# Biology S4 Student book

First Edition

Kigali, September 2021

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## **FOREWORD**

#### Dear Student,

Rwanda Basic Education Board is honoured to present to you this Biology Book for Senior Four which serves as a guide to competence-based teaching and learning to ensure consistency and coherence in the learning of Biology subject. The Rwandan educational philosophy is to ensure that you achieve full potential at every level of education which will prepare you to be well integrated in society and exploit employment opportunities.

The government of Rwanda emphasizes the importance of aligning teaching and learning materials with the syllabus to facilitate your learning process. Many factors influence what you learn, how well you learn and the competences you acquire. Those factors include the instructional materials available among others. Special attention was paid special attention to the activities that facilitate the learning process in which you can develop your ideas and make new discoveries during concrete activities carried out individually or with peers.

In competence-based curriculum, learning is considered as a process of active building and developing knowledge and meanings by the learner where concepts are mainly introduced by an activity, a situation or a scenario that helps the learner to construct knowledge, develop skills and acquire positive attitudes and values. For effective use of this textbook, your role is to:

- Work on given activities which lead to the development of skills
- Share relevant information with other learners through presentations, discussions, group work and other active learning techniques such as role play, case studies, investigation and research in the library, from the internet or from your community;
- Participate and take responsibility for your own learning;
- Draw conclusions based on the findings from the learning activities.

I wish to sincerely extend my appreciation to the people who contributed towards the development of this book, particularly REB staff who organized the whole process from its inception. Special gratitude goes to the University of Rwanda which provided experts in design and layout services, illustrations and image antiplagiarism, lecturers and teachers who diligently worked to successful completion of this book. Any comment or contribution would be welcome for the improvement of this textbook for the next edition.

Dr. MBARUSHIMANA Nelson

Director General, REB

## **ACKNOWLEDGEMENT**

I wish to express my appreciation to all the people who played a major role in development of this Biology textbook for Senior Four. It would not have been successful without active participation of different education stakeholders.

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Joan MURUNGI,

Head of Curriculum, Teaching and Learning Resources Department

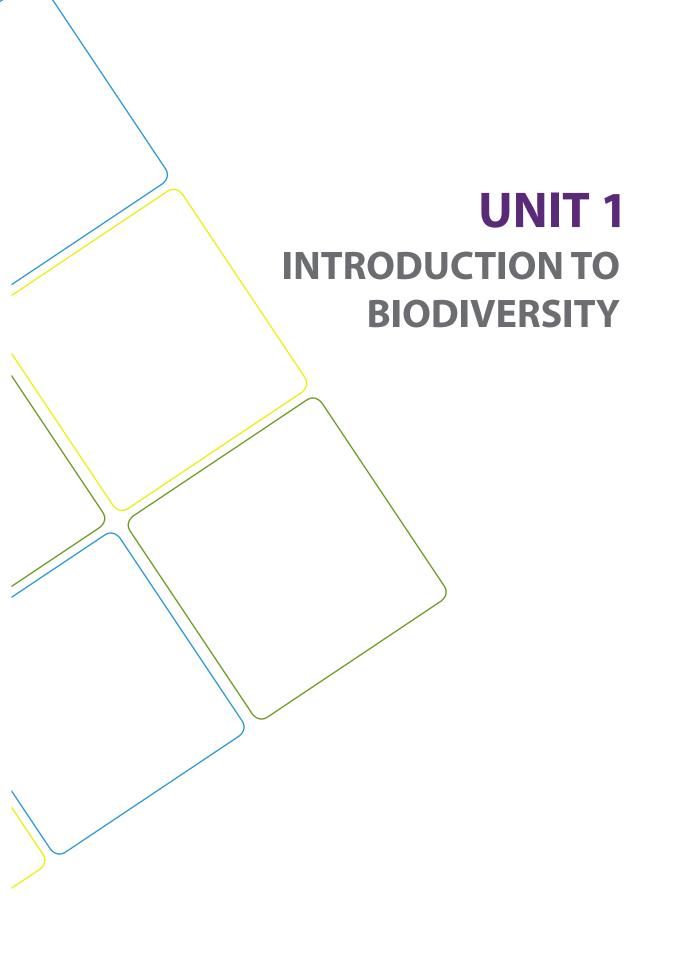
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## **UNIT 1: INTRODUCTION TO BIODIVERSITY**

