

SURFING

NSW BIOLOGY

1&2

Module 1 Cells As the Basis Of Life

Module 2 Organisation Of Living Things

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Introduction

This book covers the Biology content specified in the NSW Biology Stage 6 Syllabus. Sample data has been included for suggested experiments to give you practice to reinforce practical work in class.

Each book in the *Surfing* series contains a summary, with occasional more detailed sections, of all the mandatory parts of the syllabus, along with questions and answers.

All types of questions – multiple choice, short response, structured response and free response – are provided. Questions are written in exam style so that you will become familiar with the concepts of the topic and answering questions in the required way.

Answers to all questions are included.

A topic test at the end of the book contains an extensive set of summary questions. These cover every aspect of the topic, and are useful for revision and exam practice.

Words To Watch

account, account for State reasons for, report on, give an account of, narrate a series of events or transactions.

analyse Interpret data to reach conclusions.

annotate Add brief notes to a diagram or graph.

apply Put to use in a particular situation.

assess Make a judgement about the value of something.

calculate Find a numerical answer.

clarify Make clear or plain.

classify Arrange into classes, groups or categories.

comment Give a judgement based on a given statement or result of a calculation.

compare Estimate, measure or note how things are similar or different.

construct Represent or develop in graphical form.

contrast Show how things are different or opposite.

create Originate or bring into existence.

deduce Reach a conclusion from given information.

define Give the precise meaning of a word, phrase or physical quantity.

demonstrate Show by example.

derive Manipulate a mathematical relationship(s) to give a new equation or relationship.

describe Give a detailed account.

design Produce a plan, simulation or model.

determine Find the only possible answer.

discuss Talk or write about a topic, taking into account different issues or ideas.

distinguish Give differences between two or more different items.

draw Represent by means of pencil lines.

estimate Find an approximate value for an unknown quantity.

evaluate Assess the implications and limitations.

examine Inquire into.

explain Make something clear or easy to understand.

extract Choose relevant and/or appropriate details.

extrapolate Infer from what is known.

hypothesise Suggest an explanation for a group of facts or phenomena.

identify Recognise and name.

interpret Draw meaning from.

investigate Plan, inquire into and draw conclusions about.

justify Support an argument or conclusion.

label Add labels to a diagram.

list Give a sequence of names or other brief answers.

measure Find a value for a quantity.

outline Give a brief account or summary.

plan Use strategies to develop a series of steps or processes.

predict Give an expected result.

propose Put forward a plan or suggestion for consideration or action.

recall Present remembered ideas, facts or experiences.

relate Tell or report about happenings, events or circumstances.

represent Use words, images or symbols to convey meaning.

select Choose in preference to another or others.

sequence Arrange in order.

show Give the steps in a calculation or derivation.

sketch Make a quick, rough drawing of something.

solve Work out the answer to a problem.

state Give a specific name, value or other brief answer.

suggest Put forward an idea for consideration.

summarise Give a brief statement of the main points.

synthesise Combine various elements to make a whole.

CELLS AS THE BASIS OF LIFE



In this module you will:

- Examine the structure and function of organisms at both the cellular and tissue levels.
- Describe how organisms facilitate the efficient provision and removal of materials to and from all cells.
- Explore biochemical processes through the application of the Working Scientifically skills processes.
- Investigate the study of microbiology and the tools that scientists use in this field.
- Use these tools to assist in making predictions and solving problems of a multidisciplinary nature.
- Engage with all the Working Scientifically skills for practical investigations involving the focus content to collect, process and analyse data and identify trends, patterns and relationships related to cell structure and function.



1 Assumed Knowledge Module 1

QUESTIONS

- Identify seven properties of living organisms.
- The cell is the basic unit of life. What structural features of cells are possessed by all living things?
- Draw a fully labelled diagram of a plant cell as seen under a light microscope.
- Draw a fully labelled diagram of an animal cell as seen under a light microscope.
- Identify the following parts of a light microscope and use by a person.

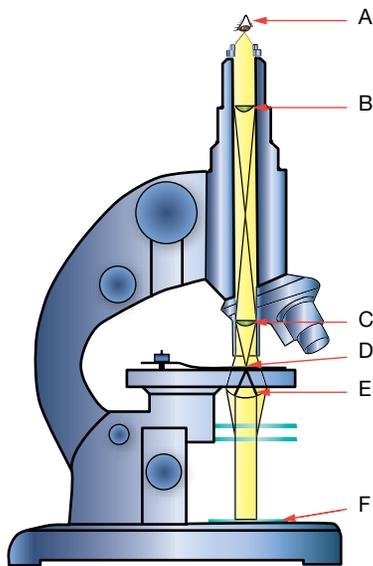


Figure 1.1 Light microscope.

- Describe one safety precaution you should follow while using a light microscope.
- What is the function of the nucleus of a cell?
- What is the function of the cell membrane?
- What is cytoplasm?
- Define protoplasm.
- Describe a chloroplast.
- Define photosynthesis.
- Which group of organisms can photosynthesise?
- Identify the materials required by multicellular organisms for photosynthesis.
- Why is photosynthesis an important process in ecosystems?
- The diagram shows the cellular structure of a prokaryote. Label the parts A to E.

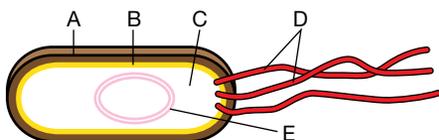


Figure 1.2 Typical prokaryote cell.

- Name the four basic groups of organic compounds.
- What are inorganic compounds?
- What is a trace element?
- What are the approximate proportions of these elements in living things?
 - Oxygen
 - Carbon
 - Hydrogen
 - Nitrogen
- What is the main difference between prokaryotic cells and eukaryotic cells?
- What is an ion?
- The graph shows the change in mass of 100 plant cells over several days.

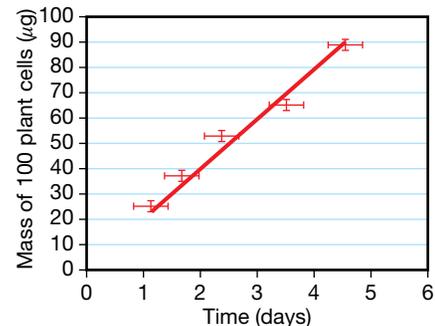


Figure 1.3 Change in mass of plant cells over time.

- Explain why there are bars with the plotted points.
 - Why are these bars used to show results?
- When gathering data, how do you calculate the mean?
 - The diagram shows diffusion of a gas across a room.

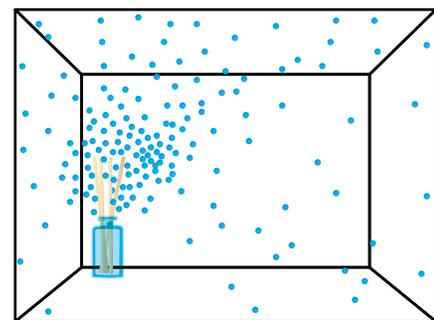


Figure 1.4 Diffusion of a gas in a room.

Define diffusion.

- The diagram shows a water molecule.

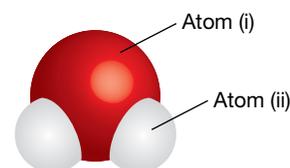


Figure 1.5 Water molecule.

- Identify atom (i) and atom (ii).
- What is the formula for water?

2 Prokaryotes

Prokaryotic organisms do not have membrane bound organelles, i.e. they do not have a nucleus, mitochondria or chloroplasts. They are the simplest of organisms on Earth, are very small, e.g. most have a diameter between 1 and 10 μm , are extremely abundant and survive in a wide variety of environments. The prokaryotic genome is in a circular DNA/protein single chromosome called the **nucleoid**. The nucleoid is in the **cytosol** (the intracellular fluid). Some prokaryote genes are found in **plasmids** which are small circular double stranded DNA molecules also in the cytosol. In prokaryotes most metabolic reactions occur in the cytosol. Many prokaryotes have **fimbriae** which are short hair-like structures on the surface that help the prokaryote stick to other cells or to a substrate. Many prokaryotes are surrounded by a **capsule** which is a sticky layer of protein or polysaccharide that can help the cell evade a host's immune system or help the cell adhere to other cells or surfaces. **Flagella** are used by motile bacteria for movement to move either towards or away from stimuli.

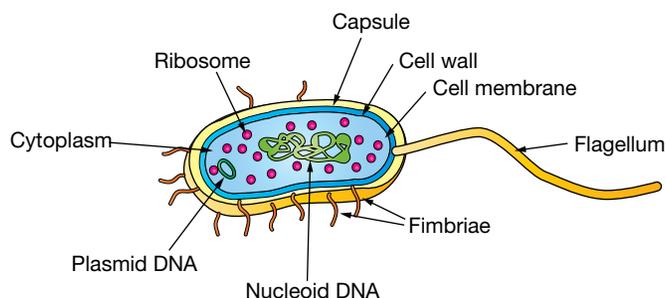


Figure 2.1 Generalised structure of a bacterial cell.

Classification of living things

Up until the 20th century people classified organisms into two groups – the **plant kingdom** and the **animal kingdom**. However, the invention of the electron microscope and new biochemical techniques has led to changes in our classification, especially with respect to the prokaryotes. The electron microscope has shown finer details of prokaryote cell structure and the analysis of DNA sequences has shown distinct differences between different types of prokaryotes.

In the 1970s a five kingdom system of classification, was introduced and the prokaryotes were placed in their own kingdom called **Monera**, which was subdivided into two groups – the bacteria (Schizophyta) and the cyanobacteria (Cyanophyta).

In 1977, Carl Woese found that the genetic make-up of the DNA sequences in bacteria separated the bacteria into two distinct groups. He named these two groups the

Bacteria (true bacteria) and the Archaeobacteria, which he later renamed the **Archaea**. The Archaea were once thought to only live in extreme condition environments, e.g. high temperatures (higher than 100°C), deep sea trenches, alkaline or acidic waters, digestive tracts of cows and termites, or produce methane when in a group cluster. The Archaea have since been found in all types of habitats. Woese then suggested that all life should be divided into three **domains** – the Eukaryota (have membrane bound organelles), the Bacteria and the Archaea.

