

Measurement, Dimensions and Units

Standards of Comparison

The US standard railroad gauge (distance between the rails) is 4 feet, 8.5 inches. That's a very strange number, why was it used? Because the first railroads were built in Britain, and the North American railroads were built by British immigrants.

Why did they build them like that? Because the first railways (lines and rolling stock) were built by the same companies that built the pre-railroad tramways, and they used the same old gauge. All right, why did 'they' use that gauge? Because the tramways used the same jigs and tools that had been used for building wagons, and the wagons used that wheel spacing.

Are we getting anywhere? Why did the wagons use that strange wheel spacing? Well, if they tried to use any other spacing the wagons would break down on some of the old long distance roads, because that's the spacing of the old wheel ruts.

So who built these old rutted roads? The first long distance roads in Europe were built by Imperial Rome for the purposes of the Roman Legions. These roads were still widely used in the 19th century. And the ruts? The initial ruts, which everyone else had to match in case they destroyed their wagons, were made by Roman war chariots. Since the chariots were made for Imperial Rome they were all alike, including the wheel spacing. So now we have an answer to the original question. The US standard railroad gauge of 4 feet, 8.5 inches is derived from the original specification for an Imperial Roman army war chariot.

The next time you are struggling with conversion factors between units and wonder how we ended up with all this nonsense, you may be closer to the truth than you knew. Because the Imperial Roman chariots were made to be just wide enough to accommodate the south ends of two war horses heading north.

And this is not yet the end! The US space shuttle has two big booster rockets attached to the sides of the main fuel tank. These are solid rocket boosters (SRBs) made in a factory in Utah. It has been alleged that the engineers who designed the SRBs would have preferred to make them a bit fatter, but the SRBs had to be shipped by train from the factory to the launch site. The railroad line from the factory happens to run through a tunnel in the mountains, and the SRBs had to fit through that tunnel. The tunnel is only slightly wider than the railroad track, and we now know the story behind the width of the track!

So, limitations on the size of crucial components of the space shuttle arose from the average width of the Roman horses' rear ends.

1.1 Introduction

All quantitative measurements are really comparisons between an unknown quantity (such as the height of a person) and a measuring instrument of some kind (e.g., a measuring tape). But to be able to communicate the results

of our measurements among one another we have to agree on exactly what we are comparing our measurements to. If I say that I measured my height and the reading on the tape was 72, that does not tell you much. But if I say the value was 72 inches, that does provide some meaningful

information provided that you know what an inch is (tradition tells us that the inch was originally defined as the length of part of the thumb of some long-forgotten potentate but that does not help us much). But even that information is incomplete as we do not know the uncertainty in the measurement. Most people understand in a general way the concepts of accuracy (deviation of the measured value from the 'true' value) and precision (a measure of how close is the agreement among repeated measurements of the same quantity) as different aspects of total uncertainty, and such a general understanding will suffice for the first few chapters of this book. However, the result of a measurement without an accompanying estimate of its uncertainty is of little value, and a more complete discussion of experimental uncertainty is provided in Chapter 8 in preparation for the practical discussions of Chapters 9 and 10.

Actually, the only correct answer to the question 'what is an inch' is that one inch is defined as exactly 2.54 centimeters (zero uncertainty in this defined conversion factor). So now we have to ask what is a centimeter, and most of us know that a centimeter is 1/100 of a meter. So what is a meter? This is starting to sound about as arbitrary as the Roman horses' hind quarters mentioned in the text box but in this case we can give a more useful

if less entertaining answer: The meter is the length of the path traveled by light in vacuum during a time interval of $1/299\,792\,458$ of a second. Note that the effect of this definition is to fix the speed of light in vacuum at exactly $299\,792\,458$ meters per second, and that we still have not arrived at a final definition of the meter until we have defined the second (Table 1.1). This is the internationally accepted definition of the meter, established in 1983, and forms part of the International System of Units (Système Internationale d'Unites, known as SI for short). The SI establishes the standards of comparison used by all countries when the measured values of physical and chemical properties are reported. Such an international agreement is essential not only for science and technology, but also for trade. For example, consider the potential confusion arising from the following example:

- 1 US quart (dry) = 1.10122 litres
- 1 US quart (liquid) = 0.94635 litres
- 1 Imperial (UK/Canada) quart (liquid) = 1.136523 litres

(The litre is defined in the SI as $1/1000$ of a cubic meter: $1\text{L} = 10^{-3}\text{m}^3$). Many other examples of such ambiguities can be given (see, for example, the unit conversions

Table 1.1 SI Base Quantities and Units

Quantity	Name of unit	Symbol	Definition
Length	meter	m	The meter is the length of the path travelled by light in vacuum during a time interval of $1/299\,792\,458$ of a second.
Mass	kilogram	kg	The kilogram is the unit of mass; it is equal to the mass of the international prototype of the kilogram.
Time	second	s	The second is the duration of $9\,192\,631\,770$ periods of the radiation corresponding to the transition between the two hyperfine levels of the ground state of the caesium 133 atom.
Electric current	ampere	A	The ampere is that constant current which, if maintained in two straight parallel conductors of infinite length, of negligible circular cross-section, and placed one meter apart in vacuum, would produce between these conductors a force equal to 2×10^{-7} newton per meter of length.
Thermodynamic temperature	kelvin	K	The kelvin, unit of thermodynamic temperature, is the fraction $1/273.16$ of the thermodynamic temperature of the triple point of water.
Amount of substance	mole	mol	The mole is the amount of substance of a system which contains as many elementary entities as there are atoms in 0.012 kilogram of carbon 12; its symbol is 'mol.' When the mole is used, the elementary entities must be specified and may be atoms, molecules, ions, electrons, other particles, or specified groups of such particles.
Luminous intensity	candela	cd	The candela is the luminous intensity, in a given direction, of a source that emits monochromatic radiation of frequency 540×10^{12} hertz and that has a radiant intensity in that direction of $1/683$ watt per steradian.

at: <http://www.megaconverter.com/Mega2/index.html>). Such discrepancies may not seem to be very important when only a single quart is considered, but in international trade where literally millions of quarts of some commodity might be traded, the 19% difference between the two definitions of the liquid quart could lead to extreme difficulties if the ambiguity were not recognized and taken into account. In a lecture on ‘Money as the measure of value and medium of exchange’, delivered in 1763 at the University of Glasgow, Adam Smith commented (Smith 1763, quoted in Ashworth 2004):

‘Natural measures of quantity, such as fathoms, cubits, inches, taken from the proportion of the human body, were once in use with every nation. But by a little observation they found that one man’s arm was longer or shorter than another’s, and that one was not to be compared with the other; and therefore wise men who attended to these things would endeavour to fix upon some more accurate measure, that equal quantities might be of equal values. Their method became absolutely necessary when people came to deal in many commodities, and in great quantities of them.’

It is precisely this kind of uncertainty that the SI is designed to avoid in both science and in trade and commerce. In this regard it is unfortunate to note that even definitions of words used to denote numbers are still subject to ambiguity. For example, in most countries ‘one billion’ (or the equivalent word in a country’s official language) is defined as 10^{12} (a million million), but in the USA (and increasingly in other English-speaking countries) a billion is used to represent 10^9 (a thousand million) and 10^{12} is referred to as a ‘trillion’. In view of this ambiguity it is always preferable to use scientific numerical notation.

