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Second Edition

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ALWAYS LEARNING

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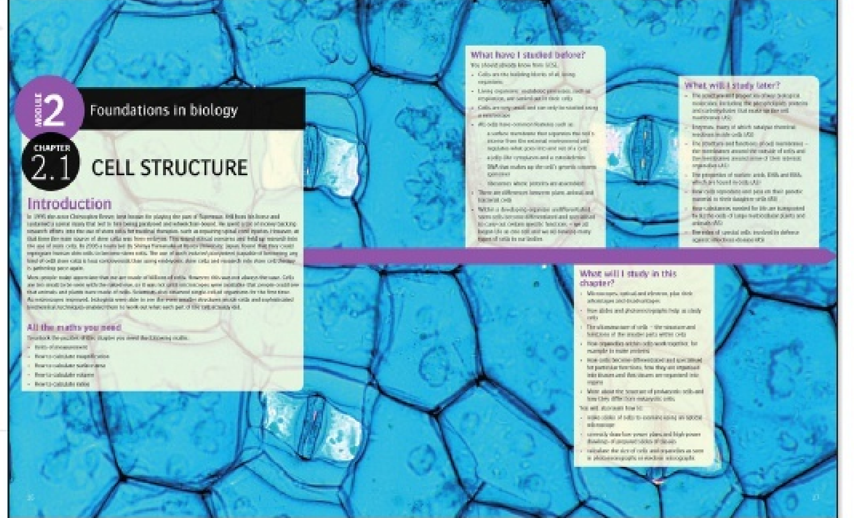

MODULE 2 Foundations in biology

CHAPTER 2.1 CELL STRUCTURE

Introduction

All the maths you need

What have I studied before?

What will I study later?

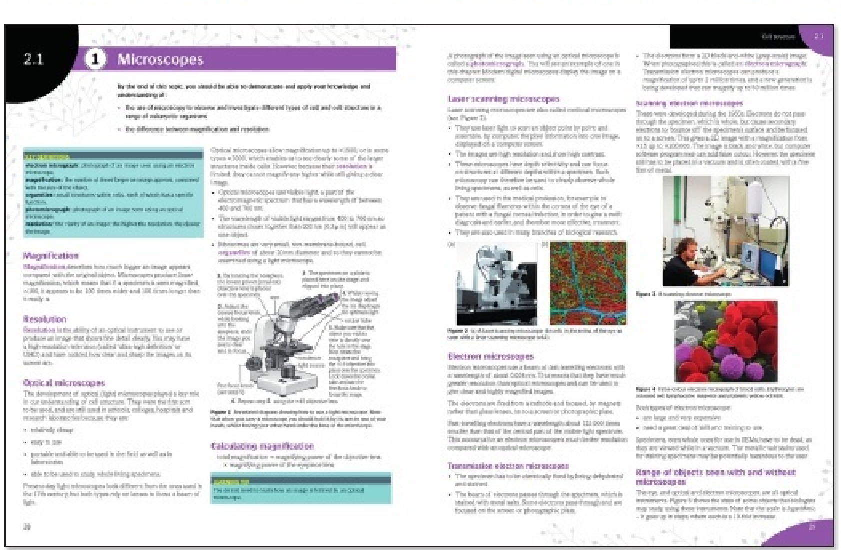
What will I study in this chapter?

Welcome to your OCR AS/A Level Biology A student book. In this book you will find a number of features designed to support your learning.

Chapter openers

Each chapter starts by setting the context for that chapter's learning:

- Links to other areas of Biology are shown, including previous knowledge that is built on in the chapter and future learning that you will cover later in your course.
- The **All the maths I need** checklist helps you to know what maths skills will be required.


2.1 1 Microscopes

Magnification

Resolution

Optical microscopes

Calculating magnification

Laser scanning microscopes

Electron microscopes

Transmission electron microscopes

Scanning electron microscopes

Range of objects seen with and without microscopes

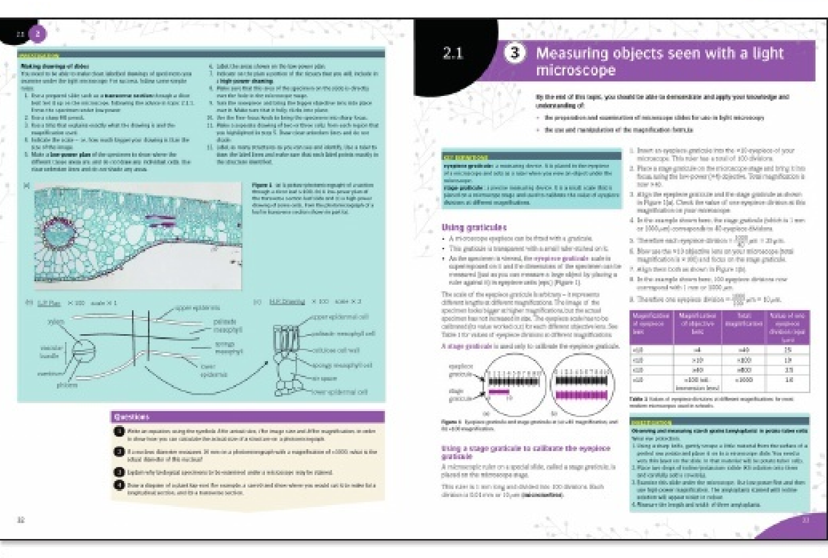
Main content

The main part of the chapter covers all of the points from the specification you need to learn. The text is supported by diagrams and photos that will help you understand the concepts.

Within each topic, you will find the following features:

- **Learning objectives** at the beginning of each topic highlight what you need to know and understand.
- **Key terms** are shown in bold and defined within the relevant topic for easy reference.
- **Worked examples** show you how to work through questions, and how your calculations should be set out.
- **Investigations** provide a summary of practical experiments that explore key concepts.
- **Learning tips** help you focus your learning and avoid common errors.
- **Did you know?** boxes feature interesting facts to help you remember the key concepts.

At the end of each topic, you will find **questions** that cover what you have just learned. You can use these questions to help you check whether you have understood what you have just read, and to identify anything that you need to look at again.


2.1 3 Measuring objects seen with a light microscope

Using graticules

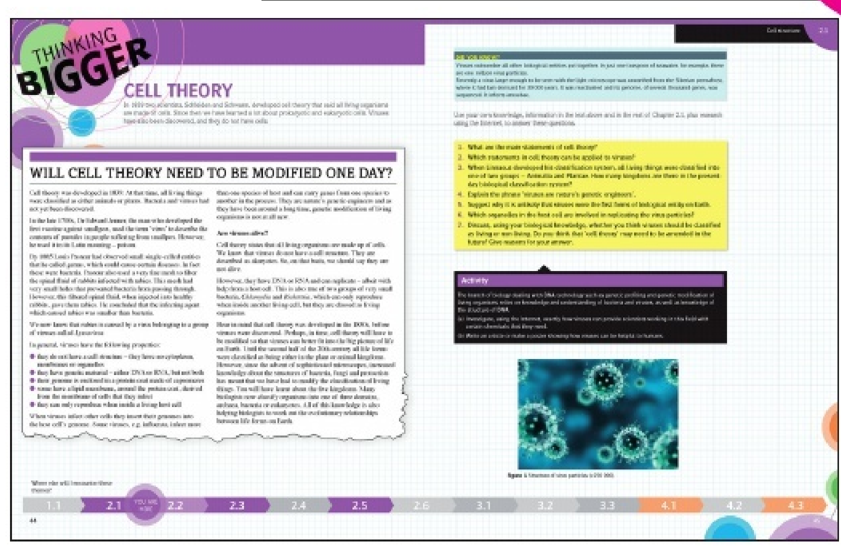
Questions

Thinking Bigger

At the end of each chapter there is an opportunity to read and work with real-life research and writing about science. These sections will help you to expand your knowledge and develop your own research and writing techniques. The questions and tasks will help you to apply your knowledge to new contexts and to bring together different aspects of your learning from across the whole course. The timeline at the bottom of the spread highlights which other chapters of your book the material relates to.

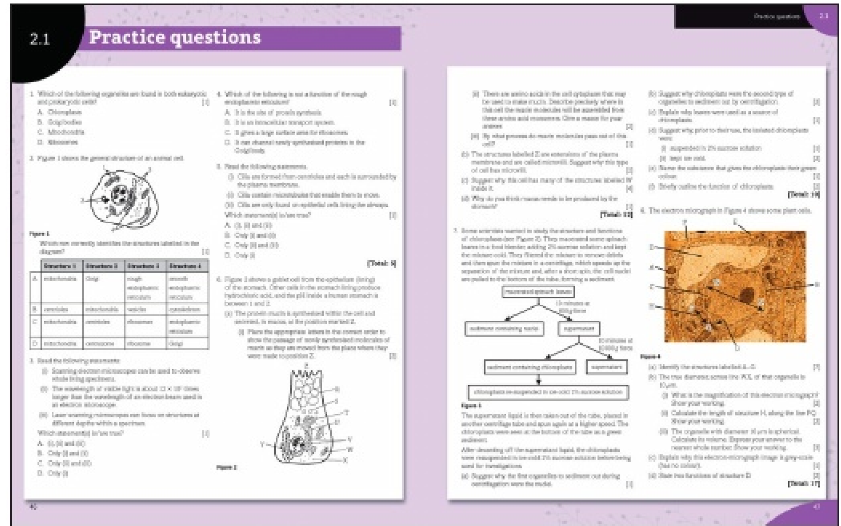
These spreads will give you opportunities to:

- read real-life material that's relevant to your course
- analyse how scientists write
- think critically and consider relevant issues
- develop your own writing
- understand how different aspects of your learning piece together.



Practice questions

At the end of each chapter, there are **practice questions** to test how fully you have understood the learning.



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Your ActiveBook is the perfect way to personalise your learning as you progress through your OCR AS/A Level Biology A course. You can:

- access your content online, anytime, anywhere
- use the inbuilt highlighting and annotation tools to personalise the content and make it really relevant to you
- search the content quickly.

Highlight tool

Use this to pick out key terms or topics so you are ready and prepared for revision.

Annotations tool

Use this to add your own notes, for example, links to your wider reading, such as websites or other files. Or make a note to remind yourself about work that you need to do.

