1 CELL BIOLOGY

Introduction

There is an unbroken chain of life from the first cells on Earth to all cells found in organisms alive today. Eukaryotes have a much more complex cell structure than prokaryotes. The evolution of multicellular organisms allowed cell specialization and cell replacement. Cell division is essential but is carried out differently in prokaryotes and eukaryotes. While evolution has resulted in a biological world of enormous diversity, the study of cells shows us that there are also universal features. For example, the fluid and dynamic structure of biological membranes allows them to control the composition of cells.

1.1 Introduction to cells

Understanding

- According to the cell theory, living organisms are composed of cells.
- Organisms consisting of only one cell carry out all functions of life in that cell.
- Surface area to volume ratio is important in the limitation of cell size.
- Multicellular organisms have properties that emerge from the interaction of their cellular components.
- Specialized tissues can develop by cell differentiation in multicellular organisms.
- Differentiation involves the expression of some genes and not others in a cell's genome.
- The capacity of stem cells to divide and differentiate along different pathways is necessary in embryonic development. It also makes stem cells suitable for therapeutic uses.

Applications

- Questioning the cell theory using atypical examples, including striated muscle, giant algae and aseptate fungal hyphae.
- Investigation of functions of life in *Paramecium* and one named photosynthetic unicellular organism.
- Use of stem cells to treat Stargardt's disease and one other named condition.
- Ethics of the therapeutic use of stem cells from specially created embryos, from the umbilical cord blood of a new-born baby and from an adult's own tissues.

Nature of science

- Looking for trends and discrepancies: although most organisms conform to cell theory, there are exceptions.
- Ethical implications of research: research involving stem cells is growing in importance and raises ethical issues.

Skills

- Use of a light microscope to investigate the structure of cells and tissues.
- Drawing cell structures as seen with the light microscope.
- Calculation of the magnification of drawings and the actual size of structures shown in drawings or micrographs.

The cell theory

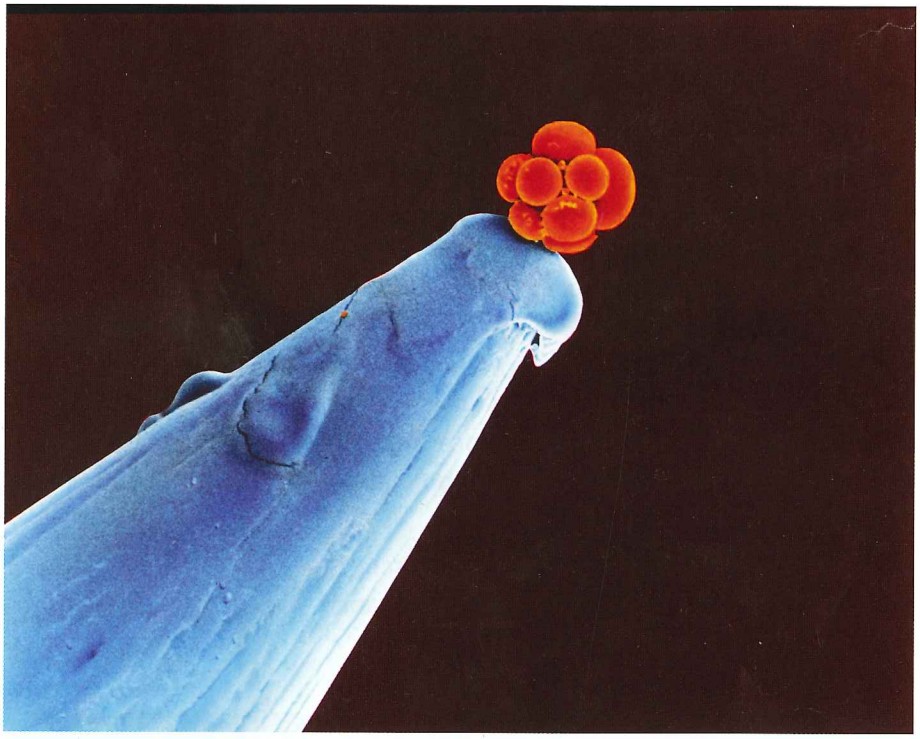
Living organisms are composed of cells.

The internal structure of living organisms is very intricate and is built up from very small individual parts. Organs such as the kidney and the eye are easily visible. If they are dissected we can see that large organs are made of a number of different tissues, but until microscopes were invented little or nothing was discovered about the structure of tissues. From the 17th century onwards biologists examined tissues from both plants and animals using microscopes. Although there was much variation, certain features were seen again and again. A theory was developed to explain the basic features of structure – the cell theory. This states that cells are the fundamental building blocks of all living organisms. The smallest organisms are unicellular – they consist of just one cell. Larger organisms are multicellular – they are composed of many cells.

