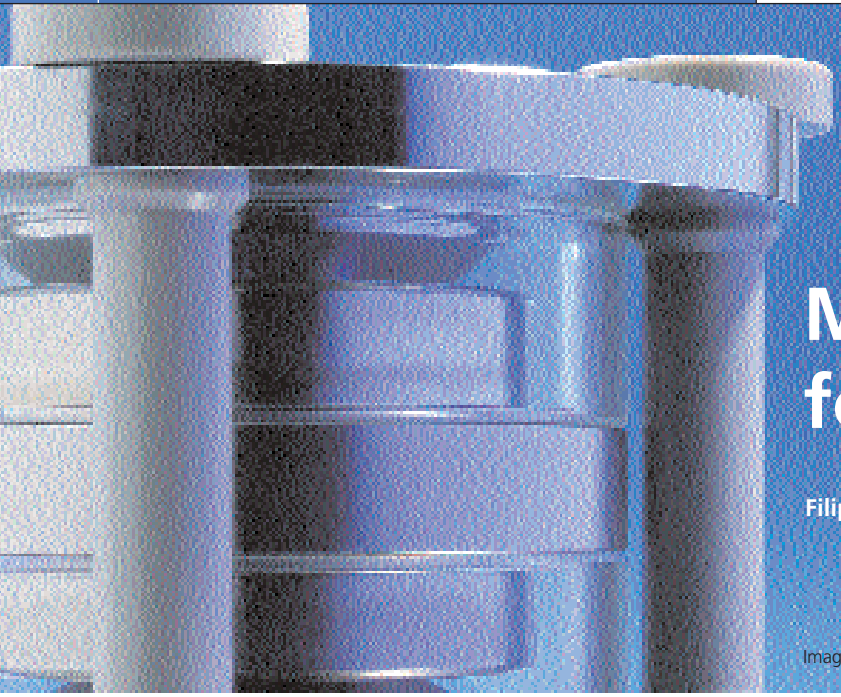

# Mass Analysers for LC–MS

**Filip Lemière,** Dept of Chemistry, University of Antwerp, Belgium.

Image courtesy of Bruker Daltonik GmbH

## Introduction

The major technical challenge in LC–MS is interfacing the chromatographic and spectrometric components (see "Interfaces for LC–MS" section). This observation implies that any mass spectrometer can potentially be coupled to an LC or other separation system. Understanding the different working principles and technical properties of different MS instruments gives an insight into the technical possibilities and limitations that must be acknowledged when coupling with LC. The characteristics of different MS instruments also determine their applications in the different LC–MS research fields, and the requirements imposed by the analytical question determine the type of MS to couple to the LC system. Ultimately, the only limit to the coupling of LC to MS is the imagination of the analyst setting up the hyphenated experiment.

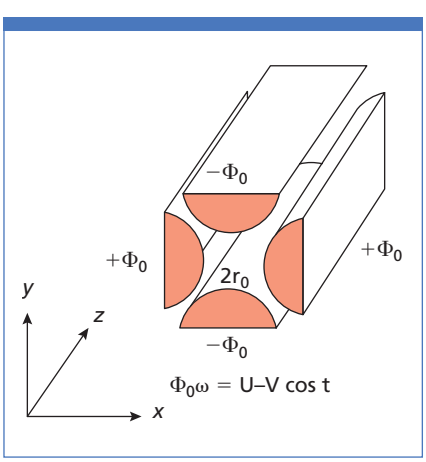


**Figure 1:** *Layout of the four rods used in a quadrupole MS system.*

This section will give a short overview of the most common MS instruments and their working principles. However, this is not intended to be an exhaustive presentation of all possible techniques and variants. For more details we refer the reader to dedicated literature on MS (e.g., references 1–3).

## Quadrupole Systems

**Introduction:** Both quadrupole and ion trap systems use RF and DC voltages for the separation of ions. These MS systems are probably the most widespread mass spectrometers because of their relatively low price and ease of operation.

**Working principle:** In a quadrupole mass spectrometer (4) the RF and DC potentials are applied to four rods arranged in a square array (Figure 1). Ideally, rods with hyperbolic cross-sections should be used; however, in practice more economical cylindrical rods give results that are more than sufficient for most applications. Opposite rods are electrically connected. Application of voltages on these pairs of rods creates a hyperbolic field within the rods given by

$$\Phi = (V_{dc} + V_{rf} \cos(\omega.t) \frac{x^2 - y^2}{r_0} \qquad [1]$$

resulting in a force on the ion at position ($x$, $y$) proportional to the distance from the axis ($z$) of the quadrupole. Whether ions will succeed in passing through this mass filter depends on their $m/z$ value.

When $V_{rf} > V_{dc}$ the low mass ions are lost in, for example, the $x$-direction because they readily follow the rapidly fluctuating RF potential and collide with the rods, such that only the heavier ions pass through (quadrupole working as high pass filter). At the same time in the $y$ direction the heavy atoms are lost because of their inertia towards the fast altering RF field (i.e., quadrupole working as a low pass filter). The combination of both yields a stability window defined by the frequency ($\omega$) of $V_{rf}$ and the ratio $V_{rf}/V_{dc}$.

This is described in the equations of motion. For each axis ($u = x$, $y$ or $z$) we know that

$$m \frac{d^2u}{dt^2} = ze \frac{d\Phi}{du} = \frac{ze\Phi_0 u}{r_0^2} \qquad [2]$$

where $r_0$ is the distance between the rods.