It is believed the Bacteria and Archaea evolved from a common ancestor around 4 billion years ago.

Archaea

Some Archaea are called ‘extremophiles’ as they exist in extreme conditions and they can make a variety of protective molecules and enzymes to help them survive in these conditions, e.g. defensive molecules on their cell surface to prevent the action of acids, or enzymes to survive extreme hot or cold temperatures. **Methanogens** are Archaeans that produce methane gas as a waste product. They can live in an anaerobic environment (no oxygen) such as the intestinal tract of animals, or mud at the bottom of lakes and swamps. **Halophiles** live in salty environments, **thermophiles** live at extremely hot temperatures and **psychrophiles** live at unusually cold temperatures. Archaeans can use a variety of materials as a source of energy, e.g. hydrogen gas, carbon dioxide, sulfur and one group of halophiles contain bacteriorhodopsin which is a light sensitive pigment that can use light energy to provide chemical energy in a way different to photosynthetic plants.

Bacteria

Until recently the Bacteria were classified on shape (bacilli, cocci, spirilla), their reaction to the Gram stain, their ability to form spores and their method of energy production (heterotroph, autotroph, chemotroph).

The **Gram stain** was instigated by Christian Gram in 1884 and shows two different kinds of bacterial cell walls – Gram negative stain red, Gram positive stain purple.

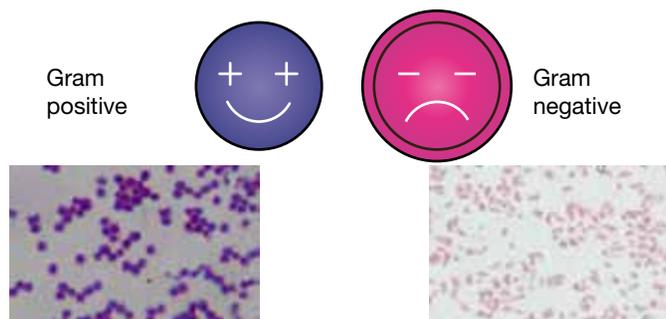


Figure 2.2 Gram stain.

Bacteria include cyanobacteria and nitrogen fixing bacteria.

Cyanobacteria (blue-green bacteria) are photosynthetic, containing the pigments chlorophyll *a* (like plants), phycocyanin (making them 'blue-green' and phycoerythrin (red colour) and they can fix nitrogen from the air. They can form stromatolites making them one of the oldest fossil organisms. An overabundance of cyanobacteria in a lake or river can lead to eutrophication when the decomposition of the dead cyanobacteria uses up all the available oxygen in the water and thus causes animal life to die. Cyanobacteria can also have mutualistic relationships with other organisms, e.g. with a fungus and form a lichen.

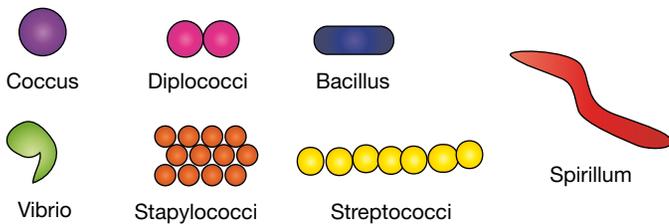


Figure 2.3 Naming bacteria.

Nitrogen fixing bacteria include both bacteria and cyanobacteria. *Rhizobia* bacteria live in the roots of legumes in a mutualistic relationship with the plant where they 'fix' nitrogen (N_2) in the air into compounds able to be used by the plant. The plant receives these nitrogen compounds and gives protection to the *Rhizobia*.

Deep sea bacteria have been found in many locations especially near the rift vents at mid ocean ridges. Both Bacteria and Archaea have been found in deep sea locations. Many bacteria which live in the deep sea are not necessarily found in the floor sediments or water – they live in the guts of deep dwelling animals. These bacteria release enzymes that degrade the food eaten by their host. The biotechnology industry is interested in deep sea bacteria as many produce degradative enzymes used to get food and there is large commercial potential for enzymes, e.g. in the laundry industry for enzymes that can break down and remove organic materials.

QUESTIONS

1. What is a prokaryote?
2. Distinguish between the nucleoid, the plasmid and the cytosol.
3. Outline the function of fimbriae.
4. What is the function of flagella?
5. How were the prokaryotes classified and then subdivided in the five kingdom system of classification?
6. Describe how technological advances have increased our understanding of prokaryotes.
7. Identify some of the extreme conditions where the Archaea are found.
8. What classification system was suggested by Carl Woese?
9. How are the Bacteria and Archaea related?
10. Describe one way in which the Eukaryota, Archaea and the Bacteria are different.
11. What is the difference between methanogens, thermophiles and halophiles?
12. The halophile, *Halobacterium*, which contains the light sensitive pigment bacteriorhodopsin, has been studied in depth by scientists. Suggest why this Archaea is of such interest to scientists.
13. The Gram stain was introduced in the 19th century when scientists knew very little about bacteria. Suggest why a staining method is still used as a part of bacterial taxonomy.
14. Describe cyanobacteria.
15. Use an example to show how the study of prokaryotic organisms can affect technology and society.
16. The Red Sea has periodic blooms of cyanobacteria that gave the sea its name. Which pigment in cyanobacteria would cause this effect?
 - (A) Chlorophyll.
 - (B) Phycoertherin.
 - (C) Phycocyanin.
 - (D) Saffarin.
17. Europa is one of the moons of Jupiter. It is covered in ice and recent evidence suggests there may be water underneath the ice kept liquid by hydrothermal vents. If life exists on Europa, of the following life forms, which would be the mostly likely to be present?
 - (A) Algae.
 - (B) Cyanobacteria.
 - (C) Worms.
 - (D) Archaea.
18. If an ancient halophile was found fossilised in a rock, what environment could be inferred to have existed at this location when this organism was alive?
 - (A) Tropical rainforest.
 - (B) Sulfur springs.
 - (C) High salt area.
 - (D) Deep sea vent.
19. When scientists decided to classify life into three domains, what were these domains?
 - (A) Eukaryota, Archaea, Bacteria.
 - (B) Plant, Animalia, Monera.
 - (C) Eukaryota, Protista, Monera.
 - (D) Archaea, Cyanobacteria, Eukaryota.
20. Many marine bacteria are aerobic, Gram negative, heterotrophic and bacilli shaped. Which of the following does *not* fit in with this description?
 - (A) Have rod shape.
 - (B) Stain purple with Gram stain.
 - (C) Use oxygen.
 - (D) Require organic materials to consume.

3 Eukaryotes

A eukaryote has membrane organelles, e.g. nucleus, mitochondria. Eukaryotes include single-celled organisms, e.g. some protista and some fungi and multicellular organisms, e.g. plants, animals, some protists and some fungi.

The oldest fossils of eukaryotes are around 2.1 billion years old. Analyses of DNA sequences suggest that the common ancestor of multicellular eukaryotes lived around 1.5 billion years ago.

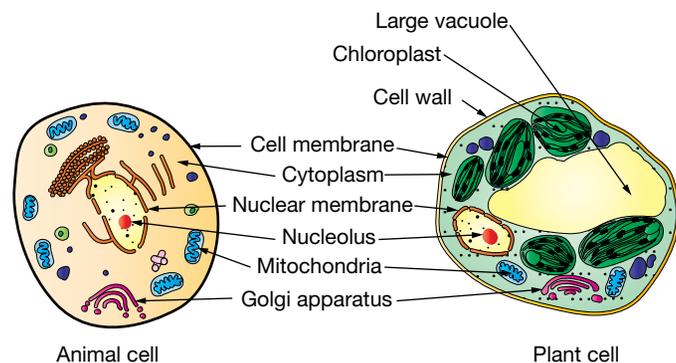


Figure 3.1 Generalised eukaryote cells.

Endosymbiont theory

Several areas of evidence support the **endosymbiont theory** which proposes that mitochondria and chloroplasts (and other plastids) were once small prokaryotes that were engulfed by larger cells and as either undigested prey or as a parasite lived in a symbiotic relationship with its host. Eventually the host and the endosymbiont became inseparable and eukaryotes evolved from this relationship.

Endomembrane system

The **endomembrane system** of eukaryotic cells includes the cell membrane, the nuclear membrane, the endoplasmic reticulum, the Golgi apparatus, lysosomes and other vesicles and vacuoles found within the cell. The arrangement of the internal membranes inside a eukaryotic cell divides the cell into compartments which have different environments suitable for different metabolic reactions. Enzymes are found on membranes, including the cell membrane, which means the membranes are directly involved in cellular reactions. The type of biochemical process that occurs depends on:

- The nature of the internal membrane.
- The arrangement of the internal membrane.
- The presence of specific enzymes.
- Environmental factors.

Eukaryotes have specialised organelles that carry out biochemical processes, e.g. chloroplasts carry out photosynthesis, mitochondria carry out cellular respiration, lysosomes contain hydrolytic enzymes that break down macromolecules and recycle the cell's own organic material. Smooth endoplasmic reticulum is involved in the synthesis of lipids, metabolism of carbohydrates and the detoxification of drugs and poisons and rough endoplasmic reticulum is involved in the production of proteins.

The **nucleus** contains most of the genes of eukaryotic cells. Chloroplasts and mitochondria also contain DNA which is like the chromosomes of bacteria.

QUESTIONS

1. What is meant by a eukaryote cell?
2. Which kingdoms have organisms that are eukaryotes?
3. What is the age of the oldest eukaryote fossils?
4. When did the common ancestor of multicellular eukaryotes most likely live on Earth?
5. Outline the endosymbiont theory.
6. Identify the parts of the endomembrane system of eukaryotes.
7. List the factors that control biochemical processes in cells.
8. Identify the organelle that is responsible for the following functions.
 - (a) Cellular respiration.
 - (b) Photosynthesis.
 - (c) The breakdown of macromolecules and recycling of organic material in the cell.
 - (d) Metabolism of carbohydrates and detoxification of drugs and poisons.
9. Where is DNA found in a eukaryotic cell?
10. The diagram shows the structure of *Euglena*, a single celled organism commonly found in pond water.