1.2 The International System of Units (SI)

An excellent source of information about the SI can be found at the website of the US National Institute for Standards and Technology (NIST): <http://physics.nist.gov/cuu/Units/index.html>

Here we shall be mainly concerned with those quantities that directly affect quantitative measurements of amounts of chemical substances by mass spectrometry. However, it is appropriate to briefly describe some general features of the SI.

Early History of the SI

There is a strong French connection with the SI, including its name and the location in Paris of the central organization that coordinates this international agreement (Bureau International des Poids et Mesures, or BIPM), and the international guiding body CIPM (Comité International de Poids et Mesures, i.e., International Committee for Weights and Measures). This connection was established at the time of the French Revolution when the revolutionary government decided that the chaotic state of weights and measures in France had to be fixed. The intellectual leader in this initiative, that resulted in the so-called Metric System, was the chemist Antoine Lavoisier, famous for his demonstration that combustion involves reaction with oxygen and that water is formed by combustion of two parts of hydrogen with one of oxygen. His efforts resulted in the creation of two artifacts



Antoine Lavoisier

made of platinum (chosen because of its resistance to oxidation), one representing the meter as the new unit of length between two scratch marks on the platinum bar, and the other the kilogram. These artifacts were housed in the Archives de la République in Paris in 1799, and this represents the first step taken towards establishment of the modern SI.

Sadly, Lavoisier did not live to see this realization of his ideas. Despite his fame, and his services to science and his country (he was a liberal by the standards of pre-revolutionary France and played an active role in the events leading to the Revolution and, in its early years, formulated plans for many reforms), he fell into disfavour because of his history as a former farmer-general of taxes, and was guillotined in 1794. After his arrest and a trial that lasted less than a day, Lavoisier requested postponement of his execution so that he could complete some experiments, but the presiding judge infamously refused: ‘L’état n’a pas besoin de savants’ (the state has no need of intellectuals).

Any system of measurement must decide what to do about the fact that there are literally thousands of physical properties that we measure, each of which is expressed as a measured number of some well-defined unit of measurement. It would be impossible to set up primary standards for the units of each and every one of these thousands of physical quantities, but fortunately there is no need to do so since there are many relationships connecting the measurable quantities to one another. A simple example that is of direct importance to the subject of this book is that of volume; as mentioned above, the SI unit of volume (cubic meter) is simply related to the SI unit for length via the physical relationship between the two quantities. So the first question to be settled concerns how many, and which, physical quantities should be defined as SI base quantities (sometimes referred to as dimensions), for which the defined units of measurement can be combined appropriately to give the SI units for all other measurable quantities.

At one time it was thought to be more 'elegant' to work with a minimum possible number of dimensions and their defined units of measurement, and this pseudo-esthetic criterion gave rise to the three-dimensional centimeter-gram-second (cgs) and meter-kilogram-second (MKS) systems. However, it soon became apparent that utility and convenience were more important than perceived elegance! As a simple example, consider Coulomb's Law for the electrostatic force F between two electric charges q_1 and q_2 separated by a distance r in a vacuum:

$$F = k_o \cdot q_1 \cdot q_2 / r^2$$

In the simple form of Coulomb's Law as used with the cgs system, the Coulomb's Law Constant k_o is treated as a dimensionless constant with value 1. (This is not the case

in the SI, where $k = 1/(4\pi\epsilon_o)$ where ϵ_o is the permittivity of free space $= 8.854187817 \times 10^{-12} \text{ s}^4 \text{ A}^2 \text{ kg}^{-1} \text{ m}^{-3}$). By Newton's Second Law of Motion, force is given as (mass \times acceleration), i.e., (mass \times length \times time⁻²), so in the cgs system $q_1 \cdot q_2$ corresponds to (mass \times length³ \times time⁻²); thus, in such a three-dimensional measurement system, electrical charge q corresponds to (mass^{1/2} \times length^{3/2} \times time⁻¹). This very awkward (and inelegant!) result involving fractional exponents becomes even more cumbersome when magnetism is considered. Once it was accepted that usefulness was the only criterion for deciding on the base physical quantities (dimensions) and their units of measurement, it was finally agreed that the most useful number of dimensions for the SI was seven. Some of these seven are of little or no direct consequence for this book, but for the sake of completeness they are all listed in Table 1.1. Some important SI units, that are derived from the base units but have special names and symbols, are listed in Table 1.2.

The two base quantities (and their associated SI units) that are most important for quantitative chemical analysis are amount of substance (mole) and mass (kilogram), although length (meter) is also important via its derived quantity volume in view of the convenience introduced by our common use of volume concentrations for liquid solutions. (Note, however, that the latter will in principle vary with temperature as a result of expansion or contraction of the liquid).

The kilogram is unique among the SI base units for two reasons. Firstly, the unit of mass is the only one whose name contains a prefix (this is a historical accident arising from the old centimeter-gram-second system of measurement mentioned above). Names and symbols for decimal multiples and submultiples of the unit of mass are formed

Table 1.2 Some SI Derived units with special names and symbols^a

Derived quantity	Name of unit	Symbol	Relationship to SI base units	Relationship to other SI units
Plane angle	radian	rad	$\text{m} \cdot \text{m}^{-1} (= 1)$	—
Solid angle	steradian	sr	$\text{m}^2 \cdot \text{m}^{-2} (= 1)$	—
Frequency	hertz	Hz	s^{-1}	—
Force	newton	N	$\text{m} \cdot \text{kg} \cdot \text{s}^{-2}$	—
Pressure	pascal	Pa	$\text{m}^{-1} \cdot \text{kg} \cdot \text{s}^{-2}$	$\text{N} \cdot \text{m}^{-2}$
Energy	joule	J	$\text{m}^2 \cdot \text{kg} \cdot \text{s}^{-2}$	$\text{N} \cdot \text{m}$; $\text{Pa} \cdot \text{m}^3$
Power	watt	W	$\text{m}^2 \cdot \text{kg} \cdot \text{s}^{-3}$	$\text{J} \cdot \text{s}^{-1}$
Electric charge	coulomb	C	A.s	—
Electric potential difference	volt	V	$\text{m}^2 \cdot \text{kg} \cdot \text{s}^{-3} \cdot \text{A}^{-1}$	$\text{W} \cdot \text{A}^{-1}$
Electric resistance	ohm	Ω	$\text{m}^2 \cdot \text{kg} \cdot \text{s}^{-3} \cdot \text{A}^{-2}$	$\text{V} \cdot \text{A}^{-1}$
Magnetic flux density	tesla	B	$\text{kg} \cdot \text{s}^{-2} \cdot \text{A}^{-1}$	—
Celsius temperature	degree Celsius	$^{\circ}\text{C}$	K^{b}	—

^a for a complete list and discussion, see Taylor (1995) and Taylor (2001).

^b the size of the two units is the same, but Celsius temperature ($^{\circ}\text{C}$) = thermodynamic temperature (K) – 273.15 (the ice point).

by attaching prefix names to the unit name ‘gram’ and prefix symbols to the unit symbol ‘g’, not to the ‘kilogram’. (A list of SI prefixes denoting powers of 10 is given in Table 1.3). The other unique aspect of the kilogram is that it is currently (2007) the only SI base unit that is defined by a physical artifact, the so-called international prototype of the kilogram (made of a platinum–iridium alloy and maintained under carefully controlled conditions at the BIPM in Paris (Figure 1.1)). This international prototype is used to calibrate the national kilogram standards for the countries that subscribe to the SI.