When the known function of the electric field is substituted in the equations of motion we obtain the so called Mathieu equations.

$$\frac{d^2x}{dt^2} + \frac{e}{mr_0}(V_{dc} + V_{rf} \cos \omega t)\, x = 0 \qquad [3]$$

$$\frac{d^2y}{dt^2} - \frac{e}{mr_0}(V_{dc} + V_{rf} \cos \omega t)\, y = 0 \qquad [4]$$

For a given quadrupole instrument, $r_0$ is a fixed value and the frequency of the RF potential ($\omega$) is constant. These equations are solved numerically and plotted (Figure 2) using two parameters:

$$a = \frac{4eV_{dc}}{m\omega^2 r_o^2} \qquad [5]$$

$$q = \frac{2eV_{rf}}{m\omega^2 r_0^2} \qquad [6]$$

All areas in the plot where $x$ and $y$ are smaller than $r_0$, (i.e., the ion stays within the rods), represent a stable path through the mass spectrometer. The two

experimental variables are the potentials $V_{rf}$ and $V_{dc}$. In most experiments the ratio $V_{dc}/V_{rf}$ is constant. This defines a line in the a-q plot (see Figure 2). All ions with an *m/z* value that fits with points on this line within a stable region will move through the mass spectrometer. The intersection with the edges of the stability region defines the window of *m/z* values that are stable. Generally, the ratio $V_{dc}/V_{rf}$ is chosen such that a 1 Da window is selected resulting in unit resolution over the entire mass range. Modern quadrupole instruments have an upper mass limit between 3000 and 4000 Da. The separation of ions in a quadrupole does not depend on the kinetic energy of the ions entering the analyser or the initial spatial distribution of the ions. However, the residence time between the rods has to be long enough to allow for the RF potential to influence them; that is, the residence time must be longer than the period of the RF voltage. This poses limits on the kinetic energy of the ions leaving the ion source/LC interface and entering the quadrupole to approximately 100 eV. Also, collision energies used in tandem MS instruments employing quadrupoles are limited to approximately 100 eV for the same reason (see *Tandem Instruments*).

**Pros**

- energy and spatial distribution of ions produced in the source and entering the mass analyser are not critical (important for coupling to LC because LC–MS interfaces generally produce ions with a relatively wide energy and spatial distribution).
- simple scanning method $V_{dc}/V_{rf} = C^{te}$
- low potentials allow relatively high pressures and simple vacuum systems
- low cost
- easy to couple multiple quadrupoles to one another or other MS analysers (see *Tandem Instruments*).

**Cons**

- low-resolution systems, typically 1 Da
- mass range limited to approximately *m/z* 4000.

**Applications**

- general low-resolution MS instrument
- molecular weight determination.

## Ion Traps

**Introduction:** Together with the quadrupole instruments, ion traps are among the most widespread mass analysers. Their small size and inherent tandem MS capabilities make quadrupole ion traps or "quistors" ideal for benchtop applications and $MS^n$ experiments.

**Working principle:** The fundamental working principles of the ion trap (5) are the same as described for the linear quadrupole (see *Quadrupole Systems*). One can describe a quistor as a linear quadrupole bent to a closed loop. The outer rod forms a ring, and the inner rod is reduced to a mathematical point in the centre of the trap. The top and bottom rods form end-caps above and below the ring electrode (see Figure 3). These end-caps are perforated to allow the injection and detection of ions. Within this three-dimentional quadrupole the ions travel on a Lissajous figure–shaped path. Similar equations of motion as used for the quadrupole are used to describe the behaviour of ions in a quistor, except that $r$ and $z$ are used instead of $x$ and $y$.

Ions of different mass are stored together in the trap and released one at a time by scanning the applied voltages. Ions of selected mass can be ejected through the end-caps and detected by applying an RF voltage with a frequency corresponding to the characteristic frequency of the ion moving through the ion trap or by scanning the amplitude of the applied RF voltage.

By allowing a $10^{-3}$ Torr pressure of gas (often He), the motion of ions in the trap is dampened and the ions move closely around the centre of the trap. This avoids ion loss by collisions with the electrodes and improves the resolution because all ions remain in a small space (limited spatial distribution), and field imperfection is minimal at the centre of the trap. Without the damping gas, ions have a tendency to increase the radius of their motion because of the electrostatic repulsion of equally charged particles. As He or $H_2$ is used in the trap this makes ion traps natural detectors for coupling with gas chromatography (GC) in which the ions are formed by electron impact (EI) with a short pulse of electrons in the ion trap. When applied in LC–MS, the ions are formed outside the ion trap and then transported into the cell for mass analysis.

