

# CHROMacademy

e-learning for the analytical chemistry community

## Mass Spectrometry

## Fundamental LC-MS

## Introduction



Wherever you see this symbol, it is important to access the on-line course as there is interactive material that cannot be fully shown in this reference manual.



## Aims and Objectives

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### Aims

- Introduce fundamental LC/MS concepts
- Explain the function of each major component of the LC/MS system
- Indicate the major advantages of LC/MS and the application areas in which it is used

### Objectives

*At the end of this Section you should be able to:*

- Describe the function of the various elements that are present in a typical LC/MS system
- List and explain the two main considerations common to all interface types
- List the most common interfaces and be able to clearly describe the differences between them
- List the most common mass analyzer types

## Content

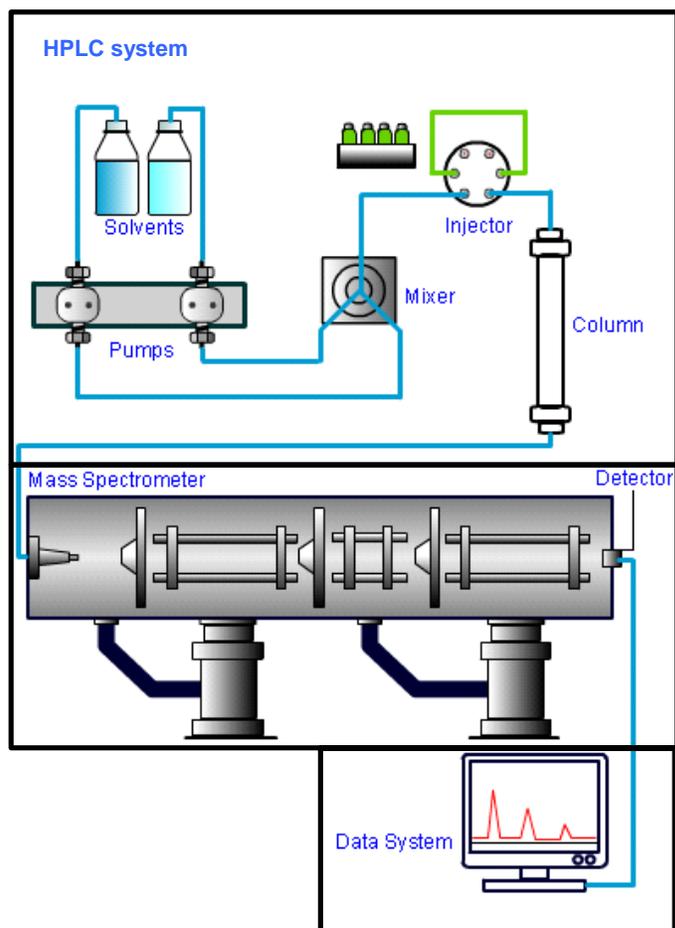
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## Definitions

LC/MS is a hyphenated technique, which combines the separating power of High Performance Liquid Chromatography (HPLC), with the detection power of mass spectrometry.

Mass Spectrometry is a wide-ranging analytical technique, which involves the production and subsequent separation and identification of charged species.

The associated acronym, LCMS (Liquid Chromatography-Mass Spectrometry) covers a broad range of application areas. This module will explore the instrument acquisition methods used, and examine the type of data that can be produced from such systems.



LC/MS diagram

For more information about HPLC you can refer to the HPLC Channel.<sup>[1]</sup>

## Instrument Fundamentals

The mass spectrometer is an instrument designed to separate gas phase ions according to their  $m/z$  (mass to charge ratio) value.

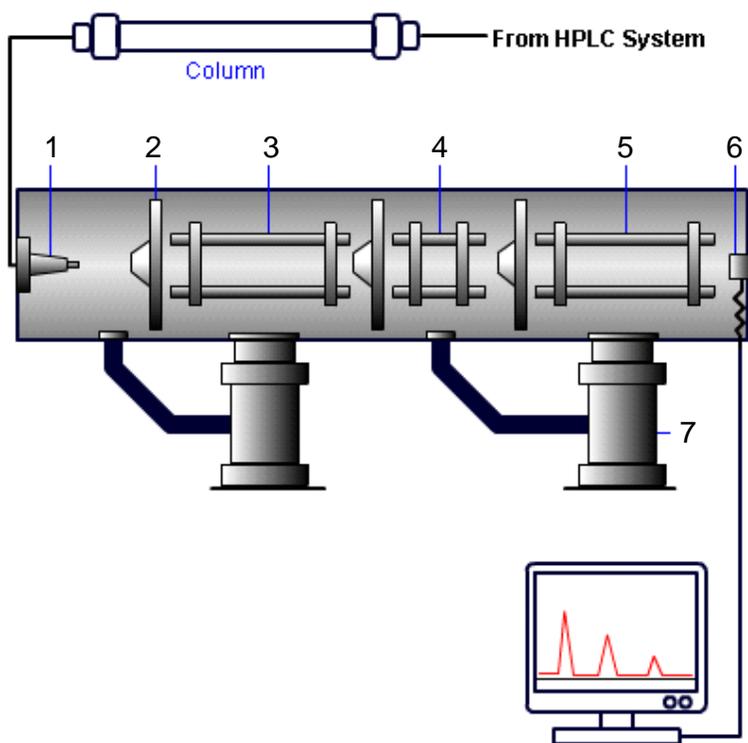
Mass spectrometry involves the separation of charged species which are produced by a variety of ionisation methods in LC-MS. These include:

- Electrospray Ionisation (EI)
- Atmospheric Pressure Chemical Ionisation (APCI)

In all cases the charged species are produced as gas phase ions under atmospheric pressure conditions.

The separation of the gas phase ions is achieved within the mass spectrometer using electrical and/or magnetic fields to differentiate ions.

In addition to the analyser, the mass spectrometer also includes an atmospheric ionisation chamber, a vacuum system and a detector.



Main elements of an LC/MS equipment

Where:

- 1. Ion source:** The HPLC eluent is sprayed into the atmospheric pressure region
- 2. Skimmer Cone:** A cone with a sampling orifice of reduced diameter to preferentially sample gas phase ions and reduce the gas load entering the vacuum system of the mass analyser device.
- 3. Quadrupole:** Device that uses electric fields in order to separate ions according to their mass to charge ratio ( $m/z$ ) as they pass along the central axis of four parallel equidistant rods.
- 4. Collision Cell:** Ions emerging from the first mass analyser are accelerated using a potential difference and collide with neutral gas molecules such as  $H_2$ ,  $N_2$  or Ar, causing analyte fragmentation.
- 6. Detector:** Once produced and separated, the ions need to be detected and transformed into a usable signal. Electron multiplier, Dynode, Photodiode, and Multi Channel Plate (MCP) ion detection systems are widely employed in most modern mass spectrometer systems.
- 7. Vacuum system:** Mass analysers require high levels of vacuum in order to operate in a predictable and efficient way. The vacuum systems of most modern LC-MS systems consist of two or more differentially pumped vacuum chambers, separated by baffles or orifice plates of varying design depending upon the instrument manufacturer.

## Process

There are several discrete stages in LC-MS analysis, typically these include:

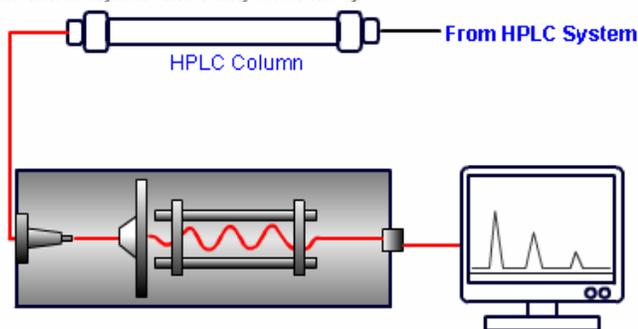
Separation of the sample components using an HPLC column where the analytes are differentially partitioned between the mobile phase (eluent) and the stationary phase (coated onto a support material and packed into the column). The mechanism of retention and separation will depend on the mode of chromatography but may include, Hydrophobic Interaction, Ion Exchange, Ion-Pair, Surface Localisation, etc.