Table 1.3 SI Prefixes

Factor	Name	Symbol	Factor	Name	Symbol
10^{24}	yotta	Y	10^{-1}	deci	d
10^{21}	zetta	Z	10^{-2}	centi	c
10^{18}	exa	E	10^{-3}	milli	m
10^{15}	peta	P	10^{-6}	micro	μ
10^{12}	tera	T	10^{-9}	nano	n
10^9	giga	G	10^{-12}	pico	p
10^6	mega	M	10^{-15}	femto	f
10^3	kilo	k	10^{-18}	atto	a
10^2	hecto	h	10^{-21}	zepto	z
10^1	deka	da	10^{-24}	yocto	y

By convention, multiple prefixes (e.g., dekakilo) are not allowed. Thus in the case of the SI unit of mass (kilogram), that for historical reasons contains a prefix in its name, the SI prefix names are used with the unit name ‘gram’ and the prefix symbols with the corresponding symbol ‘g’.

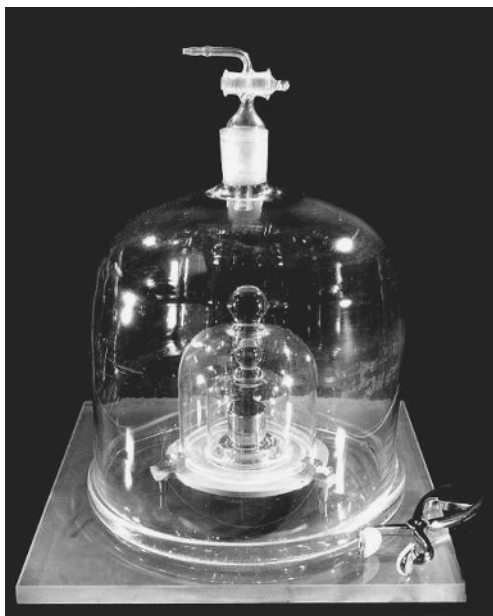


Figure 1.1 The International prototype of the kilogram.

The addition of amount of substance as the seventh SI base unit, the ‘chemical’ unit, was achieved only after considerable dispute between chemists and physicists (McGlashan 1970), and was officially adopted only in 1971 about 17 years after adoption of the ampere, the kelvin and the candela (Table 1.1). Essentially the physicists felt that mass was a perfectly adequate quantity for all quantitative chemical purposes, since for all practical purposes mass is conserved in chemical reactions. Note that this can not be exactly correct since chemical reactions involve energy changes, e.g., energy loss in the case of exothermic reactions, and this energy corresponds to a change in mass via Einstein’s famous relationship $E = mc^2$. However, for a typical reaction enthalpy of $10^5 \text{ J} \cdot \text{mol}^{-1}$, the corresponding change in mass is given as:

$$\Delta m \sim 10^5 \text{ J} / (3 \times 10^8 \text{ ms}^{-1})^2 \sim 10^{-12} \text{ kg}$$

A good laboratory balance can measure mass routinely to within 10^{-7} kg , and use of a microbalance with considerable precautions can lead to mass measurements to within 10^{-9} kg or so (Section 2.3). This is still three orders of magnitude larger than the mass changes equivalent to heats of reaction, so the physicists’ argument is valid from this point of view. (Note that the above calculation of Δm exemplifies an important property of the SI, its coherence, by which we mean that if all quantities in a formula are expressed in SI units without prefixes the result of the calculation is also expressed in the appropriate SI unit, with no need for conversion factors).

However, the guiding principle in choice of the base quantities in any measurement system is that of usefulness and convenience, and since chemistry involves interactions among individual discrete molecules it is simply commonsense to adopt a quantity (and a corresponding unit) that reflects this reality.

The definition of the mole (Table 1.1) refers to the number of atoms in 0.012 kilogram of carbon 12 (^{12}C). This number is the Avogadro Constant $N_A = 6.0221479 (\pm 0.0000030) \times 10^{23} \text{ mol}^{-1}$, formerly known as ‘Avogadro’s Number’ but now in the SI not a dimensionless number but a quantity that must be expressed in SI units; the Avogadro Constant defines the number of molecules of a compound in 1 mol of that compound. Since different molecules interact chemically on the basis of small integral numbers of each type, it makes sense on a purely utilitarian basis to define such a base quantity and a corresponding unit, e.g., since one milligram of glucose contains 30 times as many molecules as one milligram of insulin, it makes no *physical* sense to discuss *chemical* interactions between these two compounds in terms of mass only!

The Mole and the Avogadro Constant

An interesting historical account of the origins of the concept and the name of the mole has been published by Gorin (1994).

Definition of base quantities and their respective units is a serious business, absolutely necessary for the unambiguous sharing of quantitative experimental data among scientists and engineers around the world. However, it can become a somewhat dry and even boring subject for chemists, who are never reluctant to look for ways to spice up their professional business with a little self-deprecating humour.

For example, the amazing reduction in detection limits for mass spectrometry that has been possible over the last 20 years or so has led to a proposal that a new SI prefix (see Table 1.3) will be required before long. The name of the proposed new prefix is the guaco, referring to a factor of 10^{-25} , since ambitions for guacamole sensitivity were thought to be a suitable target for instrument designers. However, this proposal was abandoned when it was realized that guacamole sensitivity was intrinsically impossible as a result of the value of the Avocado Constant.

Slightly less nonsensical is the introduction of 'Mole Day', created as a way to foster interest in chemistry. Schools throughout the United States of America and around the world celebrate Mole Day on October 23 from 6:02 a.m. to 6:02 p.m., to commemorate the Avogadro Constant (6.02×10^{23}), with various activities related to chemistry and/or moles (see www.moleday.org).

However, the physicists did have a point in their argument with respect to the importance of measurements of mass in how chemists actually set about performing quantitative analyses of amount of substance, and this will be discussed in Section 2.3.

1.3 'Mass-to-Charge Ratio' in Mass Spectrometry

How does the foregoing discussion relate to the so-called mass-to-charge ratios, universally denoted as m/z , that are used to mark the abscissa of a mass spectrum? It must be emphasized that the question of the meaning of the quantity m/z appears to be highly contentious among mass spectrometrists, and the following discussion represents only the best efforts of the present writers to devise a self-consistent interpretation that will be used where appropriate in the rest of the book.

In this book ' m/z ' is best regarded as a three-character symbol, not a mathematical operation. Although no units are ever given for m/z in published spectra, within this three-character symbol ' m ' does indeed denote the mass of a single atom or molecule that has been transformed into an ion so that it is amenable to analysis by the mass spectrometer. For purposes of mass spectrometry, however, it is convenient to not use the kilogram as the unit of mass, but instead to express the mass in terms of the unified atomic mass unit u (or sometimes m_u) defined as:

$$u(\text{or } m_u) = (\text{mass in kilograms of one atom of } ^{12}\text{C})/12$$

Since the symbol ' u ' is used for other purposes in this book, ' m_u ' will be used in the following discussion. The connection of this unit of mass, convenient for expressing masses of single molecules, to the definition of the mole and of the Avogadro Constant, is:

$$12 \times m_u(\text{kg}) \times N_A(\text{mol}^{-1}) = 0.012 \text{ kg mol}^{-1}$$

whence

$$m_u(\text{or } u) = 0.001 / (6.02214179 \times 10^{23}) \text{ kg} = 1.660538782 \\ (\pm 0.000000083) \times 10^{-27} \text{ kg}$$