*"Together with the quadrupole instruments, ion traps are among the most widespread mass analysers."*

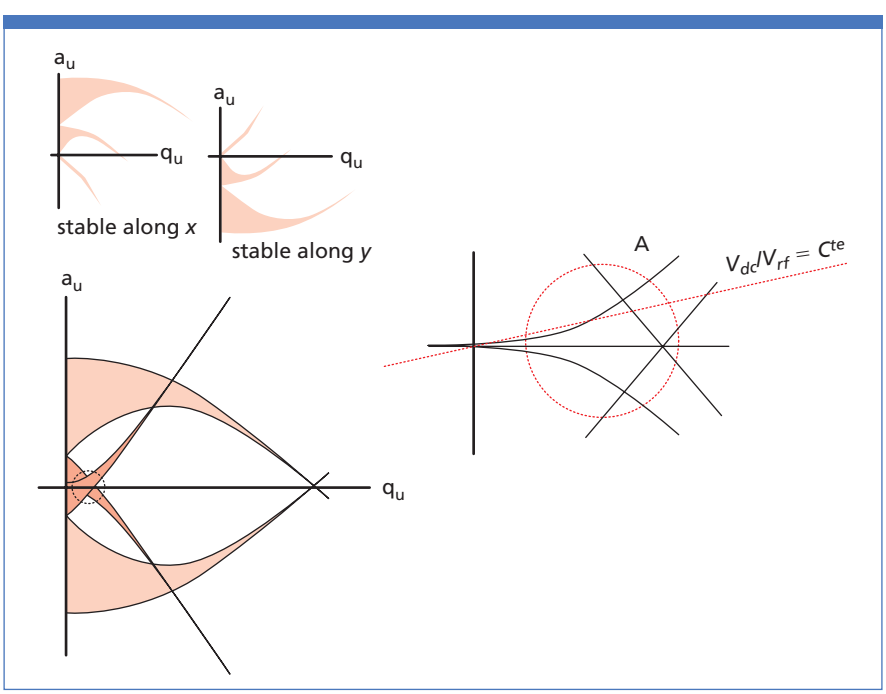


**Figure 2:** *Stability diagrams of a linear quadrupole.*

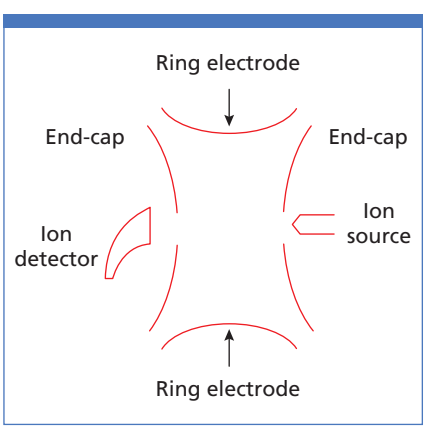


**Figure 3:** *Overview of a quadrupole ion trap.*

**Pros**

- energy and spatial distribution of ions produced in the source and entering the mass analyser are not critical (important for coupling with LC because LC–MS interfaces generally produce ions with a relatively wide energy and spatial distribution)
- low potentials allow relatively high pressures and simple vacuum systems
- low cost
- small instruments (most systems fit on benchtop)
- inherent tandem MS capabilities (see *Tandem Instruments*).

**Cons**

- low-resolution systems, typically 1 Da; however, using specific scan methods higher resolutions can be obtained.

**Applications**

- general low-resolution MS instrument
- molecular weight determination.

***"Current state-of-the-art electronics use detectors with GHz sampling rates."***

### Time-of-Flight Analysers

**Introduction:** The initial idea of measuring the mass of an ion by its flight time was put forward by Stephens in 1946 (6, 7). The very simple concept makes time-of-flight (TOF) an attractive method, but high demands on electronics and limited resolution constrained its applications. The technique was improved with the introduction of an electrostatic reflector (8). Enhanced ion optics to focus the ion beam from the LC–MS source into a pusher made coupling with LC available, while the development of fast electronics further improved the results of these instruments. When the ion-beam focusing lenses are replaced by a mass spectrometer (quadrupoles, sectors), TOF analysers are readily incorporated in tandem MS instruments.

**Working principle:** The basic working principle of a TOF mass spectrometer involves measuring the flight time of an ion through the mass spectrometer (9, 10). Because the dimensions of the mass spectrometer are known, as is the energy of the ion, a straightforward calculation is required to obtain the *m/z* value of the ion.

We give the ion a well-defined kinetic energy by acceleration of the ion in a known electrostatic field.

The kinetic energy is given by

$$\frac{mv^2}{2} = qV \qquad [7]$$

Where the charge is:

$$q = ze \qquad [8]$$

The flight time of the ion through the mass spectrometer is

$$t = \frac{d}{v} \qquad [9]$$

Substituting $v$ yields

$$\frac{m}{z} = t^2 \left(\frac{2Ve}{d^2}\right) \qquad [10]$$

For a given setting of $V$ and a given apparatus ($d$ is fixed) the value between brackets is a constant. Measurement of the flight time, $t$, yields the *m/z* value.

As a defined start/stop signal is essential for the time measurement the ion input must be pulsed. This requires the introduction of some kind of pulsing device to convert the continuous flow of an LC system to a discrete series of ion packets. Often this is performed by focusing the ion beam orthogonal to the flight tube and using a pulsed electric field to push the ions into the TOF analyser. This complies both with the limitations on energy and spatial distribution. The ion beam has little or no radial velocity component and a well-focused beam orthogonal to the TOF analyser has a well-defined position.

The correctness of the time measurement immediately influences the accuracy of the mass assignment. Also the initial kinetic energy and spatial distribution of the ions is important. Variations of the kinetic energy and/or position of the ion when entering the acceleration region will influence the energy of the ions when starting their flight through the flight tube. These variations themselves will result in variations in the measured flight time and *m/z* value reducing the mass resolution of the system. As the energy and spatial distribution of most LC interfaces is not well defined, special measures must be taken in order to get good resolution using TOF instruments. The introduction of a reflector in the flight tube and the use of orthogonal injection techniques improved the mass spectrometric resolution by controlling or compensating for the initial energy spread and spatial distributions.