Cells vary considerably in size and shape but they share certain common features:

- Every living cell is surrounded by a membrane, which separates the cell contents from everything else outside.
- Cells contain genetic material which stores all of the instructions needed for the cell's activities.
- Many of these activities are chemical reactions, catalysed by enzymes produced inside the cell.
- Cells have their own energy release system that powers all of the cell's activities.

So, cells can be thought of as the smallest living structures – nothing smaller can survive.



▲ Figure 1 Coloured scanning electron micrograph (SEM) of a human embryo on the tip of a pin

Exceptions to the cell theory

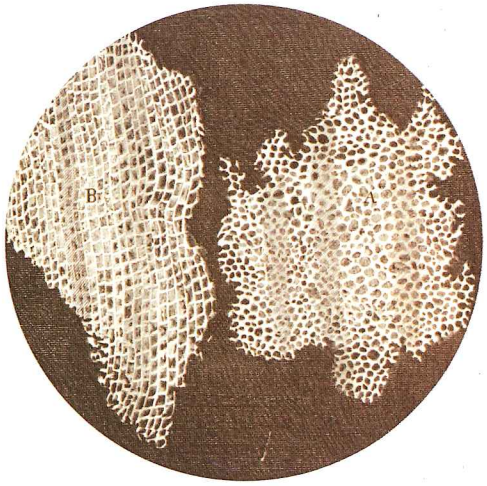
Looking for trends and discrepancies: although most organisms conform to cell theory, there are exceptions.

An early stage in scientific investigation is to look for trends – things that appear to be found generally rather than just in specific cases. These trends can lead to the development of a theory. A scientific theory is a way of interpreting the natural world. Theories allow us to make predictions. Sometimes exceptions to a general trend are found. These are called discrepancies. Scientists have to judge whether the discrepancies are common or serious enough to make predictions too unreliable to be useful. The theory is then discarded.

The cell theory is an example of where scientists have looked for trends and discrepancies. Robert Hooke was the first to use the word cell for structures in living organisms. He did this in 1665 after examining cork and other parts of plants. After describing cells in cork he wrote this:

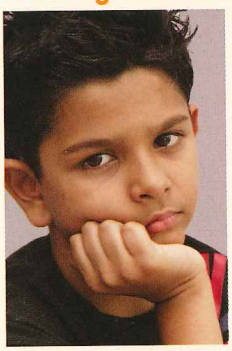
> *Nor is this kind of texture peculiar to cork only, for upon examination with my microscope I have found that the pith of the Elder or almost any other tree, the inner pith of the Cany hollow stems of several other vegetables: as of Fennel, Carrets, Daucus, Bur-docks, Teasels, Fearn, some kind of Reeds etc. have much such a kind of Schematisme, as I have lately shown that of cork.*

So Hooke wasn't content with looking at just one type of plant tissue – he looked at many and discovered a general trend. Since Hooke's day biologists have looked at tissues from a huge variety of living organisms. Many of these tissues have been found to consist of cells, so the cell theory has not been discarded. However, some discrepancies have been discovered – organisms or parts of organisms that do not consist of typical cells. More discrepancies may be discovered, but it is extremely unlikely that the cell theory will ever be discarded, because so many tissues do consist of cells.



▲ Figure 2 Robert Hooke's drawing of cork cells

Activity



▲ Figure 3 What is the unit of life: the boy or his cells?

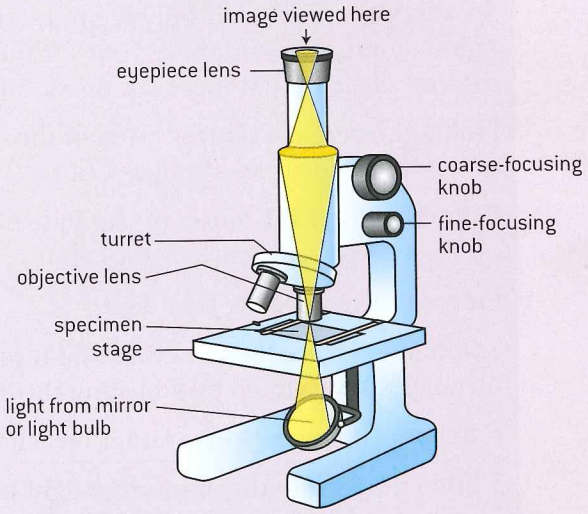
These two answers represent the holistic and the reductionist approach in biology.

Using light microscopes

Use of a light microscope to investigate the structure of cells and tissues.

Try to improve your skill at using microscopes as much as you can.

- Learn the names of parts of the microscope.
- Understand how to focus the microscope to get the best possible image.
- Look after your microscope so it stays in perfect working order.
- Know how to troubleshoot problems.



▲ Figure 4 Compound light microscope

Focusing

- Put the slide on the stage, with the most promising region exactly in the middle of the hole in the stage that the light comes through.
- Always focus at low power first even if eventually you need high power magnification.
- Focus with the larger coarse-focusing knobs first, then when you have nearly got the image in focus make it really sharp using the smaller fine-focusing knobs.
- If you want to increase the magnification, move the slide so the most promising region is exactly in the middle of the field of view and then change to a higher magnification lens.