The separated sample species are then sprayed into an Atmospheric Pressure Ion Source (API) where they are converted to ions in the gas phase and the majority of the eluent is pumped to waste.

The mass analyser is used to sort ions according to their mass to charge ratio. Most popular analyser types include Quadrupole (shown opposite), Time of Flight, Ion Trap and Magnetic Sector. The mass analyser may be used to isolate ions of specific mass to charge ratio or to 'scan' over all ion  $m/z$  values present.

The detector is used to 'count' the ions emergent from the mass analyser, and may also amplify the signal generated from each ion. Widely used detector types include: electron multiplier, dynode, photodiode and multi-channel plate.

All mass analysis and detection is carried out under high vacuum –established using a combination of foreline (roughing) and turbomolecular pumps.



LC/MS system

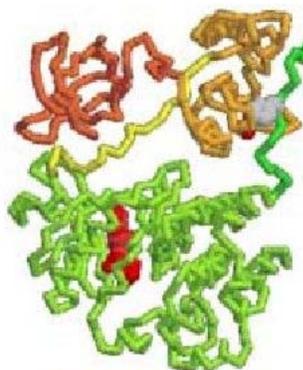
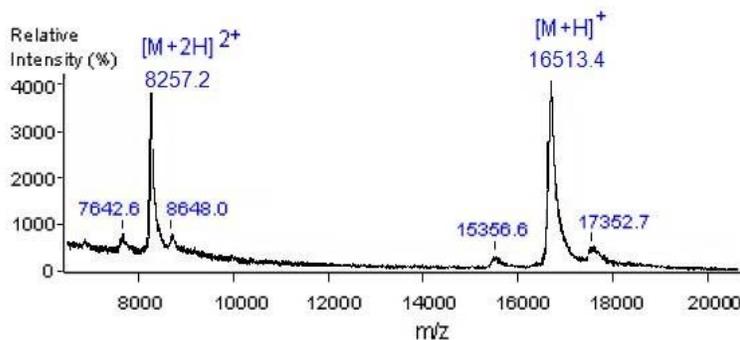


## Why and when to use LC/MS

The use of LC-MS in many application areas within analytical science continues to grow almost exponentially. Listed below are some pointers as to the applicability of both HPLC as a separative technique and MS as a means of detecting analyte species.

### HPLC separations

- For HPLC analysis the analyte must be soluble in the mobile phase
- HPLC can analyse samples over a wide polarity range including ionic samples
- HPLC has no real upper molecular weight limit and large proteins of many thousands of Daltons may be analysed. Solubility in the mobile phase may preclude the analysis of very large molecules. The ESI LC/MS spectrum of the *Locusta migratoria* cuticular protein<sup>[2]</sup> (16512.4 Da) is presented below
- HPLC samples are prepared in a solvent system that has the same or less organic solvent than the mobile phase and injection volumes of 1 to 50  $\mu$ l are common (1-10 $\mu$ g of analyte per 1g packing material)



*Locusta migratoria*

ESI LC/MS spectrum of *Locusta migratoria* cuticular protein (16512.4 Da)

### MS detection

- Allows specific compound identification (structural elucidation via spectral interpretation combined with elemental composition from accurate mass analysers is possible)
- Very sensitive (femto-gram amounts have been detected by certain mass analyzer types)
- Highly selective (certain analyzer and experiment combinations can lead to highly selective and sensitive analysis of a wide range of analytes)

## Ionisation

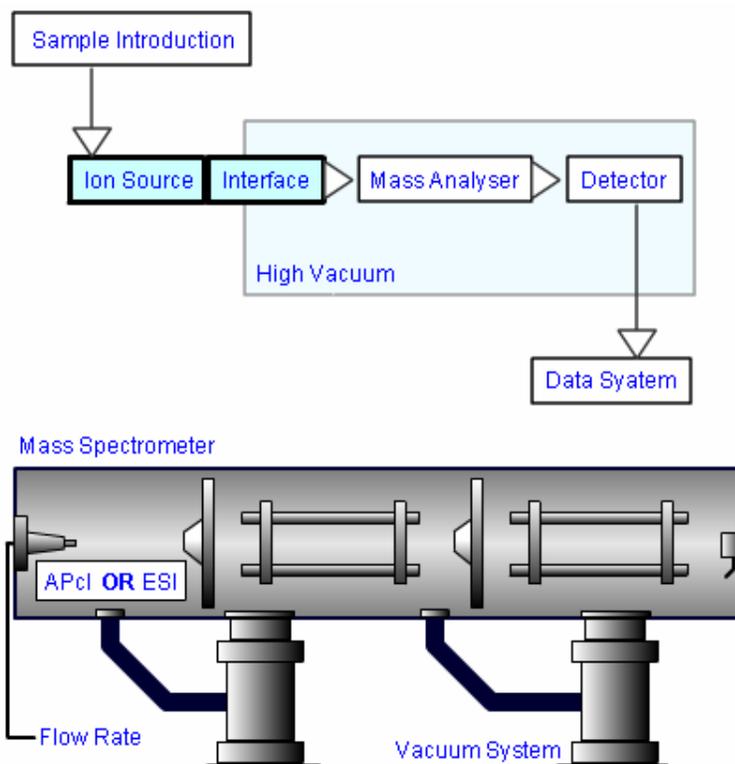
### Overview

Ionisation is the process whereby electrons are either removed or added to atoms or molecules to produce ions. In LC-MS charge may also be applied to the molecule via association with other charged molecules –for example a proton ( $H^+$ ).

Such ions are produced in LC/MS systems by the use of strong electric fields in the vapour or condensed phase. Interfaces whereby the sample is ionised or desolvated under atmospheric pressure conditions are termed **Atmospheric Pressure Ionisation (API)**.

The most common ionisation methods in LC-MS include:

- Electrospray Ionisation (ESI) –ionisation in the condensed phase
- Atmospheric Pressure Chemical Ionisation (APCI) –ionisation in the gas phase
- Atmospheric Pressure Photo Ionisation (APPI) –ionisation in the gas phase



Where the “Flow rate” label denotes the effluent (analyte plus eluent and additives) coming from the HPLC system.

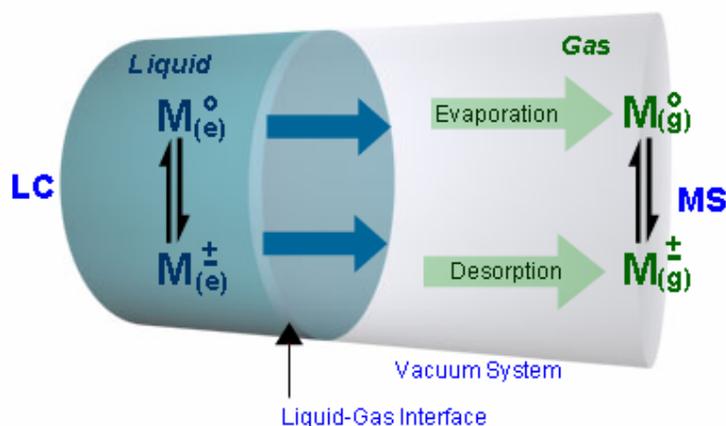
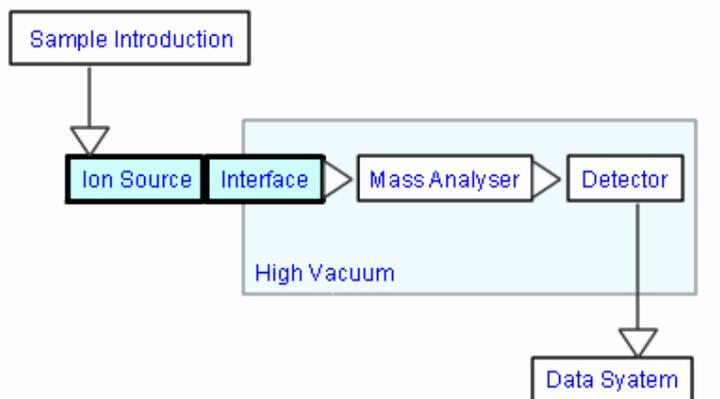
## Atmospheric Pressure Ionisation (API)

In Atmospheric Pressure Ionisation (API), the solvent elimination and ionisation steps are combined and take place in the ion source.