The electronics of the detector must be capable of recording the complete mass spectrum within the flight time of the ions (typically in the 1–100 µs range), with peak widths in the ns range. Current state-of-the-art electronics use detectors with GHz sampling rates. Even at these high speeds detectors are still a limiting factor on the resolution of TOF instruments (resolutions 10 000 to 20 000 are routinely obtainable), given that all other components and experimental parameters are tuned to an optimum. Note that no instrumental parameter limits the detection of high-mass ions. Theoretically the highest detectable mass is only determined by the observed flight time. In practice, production and stability of the ions and response differences of the detector for different masses are the limiting factors for the detection of high *m/z* values.

**Pros**

- high 'scan rate' (up to 20 000 scans/s), allows for the detection of narrow/transient chromatographic signals
- high resolution when reflectron and high-speed electronics are used
- virtually no limit on mass range
- high sensitivity, no ion loss because of scanning.

**Cons**

- strict demands on initial energy and spatial distribution of ions
- high-performance electronics needed
- low resolution in linear mode.

**Applications**

- high-resolution measurement using reflectron
- high-sensitivity measurements
- detection of transient signals (narrow chromatographic peaks).

### Sector Instruments

**Introduction:** The very first mass spectrometer used by Sir J.J. Thomson was a sector instrument; that is, he used the same physical forces still applied in modern instruments. When studying the nature of cathode rays (electrons) he applied magnetic and electric fields to a beam of electrons and thus obtained an estimate for the m/e value of the charged particles in these beams (11). The modern descendants of Thomson's devices are now highly sophisticated mass spectrometers working at high resolution, often in complex set-ups using multiple sectors (magnetic and/or electrostatic), and other mass analysers (quadrupoles, TOF).

**Working principle:** In sector instruments two types of sectors can be encountered: magnetic and electric, abbreviated

respectively as B and E. For an ion (charge $ze$) moving in a magnetic field ($B$) we know that the charged particle moves on a circular path (radius $r_m$), where the centrifugal force equals the force of the magnetic field on the ion.

$$zeBv = \frac{mv^2}{r_m} \qquad [11]$$

which yields

$$mv = zeBr_m \qquad [12]$$

Note that the magnetic field works as a momentum ($mv$) separator! If we want ions with equal mass and charge to follow the same path ($r_m$) through the fixed magnetic field ($B$), the momentum (or for a given mass the velocity $v$ or kinetic energy $mv^2/2$) must be constant.

If we give the ions a known kinetic energy by accelerating them in an electric field ($V_a$) we know that

$$\frac{mv^2}{2} = zeV_a \qquad [13]$$

When we substitute velocity into the previous equation we can measure the $m/z$ value of an ion as a function of the experimental parameters $B$ and $V_a$.

$$\frac{m}{z} = \frac{eB^2r_m^2}{2V_a} \qquad [14]$$

The kinetic energy of the ions depends on their initial energy and spatial distribution prior to acceleration. Therefore, an electrostatic sector is introduced. When the ions move through a radial electrical field, their path is determined by

$$\frac{mv^2}{r_e} = \frac{zeV_e}{d} \qquad [15]$$

where $V_e$ is the potential applied on the curved plates, $d$ the distance between both plates and $r_e$ the radius of the path the ions follow in the electric field. For a given geometry, $r_e$ and $d$ are fixed, the setting of $V_e$ now selects the kinetic energy $mv^2/2$.

As the ions move with the same velocity through both the magnetic and electrostatic sectors, we can equate the velocities used for electric and magnetic sectors which yield

$$\frac{zeBr_m}{m} = \sqrt{\frac{zeV_er_e}{md}} \qquad [16]$$

From this equation we can measure a mass spectrum by scanning the magnetic field B and/or the electrostatic field $V_e$.

$$\frac{m}{z} = \frac{ed}{V_er_e}\,B^2r_m^2 \qquad [17]$$

From this equation we see that $m/z$ is independent of the initial acceleration potential ($V_a$). However, by stating that the final velocity after acceleration is equal to the velocity in the electrostatic sector we obtain an instrument-defined correlation between the acceleration potential and the electric field strength ($V_e/d$) in the electrostatic sector.

$$V_a = \frac{V_er_e}{2d} \qquad [18]$$

In a double-focusing magnetic sector mass spectrometer, both a magnetic and electrostatic sector are used. Proper combination of both analysers, several different geometries are used (see Figure 4), yields an instrument that corrects for the spatial and velocity (energy) heterogeneities of the incoming ions. These instruments give mass spectra with high resolution (over 10 000), high accuracy of mass and a wide mass range (over 15 000 Da). Both can be increased (resolution to ±100 000 and mass range to over 100 000 Da), but at the cost of sensitivity loss because of reduced voltages and narrower slit openings. These instruments are used most often in fundamental studies.

To reduce the energy spread of the ion beam and improve ion extraction, the ion source is operated at high voltage (up to 10 kV). This high energy allows high-energy CID studies. However, these high voltages yield practical problems when coupled to an LC system. At the pressures used in LC–MS techniques, such as thermospray and electrospray (see "Interfaces for LC–MS" section) arcing is a serious problem, although good commercial solutions are available.