MODULE 1

Development of practical skills in biology

CHAPTER 1.1

PRACTICAL SKILLS ASSESSED IN A WRITTEN EXAMINATION

Introduction

In 2010 Ruth Brooks began an experiment. She was fed up with the snails that ate her flowers and lettuces but did not like killing them. Although scientists thought that snails were too simple to have a homing mechanism she wondered if they would return when moved, and over what distance they could find their way home. She marked the snails' shells with nail varnish and put them into her neighbour's garden. She asked her neighbour to mark snails and these were released into Ruth's garden. Both batches of snails returned to their original gardens, showing a strong homing instinct for distances up to 10 metres, with one snail travelling 100 metres. The conclusion was that if you want to move snails away from your garden, put them more than 100 metres away and where there is food for them. The retired special-needs teacher won the BBC Radio 4 amateur scientist of the year competition, and a senior lecturer in ecology at Exeter University, who was amazed by her findings, is conducting further research into this topic to find the mechanism that snails have.

Ruth had a question and used scientific method to investigate it. Her findings are likely to lead to a reassessment of snail behaviour. The development of practical skills is fundamental to the study and understanding of all aspects of Biology. All the theories you will learn, about how scientists think living organisms function and interact with each other and their environments, are based upon practical investigations carried out by many scientists over time. The theory and practical are intertwined.

All the maths you need

To unlock the puzzles of this chapter you need the following maths:

- Perform arithmetic and numerical calculations (e.g. finding the arithmetic mean of data set replicates)
- Be able to use an appropriate number of significant figures
- Construct and interpret tables and graphs
- Calculate the rate of change from a graph
- Understand the terms mean, median and mode
- Make order of magnitude calculations
- Understand the principles of sampling
- Select and use an appropriate statistical test, such as the chi squared test, Student's *t*-test or correlation coefficient, to analyse data
- Understand standard deviation and range
- Calculate percentage error

What will I study later?

Practical skills are embedded throughout the content of your course. You will carry out various practical activities for each module and may develop different skills with each one. Some skills will underlie and be used for many of the practical activities.

You will:

- use a microscope to observe, measure and make annotated and labelled drawings of specimens (AS and AL)
- carry out qualitative and quantitative assays for biological matter, including colorimetry (AS)
- use chromatography or electrophoresis (AS)
- investigate the factors affecting metabolic and physiological processes (including enzyme-catalysed reactions) in plants and animals (AS and AL)
- dissect animal and plant organs and observe whole specimens to see how they are adapted to their environments (AS and AL)
- use sampling to measure biodiversity and investigate the factors affecting it (AS)
- sample a variety of ecosystems and record distribution and abundance of organisms (AS and AL)
- investigate genetic inheritance patterns and use the chi-squared test to analyse results (AS and AL)
- use electrophoresis to separate DNA fragments (AL)

What have I studied before?

- How to safely use a range of apparatus and equipment to carry out investigations and observations
- How to display and analyse data
- How to recognise limitations in experimental design and suggest improvements
- How to evaluate the quality and usefulness of data

What will I study in this chapter?

- The principles of experimental design, including how to solve problems in a practical context,
- Identification of control variables and evaluating the methodology
- How to process, analyse and interpret qualitative and quantitative data
- How to interpret graphs
- How to evaluate results and draw conclusions
- How to recognise the limitations in experimental procedures and suggest improvements to the experimental design

Planning

By the end of this topic, you should be able to demonstrate and apply your knowledge and understanding of:

- experimental design, including how to solve problems set in a practical context
- identification of variables that must be controlled, where appropriate
- evaluation that an experimental method is appropriate to meet the expected outcomes

Solving problems in a practical context

In your written examination you may be asked, as part of a particular question, how you could test a prediction or investigate a hypothesis or question. This is an example of solving problems in a practical context.

Although this would be a written paper and you would not have to actually carry out the investigation, you should suggest a procedure that is possible.

You should:

- be able to state which apparatus, equipment and techniques would be needed for the proposed experiment.
- apply your scientific knowledge relating to that topic.
- identify and state the independent and dependent variables and the variables that need to be controlled.
- evaluate the proposed method to see if it would do the job and provide an answer to the question. It is quite likely that your proposed method would not provide a full answer and that is fine as long as you can recognise this and say so in your evaluation.

An example of a problem

Is the growth of the single-celled green alga *Pleurococcus* affected by its geographical position, e.g. north-facing or south-facing aspect? What factors might influence its distribution?

Applying some biological knowledge to the problem

Many living things are unevenly distributed both between and within ecosystems. Many factors affect their distribution. These may be temperature; habitat; availability of water, minerals, food, space and mates; light intensity; pollution and competition with other organisms for those limited resources.

Pleurococcus is a single-celled, photosynthetic green alga. It looks like green dust and you see it on vertical surfaces such as walls and tree trunks. You may notice that there is often more on the north-facing side of these surfaces or it may be more abundant in shaded and damp areas.

As it is photosynthetic you might expect it to grow more where light intensity is greater. However, it may be damaged by high light intensities or high temperatures, or be susceptible to desiccation, in which case it would grow more in shaded areas. It is living and so will need some water.

Observations have indicated that *Pleurococcus* may have greater abundance and distribution in cooler areas with lower light intensity, i.e. in areas with a north-facing aspect. However, you cannot draw any conclusions unless you carry out some systematic investigations.

Experimental design

Think about the type of data you will be collecting and whether you have a suitable statistical test for analysing that type of data. You would need to sample many trees in different locations. If you tied a piece of string, to form a **transect**, around the tree trunk and then used a compass to find North, you could sample around the trunk, by placing mini **quadrats** (of sides 10 cm) at intervals around the circumference where the string is, and give a score of 0–10 for density of *Pleurococcus* (see topic 4.2.2 for more on using transects and quadrats for sampling plants).

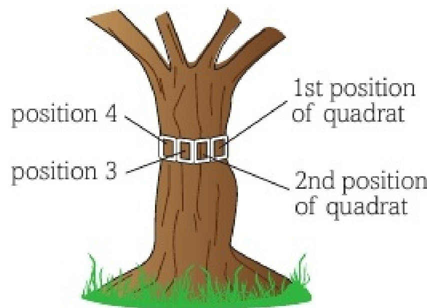


Figure 1 Using a transect and quadrat to sample the densities of *Pleurococcus* around a tree trunk.

Variables

- The independent variable (IV) is the aspect – whether north-, south-, east- or west-facing tree surface.
- The dependent variable (DV) is the density of *Pleurococcus* resulting from the different aspects.

Variables to be controlled

For example:

- species of tree
- ecosystem, whether a field or a wood
- sampling height above ground
- time of day/same day, so the weather and ambient temperature are the same
- the same person to assess density, as it is subjective.

What will you do with the data?

You could visually represent the data by constructing a bar chart for each tree, as shown in Figure 2.

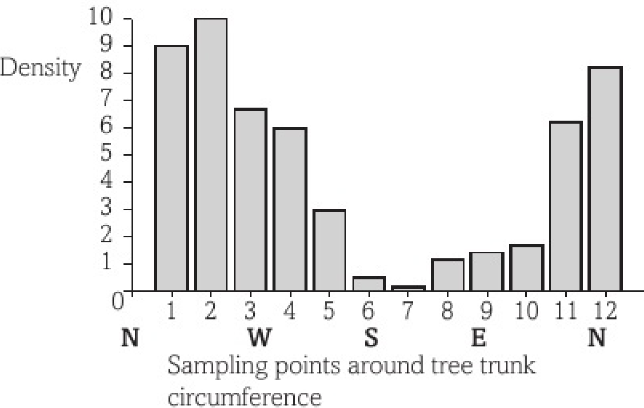


Figure 2 The distribution of *Pleurococcus* around an oak tree in a field, measured at noon during June.

Evaluation of the experimental method

There are limitations in this design:

- We have only sampled one tree, of one species, in one location.
- We have not sampled any other vertical surfaces such as walls.
- We have not used data loggers that can be left for a period of time to monitor the varying conditions.
- The data have not been analysed statistically to see if the difference between density on the north- and south-facing sides of the trees is significant.
- Even if we see a correlation between variables, for example light intensity and *Pleurococcus* distribution, correlation between two variables does not necessarily mean that one is causing the other.

Further investigations

Many experimental investigations lead to other questions that need investigating.

The data here show that the distribution of *Pleurococcus* is uneven but this does not solve the problem of what factors may *cause* this uneven distribution. We can make educated guesses, or hypotheses, as to the causes but we would need to investigate further. Those further investigations would also have to be evaluated.

Could it be light intensity? We could use a light meter to measure light intensity at the sampling areas and also look at the data on the bar chart to see if there is any pattern or correlation between light intensity and *Pleurococcus* distribution. Evaluation points: This would have to be done on the same day and at the same time of day, on a cloudy day and on a sunny day, and at the same sampling height.

Could it be temperature? Light heats surfaces so we might expect the temperature to be higher on the south-facing side of the tree trunk. Evaluation points: We could measure the temperature at each sampling area around the trunk, at the same sampling height, at the same time of day; this could be done for a cloudy day and a sunny day.

Could it be water availability? We could tape test tubes around the tree trunk and leave them to collect rain water that runs off the tree trunk. Evaluation points: Each tube would have to be left in place for the same length of time, at the same sampling height, and on the same days of the year. The tubes would have to be collected at the same time and covered to prevent evaporation, and then the water content measured by mass or volume.

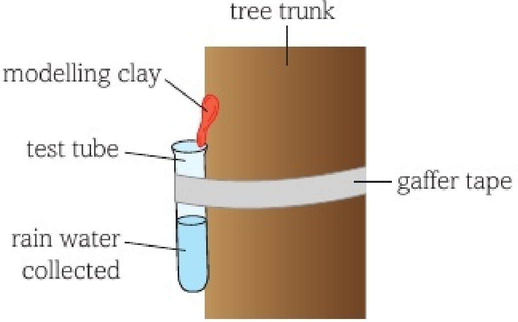


Figure 3 Collecting the water running off a tree trunk.

Could it be predation or infection? Does anything eat *Pleurococcus*? Do any microorganisms infect *Pleurococcus*? We might need to research to find this out and then examine the tree or its location to see if organisms might be infecting or eating *Pleurococcus*.

