# SECTION 3.3

## Elemental microanalysis and mass spectrometry

**Elemental microanalysis**

Elemental analysis (or combustion analysis) is used to determine the masses of the elements in a sample of an organic compound in order to calculate its empirical formula.

The carbon, hydrogen and nitrogen contents are measured using an instrument known as a CHN analyser. For analysis approximately 2 mg of the solid sample is placed in a small tin capsule and weighed to $\pm$ 0.0001 g. This sample is then sealed before being put into a furnace where it is combusted in an atmosphere of oxygen at almost 1000$^{\circ}$C. At this temperature, the carbon, hydrogen and nitrogen react with oxygen to form $CO_2$, $H_2O$ and $N_xO_y$ respectively. These gases are carried via a stream of helium gas through a chromatography column to a detector. The detector sends data to a computer that compares it with that of a known value of a standard. Acetanilide is generally used as the standard since it conveniently contains the elements carbon, hydrogen and nitrogen.

Samples are usually analysed in duplicate to give more reliable results. The results are then given as a percentage, by mass, of each of the three elements.

If the sample is a liquid it is aspirated through a capillary glass tube before being weighed and put into the furnace.

To measure the sulphur content of a sample, a similar instrument with slight modifications needs to be used.

Compounds containing chlorine, bromine and iodine are titrated to determine the halide concentration. The sample is combusted and the gases passed through a halide-absorbing solution, which is then placed in an auto-titrator where a computer calculates the percentage of each halide. All three of these halides can be analysed simultaneously. Another method must be used to detect and measure the fluorine content of a sample.

**Questions**

1.   A sample of an organic compound with a mass of 1.224 g was completely burned in oxygen and found to produce 2.340 g of $CO_2$ and 1.433 g of water only. Calculate the empirical formula of the organic compound.

2.   Oxalic acid is found in rhubarb and contains only the elements carbon, hydrogen and oxygen. When 1.540 g of oxalic acid was burned in oxygen, 1.504 g of $CO_2$ and 0.310 g of water were formed. Calculate the empirical formula for oxalic acid. If the molecular mass of oxalic acid is 90.0, what is its molecular formula?

3.   An organometallic compound known as ferrocene contains only the elements Fe, C and H. When 1.672 g of ferrocene was combusted in oxygen, 3.962 g of $CO_2$ and 0.810 g of water were formed. Calculate the empirical formula of ferrocene.

**Mass spectrometry**

Mass spectrometry can be used to determine the accurate molecular mass and structural features of an organic compound. Figure 106 shows a diagram of a simple mass spectrometer.

*Figure 106*
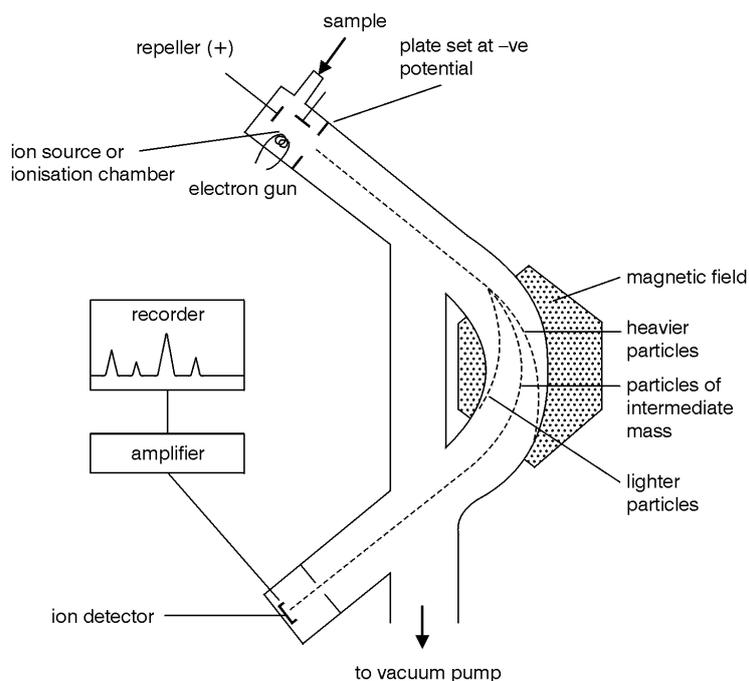
This type of mass spectrometer functions in the following manner:
1. The vacuum pump is switched on to reduce the pressure in the spectrometer.
2. A sample is introduced into the spectrometer and vaporised.
3. The vaporised sample is then bombarded with an electron beam in the ionisation chamber. These electrons 'knock off' some of the outer electrons from the molecules in the sample and positive ions are formed.
4. Fragmentation of the molecules can also occur when the energy available is greater than the molecular ionisation energy. Some of these different fragments are also positive ions.
5. The parent ion and ion fragments are accelerated by an electric field until they have similar kinetic energy and form an ion beam.
6. The ions are deviated by a magnetic field in such a way that ions of one particular mass/charge ratio continue along the spectrometer and hit the detector.
7. The strength of the magnetic field is varied to enable the ions of all different mass/charge ratios to be detected in turn.
8. The mass spectrum is plotted automatically by the instrument. The mass/charge ratio is labelled as *m/z*. Doubly charged ions can be formed in the mass spectrometer and they are detected at half their mass value on the mass spectrum.

To provide an accuracy of up to 1 in $10^6$ of an atomic mass unit, a double focusing spectrometer is used. This has greater resolving power and can distinguish between molecules of compounds with similar molecular masses, as in Table 8.