There are two main considerations common to all interface types:

1. Desolvation of the analyte molecule: The solvent molecules must be removed from the HPLC eluent to produce gas phase analyte ions.
2. Charging of the Analyte Molecule: Ions must be formed in order to transmit the analyte (or analyte derivatives), from the interface into the mass spectrometer where they will be filtered from other masses and subsequently detected.

The two main modes of ionisation used in API LC/MS are Electrospray Ionisation (ESI) and Atmospheric Pressure Chemical Ionisation (APCI). The schematic diagram BELOW indicates the main processes used in API ionisation processes.



Ion production process

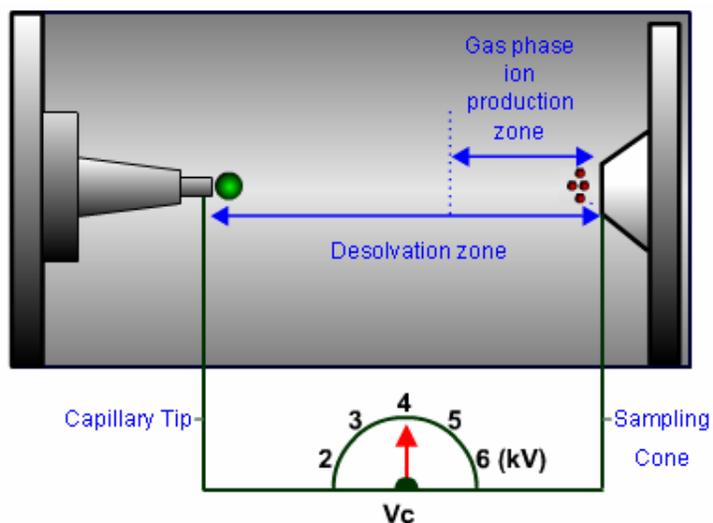
In Electrospray ionisation analyte ions are pre-formed in the mobile phase prior to entering the API interface. For Atmospheric Pressure Chemical Ionisation, ions are formed via charge transfer processes in the gas phase within the API interface.

## Electrospray Ionisation

Electrospray Ionisation (ESI) uses condensed phase (liquid) charge separation and ion evaporation techniques to produce vapour phase analyte ions. Primarily – analyte molecules of interest must be in the ionized form prior to spraying into the Electro spray interface in order to achieve a reasonable response. This dictates that analytes are ionised by manipulation of the HPLC eluent pH either before or after separation in the HPLC column.

In electrospray ionisation there are three important processes that occur in order to transfer sample ions from the HPLC eluent into the gas phase within the mass spectrometer. These processes are:

- Production of charged droplets at the capillary tip
- Desolvation of the droplets
- Production of gas phase ions from small / highly charged droplets



- Sprayed eluent droplet –desolvated using heated drying gas within the interface.
- Gas phase analyte molecule ( $[M+H]^+$ ) or small solvated droplet containing a higher number of rudimentary charges than the initial sprayed droplet.

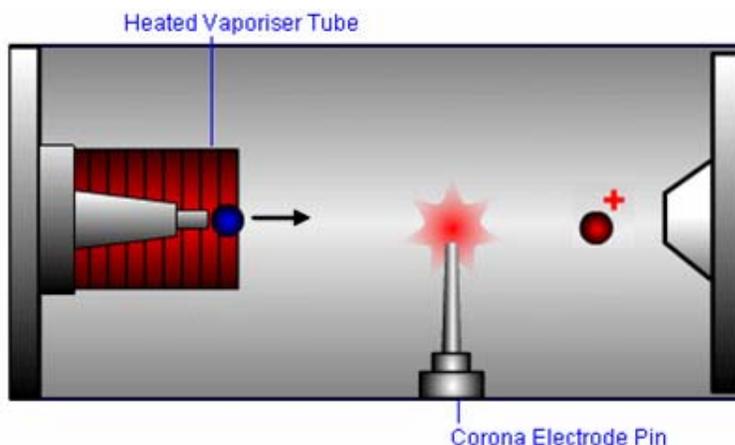
## Atmospheric Pressure Chemical Ionisation (APCI)

Atmospheric Pressure chemical Ionisation uses analyte desolvation and charge transfer reactions in the vapour phase to produce vapour phase analyte ions.

In APCI the eluent is introduced into the interface using a capillary of similar design to the ESI source. However, no potential is applied to the capillary but instead the liquid emerges from the capillary surrounded by a flow of inert, nebulising gas into a heated region.

The combination of nebulising gas and heat forms an aerosol that begins to rapidly evaporate. A pin is placed within the heated region that has a high potential applied to it and produces an electrical discharge that ionizes eluent molecules, these ionized molecules impart charge to the analyte molecules via charge transfer reactions or molecular association.

Both ESI and APCI are termed “soft” ionisation methods. This means that in the process of producing ions there is negligible energy transferred to the ion. As a consequence the ion formed does not fragment to small mass ions. The resultant mass spectrum therefore consists predominantly of **pseudomolecular** ions, either  $[M+H]^+$  or  $[M-H]^-$  or adduct ions like  $[M+Na]^+$ .



APCI process

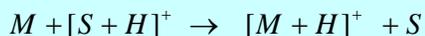
The ionised form of the molecule:



The  $M^+$  is known as the molecular ion. Note that molecular ions do not typically occur in LC/MS

### Pseudomolecular ion formation

If the analyte (M) has a larger proton affinity than the solvent (S), then:



If the analyte (M) has a lower proton affinity than the solvent (S), then:

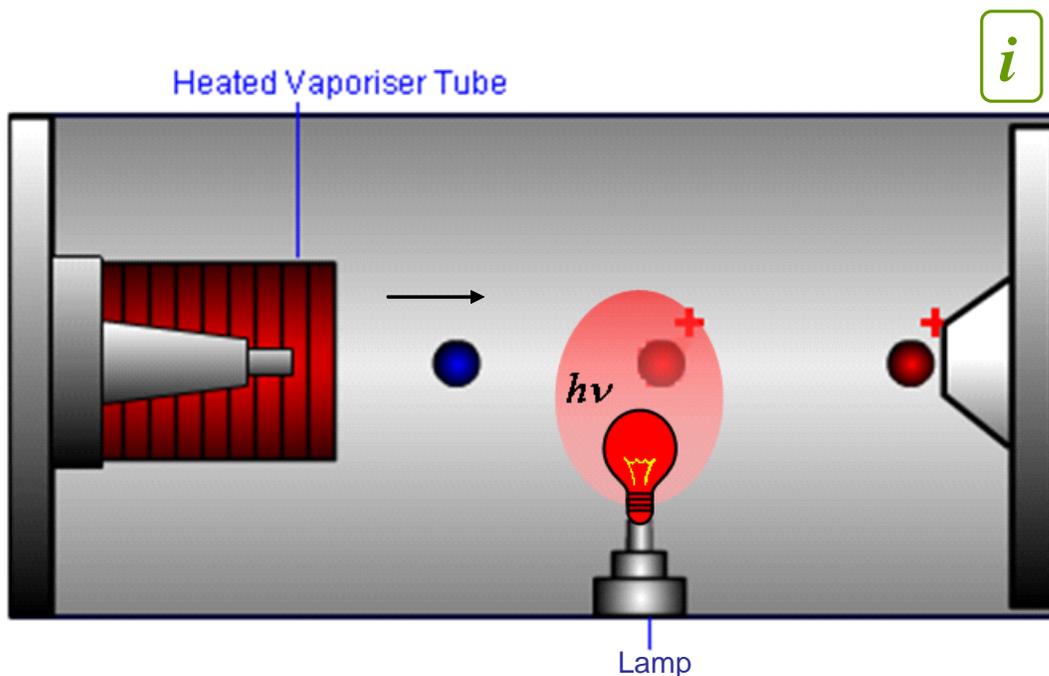


### Atmospheric Pressure Photo Ionisation (APPI)

APPI, is a complement to ESI and APCI and has been developed to broaden the range of ionizable analytes at atmospheric pressure.