LEARNING TIPS

If you are stating that light is a possible factor that affects an organism's distribution, refer to the *intensity* of light.
Note that italics are used for proper names of living organisms. If you were writing *Pleurococcus*, for example in your field note book, by hand, you would underline it. As this is the generic name, it begins with an upper case letter.

DID YOU KNOW?

Pleurococcus is a genus of algae and has been said to be the most abundant organism on the planet.
If you use an artist's fine paint brush you can put a little of the green powdery *Pleurococcus* onto a microscope slide and examine it under low and high power. This is a eukaryotic organism; what features of its cell structure can you identify?

Questions

1. Suggest a more objective way of assessing the density of *Pleurococcus* on the bark of tree trunks.
2. Write a list of equipment you would need to carry out the investigation outlined above on *Pleurococcus* distribution.
3. Suggest improvements to this investigation, to reduce its limitations.
4. What are the possible sources of errors in this investigation?

1.1 2 Implementing an investigation

By the end of this topic, you should be able to demonstrate and apply your knowledge and understanding of:

- how to use a wide range of practical apparatus and techniques correctly
- appropriate units for measurement
- presenting observations and data in an appropriate format

Using practical apparatus and techniques correctly

Throughout your course, you will carry out several practical investigations, some of which are outlined in this book. Bear in mind that our knowledge and ideas about biology stem from practical investigations that gather data to support hypotheses that then become theories or models.

You will already have carried out practical investigations for GCSE Science and will be familiar with a range of equipment and apparatus and be aware of how to use it safely.

In your written examination you may be asked about the use of apparatus in a practical investigation. Table 1 lists some of the apparatus and techniques that you should use during your course, as well as giving examples of suitable practical activities and areas of the specification that they cover.

Type of practical activity	Skills and techniques	Example(s) of suitable practical activities	Specification section
Light microscopy	• Prepare and stain material for slides • Use microscopes at a range of magnifications • Use a graticule and measure specimens • Produce annotated scientific drawings	• Study structure of plant, animal and prokaryotic cells • Study stages of mitosis • Observe plasmolysis and crenation • Observe a range of tissues	• Cells • Exchange and transport • Homeostasis (A Level only) • Respiration (A Level only) • Photosynthesis (A Level only)
Dissection	• Safely use dissecting instruments • Make annotated drawings	• Dissect mammalian heart • Dissect mammalian kidney • Dissect plant stems	• Homeostasis (A Level only) • Exchange and transport
Sampling techniques	• Sampling techniques used in fieldwork • Make annotated scientific drawings	• Calculate species diversity	• Biodiversity • Ecosystems (A Level only)
Rates of enzyme-controlled reactions	• Use a range of apparatus to record quantitative measurements • Use a range of glassware to make serial dilutions • Use data loggers to collect data or use computer software to process data	• Effects of temperature, pH, substrate and enzyme concentration on rate of enzyme-catalysed reactions	• Enzymes • Homeostasis (A Level only)
Colorimeter or potometer	• Use colorimeter to record quantitative data • Use potometer	• Effect of temperature on membrane permeability • Rate of enzyme-catalysed reaction • Investigate the factors affecting rate of transpiration	• Enzymes • Membranes • Exchange and transport

Table 1 Apparatus and techniques used in A Level Biology.

continued

Type of practical activity	Skills and techniques	Example(s) of suitable practical activities	Specification section
Chromatography or electrophoresis	• Thin layer or paper chromatography to separate biological compounds • Gel electrophoresis	• Analyse chlorophyll • Separate and identify a mixture of amino acids • Separate DNA fragments produced by treatment with restriction enzymes	• Biological molecules • Photosynthesis (A Level only) • Nucleic acids, genetic manipulation
Microbiological techniques	• Aseptic techniques • Use of solid and liquid culture media • Colorimetry • Serial dilutions	• The effect of antibiotics on microbial growth	• Cloning and biotechnology (A Level only) • Genetic manipulation (A Level only)
Transport into and out of cells	• Serial dilutions • Data logging	• Investigate water potential of plant tissue, such as potato tuber	• Cells • Membranes
Qualitative testing	• Use qualitative reagents to identify biological molecules	• Test for biological molecules, such as proteins, lipids, sugars and starch	• Biological molecules
Investigation using a data logger or computer modelling	• Use ICT	• Investigate DNA structure using RasMol	• Nucleic acids
Investigate plant and animal responses	• Safe and ethical use of organisms to measure plant and animal responses and physiological functions • Use spirometer	• Investigate tropism in plants • Investigate growth requirements of bacteria • Measure human pulse rate at rest and after exercise • Investigate breathing rate and oxygen uptake by human at rest and during exercise • Use *Drosophila* for genetic investigations	• Plant and animal responses (A Level only) • Exchange and transport
Research skills	• Use online sources and books to research topics • Correctly cite sources of information	• Investigate respiration in yeast, *Saccharomyces cerevisiae*	• All topic areas

Table 1 Apparatus and techniques used in A Level Biology (*continued*).
Note that the types of practical activity listed are organised according to the practical activity groups (PAGs) referred to in the specification.

Appropriate units for measurement

In many practical investigations you are likely to be measuring something. It is important that you use the correct units and the correct symbols or abbreviations.

Below are some of the units you may use, with their correct symbols, e.g. kilograms (kg), metres (m), seconds (s), joules (J) or kilojoules (kJ) for energy, kilopascals (kPa) for pressure or water potential. However, the actual unit used depends on what you are measuring. If you are measuring the diameter of a cell, micrometres (μm) would be appropriate, but if measuring the height of a tree, metres would be a more appropriate unit. For certain studies involving energy flow through ecosystems, the units might be gigajoules per hectare per year (GJ ha^{-1} yr^{-1}).

Prefix	Order of magnitude
nano-	10^{-9}
micro-	10^{-6}
milli-	10^{-3}
centi-	10^{-2}
kilo-	10^{3}
mega-	10^{6}
giga-	10^{9}
tera-	10^{12}
peta-	10^{15}

Table 2 Prefixes denoting orders of magnitude.

DID YOU KNOW?

A googol is the name given to a number of order magnitude 10^{100}, which is 10 with 100 zeros after it.

And 10^{googol} is called a googolplex. Both these names were invented by a nine-year-old child, the son of a mathematician.

Unit	Abbreviation	Number of metres
kilometre	km	1000
metre	m	1
centimetre	cm	0.01
millimetre	mm	0.001
micrometre	μm	0.000 001
nanometre	nm	0.000 000 001

Table 3 Units for length: SI base unit = metre.

Unit	Abbreviation	Number of square metres
kilometres squared	km^2	1 000 000
hectare	ha	10 000
centimetres squared	cm^2	0.0001
millimetres squared	mm^2	0.000 001

Table 4 Units for area.

Unit	Abbreviation	Number of centimetres cubed
cubic decimetres	dm^3	1000
cubic centimetres – also called millilitres	cm^3 or ml	1
cubic millimetres – also called microlitres	mm^3 or μl	0.001

Table 5 Units for volume.

Unit	Abbreviation	Number of grams
metric tonne	t	1 000 000
kilogram	kg	1000
gram	g	1
milligram	mg	0.001
microgram	μg	0.000 001

Table 6 Units for mass.

LEARNING TIP

Always state the units you are using when describing a quantity.

Presenting your observations and data

If you have been observing a structure, such as an organ or organ system via dissection, a labelled drawing is the way to present this. When you study transport in animals (Chapter 3.2) you will have the opportunity to dissect a mammalian heart and make annotated drawings of your observations.

A labelled drawing is also the way to present observations of cells or tissues on a microscope slide. In Chapter 2.1 you will have several opportunities to make such annotated drawings from microscope slides, in the correct way.

Besides drawings, figures, graphs and diagrams are also visual representations of observations and results of investigations. Topics 1.1.3 and 1.1.4 deal with different types of graphs and diagrams.

Tables

Often the best way to present initial data from an investigation is in a table – see Table 7 for an example:

- The table must have a clear title to inform the reader.
- The table should be ruled off.
- The independent variable should be in the first column (to the left side of the table).
- Each column should have an informative heading and the units for the quantities shown should be in the column heading, not in the column itself.

- You can tabulate data that are not quantitative, such as colour of reagents used in tests and the inference (what it tells you).
- If the data are quantitative, the same number of decimal places should be used for all the values in one particular column.
- If replicates have been carried out there should be a column for each and a column for the calculated mean values.
- The mean values should be calculated to the same number of decimal places or to one more decimal place than those of the raw data values, but all the mean values in a column must be to the same number of decimal places.

Temperature (°C)	Rate of hydrolysis of starch (mg s^{-1})			Mean rate of hydrolysis of starch (mg s^{-1})
	1	2	3	
10	11.54	11.36	11.43	11.44
20	21.90	21.59	22.01	21.83
30	35.30	36.00	35.85	35.72
40	36.54	37.01	36.97	36.84

Table 7 Rates of digestion of starch by the enzyme amylase, obtained from goat saliva, at different temperatures.

Questions

A student investigated the digestion of triglyceride (fat) by the enzyme lipase. He wanted to investigate the effect of increasing temperature on the rate of reaction. The enzyme-catalysed reaction produces fatty acids and these lower the pH. This change in pH can be detected by an indicator, such as bromothymol blue, which is blue at pH 7.6, green at pH 7.0 and yellow at pH 6.0. The time taken for the indicator to change to yellow can be measured and so the rate of digestion can be determined. The student presented his data in a table as shown below.

Time taken for indicator to become yellow (secs)			Temperature
1	2	3	
454	476	468	10 °C
287	295	305	15 °C
210	208	212	20 °C
121	123	126	25 °C
105	110	109	30 °C
68	63.5	65.5	35 °C

1. State six ways in which this table can be improved.
2. Calculate the mean rates of reaction for these data. Calculate rate as 1000 divided by time taken for indicator to become yellow. (We use $1000/t$ rather than $1/t$ to calculate the rate, so that the numbers in the calculation are more user friendly. As long as all values are treated in this way, the relative rate of reaction is the same, in effect $1/t \times 10^3$.)
3. Present these data in a properly constructed table.
4. Comment on the range of temperatures used in this investigation.
5. What are the limitations of this investigation in terms of determining the end point of the indicator?
6. Suggest how this investigation could be improved and include suggestions for other ways of measuring the fall in pH.