**Pros**

- high resolution and mass accuracy
- high-energy CID
- tandem MS experiments are possible.

**Cons**

- expensive instruments
- scan speeds limited by hysteresis and heating of the magnets
- limited sensitivity, especially at high resolution
- LC–MS coupling technically demanding (sparking in the source)
- complex instruments.

**Applications**

- high-resolution measurements
- fundamental MS studies (e.g., studies using fragmentation in the different field free regions.)

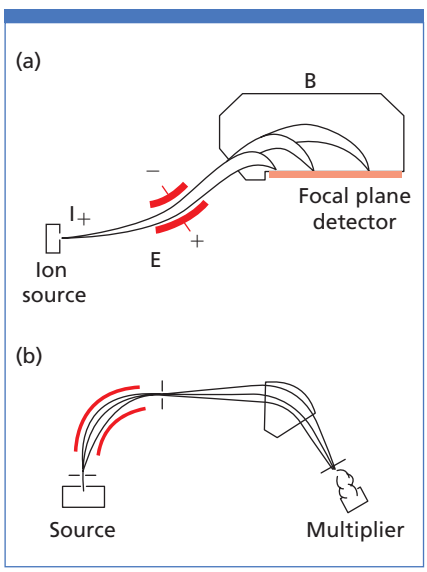


**Figure 4:** *Two double-focusing instruments. (a) Mattauch-Herzog geometry — 1935 and (b) Nier-Johnson geometry — 1953.*

## Fourier Transform Instruments

**Introduction:** The Fourier transform mass spectrometer (12) is the currently practised version of ion cyclotron resonance (ICR) mass spectrometry. Though being the *nec*

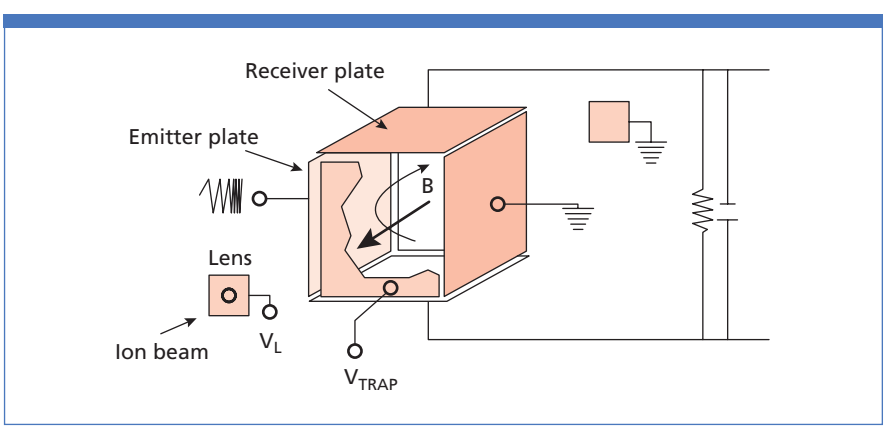


**Figure 5:** *Schematic of an ICR cell.*

*plus ultra* (nothing is higher) in terms of resolution (in excess of 106), detection efficiency (1 molecule detection (13)), scans speed and the $MS^n$ capabilities, its technical demands, especially in terms of vacuum technology, and its high price limits its use mostly to fundamental studies.

**Working principle:** As in sector instruments a magnetic field is used to curve the motion of the ions. However, the applied magnetic field B is so intense that the radius of the ion's trajectory becomes smaller than the inner dimensions of the mass spectrometer, effectively trapping the ions on a circular path inside the ICR cell. An electric potential on the trapping plates prevents the ions from escaping laterally (Figure 5).

The ions circle in the ICR cell with an angular velocity given by

$$\omega = 2\pi\nu = \frac{zeB}{m} \qquad [19]$$

The mass spectrum is given by

$$\frac{m}{z} = \frac{eB}{\omega} \qquad [20]$$

For a magnetic field of ±5 T and a mass range of 15–1500 amu, radio frequencies in the kHz to MHz range result. When a radiofrequent potential is applied to the emitter plates (Figure 5) ions with the corresponding angular frequency will absorb energy and their trajectory radius will increase. Thus, ions first randomly distributed over the circular path will become focused into a packet of ions circling together. Once the ion trajectory comes close enough to the receiver plates, an "image current" will be induced that can be detected. Scanning the frequency will yield a signal if an ion with the corresponding *m/z* value is present. This method can be thought of as pushing the keys of a piano one-by-one and listening to hear a note (i.e., a string is present — ion of corresponding *m/z* in the ICR). A faster method involves hitting the piano with a big hammer and listening to all the notes produced, and from that deriving which strings are present. In the ICR this is done by exciting all ions present using a fast sweep of frequencies over a broad range. The receiver plates pick up the signals of all the ions present in the cell. This complex signal can be reduced to its component frequencies by Fourier transformation. The individual frequencies can then be converted to corresponding *m/z* values by the equation given above. Note that the ions are not destroyed by the detection. Because they remain in the ICR cell, they can be re-observed (ion stability experiments) or fragmented for use in a second MS experiment (tandem MS).

At high frequencies/low *m/z* detection is limited by the speed of the digitizers; at low frequencies/high *m/z* the amplifiers and low frequency noise in electronics limits the mass range. The dynamic range is limited by the maximum number of ions that the ICR cell can hold. About 106 ions are allowed in the cell. Larger numbers result in repulsion between ions and, subsequently, lower resolution.

