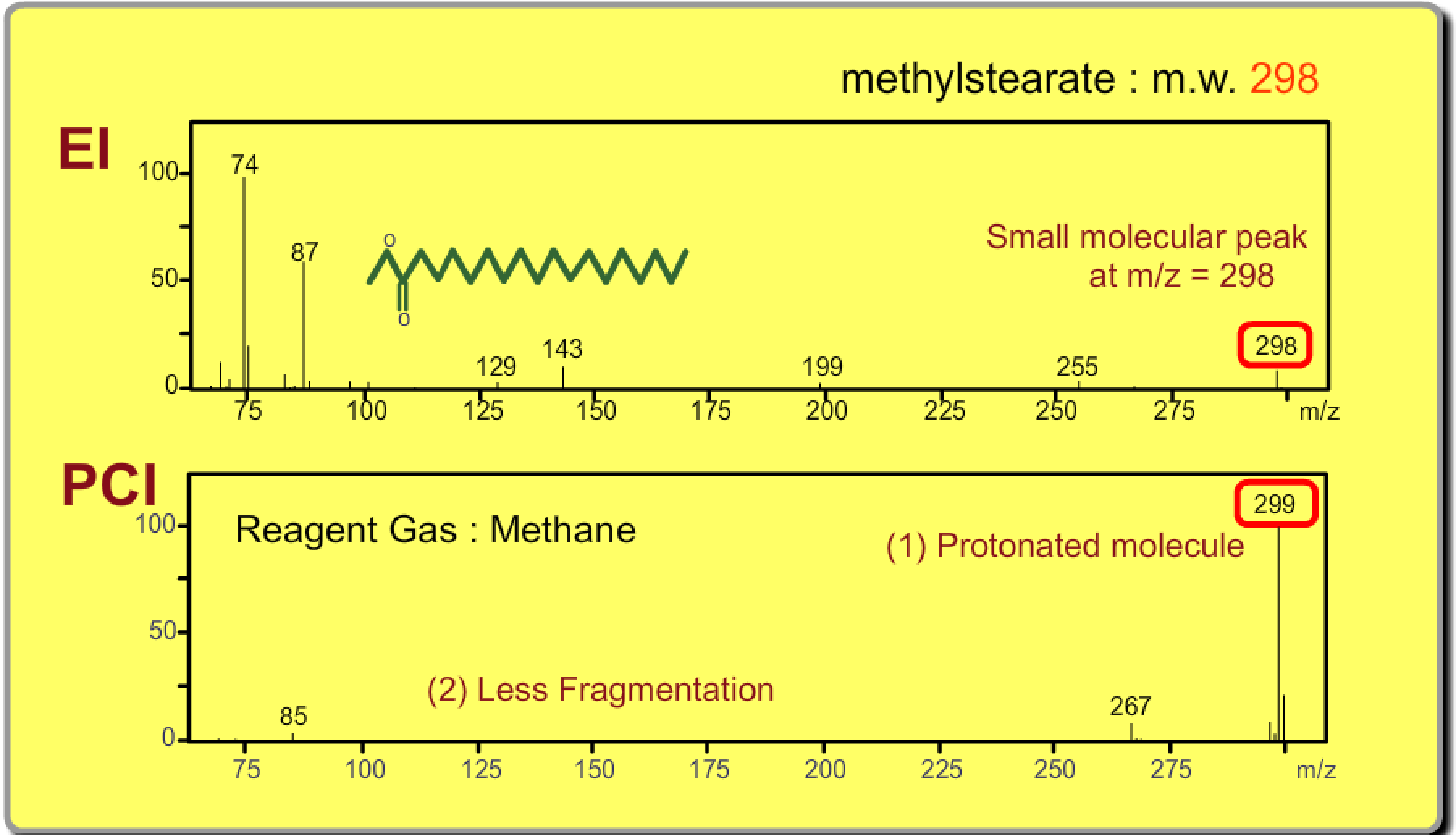
- Low energy electrons and positive fragment ion are produced when high energy electrons from the filament strike the reagent gas.



PCI, Positive chemical Ionization

- Through collision and chemical reaction, the reagent gas ions transfer energy to, and ionize the sample molecules.
- As a result, molecular ions are formed with much less excess internal energy, which leads to less fragmentation.
- The molecular ion thus appears as a strong, if not the major, fragment in the mass spectrum.
- This fragment can be used as quick identification of the molecular weight of the sample molecule.

PCI- Spectrum of PCI



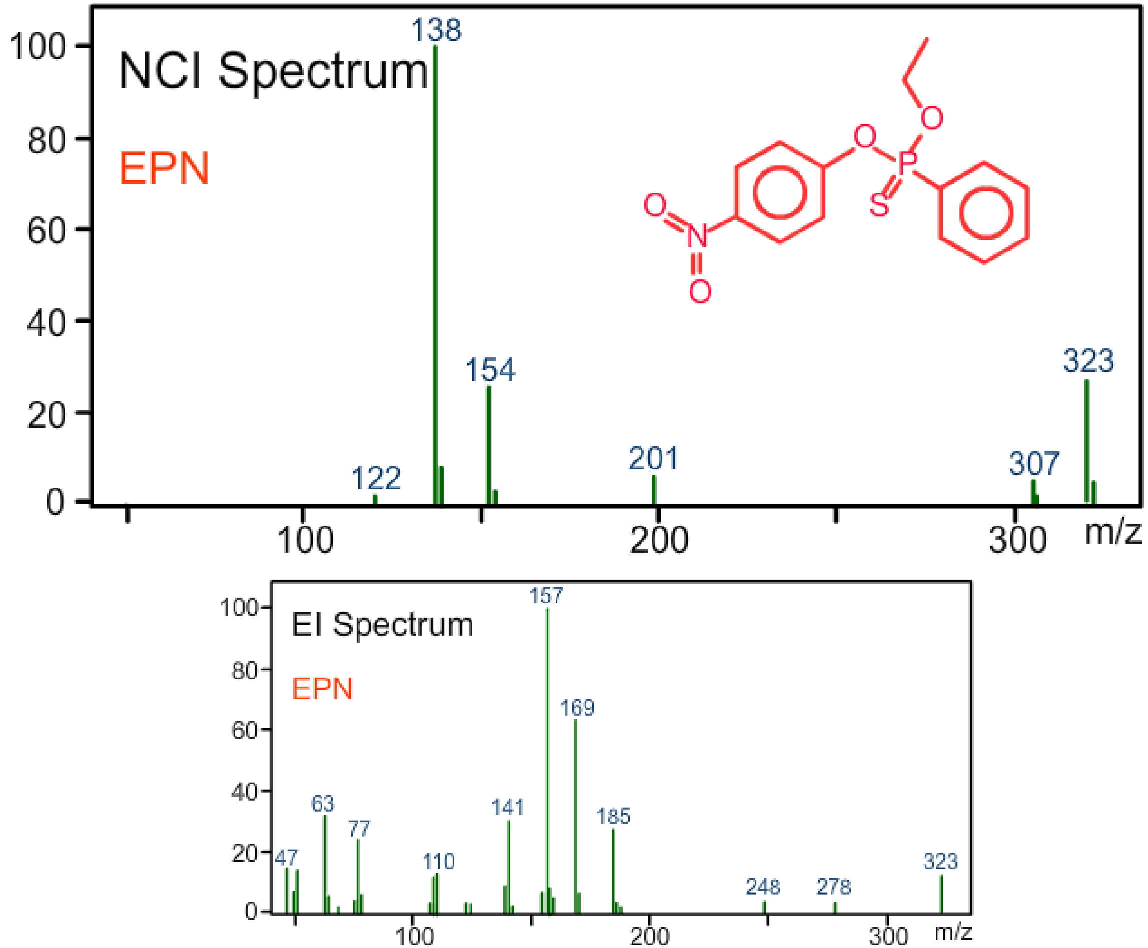
PCI- Reaction Ionizing sample molecule.

Proton Transfer	The major reaction of PCI. A proton (Hydrogen ion) H^+ is transferred from the reactant ion $[BH]^+$ to sample molecule M to form protonated molecule $[M+H]^+$ $M+[BH]^+ \rightarrow [M+H]^+ + B$
Addition of Reactant Ion	The reactant ion X+ is added to sample molecule to form adduct ion $[M+X]^+$. $M+X^+ \rightarrow [M+X]^+$
Hydride Abstraction	A Hydride ion is abstracted from the sample molecule to form the ion $[M-H]^+$ $M+[BH]^+ \rightarrow [M-H]^+ + BH_2$
Charge Exchange	Charge exchange between reactant ion $[BH]^+$ and sample molecule M. $M+[BH]^+ \rightarrow M^{+\cdot} + BH^{\cdot}$

NCI, Negative chemical Ionization

- Negative Chemical Ionization is used for selective detection of compounds that can produce negative ions.
- Electron is captured or attached to a neutral molecule, giving molecular anion.
- For electron capture, energy of electrons must be very low.

NCI, Spectrum of NCI.



NCI, Negative Ion Production Processes.