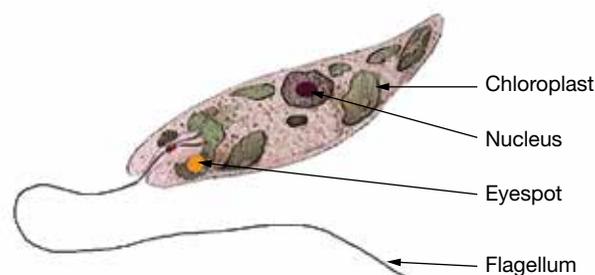


Figure 3.2 *Euglena*.

What would be the best description of *Euglena*?

- (A) A prokaryote.
- (B) A bacteria.
- (C) A plant.
- (D) A eukaryote.

4 The Modern Light Microscope

The compound light microscope operates on the main principle that an objective lens with a very short focal length can form a highly magnified real image of the object. Visible light passes through the specimen and then a series of lenses. The resolution of the microscope is limited by the shortest wavelength of light used to view the specimen.

Images from a light microscope can be captured with a camera to produce a **photomicrograph**. Digital images can be shown directly on a computer screen.

Table 4.1 Features of the modern light microscope.

Feature	Light microscope
Magnification	Effective up to 1000×
Resolution	Up to 0.2 μm
Stains	Allows the use of many different coloured stains to identify substances, structures and provide contrast for easier viewing.
Living specimen	The light microscope allows viewing of living specimen and processes occurring within a cell or within an organism.
Mounting	The specimen is mounted on a glass slide in air.
Focusing	By glass lenses.
Energy source for viewing	A beam of light is passed through the specimen.

How a light microscope works

The objective lens is brought close to the specimen to create an enlarged image of the object. The image is inverted. In most modern light microscopes the eyepiece is a compound lens near the back of an eyepiece tube. Light travels from the light source up the microscope to form an image at the eye.

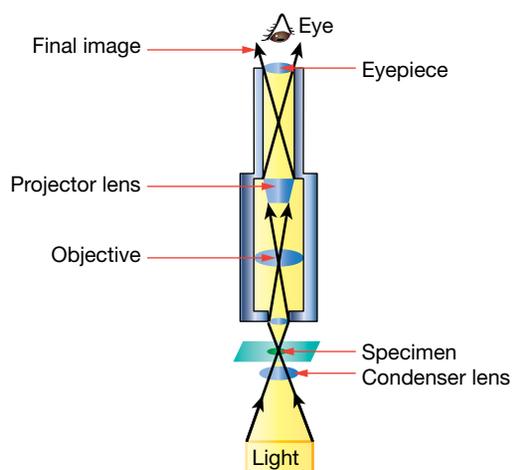


Figure 4.1 How light travels through a light microscope.

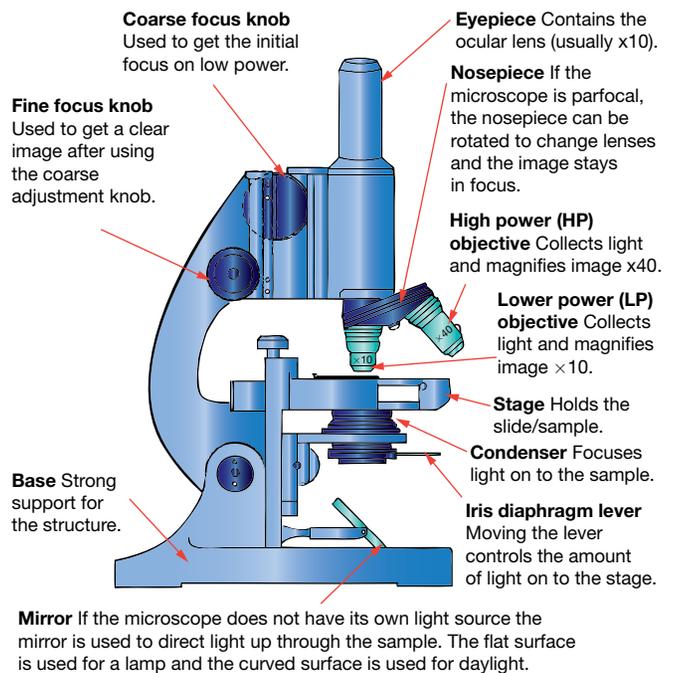


Figure 4.2 Features of the modern light microscope.

Advantages of a light microscope

The main advantages of light microscopes are that:

- Living cells can be observed.
- Coloured stains can be used.
- The specimens are easy to prepare.
- The microscopes are relatively inexpensive (compared to the cost of an electron microscope).
- Their size means they are relatively easy to store.

Disadvantages of a light microscope

The main disadvantages of the light microscope are:

- Its limited magnification (effective magnification begins to reduce after 1000×
- Its limited resolution.

During the 20th century many different illumination techniques and other developments have increased the detection power of the light microscope for observing living cells.

Phase contrast microscopes

The phase contrast microscope uses interference rather than absorption of light to increase the contrast in unstained cells by amplifying variations in density within the cell. It improves our ability to study living, unpigmented cells in biological and medical research. Many dyes and stains stop chemical processes in cells which means the phase contrast microscope has improved our ability to see detail in living cells, e.g. the process of cell division. Frits Zernike was awarded with the Nobel Prize in Physics, 1953 for the development of phase contrast illumination.

Fluorescence microscopes

In the fluorescence microscope the specimen is illuminated through objective lenses with a narrow set of light wavelengths. The specimen either fluoresces in its natural form, e.g. chlorophyll or has been treated with fluorescing chemicals or antibodies. The fluorescent substances absorb UV light and emit visible light so that the fluorescence shows the location of specific molecules in the cell.

Other illumination techniques

Other illumination techniques include:

- **Bright field** – passes light through the specimen and contrast comes from the absorbance of light in the specimen. If the specimen is unstained or unpigmented there is little contrast.
- **Cross-polarised** – contrast occurs when polarised light is rotated through the sample.
- **Confocal** – is a type of fluorescence microscopy using optical sectioning by scanning lasers of fluorescently stained specimens.

Stains and dyes used with the light microscope

There are many stains that are used to highlight structures being viewed under a light microscope.

- **Gram staining** – uses several stains, e.g. crystal violet, iodine, fuchsin or safranin to stain cell cells to differentiate bacteria into Gram positive (purple/blue colour occurs) and Gram negative (pink/red colour occurs). This is usually the first step in identifying bacteria.
- **DAPI** – is a fluorescent nuclear stain that shows a blue fluorescence when bound to DNA and viewed with ultraviolet light.
- **Eosin** – is used as a counterstain with haematoxylin in H&E staining and gives a pink/red colour to cytoplasmic material, red blood cells and cell membranes.

QUESTIONS

1. Outline the basic principle behind the operation of a light microscope.
2. When using a light microscope identify the function of:
 - (a) The condenser.
 - (b) The iris diaphragm.
3. What restricts the resolution of the light microscope?
4. Construct a table to compare the advantages and disadvantages of a light microscope.
5. What is a photomicrograph?
6. Outline the benefit of the phase contrast microscope.

7. Outline the benefit of fluorescence microscopy.
8. The table shows some objects and their sizes.

Object	Size (μm)
Frog egg	1100
<i>Paramecium</i>	100
Plant epithelial cell	60
Human ovum	10
Red blood cell	7 to 8
Mitochondrion	1.1
Nanobes	0.025

Identify which objects could be seen with the naked eye and/or light microscope.

9. Use an example to show how the use of stains and the light microscope have increased our understanding of processes in cells.
10. Most modern light microscopes in schools have at least two different objectives. Name objectives you have used and state the purpose of each objective.
11. What is a parfocal microscope?
12. The diagram shows red blood cells as seen and as drawn by a biology student using a light microscope.

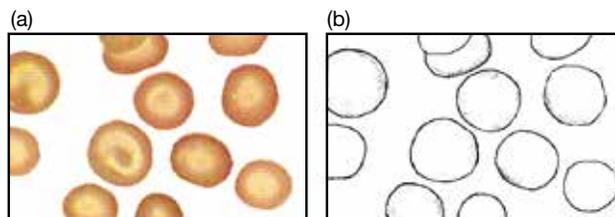


Figure 4.3 Red blood cells seen using a light microscope. (a) Seen under a light microscope. (b) As drawn by a student.

Red blood cells are usually about 6 to 8 micrometres in diameter.

Which objective would the student most likely have been using to view these cells as seen in the diagram? Explain your answer.

13. Explain why the Gram stain is used.
14. What is the correct order of the movement of light when an object is viewed under a light microscope?
 - (A) Light \rightarrow condenser \rightarrow objective \rightarrow eyepiece \rightarrow eye
 - (B) Light \rightarrow objective \rightarrow condenser \rightarrow eyepiece \rightarrow eye
 - (C) Light \rightarrow objective \rightarrow eyepiece \rightarrow condenser \rightarrow eye
 - (D) Eye \rightarrow eyepiece \rightarrow objective \rightarrow condenser \rightarrow light
15. What is the limit of resolution of a light microscope?
 - (A) 0.2 centimetres
 - (B) 0.2 millimetres
 - (C) 0.2 micrometres
 - (D) 0.2 nanometres

5 The Electron Microscope

The electron microscope provides greater detail about cell structure. The electron microscope sends a stream of electrons through a vacuum. The electron beam is focused by electromagnets, magnified by an objective lens and projected onto a fluorescent screen or photographic film. Since the beam of electrons has a much shorter wavelength than visible light resolution is greatly improved, e.g. approximately 0.002 nm, although in practical situations it can be limited to 2 nm. Typically they provide a resolution of 0.5 nm (400 times better than a light microscope) and magnify up to 500 000 times.