1.1

Analysis of data 1: Qualitative and quantitative data

By the end of this topic, you should be able to demonstrate and apply your knowledge and understanding of:

- processing, analysing and interpreting qualitative and quantitative experimental results
- use of appropriate mathematical skills for analysis of quantitative data
- appropriate use of significant figures

KEY DEFINITIONS

qualitative data: data that does not involve quantity (numbers).
quantitative data: data that does involve quantity (numbers).
significant figures: the digits of a number that have a meaning and contribute to the number's precision.

Processing, analysing and interpreting results

When you carry out tests to indicate the presence of glucose, starch, lipids or proteins (see topic 2.2.12 for more about these food tests), you will obtain **qualitative data**. You can represent such findings in a table and indicate the colour observed and the inference – this tells us whether a substance is present or not.

Benedict's reagent is used to test for reducing sugar (if positive, reagent changes from blue to red when heated); iodine/KI solution tests for starch (if positive, a blue-black colour is seen); ethanol emulsion test indicates the presence of lipids if a white emulsion is seen; biuret reagent indicates the presence of protein by a purple/mauve colour.

To make your data **quantitative**, for example to see how much glucose is in a particular drink, you would need to make up a range of glucose solutions of known concentrations, using serial dilution. You would then carry out a Benedict's test, keeping certain variables constant, such as:

- volume of reagent
- volume of solution being tested
- temperature at which heated
- length of time for heating.

You would then see a range of colours showing the positive Benedict's test result, from brick red for a high concentration, through orange, yellow to green for a very low concentration, corresponding to specific concentrations of glucose in solution.

You could use these, or a photograph of them, as standards against which to compare the results of carrying out a Benedict's test on solutions of glucose of unknown concentration.

Using mathematical skills to analyse quantitative data

Think about the measurement of water uptake by a potometer, as described in Chapter 3.3. If measurements are taken at different ambient temperatures we can see the effect of temperature on the rate of water uptake and therefore on the rate of transpiration.

Ambient temperature (°C)	Distance travelled by air bubble in 10 min (mm)				Rate of uptake of water ($\mu l\ s^{-1}$)
	1	2	3	mean	
10	12.5	13.0	13.5	13.0	0.11
20	28.0	27.5	27.3	27.6	0.23
30	45.0	47.0	46.0	46.0	0.38
40	55.5	56.5	55.7	55.9	0.46

Table 2 Mean rate of water uptake ($\mu l\ s^{-1}$) in a leafy sycamore maple, *Acer pseudoplatanus*, shoot in a potometer, at different ambient temperatures.

Calculating the volume of water taken up

If you are told, for example, that the diameter of the bore of the capillary tube is 2.5 mm and the air bubble travelled 24 mm in 10 minutes, you can calculate the rate of uptake of water in μl per minute or per second.

Food tested	Colour/observation at end of test				Inference
	Benedict's reagent at 80 °C for 10 min	Iodine/KI solution	Ethanol emulsion test	Biuret reagent	
bread	blue	black	colourless	mauve	contains starch and protein
potato	blue	black	colourless	mauve	contains starch and protein
apple	red	brown	colourless	blue	contains reducing sugar and protein
cheese	blue	brown	white	mauve	contains lipid and protein
chicken	blue	brown	white	mauve	contains lipid and protein

Table 1 Results of tests carried out on a variety of foods.

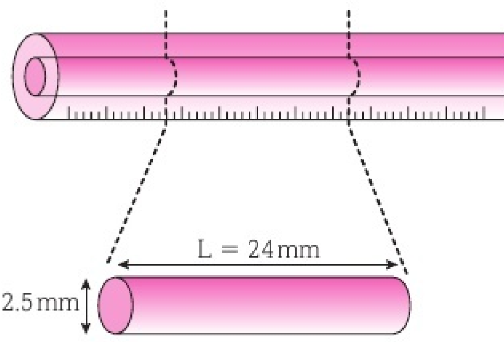


Figure 1 Calculating the volume of water in a section of capillary tube.

If the diameter is 2.5 mm then the radius is 1.25 mm.

L indicates the length moved by the air bubble, so the space in this cylinder is the same as the volume of water taken up by the shoot.

The formula for calculating the volume of a cylinder, *V*, is $V = \pi r^2 L$
So the volume of water taken up by the shoot in 10 minutes is
$[3.142 \times (1.25)^2 \times 24]$ mm^3
$= 117.825$
$= 118\ \mu l$

LEARNING TIP

Notice that the number in this calculated example has been rounded to a whole number. This is because you can only read this scale to one decimal place. You could express the answer as 117.8 µl, but to no more than one decimal place. mm^3 is not incorrect as a unit but µl is more often used.

Now to calculate rate of uptake, which is volume taken up per unit time.

If $118\ \mu l$ is taken up in 10 minutes, then the rate of uptake is $118/10 = 11.8\ \mu l\ min^{-1}$.

You could also express this in terms of volume taken up per second, which would be $118/600 = 0.20\ \mu l\ s^{-1}$.

Calculating a median value

Suppose you measure the lengths of the leaves on a branch of a shrub. Their measurements in mm are:

62, 65, 75, 83, 55, 78, 77, 68, 57, 58, 54, 66, 72, 80, 48, 71, 72, 62, 49, 81.

The **arithmetic mean** is 66.7 mm.

The range is from 48 to 83 mm.

There are 10 numbers from 48 to 66 and 10 numbers from 68 to 83. The **median** is therefore 67 (between 66 and 68). This is correct even though there are no leaves of 67 mm in the sample.

Appropriate use of significant figures

In some cases we do not need a detailed answer or very precise number. When you work out an answer on your calculator you do not need to express it to 10 decimal places so you round it off to a certain number of decimal places.

Another method is to round it off using **significant** (meaningful) **figures**.

From the column in Table 2 showing the rate of transpiration, in the second row where the rate is 0.23, 2 is the most significant digit because it tells you that the rate is about $0.2\ \mu l\ s^{-1}$. The second number, 3, is the next significant figure. It tells us that the rate is faster than $0.2\ \mu l\ s^{-1}$. This therefore gives a more accurate and precise indication of the value of the rate calculated. Because this is a calculated value, it can be expressed to one more decimal place than the values in the other columns that were obtained by reading the apparatus and were therefore limited by the precision of the apparatus. The calculated values in this column in Table 2 are all to two significant figures.

As a general rule, the calculated values, in order to be significant, can be to one more decimal place than the values in the columns from which the calculation was made.

The following are not significant figures: leading zeros, trailing zeros and digits derived by calculation and giving several decimal places, which therefore give *far* greater precision than the original data or the instrument used for measurement.

Questions

1. Express the following to two significant figures:
   (a) 5 374 641
   (b) 1.645 783 6
   (c) 0.985 342 1
   (d) 15.0
   (e) 0.678 000 0.

2. In an investigation using a potometer, the bubble of air moved 65 mm along the capillary tube in 15 minutes. The diameter of the bore of the capillary tube was 2 mm. Calculate the rate of water uptake by the plant in $mm^3\ s^{-1}$ ($\mu l\ s^{-1}$).

3. Suggest how you could adapt the use of the biuret test for protein to make it quantitative.

Analysis of data 2: Graphs

By the end of this topic, you should be able to demonstrate and apply your knowledge and understanding of:

- plotting and interpreting suitable graphs from experimental results

There is a variety of graphs and each type has specific uses, but each communicates information visually.

In a written examination you may be given a table of data and be asked to graph those data.

You may also be asked to:

- make deductions from graphical data
- draw conclusions from graphical data
- evaluate the data or its presentation (see next topic).

Line graphs

Line graphs are used to see if there is any correlation between two variables where the data are continuous.

strong positive correlation

DV

IV

weak positive correlation

DV

IV

strong negative correlation

DV

IV

Figure 1 Examples of correlation between two variables.

- They involve a vertical *y*-axis and a horizontal *x*-axis forming a grid.
- Each axis should have a suitable linear scale and be labelled with quantities and units.
- The independent variable (IV) is usually plotted along the *x*-axis and the dependent variable (DV) along the *y*-axis.
- For biological data it is often best to join the plotting points with straight lines. If you do this then the line should go through the centre of each plot. Sometimes a smooth line of best fit can be drawn that goes through or very near to the points. Whichever type of line is drawn, it should *not* be extrapolated, that is, it should not be extended outside of the minimum and maximum value plot points.

LEARNING TIPS

A line on a graph is called a curve, even if it is a straight line.
When you draw a graph, make it large and make sure you use a suitable scale and label each axis. Take care to plot the points accurately.

DID YOU KNOW?

Sometimes wrong conclusions have been drawn by extrapolating biological data. Data on high doses of ionising radiation were collected by physicists in the 1940s and 1950s and used to assess the risk to human health. When plotted and extrapolated, they suggest that low levels of radiation are harmful (Figure 2). However, more recent evidence suggests that lower levels of radiation are harmless (Figure 3).

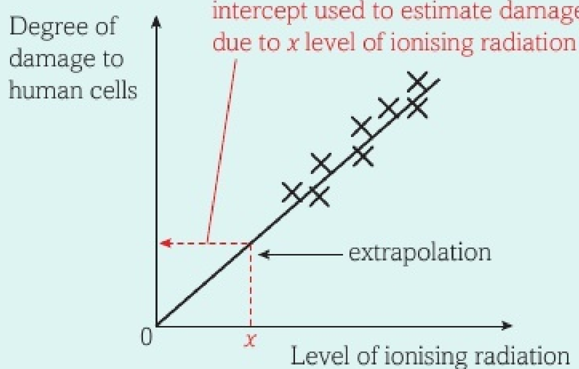


Figure 2 Predicted damage to human cells with increasing levels of ionising radiation.

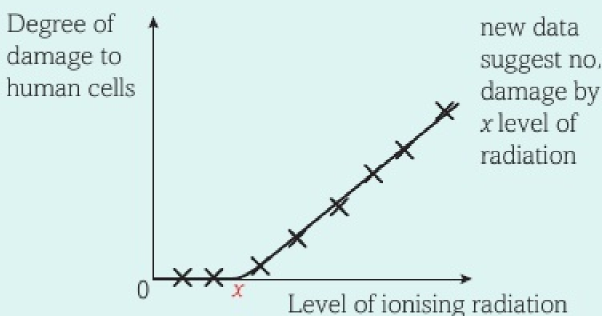


Figure 3 Actual damage to human cells with increasing levels of ionising radiation.