*Table 8*

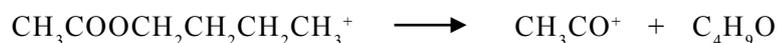| Formula | Molecular mass | Exact mass |
|---|---|---|
| $C_4H_4N_5$ | 122 | 122.046668 |
| $C_4H_{10}O_4$ | 122 | 122.057903 |
| $C_6H_4NO_2$ | 122 | 122.024201 |
| $C_6H_6N_2O$ | 122 | 122.048010 |
| $C_6H_8N_3$ | 122 | 122.071819 |
| $C_7H_8NO$ | 122 | 122.060585 |

When an organic molecule loses an electron a molecular ion is formed since one electron has been removed from a pair in a filled orbital:
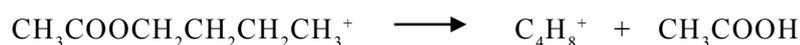
$$M(g) \longrightarrow M^{\bullet+}(g) + e^-$$

By convention the radical electron ($\bullet$) is omitted from the representation of these ions.

When a molecular ion fragments, it forms other ions and neutral molecules. This gives rise to a number of peaks in the mass spectrum. The arrangement and sizes of the peaks formed when the molecular ion of an organic compound breaks down in the mass spectrometer is known as the fragmentation pattern.

In some compounds the molecular ion forms a large peak in the mass spectrum, e.g. the peak at $m/z$ 122 in the mass spectrum of benzoic acid (Figure 108). In other compounds the molecular ion fragments so easily that it produces an insignificant peak in the mass spectrum, e.g. butyl ethanoate (Figure 107). The molecular ion of butyl ethanoate fragments as follows:
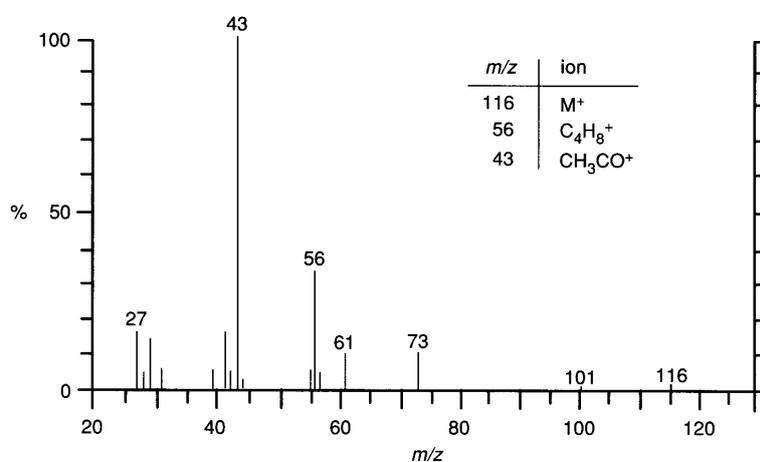
$$CH_3COOCH_2CH_2CH_2CH_3{}^+ \longrightarrow CH_3CO^+ + C_4H_9O$$

and

$$CH_3COOCH_2CH_2CH_2CH_3{}^+ \longrightarrow C_4H_8{}^+ + CH_3COOH$$

The remaining ions fragment further to give the mass spectrum shown in Figure 107.

**Figure 107:** *Mass spectrum of butyl ethanoate*



| $m/z$ | ion |
|-------|-----|
| 116 | $M^+$ |
| 56 | $C_4H_8{}^+$ |
| 43 | $CH_3CO^+$ |

The fragmentation pattern of an organic compound can be used to help identify the type of organic compound present.

**Question**

Look at the mass spectra of benzoic acid and methyl benzoate and identify the ions responsible for the major peaks in each case.
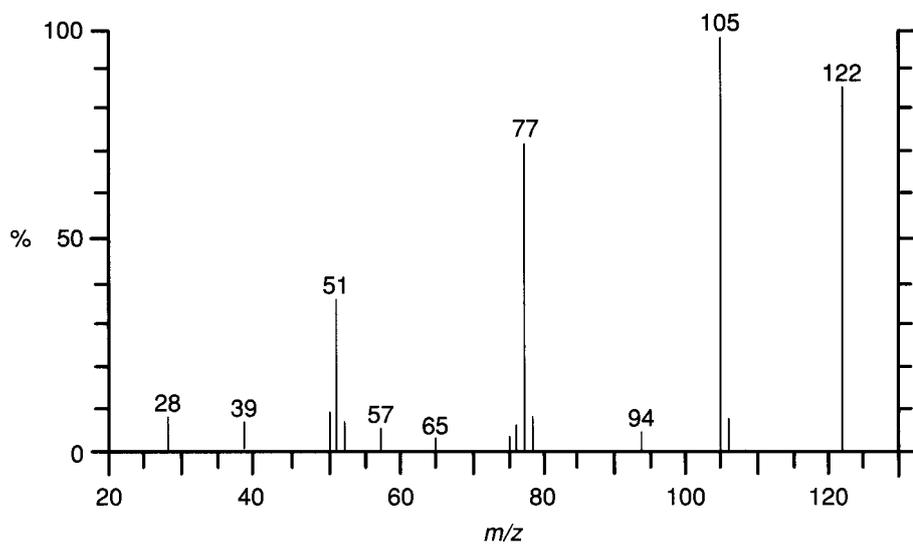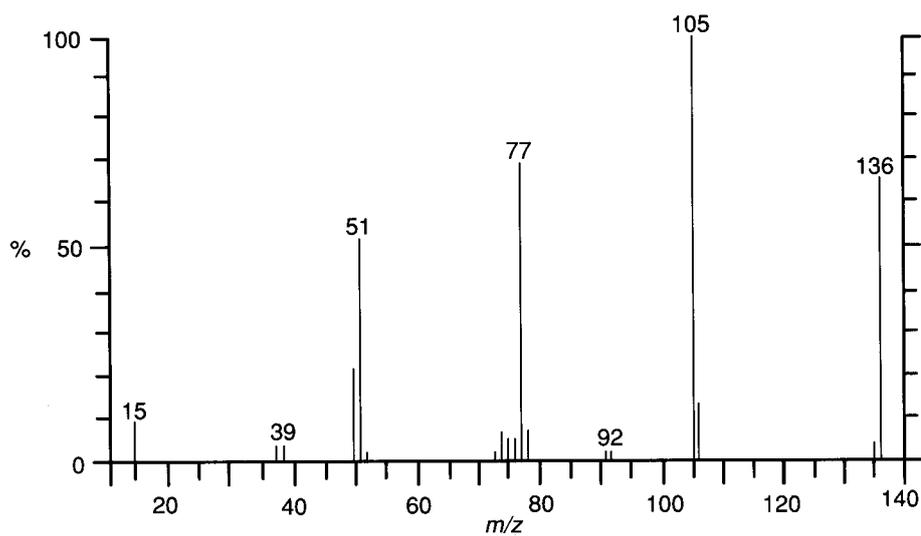
*Figure 108: Mass spectrum of benzoic acid*