APPI is important in the analysis of certain compounds that are not easily ionisable by ESI or APCI like low- and non-polar compounds (APPI has been used in the analysis of polycyclic aromatic hydrocarbons).

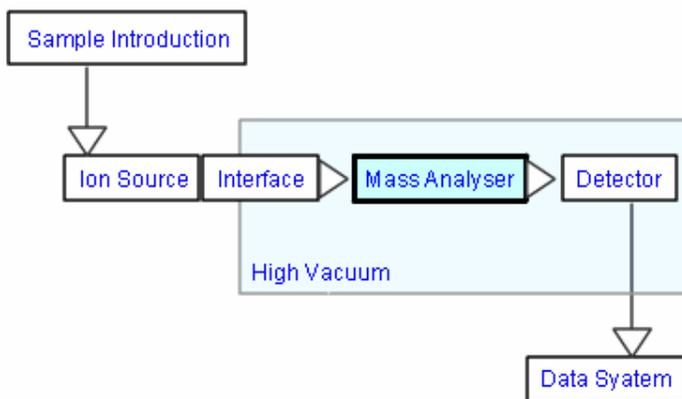
In APPI, the ionisation process is accomplished by exposing an aerosol of droplets to photoirradiation. A molecular radical ion is formed when the molecule absorbs a photon. This process is possible only when the irradiating photon (of energy  $h\nu$ ) exceeds the ionization potential (IP) of the molecule. APPI, allows the formation of charged species in positive or negative ion mode, these two mechanisms will be explained in detail in a subsequent chapter.



APPI process

## Mass analysers

In its simplest form the process of mass analysis in LC/MS involves the separation or filtration of analyte ions or fragments of analyte ions created in the Atmospheric Pressure Ionisation (API) interface or in the regions between the API interface and the high vacuum region of the mass analyser (products of collision-induced dissociation etc.).



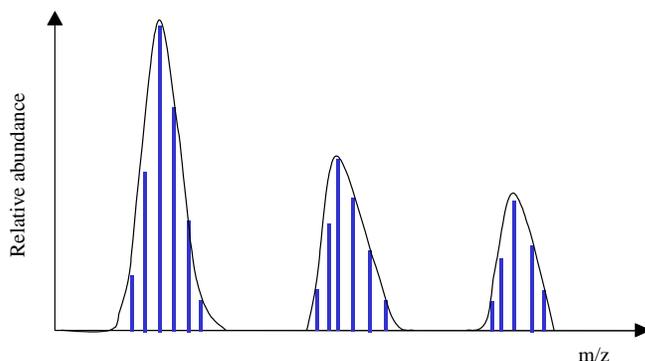
There are several very popular types of mass analyser associated with routine liquid chromatography mass-spectrometric analysis and all differ in the fundamental way in which they separate species on a mass-to-charge basis:

**Quadrupole and Ion Trap Mass analysers:** ions are filtered using electrostatic potentials applied to the elements of the mass analysers which are used to 'select' ions according to their mass to charge ratio –non-selected ions are ejected from the mass analysing device and are not detected.

**Time of Flight (TOF) mass analysers:** use differences in flight times of accelerated ions through an extended flight path to separate ions.

**Magnetic Sector Mass Analysers:** use magnetic fields to select ions by directing the beam of ions of interest towards the detector.

The analyte and fragment ions are plotted in terms of their mass-to-charge ratio ( $m/z$ ) against the abundance of each mass to yield a mass spectrum of the analyte as shown.

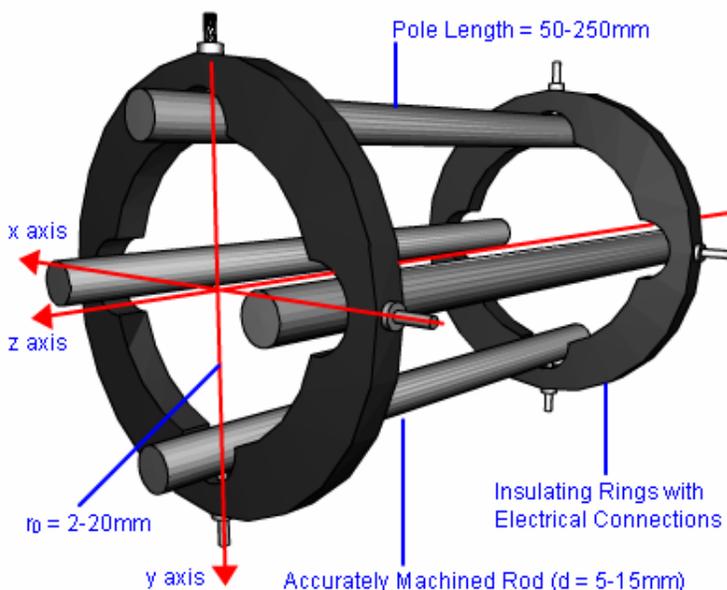


Mass spectrum

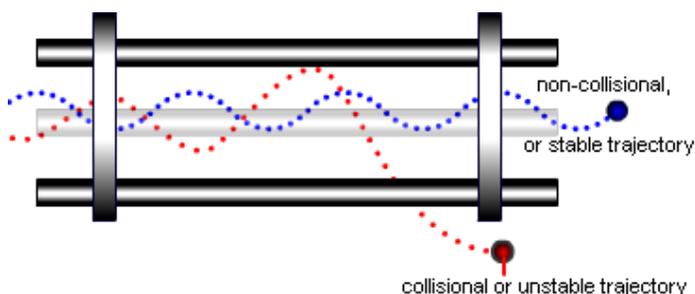
## Quadrupole

In quadrupole mass analysing devices, electric fields are used to separate ions according to their mass-to-charge ratio ( $m/z$ ) as they pass along the central axis of four parallel equidistant rods (or poles). Ion separation is performed by using controlled voltages applied to the mass analyser rods which impart an electrostatic field inside the analysing device.

As long as  $x$  and  $y$ , which determine the position of an ion from the centre of the rods, remains less than  $r_0$ , the ion will be able to pass through the quadrupole without touching the rods. This is known as a non-collisional or stable trajectory.



Where the ion is caused to oscillate with a trajectory whose amplitude exceeds  $r_0$  it will collide with a rod, and become discharged and subsequently pumped to waste. This is known as an unstable or collisional trajectory.

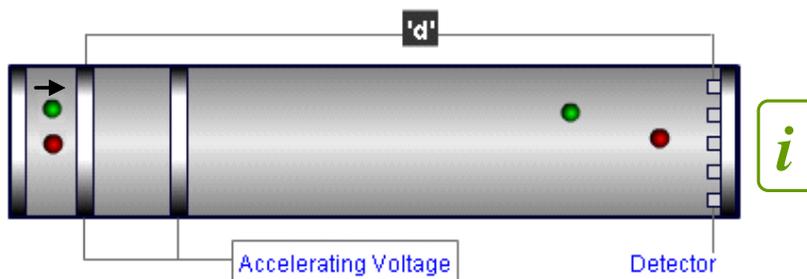


Quadrupole mass analyser	
Advantages	Disadvantages
<ul style="list-style-type: none"> <li>• Reproducibility</li> <li>• Low cost</li> </ul>	<ul style="list-style-type: none"> <li>• Low resolution</li> <li>• Mass discrimination. Peak height vs. mass response must be 'tuned'</li> </ul>

## Time-of-flight (TOF)

The basic principles of mass analysis using time-of-flight mass analysers are relatively straightforward in comparison to many of the other typical mass analysing devices.