More than one curve can be drawn on the same set of axes, so comparisons can be made and a picture of what is happening during an investigation or observed phenomenon can be seen.

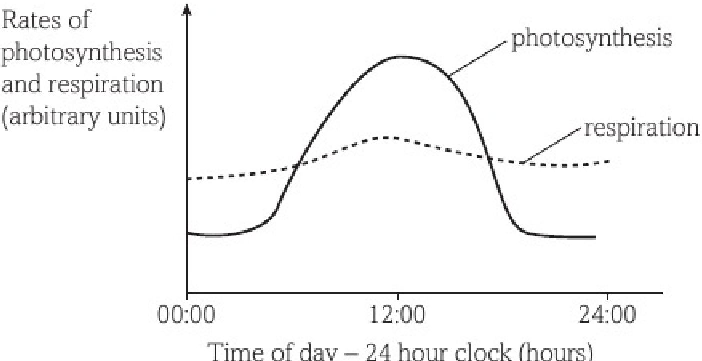


Figure 4 Graph showing the changes in rates of photosynthesis and respiration in a small pond over a 24 hour period during May.

- The rate of reaction can be calculated from the slope of a curve showing the progress of the reaction over time.

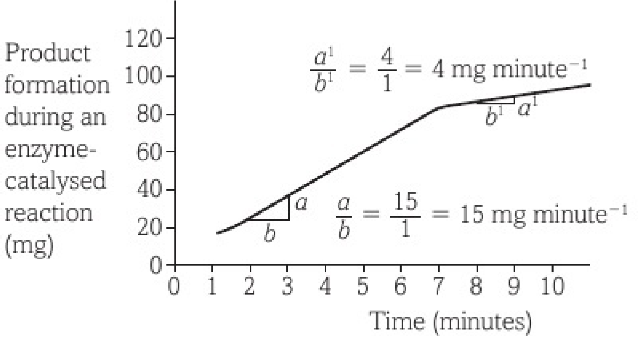


Figure 5 Calculating the rate of an enzyme-catalysed reaction from the slope of a graph

Scattergrams

Also called scatter diagrams or scatter plots, scattergrams are used when investigating the relationship between two naturally changing variables. For example, several plots can be made showing mean blood cholesterol level and death rates from heart disease and stroke in various countries. No line needs to be drawn, but the pattern of the plots can show if there is any correlation.

Bar graphs

Bar graphs are used to investigate relationships when the independent variable is categorical and the dependent variable is continuous, e.g. the concentration of Vitamin C (DV) in different fruit drinks (IV).

- The bars should be of the same width and equally spaced.
- If mean values are shown on the bars, the range bars can also be shown.
- If the data sets being compared have been analysed statistically, the error bars can be shown. If there is overlap it indicates that any apparent difference is not significant.

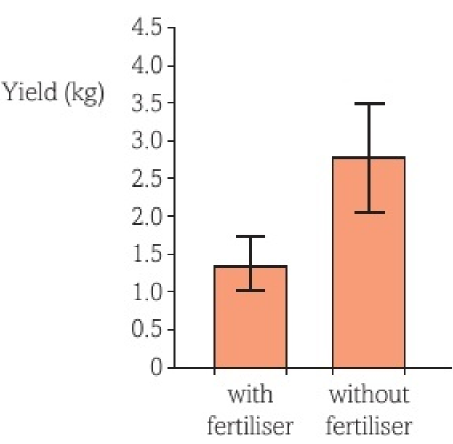


Figure 6 Comparison of yield of tomatoes grown with and without fertiliser. Error bars do not overlap, showing that the difference between these two data sets is significant.

Histograms

Histograms can be used for showing quantitative data organised into classes. For example, if we measured the height of a large number of human adults we may categorise the data, for example those between 140 and 149 cm and those between 150 and 159 cm. The number of people within each class shows the frequency. The class or category that contains the greatest frequency is the **mode.**

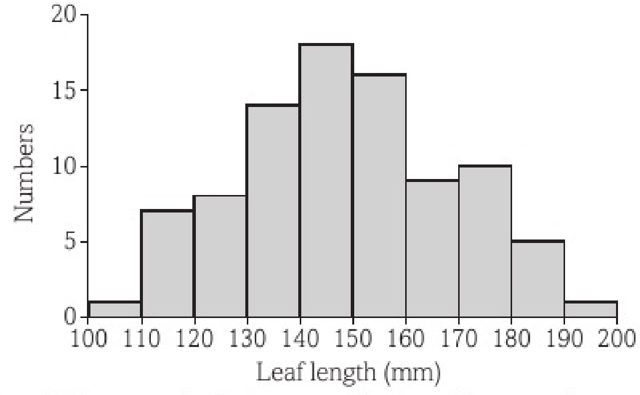


Figure 7 Histogram showing frequency of leaf length in sweet chestnut.

Questions

Which type of graph would you draw to display each of the following types of data?

1. Lengths of leaves on a tree branch.
2. Effect of changing pH on enzyme activity.
3. Sugar content of different types of biscuits.
4. Effect of light intensity on rate of photosynthesis.
5. Collagen content of skin and age in humans.
6. Amino acid content of beef and cheese.

Evaluation

By the end of this topic, you should be able to demonstrate and apply your knowledge and understanding of:

- how to evaluate results and draw conclusions
- the identification of anomalies in experimental measurements
- limitations in experimental procedures
- the refining of experimental design by suggestion of improvements to the procedures and apparatus
- precision and accuracy of measurements and data, including margins of error, percentage errors and uncertainties in apparatus

KEY DEFINITIONS

accuracy: how close a measured or calculated value is to the true value.

anomaly: result that does not fit the expected trend or pattern.

precision: the closeness of agreement between measured values obtained by repeated measurements.

Evaluating results and drawing conclusions

You may be shown data and asked to evaluate them or to comment on a conclusion drawn from the data.

For example, in Table 1 are data about changes in blood cholesterol levels. One group of patients was given cholesterol-lowering drugs, called statins. Another group of patients within the same GP practice decided to try to lower their blood cholesterol levels by taking more exercise and altering their diet.

Patient group	Mean blood cholesterol level (mmol dm^{-3} [±SD])	
	Before treatment	6 months after treatment began
Group A – treated with statins ($n = 12$)	6.36 (±1.58)	4.21 (±0.19)
Group B – treated with lifestyle change ($n = 12$)	5.95 (±1.34)	4.87 (±1.60)

Table 1 Blood cholesterol levels of groups of patients in a GP practice.

Statins inhibit an enzyme in the liver from making cholesterol. The guidelines set by NICE (National Institute for Health and Care Excellence) in 2008 stated that people should have a blood cholesterol level of 5.2 mmol dm^{-3} or less. In 2014 the guidelines were changed to 4.0 mmol dm^{-3}.

What can we conclude from these data?

- It would appear that statins are more effective at lowering blood cholesterol level than a patient making lifestyle changes.
- Lifestyle changes appear to lower blood cholesterol, although the improvement was not as marked as in the group taking statins. However, the SD of this group was greater after treatment so some may not have shown any improvement.
- In group A the **standard deviation**, **SD** (indicating the variability of the data, or the size of its spread about the mean) is much larger before treatment than after, which shows that there was quite a high range of blood cholesterol values among these patients before treatment, but their values showed a much narrower range (they were all closer to the mean value) after treatment. This indicates that most of their blood cholesterol levels were probably brought close to the new guideline levels.
- However, we do not know if they suffered any side effects. Some people suffer muscular pains when taking statins and these prevent them from exercising, which is another way of helping to lower blood cholesterol.
- It is easy to monitor the dose of statins taken by each patient in group A, whereas lifestyle changes are harder to monitor as they depend on subjectivity of those making the changes.
- Group A could have also made some lifestyle changes; if they are concerned about their health and willing to take statins, they may also decide to eat more fruit and vegetables and take more exercise.
- There is no information here about the age, gender or family history of patients in each group.
- The two groups had different mean starting levels of blood cholesterol.
- The initial SD for group A is larger than for group B, so there may be more patients in group A with very high blood cholesterol levels.
- These are small groups and this is only one study. It would have to be replicated before any valid conclusions could be drawn.

Identifying anomalies in data

You have been trained to identify **anomalies** in data. These are results that do not fit the expected pattern. Seeing an anomaly can be an exciting moment, providing evidence that your expectation is wrong and a scientific breakthrough could be staring you in the face. On the other hand it could be due to a piece of grit in your detector or a leaky flask in your incubator. If you are certain that an anomalous piece of data was produced due to a failure in the experimental procedure, you might be justified in removing it before analysing the data. However, you must never discard data simply because they do not correspond with your expectation. By repeating the experiment and amassing more data one of two things could happen. If the anomaly was the result of an experimental error or was simply a very unusual result from naturally-occurring variation it will 'disappear' as the repeat measurements produce a mean in line with expectation. On the other hand, if the anomaly was in fact telling you something surprising about the system you are investigating it will be confirmed by repeat observations and your Nobel Prize is just around the corner.

Limitations in experimental procedures

- It is not always possible to control all extraneous variables.
- Some investigations would be unethical, such as deliberately damaging an area of children's brains to study the effects on their development.
- Results obtained from studying a small population cannot be generalised to the whole population.
- The resolution of the instruments and equipment used may impose limitations.
- The degree of **accuracy** of measurements may lead to limitations.
- Using a small sample size or having too few replicates is also a limitation, as it is difficult to see if the data are reliable; therefore a large enough sample or enough replicates should be used where possible.
- Not leaving a reaction for long enough to fully complete will give misleading data; therefore we should make sure that reactions are given long enough to complete.
- Not allowing reactants to reach the required temperature before adding them together will reduce validity; reactants should be placed, in their tubes, into a water bath to reach the required temperature before they are mixed.
- Some investigations that rely on questioning people or observing them in particular situations may be limited, because only certain types of people will volunteer to take part or people will behave differently when they think they are being observed.
- Lack of equipment to objectively measure something, such as a colour change, is a limitation as the observation is subjective and may change depending on the investigator.
- Limitations in equipment such as using a beaker of hot water for a waterbath; the investigation can be improved by using a thermostatically controlled water bath, with a thermometer to check the temperature, so as to maintain the desired temperature throughout the reaction.

Errors

Errors or experimental uncertainties arise because there are:

- inadequacies and imperfections in experimental procedures
- lapses of judgement by the experimenter
- limits to resolution, **precision** or accuracy of measuring apparatus.