#### **Key Unit Competence**

Explain how diversity is threatened by climate change and human activities

## **Learning objectives**

By the end of this unit, I should be able to:

- Define the terms: species, ecosystem and niche.
- Explain that biodiversity is considered at three different levels
- Evaluate the consequences of loss of biodiversity.
- Characterize the biotic and abiotic components that define Rwanda's ecosystems (example: freshwater, marine, and terrestrial).
- Apply Simpson's Index of Diversity.
- Explain the importance of random sampling in determining the biodiversity of an area.
- Use suitable survey methods such as frame quadrats, line and belt transects to assess the distribution and abundance of organisms in a local area.
- Use Pearson's linear correlation to analyze the relationships between the distribution and abundance of species and abiotic or biotic factors.
- Recognize that the biodiversity of the earth is threatened by human activities and climate change

## **Introductory activity: Biodiversity of Rwanda**

## Read the following text and answer the questions that follow

Rwanda is located at the heart of the Albertine Rift eco-region in the western arm of the Africa's Rift Valley. Habitats of Rwanda are equally varied, ranging from Afro-Montana ecosystems in the northern and western regions to lowland forests, savannah woodlands and savannah grasslands in the southern and eastern regions. There are other habitats around volcanic hot springs and old lava flows, especially in the northern and western part of the country.

Rwanda also has several lakes and wetlands which are rich in different species. Though not yet well surveyed, all these ecosystems host a rich variety of fauna and flora and micro-organisms. This rich biodiversity is mainly conserved in protected areas including three national parks, natural forests and wetlands. These cover almost 10 percent of the national territory while the rest of the country is densely populated (507 people per square kilometer in 2018).

Many tourists visit Rwanda for its beautiful environment and biodiversity made of different species of plants and animals such as Aloe vera (Igikakarubamba), Muringa oleifera (Muringa), Phaseolus vulgaris (common bean), Nymphaea thermarum (Endemic plant species that cannot be met elsewhere in the world, only found in Mashyuza minor locality harbors),

Colobus polykoma (White-black colobus monkey), Gorilla gorilla (mountain gorilla) bird Laniarius mufumbiri (Bird species mainly found in Rweru- Mugera wetland),etc.

The most attracting species in Rwanda is Gorilla gorilla whose habitat is the mountains of Birunga where they make a large population. Another natural forest, Nyugwe National Park is a terrestrial ecosystem that contains a large community of different plants and animals.

Rwanda also has different lakes such as Muhazi and Rumira. They are aquatic ecosystems made of few species of fish, such as tilapias. Tilapias from Lake Muhazi are small, black and bony fish while those from Lake Rumira look red, big and soft. Tilapias from both lakes still belong in the same species but show variations.

Many species of animals and plants have been discovered in Rwanda but some species also disappeared. Today the big garden snails known as Achatina achatina have become rare in Bugesera. Other people poached Rhinoceros alba living in Savanah of Akagera National Park.

Honey bees, butterflies and grasshoppers are small in size but still important for different ecosystem services. Each organism is important for its niche in ecosystem. We need to identify and protect the biodiversity of our ecosystem. Many tourists enjoy visiting Rwanda for its biodiversity.

- 1. Name the species not found elsewhere that attract the tourists and locate where it is found.
- 2. Mashyuza is a minor locality in western province in Rusizi district that contributes to biodiversity of Rwanda. Give any other two locations.
- 3. Define each of the following biological terms and give an example from the text
  - above:(a) Species (b) Population (c) Community (d) Habitat (e) Ecosystem (f)Variation (g) Niche
- 4. What causes some species to become extinct?
- 5. What can be the consequences of the loss of some species from our biodiversity?
- 6. Do you support tourism in Rwanda? Give a reason to justify your answer.

## 1.1. Meaning of key ecological terms and biodiversity

## **Activity 1.1**

Using addition resources to your textbook available in your school such as the books from the school library and search further information from the internet:

1. Describe the following terms: biodiversity, species, niche, population, and community?

#### 2. Differentiate between ecological niche and habitat.

#### 1.1.1. Key ecological terms

Species is a group of closely related organisms which are capable of interbreeding to produce fertile offspring. Occasionally two organisms which are genetically closely related but not of the same species can interbreed to produce infertile offspring. For example:

- A cross between a donkey and a horse produces a mule, which is infertile.
   Thus, a donkey and a horse do not belong to the same species
- Lions and tigers belonging to different species. However, when a male tiger mates with a female lion they can have fertile offspring called tiglons, although the offspring of female tigers and male lions called ligers are not fertile

Note that normally, tigers are forest dwellers and lions are plains dwellers and they are ecologically isolated. Breeding has only been observed in captivity.