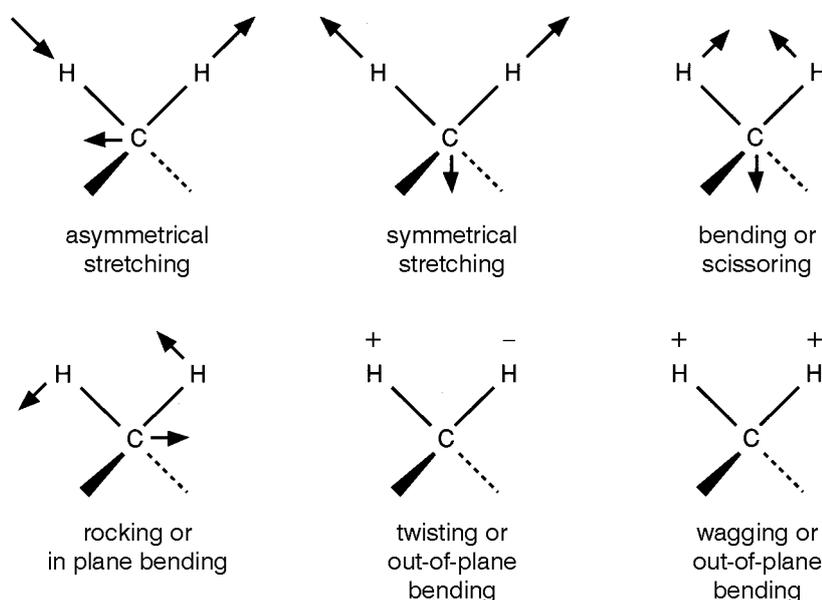*Figure 109: Mass spectrum of methyl benzoate*

# Infra-red and nuclear magnetic resonance spectroscopy and X-ray crystallography

## Infra-red spectroscopy

Infra-red spectroscopy can be used to identify certain functional groups in an organic compound. The atoms in molecules vibrate even in the solid state. Each atom vibrates with a frequency that depends on its mass and the length and strength of its bonds. The natural vibrational frequency of chemical bonds lies within the infra-red region of the electromagnetic spectrum and so molecular vibrations are stimulated by bonds absorbing infra-red radiation. In simpler terms it can be stated that infra-red radiation causes parts of a molecule to vibrate. The wavelengths that are absorbed and cause these vibrations depend on the nature of the chemical bonds and the groups or atoms at the ends of the bonds.

Examples of different types of vibration are shown in Figure 110.

*Figure 110*



|  |  |  |
|---|---|---|
| asymmetrical stretching | symmetrical stretching | bending or scissoring |
| rocking or in plane bending | twisting or out-of-plane bending | wagging or out-of-plane bending |

+ and – mean here a movement out of and into the plane of the paper respectively

These different vibrations absorb different frequencies of infra-red radiation. The absorption is usually given in wavenumbers, units $cm^{-1}$. The relationship between frequency, wavelength, wavenumber and energy dealt with in Unit 1 is repeated here. The relationship between wavelength, frequency and velocity is:

$$\text{velocity} = \text{wavelength} \times \text{frequency}$$
$$(\text{m s}^{-1}) \qquad (\text{m}) \qquad (\text{s}^{-1})$$

$$c = \lambda \times v \text{ and hence } v = c/\lambda$$

Wavenumber is the reciprocal of wavelength and is given the symbol $\bar{v}$ :

$\bar{v} = 1/\lambda$ and hence $\bar{v} = cv$

$E = Lhv$ or $E = \dfrac{Lhc}{\lambda}$ or $E = Lhc\bar{v}$

where L is Avogadro's constant.

These relationships are summarised in Table 9.

*Table 9*

| Quantity | Symbol | Units | Description |
|---|---|---|---|
| Wavelength | $\lambda$ (lambda) | Metres (m) | Distance between adjacent wave crests  |
| Velocity | $c$ | m s$^{-1}$ | Rate of advance of any one wave crest  |
| Frequency | $v$ (nu) | Hertz (Hz) | The number of wavelengths or cycles passing a point in 1 second. 1 cycle per second = 1 Hz $v = c/\lambda$ or $v \propto 1/\lambda$ (for all electromagnetic radiation $c$ is constant, $3 \times 10^8$ m s$^{-1}$) |
| Wavenumber | $\bar{v}$ | Waves per metre (m$^{-1}$) but many spectrometers are calibrated in waves per cm (cm$^{-1}$). | Number of waves in 1 metre or 1 centimetre of radiation. Often used in place of frequency. $\bar{v} = \dfrac{1}{\lambda}$ |
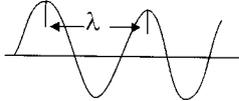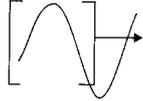
Compared to visible light, infra-red radiation has lower frequency and thus longer wavelength and lower energy.  A typical infra-red spectrometer measures absorbance in the range 650–4000 cm$^{-1}$.
Conventional infra-red spectrometers have two beams of radiation: one passes

through the sample and the other passes through a reference cell. When a particular frequency is absorbed by the sample, less radiation is transmitted and the detector compares the intensity passing through the sample with that passing through the reference.