Ions are extracted (or produced) in short bursts or packets within the ion source and subjected to an accelerating voltage. The ions then 'drift' or 'fly' down an evacuated tube of a set length ('d'). Once free from the region of accelerating voltage the speed at which the ions travel down the tube is dependant upon their mass (m) and charge (z). This mass analyser is useful as all ions are detected (almost) simultaneously. Scanning the mass range of all ions is very rapid and as such the inherent sensitivity of the instrument is increased.



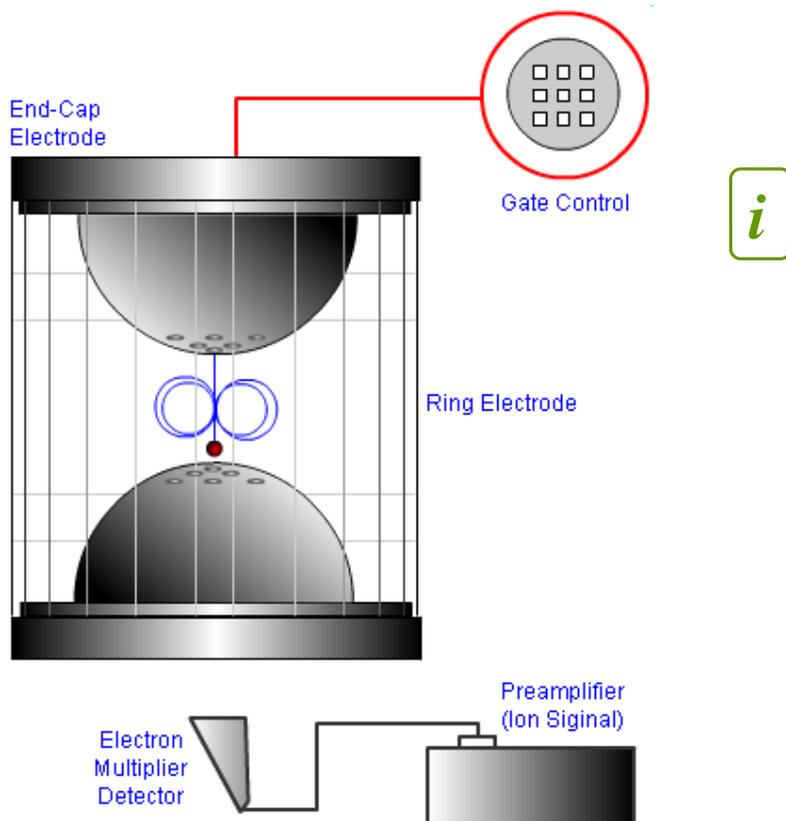
### Time-of-flight (TOF)

Advantages	Disadvantages
<ul style="list-style-type: none"> <li>• High ion transmission</li> <li>• Highest practical mass range of all MS analyzers</li> <li>• Detection limit</li> </ul>	<ul style="list-style-type: none"> <li>• Fast digitizers used in TOF can have limited dynamic range</li> </ul>

## Ion Trap Mass Analyser

Ion trap mass spectrometers work on the basis of storing ions in a "trap", and manipulating the ions by using applied DC and RF fields. The amplitude of the applied voltages enables the analyser to trap ions of specified mass to charge ratio within the analysing device. Non-selected ions are given a trajectory by the electrostatic field that causes them to exit the trap. By filling the trap with an inert gas fragmentation of selected ions is possible. This is useful when structural information is required.

The system has some unique capabilities, including being able to perform, multiple product ion scans with very good sensitivity ( $MS^n$ ). It should be noted that the spectra acquired with an ion trap mass analyser may be significantly different to those acquired from a triple quadrupole system due to the different collision regimes within the systems (collision energy/gas).



Ion trap mass analyser

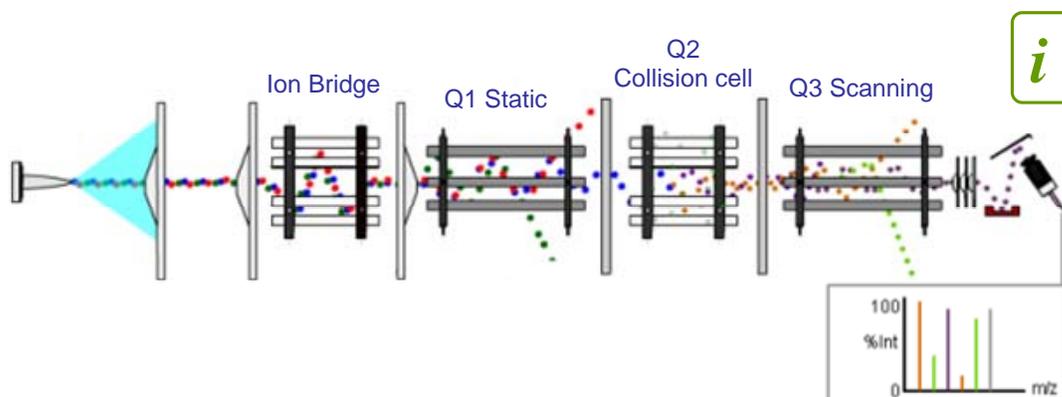
Ion trap mass analyser	
Advantages	Disadvantages
<ul style="list-style-type: none"> <li>• High sensitivity</li> <li>• Multiple Product Ion scan capability (MS)<sup>n</sup></li> <li>• High resolution</li> <li>• Good for DDA analyses</li> </ul>	<ul style="list-style-type: none"> <li>• Produces very unusual spectra if the ions are stored in the trap too long.</li> <li>• Easily saturated</li> <li>• Poor for low mass work (below 100 Da)</li> <li>• Poor dynamic range (except the most modern devices) and hence may have limited quantitative use</li> </ul>

## Tandem mass spectrometry (MS/MS)

MS/MS is the combination of two or more MS experiments. The aim is either to get structural information by fragmenting the ions isolated during the first experiment, and/or to achieve better selectivity and sensitivity for quantitative analysis by selecting representative ion transitions using both the first and second analysers.

MS/MS analysis can be achieved either by coupling multiple analysers (of the same or different kind) or, with an ion trap and carrying out successive fragmentations of trapped ions.

MS<sup>n</sup> (should read MS to the n) is an acronym that refers to multiple ion production and filtering within a single instrument. Most common instruments use a combination of quadrupoles (as shown below) with a collision cell (usually a multi-pole device) between the analyzing devices in which the emergent ions from the first analyzer are fragmented prior to secondary mass filtering. Other combinations of mass analysing devices such as quadrupoles and time of flight, or quadrupoles with magnetic sector instruments are possible.



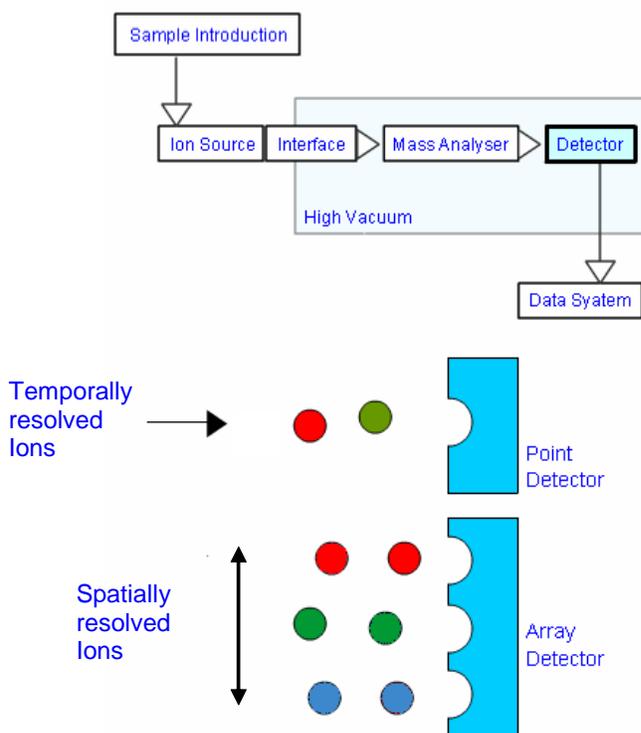
Tandem mass spectrometry

## Detectors

Once the ions have passed the mass analyser they have to be detected and transformed into a usable signal.<sup>[3]</sup> The detector is an important element of the mass spectrometer that generates a signal from incident ions by either generating secondary electrons, which are further amplified, or by inducing a current (generated by moving charges). Ion detector systems fall into two main classes:

**Point detectors:** ions are not spatially resolved and sequentially impinge upon a detector situated at a single point within the spectrometer geometry.