Resonance Electron Capture

A electron with kinetic energy 0-2 eV is directly captured by a molecule without fragmentation. This process forms molecular ion $[M]^-$

$$M+e^- \rightarrow [M]^-$$

Dissociative Electron Capture

A electron with kinetic energy 0-15 eV is captured by a molecule. Excess energy of electron leads to produce fragment ion $[M-A]^-$

$$M+e^- \rightarrow [M-A]^- + A$$

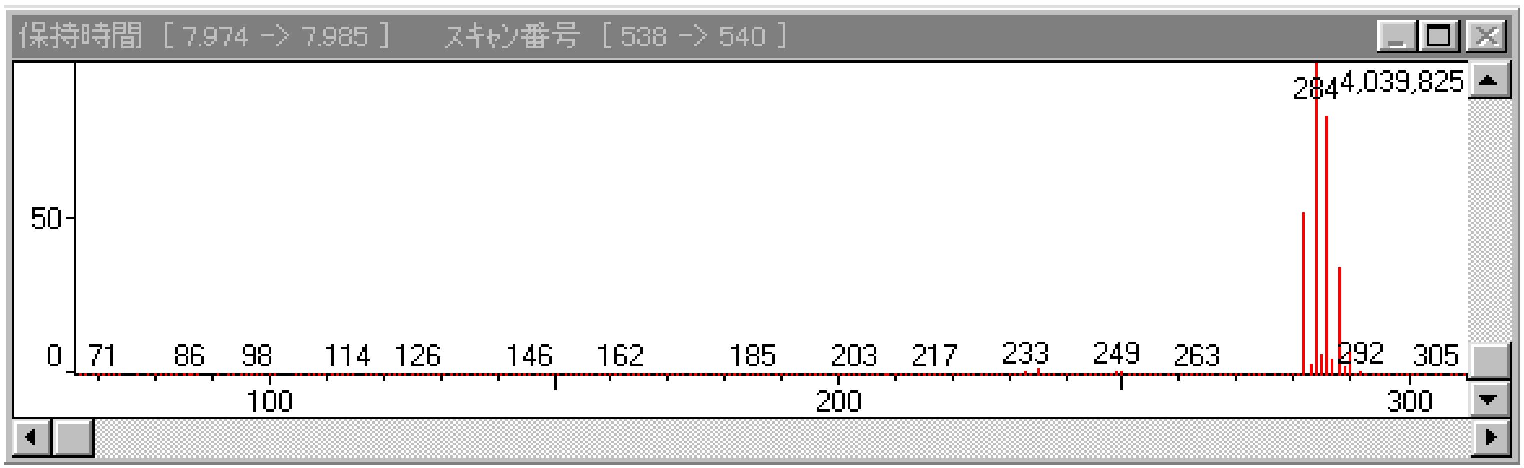
Ion Pair Formation

Bombardment of a electron with kinetic energy more than 10eV fragment a molecule to produce a pair of positive and negative ions.

$$M+e^- \rightarrow [M-B]^- + B^+ + e^-$$

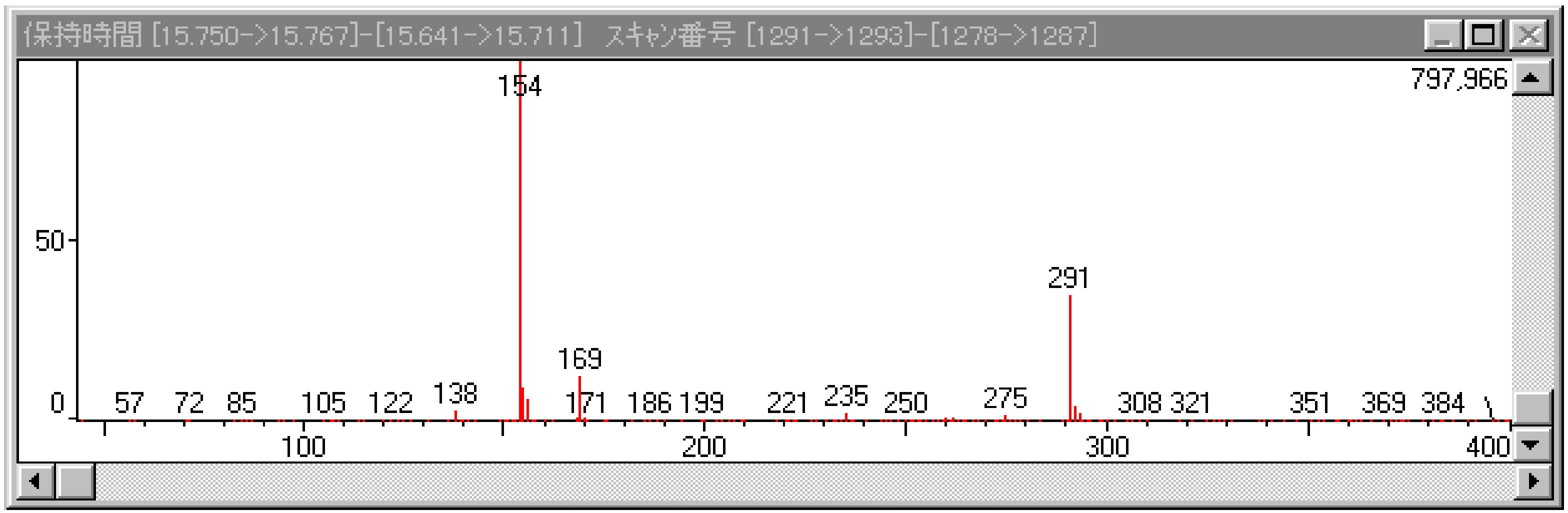
NCI Mass Spectrum of Hexachlorobenzene

- Hexachlorobenzene (MW = 284)

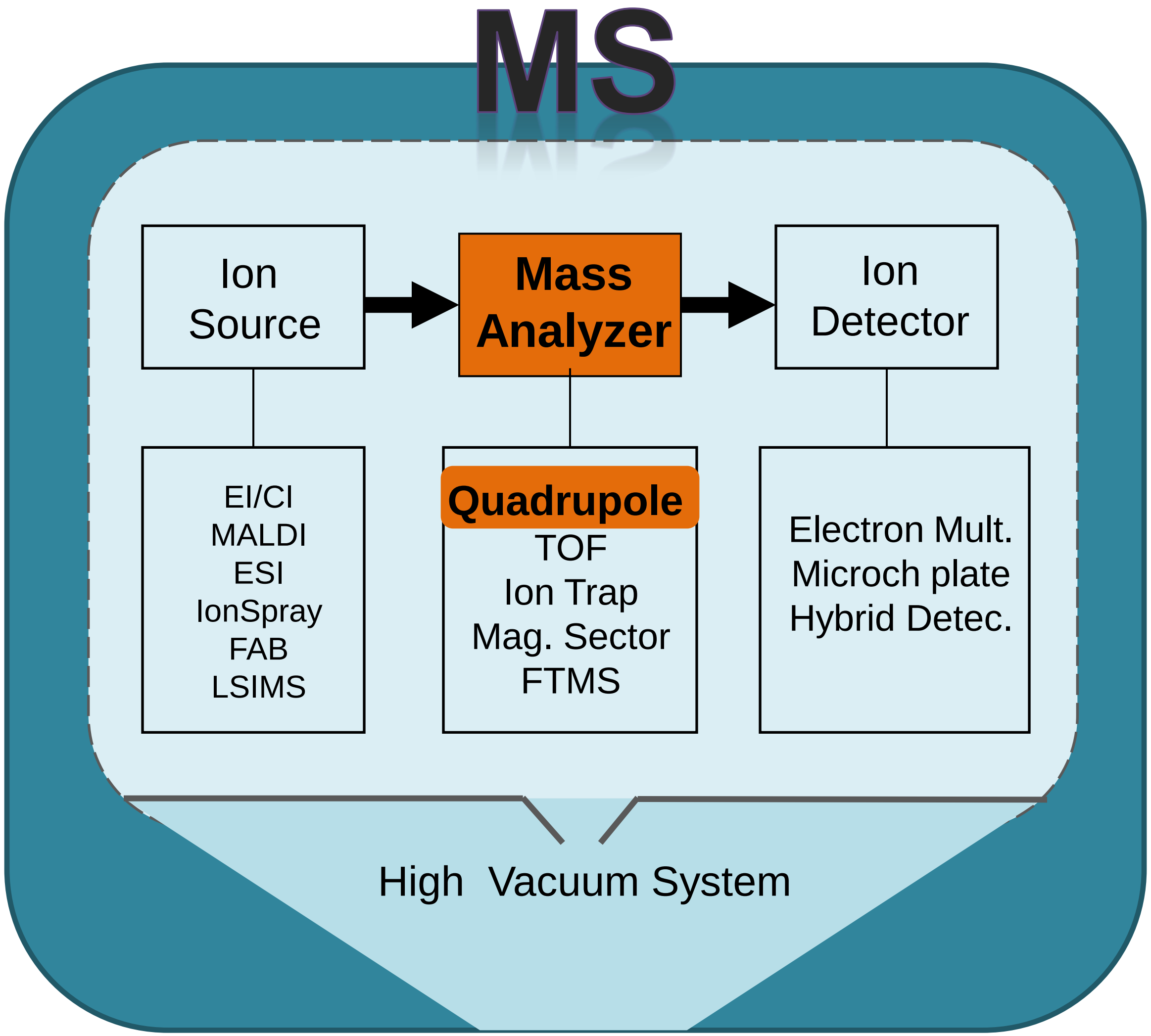


NCI Mass Spectrum of Parathion

- Parathion (MW = 291)

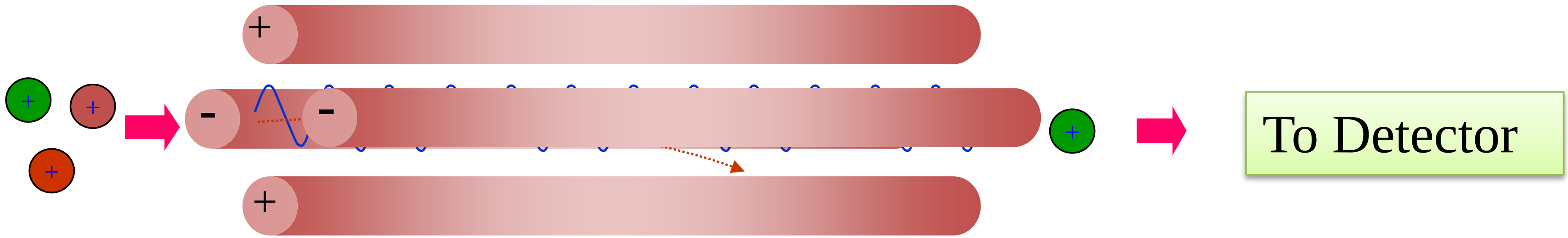


Mass Analyzer



Quadrupole Mass Spectrometer

- A quadrupole(QP) mass analyzer is most extensively used for GC/MS analysis.
- Voltages applied to four cylindrical quadrupole rods generate a quadrupole field in the center axis of the mass spectrometer
- Ions entering the field will oscillate; the oscillation characteristics depend on the mass-to-charge ratio of the ions and the voltage applied
- Voltage applied to the quadrupole can be varied/selected so that only ions with specific m/z will have stable oscillations in the center axis, and will reach the ion detector
- Quadrupole MS can separate ions whose mass number differ by 1 unit mass



How a Quadrupole MS operates in GC/MS

- Voltages applied to the quadrupole are varied so that the mass range specified are scanned repeatedly at high frequency.
- Scan Mode and SIM Mode are the generally used analysis mode.