Transmission electron microscopes

The transmission electron microscope (TEM) uses the same basic principles as the light microscope with a beam of electrons passing through the specimen instead of a beam of light. The TEM is used to study the internal ultrastructure of cells. The specimen is stained with heavy metal atoms which attach to particular cellular structures, preserved by a chemical fixative, embedded in plastic then cut into exceedingly thin slices (50 to 100 nm). You can see objects to the order of several nanometres (10^{-9} m) increasing the capacity for medical, biological and materials research. Many organelles were discovered using the TEM, e.g. ribosomes. One of the main disadvantages of the TEM is that the method of specimen preparation kills the cells and the specimen is viewed in a vacuum. This means living cells cannot be viewed. Specimen preparation also produces artefacts and structural features that do not exist in living cells.

Table 5.1 Features of the electron microscope.

Feature	Electron microscope
Magnification	Up to 300 000 \times .
Resolution	Approximately 0.0005 μ m.
Stains	Heavy metal stains identify substances, structures and provide contrast for easier viewing. No colour stains.
Living specimen	The electron microscope does not allow viewing of living cells.
Mounting	Metal background in a vacuum chamber.
Focusing	By electromagnetic lenses.
Energy source for viewing	A beam of electrons is passed through the specimen.

Scanning electron microscopes

The scanning electron microscope (SEM) emits an electron beam which is rapidly passed back and forth over the surface of the specimen. Surface variations alter the pattern of the scattering of the electrons and the pattern is recorded, amplified and transmitted to a TV monitor.

This gives a three-dimensional detailed view of the surface of the specimen. The image has great depth of field.

QUESTIONS

- Outline why the electron microscope has better resolution than the light microscope.
- Outline the basic principle behind the operation of an electron microscope.
- Construct a table to compare the advantages and disadvantages of a light microscope.
- Discuss the importance of the transmission electron microscope.
- Discuss the importance of the scanning electron microscope.
- Identify the type of microscope that was used to view each of the following.

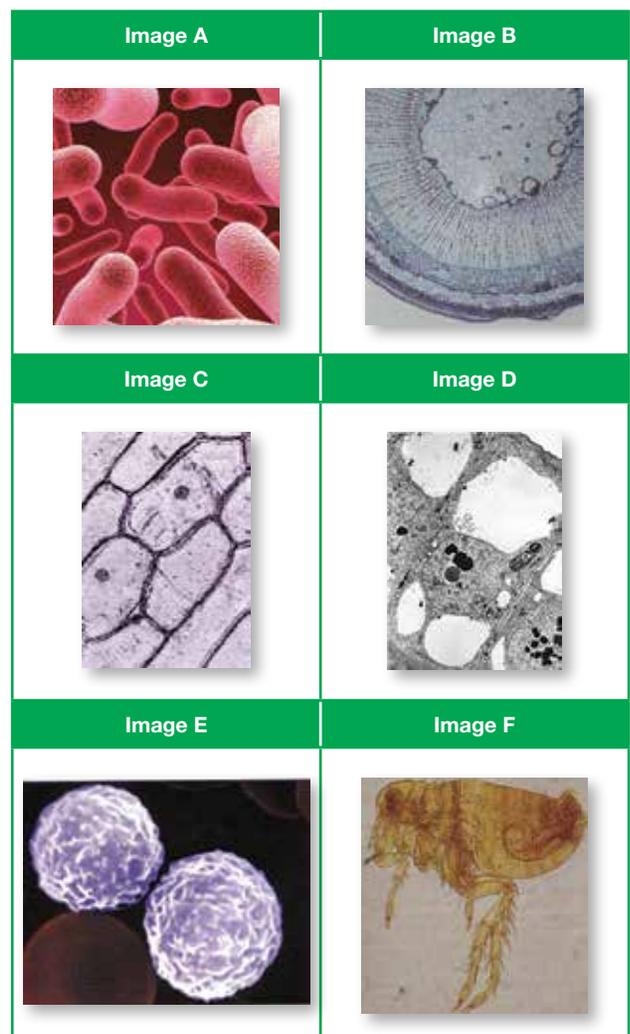


Figure 5.1 Images from different types of microscopes.

- What is the limit of resolution of a transmission electron microscope?
 - 0.5 centimetres.
 - 0.5 millimetres.
 - 0.5 micrometres.
 - 0.5 nanometres.

6 The Stereo Microscope

The **stereo microscope** has two ocular lens so that the left eye and the right eye each view the object from different angles. This sends two images to the brain. The brain then produces a three-dimensional image of the object. Thus the microscope gives a stereoscopic view or 'stereo' view of an object.

Some compound microscopes have double eyepieces which make viewing easier but give both eyes the same image and do not provide the three-dimensional images provided by stereo microscopes.

To use a stereo microscope the eyepieces need to be adjusted. There is variation in the distance between the pupils of the eyes of each person and this means that the position of the eyepieces of the stereo microscope need to be corrected to suit the requirements of each person.

Different parts of a specimen come into focus when focusing up and down while using a stereo microscope. Organisms are three-dimensional and the stereo microscope allows viewing of the different levels of the specimen. For larger specimens every feature cannot be seen clearly at the same time.

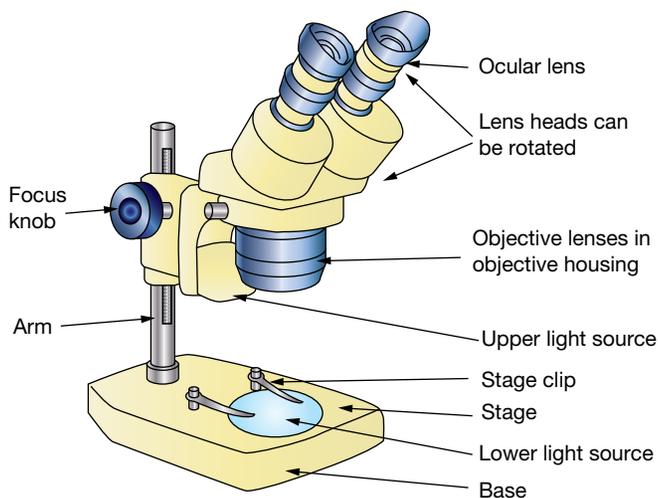


Figure 6.1 Stereo microscope.

Resolution, illumination and magnification of a stereo microscope

A stereo microscope usually has poor resolution as the lenses are a distance away from the object.

Most stereo microscopes use reflected light from an upper and/or lower light source. It is important when observing live organisms, e.g. worms and insects not to leave the light source shining on the organism for too long. The heat can kill the organism.

Most stereo microscopes have magnifications from $10\times$ to $40\times$.

Uses of a stereo microscope

A stereo microscope is often used when dissecting small organisms and is very useful when you need to carefully observe the external features of a specimen.

If a camera and/or computer is attached to the stereo microscope the image can be recorded and displayed on a screen. Since the camera is only connected to one eyepiece the result will be a 2-D image.

QUESTIONS

1. What is the distinguishing feature of a stereo microscope?
2. When are you likely to use a stereo microscope?
3. The diagram shows a side view of a stereo microscope.

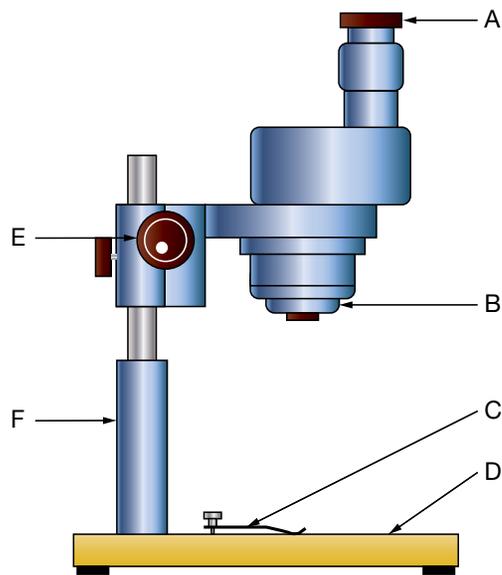


Figure 6.2 Side view of a stereo microscope.

Identify the parts of the microscope labelled, A, B, C, D, E and F.

4. Explain how a compound microscope with double eyepieces may not provide a three-dimensional image to form a 'stereo microscope'.
5. Explain why many stereo microscopes have poor resolution.
6. Compare the magnification of a typical stereo microscope with a typical monocular microscope found in schools.
7. When studying crystals, e.g. salt crystals under a stereo microscope, explain why it is advisable to put a piece of black paper beneath the crystal.
8. Explain why some parts of an organism may appear out of focus while studying a specimen with a stereo microscope.
9. Which of the following would *not* be suitable to be observed using a stereo microscope?
(A) Red blood cells. (B) Honey bee.
(C) Eucalypt leaf. (D) Salt crystal.

7 Technology and the Development Of the Cell Theory

The historical development of the cell theory has often depended on major advances in technology, taking over 300 years (1665-1838) to be fully established. Advances in technology still influence the development of knowledge about cells, organelles, biochemical reactions and how each structure contributes to life processes.

Development of the light microscope

The grinding of glass lenses with the invention of glass spectacles in the 14th century in Italy was one of the first steps leading to our ability to see cells. In the 1590s Hans and Zacharias Janssen are believed to have created the first compound light microscope. They placed several lenses in a tube and realised the object near the end of the tube could be magnified more than using a single lens in a magnifying glass.

But it was not until 1665 that **Robert Hooke** (1635-1703) with his compound microscope, first observed cells. In 1674 **Anton van Leeuwenhoek** (1632-1723) saw micro-organisms, '*animalcules*' using a new single lens microscope he had developed. Leeuwenhoek made better lenses with greater curvature which gave better magnification. His grinding and polishing techniques produced lenses able to magnify up to 270 diameters. **Jan Swammerdam** (1637-1680) was one of the first people to use a microscope in dissections and discovered animal cells describing the blood cells of a louse.

Joseph Jackson Lister (1786-1869) produced achromatic lenses and placed them at suitable distances to give better resolution without spherical aberrations that blurred the image. He published his findings in 1830 and by 1832 was one of the best microscope makers of his time.

Henri Dutrochet (1776-1847) studied plant cells using a microscope and named the process of osmosis and noted that the processes in all living things are similar. In 1824 he suggested that living things might be made of cells and therefore that cells may be the basic unit of all living things. In 1833 **Robert Brown** (1773-1858) was the first person to recognise the nucleus in plant cells and, although the structure had been observed in animal cells, he gave it a name.