Looking after your microscope

- Always focus by moving the lens and the specimen further apart, never closer to each other.
- Make sure that the slide is clean and dry before putting it on the stage.
- Never touch the surfaces of the lenses with your fingers or anything else.
- Carry the microscope carefully with a hand under it to support its weight securely.

Troubleshooting

Problem: Nothing is visible when I try to focus.

Solution: Make sure the specimen is actually under the lens, by carefully positioning the slide. It is easier to find the specimen if you focus at low power first.

Problem: A circle with a thick black rim is visible.

Solution: There is an air bubble on the slide. Ignore it and try to improve your technique for making slides so that there are no air bubbles.

Problem: There are blurred parts of the image even when I focus it as well as I can.

Solution: Either the lenses or the slide have dirt on them. Ask your teacher to clean it.

Problem: The image is very dark.

Solution: Increase the amount of light passing through the specimen by adjusting the diaphragm.

Problem: The image looks rather bleached.

Solution: Decrease the amount of light passing through the specimen by adjusting the diaphragm.

Types of slide

The slides that we examine with a microscope can be permanent or temporary.

Making permanent slides is very skilled and takes a long time, so these slides are normally made by experts. Permanent slides of tissues are made using very thin slices of tissue.

Making temporary slides is quicker and easier so we can do this for ourselves.

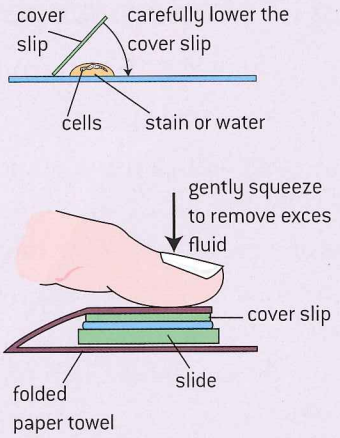
Examining and drawing plant and animal cells

Almost all cells are too small to be seen with the naked eye, so a microscope is needed to study them.

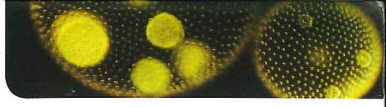
It is usually easy to see whether a cell is from a plant or an animal, even though there are many different cell types in both the plant and animal kingdoms.

- Place the cells on the slide in a layer not more than one cell thick.
- Add a drop of water or stain.
- Carefully lower a cover slip onto the drop. Try to avoid trapping any air bubbles.
- Remove excess fluid or stain by putting the slide inside a folded piece of paper towel and pressing lightly on the cover slip.

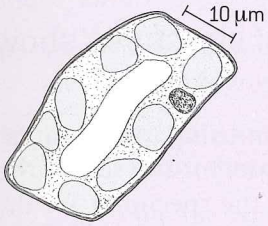
It is best to examine the slide first using low power. Move the slide to get the most promising areas in the middle of the field of view and then move up to high power. Draw a few cells, so you remember their structure.



▲ Figure 5 Making a temporary mount

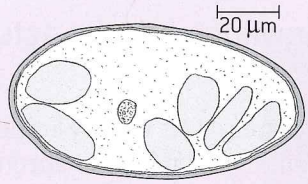


1 Moss leaf



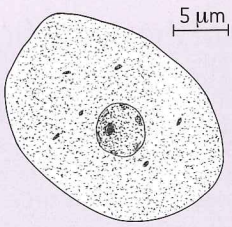
Use a moss plant with very thin leaves. Mount a single leaf in a drop of water or methylene blue stain.

2 Banana fruit cell



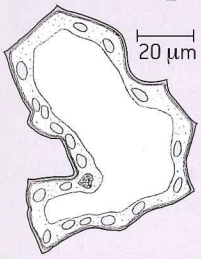
Scrape a small amount of the soft tissue from a banana and place on a slide. Mount in a drop of iodine solution.

3 Mammalian liver cell



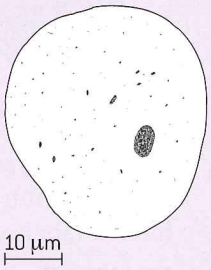
Scrape cells from a freshly cut surface of liver (not previously frozen). Smear onto a slide and add methylene blue to stain.

4 Leaf lower epidermis



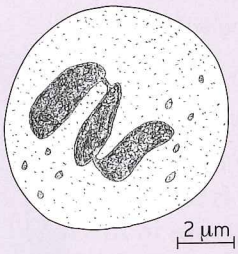
Peel the lower epidermis off a leaf. The cell drawn here was from *Valeriana*. Mount in water or in methylene blue.

5 Human cheek cell



Scrape cells from the inside of your cheek with a cotton bud. Smear them on a slide and add methylene blue to stain.

