

Analysis techniques:

'Normal' mass spectra:

The most basic operation of a mass analyser is to record a 'normal' mass spectrum, that is to say a scan through a given appropriate range of masses. All peaks which appear in the chosen mass range are swept past the detector slit, giving peaks of gaussian shape which can be converted using software into data pairs of mass and intensity. The position of the peak on the time axis is assigned by the position of the centroid of the peak, and its intensity is usually given by a peak's maximum height (though sometimes the area is used).

A relationship has to be established between the time from the start of a scan and mass. This is achieved by means of a 'reference' compound which gives peaks of known mass over the range of interest. The scan is performed with the reference compound introduced into the source, and a relationship (known as the 'scan law') is established between time and mass. This relationship may subsequently be used to convert times into masses when unknown compounds are run.

Accurate mass measurements:

The above scanning procedures afford sufficient mass measurement accuracy (typically 100-200ppm) for integer or 'nominal' mass measurement.

If we introduce a reference compound into the ion source at the same time as the unknown sample, we can use peaks of known mass from this reference compound to calibrate the mass scale within the same scan.

This can produce mass measurement accuracy of better than 5ppm, which is sufficient to establish elemental composition for the measured ion.

Tandem Mass Spectrometry (MS/MS)

Metastable ions:

These ions may be formed by decomposition of ions in any of the field-free regions of a magnetic sector mass spectrometer.

When an ion of given energy fragments, the energy possessed by it is divided between the fragments in the inverse ratio of their masses.

For a transition $M_1^+ \rightarrow M_2^+$, the metastable peak will appear at mass M^* on the mass scale, where

$$M^* = M_2^2 / M_1$$

Metastable ions are recorded as very weak, broad peaks in the normal mass spectrum. The width of the peaks is due to the release of energy during fragmentation. Because these peaks are so wide and of such low intensity, they are unsuitable for analytical use. Also the equation relating the masses cannot be solved uniquely for M_1 and M_2 , so unambiguous mass assignment is not possible.

Using a double-focussing magnetic sector mass spectrometer, specific ion fragmentations can be examined by employing special scanning modes. In the following descriptions, the "electric sector" is the electrostatic analyser (ESA), B is the magnetic field, and E the electric sector voltage. The most commonly used scan modes are:

- 1) Daughter ion scans. In this mode a parent or precursor ion is selected, and the scan gives all the daughter ions resulting from fragmentation of that ion. The scan is achieved by scanning the magnetic and electric sectors together such that B/E is a constant.
- 2) Parent ion scans. In this mode a fragment ion is selected, and the scan shows all the ions which have fragmented to give that daughter ion. The scan is achieved by scanning the magnetic and electric sectors together such that B^2/E is a constant. Resolution of the spectrum is very poor due to the energy released during fragmentation.
- 3) Constant neutral loss scans. In this mode, a spectrum is recorded showing all the ions which have fragmented by the loss of a specific neutral mass. The scan is achieved by scanning the magnetic and electric sectors such that $B/E(1-E)^{1/2}$ is a constant.

All the above scan modes observe fragmentations which occur in the first field free region of the double focusing mass spectrometer. (between the source and the ESA in forward-geometry machines).

Metastable ions to be observed in this way are often of low abundance if the fragmentations occur with rate constants which favour dissociation in the ion source rather than in the field free region. The abundance can be enhanced by the employment of collision-induced dissociation (CID) or collisional activation (CA).

In CID, the ion beam is passed through a small region (the collision cell), which contains a high pressure ($\sim 10^{-3}$ mbar) of an inert gas (typically helium). This cell ideally is placed at or near to the focal point of the field-free region. As the ions undergo collisions with the inert gas, part of the kinetic energy of the ions is converted to internal energy, resulting in fragmentation of the ion.

As the pressure is increased, the fragmentation efficiency increases, but the collection efficiency decreases. Overall efficiency is the product of the fragmentation & collection efficiencies. Typically, the best efficiency is achieved when the precursor ion beam is attenuated by about 70% in the collision cell.

Tandem mass spectrometer instrument configurations.

Instruments designed for the examination of ion fragmentations are available, which consist of two mass analysers separated by a collision cell. The most common configurations for tandem mass spectrometry are:

1) Triple quadrupole mass spectrometer. This instrument consists of two conventional quadrupole mass analysers (Q_1 and Q_3) separated by an rf-only quadrupole (Q_2), which acts as a collision cell. The rf-only quadrupole will transmit ions of all masses. Because quadrupoles are true m/z analysers, peak distortion due to energy release is absent. Scan laws are very simple to understand and control.

Modes available are:

- 1) Setting Q_1 to transmit a precursor ion while scanning Q_3 gives a daughter ion scan of the selected precursor.
- 2) Setting Q_3 to transmit a daughter ion while scanning Q_1 gives a precursor ion scan of the selected daughter.
- 3) Scanning Q_1 and Q_3 with a fixed difference in mass gives a constant neutral loss scan.
- 4) Setting Q_1 and Q_3 to transmit selected masses will monitor for a specific fragmentation pathway.

2) Hybrid mass spectrometers. These instruments use a combination of magnetic sector / ESA geometry as a high-resolution first analyser, and a lower resolution second analyser. Quadrupole, time-of-flight, and ion trap analysers can all be used as the second analyser.

3) Ion trapping analysers. Both ion trap and FT/ICR analysers can be used for MS/MS. Ions of the required mass are trapped in the analyser and collisions induced with an inert gas introduced. Because the ions are trapped, sequential MS/MS experiments can be performed. Precursor or neutral loss scans are not possible with these analysers.

4) Multi-sector mass spectrometers. These instruments consist of combinations of ESA and magnetic sectors arranged such as to give two mass analysers, separated by a collision cell. They can be used for high-resolution MS/MS studies. Many configurations are available, including EBEB, BEEB and BEB geometry.