Random errors due to judgement errors made by the experimenter are reduced when the procedure is repeated several times.

Systematic errors may be inherent in the equipment and are repeated at every replicate. However, if the percentage error is known, a calculation can be done to determine the margin of error.

LEARNING TIP

Be clear about the difference between accuracy and precision. A thermometer is inaccurate if it gives readings that are 5 °C above the true temperature but it could still be precise if it gives very consistent readings. By recalibrating an inaccurate instrument you can correct this and make accurate measurements. Still confused? An analogy might help: A precise archer will have her arrows tightly clustered somewhere on the target. By recalibrating her sight she can become accurate *and* precise and will have her arrows clustered on the bullseye!

Questions

1. A digital stopwatch can measure to the nearest 0.1 s. Explain why using this stopwatch to measure a reaction for 5 minutes is more accurate than using it to measure the reaction times of humans, which are around 0.3 s duration.
2. In school laboratories, thermometers filled with alcohol rather than mercury are used for safety reasons. They are precise and have an impressive resolution of 0.2 °C. However, the overall calibration could be up to 1°C out. If you used one of these thermometers to measure the temperature of a water bath at 38 °C, within what range would the real temperature be?
3. Explain why using a gas syringe to collect oxygen given off from a well-illuminated aquatic plant, for 5 minutes, is better than counting the bubbles of oxygen produced during 5 minutes.

HOW SCIENCE CAN GO WRONG

Scientific research has changed how we perceive the world, and the scientific method has evolved to try and prevent flawed research that could misinform us. However, now it may itself need to change.

HOW SCIENCE GOES WRONG

A simple but powerful idea that underpins science: 'trust but verify', has generated a vast body of knowledge. Since its birth in the 17th century, modern science has changed the world beyond recognition, and overwhelmingly for the better. Results should always be subject to challenge from experiment.

However, success can breed complacency. There are many published academic studies that are the result of shoddy experiments or poor analysis. Less than half the published research on biotechnology can be replicated.

In the 1950s, following many successful applications of science during World War II, academic research was seen as important, and a few hundred thousand scientists were carrying out research. Since then, the numbers have grown to 6–7 million active researchers, and there is great pressure on them to 'publish or perish'. This overriding demand has led to a loss of self-policing and quality control. Many journals will not print studies that verify previous findings, so researchers see little value, in terms of advancing their careers, in replicating the studies of other scientists.

Failures to support a hypothesis are rarely offered for publication, and 'negative' results account for only 14% of published studies, down from 30% in 1990. However, in science, knowing what is false is as important as knowing what is true. The failure to report failures means that other researchers waste money and time exploring blind alleys that have already been explored. It may also cost lives. In March 2006, six healthy young men volunteered to take part in a clinical trial for a new drug TGN1412 that had not previously been given to humans. Within a day, all six were extremely unwell and in intensive care. With heroic efforts on the part of medical personnel over several weeks, the men all recovered, but lost fingers and toes. TGN1412 is an antibody molecule that attaches to a CD28 receptor on white blood cells of the immune system and interferes with the immune system in ways that are poorly understood. In 1996, a similar study using an antibody that attached to CD28 (as well as to CD3 and CD2) receptors, using one human subject, had similar results, but was not published. Had it been published then it may have prevented the ordeal of the six volunteers in 2006.

Even if flawed research does not always put people's lives at risk, it squanders money and effort.

The hallowed process of peer review may not be all it is cracked up to be. A prominent medical journal ran research past other experts in the field, and they failed to spot some deliberately inserted mistakes, even though they knew that they were being tested.

Ideally, research protocols should be registered in advance to prevent fiddling with experimental design midstream. Trial data should also be open to others to inspect and test. The most enlightened journals are becoming less averse to publishing less interesting papers, and some are encouraging replication studies. Younger scientists have a better understanding of statistics, and their use in analysing data needs to be extended. Peer review needs to be tightened so that science can correct its own mistakes and continue to command respect, rather than create barriers to understanding by shoddy research.

Sources

- Leader article: How science goes wrong, *The Economist*, 19 October 2013, p.11.
- Goldacre, B. (2012) *Bad Pharma*. Fourth Estate.
- http://www.newscientist.com/issue/2004.

DID YOU KNOW?

During the 19th century, there were many pseudoscientific claims, such as claims by Sylvester Graham, inventor of Graham crackers, that ketchup and mustard can cause insanity. Unfortunately, such pseudoscience still exists today. One 'celebrity' nutritionist has claimed that dark green leaves such as spinach are good for you, as they contain lots of chlorophyll, which is high in oxygen and so will give you more oxygen! She also says that certain foods are good sources of digestive enzymes. Another nutrition journalist has claimed that fructose is digested in the liver.

Where else will I encounter these themes?

1.1 YOU ARE HERE | 2.1 | 2.2 | 2.3 | 2.4 | 2.5 | 2.6

Let's start by considering the nature of the writing in the article. This extract is from the magazine *The Economist*, which is aimed at the general public.

1. Discuss the style of writing – has the writer made too many assumptions about the knowledge of readers? Do you think that the 'scientific method' should have been explained? Do you think that all readers understand why replicating scientific investigations is important?
2. Is it clear what makes an experiment 'shoddy'?

Now let's look at the concepts about scientific methodology and the biology underlying the information in the article.

3. Explain why an investigation that fails to support a hypothesis is not a failed investigation.
4. Describe the process of peer review.
5. The results of many clinical trials, where the new treatment does not appear to be more effective than the present available treatment or a placebo, are not published. However, trials that show the new treatment in a good light are. What do you think are the disadvantages of such 'cherry-picking'?
6. The discovery of the structure of DNA was a piece of curiosity-driven research. At the time, it was not envisaged that it would have any practical applications. In today's economic climate, scientists have to show that their proposed research will have some economic benefits, in order to obtain funding. Briefly outline the disadvantages of this approach.
7. State two problems that are faced by humans today, that will need science to help find the solutions.
8. Discuss how the statements (a) made by a celebrity nutritionist – that eating foods rich in chlorophyll gives us more oxygen, and that some foods are sources of digestive enzymes; and (b) made by a journalist – that fructose is digested in the liver, are incorrect.

Activity

Complete ONE of the following two activities.

1 When scientific research challenges major dogma (accepted theory) it is said to be revolutionary and cause a paradigm shift. Examples of paradigm shifts are the Darwin–Wallace theory of evolution by natural selection; Pasteur's germ theory of disease; the theory of biogenesis; and Mendel's theory of inheritance.

Select one of the above paradigm shifts, and do research, using the Internet, to find out more about it. Prepare and deliver to others in your class a short (three-minute maximum) presentation outlining the essence of the displaced dogma and the now-accepted theory that replaced it.

2 There have been recent examples of flawed scientific investigations that have done considerable harm, such as the suggested link between the measles vaccine and autism, and the investigation that showed that rats fed on GM potatoes were harmed. Choose one of these studies, or another flawed study that you are aware of, and do research, using the Internet, to find out more about it. Prepare and deliver to others in your class a short (three-minute maximum) presentation detailing what the study purported to show; in what way it was found to be flawed; the harm that it caused; and what happened to the researcher.

1.1 Practice questions

At A level, assessment of your practical skills is carried out through the practical endorsement. However, you will also find questions in your written examinations that test your knowledge and understanding of practical procedures. These are likely to form part of a whole question rather than entire questions.

1. Which row shows the most appropriate units to use when measuring the size of different samples? [1]

	height of person	length of leaf	diameter of artery	length of cell
A	metres	millimetres	centimetres	micrometres
B	metres	centimetres	millimetres	micrometres
C	metres	centimetres	micrometres	millimetres
D	metres	centimetres	millimetres	nanometres

2. A student estimated the percentage cover of four different species in a field. What type of graph would be the best way to present these results? [1]
   A a histogram
   B a kite graph
   C a line graph
   D a pie chart

3. A student investigated the effect of temperature on the rate of enzyme action. Which row correctly identifies the dependent variable, independent variable and control variables? [1]

	dependent variable	independent variable	control variable	control variable
A	rate of enzyme action	temperature	volume of solutions	substrate concentration
B	temperature	rate of enzyme action	volume of solutions	substrate concentration
C	rate of enzyme action	temperature	volume of solutions	intensity of light
D	volume of solutions	rate of enzyme action	temperature	substrate concentration

4. Figure 1 shows the scale on a graticule. What is the level of precision in the measurement *s* that can be made using this scale? [1]

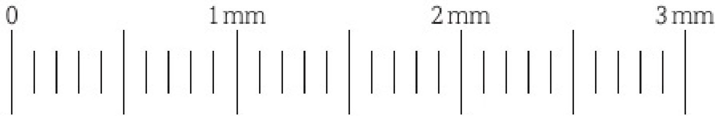


Figure 1 The scale on a graticule

   A 1 mm
   B 0.5 mm
   C 0.15 mm
   D 0.05 mm

5. A student investigated the rate of transpiration from a leafy shoot using a potometer. The student removed leaves from the shoot one at a time to investigate the effect of reduced surface area. As each leaf was removed, the student covered the damaged stem with petroleum jelly. The student identified a number of variables in this investigation:
   (i) leaf size
   (ii) loss of water from damaged stem
   (iii) light intensity
   (iv) temperature
   (v) species of plant used

   Which of these variables could be limiting factors for transpiration? [1]

   [Total: 5]

6. You are provided with seven beakers containing solutions of reducing sugar in beakers labelled 0.0, 0.2, 0.4, 0.6, 0.8, 1.0% and *X*. You are also provided with suitable reagents and glassware.
   (a) Outline how you would use the solutions provided to estimate the concentration of reducing sugar in solution *X*. [6]
   (b) State two limitations of your plan. [2]
   (c) Describe how you could modify your plan to make it more accurate. [5]

   [Total: 13]

7. Figure 2 shows a potometer.

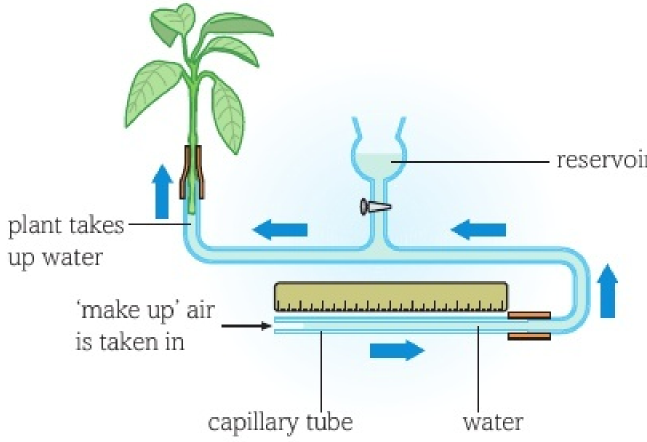


Figure 2

(a) Describe three precautions that should be taken to ensure the apparatus works properly. [3]

(b) You are asked to use the apparatus to investigate the effect of increasing air movement on the rate of transpiration. Draw a suitable blank results table, assuming you have a fan with three speed settings. [6]

(c) State the expected effect of increasing wind speed. [1]

(d) Suggest a suitable statistical test to assess whether your results match the expected results. [1]

[Total: 11]

8. You have been asked to measure the biodiversity of a field near your school. You decide to use a random sampling technique.