Only vibrations resulting in a change in the dipole moment absorb infra-red radiation. In simple molecules such as $SO_2$, it is relatively easy to assign each absorption to a particular vibration. With more complex molecules each absorption can be assigned to the vibrations of particular functional groups within the molecule. Since particular types of vibration always occur at a similar frequency it is possible to build up a table of characteristic absorption frequencies (see page 13 of the Data Booklet). This means that a study of the infra-red spectrum of a compound will enable the functional groups present in that compound to be identified.

The infra-red spectrum of propanal is shown in Figure 111. Note that the major absorption peak at around 1730 $cm^{-1}$ is due to the carbonyl group.

*Figure 111*
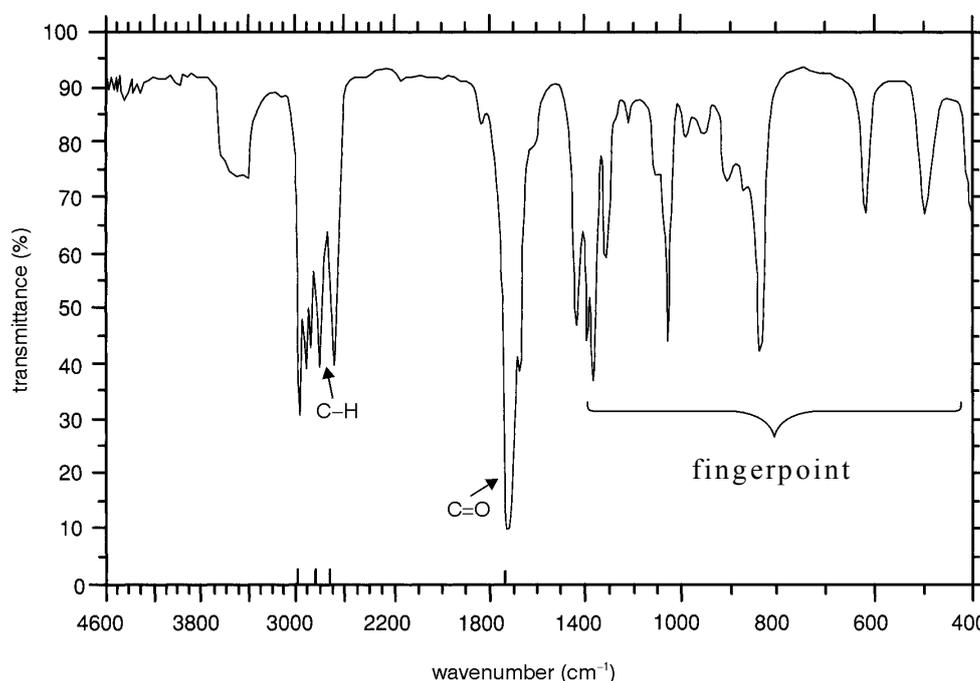


The pattern of absorptions below 1400 $cm^{-1}$ is characteristic of a particular organic compound and can be used in its identification. This region is often called the 'fingerprint' region.

**Question**

The spectra below are of ethanoic acid, $CH_3COOH$ (Figure 112), and ethanoic anhydride, $(CH_3CO)_2O$ (Figure 113). Draw the full structural formula for both compounds and then determine, giving reasons, which spectrum is due to which compound.
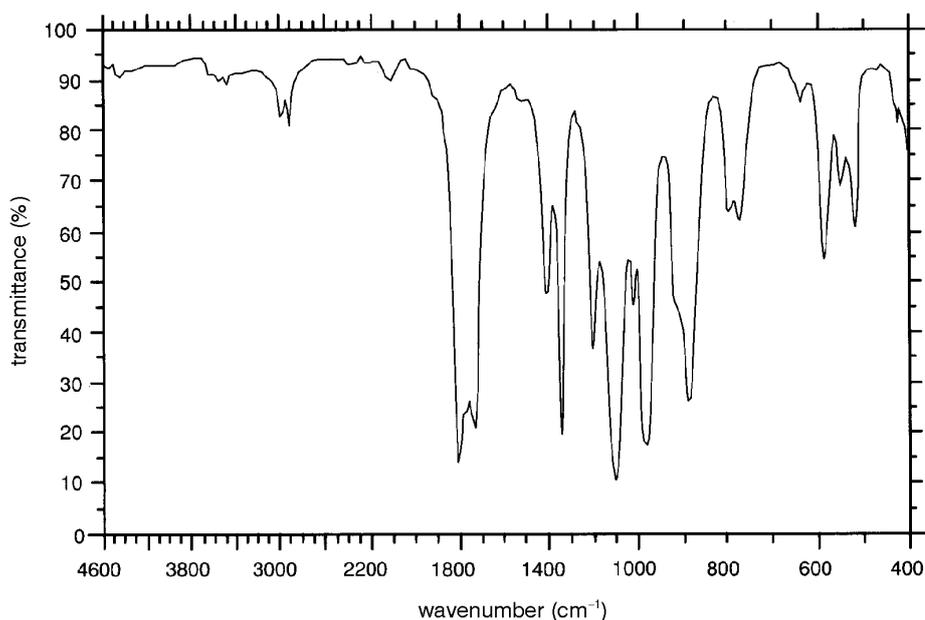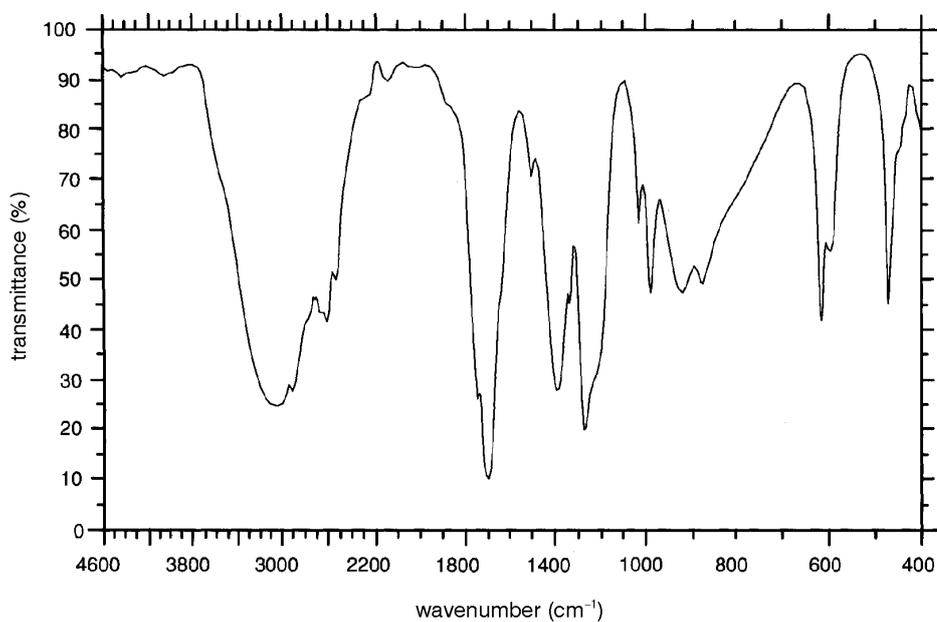
*Figure 112*