An ecological population is a group of individuals of the same species which live in a particular area at any given time.

An ecological community consists of populations of different species which live in the same place at the same time, and interact with each other.

A habitat is a specific area or place in which an individual organism lives. When a habitat is very small it is regarded as a microhabitat. Most ecosystems contain several habitats, and one species can have more than one habitat constituting its geographic range.

An ecological niche is the status or the role of an organism in its habitat or the mode of life of an organism within its habitats. For example, insects are pollinating agents and preys of insectivores.

Abiotic factor are non-living physical aspects of the environment such as the sunlight, soil, temperature, wind, water, and air.

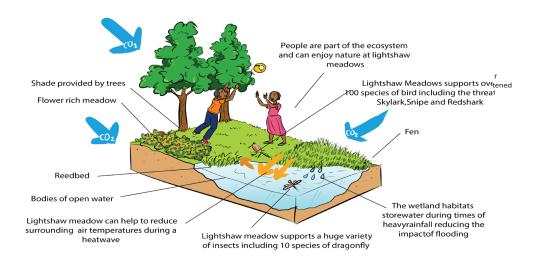
Biotic factors are the living organisms in the environment. They include organisms and their interactions with each other.

An ecosystem is a natural unit consisting of biotic and abiotic factors through which energy flows and nutrients recycle. In an ecosystem, nutrients pass between different organisms in definite pathways. For example, nutrients in the soil are taken up by plants, which are then eaten by herbivores, which in turn may be eaten by carnivores and recycled by decomposers.

A biome is a group of ecosystems that have the same climate and similar dominant communities. The highest level of organization is the entire biosphere.

The Biosphere is the whole of the earth's surface, the sea and the air that is inhabited by living organisms. The biosphere is made up of all ecosystems.

## 1.1.2. Biodiversity



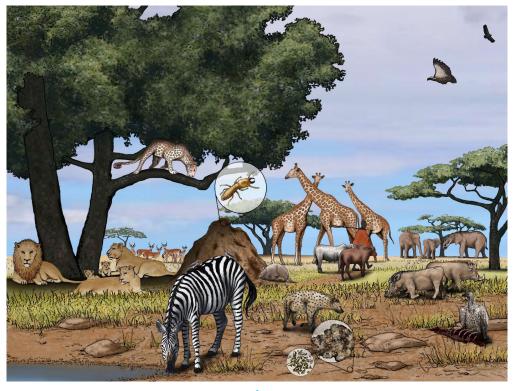


Figure 1.1: Varieties of species in ecosystems

Biodiversity is defined as the full range of variety and variability within and among living organisms and the ecological complexes in which they occur.

#### Self-assessment 1.1

- 1. Describe the two main components of an ecosystem.
- 2. Hippopotamus has different habitats. It was found that the resting habitat is different from the mating habitat, and these two habitats are different from the area where this animal gets food. Explain the ecological term given to this set of habitats.

## 1.2. Identification of biodiversity

## **Activity 1.2**

Use books or other sources of information to answer the followings questions:

- 1. What kinds of initiatives and incentive mechanisms are put in place by the Government of Rwanda to motivate local community in biodiversity conservation?
- 2. Describe different ways used to identify biodiversity.
- 3. Discuss the values of biodiversity and ecosystem services in Rwanda.
- 4. Evaluate the contribution of biodiversity to human well-being.

### 1.2.1. Categories of biodiversity

Biodiversity can be categorized into three groups:

- Genetic diversity: the combination of different genes found within a population of a single species, and the patterns of variation found within different populations of the same species.
- Species diversity: the variety and abundance of different types of organisms which inhabit an area.
- Ecosystem diversity: the variety of habitats that occur within a region, or within the mosaic of patches found within a landscape.

## 1.2.2. Importance of biodiversity

Biodiversity contributes to ecosystem goods and services. The ecosystem goods and services include:

- Provision of food, air, fire wood, medicines(Fig.1.2), energy, fresh water.
- Nutrient cycling such carbon, water and nitrogen cycles by microorganisms and primary production by photosynthesis.
- Cultural or aesthetic service recreation, ecotourism, cultural andreligious inspiration.