(a) Explain why you should use random samples. [2]

(b) Describe how you could generate random sample sites. [3]

(c) List the apparatus that you would need to take with you. [5]

(d) When you visit the site you realise that there are three distinct patterns of vegetation. Suggest how you should modify your plan. [3]

[Total: 13]

9. The table below shows the results of an investigation into the effect of reducing the number of leaves on the rate of transpiration.

number of leaves	rate of transpiration		
	1	2	3
8	65	62	64
7	53	27	56
6	42	45	44
5	45	31	32
4	25	26	27

(a) How could the table of results be improved? [1]

(b) One limitation of this experiment is that the leaves are not all the same size. How could you modify the experiment to overcome this limitation? [1]

(c) Identify one anomalous result and give a reason for your answer. [2]

(d) Do you think the results are reliable? Justify your answer by referring to the data. [9]

(e) Explain why the student collected three sets of data. [2]

[Total: 15]

10. Figure 3 shows an eyepiece graticule and a stage graticule.

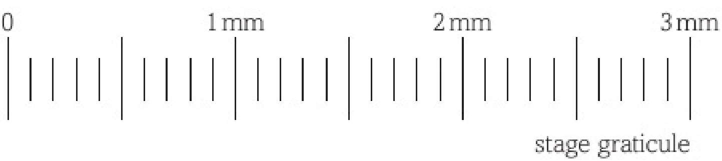


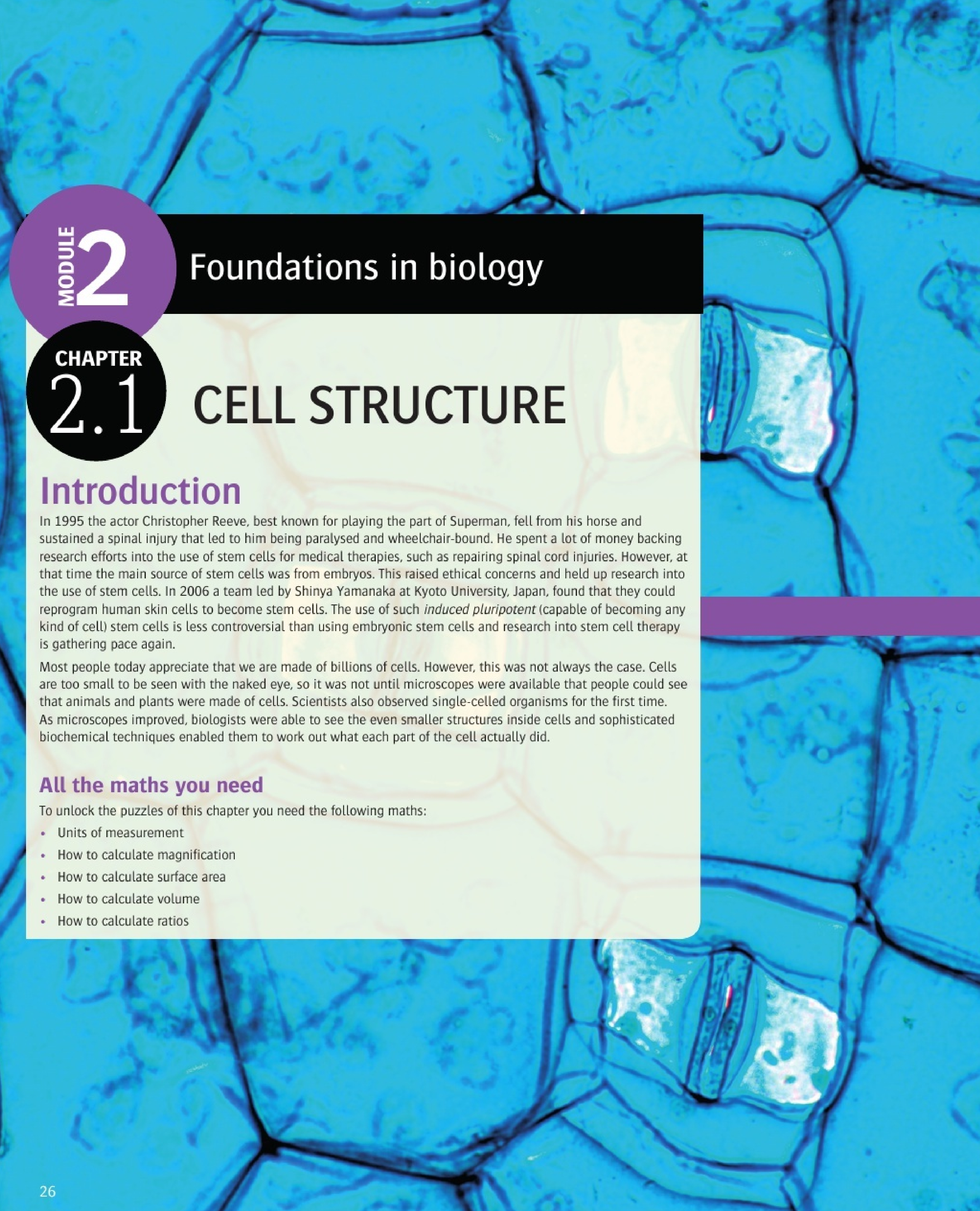
Figure 3

(a) What is the value of one eyepiece unit (epu)? [1]

(b) The diameter of a small artery was measured using the eyepiece graticule. It measured as 24 epu. What is the diameter of the artery? [2]

(c) Explain why it is important to use the same objective lens throughout the measuring process. [2]

[Total: 5]



MODULE 2 Foundations in biology

CHAPTER 2.1

CELL STRUCTURE

Introduction

In 1995 the actor Christopher Reeve, best known for playing the part of Superman, fell from his horse and sustained a spinal injury that led to him being paralysed and wheelchair-bound. He spent a lot of money backing research efforts into the use of stem cells for medical therapies, such as repairing spinal cord injuries. However, at that time the main source of stem cells was from embryos. This raised ethical concerns and held up research into the use of stem cells. In 2006 a team led by Shinya Yamanaka at Kyoto University, Japan, found that they could reprogram human skin cells to become stem cells. The use of such *induced pluripotent* (capable of becoming any kind of cell) stem cells is less controversial than using embryonic stem cells and research into stem cell therapy is gathering pace again.

Most people today appreciate that we are made of billions of cells. However, this was not always the case. Cells are too small to be seen with the naked eye, so it was not until microscopes were available that people could see that animals and plants were made of cells. Scientists also observed single-celled organisms for the first time. As microscopes improved, biologists were able to see the even smaller structures inside cells and sophisticated biochemical techniques enabled them to work out what each part of the cell actually did.

All the maths you need

To unlock the puzzles of this chapter you need the following maths:

- Units of measurement
- How to calculate magnification
- How to calculate surface area
- How to calculate volume
- How to calculate ratios

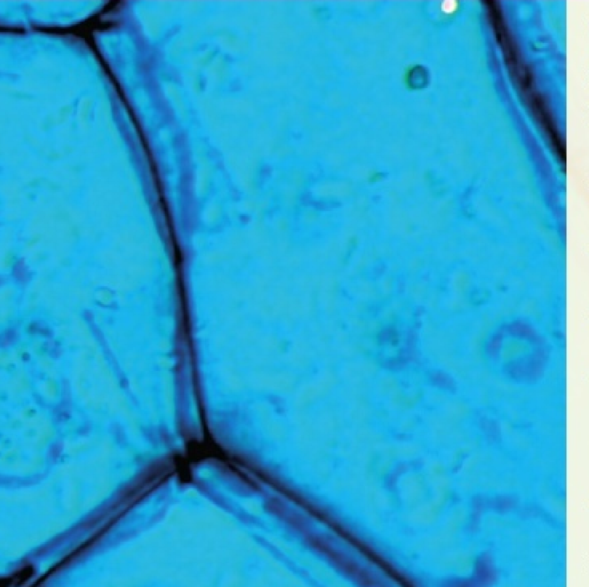
What have I studied before?

You should already know from GCSE:

- Cells are the building blocks of all living organisms
- Living organisms' metabolic processes, such as respiration, are carried out in their cells
- Cells are very small and can only be studied using a microscope
- All cells have common features such as:
  - a surface membrane that separates the cell's interior from the external environment and regulates what goes into and out of a cell
  - a jelly-like cytoplasm and a cytoskeleton
  - DNA that makes up the cell's genetic content (genome)
  - ribosomes where proteins are assembled
- There are differences between plant, animal and bacterial cells
- Within a developing organism undifferentiated stem cells become differentiated and specialised to carry out certain specific functions – we all began life as one cell and we all develop many types of cells in our bodies

What will I study later?

- The structure and properties of key biological molecules, including the phospholipids, proteins and carbohydrates that make up the cell membranes (AS)
- Enzymes, many of which catalyse chemical reactions inside cells (AS)
- The structure and functions of cell membranes – the membranes around the outside of cells and the membranes around some of their internal organelles (AS)
- The properties of nucleic acids, DNA and RNA, which are found in cells (AS)
- How cells reproduce and pass on their genetic material to their daughter cells (AS)
- How substances needed for life are transported to all the cells of large multicellular plants and animals (AS)
- The roles of special cells involved in defence against infectious disease (AS)



What will I study in this chapter?