In 1838 **Matthias Schleiden** (1804-1881) and **Theodor Schwann** (1810-1882) proposed that all living things are made of cells. They suggested that each cell functions not only independently of other cells but also in cooperation with them so that organisms can function as a whole. Schleiden studied different plant structures under a microscope and recognised the importance of the nucleus. Schwann viewed animal tissues under the microscope, especially nervous and muscle tissues.

Rudolf Virchow (1821-1902) encouraged his students to use microscopes, especially in pathological anatomy and in animal experiments. He found cells in bone and in connective tissue and in 1855 published a work that proposed that the origin of cells was the division of pre-existing cells. This is the last point of the cell theory.

In 1893 **August Kohler** developed a new way to illuminate the field of view in a light microscope. The Kohler illumination scheme provided even lighting which enabled high quality photomicrographs to be taken.

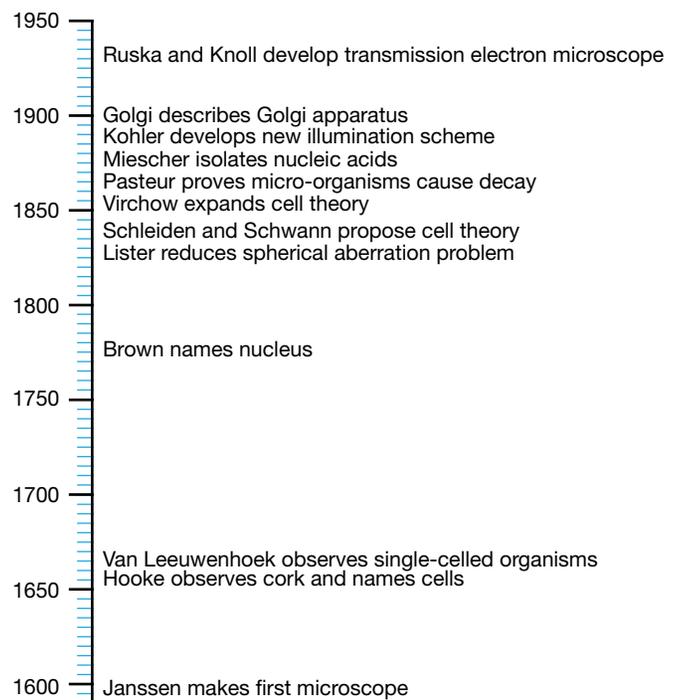


Figure 7.1 Timeline for the development of technology and the cell theory.

Development of staining techniques

The development of specific dyes was needed to help scientists discover the internal structures of a cell. Many dyes will stain specific chemicals, assisting in the identification of chemicals in cells, and the contrast in colour helps to outline different structures. For example, in 1849 the carminic acid procedure was developed by Hartung and in 1863 the haematoxylin colouring was developed by Waldeyer. These methods led to the observation of cell and nuclear division. Carmine is a bright red pigment obtained from scale insects, e.g. cochineal scale and is used to stain glycogen or animal starch red and is used to colour nuclei and chromosomes. Haematoxylin stains cell nuclei blue.

Iodine is used in chemistry as a test for starch as the starch/iodine complex is a dark blue-black colour. Lugol's iodine solution is a cell stain used in biology to make the cell nuclei more visible.

New methods in making slides have also enabled cells to be seen more clearly. Fixing specimens, e.g. in formalin will harden tissues before embedding in wax and then cutting. New instruments, e.g. **microtomes** have enabled extremely thin slices of specimens to be cut more easily.

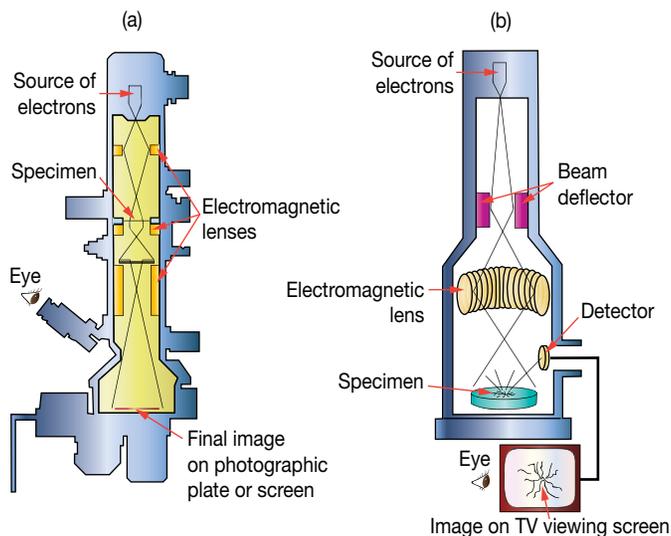


Figure 7.2 Comparing electron microscopes. (a) Transmission electron microscope. (b) Scanning electron microscope.

Development of the electron microscope

In 1932 **Ernst Ruska** (1906-1988) and **Max Knoll** (1897-1969) developed the first transmission electron microscope (TEM). The development of the electron microscope with its greater resolution and greater magnification compared to the light microscope has enabled scientists to see more details within the cell, e.g. the structure of the nuclear membrane, as well as discovering new parts, e.g. ribosomes. The transmission electron microscope uses a beam of electrons instead of a beam of light and uses electromagnets instead of glass lenses. It shows the internal appearance of cells.

Magnification refers to making things appear larger and **resolution** is the ability to distinguish between two points and makes detail appear more clearly.

QUESTIONS

- Why is the development of the light microscope associated with the development of glass lenses?
- When were cells first observed and why was this possible?
- Outline the contribution of Anton van Leeuwenhoek to the development of the microscope.
- What was the significance of Kohler's development of a new illumination method?
- Which aspect of the cell theory was not correctly understood by Schleiden and Schwann?
- What was the contribution of Virchow?

- Construct a table to show the contributions of at least six people to the cell theory giving the year, the person and their contribution.
- Describe the evidence used by Hooke to support the beginning of the cell theory.
- Discuss how the electron microscope has assisted our understanding of cell structure.
- Ernst Abbe (1840-1905) developed many optical instruments and from experiments worked out a formula to determine the resolution limit of a microscope. He showed resolution was inversely proportional to the wavelength of light. Use this information and the fact that electron beams have much shorter wavelengths than the wavelengths of visible light to compare the resolution of a light microscope with the resolution of an electron microscope.
- Distinguish between magnification and resolution.
- Draw a timeline to show the stages in the development of the cell theory including the work of Hooke, Leeuwenhoek, Dutrochet, Brown, Schleiden and Schwann and Virchow.
- Assess the impact of the development of technology on the development of the cell theory.
- The diagram shows a cross-section of a leaf as seen under low power light microscope.

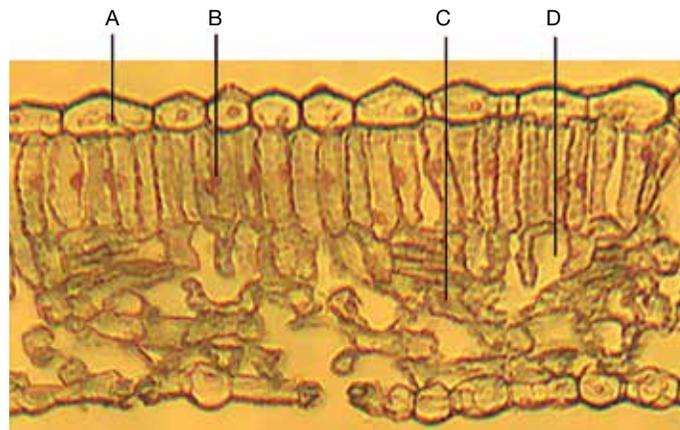


Figure 7.3 Cross-section of a leaf, low power light microscope.

- Which arrow does *not* point to a nucleus of a cell?
 (A) A (B) B (C) C (D) D
- Which statement is *not* part of the cell theory?
 (A) All living things are made of cells.
 (B) All cells have a nucleus.
 (C) All cells come from pre-existing cells.
 (D) The cell is the basic unit of life in which the processes of living occur.
 - Name the scientist who proposed that all cells come from pre-existing cells.
 (A) Hooke.
 (B) Brown.
 (C) Leeuwenhoek.
 (D) Virchow.

8 Developing the Cell Theory

The cell theory states the following.

- All living things are made of cells and of substances produced by cells.
- All cells come from pre-existing cells.
- The cell is the basic unit in which the processes of living take place.

The development of the cell theory is linked with the invention of technology that enabled scientists to see cells and to investigate the properties of cells.

Humans have made observations and recorded their findings about living things since the first mark was put on a cave wall, clay tablet or sheet of papyrus paper. These observations record the **macroscopic appearance** of living things – the features observed with the naked eye.

The development of glass lenses and the construction of the first microscope using a glass lens enabled scientists to observe the **microscopic appearance** of living things. The first cells were observed and the ideas about the structure of the building blocks of life were forever changed.

Hans and Zacharias Janssen

Hans and Zacharias Janssen are believed to have created the **first compound light microscope** around the 1590s. They placed several lenses in a tube and realised the object near the end of the tube could be magnified more than using a single lens in a magnifying glass.



Zacharias
Janssen

Francesco
Redi

Anton van
Leeuwenhoek

Robert
Brown

Figure 8.1 Janssen, Redi, van Leeuwenhoek and Brown.

Robert Hooke

In 1663 Robert Hooke observed cork under a microscope and introduced the term ‘**cell**’. He published his microscopical observations in 1665 in his book *Micrographia*. This book led to public interest in microscopy.

Francesco Redi

In 1668 Francesco Redi published the results of his experiment with insects which was one of the first steps in proving that living things do not arise from **spontaneous generation**. He showed that fly maggots did not spontaneously arise from dead meat as meat kept in jars covered with gauze did not get maggots.