Collisions with residual gas in the ICR cell reduce the amplitude of the cyclotron motion of the ions. As such, a high vacuum must be used ($10^{-8}$ Torr). This poses a technical challenge when coupling with LC interfaces. However successful couplings have been made (14). Because of the number of calculations needed for Fourier transform, dedicated high-performance computers are needed to cope with the data flow generated by an LC separation.

**Pros**

- unsurpassed resolution
- high detection efficiency
- detection is not destructive (ion stability experiments)
- inherent tandem MS capabilities.

**Cons**

- high price, both in purchase and use (cooling of super conducting magnets!)
- high vacuum, difficult to couple with high pressure ion sources (LC–MS)
- demanding on computing facilities, problems with fast signal (narrow LC peaks).

**Applications**

- extreme high-resolution measurements
- fundamental ion chemistry studies
- detection of single ions

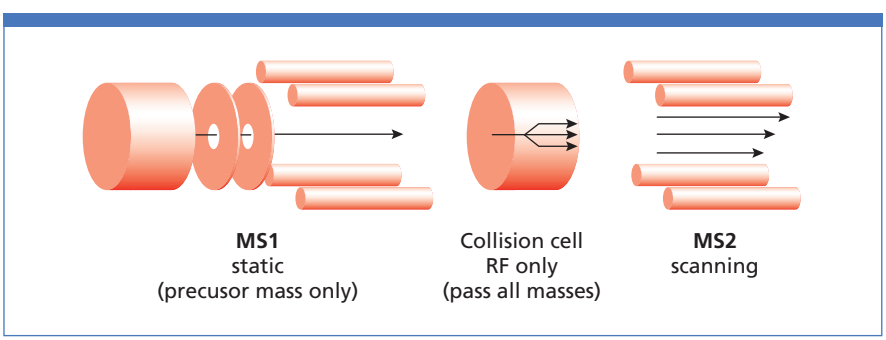


**Figure 6:** *The product ion scan.*

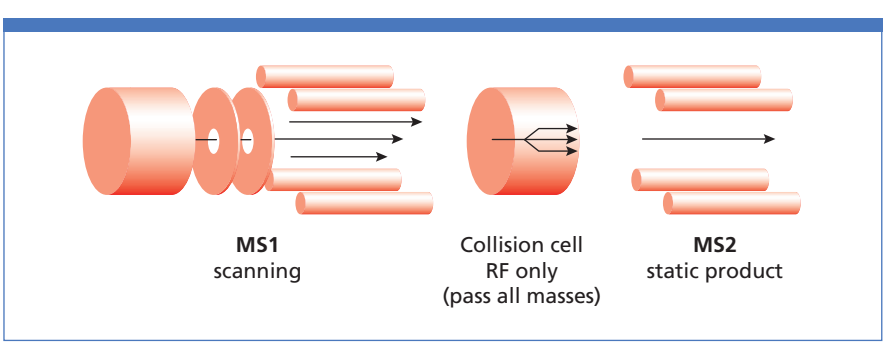


**Figure 7:** *The precursor ion scan.*

## Tandem Instruments

Tandem mass spectrometry employs two or more stages of mass spectrometric analysis. Each mass spectrometer might scan, select one ion or transmit all ions. In between two MS experiment the ions can be subjected to different actions; collision with neutral or reactive gases, collision with surfaces, interaction with light, electrons or other ions, acceleration, deceleration, neutralization, decomposition etc. Selection from these scanning or non-scanning MS analysers and the different actions on the ions between the analysers yields a panoply of possible tandem MS experiments. Describing all of them is far beyond the scope of this text. Therefore, focus will be put on the most used instruments and scanning methods.

## Tandem MS Scanning Methods

**Product ion scan:** This is one of the most widespread scanning modes in MS–MS, often called 'daughter ion scanning'. The first mass analyser (MS1) is used to select an ion, called the precursor or parent ion, which is provided with extra energy in a reaction region to induce fragmentation.

This energy can be high (kEV range) or low (eV range), and various techniques can be used to provide it.

The most common method is to collide the precursor ion with an inert gas (often a noble gas) introduced into the reaction region, here called the "collision cell". This process is termed collision-activated dissociation (CAD), and is the technique used in this study. Collisions with a solid surface result in surface-induced fragmentation. Alternatives include excitation of the ions by photon absorption using, for example, a laser, resulting in photo-dissociation, or the use of an electron beam to excite the ions. The excited ions fall apart into a charged fragment ion, the product ion, and one or more neutral parts. The product ions are then sampled into the next mass filter (MS2) scanning over the mass-to-charge (*m/z*) range of interest and detected yielding a product ion spectrum (See Figure 6).

**Precursor ion scan:** In terms of activity of the mass filters, this scanning mode, also named parent ion scan, is the mirror image of the product ion scan. MS1 scans while MS2 is fixed at a chosen *m/z* value. Because there is only a response at the detector when a precursor of the selected ion passes through MS1, here the precursor ions of a selected product are detected (see Figure 7). This method can be applied to detect the nucleotides present in a more or less complex mixture. Selecting in MS2 a fragment typical for the presence of a phosphate moiety ($PO3^-$, *m/z* 79 in ES(-)), and scanning MS1 yields only a signal for those precursor ions bearing a phosphate group, thus reducing the interference (chemical noise) of other compounds in the mixture (15).