Scan mode & SIM mode

SCAN MODE

Qualitative Analysis and Quantitative Analysis

Scan mode is used for the identification of chemical components using a mass spectrum, quantitative analysis and determination of some parameters for SIM analysis.

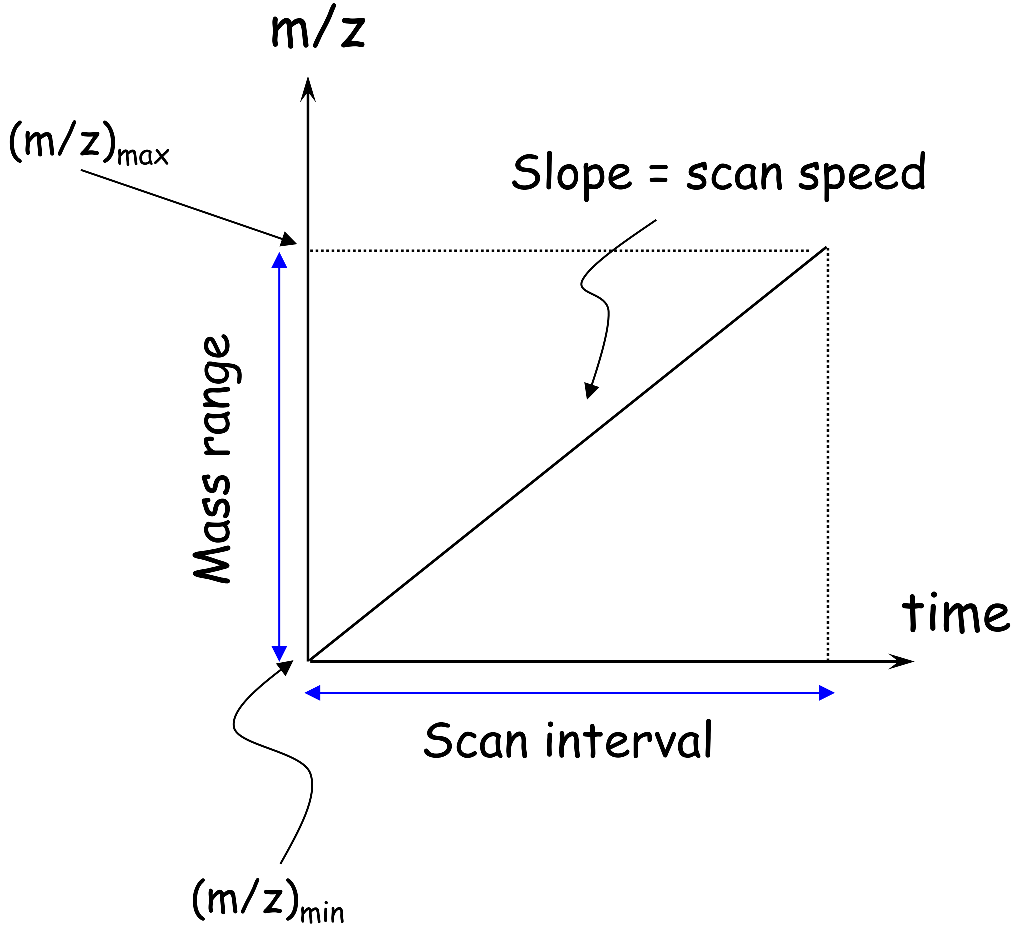
SIM MODE

Quantitative Analysis

Lower detection limits can be obtained with the SIM mode than the scan mode for quantitative analysis, because the sensitivity is tens to hundreds times better.

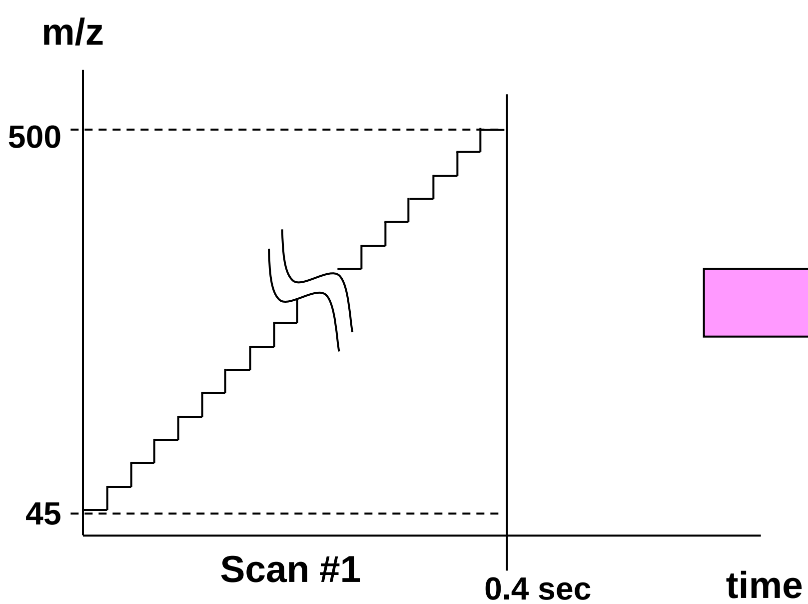
Scan Mode

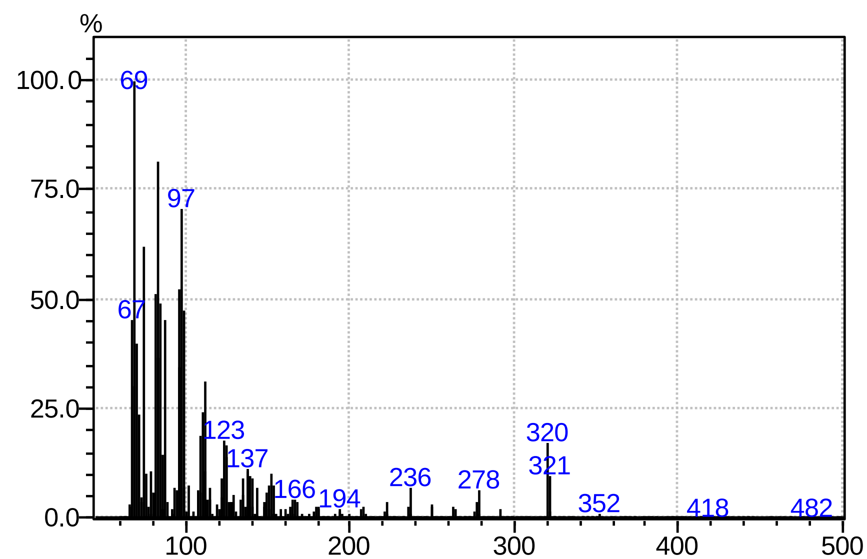
- The mass analyzer is scanned from low to high field strength within a scan interval
- Ions having m/z within the mass range are scanned and detected
- The result is called a full-scan mass spectrum



Full Scan or SCAN Mode

- Mass numbers are scanned in steps of 1 u
- Good for qualitative analysis (identification)