Anton van Leeuwenhoek

Anton van Leeuwenhoek produced higher quality lenses that gave greater magnification and aided the development of the light microscope. He is considered to be the ‘Father of Microbiology’. In 1674 he was the first to observe and describe **single celled organisms** which he called *animalcules*. In 1676 when he sent his drawings of single-celled organisms to the Royal Society of London, his credibility was questioned. In 1680 his observations were vindicated after others observed the unicells. He discovered and made drawings of protozoa, bacteria, the vacuole of the cell, the banded pattern of muscle fibres and spermatozoa.

Robert Brown

Robert Brown described the **nucleus** in cells of the orchid and gave the structure its name. He travelled to Australia in 1801 on the HMS *Investigator* as the naturalist at the request of the commander of the vessel, Matthew Flinders. He collected many specimens and left Australia in 1805. In 1831 he read a paper about the cell nucleus to the Linnaean Society and published this work in 1833. Although the nucleus had been drawn by others, e.g. van Leeuwenhoek and Franz Bauer, Brown gave the structure its name. His observations of the random movement of pollen grains led to the naming of the phenomena now known as Brownian motion.

Matthias Schleiden

In 1838 Matthias Schleiden wrote *Contributions to Phytogenesis* and proposed that different parts of plants are made of cells. With Schwann he was the first to propose the **cell theory**. He also recognised the importance of the cell nucleus and its possible relationship with cell division.

Theodor Schwann

Theodor Schwann noted that parts of animals are made of cells and that non-cellular parts, e.g. nails, feathers and tooth enamel had a cellular origin. In 1839 he extended Schleiden’s cell theory to animals and proposed that all living things are made of cells and cell products. The cell was the basic unit of life. This is now called the **Schleiden and Schwann cell theory**. Schwann also observed the cells associated with nerve fibres which are now called Schwann cells.

Rudolf Virchow

In 1855 Rudolf Virchow published a work that proposed that the origin of cells was the division of pre-existing cells and the cell theory was expanded to include the point that every cell originated from another living cell like it. This rejected the concept of spontaneous generation. Virchow is known as the ‘father of modern pathology’ and he developed a standard method of autopsy procedure.



Matthias Schleiden



Theodor Schwann



Rudolf Virchow

Figure 8.2 Matthias Schleiden, Theodor Schwann and Rudolf Virchow.

Louis Pasteur

In 1861 Louis Pasteur published his experiments demonstrating that fermentation was caused by micro-organisms which finally disproved the theory of spontaneous generation. The experiment also supported the germ theory.

Friedrich Miescher

In 1869 Friedrich Miescher isolated nucleic acids which he called *nuclein* from the nuclei of white blood cells. This was the first time DNA had been purified and led to investigations into its composition, properties and structure.

Camillo Golgi

In 1898 Camillo Golgi described the Golgi apparatus by staining cells with silver nitrate. At first some believed the structure was an optical illusion caused by the staining technique. The invention of the electron microscope in the 20th century proved the existence and shape of this organelle.



Louis Pasteur



Friedrich Miescher



Camillo Golgi

Figure 8.3 Louis Pasteur, Friedrich Miescher and Camillo Golgi.

Max Knoll and Ernst Ruska

In 1932 Max Knoll and Ernst Ruska invented the transmission electron microscope. The higher magnification and higher resolution meant greater details of the ultrastructure of cells could be observed and analysed and new structures were discovered, e.g. ribosomes.

QUESTIONS

- State the cell theory.
- Construct a table to summarise the historical development of the cell theory.
- Explain how the invention of the light microscope is linked with the development of the cell theory.
- Explain why Anton van Leeuwenhoek is known as the ‘Father of Microbiology’.
- Suggest why Leeuwenhoek’s discovery of animalcules was at first disbelieved and explain why it was finally accepted.
- Outline the discovery and naming of the nucleus.
- What are the two points of the cell theory proposed by Schleiden and Schwann?
- What is the theory of spontaneous generation?
- Discuss how the theory of spontaneous generation was finally disproved.
- How is the theory of spontaneous generation linked with the development of the cell theory?
- (a) Why did people question the actual existence of the Golgi body in cells?
(b) What evidence proved the existence of the Golgi body?
- Explain how the invention of the electron microscope aided the development of knowledge about cell structure.
- Who was the first person to isolate DNA?
(A) Robert Hooke. (B) Robert Brown.
(C) Rudolf Virchow. (D) Friedrich Miescher.
- The timeline shows events in the development of the cell theory. From this timeline which event occurred before the nucleus was named?
(A) Single celled organisms were observed.
(B) Golgi apparatus was described.
(C) Nucleic acids were isolated.
(D) Cell theory was expanded.

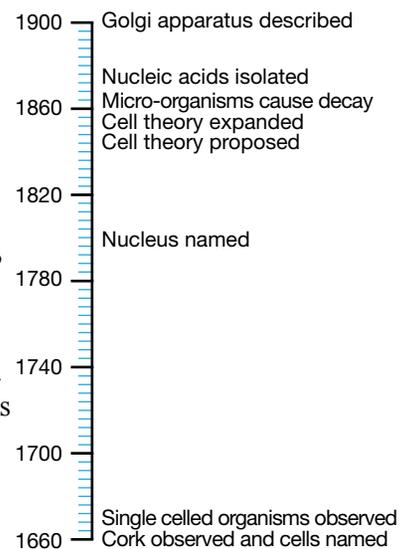


Figure 8.4 Cell theory timeline.

9 Experiment – The Light Microscope

Biology students need to be able to use a monocular light microscope. The light microscopes found in schools are compound microscopes with magnifying lenses used in series to provide an enlarged image of a specimen. Light is transmitted through the specimen to reach the eye of the observer. The specimen needs to be very thin to allow light to pass through. A simple light microscope like the one used by van Leeuwenhoek has one lens.

The eyepiece lens on most microscopes is $10\times$ and some microscopes have interchangeable eyepieces, e.g. $5\times$ or $10\times$. There is usually a choice of at least two objective lenses, for example $10\times$ and $40\times$. The overall magnification is calculated by multiplying the degree of magnification of the objective lens by the degree of magnification of the eyepiece lens. The $100\times$ objective is an oil immersion lens and can only be used with a special oil between the slide and the lens.

Many school light microscopes are **parfocal** which means that a specimen viewed under low power will automatically be in focus or near focus when the nosepiece is rotated from low power to high power.

Rules for using a light microscope

There are several important **rules** to follow when using a light microscope.

1. You must always use two hands when carrying a microscope. One should hold the arm and the other hand needs to be beneath the base.
2. Never touch the lenses with your fingers.
3. When you have finished using your microscope the nosepiece needs to be rotated so the low power objective is facing the stage and then lower the objective down to the stage.
4. Never leave a slide on the stage.
5. Place the dust cover over the microscope and return the microscope to its proper location.
6. If you made a wet mount slide, make sure your work area is clean and dry and all glass slides and cover slips are returned to their proper location at the end of the experiment.

Setting up a microscope

1. Place the microscope on the desk or bench with the arm *towards* you. This means that the stage is facing *away* from you.
2. Turn the coarse adjustment knob and raise the nosepiece.
3. Rotate the nosepiece so that the low power objective is facing the stage.

4. Look through the eyepiece and adjust the mirror and the diaphragm so that there is a bright circle of light visible.
5. Place the slide on the stage. Stage clips can be used.
6. Look at the stage from the side and lower the low power objective using the coarse adjustment knob. Stop before the objective touches the slide.
7. Look through the eyepiece and slowly turn the coarse adjustment knob until the image comes into focus.
8. Use the fine adjustment knob to bring the image into sharp focus.

QUESTIONS

1. Identify each part of the compound light microscope shown in Figure 9.1.

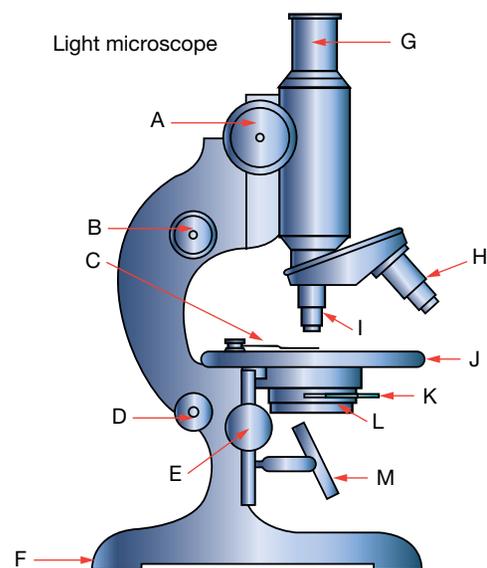


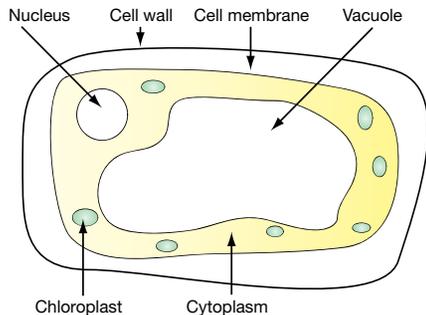
Figure 9.1 Compound light microscope.

2. Explain why it is necessary to always use two hands when carrying a light microscope.
3. Explain why it is important to view the stage from the side when you lower the low power objective and then always move the objective lens away from the microscope slide.
4. If an eyepiece has a magnification of $10\times$, what is the overall magnification for each of these objective lenses?
 - (a) Low power objective $10\times$.
 - (b) High power objective $40\times$.
5. Outline the function of the iris diaphragm.
6. Explain why it is usually necessary to adjust the amount of light after changing objective lenses.
7. How does a stereo microscope vary from a light compound microscope?
8. Explain why it is important not to touch the lens with your fingers and discuss what you would do if you found a fingerprint on the lens.

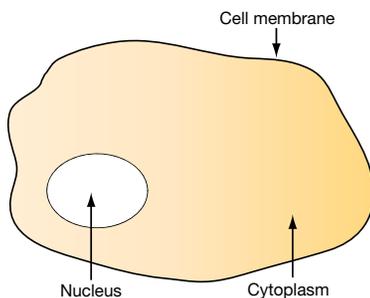
Answers

1 Assumed Knowledge Module 1

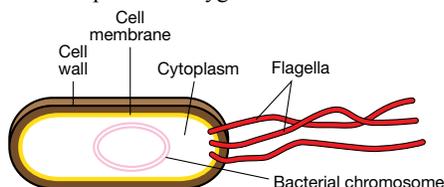
- Living organisms can: 1. Breathe. 2. Assimilate food and synthesise organic molecules. 3. Grow. 4. Reproduce. 5. Respond to stimuli from their environment. 6. Excrete. 7. Locomotion
- The cells of living things have a cell membrane, cytoplasm and DNA.
- Plant cell.