*Figure 113*

**Nuclear magnetic resonance spectroscopy**

Nuclear magnetic resonance (NMR) spectroscopy is one of the most useful analytical tools available to the modern organic chemist. It is possible to derive a huge amount of information concerning the environment of the carbon and hydrogen atoms in an organic molecule from a single NMR spectrum.

**Theory**

It has been discovered that many nuclei behave as if they were spinning about an axis. For organic chemists, the most important of these nuclei are $^1$H and $^{13}$C. We will only consider $^1$H at Advanced Higher level. Since the nucleus of an $^1$H atom is simply a proton, the technique is often described as proton NMR spectroscopy.

Any spinning charged particle produces a magnetic field. In other words it will behave like a tiny magnet. Any $^1$H nucleus will have either of two possible spin states (Figure 114). Compare this with the similar situation that arises with electrons (see Unit 1).

*Figure 114*

In the absence of a magnetic field, the spin states have the same energy. However, if the nuclei are placed in an external magnetic field (e.g. between the poles of a powerful magnet), the nuclei become orientated in much the same way as a compass needle orientates itself in the earth's magnetic field. The nuclei with one spin state will align themselves in the same direction as the magnetic field (parallel to the field) while the nuclei with the other spin state will oppose the magnetic field (Figure 115).

*Figure 115*



(a) no magnetic field        (b) external magnetic field applied
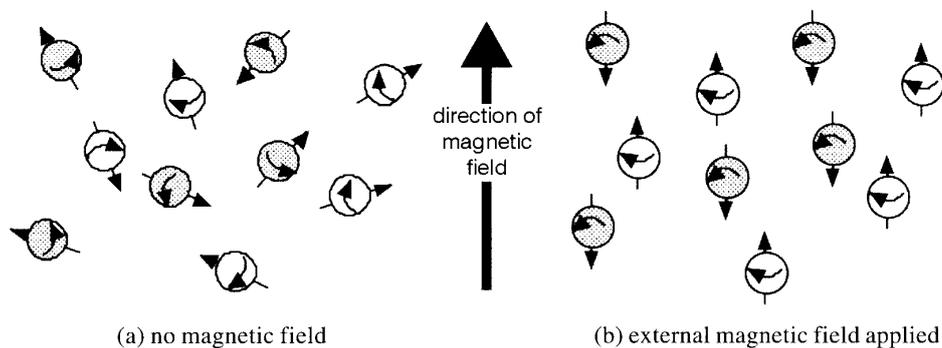
Those nuclei that are parallel to the magnetic field will have lower energy than those that oppose the field (Figure 116).

*Figure 116*



(a) no magnetic field      (b) external magnetic field applied
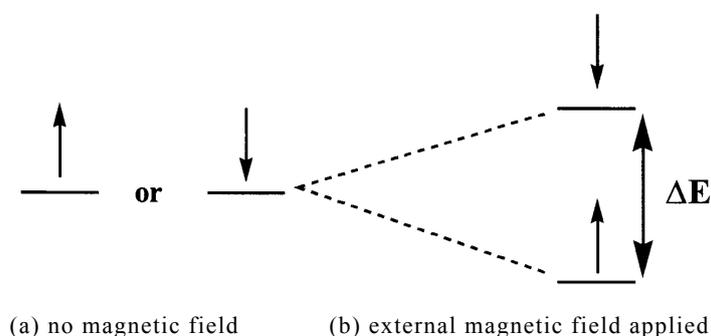
The stronger the applied magnetic field, the greater will be the energy difference( $\Delta E$ ) between the two states. If nuclei that are parallel to the field absorb sufficient energy, the spin can be 'flipped' to the higher energy state. The energy difference between the two states corresponds to the radio frequency region of the electromagnetic spectrum, typically in the VHF region, 60–1000 MHz. When the nuclei fall back to the lower state, the energy emitted can be detected.

Electrons also have the property of spin (see Unit 1). The electron cloud around a nucleus will therefore generate its own magnetic field. When an external magnetic field is applied, the $^1H$ nucleus does not experience its full effect. Instead it will experience a different field because it is 'shielded' by the magnetic field of the electron cloud. In any organic molecule, the density of the electron cloud will vary from one part of the molecule to another. Different hydrogen nuclei will experience slightly different magnetic fields

and will therefore have slightly different energy gaps ($\Delta E$) between their spin states. The radiation absorbed or emitted by an $^1$H nucleus as it changes from one state to another therefore depends on its local environment in the molecule. This variation of absorption frequency with chemical environment is called **chemical shift**.