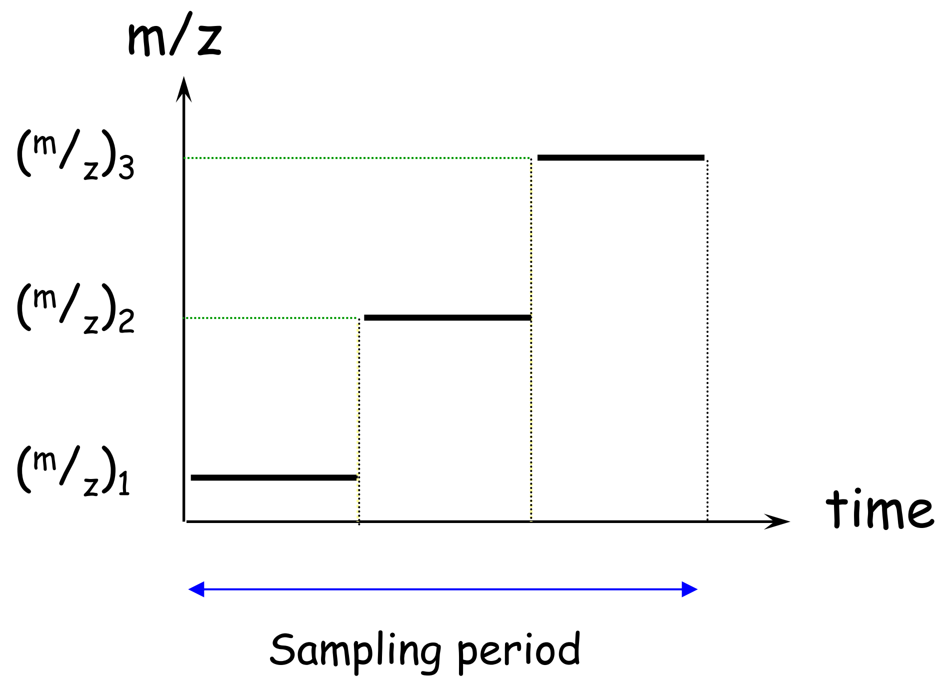




SIM (Selected Ion Monitoring) Mode

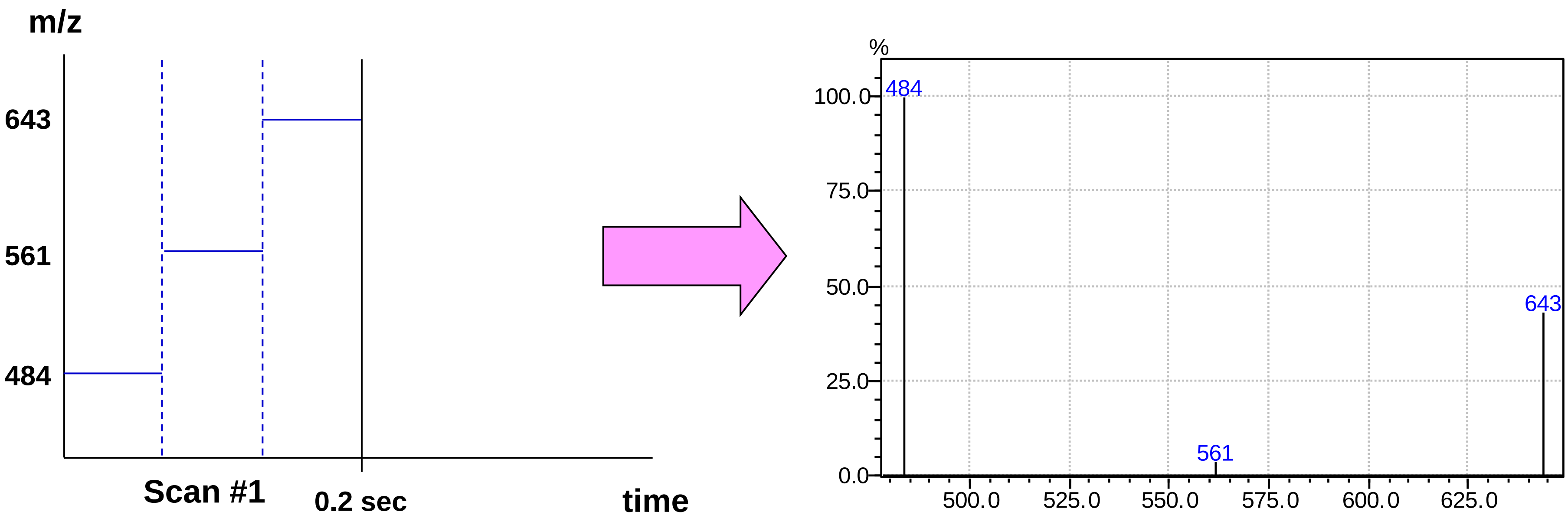
- Detect only a small number of ions (pre-determined m/z values)

- Increase the sensitivity of detection
  - Detector has a longer time to measure the abundance of ion fragment of a particular m/z

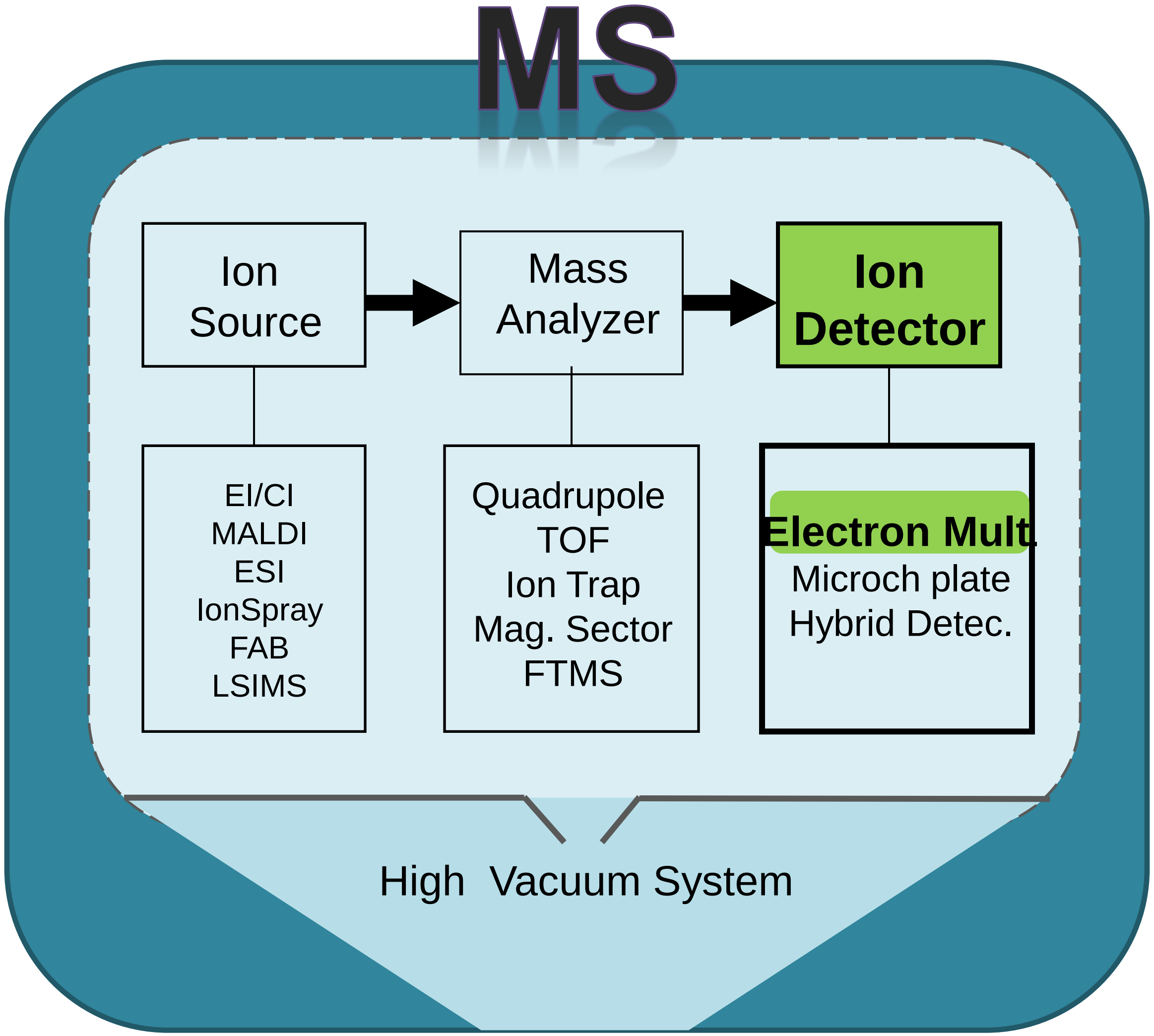


Selected Ion Monitoring Mode

- Only pre-determined (user-specified) mass numbers are scanned
  - E.g. m/z 200, m/z 215
- More sensitive than Scan mode

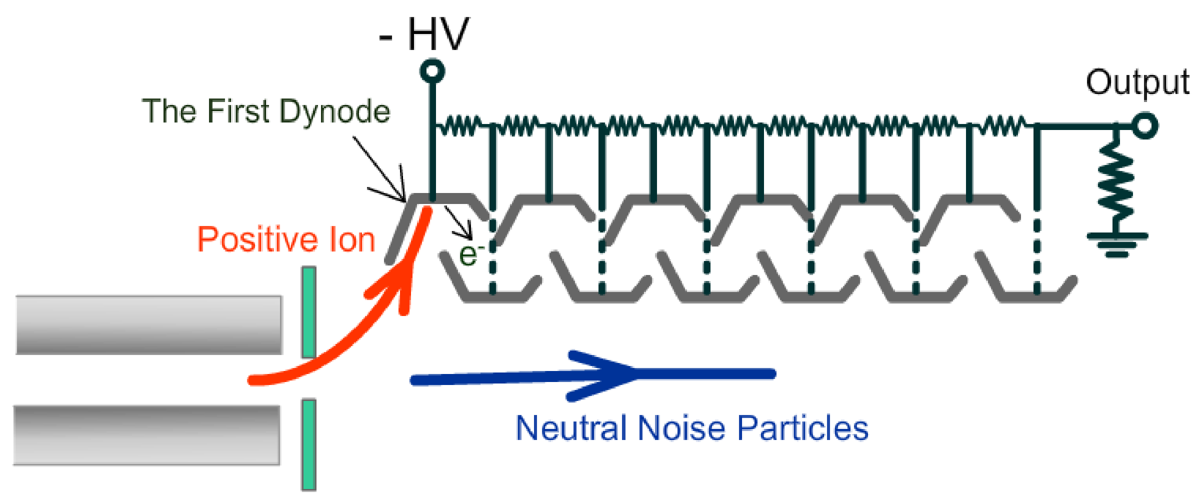


Ion Detector



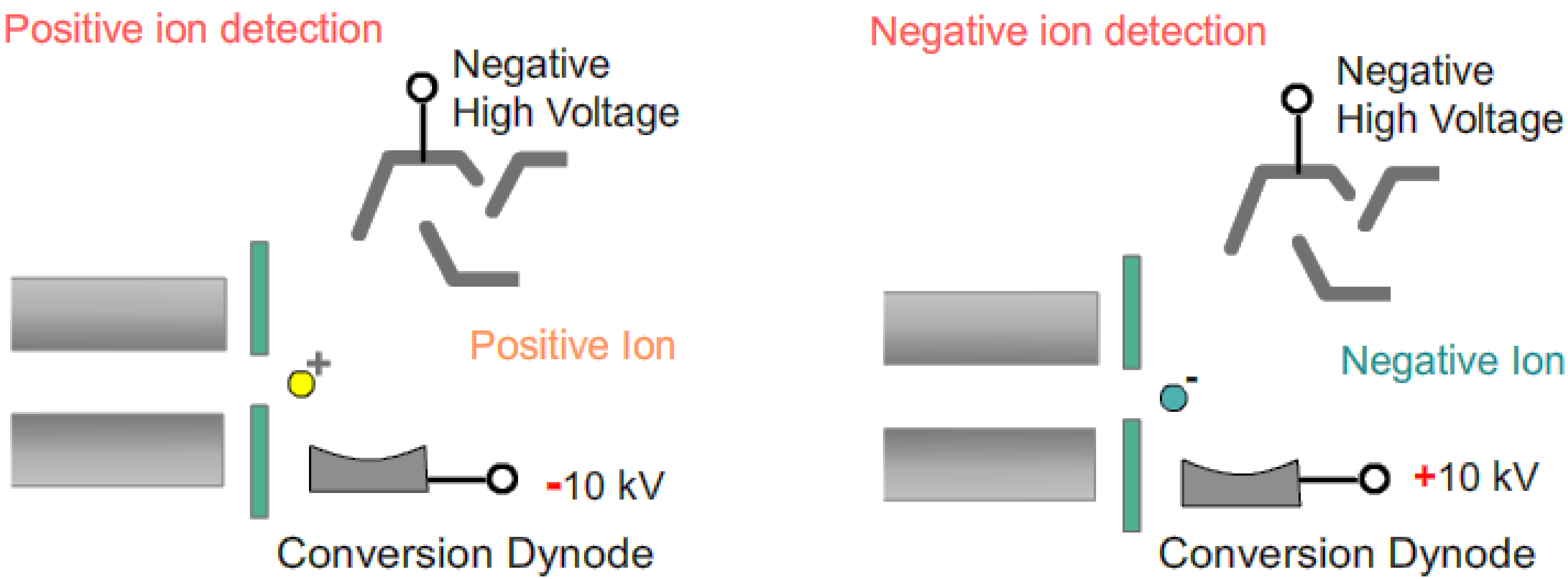
Electron Multiplier

- Ions that exit the mass analyzer are detected and the abundance / intensities are measured
  - Bench-top GC-MS usually use electron multiplier detector