6 White blood cell



A thin layer of mammalian blood can be smeared over a slide and stained with Leishman's stain.

▲ Figure 6 Plant and animal cell drawings

Drawing cells

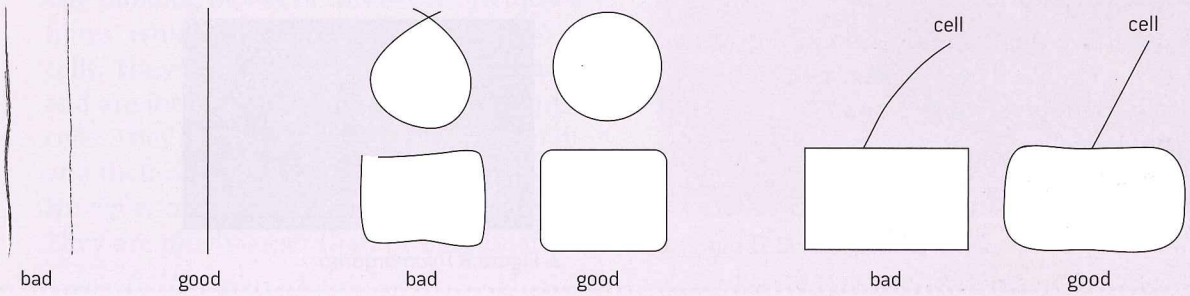
Drawing cell structures as seen with the light microscope.

Careful drawings are a useful way of recording the structure of cells or other biological structures. Usually the lines on the drawing represent the edges of structures. Do not show unnecessary detail and only use faint shading. Drawings of structures seen using a microscope will be larger than the structures actually are – the drawing shows them magnified. On page 6 the method for calculating the magnification of a drawing is explained. Everything on a drawing should be shown to the same magnification.

a) Use a sharp pencil with a hard lead to draw single sharp lines.

b) Join up lines carefully to form continuous structures such as cells

c) Draw lines freehand, but use a ruler for labelling lines.



▲ Figure 7 Examples of drawing styles

Calculation of magnification and actual size

Calculation of the magnification of drawings and the actual size of structures shown in drawings or micrographs.

When we look down a microscope the structures that we see appear larger than they actually are. The microscope is magnifying them. Most microscopes allow us to magnify specimens by two or three different factors. This is done by rotating the turret to switch from one objective lens to another. A typical school microscope has three levels of magnification:

- × 40 (low power)
- × 100 (medium power)
- × 400 (high power)

If we take a photo down a microscope, we can magnify the image even more. A photo taken down a microscope is called a micrograph. There are many micrographs in this book, including electron micrographs taken using an electron microscope.

When we draw a specimen, we can make the drawing larger or smaller, so the magnification of the drawing isn't necessarily the same as the magnification of the microscope.

To find the magnification of a micrograph or a drawing we need to know two things: the size of the image (in the drawing or the micrograph) and the actual size of the specimen. This formula is used for the calculation:

$$\text{magnification} = \frac{\text{size of image}}{\text{actual size of specimen}}$$

If we know the size of the image and the magnification, we can calculate the actual size of a specimen.

It is very important when using this formula to make sure that the units for the size of the image and actual size of the specimen are the same. They could both be millimetres (mm) or micrometres (μm) but they must not be different or the calculation will be wrong. Millimetres can be converted to micrometres by multiplying by one thousand. Micrometres can be converted to millimetres by dividing by one thousand.

Scale bars are sometimes put on micrographs or drawings, or just alongside them. These are straight lines, with the actual size that the scale bar represents. For example, if there was a 10 mm long scale bar on a micrograph with a magnification of ×10,000 the scale bar would have a label of 1 μm.

EXAMPLE:

The length of an image is 30 mm. It represents a structure that has an actual size of 3 μm. Determine the magnification of the image.

Either:

$30\,\text{mm} = 30 \times 10^{-3}\,\text{m}$

$3\,\mu\text{m} = 3 \times 10^{-6}\,\text{m}$

$$\text{Magnification} = \frac{30 \times 10^{-3}}{3 \times 10^{-6}}$$

$= 10{,}000\times$

Or:

$30\,\text{mm} = 30{,}000\,\mu\text{m}$

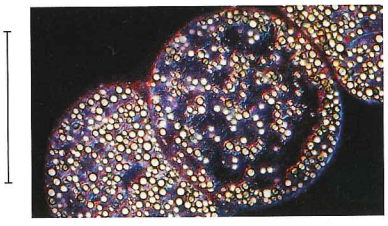
$$\text{Magnification} = \frac{30{,}000}{3}$$

$= 10{,}000\times$

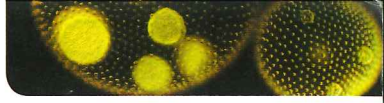
Data-based questions

1 **a)** Determine the magnification of the string of *Thiomargarita* cells in figure 8, if the scale bar represents 0.2 mm [3]

b) Determine the width of the string of cells. [2]