Values of fundamental constants like m_u , N_A , etc., together with related information concerning their relationships, can be found at <http://physics.nist.gov/cuu/Constants/index.html>. Values of m for all isotopes of the elements (including radioactive nuclides) are constantly being refined; the International Union of Pure and Applied Chemistry (IUPAC) publishes frequent revised tables of isotope-averaged atomic weights (Loss 2003), and extensive updated information on individual isotopic masses can be found at <http://ie.lbl.gov/toimass.html>, while similarly updated information on natural isotopic abundances (Rosman 1998) is also available at www.iupac.org/reports/1998/7001rosman/iso.pdf. Such detailed high-precision information is not usually important for the kinds of measurements discussed in this book. Lists of atomic masses and isotope abundances of adequate quality for trace chemical analysis are available from several sources, e.g., http://physics.nist.gov/cgi-bin/Compositions/stand_alone.pl?ele=&ascii=html&isotope=some where the elements are listed in order of atomic number, and

Table 1.4 Relative atomic masses and relative abundances of some stable isotopes

Element (Atomic number)	Isotope	Relative atomic mass	Abundance (%)	Isotope-averaged atomic weight
H (1)	H	1.00782503	99.9885	1.00794
	² H (D)	2.01410178	0.0115	
B (5)	¹⁰ B	10.0129370	19.9	10.81
	¹¹ B	11.0093055	80.1	
C (6)	¹² C	12.0000000 (by definition)	98.93	12.0107
	¹³ C	13.0033548	1.08	
N (7)	¹⁴ N	14.0030740	99.632	14.0067
	¹⁵ N	15.0001089	0.368	
O (8)	¹⁶ O	15.9949146	99.757	15.9994
	¹⁷ O	16.9991315	0.038	
	¹⁸ O	17.9991605	0.205	
F (9)	¹⁹ F	18.9984032	100	18.9984032
Na (11)	²³ Na	22.9897696	100	22.9897696
Si (14)	²⁸ Si	27.9769265	92.2297	28.0855
	²⁹ Si	28.9764947	4.6832	
	³⁰ Si	29.9737702	3.0872	
P (15)	³¹ P	30.9737615	100	30.9737615
S (16)	³² S	31.9720707	94.93	32.065
	³³ S	32.9714585	0.76	
	³⁴ S	33.9678668	4.29	
	³⁶ S	35.9670809	0.02	
Cl (17)	³⁵ Cl	34.9688527	75.78	35.453
	³⁷ Cl	36.9659026	24.22	
K (19)	³⁹ K	38.9637069	93.2581	39.0983
	⁴⁰ K	39.9639987	0.0117	
	⁴¹ K	40.9618260	6.7302	
Br (35)	⁷⁹ Br	78.9183376	50.69	79.904
	⁸¹ Br	80.9162913	49.31	
I (53)	¹²⁷ I	126.9044684	100	126.9044684

(Note that the relative abundances are terrestrial averages; small deviations (a few parts per thousand) contain information that is valuable in several fields of science, and are measured using Isotope Ratio Mass Spectrometry.)

at www.sisweb.com/referenc/source/exactmaa.htm where the elements are listed in alphabetical order. An abbreviated list covering the elements of most interest for organic analyses in this book is given in Table 1.4.

It is important to note that m_u is the convenient unit adopted to express the mass of one unique molecule containing specified numbers of isotopes of the elements (e.g., $^{12}\text{C}^{35}\text{Cl}^{37}\text{Cl}_2^1\text{H}$ for an isotopically specified form of chloroform). The m_u unit is conventionally not used for the average mass of the molecules of the same compound, still specified as the same numbers of atoms of the elements (CCl_3H for chloroform), but now assuming the various isotopic distributions to be the average values observed on the surface of our planet Earth. (Such

quantities have been referred to in the past as the ‘molecular weight’ of the compound, but this usage can be misleading because the ‘weight’ of an object is by definition the gravitational force on that object, i.e., it depends on both the mass of the object and its position in space). Chemists and biochemists have in the past used the dalton (Da) as an atomic mass unit derived from m_u but adjusted for each element according to the average isotopic distribution of that element. This is not an official SI unit but is useful because it directly relates the mass of a macroscopic sample of a real natural compound, determined by weighing (see Section 2.3), to the molecular formula of the compound. Of course this relationship does not hold for a variant of the compound in which one or more

of the atoms have been synthetically specified with a non-natural isotopic composition, for use as an internal standard in isotope dilution approaches, as discussed in Section 2.2.3). However, in actual practice the dalton is increasingly being used by mass spectrometrists as an alternative to the ‘unified atomic mass unit’ m_u (or u), and usually the context makes clear which interpretation of this chemical unit (i.e., the isotopic specification) is intended.

In summary, the quantity m (italicized) in m/z is related to the mass m (in kilograms) of a single isotopically-specified (ionized) molecule, and is strictly defined as:

$$m = m(\text{kg})/m_u(\text{kg})$$

where m is the actual mass of one of the specified molecules; m can thus be regarded as a dimensionless quantity (ratio) that requires no units, although it is intimately related to the kilogram via the Avogadro Constant and m_u .

Just as for the mass of an ion, the charge on the ion is conveniently expressed not in terms of the SI unit for electric charge, i.e., the Coulomb (C, see Table 1.2), but relative to the elementary charge (e), one of the fundamental physical constants and equal to the magnitude of the charge (i.e. without sign) on the electron:

$$e = 1.602176487 (\pm 0.000000040) \times 10^{-19} \text{C}$$

Then, the quantity z (italicized) in m/z is the number of elementary charges on the ion (usually quoted irrespective of sign as the context almost always makes the latter clear):

$$z = \text{magnitude of charge on the ion (C)} / e(\text{C})$$

and is thus also, like m , a dimensionless number!

Thus, the ‘mass-to-charge ratio’ of mass spectrometry, conveniently denoted by the symbol m/z , is a dimensionless ratio of two dimensionless quantities that is nonetheless intimately related to both the kilogram (via m_u) and to the coulomb (via e). It is possible that the ‘ m ’ in ‘ m/z ’ could be misinterpreted as the same (but nonitalicized) symbol used in the SI to represent the base quantity ‘mass’ and possibly also the SI symbol for the meter (unit of length)! The letter ‘ m ’ is greatly overused in metrological notation! However, in the mass spectrometric sense, ‘ m ’ NEVER appears without ‘ $/z$ ’, so it seems that the context should never give rise to ambiguity or confusion. In fact, as mentioned above, the notation ‘ m/z ’ is best regarded as a three-character symbol for the dimensionless quantity

defined above, rather than as a mathematical operation on two different quantities. Otherwise the abscissa of a mass spectrum, invariably labeled as m/z (with no units specified!) would be labeled in units of, e.g., kilograms per coulomb (for mass:charge ratio)! In Section 6.2 we shall see how this dimensionless quantity m/z is incorporated into quantitative calculations of physical quantities that are important in describing the instruments used to separate ions according to their m/z values.