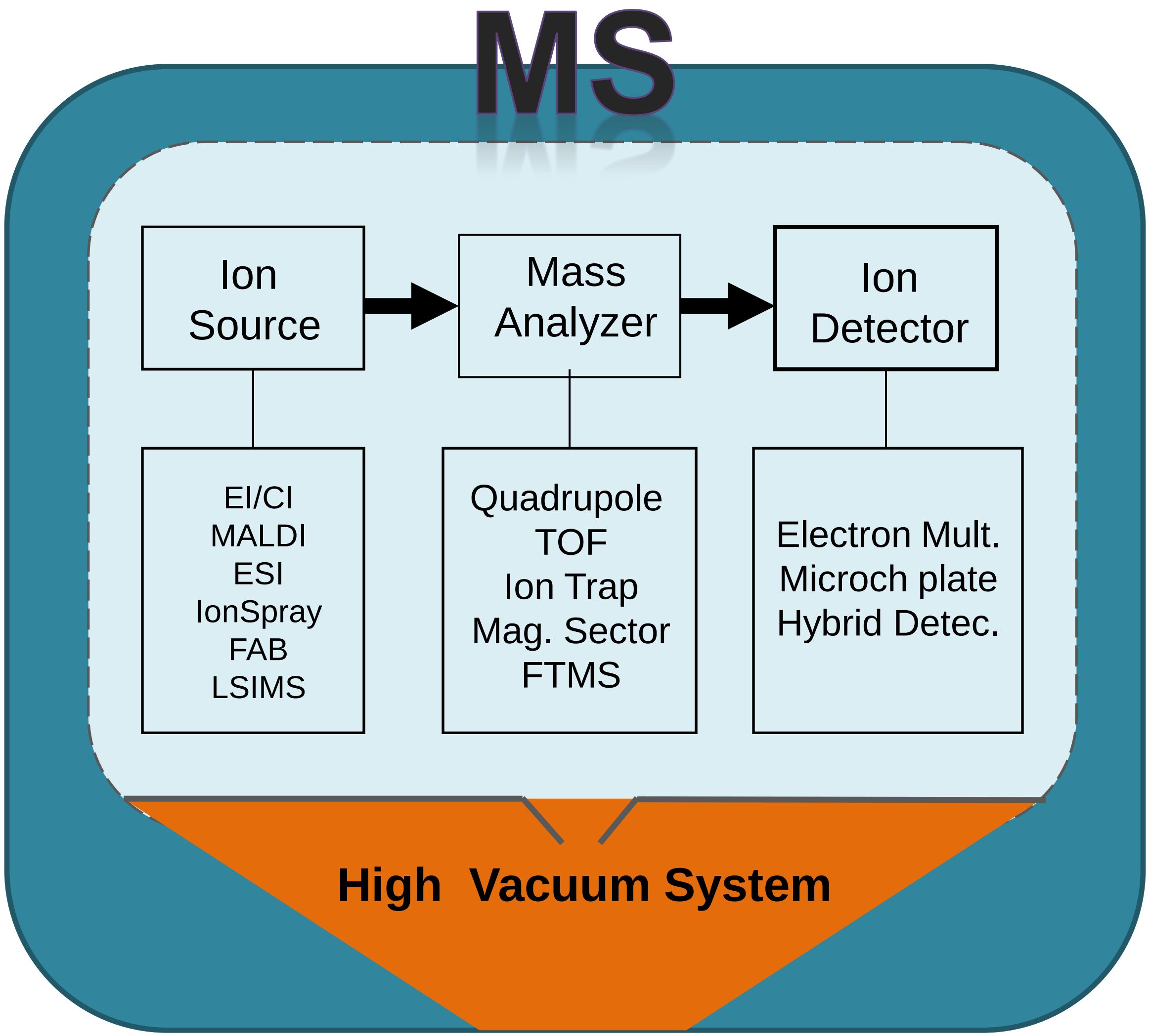


Discrete Dynode Type

Positive & Negative Ion Detection



High Vacuum System

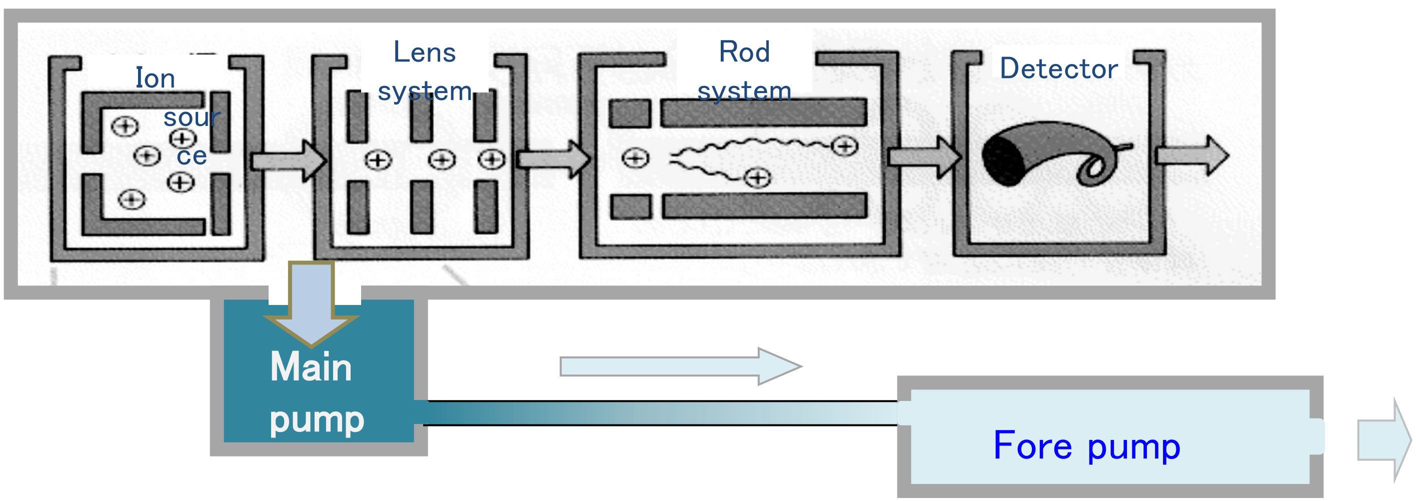


Vacuum System

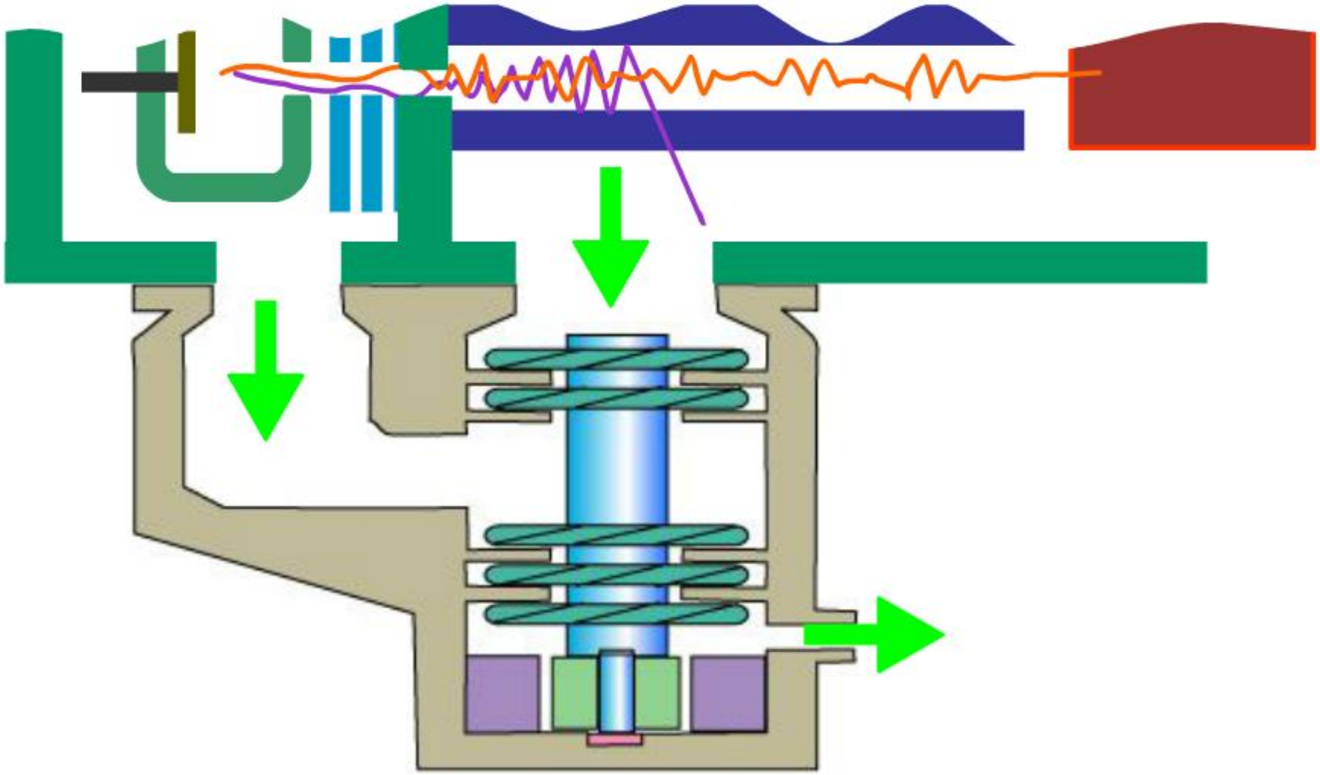
- Two types normally used for bench-top GC/MS:
  - High vacuum pump
    - Reduces and maintain pressure in MS at the operating pressure

      (e.g.: Turbo Molecular Pump)
  - Low vacuum pump
    - Reduces the pressure in MS from atmospheric pressure to the outlet pressure of the high vacuum pump

    (e.g.: Rotary Pump)



Vacuum System



TMP in QP2020

Why is Vacuum Necessary?