▲ Figure 8 *Thiomargarita*

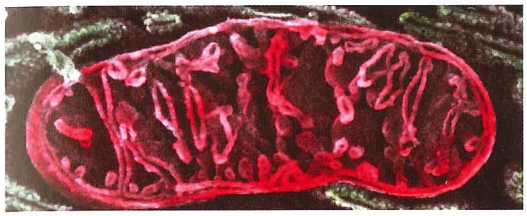


2 In figure 9 the actual length of the mitochondrion is 8 μm.

a) Determine the magnification of this electron micrograph. [2]

b) Calculate how long a 5 μm scale bar would be on this electron micrograph. [2]

c) Determine the width of the mitochondrion. [1]

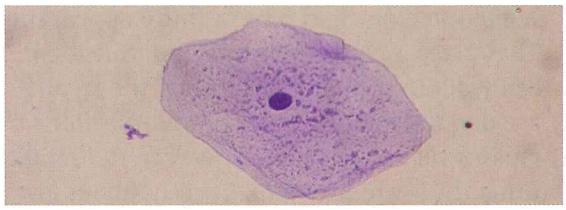


▲ Figure 9 Mitochondrion

3 The magnification of the human cheek cell from a compound microscope (figure 10) is 2,000 ×.

a) Calculate how long a 20 μm scale bar would be on the image. [2]

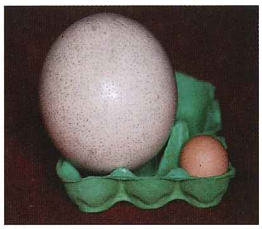
b) Determine the length of the cheek cell. [2]



▲ Figure 10 Human cheek cell

4 a) Using the width of the hen's egg as a guide, estimate the actual length of the ostrich egg (figure 11). [2]

b) Estimate the magnification of the image. [2]



▲ Figure 11 Ostrich egg

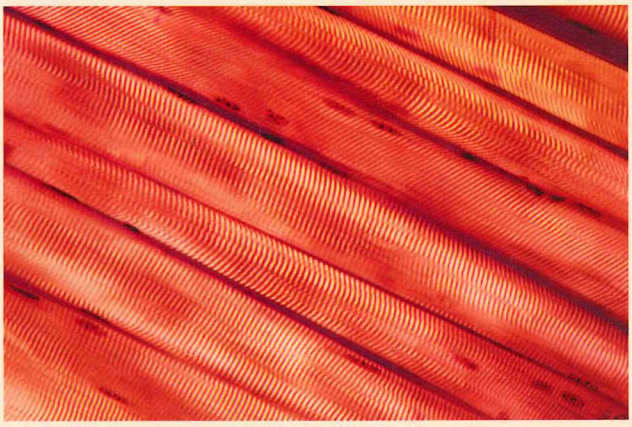
Testing the cell theory

Questioning the cell theory using atypical examples, including striated muscle, giant algae and aseptate fungal hyphae.

To test the cell theory you should look at the structure of as many living organisms as you can, using a microscope. Instructions for microscope use are given on page 4. In each case you should ask the question, "Does the organism or tissue fit the trend stated in the cell theory by consisting of one or more cells?"

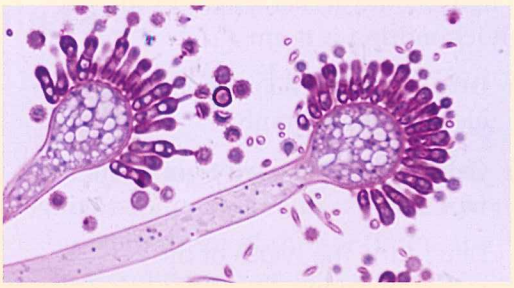
Three atypical examples are worth considering:

- Striated muscle is the type of tissue that we use to change the position of our body. The building blocks of this tissue are muscle fibres, which are similar in some ways to cells. They are surrounded by a membrane and are formed by division of pre-existing cells. They have their own genetic material and their own energy release system. However muscle fibres are far from typical. They are much larger than most animal cells. In humans they have an average length of about 30 mm, whereas other human cells are mostly less than 0.03 mm in length. Instead of having one nucleus they have many, sometimes as many as several hundred.



▲ Figure 12 Striated muscle fibres

- Fungi consist of narrow thread-like structures called hyphae. These hyphae are usually white in colour and have a fluffy appearance. They have a cell membrane and, outside it, a cell wall. In some types of fungi the hyphae are divided up into small cell-like sections by cross walls called septa. However, in aseptate fungi there are no septa. Each hypha is an uninterrupted tube-like structure with many nuclei spread along it.
- Algae are organisms that feed themselves by photosynthesis and store their genes inside nuclei, but they are simpler in their structure and organization than plants. Many algae consist of one microscopic cell. There are vast numbers of these unicellular algae in the oceans and they form the basis of most marine food chains. Less common are some algae that grow to a much larger size, yet they still seem to be single cells. They are known as giant algae. *Acetabularia* is one example. It can grow to a length of as much as 100 mm, despite only having one nucleus. If a new organism with a length of 100 mm was discovered, we would certainly expect it to consist of many cells, not just one.