### *Producing an $^1$H NMR spectrum*

The sample to be analysed is dissolved in a solvent that contains no hydrogen atoms, e.g. $CDCl_3$ or $CD_3COCD_3$. D is deuterium ($^2$H), which has an NMR frequency that is a long way from $^1$H. The applied magnetic field is kept constant and a pulse of radio frequency is supplied which promotes some nuclei to the higher energy state. Immediately after the pulse, some nuclei will drop back and emit radiation of a frequency corresponding to $\Delta E$. This radiation can be detected but it is very weak. The process is repeated many times and the data is repetitively added up and stored and then analysed by computer.

When producing an $^1$H NMR spectrum, an internal reference standard is included in the solution. Tetramethylsilane [$Si(CH_3)_4$, normally called TMS] is chosen because it produces a sharp signal in a region well away from the signals produced by almost all other organic hydrogen atoms. TMS is assigned a chemical shift of zero. For any $^1$H atom, the chemical shift is expressed as the difference between its signal and that of TMS and is denoted by the symbol $\delta$ (delta). Notice that the units of $\delta$ are parts per million, emphasising how small the effect is.

### *Interpreting NMR spectra*

In an organic molecule, different hydrogen atoms will be in different environments and have different chemical shifts. Each different environment will give rise to a different signal, e.g.
- in benzene, $C_6H_6$, all the hydrogen atoms are identical and only one peak appears in the spectrum.
- in ethanal, $CH_3CHO$, there are two different kinds of environment for the hydrogen atoms (the methyl group and the aldehyde group) and two peaks are produced.

Figure 117 shows the low-resolution NMR spectrum for ethanol.

*Figure 117*
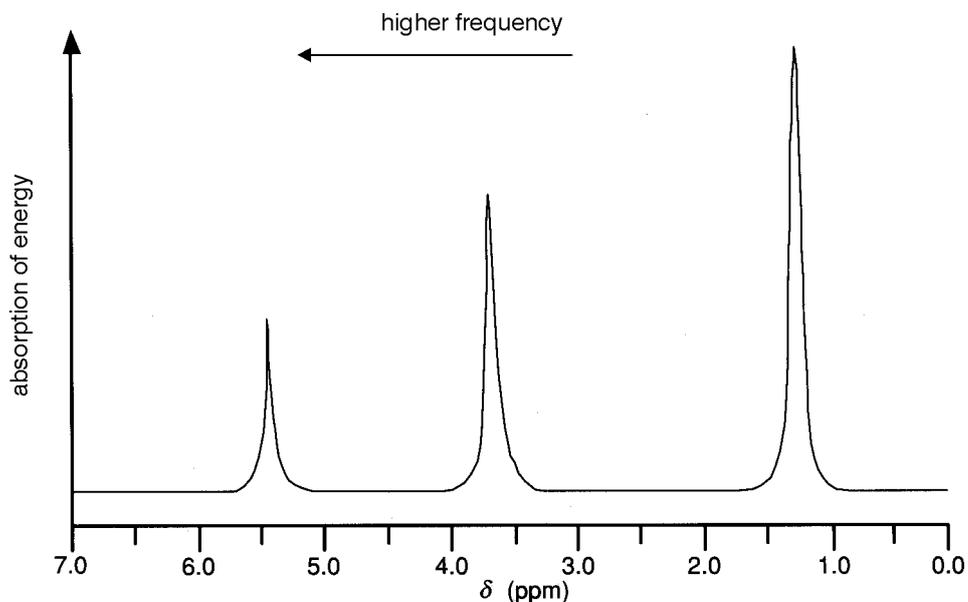


There are three peaks because there are three different environments for the $^1$H atoms in ethanol. The peak at about 1.2 $\delta$ is caused by the $^1$H atoms in the methyl group ($CH_3$). The peak at about 3.7 $\delta$ is caused by the hydrogen atoms in the $CH_2$ group. The smallest peak at about 5.4 $\delta$ is caused by the $^1$H atom of the hydroxyl group (OH).

Tables are available which show the chemical shifts of $^1$H atoms in different environments, e.g. page 15 of the Data Booklet. If the NMR spectrum of an unknown substance is produced, the chemical shifts of the peaks can be compared with the values in the tables to predict the different environments for the $^1$H atoms in the sample.

However, the spectrum can provide yet more information. Look again at the spectrum for ethanol (Figure 117). The peaks are clearly different in size. In fact, the area under each peak is proportional to the number of $^1$H atoms causing the absorption. In an ethanol molecule, there are three identical hydrogen atoms in the methyl group but only one hydrogen atom in the hydroxyl group. Consequently, the area under the methyl peak (1.2 $\delta$ ) is three times the area under the hydroxyl peak (5.4 $\delta$ ). Estimating areas is not easy so in practice an integration curve is plotted on the NMR spectrum (see Figure 118).