- Sample ions must travel from ion source to the detector without, or with minimum collisions with other particles

- Reduce ion-molecule reactions
- Reduce background interference
- Increase sensitivity

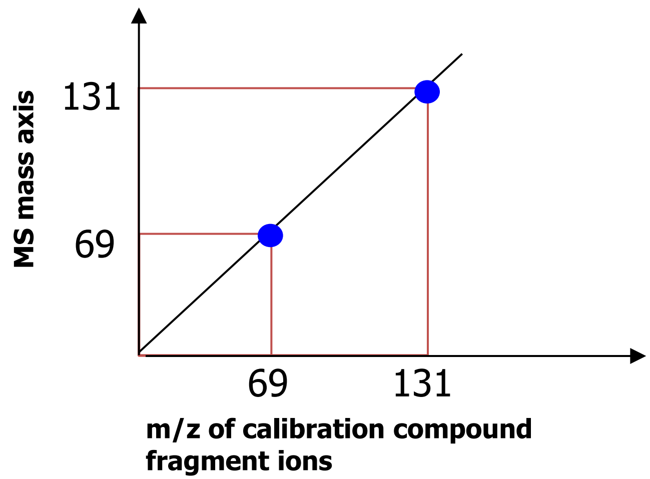
- Increase filament lifetime

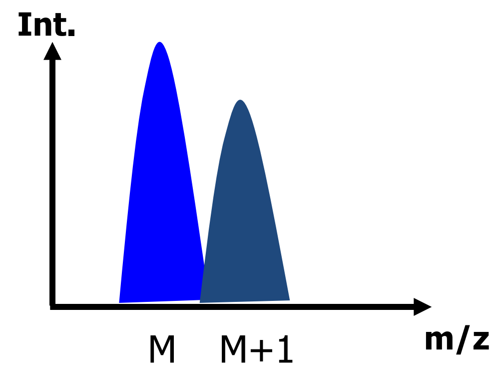
Mass Spectrometer Calibration & Tuning

- MS calibration (calibration of mass number) means

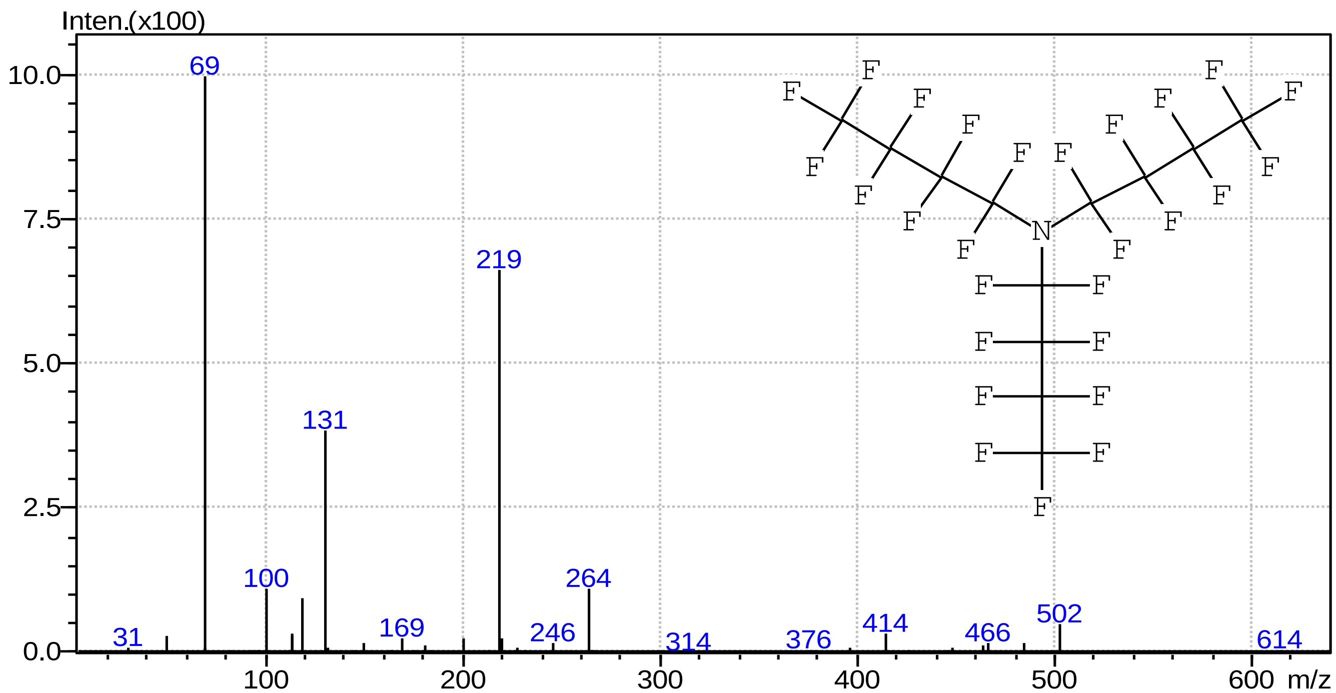
  adjusting the quadrupole signal frequency so that mass axis points correspond to expected fragments of calibration compound

- Tuning is the adjustment of ion optics voltages so that
  - adjacent mass peaks have little overlap (resolution adjustment),
  - ion abundances are optimised (sensitivity adjustment), and/or
  - fragment ions have the expected relative abundance (mass pattern adjustment)





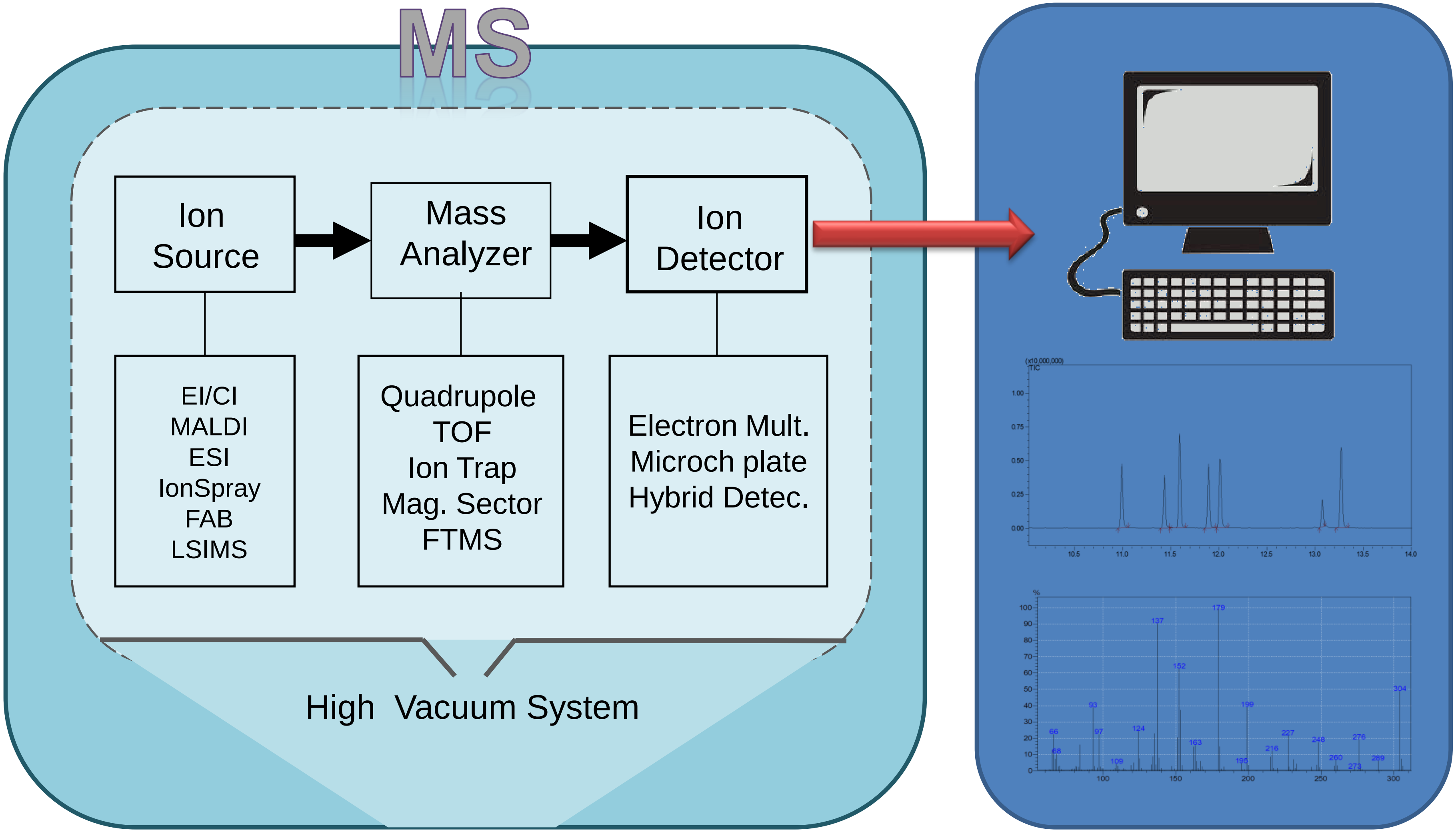
Calibration Compound (Standard)