More recently, with the introduction of electrospray ionization (Section 5.3.6) to mass spectrometry, it has become much more common to observe multiply-charged ions with $z > 1$. This does not introduce any fundamental difficulty into the established measurement system for ions, but a question of convenience does arise when describing changes in, or differences between, mass spectra. For example, if it is observed that two mass spectra differ only with respect to one peak that appears at different m/z values, how does one describe the magnitude of the shift? Some authors say that the peak was shifted by ‘ $X m/z$ units’, which is clumsy but does transmit the desired message. It has been suggested (Cooks 1991) that the mass spectrometry community should, purely for convenience, adopt a unit for m/z defined as above, to be called the thomson (Th) in honour of J.J. Thomson; then we could speak of a peak shift by ‘ $X \text{Th}$ ’. This suggestion has not been approved by any international body, but has come into common use simply because it is convenient to do so under circumstances such as those mentioned above. Unfortunately a quantity (not a unit) named the Thomson cross-section (and indeed also named in honor of J.J. Thomson) already exists; this quantity describes the probability that electromagnetic radiation will be scattered by a charged particle, and its unit (symbol σ_e) is the value of this cross section for an electron ($0.665245873 \times 10^{-28} \text{m}^2$, see: http://physics.nist.gov/cgi-bin/cuu/Value?sigmae|search_for=atomnuc!). In the context of mass spectrometry the proposal to name a *unit* (symbol Th) for m/z in honor of Thomson (Cooks 1991) has been criticized on the basis that this would create confusion with the physical quantity (Thomson cross-section, with its own unit σ_e). This seems unlikely given the very different contexts in which the two will appear, quite apart from the fundamental difference between a physical quantity and a unit of measurement. It should also be recalled that convenience is an important criterion in deciding upon details of any measurement system, even the number of base quantities (and thus units) to be adopted! Accordingly the Thomson (Th) is used where appropriate as the unit of m/z in this book.

For convenience a list of some fundamental physical constants is provided in Table 1.5.

Table 1.5 Values of some fundamental physical constants, given to a number of significant figures sufficient for purposes of this book

Physical quantity	Symbol	Value
Elementary charge	e	1.60218 C
Unified atomic mass unit (also Da)	u or m_u	1.66054×10^{-27} kg
Mass of electron	m_e	9.10938×10^{-31} kg
Avogadro Constant	N_A	6.02214×10^{23} mol ⁻¹
Boltzmann Constant	k_B	1.38065×10^{-23} J.K ⁻¹
Gas Constant	$R (= N_A \cdot k_B)$	$8.31447 \text{ J.K}^{-1} \cdot \text{mol}^{-1} = 0.0820575 \text{ L.atm.K}^{-1} \cdot \text{mol}^{-1}$
Planck Constant	h	6.62607×10^{-34} J.s ⁻¹
Permittivity of vacuum	ϵ_0	8.85419×10^{-12} J ⁻¹ .C ² .m ⁻¹
Permeability of vacuum	μ_0	$4\pi \times 10^{-7}$ J.s ² .C ⁻² .m ⁻¹ (or T ² .J ⁻¹ .m ³)

(Note: T = tesla (unit of magnetic flux density, Table 1.2).)

1.4 Achievable Precision in Measurement of SI Base Quantities

The achievable precision in measuring quantities like time, length, mass and the other SI base quantities, and thus in the definitions of their units, is intimately dependent on developments in the technologies used to measure them. Once the recommendations of Adam Smith (see the introductory paragraphs of this chapter) had been adopted, and ‘natural’ rather than ‘anthropological’ units of measurement were sought, early attempts used the size of the earth (the meter was originally defined as the length of a platinum bar designed to be 10^{-7} of the length of a quadrant of the Earth), the rotation rate of the Earth to define 24 hours and thus the second, and the density of water as a link between the meter and the gram. Clearly all of these standards are subject to variation and/or uncertainty, and it became evident that standards based on atomic phenomena would be much more reproducible and constant. For example, James Clerk-Maxwell commented (Clerk-Maxwell, 1890):

‘The earth has been measured as a basis for a permanent standard of length, and every property of metals has been investigated to guard against any alteration of the material standards when made. To weigh or measure anything with modern accuracy, requires a course of experiment and calculation in which almost every branch of physics and mathematics is brought

into requisition. Yet, after all, the dimensions of our earth and its time of rotation, though, relative to our present means of comparison, very permanent, are not so by any physical necessity. The earth might contract by cooling, or it might be enlarged by a layer of meteorites falling on it, or its rate of revolution might slowly slacken, and yet it would continue to be as much a planet as before. But a molecule, say of hydrogen, if either its mass or its time of vibration were to be altered in the least, would no longer be a molecule of hydrogen. If, then, we wish to obtain standards of length, time and mass which shall be absolutely permanent, we must seek them not in the dimensions, or the motion, or the mass of our planet, but in the wavelength, the period of vibration and the absolute mass of these imperishable and unalterable and perfectly similar molecules’.

An excellent review (Flowers 2004) has described the modern advances in achieving this objective. The most spectacular achievements have been in the measurement of time, for which modern cesium atom beam atomic clocks can subdivide time to better than one part in 10^{15} and thus achieve a measurement precision (and thus a definition of the second) of this order (Diddams 2004). As mentioned above, the meter is now defined as the length of the path travelled by light in vacuum during a time interval of *exactly* (1/299 792 458) of a second. The kilogram is still defined in terms of a man-made artifact (Figure 1.1), but efforts are in progress to devise a scheme of measurement and definition that will allow establishment of a unit of mass that is based on some atomic property or perhaps the mass equivalent of energy. It is interesting to note in passing that recent developments (Rainville 2004) in the measurements of cyclotron frequencies of isolated ions (either one or two ions to avoid space-charge effects, see Chapter 6) in a Penning trap have resulted in a precision of better than one in 10^{11} in measurements of ion masses. In addition to the many applications of this technology to fundamental physics (Rainville 2004), measurement precision of this order is only 1–2 orders of magnitude below that required to be able to ‘weigh’ chemical bond strengths to a useful degree of accuracy and precision via $E = m \cdot c^2$!

These spectacular achievements in the precision of physical metrology are to be compared with the levels of precision that can be achieved in, for example, high-throughput measurements of the amounts of a target analyte present at levels of one part in 10^9 – 10^{12} in a complex matrix, a common circumstance faced in laboratories inhabited by readers of this book! In such cases,

within-day and between-day precision of 10% (one in 10^1) would be considered acceptable for organic or speciated inorganic analytes. (The precision can be improved to 1–2% in cases where an isotope-labeled internal standard can be used with isotope dilution mass spectrometry, provided that meticulous precautions are taken as in certification of a reference material for example, see Section 2.2.2). At first sight this performance of chemical metrology would appear to be miserable compared with that of our physicist colleagues.

A first perspective on this enormous discrepancy can be found by considering the problems in measuring another physical base quantity, temperature. The definition of the kelvin, the unit of thermodynamic temperature (Table 1.1), is the fraction (1/273.16) of the

thermodynamic temperature of the triple-point of water (the unique temperature at which all three phases – solid, liquid and vapor – can co-exist, see Section 4.3.2e). The implied measurement precision of ~ 1 in 10^5 is far below that currently achievable with respect to time/frequency, for example. A major reason for this is the intrinsic complexity of chemistry! The principal barrier to improving this situation, based on the current definition, lies in uncertainties in the chemical purity and isotopic composition of the water sample used (Flowers 2004). Such complexity, that is part and parcel of chemical science, is somewhat alien to many physicists as exemplified in an extreme form by the so-called polywater scandal (see the accompanying text box).

Chemical Complexity in Physics: the Polywater Story

In the early 1960s a Soviet physicist, N.N. Fedyakin, discovered what he believed to be a new form of water with anomalous properties, e.g., higher boiling point, lower freezing point, higher viscosity, compared with ‘normal’ liquid water. In 1966 a senior colleague from a prestigious institute in Moscow, B.V. Derjaguin, presented a lecture on this ‘anomalous water’ at the Discussions of the Faraday Society in the United Kingdom.