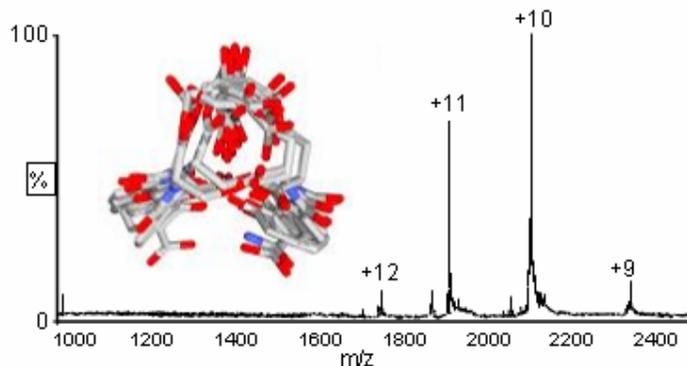
**Array detectors:** ions are spatially resolved and all ions arrive simultaneously (or near simultaneously) and are recorded along a plane using a bank of detectors.



## Applications

To give a full list with the applications of LC/MS is simply impossible, its flexibility makes it attractive in a lot of different fields. Some interesting applications are listed below.

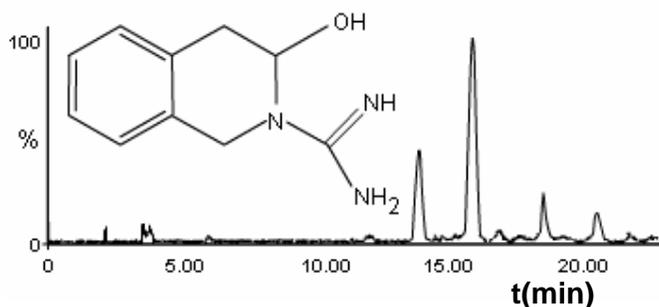
**Proteomics:**<sup>[4]</sup> systematic analysis of proteins.



Negative ESI-MS spectrum of holo-siderocalin ( $21195 \pm 1$  Da)

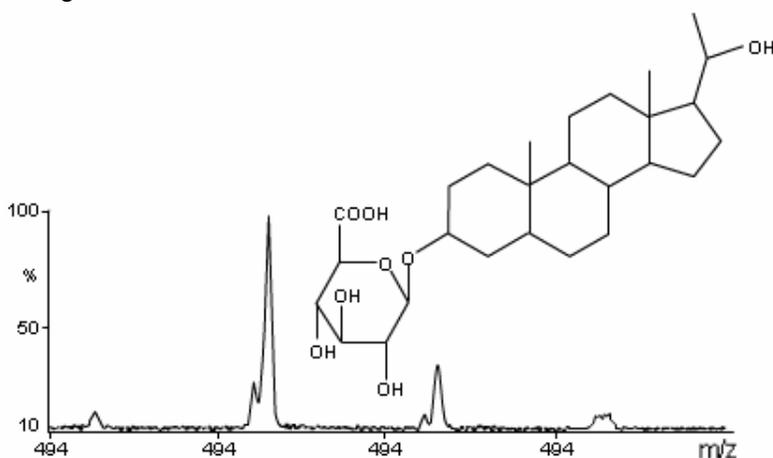
Although mass analyzing devices have a practical upper molecular weight limit of around 5KDa (5000Da), Electrospray MS is capable of imparting multiple charges onto the analyte ion. As analysing devices select ions based on mass to charge ratio (m/z), this protein is capable of holding 12 charges (12+), effectively lowering the 'apparent' molecular weight to  $21195/12 = 1766$

**Pharmacokinetics:**<sup>[5]</sup> study of the absorption, distribution, metabolism and excretion of drugs.



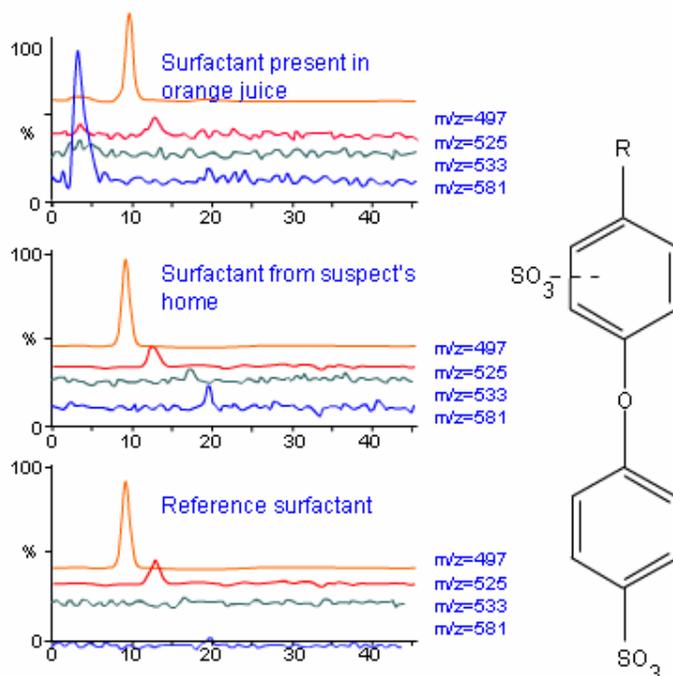
LCMS has been used to analyse and quantify drugs and metabolites contained in biological samples. Debrisoquine and its hydroxylated metabolites were analysed from urine extract. The mass spectrometer (model Q-TOF; Micromass, Manchester, UK) was equipped with an electrospray ion source operated at 70°C.

**Bioanalysis:**<sup>[6]</sup> which involves the quantification and identification of metabolites in biological fluids.



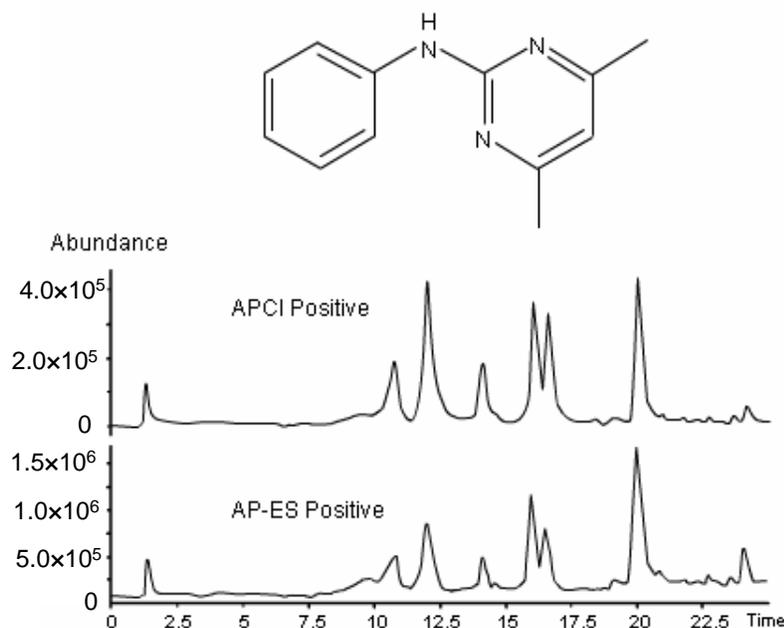
Analysis of derived sialyl oligosaccharides by negative-ion ESI have been recently performed. The mass spectrum of the weakly acidic lipid fraction extracted from the urine of a female coffee drinker suffering from intrahepatic cholestasis of pregnancy.