**Constant neutral loss:** When both analysers are scanned with a fixed mass difference (MS1 higher than MS2), only those ions that lose that specific mass in the collision cell will be detected (see Figure 8). This feature provides another opportunity to detect products of a specific class containing similar structural features. Nucleosides can be detected as a group by their loss of the (2'-deoxy)-ribose moiety ($\Delta = 116$), which is characteristic for (2'-deoxy)-nucleosides, however, a whole range of losses can be used: ($\Delta = 18$ for water, $\Delta = 44$ for carbon dioxide, $\Delta = 46$ for $NO_2$ etc). As for precursor ion scanning, constant neutral loss (CNL) enhances data by reducing chemical noise.

A related technique is constant neutral gain (CNG), essentially the same scan technique as CNL, but MS1 is a constant mass difference below MS2. For this technique to be applicable, the ions of interest passing through the collision cell should interact with the gas present there. Obviously, this requires some kind of 'reactivity' of the collision gas and the inert gases used for the techniques mentioned above are useless.

***"The selectivity of mass selection can be used to enhance the detection limits in analytical procedures."***

**Single/multiple reaction monitoring:** The selectivity of mass selection can be used to enhance the detection limits in analytical procedures. By fixing MS1 on the mass-to-charge ratio of interest, the signal at the detector is improved. The simple reason for this phenomenon is that practically all the ions of interest will be detected, whereas in scanning MS1 an important fraction of the ions of interest is lost. To eliminate interference from isobaric ions and the isotopic contribution of lighter analytes, one can select, after fragmentation, a product ion characteristic for the analyte of interest using MS2, thus removing the interference. A single reaction is monitored, yielding a highly selective detection with high sensitivity because of the removal of chemical noise. Selecting two or more characteristic ions is possible as well, a technique called multiple reaction monitoring (MRM) (see Figure 9).

**Triple Quadrupole Instruments**

A triple quadrupole instrument uses two quadrupole MS analysers for the actual MS experiments and a third quadrupole in RF-only mode, which transmits all incoming ions from MS1 to MS2 (16). This second intermediate quadrupole, (in some commercial instruments higher multipoles are applied) is placed in a partially closed area filled with a collision gas. In the majority of experiments a neutral gas, often a noble gas, is used.

These instruments have all the advantages of the quadrupoles (see earlier). As both MS experiments are separated in space; that is, MS1 and MS2 are two different mass analysers (for comparison see *Ion Traps* and *Fourier Transform Instruments*), both mass spectrometers can be scanned simultaneously. This allows CNL and CNG experiments.

**Pros**

- unit resolution tandem MS experiments
- relatively low price
- ease of operation
- high sensitivity in MRM experiments
- allows parent ion scanning, CNL and CNG.

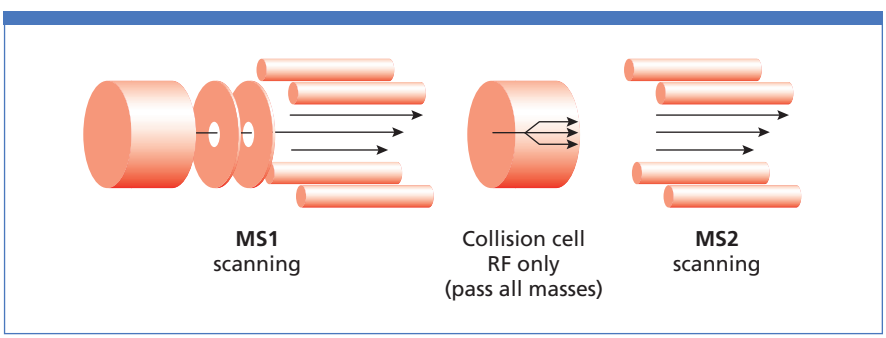


**Figure 8:** *The constant neutral loss/gain scan.*

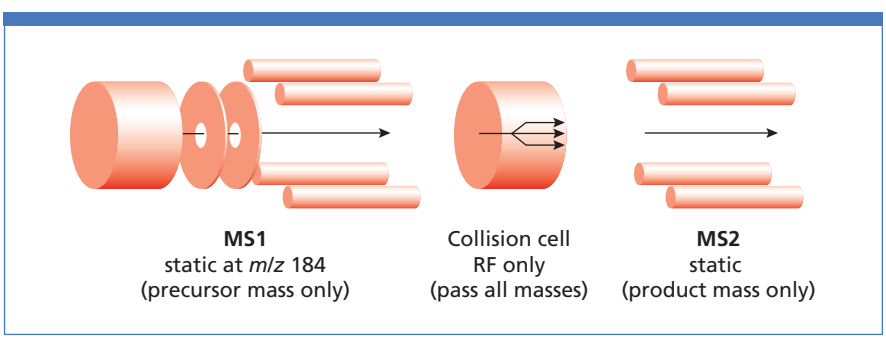


**Figure 9:** *Single/multiple reaction monitoring.*

**Cons**
- limited to MS2 experiments.

**Applications**
- general tandem MS instrument
- sensitive detection system (MRM).

### Quadrupole–TOF Instruments

**Introduction:** The quadrupole orthogonal acceleration time-of-flight instrument is derived from triple quadrupole instruments. The last quadrupole is replaced by a TOF analyser, often equipped with a reflectron. The collision cell is used to fragment the precursor ions and to decelerate the ions prior to orthogonal acceleration into the TOF analyser. As the TOF analyser is an obligatory scanning device, all tandem MS experiments requiring a non-scanning MS2 are excluded for a Q-TOF instrument. Also, experiments requiring parallel scanning of MS1 and MS2 are impossible (CNL and CNG). However, because of its fast scanning, intelligent software applications and dynamic experiments can overcome this potential handicap and produce results similar to, or even more informative than, precursor ion scanning, CNL and CNG.