- Animal cell.



- A = eye of person using the light microscope, B = ocular lens, C = objective lens, D = specimen, E = condenser lens, F = light source
- When using a light microscope, you should always wear shoes with covered toes, as the microscope is heavy and if you drop it you could damage exposed skin on your feet.
- The nucleus stores information needed to control all cell activities.
- The cell membrane surrounds the cell contents from the external environment and controls the substances that can leave or enter the cell.
- Cytoplasm is a general term for the contents of a cell outside the nucleus and within the cell membrane.
- Protoplasm is the semi-fluid transparent substance that makes up the living matter of plant and animal cells including the nucleus and cytoplasm.
- A chloroplast is a green organelle found in green tissues of plants that captures sunlight in photosynthesis to manufacture sugars from carbon dioxide and water.
- Photosynthesis is a process where the energy of sunlight is used to convert carbon dioxide and water into sugars and oxygen.
- Groups of organisms that can photosynthesise include plants, algae and photosynthetic bacteria.
- Carbon dioxide and water are needed for photosynthesis using light energy and in the presence of chlorophyll.
- Photosynthesis is important in ecosystems as it converts light energy into chemical energy to begin most food chains on Earth and also provides oxygen which is needed for respiration.



- The four basic groups of organic compounds are proteins, carbohydrates, lipids and nucleic acids.
- Inorganic compounds are molecules that do not contain carbon (excluding some carbonates and simple oxides of carbon).
- A trace element is an element essential for life but needed in small amounts.
- (a) Oxygen = 65%
(b) Carbon = 19%
(c) Hydrogen = 10%
(d) Nitrogen = 3%
- Prokaryotes do not have a nucleus while eukaryotes have a nucleus.
- Ions are charged particles eg H^+ , Na^+ , Cl^- .
- (a) The bars with the plotted points are error bars. The bars shows errors for both variables (time and mass).
(b) Error bars are used to show the uncertainty of the measurement which will depend on the instrument being used. Error bars can be used to show the statistical significance of the results. Error bars are useful when drawing the line of best fit.
- The mean is the average of the numbers obtained by adding all the numbers and then dividing by how many numbers are present.
- Diffusion is the movement of particles from an area of high concentration of particles to an area of low concentration of particles.
- (a) Atom
(i) is oxygen and Atom
(ii) is hydrogen.
(b) Formula for water is H_2O

2 Prokaryotes

- Prokaryotic organisms do not have membrane bound organelles, i.e. they do not have a nucleus, mitochondria or chloroplasts.
- The nucleoid is the part of a prokaryotic cell where the genetic information (usually a single circular chromosome) is found. A plasmid is a small circular double stranded DNA molecule. The cytosol is the intracellular matrix of the cell.
- Fimbriae are short hair-like structures on the surface of some prokaryotes that help the cell stick to other cells or to a substrate.
- Prokaryotes use flagella for movement, e.g. to either move towards or away from stimuli.
- In the five kingdom system of classification, the prokaryotes are placed in their own kingdom called Monera, which is subdivided into two groups – the bacteria (Schizophyta) and the cyanobacteria (Cyanophyta).
- Up until the 20th century people classified organisms into two groups – the plant kingdom and the animal kingdom. However, the invention of the electron microscope and new biochemical techniques has led to changes in our classification, especially with respect to the prokaryotes. The electron microscope has shown finer details of prokaryote cell structure and the analysis of DNA sequences has shown distinct differences between different types of prokaryotes. This has led to a reclassification of the prokaryotes, based on this new evidence.
- The Archaea live under extreme conditions, e.g. high temperatures, deep sea trenches, alkaline or acidic waters, digestive tracts of cows and termites, or produce methane when in a group cluster.
- Woese suggested that all life should be divided into three domains – the Eukaryota (have membrane bound organelles), the Bacteria and the Archaea.
- It is believed the Bacteria and Archaea are related in that they both evolved from a common ancestor around 4 billion years ago.
- The Eukaryota, Archaea and Bacteria are different in the composition of the cell wall. Bacteria have a wall made of peptidoglycan, Archaea have a wall made of pseudopeptidoglycan or protein only and Eukaryotic plants have a wall made of polysaccharide and Eukaryotic fungi have a cell wall made of chitin.
- Methanogens produce methane gas as a waste product and live in anaerobic environments such as the intestinal tract of animals, or mud at the bottom of lakes and swamps. Halophiles live in salty environments and thermophiles live at extremely hot temperatures.

12. *Halobacterium* is a halophile that can use its light sensitive pigment bacteriorhodopsin to use light energy to provide chemical energy in a way different to photosynthetic plants. This is of great interest to scientists as it shows that photosynthetic organisms do not necessarily have to start food chains and food webs. There are other organisms capable of performing this feat without chlorophyll.
13. The Gram stain shows two different kinds of bacterial cell walls – Gram negative stain red and Gram positive stain purple and hence it is still useful in bacterial taxonomy as it is a way of showing a natural division of bacteria into two distinct groups.
14. Cyanobacteria are photosynthetic bacteria, containing the pigments chlorophyll, phycocyanin and phycoerythrin and they can fix nitrogen from the air.
15. Prokaryotic organisms are the bacteria. A study of the bacteria found in deep sea locations shows that many produce degradative enzymes that are used to get food. The biotechnology industry is interested in these bacteria as there is large commercial potential for enzymes, e.g. in the laundry industry for enzymes that can break down and remove organic materials. Thus the study of these bacteria could have a dramatic impact not only on society and our attitude to be able to clean and launder clothing, but also have an economic impact providing jobs in the workforce and company income.
16. B
17. D
18. C
19. A
20. B

3 Eukaryotes

1. A eukaryote cell has membrane bound organelles, e.g. it has membrane bound nucleus.
2. Eukaryotes include the animal kingdom, the plant kingdom, the protista kingdom and the fungi kingdom.
3. The oldest fossils of eukaryotes are around 2.1 billion years old.
4. The common ancestor of multicellular eukaryotes most likely lived on Earth around 1.5 billion years ago.
5. The endosymbiont theory proposes that mitochondria and chloroplasts (and other plastids) were once small prokaryotes that were engulfed by larger cells and as either undigested prey or as a parasite lived in a symbiotic relationship with its host. Eventually the host and the endosymbiont became inseparable and eukaryotes evolved from this relationship.
6. The endomembrane system of eukaryotes includes the cell membrane, the nuclear membrane, the endoplasmic reticulum, the Golgi apparatus, lysosomes and other vesicles and vacuoles found within the cell.
7. Biochemical processes in cells are controlled by:
 1. The nature of internal membranes.
 2. The arrangement of internal membranes.
 3. The presence of specific enzymes.
 4. Environmental factors.
8.
 - (a) Mitochondria are responsible for cellular respiration.
 - (b) Chloroplasts are responsible for photosynthesis.
 - (c) Lysosomes break down macromolecules and recycle organic materials in the cell.
 - (d) Smooth endoplasmic reticulum is involved in the metabolism of carbohydrates and the detoxification of drugs and poisons.
9. In eukaryotic cells DNA is found in the nucleus, mitochondria, chloroplasts and other plastids.
10. D

4 The Modern Light Microscope

1. The compound light microscope passes visible light through the specimen and then through a series of lenses. It operates on the main principle that an objective lens with a very short focal length can form a highly magnified real image of the object.
2.
 - (a) The condenser focuses light on to the sample.
 - (b) The iris diaphragm controls the amount of light on to the stage.
3. The resolution of the microscope is limited by the shortest wavelength of light used to view the specimen (wavelengths of visible light are 400 to 700 nm).

Advantages of light microscope	Disadvantages of light microscope
Can view living specimens and observe processes in cells and within an organism.	Limited magnification, e.g. effectively up to 1000 \times .
Allows the use of many different coloured stains to identify substances, structures and provide contrast for easier viewing.	Resolution limited to 0.2 micrometres.
Specimens are easy to prepare, stain and observe in a short time frame.	
Relatively inexpensive and class sets can be purchased for use in schools.	
Light microscopes are not particularly large and are easy to store, e.g. in schools.	

5. A photomicrograph is a photograph made through a microscope.
6. The main benefit of the phase contrast microscope is that it increases the contrast in unstained cells. It improves our ability to study living, unpigmented cells in biological and medical research. Many dyes and stains stop chemical processes in cells which means the phase contrast microscope has improved our ability to see detail in living cells, e.g. the process of cell division.
7. The main benefit of fluorescence microscopy is that the fluorescence shows the location of specific molecules in the cell. Natural fluorescing substances, e.g. chlorophyll can be located or the specimen can be treated with fluorescing chemicals or antibodies to locate other particular section or substances in cells.