Over the next several years, hundreds of papers from around the world, describing the properties of what soon came to be known as ‘polywater’, appeared in the scientific literature. Theorists developed models, supported by some experimental measurements, in which strong hydrogen bonds were causing water to polymerize. There were even some who expressed concerns that, if polywater escaped from the laboratory, it could autocatalytically polymerize all of the world’s water. It is interesting that the famous American author Kurt Vonnegut published his novel *Cat’s Cradle* (Vonnegut 1963) in this same time period. In this novel a substance called ‘ice-nine’, created by a fictitious Nobel Laureate in Physics, is an alternative structure of water that is solid at room temperature. When a crystal of ‘ice-nine’ is brought into contact with liquid water, it becomes a seed that indeed autocatalyzes the conversion of liquid water to the solid ‘ice-nine’ that unfortunately has a melting point of 45.8°C , thus potentially ending all life on the planet. (Note that Vonnegut’s fictional ice-nine is not to be confused with the real substance Ice IX, also pronounced ‘ice-nine’, an exotic form of solid water that exists at temperatures below 140 K and pressures between 200 and 400 MPa).

It was argued by some, early on in the story, that ‘polywater’ was simply impure water since boiling point elevation and freezing point depression are colligative properties characteristic of solutions. However, the flurry of papers emphasized the precautions that had been taken to avoid contamination, so for several years the existence of ‘polywater’ was taken for granted as a real phenomenon. However, eventually the case for ‘polywater’ began to crumble. Because it could only be formed in quartz capillaries of very small internal diameter, very little was available for analysis. When eventually small samples could be subjected to trace chemical analysis, ‘polywater’ was shown to be contaminated with a variety of substances from silica to phospholipids. Moreover, electron microscopy revealed that ‘polywater’ also contained finely divided colloidal particulates in suspension.

At this point the experiments that had produced polywater were repeated with extreme precautions, including rigorous cleaning of glassware. As a result the anomalous properties of the resulting water vanished, and even the scientists who had originally advanced the case for ‘polywater’ agreed that it did not exist. There was no question of scientific fraud. In retrospect it was simply a case where meticulous physical experiments turned out to be of no value because the subject of the experiments was not a simple pure substance, but a complex chemical mixture albeit one in which the components that caused the problem were present at ultra-trace levels.

A book describing the ‘polywater’ story has been published (Franks 1981).

The first step in trace chemical analyses is to determine whether or not the target analyte really is present (we might call this a ‘binary quantitative analysis’ with possible values of 0 or 1). An implication of this step is that we are ensuring that the signals that we eventually record using our analytical apparatus really do arise from the target analyte, and only from that compound. In order to achieve this it is necessary to selectively remove most of the other 10^9 – 10^{12} molecules of many kinds so that they do not interfere with our recorded signals, and do this while at the same time discarding only a small fraction (that must be measured as accurately and precisely as possible) of the analyte itself. Moreover, in real life, different methodologies must be devised for each combination of analyte and matrix, and in most real-world cases must be capable of delivering measurements that are ‘fit for purpose’, often on a scale of hundreds or even thousands of samples per week. An admittedly poor analogy with the approach used to achieve the spectacular successes of physical metrology would involve several generations of trace analytical chemists working on analysis of the same single analyte in the same batch of matrix (that might be a homogeneous liquid or a heterogeneous powder), in order to achieve the ultimate levels of accuracy and precision.

Naturally the likelihood of any such project being undertaken is essentially zero; in real life chemists are faced with one or several of literally hundreds of thousands of possible compounds present in a wide range of matrices; in Chapter 11 a handful of examples out of literally tens of thousands of possibilities are discussed. There is no useful purpose to a ‘physics-style’ effort to measure a trace level amount of substance to the utmost precision and accuracy that analytical technologies could conceivably provide. In fact, the spectacular success of chemical metrology applied to trace analysis is not the ~ 1 in 10^1 precision that is routinely achieved, but the analyte levels of one in 10^9 – 10^{12} for which such levels of precision (and accuracy) can be achieved on a high throughput basis for a wide range of, e.g., biomedical and environmental samples, despite their inherent complexity.

The mass spectrometer is a key component in this quest for accuracy and precision in the midst of complexity as a result of its unique combination of universality, sensitivity and selectivity for chemical analysis, but only as an important component in a series of integrated steps ranging from proper sampling of the material to be analyzed to interpretation and evaluation of the experimental data. This aspect is emphasized in Figure 1.2, and this book represents an attempt to describe the current state of the art in each step and some of the difficulties faced in

achieving a best overall compromise if optimized conditions for one particular step are not compatible with the others. But even a mass spectrometer is itself composed of several identifiable components that must be properly integrated with one another if the most reliable experimental data are to be obtained. A schematic sketch of the major components of any analytical mass spectrometer is shown in Figure 1.3, emphasizing the complexity not only the analytical samples themselves, but also of the apparatus and techniques that need to be integrated together to achieve acceptable levels of accuracy and precision in measurement of amount of substance.

1.5 Molecular Mass Limit for Trace Quantitation by Mass Spectrometry

It is important to realize that, as a consequence of the non-negligible natural abundances of higher mass isotopes of important elements (particularly carbon, Table 1.4), an upper limit of molecular mass (NOT m/z) for analytes that can be quantitated at trace levels using the techniques described here is generally around 2000 Da. One major reason for this limit arises from the natural abundance of the ^{13}C isotope (~ 0.011 , Table 1.4). As an organic molecule becomes larger, the number of carbon atoms that make the largest contribution to the distribution of isotopologs (a composite word derived from ‘isotopic’ and ‘analog’) of a molecule (in the absence of chlorine or bromine) also becomes larger; as a result the relative importance of isotopologs containing one or more ^{13}C atoms increases (see Section 2.2.3 for a discussion of ‘isotopolog’ vs ‘isotopomer’). We can get a quantitative feel for the effect by considering the probabilistic expression (binomial expansion) for the relative abundances of isotopologs $^{12}\text{C}_{(n-k)}^{13}\text{C}_k$ where n is the total number of carbon-atoms in the molecule (see isotope text box in Chapter 8 for discussion of the coefficients in the binomial expansion):

$$(a + b)^n = \sum_k \{n!/[k!(n - k)!]\} . a^{(n-k)} . b^k$$

where for carbon $a = 0.989$, $b = 0.011$, and $n!$ is ‘factorial n ’, a continued product:

$$n! = n.(n - 1).(n - 2). \dots .3.2.1$$

Suppose we find the value of n for which the natural abundances of the $^{12}\text{C}_n$ and $^{12}\text{C}_{(n-1)}^{13}\text{C}_1$ isotopologs (i.e., those with $k = 0$ and $k = 1$) are equal; the corresponding molecular mass is sometimes referred to as the ‘cross-over’ value. All we have to do is equate the first two terms