- Microscopes, optical and electron, plus their advantages and disadvantages
- How slides and photomicrographs help us study cells
- The ultrastructure of cells – the structure and functions of the smaller parts within cells
- How organelles within cells work together, for example to make proteins
- How cells become differentiated and specialised for particular functions, how they are organised into tissues and that tissues are organised into organs
- More about the structure of prokaryotic cells and how they differ from eukaryotic cells

You will also learn how to:

- make slides of cells to examine using an optical microscope
- correctly draw low-power plans and high-power drawings of prepared slides of tissues
- calculate the size of cells and organelles as seen in photomicrographs or electron micrographs

Microscopes

By the end of this topic, you should be able to demonstrate and apply your knowledge and understanding of:

- the use of microscopy to observe and investigate different types of cell and cell structure in a range of eukaryotic organisms
- the difference between magnification and resolution

KEY DEFINITIONS

electron micrograph: photograph of an image seen using an electron microscope.
magnification: the number of times larger an image appears, compared with the size of the object.
organelles: small structures within cells, each of which has a specific function.
photomicrograph: photograph of an image seen using an optical microscope.
resolution: the clarity of an image; the higher the resolution, the clearer the image.

Magnification

Magnification describes how much bigger an image appears compared with the original object. Microscopes produce *linear* magnification, which means that if a specimen is seen magnified ×100, it appears to be 100 times wider and 100 times longer than it really is.

Resolution

Resolution is the ability of an optical instrument to see or produce an image that shows fine detail clearly. You may have a high-resolution television (called 'ultra-high definition' or UHD) and have noticed how clear and sharp the images on its screen are.

Optical microscopes

The development of optical (light) microscopes played a key role in our understanding of cell structure. They were the first sort to be used, and are still used in schools, colleges, hospitals and research laboratories because they are:

- relatively cheap
- easy to use
- portable and able to be used in the field as well as in laboratories
- able to be used to study whole living specimens.

Present-day light microscopes look different from the ones used in the 17th century, but both types rely on lenses to focus a beam of light.

Optical microscopes allow magnification up to ×1500, or in some types ×2000, which enables us to see clearly some of the larger structures inside cells. However, because their **resolution** is limited, they cannot magnify any higher while still giving a clear image.

- Optical microscopes use visible light, a part of the electromagnetic spectrum that has a wavelength of between 400 and 700 nm.
- The wavelength of visible light ranges from 400 to 700 nm so structures closer together than 200 nm (0.2 μm) will appear as one object.
- Ribosomes are very small, non-membrane-bound, cell **organelles** of about 20 nm diameter, and so they cannot be examined using a light microscope.

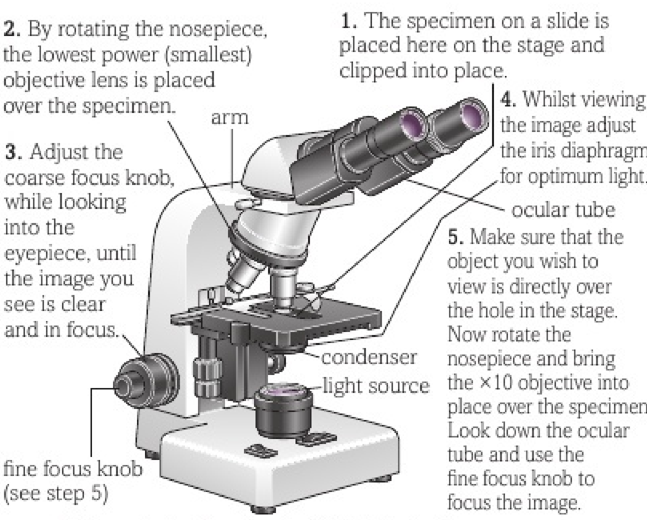


Figure 1 Annotated diagram showing how to use a light microscope. Note that when you carry a microscope you should hold it by its arm in one of your hands, whilst having your other hand under the base of the microscope.

Calculating magnification

total magnification = magnifying power of the objective lens × magnifying power of the eyepiece lens

LEARNING TIP

You do not need to learn how an image is formed by an optical microscope.

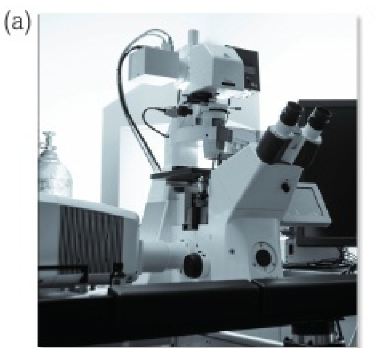
A photograph of the image seen using an optical microscope is called a **photomicrograph**. You will see an example of one in this chapter. Modern digital microscopes display the image on a computer screen.

Laser scanning microscopes

Laser scanning microscopes are also called confocal microscopes (see Figure 2).

- They use laser light to scan an object point by point and assemble, by computer, the pixel information into one image, displayed on a computer screen.
- The images are high resolution and show high contrast.
- These microscopes have depth selectivity and can focus on structures at different depths within a specimen. Such microscopy can therefore be used to clearly observe whole living specimens, as well as cells.
- They are used in the medical profession, for example to observe fungal filaments within the cornea of the eye of a patient with a fungal corneal infection, in order to give a swift diagnosis and earlier, and therefore more effective, treatment.
- They are also used in many branches of biological research.

(a)



(b)

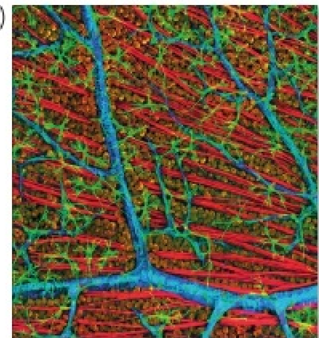


Figure 2 (a) A laser scanning microscope; (b) cells in the retina of the eye as seen with a laser scanning microscope (×64).

Electron microscopes

Electron microscopes use a beam of fast-travelling electrons with a wavelength of about 0.004 nm. This means that they have much greater resolution than optical microscopes and can be used to give clear and highly magnified images.

The electrons are fired from a cathode and focused, by magnets rather than glass lenses, on to a screen or photographic plate.

Fast-travelling electrons have a wavelength about 125 000 times smaller than that of the central part of the visible light spectrum. This accounts for an electron microscope's much better resolution compared with an optical microscope.

Transmission electron microscopes

- The specimen has to be chemically fixed by being dehydrated and stained.
- The beam of electrons passes through the specimen, which is stained with metal salts. Some electrons pass through and are focused on the screen or photographic plate.
- The electrons form a 2D black-and-white (grey-scale) image. When photographed this is called an **electron micrograph**. Transmission electron microscopes can produce a magnification of up to 2 million times, and a new generation is being developed that can magnify up to 50 million times.

Scanning electron microscopes

These were developed during the 1960s. Electrons do not pass through the specimen, which is whole, but cause secondary electrons to 'bounce off' the specimen's surface and be focused on to a screen. This gives a 3D image with a magnification from ×15 up to ×200 000. The image is black and white, but computer software programmes can add false colour. However, the specimen still has to be placed in a vacuum and is often coated with a fine film of metal.

Figure 3 A scanning electron microscope.

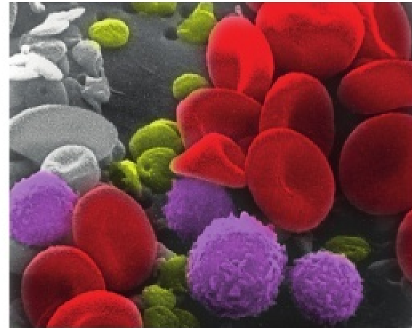


Figure 4 False-colour electron micrograph of blood cells. Erythrocytes are coloured red, lymphocytes magenta and platelets yellow (×1900).

Both types of electron microscope:

- are large and very expensive
- need a great deal of skill and training to use.

Specimens, even whole ones for use in SEMs, have to be dead, as they are viewed while in a vacuum. The metallic salt stains used for staining specimens may be potentially hazardous to the user.

Range of objects seen with and without microscopes

The eye, and optical and electron microscopes, are all optical instruments. Figure 5 shows the sizes of some objects that biologists may study, using these instruments. Note that the scale is *logarithmic* – it goes up in steps, where each is a 10-fold increase.

Electron microscope
Light microscope
Human eye

0.1 nm atom
1 nm lipids
10 nm ribosome protein
100 nm influenza virus
1 μm mitochondrion chloroplast bacterium
10 μm human cheek cells
human ovum
100 μm onion epidermis cell
1 mm *Amoeba*
10 mm
100 mm hen's egg
1 m

Figure 5 Relative sizes of some biological structures on a logarithmic scale, showing the scope of the electron microscope, light microscope and human eye for studying them.

DID YOU KNOW?

Your eye can distinguish objects that are about 0.3–0.5 mm apart. This is the limit of its resolution, but it gives you quite good visual acuity for 'everyday' objects. In the retina, at the back of the eye, are photosensitive cells called cones that work in bright light and produce this visual acuity (sharpness). You have about 200 000 cones per mm^2. Eagles and hawks have many more cones in their retinas, around 1 million per mm^2, and therefore have greater resolution and visual acuity. When you see a hawk hovering 20 m high over a roadside grass verge, it can clearly see an insect scurrying amongst that vegetation. An eagle can spot a rabbit 2 miles away and, although it is much smaller than you, its eyes are about the same size as yours.

LEARNING TIP

You need to really get to grips with the units mm, μm and nm and be able to convert one to the other.
In the next topic you will carry out some maths exercises that require you to use and convert these units.

Questions

1. If you were to examine a slide of a protoctist, using a ×40 objective lens and a ×15 eyepiece lens, what would be the total magnification of the protoctist?
2. List or make a table to show the advantages and disadvantages of optical microscopes.
3. Suggest the most useful type of microscope to observe each of the following:
   (a) living water-fleas in pond water during a biology field trip
   (b) cells taken from a cervical smear to be examined for abnormalities that may indicate cancer
   (c) virus particles
   (d) the inner structure of a mitochondrion
   (e) the ribosomes in a liver cell.
4. List or make a table to show the advantages and disadvantages of electron microscopes.
5. The wavelength of red light is 700 nm. How many times larger is this than the wavelength of electrons?
6. What is a logarithmic scale? Why do you think it is used for comparing sizes of biological structures?