*Figure 118*
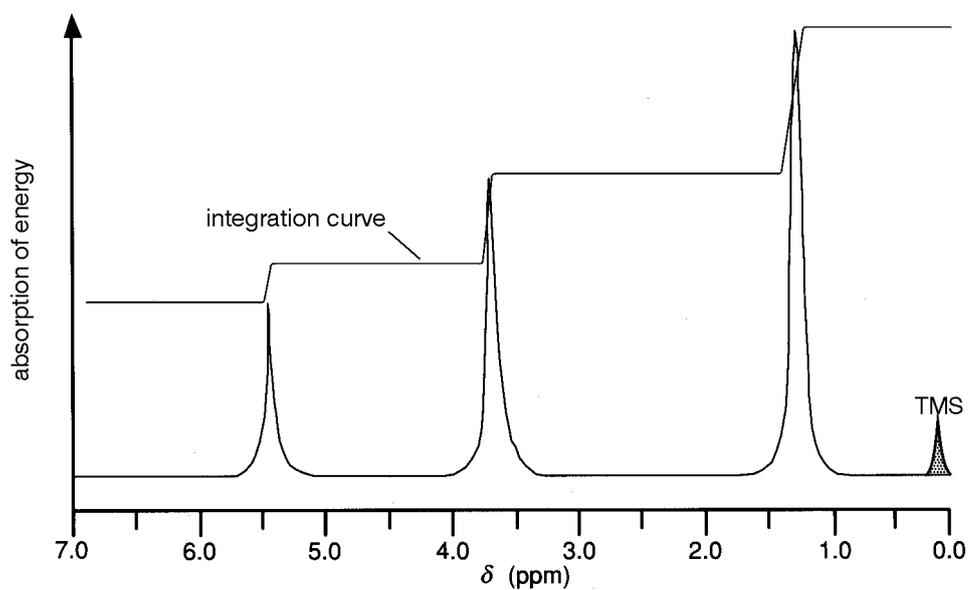


By measuring the height of the step in the integration curve for each signal, the ratio of $^1H$ atoms can be determined. Usually the spectrometer's computer will give a numerical printout of the area under each peak. As well as giving information about the number of different environments for the $^1H$ atoms in a molecule, an NMR spectrum therefore gives information about the ratio of the number of hydrogen atoms in each environment.

Under higher resolution, the peak arising from a particular hydrogen atom may be split into a double, triple or multiple peak. This provides a great deal of information about the structure of the molecule but is beyond the scope of Advanced Higher.
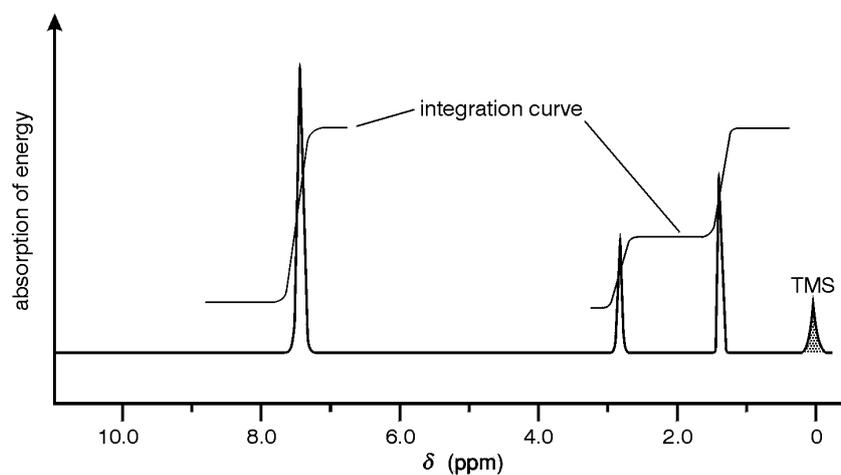
**Questions**

From the following low-resolution NMR spectra and the other information given, suggest a possible structure for each substance.
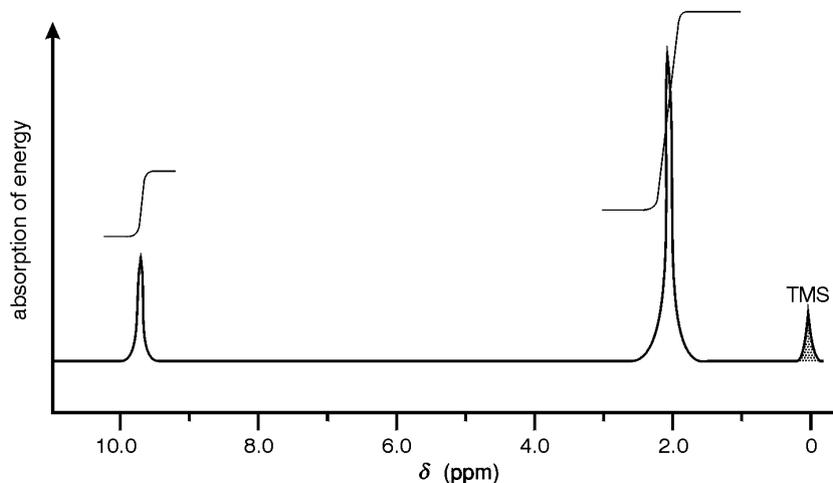
1.  Figure 119 is the spectrum for a hydrocarbon.

*Figure 119*



2.  Figure 120 is the spectrum produced by a compound of molecular formula, $C_2H_4O$.

*Figure 120*



3.  Analysis of a sweet-smelling, neutral compound of carbon, hydrogen and oxygen produced the following results: %C = 54.5; %H = 9.1.

From its mass spectrum, the molecular ion had a mass/charge ratio of 88.

Its infra-red spectrum showed a prominent peak at 1735 cm$^{-1}$.

Figure 121 shows the NMR spectrum of the compound.