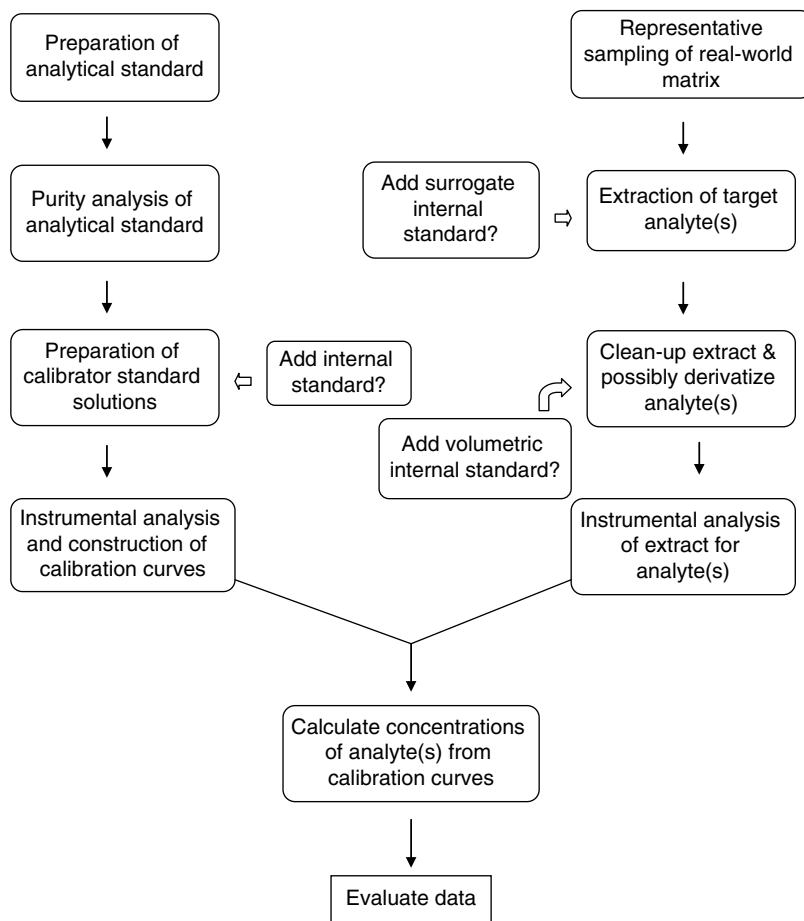


Figure 1.2 Summary of the several steps that must be integrated in an overall optimized quantitative analytical procedure. (Note that a mass spectrometer, although a key component, appears only in the ‘Instrumental Analysis’ steps (and to some extent in the purity analysis of the calibration standard)).

($k = 0$ and 1 , corresponding to $^{12}\text{C}_n$ and $^{12}\text{C}_{(n-1)}\ ^{13}\text{C}_1$) in the above expression for the binomial distribution $(a + b)^n$:

$$\begin{aligned} & [n!/[0!(n-0)!] \cdot (0.989)^n \cdot (0.011)^0 \\ &= [n!/[1!(n-1)!] \cdot (0.989)^{(n-1)} \cdot (0.011)^1 \end{aligned}$$

Since $0! = 1 = 1!$ (by definition), and any number raised to the power zero = 1, cancellation of terms gives:

$$(0.989)^n = n \cdot (0.989)^{n-1} \cdot (0.011)$$

whence rearrangement gives $n = 0.989/0.011 \approx 90$, corresponding to a carbon contribution to this ‘cross-over’ molecular mass of 1080 Da. If we assume that elements other than carbon (hydrogen, nitrogen, oxygen etc.) can

contribute up to 25 % of the total molecular mass, then the ‘cross-over’ molecular mass is estimated as $\sim 1350 - 1400$ Da.

Of course this is an approximate value since we ignored isotopic contributions from ^2H , ^{18}O , ^{15}N , etc., a reasonable first approximation even though the relative abundance of ^{15}N is a relatively high 0.368 (Table 1.4) since most organic molecules contain many fewer nitrogen and oxygen atoms than carbon. More realistic assessments of the cross-over molecular mass give a value 1700–1800 Da. The effect is illustrated in Figure 1.4 for some real examples of isotope distributions for unit-mass spectra (Section 6.2.3b) of compounds containing more than one multi-isotopic element. The restriction to unit-mass spectra implies that the small mass differences between,

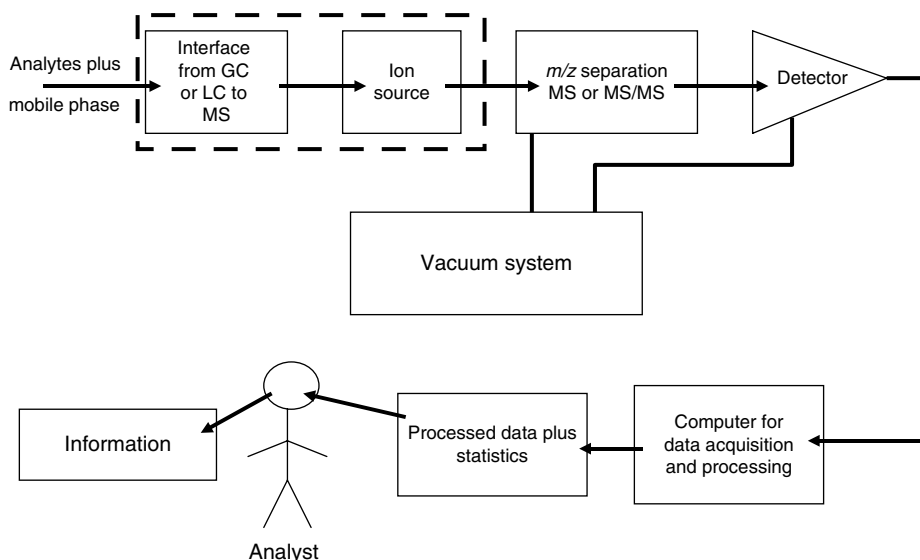


Figure 1.3 Schematic of the components integrated into a mass spectrometer used to provide selective and sensitive quantitation data. The analyte molecules entrained in the mobile phase are passed into the chromatography–MS interface and ion source where the majority of the mobile phase is selectively removed and the analyte is transformed into gaseous ions. (These two steps are sometimes incorporated into a single device, indicated by the dashed box surrounding them). The ions are then separated according to their m/z values (this can also include MS/MS analysis) and selected ions are introduced to the detector, usually an electron multiplier that can amplify the ion beam current by several orders of magnitude. This amplified current is transformed into a voltage signal that is digitized and fed into a computer where the raw data are logged in, stored, and processed. The processed data (usually with statistical assessments) are then outputted for interpretation and evaluation by the analyst who transforms the data into useful information.

e.g., $^{12}\text{C}_2\text{H}_5^{18}\text{OH}$ and $^{13}\text{C}_2\text{H}_5^{16}\text{OH}$ are not resolved so that a sample of ethanol will yield a mass spectrum in which the signal at m/z 34 contains superimposed contributions from these two species. A high resolution instrument would resolve these two signals, but for a unit-mass spectrum the combined signals would be registered and this is an underlying assumption of the calculations of isotopic distributions discussed here (Figure 1.4) and in the text box included in Chapter 8.

There are several consequences of this isotopic distribution effect for quantitative mass spectrometry. The most obvious is that sensitivity, and thus limits of detection and quantitation, will suffer as a result of the ion current arising from an analyte with naturally occurring isotopic abundances spread over several isotopologs with comparable abundances, thus lowering signal/noise (S/N) ratios (see Section 7.1) for each individual m/z value. This effect of isotopic distributions is in addition to that arising from the multi-charging phenomenon in electrospray ionization of larger molecules, which further dilutes the total ion current over more than one charge state (Section 5.3.6). Another obvious effect is the difficulties introduced when the higher isotopologs of the

natural analyte are isobaric with the lower isotopologs of an isotopically-labeled internal standard (see discussion of nonlinear calibrations in Section 8.5.2c).