Figure 1.2: Some of the common medicinal plants (A: Aloe vera, B: Spinacia oleracea, C: Datura stramonium, D: Camellia sinensis)

## 1.2.3. The threats and consequences of biodiversity loss

#### 1.2.3.1. Causes of biodiversity loss

The main causes of biodiversity loss can be attributed to the influence of human activities on ecosystems. Threats to biodiversity include:

#### a. Habitat loss and the degradation of the environment

The habitat loss and the degradation of the environment occur in different ways. The most occurring, are tree cutting, agriculture and fires (Figure 1.3). These human activities lead to the alteration and loss of suitable habitats for biodiversity. As a consequence, there is a loss of plant species as well as the decrease in the animal species associated to this plant diversity.



Figure 1.3: Deforestation and bush burning cause biodiversity loss

## b. Introduction of invasive alien species and genetically modified organisms

Species originating from a particular area are harmful to native species also called endemic species when they are introduced into new natural environments. They can lead to different forms of imbalance in the ecological equilibrium, so that endemic species may fail to compete with introduced species, and they may affect the abundance and distribution in natural habitat.

#### c. Pollution

Human activities such as excessive use of fertilizers, and increased pollutants from industries and domestic sewage affect biodiversity. They contribute to the alteration of the flow of energy, chemicals and physical constituents of the environment and hence species may die as a result of toxic accumulation.

#### d. Overexploitation of natural resources

Increased hunting, fishing, and farming in particular areas lead to the decrease and loss of biodiversity due to excessive and continuous harvesting without leaving enough time for the organisms to reproduce and stabilize in their natural habitat.

#### e. Climate change

This is a change in the pattern of weather, related changes in oceans, land surfaces and ice sheets due to global warming resulting from man's activities. Increasing global temperatures have resulted into melting of icebergs raising sea levels and so flooding coastal areas eventually affecting the niche.

#### 1.2.3.2. Consequences of loss of biodiversity

They are various consequences of loss of biodiversity that include:

- Desertification, is thought by scientists to be a consequence of climate change, has been considered to be related to deforestation. Disrupting water cycles and soil structure results into less rainfall in an area.
- Floods as a result of rising sea levels
- Habitat destruction for extensive farming, timber harvesting and infrastructure and settlement
- Decrease in food production as result of change in pattern of weather that affects productivity
- Large scale deforestation has a negative effect on nutrient recycling and can accelerates soil erosion
- Diseases that come as effects of floods and malnutrition due to famine

#### Self-assessment 1.2

- Define the term Extinction.
- 2. Suggest the causes of extinction of species in Rwanda.
- 3. Discuss the benefits of biodiversity to humans
- 4. Discuss the major factors leading to the degradation of ecosystems in Rwanda
- 5. Discuss the contribution of ecosystems to cultural traditions in Rwanda.
- 6. In Rwanda different plants are used in traditional medicine to treat different diseases. Conduct a research and list at least 20 medicinal plants and the diseases they treat. From the list above describe at least one medicinal plant and get ready to present your work. The project work should include: written content of 2 pages in minimum and 4 pages in maximum, a testimony of people that have used plant species.
- 7. Pollution is one of the causes of aquatic biodiversity loss.
  - a. What do you understand by water pollution?
  - b. Outline human activities that contribute to water pollution
  - c. Discuss how polluted water affects aquatic living organisms?

## 1.3. Calculation of Simpson's index

#### **Activity 1.3**

A survey on tree species was conducted in Gako forest by a group of students. Five tree species (A to E) were identified and counted. The numbers found during this exercise are summarized in the following table:

Tree species	Number
A	56
В	48
С	12
D	6
E	3

- 1. Describe the relative abundance of species A to E.
- 2. Based on the data in the above table, suggest how species diversity of tree species can be calculated.

There are many ways to measure diversity. The Simpson diversity index among indices used to measure diversity. It is expressed in three related indices namely Simpson index, Simpson index of diversity and Simpson reciprocal index.