***Figure 121***
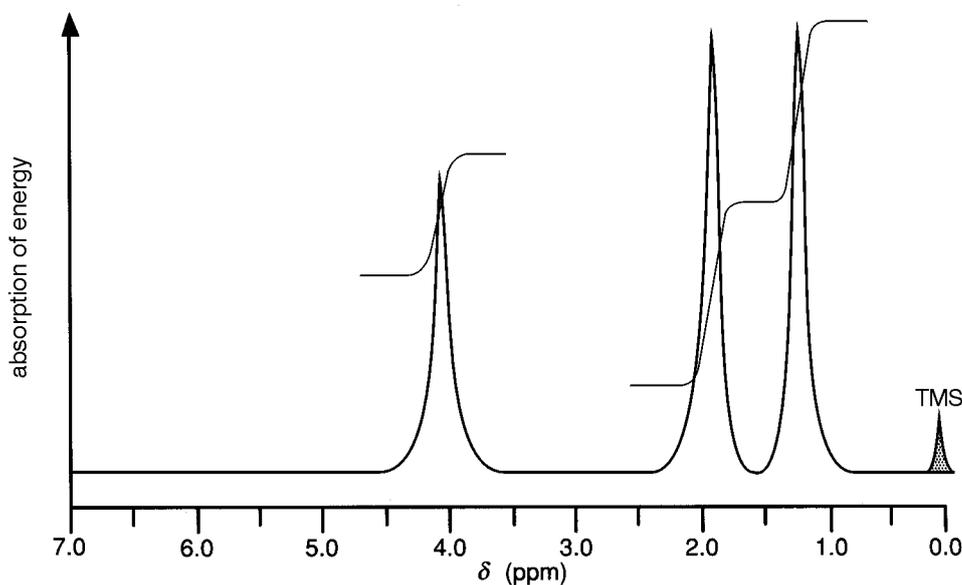


## X-ray crystallography

X-ray crystallography has developed over the last 50 years into one of the most powerful tools for determining the precise three-dimensional structure of organic compounds. Easy access to computers and electronic circuitry has transformed this technique, which gained Professor Dorothy Hodgkin the Nobel Prize in chemistry in 1964 for determining the structure of Vitamin B12. This is a very complex molecule of formula $CoC_{63}H_{88}N_{14}O_{14}P$.

The technique is based on very complex mathematical equations, which are beyond the scope of this course. Modern instruments are linked to computers and produce a large range of data very accurately and relatively quickly. A very simple explanation of the technique follows.

Organic crystals consist of an orderly arrangement of molecules in which the spacing of the atoms in the molecules is similar in size to the wavelength of X-rays (around 100 pm).

When a beam of X-rays of a single wavelength is passed through a crystal, the crystal acts as a three-dimensional diffraction grating causing the X-ray beam to be transmitted as a number of diffracted beams. A combination of diffraction and interference gives a spot spectrum (Figure 122).

*Figure 122*
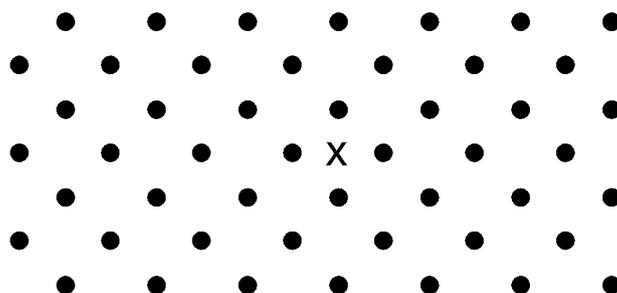


The position of the spots depends on the nature of the atoms, or more precisely the electrons in the atoms, which make up the molecules in the crystal. Thus the spectrum depends on the relative positions of the atoms in space.

From the position and intensity of the spots on the spectrum a computer can produce an electron-density map (Figure 123). This map shows contours of equal electron density.

*Figure 123*
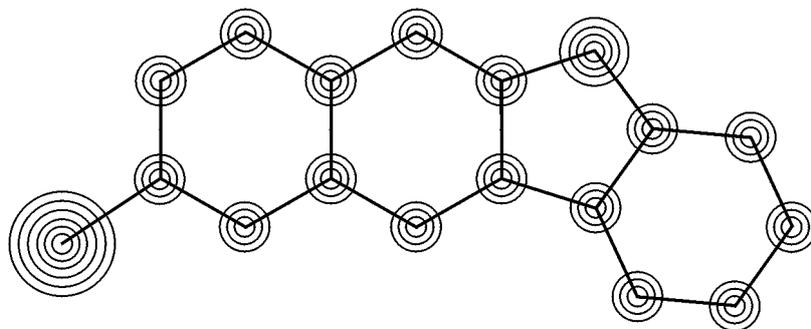


Such a map gives information about:
  • the structure of the organic molecule
  • the precise location of one atom relative to another
  • the type of atom.

The heavier the atom, the more electrons it has, therefore the greater the electron density. In fact hydrogen has such a low electron density that it is not easily detected by X-ray analysis. Figure 123 shows a simplified electron-density map of a complex organic molecule (Figure 124).

*Figure 124*
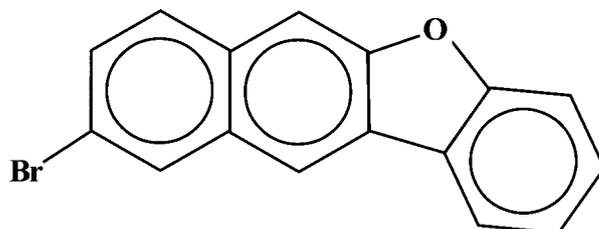


It is clear where the oxygen and bromine atoms are situated, as well as the carbon atoms. However, the position of the hydrogen atoms cannot be deduced from an electron-density map.

Figure 125 shows a simulated electron-density map in which all atoms are identifiable, even the hydrogen atoms.
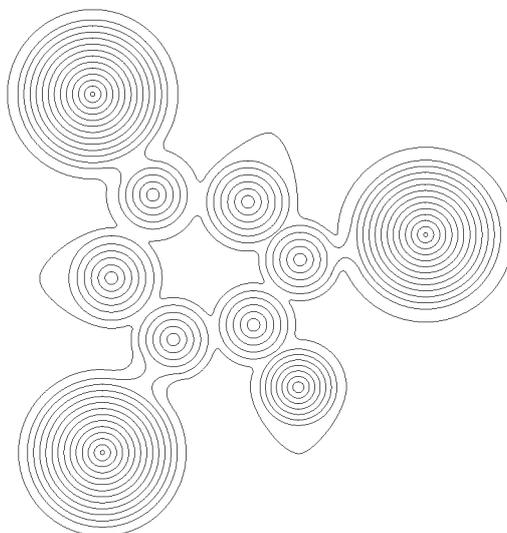
*Figure 125*



**Question**

Figure 125 is the simulated electron density map of an aromatic compound with molecular formula $C_6H_3Cl_3O$.

(a)   Draw the full structural formula for this compound.
(b)   Explain why you drew the structure in terms of electron density.
(c)   Draw an electron density map for methanoic acid.