A less obvious effect at or near the cross-over molecular mass arises for calibration of the m/z axis of the mass spectrometer when an automated calibration procedure is used to identify the ‘molecular peak’ (lowest mass isotopolog, often referred to as the ‘mono-isotopic peak’, i.e., ^1H , ^{12}C , ^{16}O , ^{14}N , etc. only), often specified in such automated algorithms as the most intense peak in a specified range; ambiguity in such an operational definition obviously arises near the cross-over range, potentially leading to a misidentification of the peak corresponding to the lowest-mass isotopolog (e.g., $k = 0$ in the $^{12}\text{C} - ^{13}\text{C}$ case discussed above) and thus a m/z calibration that is in error (~ 1 Th too high). Such uncertainties have extreme consequences in identification of target analytes. Selection of a precursor ion for MS/MS analyses (Section 6.2.9) also becomes problematic for larger molecules for which the mono-isotopic peak is no longer the most abundant natural isotopolog; selection of a higher isotopolog (containing two or more ^{13}C atoms) implies that fragment ions will also be represented by several isotopologs, further diluting

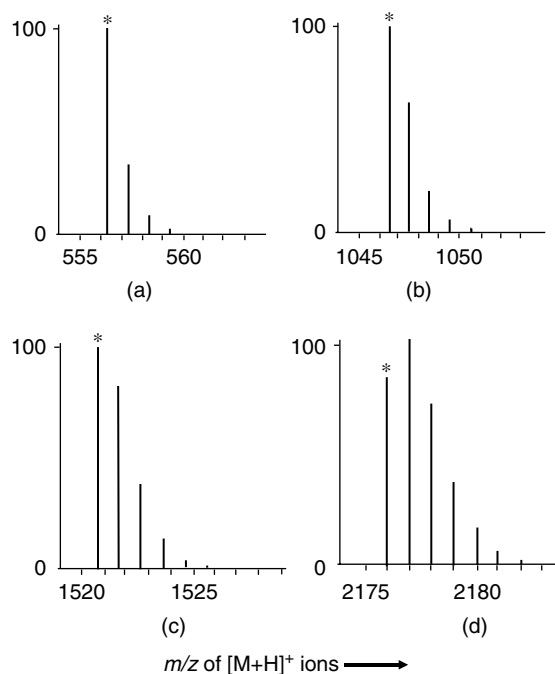


Figure 1.4 Relative abundances of isotopologs (isotopic distributions) for some peptides (containing C, H, N, S and O only), covering a range of m/z values for $[M+H]^+$ ions including the cross-over value (~ 1900 Da) at which the relative abundances are equal for the lowest-mass isotopolog (the ‘mono-isotopic peak’ containing only ^{12}C , ^1H , ^{14}N , ^{32}S , and ^{16}O , marked with an asterisk in each case) and that with the next highest mass number. Relative abundances are shown normalized to 100% for the most abundant isotopolog. (a) Leucine-enkephalin, mono-isotopic peak 556.28 Da; (b) Angiotensin II, mono-isotopic peak 1046.54 Da; (c) Fibrinopeptide A, mono-isotopic peak 1520.73 Da; (d) Fragment peptide of β -Lipotrophin, mono-isotopic peak 2175.99 Da. ‘Nominal’ m/z values, calculated using integral values for atomic masses, are (a) 556 Da, (b) 1046 Da, (c) 1520 Da, and (d) 2175 Da. ‘Isotope-averaged’ m/z values, calculated using atomic masses averaged over individual isotope masses weighted by the corresponding natural abundances within each element, are (a) 556.64 Da, (b) 1047.21 Da, (c) 1521.64 Da and (d) 2177.42 Da. Note that isotope-averaged molecular masses correspond to those used to convert weighed mass of material to moles.

the detected ion current over several m/z values and also introducing some ambiguity into the interpretation of the fragment ion spectrum.

For all of the reasons mentioned above (dilution of available ion current over too many m/z values, ambiguities in calibration and confirmation of analyte identity, choice of precursor ion in method development involving MS/MS detection), together with a significant fall-off of

response with molecular mass observed for all ionization techniques used with current analyzers (Chapter 6) and ion detectors (Chapter 7), trace quantitative analysis is usually considered to have an upper limit of ~ 2000 Da.

1.6 Summary of Key Concepts

1. The result of a measurement without an accompanying estimate of its uncertainty is of little value. The total uncertainty can be expressed in terms of **accuracy** (deviation of the measured value from the ‘true’ value) and **precision** (a measure of how close is the agreement among repeated measurements of the same quantity).

2. The **International System of Units** (Système Internationale d’Unites, SI) establishes the standards of comparison used by all countries when the measured values of physical and chemical properties are reported. There are **seven SI base quantities** (dimensions), for which the defined units of measurement can be combined appropriately to give the SI units for all other measurable quantities (i.e., the SI system is a **coherent system** of units).

3. The two base quantities that are most important for quantitative chemical analysis are **amount of substance** (measured in moles, mol) and **mass** (measured in kilograms, kg), although **length** (measured in meters, m) is also important via its derived quantity volume in view of the convenience introduced by our common use of volume concentrations for liquid solutions.

4. The **definition of the mole** (Table 1.1) refers to the number of atoms in 0.012 kilogram of carbon 12 (^{12}C). This number is the Avogadro Constant $N_A = 6.02214179 (\pm 0.00000030) \times 10^{23} \text{ mol}^{-1}$ (formerly known as ‘Avogadro’s Number’) that defines the number of molecules of a compound in one mol of that compound.

5. The **mass/charge ratios**, universally denoted as m/z , used to mark the abscissa of a mass spectrum is **best regarded as a three-character symbol, not a mathematical operation**. Within this three-character symbol, ‘ m ’ does indeed denote the mass of a single atom or molecule that has been transformed into an ion so that it is amenable to analysis by the mass spectrometer. For purposes of mass spectrometry, it is convenient to not use the kilogram as the unit of mass, but instead to express the mass in terms of the **unified atomic mass unit** u (or sometimes m_u) defined as: u (or m_u) = [(mass in kg of one atom of ^{12}C)/12]. Thus u (m_u) is the convenient unit for the mass of one single unique

molecule (ion) of a specific **isotopic composition** (e.g., $^{12}\text{C}^{35}\text{Cl}^{37}\text{Cl}_2^1\text{H}$) and is not used for the average mass of the molecule (i.e., CCl_3H). Chemists and biochemists have in the past used the **dalton (Da)** as an atomic mass unit derived from u, but adjusted for each element according to the average isotopic distribution of that element; however, the Da is increasingly used by mass spectrometrists as an alternative to u.

6. Similarly, the charge on the ion is conveniently expressed *not* in terms of the SI unit for electric charge, i.e., the Coulomb (C), but relative to the **elementary charge (e)**, equal to the magnitude of the charge (i.e., without sign) on the electron; thus the quantity z (italized) in m/z is the number of elementary charges on the ion $z = \text{magnitude of charge on the ion (C)}/e$ (C) and, as such, is also a dimensionless number.

7. The mass spectrometer is a key component in the quest for accuracy and precision in the midst of complexity as a result of its unique combination of universality, sensitivity and selectivity for chemical analysis, but only as an important component in a series of integrated steps ranging from proper sampling of the material to be analyzed to interpretation and evaluation of the experimental data (Figures 1.2 and 1.3).

8. The upper limit on the molecular mass of analytes that are amenable to trace quantitative analysis is currently considered to be $\sim 2000\text{u}$ (Da). This is the result of several factors, some of which are intrinsic to the problem, e.g., the relative abundances of higher isotopologs that increase significantly with increasing mass, and result in dilution of the total ion current derived from the analyte over too many m/z values.