▲ Figure 13 Aseptate hypha

▲ Figure 14 Giant alga

Unicellular organisms

Organisms consisting of only one cell carry out all functions of life in that cell.

The functions of life are things that all organisms must do to stay alive. Some organisms consist of only one cell. This cell therefore has to carry out all the functions of life. Because of this the structure of unicellular organisms is more complex than most cells in multicellular organisms.

Unicellular organisms carry out at least seven functions of life:

- Nutrition – obtaining food, to provide energy and the materials needed for growth.
- Metabolism – chemical reactions inside the cell, including cell respiration to release energy.
- Growth – an irreversible increase in size.
- Response – the ability to react to changes in the environment.
- Excretion – getting rid of the waste products of metabolism.
- Homeostasis – keeping conditions inside the organism within tolerable limits.
- Reproduction – producing offspring either sexually or asexually.

Many unicellular organisms also have a method of movement, but some remain in a fixed position or merely drift in water or air currents.

Limitations on cell size

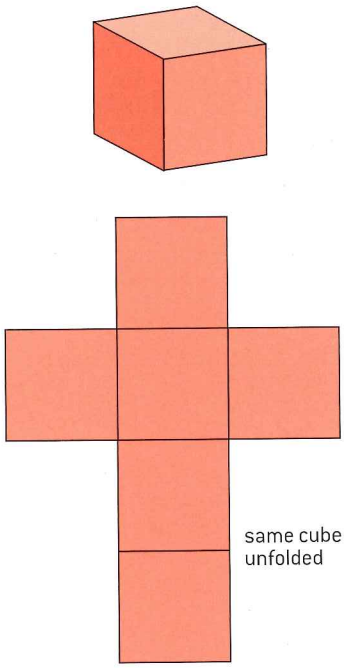
Surface area to volume ratio is important in the limitation of cell size.

In the cytoplasm of cells, large numbers of chemical reactions take place. These reactions are known collectively as the metabolism of the cell. The rate of these reactions (the metabolic rate of the cell) is proportional to the volume of the cell.

For metabolism to continue, substances used in the reactions must be absorbed by the cell and waste products must be removed. Substances move into and out of cells through the plasma membrane at the surface of the cell. The rate at which substances cross this membrane depends on its surface area.

The surface area to volume ratio of a cell is therefore very important. If the ratio is too small then substances will not enter the cell as quickly as they are required and waste products will accumulate because they are produced more rapidly than they can be excreted.

Surface area to volume ratio is also important in relation to heat production and loss. If the ratio is too small then cells may overheat because the metabolism produces heat faster than it is lost over the cell's surface.



▲ Figure 15 Volume and surface area of a cube

Functions of life in unicellular organisms

Investigation of functions of life in *Paramecium* and one named photosynthetic unicellular organism.

Paramecium is a unicellular organism that can be cultured quite easily in the laboratory. Alternatively collect some pond water and use a centrifuge to concentrate the organisms in it to see if *Paramecium* is present.

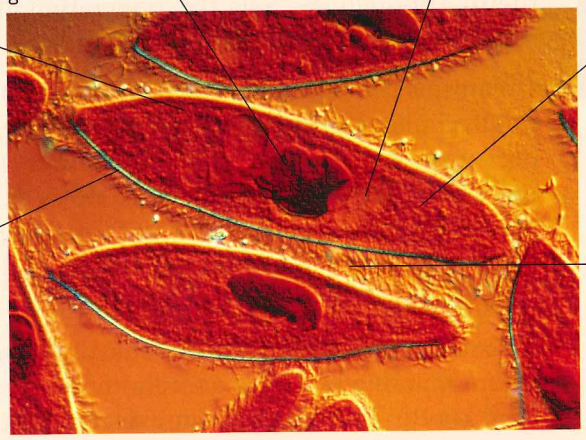
Place a drop of culture solution containing *Paramecium* on a microscope slide.

Add a cover slip and examine the slide with a microscope.

The nucleus of the cell can divide to produce the extra nuclei that are needed when the cell reproduces. Often the reproduction is asexual with the parent cell dividing to form two daughter cells.

Food vacuoles contain smaller organisms that the *Paramecium* has consumed. These are gradually digested and the nutrients are absorbed into the cytoplasm where they provide energy and materials needed for growth.

The cell membrane controls what chemicals enter and leave. It allows the entry of oxygen for respiration. Excretion happens simply by waste products diffusing out through the membrane.



The contractile vacuoles at each end of the cell fill up with water and then expel it through the plasma membrane of the cell, to keep the cell's water content within tolerable limits.

Metabolic reactions take place in the cytoplasm, including the reactions that release energy by respiration. Enzymes in the cytoplasm are the catalysts that cause these reactions to happen.