**Forensic science:**<sup>[7]</sup> such as in drug analysis, toxicology, trace analysis, etc.



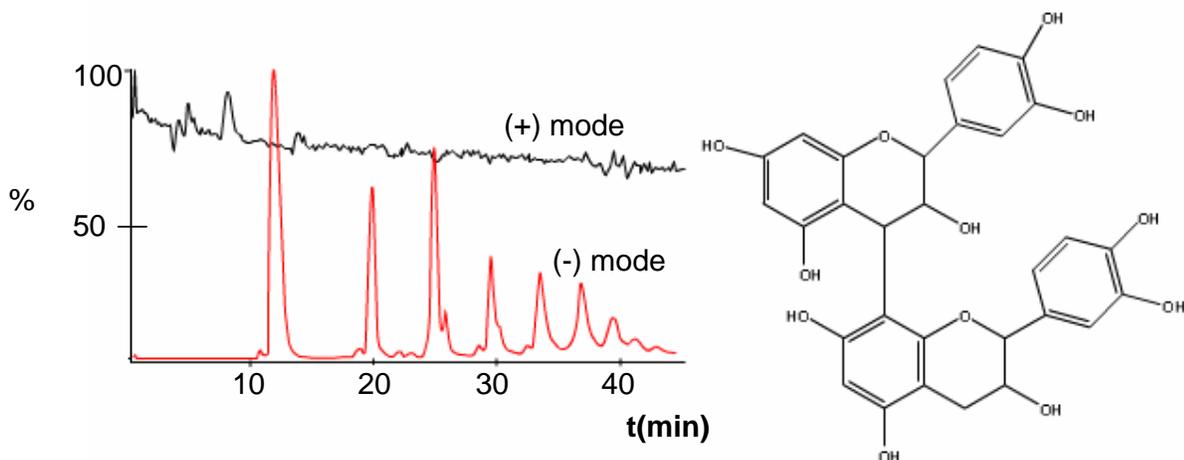
Many cases of poisoning with alkyl-diphenylether sulphonic acids (poisonous detergents) mixed into food and/or beverages have been reported. In the application the lethal mixture present in orange juice is compared against the suspect's home sample and a reference mixture.

**Agrochemistry:**<sup>[8]</sup> Involving the determination of compounds that are present in fertilizers, pesticides, etc.



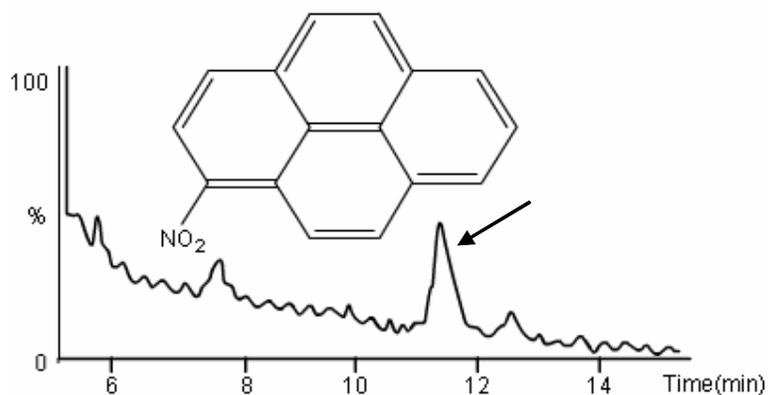
LC-MS chromatograms in APCI and AP-ES modes corresponding to a solution of Pyrimethanil (pesticide) contained in a water sample.

**Food analysis:**<sup>[9]</sup> protein characterization, and determination of organic food stuff (enzymes, flavonoids, etc).



Total ion chromatograms of cocoa procyanidins obtained after postcolumn addition of 10mM ammonium acetate in both positive and negative electrospray ionization modes.

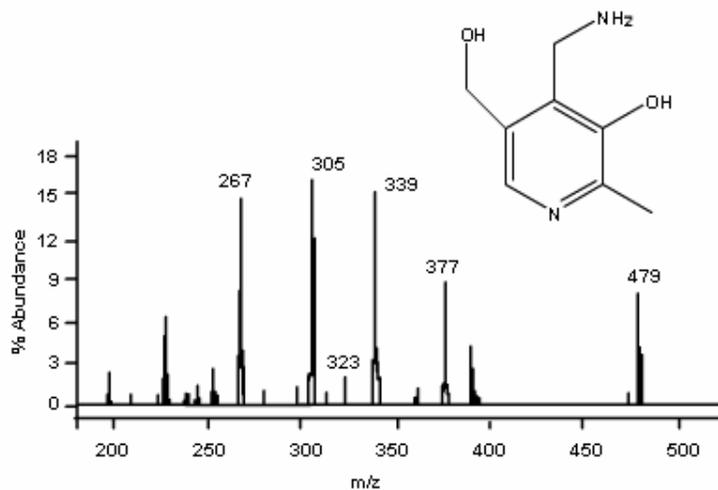
**Petrochemistry:**<sup>[10]</sup> analysis of oil-derived molecules (like hydrocarbons) and contaminants.



Analysis of nitropyrene 247 Da present in urban soil. Negative ion chemical ionisation.

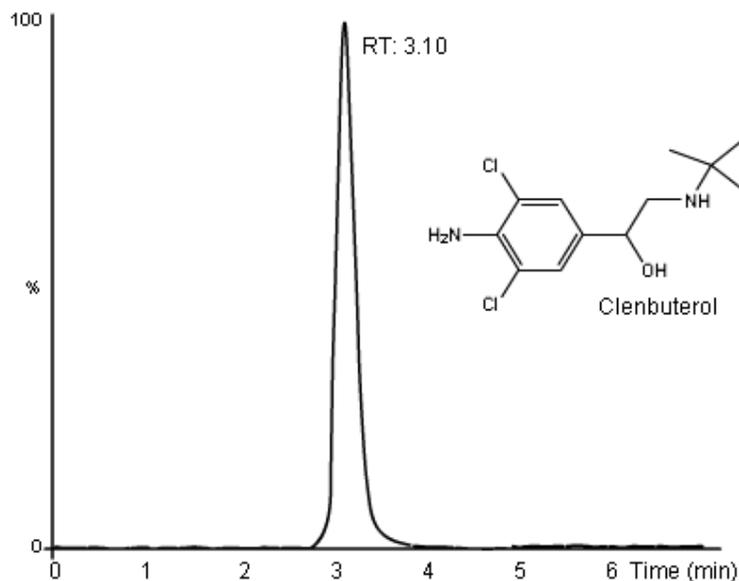
1-Nitropyrene and its isomers (environmental contaminants) are well known for its carcinogenic properties. Considerable amounts of these compounds have been detected in diesel engine exhausts.

**Cosmetics.<sup>[11]</sup>**



Piridoxamine is used in cosmetics to avoid proteins modification. The figure presents the chromatogram (direct injection positive ESI LCMS) of an aqueous solution of piridoxamine.