**Pros**
- high-resolution product ion spectra
- high sensitivity because of high transmission in MS2 (TOF).

**Cons**
- parent ion scans and CNL/CNG require extra software.

**Applications**
- proteomics.

### Ion Storage Systems

In both ion traps and Fourier transform MS instruments ions can be selected and stored in the MS, fragmented and the product ions analysed by a second MS experiment using the same mass analyser; that is, the two (or more) successive MS experiments are separated in time instead of space (two different MS analysers). Being an advantage for some types of experiments ($MS^n$ is possible) MS–MS experiments in which MS1 scans and MS2 selects a specific product ion are impossible.

**Pros**
- both ion traps and Fourier MS instruments have inherent tandem MS capabilities.

**Cons**
- not all tandem MS experiments are possible.

**Applications**
- proteomics.

### Combinations with Sector Instruments

**Introduction:** Because most sector instruments use more than one sector, it is possible to analyse fragment ions that are formed in the field-free regions before and in between the sectors. Although widely used the scanning methods are rather complex and resolutions are not very good, depending on the scan method used. As a result of the high ion velocities used in these instruments, high energy CID spectra are generally obtained (collisions in the keV region).

**Multiple sector instruments:** By adding extra sectors to the instrument (E and/or B), performance as a tandem MS dramatically increases. Several different configurations are built and used. The most successful instruments appear to be of the EBE and BEB configuration. In these configurations several different combinations of sectors and choice of reaction region can be made. Using EBE, the first two sectors can be used as a high-resolution MS selecting the precursor ion, and E analyses the product ions. Using the BEB configurations, with B as MS1, the last two sectors can work as a high-resolving MS2 that yields product ion spectra with high resolution. Further extension to a four-sector instrument EBEB, allows both MS1 and MS2 to be run at high resolution.

These complex instruments are often equipped with extra acceleration and deceleration lens stacks in between the sectors in order to control collision energies. They find their application mostly in fundamental ion chemistry and physics studies.

**Sectors combined with quadrupoles:** As quadrupoles are cheaper than sectors, the addition of quadrupoles to a sector instrument is more cost effective than adding extra sectors. Using quadrupoles also allows low-energy CID without another acceleration for MS analysis using a sector.

**Sectors combined with TOF:** Sectors have been combined with TOF measurements. However, these instruments are mostly built for fundamental studies and have few applications in the LC–MS field.

### References


**(1)** E. De Hoffman, J. Charette and V. Stroobant, *Mass Spectrometry, Principles and Applications*, (John Wiley & Sons Ltd, Chichester, UK, 1996.)
**(2)** D.M. Desiderio, Ed., *Mass Spectrometry, Clinical and Biomedical Applications*, **1**,(1992); **2**,(1994), (Plenum Press, New York, USA.)
**(3)** K.L. Busch, G.L. Glish and S.A. McLuckey, *Mass Spectrometry/Mass Spectrometry: Techniques and Applications of Tandem Mass Spectrometry*, (VCH Publishers, New York, USA, 1988.)
**(4)** P.H Dawson, *Mass Spectrom. Rev.*, **5**(1), 1–37, (1986).
**(5)** J.F.J. Todd, *Mass Spectrom. Rev.*, **10**, 3–52, (1991).
**(6)** *Phys. Rev.*, **69**, 691, (1946).
**(7)** *Bull. Am. Phys. Soc.*, **21**(2), 22, (1946).
**(8)** B.A. Mamyrin et al., *Sov. Phys. JETP*, **37**, 45–48, (1973).
**(9)** W.C. Wiley and I.H. McLaren, *Rev. Sci. Instrum.*, **16**(12), 1150–1157, (1955).
**(10)** M. Guilhaus, *J. Mass Spectrom.*, **30**, 1519–1532, (1995).
**(11)** J.J. Thomson, *Phil. Mag.*, **20**(6), 752–67, (1911).
**(12)** A.G. Marshall, C.L. Hendrickson and G.S. Jackson, *Mass Spectrom. Rev.*, **17**(1), 1–35, (1998).
**(13)** R.D. Smith et al., *Nature*, **369**, 137–138, (1994).
**(14)** L. Tang et al., *Rapid Commun. Mass Spectrom.*, **9**, 731–734, (1995).
**(15)** G. Neubauer and M. Mann, *J. Mass Spectrom.*, **32**(1), 94–98, (1997).
**(16)** R.A. Yost and C.G. Enke, *J. Am. Chem. Soc.*, **100**, 2278, (1978).



*Dr Filip Lemière works in the Nucleoside Research and Mass Spectrometry Unit, Department of Chemistry at the University of Antwerp, Belgium. His research interests include the miniaturization of LC–MS procedures particularly for the analysis of DNA adducts.*