Beating of the cilia moves the *Paramecium* through the water and this can be controlled by the cell so that it moves in a particular direction in response to changes in the environment.

▲ Figure 16 *Paramecium*

pass through the lens and our vision would be worse. While they are developing, both cell types contain the genes for making the pigment, but these genes are only used in the rod cell.

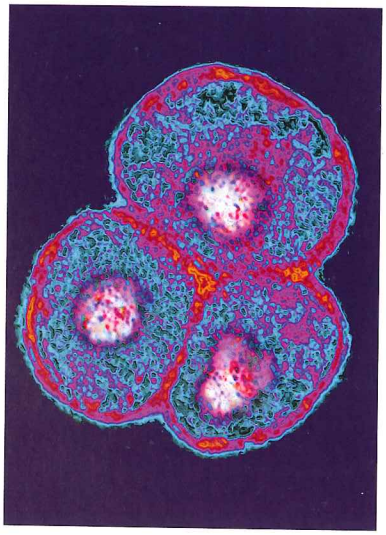
This is the usual situation – cells do not just have genes with the instructions that they need, they have genes needed to specialize in every possible way. There are approximately 25,000 genes in the human genome, and these genes are all present in a body cell. However, in most cell types less than half of the genes will ever be needed or used.

When a gene is being used in a cell, we say that the gene is being expressed. In simple terms, the gene is switched on and the information in it is used to make a protein or other gene product. The development of a cell involves switching on particular genes and expressing them, but not others. Cell differentiation happens because a different sequence of genes is expressed in different cell types. The control of gene expression is therefore the key to development.

An extreme example of differentiation involves a large family of genes in humans that carry the information for making receptors for odorants – smells. These genes are only expressed in cells in the skin inside the nose, called olfactory receptor cells. Each of these cells expresses just one of the genes and so makes one type of receptor to detect one type of odorant. This is how we can distinguish between so many different smells. Richard Axel and Linda Buck were given the Nobel Prize in 2004 for their work on this system.

Stem cells

The capacity of stem cells to divide and differentiate along different pathways is necessary in embryonic development. It also makes stem cells suitable for therapeutic uses.



▲ Figure 19 Embryonic stem cells

A new animal life starts when a sperm fertilizes an egg cell to produce a zygote. An embryo is formed when the zygote divides to give two cells. This two-cell embryo divides again to produce a four-cell embryo, then eight, sixteen and so on. At these early stages in embryonic development the cells are capable of dividing many times to produce large amounts of tissue. They are also extremely versatile and can differentiate along different pathways into any of the cell types found in that particular animal. In the 19th century, the name stem cell was given to the zygote and the cells of the early embryo, meaning that all the tissues of the adult stem from them.

Stem cells have two key properties that have made them one of the most active areas of research in biology and medicine today.

- Stem cells can divide again and again to produce copious quantities of new cells. They are therefore useful for the growth of tissues or the replacement of cells that have been lost or damaged.
- Stem cells are not fully differentiated. They can differentiate in different ways, to produce different cell types.

Embryonic stem cells are therefore potentially very useful. They could be used to produce regenerated tissue, such as skin for people who have suffered burns. They could provide a means of healing diseases such as type 1 diabetes where a particular cell type has been lost or is malfunctioning. They might even be used in the future to grow whole replacement organs – hearts or kidneys, for example. These types of use are called therapeutic, because they provide therapies for diseases or other health problems.

There are also non-therapeutic uses for embryonic stem cells. One possibility is to use them to produce large quantities of striated muscle fibres, or meat, for human consumption. The beef burgers of the future may therefore be produced from stem cells, without the need to rear and slaughter cattle.

It is the early stage embryonic stem cells that are the most versatile. Gradually during embryo development the cells commit themselves to a pattern of differentiation. This involves a series of points at which a cell decides whether to develop along one pathway or another. Eventually each cell becomes committed to develop into one specific cell type. Once committed, a cell may still be able to divide, but all of these cells will differentiate in the same way and they are no longer stem cells.

Small numbers of cells remain as stem cells, however, and they are still present in the adult body. They are present in many human tissues, including bone marrow, skin and liver. They give some human tissues considerable powers of regeneration and repair. The stem cells in other tissues only allow limited repair – brain, kidney and heart for example.

Therapeutic uses of stem cells

Use of stem cells to treat Stargardt's disease and one other named condition.

There are a few current uses of stem cells to treat diseases, and a huge range of possible future uses, many of which are being actively researched. Two examples are given here: one involving embryonic stem cells and one using adult stem cells.