**Doping.<sup>[12]</sup>**

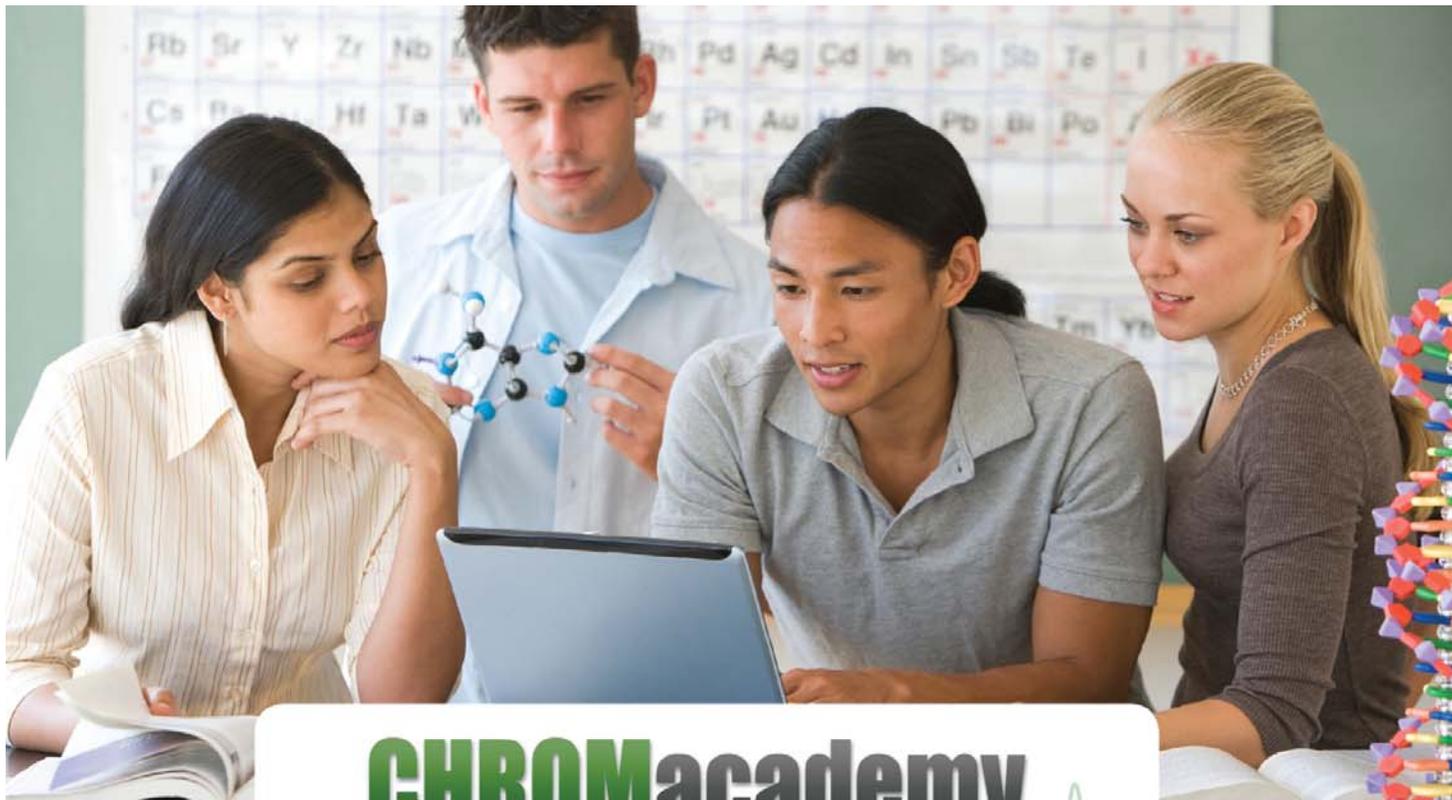


Analysis of clenbuterol from a urine sample by (+) APCI

In order to increase their performance, clenbuterol has been illegally used for some competitors.

## References

1. "The Theory of HPLC" and "Instrumentation of HPLC" from the HPLC Channel.
2. Dário Eluan Kalume, Sylvie Kieffer, Kate Rafn, Lene Skou, Svend Olav Andersen, Peter Roepstorff. "Sequence determination of three cuticular proteins and isoforms from the migratory locust, *Locusta migratoria*, using a combination of Edman degradation and mass spectrometric techniques." *Biochimica et Biophysica Acta* 1645, (2003), 152– 163.
3. De Hoffmann, J. Charette, and V. Stroobant. "Mass Spectrometry. Principles and Applications." John Wiley and Sons. Pp 91-97, 1996
4. Catalin E. Doneanu, Roland K. Strong, and William N. Howald. "Characterization of a Noncovalent Lipocalin Complex by Liquid Chromatography/Electrospray Ionization Mass Spectrometry" *Journal of Biomolecular Techniques*, volume 15, issue 3, September 2004. Pp 208 - 212
5. Birgit Eiermann, Per Olof Edlund, Agneta Tjernberg, Per Dalén, Marja-Liisa Dahl, and Leif Bertilsson. "1- and 3-hydroxylations, in addition to 4-hydroxylation, of debrisoquine are catalyzed by cytochrome p450 2d6 in humans" *drug metabolism and disposition*. Copyright © 1998 by the American Society for Pharmacology and Experimental Therapeutics. Pp 1096 – 1101
6. William J. Griffiths, Andreas P. Jonsson, Suya Liu, Dilip K. RAI and Yuqin Wang. "Electrospray and tandem mass spectrometry in biochemistry" *Biochem. J.* (2001) 355, 545±561 (Printed in Great Britain).
7. Mayumi Nishikawa, Munehiro Katagi, Akihiro Miki, and Hitoshi Tsuchihashi. "Forensic Toxicological Determination of Surfactant by Liquid Chromatography/Electrospray Ionisation Mass Spectrometry and Liquid Chromatography/Electrospray Ionisation Tandem Mass Spectrometry" *Journal of Health Science* 49(2) 138 – 148 (2003).
8. A. Agüera and A.R. Fernández-Alba. "GC-MS and LC-MS evaluation of pesticide degradation products generated through advanced oxidation processes: An overview" *EDP Sciences, Wiley-VCH. Analisis Magazine*, 1998, 26, N° 6. Pp M123 – M130.
9. Yun-leong Hong, Diane M. Barrett, and Alyson E. Mitchell. "Liquid Chromatography/Mass Spectrometry Investigation of the Impact of Thermal Processing and Storage on Peach Procyanidins" *Journal of Agricultural Food Chemistry* 2004, 52, 2366-2371.
10. Hiroshi Moriwaki, Kunihiro Funasaka and Michiko Uebori. "Direct Determination of NO<sub>2</sub>-Substituted Pyrenes by Liquid Chromatography-Atmospheric Pressure Chemical Ionization-Mass Spectrometry" *Analytical Sciences* December 2000, Vol 16, Pp 1247 - 1248. © The Japan Society for Analytical Chemistry.
11. Joelle M. Onorato, Alicia J. Jenkins, Suzanne R. Thorpe, and John W. Baynes. "Pyridoxamine, an Inhibitor of Advanced Glycation Reactions, Also Inhibits Advanced Lipoxidation Reactions. Mechanism of Action of Pyridoxamine" *The Journal of Biological Chemistry*. Vol. 275, No. 28, Issue of July 14, pp. 21177–21184, 2000.
12. Mark Churchill and Mark Harrison. "High Resolution Separation with Accurate Mass Determination of Two Co-Eluting Controlled Drugs of Abuse with Isobaric Mass" *Chromatography and Mass Spectrometry, Application Note 329*. Thermo Electron Corporation.



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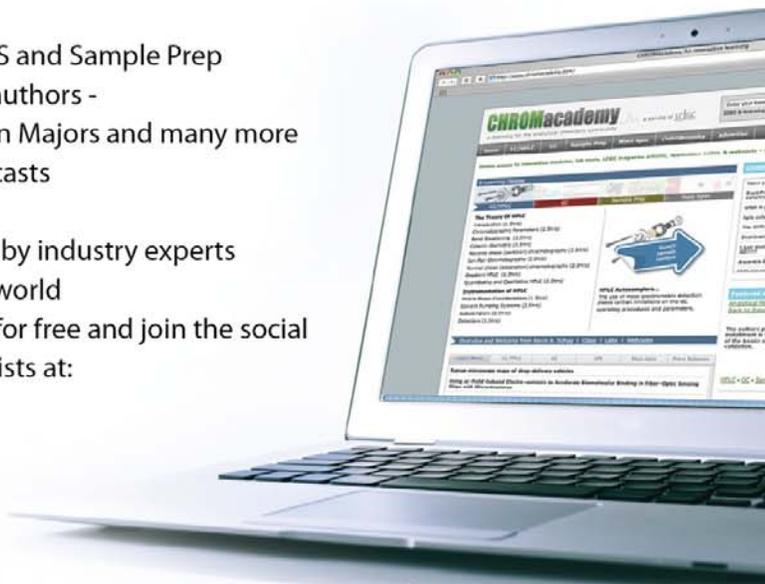
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