Object	Size (μm)	Viewing
Frog egg	1100	Naked eye and light microscope
<i>Paramecium</i>	100	Light microscope
Plant epithelial cell	60	Light microscope
Human ovum	10	Light microscope
Red blood cell	7 to 8	Light microscope
Mitochondrion	1.1	Light microscope
Nanobes	0.025	Cannot be seen under a light microscope – need an electron microscope

9. Stains and the light microscope have greatly increased our understanding of the processes in cells. For example, pH indicators which will show acidic or basic conditions can be used to show how the activity of micro-organisms change the pH balance of their environment such as bacterial action causing the souring of milk.
10. Objectives found on school light microscopes include 10 \times and 40 \times . The 10 \times objective is a low power lens used for first viewing the specimen to locate the area to be examined. The 40 \times objective is a high power lens and is used to make detailed observations of the specimen.
11. A parfocal microscope allows you to quickly focus using the lower power objective and then swivel the nosepiece to observe the specimen under a higher power objective without only minimal adjustment of the fine focus knob.
12. Red blood cells are very small – they have a diameter of 6 to 8 micrometres. This means the student would have been using a high power objective, e.g. 40 \times in the light microscope set-up to clearly observe these cells as seen in the diagram.
13. The Gram stain is usually used as the first step in identifying bacteria. It differentiates bacteria into two groups – Gram negative and Gram positive.
14. A
15. C

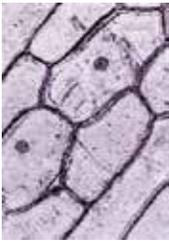
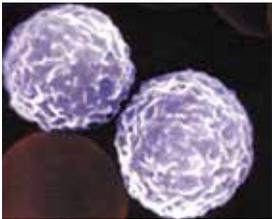
5 The Electron Microscope

1. The electron microscope has better resolution than the light microscope as it uses a beam of electrons to view the specimen. Since the beam of electrons has a much shorter wavelength than visible light resolution is greatly improved, e.g. approximately 0.002 nm, although in practical situations it can be limited to 2 nm. Resolution is inversely related to the wavelength of radiation used by the microscope for imaging.

2. The electron microscope sends a stream of electrons through a vacuum. The electron beam is focused by electromagnets, magnified by an objective lens and projected onto a fluorescent screen or photographic film.

Advantages of electron microscope	Disadvantages of electron microscope
Increased magnification, e.g. up to 300 000 \times . Increased resolution, e.g. approximately 0.0005 μm SEM shows a three-dimensional image of the specimen.	Cannot view living specimens and observe processes happening in cells. Cannot use coloured stains. Relatively expensive and are not available in schools.

4. The main benefit of the TEM is that it shows the internal ultrastructure of cells at higher magnification and resolution than the light microscope. You can see objects to the order of several nanometres (10^{-9} m) which means new organelles have been discovered using the TEM, e.g. ribosomes and there is an increased capacity for medical, biological and materials research.
5. The main benefit of the SEM is that it gives a three-dimensional detailed view of the surface of the specimen. The image has great depth of field and gives a view of the actual appearance of the specimen.

Image A	Image B
	
Scanning electron microscope	Light microscope
Image C	Image D
	
Light microscope	Transmission electron microscope
Image E	Image F
	
Scanning electron microscope	Light microscope

7. D

6 The Stereo Microscope

- The stereo microscope is easily identified as it has two ocular eyepieces that each view the object from different angles. There are two separate optical paths for viewing.
- You are likely to use a stereo microscope when dissecting small organisms or when you need to observe the external features of a specimen.

3. The parts of the stereo microscope are:

A = ocular lens
B = objective lens
C = stage clip
D = stage
E = focus knob
F = arm

- If a compound microscope has two eyepieces which give the same image then the image will not be three-dimensional and give a 'stereo' image. To provide a three-dimensional image there must be two images each from slightly different viewing angles.
- In a stereo microscope the lens is a distance away from the object and this gives poor resolution.
- A typical school stereo microscope has a magnification ranging from $10\times$ to $40\times$ while a typical school monocular microscope has a magnification from $40\times$ to $400\times$.
- Salt crystals are light coloured and usually translucent. The dark colour beneath the crystal is needed to provide contrast so that the crystal can be properly viewed.
- When using a stereo microscope some parts of an organism may not be in focus as the specimen is three-dimensional with several levels. The microscope is focusing on one level making other levels appear out of focus.
- A

7 Technology and the Development Of the Cell Theory

- The light microscope uses glass lenses to magnify objects. Thus the development of the light microscope is tightly linked with the ability to produce high quality glass lenses.
- The first cells were observed in 1665, when Robert Hooke used his compound microscope to observe cork.
- Anton van Leeuwenhoek made better lenses with greater curvature which gave better magnification. He also improved grinding and polishing techniques for making lenses.
- August Kohler's illumination method gave an even distribution of light in the field of view. This meant that better and higher quality photomicrographs could be taken.
- Schleiden and Schwann did not correctly understand how cells arose.
- Virchow proposed that all cells come from pre-existing cells which provided the last point in the cell theory.

Year	Person	Contribution
1665	Hooke	Observed cells in a slice of cork
1678	Leeuwenhoek	Observed micro-organisms
1824	Dutrochet	Living things are made of cells
1833	Brown	Observes nucleus in plant cells and names structure as nucleus
1838	Schleiden and Schwann	Tissues in all living things made of cells and cells function both independently and in cooperation
1858	Virchow	Cells come from pre-existing cells

- Hooke observed cells in a slice of cork using his compound microscope, providing evidence that the tissue consists of cells.
- The electron microscope has enabled scientists to see more details within the cell, e.g. the structure of the nuclear membrane, as well as discovering new parts, e.g. ribosomes.
- Since the resolution of a microscope is inversely proportional to the wavelength of the radiation – the smaller the wavelength, the bigger the resolution. Since the wavelength of a beam of electrons is much smaller than the wavelength of visible light the electron microscope has a much higher resolution than a light microscope.
- Magnification refers to making things bigger while resolution is the ability to distinguish between two points and see fine detail.

- 1665 Robert Hooke described cells in cork
 - 1678 Leeuwenhoek saw microbes
 - 1824 Dutrochet – living things made of cells
 - 1833 Brown named nucleus in plant cells
 - 1838 Schleiden and Schwann – cell theory
 - 1858 Virchow – cells from pre-existing cells

13. The cell theory states that: 1. All living things are made of cells. 2. All cells come from pre-existing cells. 3. The cell is the basic unit in which the processes of living take place. The development of these ideas was only possible as instruments and stains were developed so that scientists were able to see cells and their internal structure. For example, when Robert Hooke placed a slice of cork under his microscope, he was the first person to see these ‘many little boxes’, which he called cells. The discovery of staining techniques allowed the processes of cell and nuclear division to be seen. The development of the cell theory underpins our understanding of life on Earth, its structure, its physiology and why it behaves the way it does. Thus the development of technology has had a tremendous impact on the development of the cell theory as the theory could only expand as the technology allowed us to see what was present and how it behaved.
14. D
15. B
16. D

8 Developing the Cell Theory

1. The cell theory states that: 1. All living things are made of cells and of substances produced by cells. 2. All cells come from pre-existing cells. 3. The cell is the basic unit in which the processes of living take place.

Date	Person	Contribution
1590s	Hans and Zacharias Janssen	Created the first compound light microscope.
1663	Robert Hooke	Observed cork under a microscope and introduced the term ‘cell’.
1668	Francesco Redi	Published the results of his experiments with insects showing living things do not arise from spontaneous generation.
1674	Anton van Leeuwenhoek	Improved the quality of lenses and aided the development of the light microscope. Discovered unicells, e.g. protozoa and bacteria. Discovered the vacuole and drew the banded pattern of muscle fibres and spermatozoa.
1833	Robert Brown	Published a paper naming and describing the cell nucleus in orchids.
1838	Matthias Schleiden	Proposed that different parts of plants are made of cells. This became part of the Schleiden and Schwann cell theory.
1839	Theodor Schwann	Extended Schleiden’s cell theory to include animals. Schleiden and Schwann cell theory: 1. All living things are made of cells and cell products. 2. The cell is the basic unit of life.
1855	Rudolf Virchow	Extended the cell theory to include that every cell originated from a living pre-existing cell.
1861	Louis Pasteur	Fermentation experiments finally disprove the theory of spontaneous generation.
1869	Friedrich Miescher	Isolated DNA for the first time.
1898	Camillo Golgi	Described the Golgi apparatus by staining cells with silver nitrate.
1932	Max Knoll and Ernst Ruska	Invented the transmission electron microscope which gave greater resolution and magnification to study the ultrastructure of cells.

3. The invention of the light microscope is tightly linked with the development of the cell theory. When Robert Hooke used his compound microscope to view cork he discovered the cellular nature of plants and called the structures ‘cells’. When van Leeuwenhoek used improved lenses to observe unicells he was not initially believed. It was the invention and development of light microscopes with higher magnification and resolution that enabled scientists to observe and study the cellular nature of living things.

4. Anton van Leeuwenhoek produced higher quality lenses and made many microscopes. The improved resolution and magnification of his microscopes meant he was able to observe smaller objects and he was the first to draw and describe single celled organisms, e.g. protozoa and bacteria. This work led to him being called the ‘Father of Microbiology’.
5. Leeuwenhoek’s discovery of animalcules was at first disbelieved as the idea of single celled organisms did not fit in with the early 17th century concept of ‘life’. His work was finally accepted six years later when others observed unicells under the microscope.
6. Several scientists observed and drew diagrams of cells showing the presence of a nucleus, e.g. Anton van Leeuwenhoek and Franz Bauer. Robert Brown observed the nucleus in orchid cells and named the structure in 1831.
7. Schleiden and Schwann proposed that: 1. All living things are made of cells and of substances produced by cells. 2. The cell is the basic unit and building block of life.
8. The theory of spontaneous generation proposed that living things arose from non-living matter, e.g. maggots from dead flesh, rats from rubbish bins.
9. Fermentation experiments carried out by Louis Pasteur finally disproved the theory of spontaneous generation. He showed that micro-organisms were responsible for fermentation and bacteria caused the growths in boiled nutrient broths.
10. The cell theory states that all living cells come from pre-existing cells and clashes with the theory of spontaneous generation which proposes that living things can arise from non-living matter. As the cell theory developed, the theory of spontaneous generation had to be abandoned.
11. (a) Camillo Golgi discovered and used a new staining technique using silver nitrate to observe cells, e.g. in nervous tissue and to identify cell structures. Many believed that the body he found inside cells was an optical illusion caused by his staining technique.
(b) The invention of the electron microscope which had greater magnification and resolution than the light microscope proved the existence and showed the ultrastructure of the Golgi body.
12. The invention of the electron microscope has greatly aided the development of knowledge about cell structure. The greater magnification and resolution gave more detailed information about known cell organelles, e.g. Golgi body and also discovered new structures, e.g. ribosomes that can only be seen under an electron microscope.
13. D
14. A

9 Experiment – The Light Microscope

- 1.