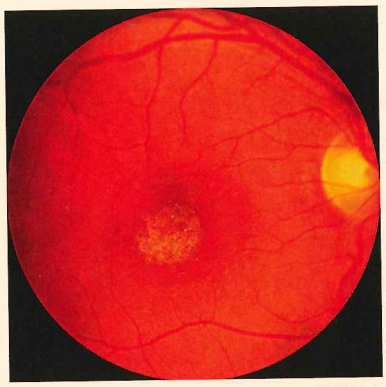
Stargardt's disease

The full name of this disease is Stargardt's macular dystrophy. It is a genetic disease that develops in children between the ages of six and twelve. Most cases are due to a recessive mutation of a gene called ABCA4. This causes a membrane protein used for active transport in retina cells to malfunction. As a consequence, photoreceptive cells in the retina degenerate. These are the cells that detect light, so vision becomes progressively worse. The loss of vision can be severe enough for the person to be registered as blind.

Researchers have developed methods for making embryonic stem cells develop into retina cells. This was done initially with mouse cells, which were then injected into the eyes of mice that had a condition similar to Stargardt's disease. The injected cells were not rejected, did not develop into tumours or cause any other problems. The cells moved to the retina where they attached themselves and remained. Very encouragingly, they caused an improvement in the vision of the mice.

In November 2010, researchers in the United States got approval for trials in humans. A woman in her 50s with Stargardt's disease was treated by having 50,000 retina cells derived from embryonic stem cells injected into her eyes. Again the cells attached to the retina and remained there during the four-month trial. There was an improvement in her vision, and no harmful side effects.

Further trials with larger numbers of patients are needed, but after these initial trials at least, we can be optimistic about the development of treatments for Stargardt's disease using embryonic stem cells.



▲ Figure 20 Stargardt's disease

Leukemia

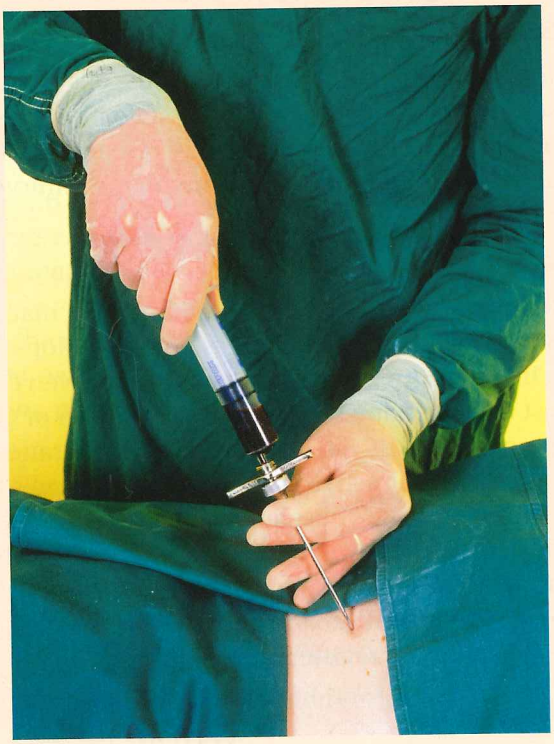
This disease is a type of cancer. All cancers start when mutations occur in genes that control cell division. For a cancer to develop, several specific mutations must occur in these genes in one cell. This is very unlikely to happen, but as there are huge numbers of cells in the body, the overall chance becomes much larger. More than a quarter of a million cases of leukemia are diagnosed each year globally and there are over 200,000 deaths from the disease.

Once the cancer-inducing mutations have occurred in a cell, it grows and divides repeatedly, producing more and more cells. Leukemia involves the production of abnormally large numbers of white blood cells. In most cancers, the cancer cells form a lump or tumour but this does not happen with leukemia. White blood cells are produced in the bone marrow, a soft tissue in the hollow centre of large bones such as the femur. They are then released into the blood, both in normal conditions and when excessive numbers are produced with leukemia. A normal adult white blood cell count is between 4,000 and 11,000 per mm^3 of blood. In a person with leukemia this number rises higher and higher. Counts above 30,000 per mm^3 suggest that a person may have leukemia. If there are more than 100,000 per mm^3 it is likely that the person has acute leukemia.

To cure leukemia, the cancer cells in the bone marrow that are producing excessive numbers of white blood cells must be destroyed. This can be done by treating the patient with chemicals that kill dividing cells. The procedure is known as chemotherapy. However, to remain healthy in the long term the patient must be able to produce the white blood cells needed to fight disease. Stem cells that can produce blood cells must be present, but they are killed by chemotherapy. The following procedure is therefore used:

- A large needle is inserted into a large bone, usually the pelvis, and fluid is removed from the bone marrow.
- Stem cells are extracted from this fluid and are stored by freezing them. They are adult stem cells and only have the potential for producing blood cells.
- A high dose of chemotherapy drugs is given to the patient, to kill all the cancer cells in the bone marrow. The bone marrow loses its ability to produce blood cells.
- The stem cells are then returned to the patient's body. They re-establish themselves in the bone marrow, multiply and start to produce red and white blood cells.

In many cases this procedure cures the leukemia completely.



▲ Figure 21 Removal of stem cells from bone marrow