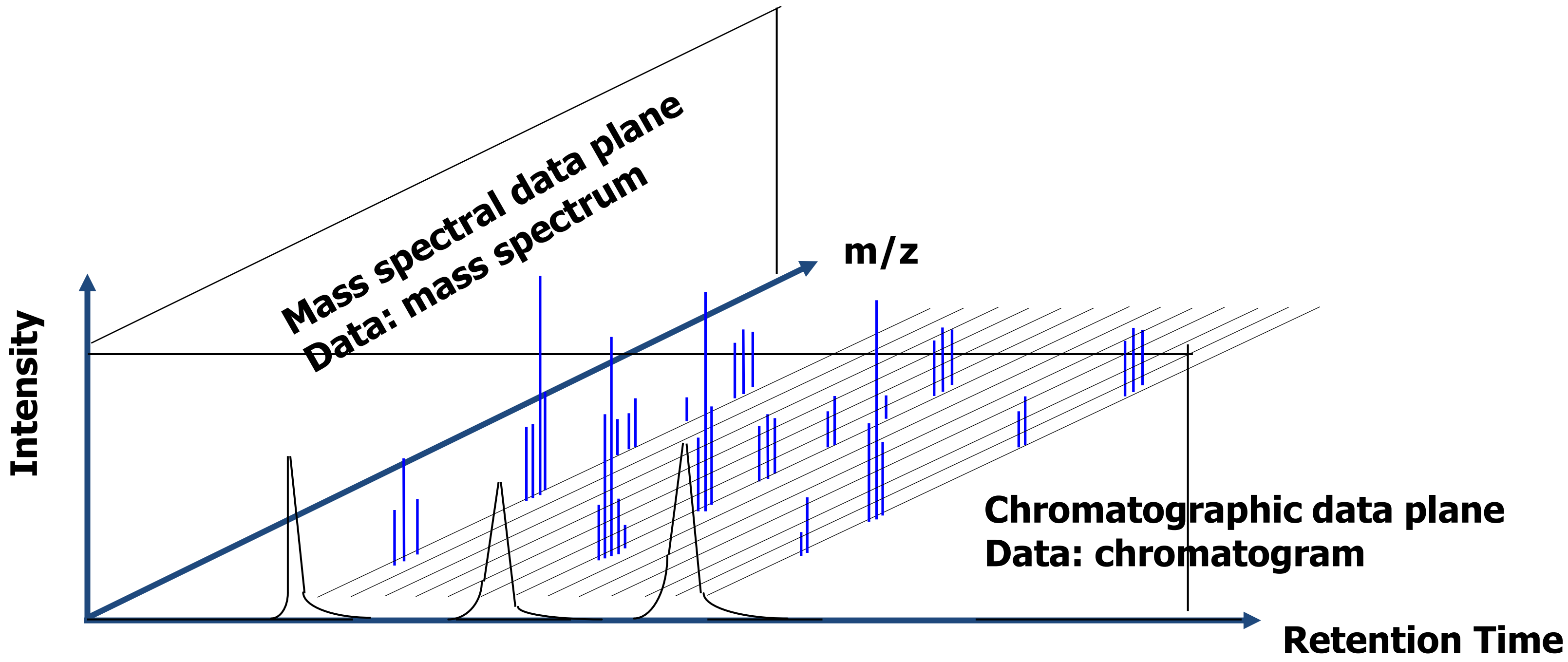
PFTBA = Perfluorotributylamine (MW = 671)

For normal tuning (mass axis calibration up to ~700 amu)

GCMS Data


MS
Ion Source
Mass Analyzer
Ion Detector
EI/CI
MALDI
ESI
IonSpray
FAB
LSIMS
Quadrupole
TOF
Ion Trap
Mag. Sector
FTMS
Electron Mult.
Microch plate
Hybrid Detec.
High Vacuum System

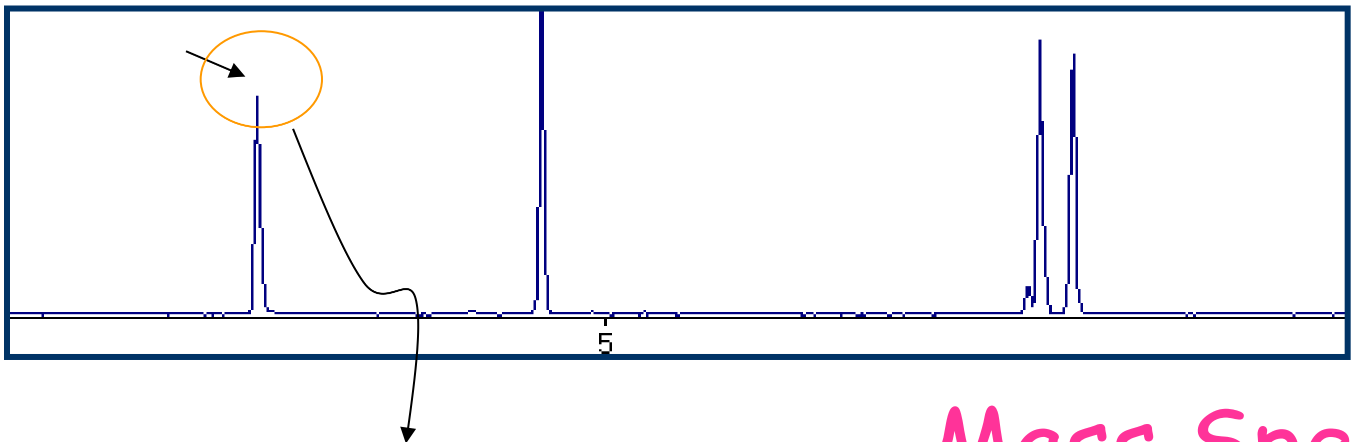
GCMS Data



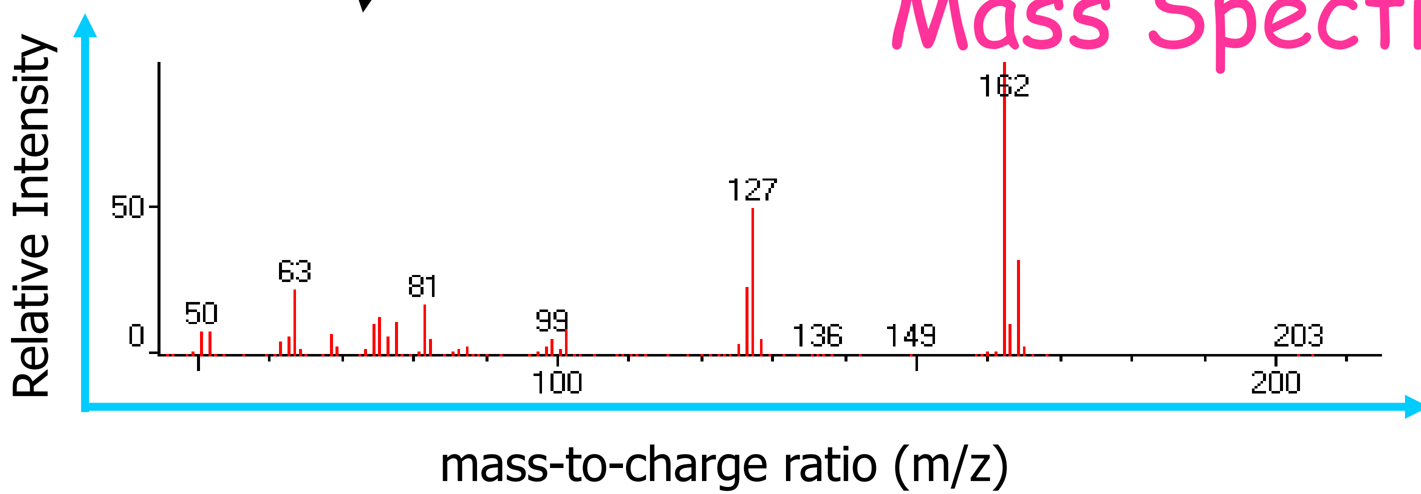
Mass spectral data plane
Data: mass spectrum
m/z
Intensity
Chromatographic data plane
Data: chromatogram
Retention Time

GCMS Data(2)

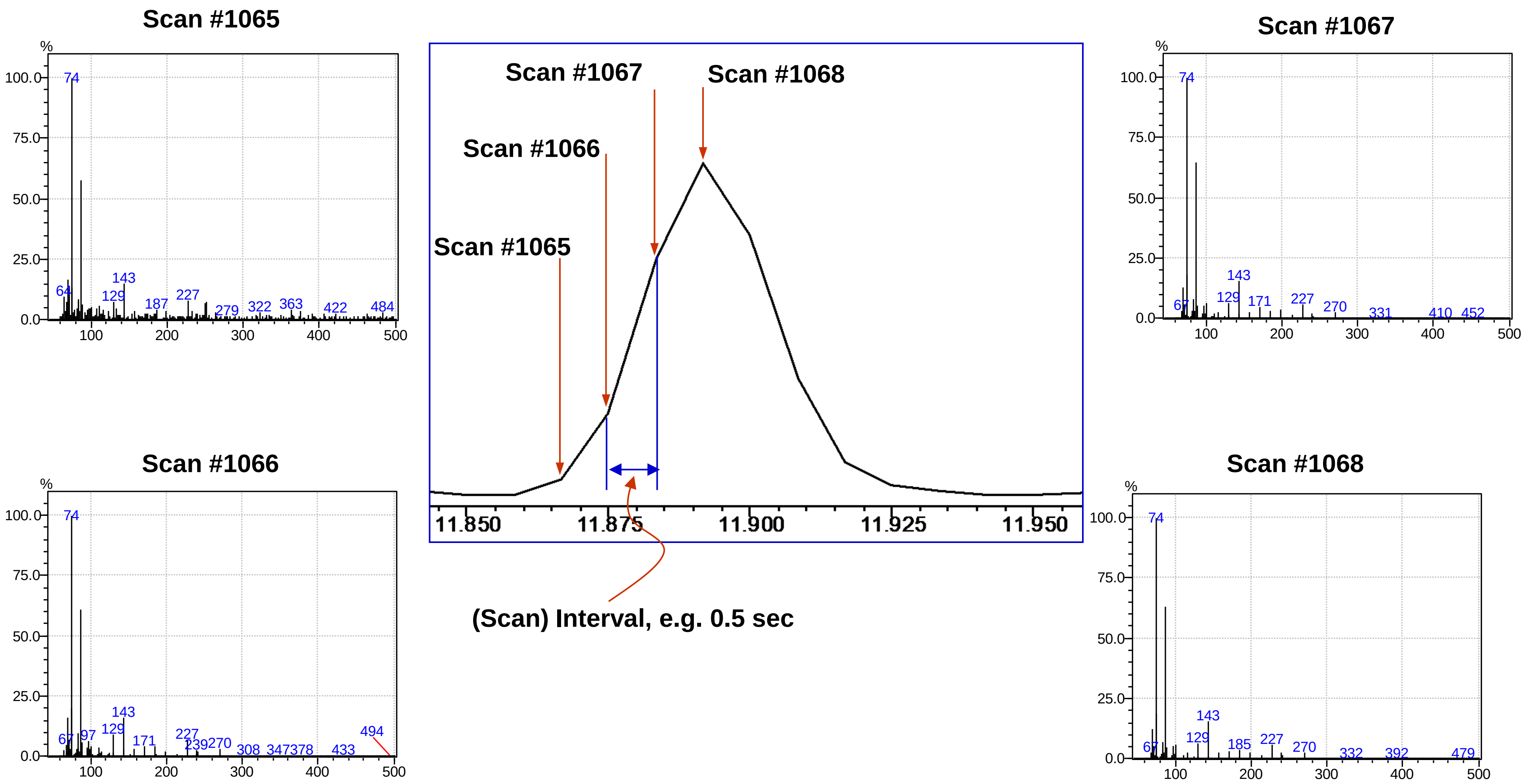
Chromatogram



Mass Spectrum

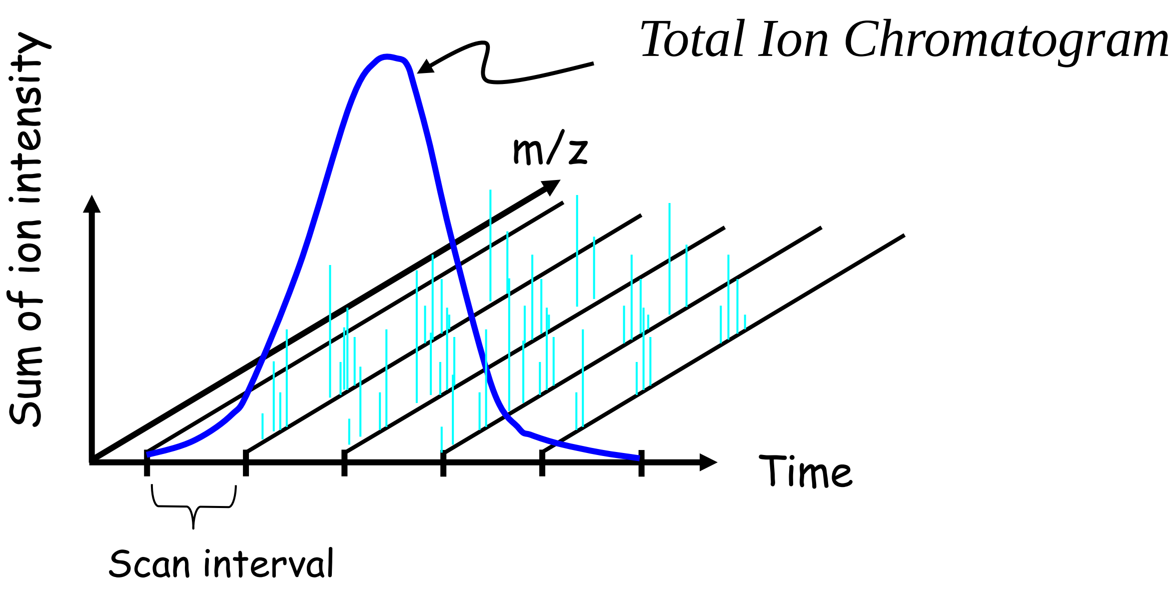


Chromatographic Peak & Mass Spectrum



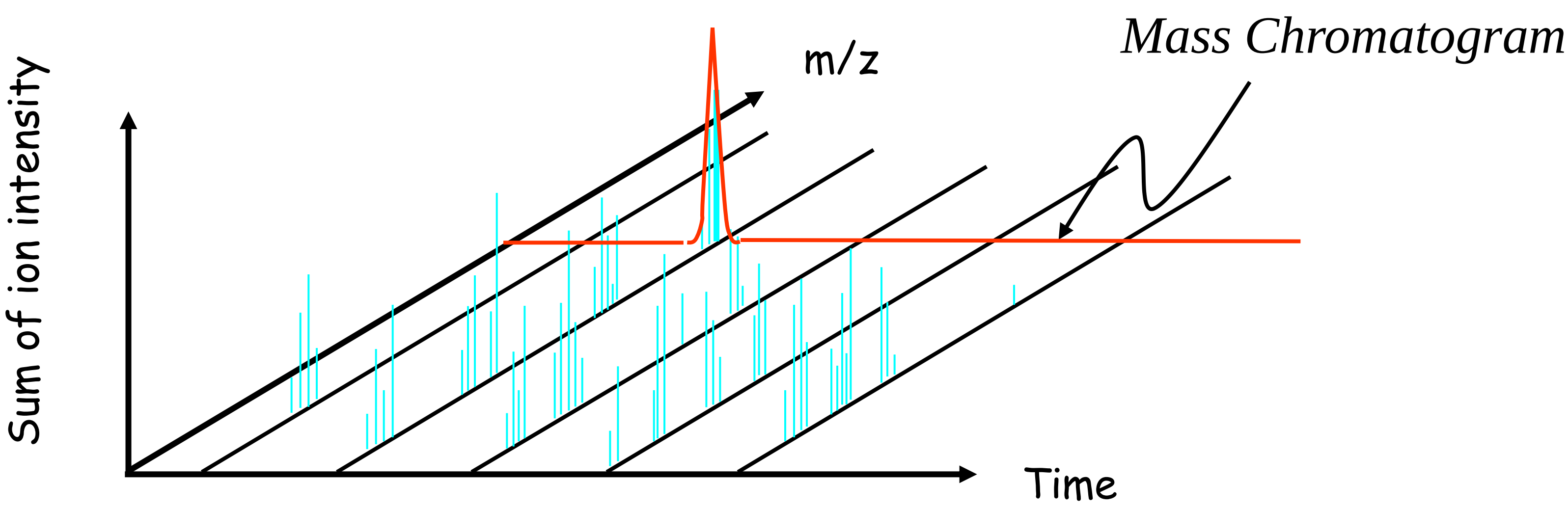
Total Ion Chromatogram (TIC)

- The summed raw signal of total ion current plotted against time

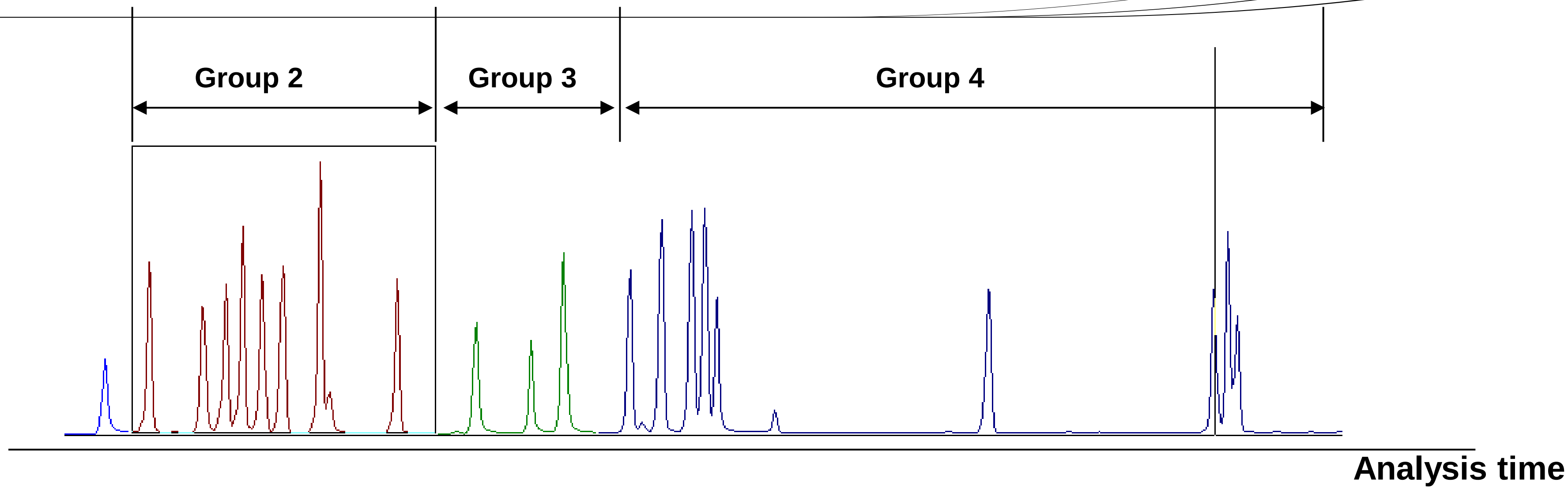


Mass Chromatogram (MC)

- A plot of the intensity of a single ion (m/z) against time



Setting of MS Acquisition Modes



	Start Time (min)	End Time (min)	Acq. Mode	Interval (sec)	Scan Speed	Start m/z	End m/z	Ch1 m/z	Ch2 m/z
1	5.00	6.50	Scan	0.40	769	70.00	370.00		
2	6.50	8.50	Scan	0.50	270	70.00	200.00		
3	8.50	10.37	Scan	0.50	357	70.00	240.00		
4	10.37	12.00	Scan	0.50	625	60.00	360.00		
5	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]	[illegible]		

	Start Time (min)	End Time (min)	Acq. Mode	Interval (sec)	Scan Speed	Start m/z	End m/z	Ch1 m/z	Ch2 m/z	Ch3 m/z	Ch4 m/z	Ch5 m/z
1	3.00	8.00	SIM	0.20				181.00	219.00	0.00	0.00	0.00
2	8.00	15.00	SIM	0.20				186.00	201.00	0.00	0.00	0.00
3	15.00	20.00	SIM	0.20				179.00	304.00	0.00	0.00	0.00
4	20.00	30.00	SIM	0.20				200.00	215.00	0.00	0.00	0.00
5	0.00	0.00	Scan	0.00	0	0.00	0.00					

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THANKS YOU

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