#### a. Simpson index D

Simpson index D can be expressed in two ways and takes into consideration the total number of organisms of a particular species and the total number of organisms of all species. It is calculated as follows:  $\mathbf{D} = \mathbf{1} - \sum_{N(N-1)}^{\infty} \mathbf{n} \cdot \mathbf{n} = 1 - \sum_{N(N-1)}^{\infty} \mathbf{n} \cdot \mathbf{n} \cdot \mathbf{n} \cdot \mathbf{n} = 1 - \sum_{N(N-1)}^{\infty} \mathbf{n} \cdot \mathbf{n} \cdot$ 

**b. Simpson index of diversity 1** – D The value of this index ranges between 0 and 1, but now, the greater the value, the greater the sample diversity. This makes more sense. In this case, the index represents the probability that two individuals randomly selected from a sample will belong to different species.

## c. Simpson reciprocal index 1 / D

Another way of overcoming the problem of the counter-intuitive nature of Simpson's index is to take the Simpson's reciprocal index 1 / D. The value of this index starts with 1 as the lowest possible figure. This figure would represent a community containing only one species. The higher is the value of Simpson reciprocal index, the greater the biological diversity.

#### **Examples**

1. In woodland, a quadrat was sampled for ground vegetation. Data collected were recorded in the table 1.3.2. Find out the value of the Simpson index and draw the conclusion about the biological diversity of the sampled area.

Table 1.3.1: Recorded data on the vegetation from a woodland

Species	Number (n)	n(n-1)
Woodrush	2	2
Holly (seedlings)	8	56
Bramble	1	0
Yorkshire Fog	1	0
Sedge	3	6
Total (N)	15	64

**Solution:** Putting the figures into the formula for Simpson's Index:

 $D = 1 - \frac{\sum n(n-1)}{N(N-1)}$  and replace each letter by its respective value, the Simpson index shall be:

$$D = 1 - \left(\frac{64}{15(15-1)}\right)$$

$$D = 1 - \left(\frac{64}{210}\right)$$

$$D = 1 - 0.7$$

$$D = 0.3$$

Based on the meaning of Simpson index, the quadrat presents a low diversity because the value of D is near zero and zero and below 0.5.

2. Calculate the value of Simpson's Diversity Index (D) for a single quadrate sample of ground vegetation in woodland from which the following sampling date was obtained:

Collected specimens	Number of individual species (n)
Woodrush	2
Holly (seedlings)	8
Bramble	1
Yorkshire Fog	1
Sedge	3

#### **Solution:**

Collected specimens	Number (n)	n/N	(n/N) <sup>2</sup>
Woodrush	2	0.13	0.017
Holly (seedlings)	8	0.53	0.284
Bramble	1	0.06	0.004

Yorkshire Fog	1	0.06	0.004
Sedge	3	0.2	0.04

Simpson's Diversity Index  $D = 1 - (\sum (n/N)^2)$ 

$$D = 1 - (\frac{2}{15})^2 + (\frac{8}{15})^2 + (\frac{1}{15})^2 + (\frac{1}{15})^2 + (\frac{3}{15})^2$$

$$D = 1 - (0.017 + 0.284 + 0.004 + 0.004 + 0.004)$$

$$D = 1 - 0.35 = 0.65$$

Because 0.65 is above 0.5 the area of study presents a moderate diversity.

#### **Self-assessment 1.3**

- 1. Differentiate between species richness and species evenness
- 2. Suggest precautions taken when measuring populations of aquatic animals or plants.
- 3. Explain why a habitat with high diversity tends to be more stable than one with lower diversity.
- 4. In a survey of trees in a tropical forest, students identified five tree species (A to E).

They counted the numbers of trees in an area 100 m  $\times$  100 m and found these results:

Tree species	Number of individual species
А	56
В	48
С	12
D	6
E	3

Calculate the Simpson's Index diversity for identified species and explain the advantage of using data on species diversity and abundance when calculating an index of diversity.

5. The Simpson's Index of diversity for vegetation in an open area inhabited by grasslands was 0.8. For a similar sized area of vegetation beneath some conifer trees it was 0.2. What do you conclude from these results?

## 1.4. Sampling techniques to assess the distribution and abundance of organisms

## **Activity 1.4**

From your school garden, sample different flowering plant species and answer the following questions:

- 1. Specify the techniques used for collecting flowers of different species.
- 2. What are the advantages of the technique you used for data collection?
- 3. Move around the school garden and collect different specimens of plant species. Name the collected species by using their names. In case you don't know their names, use letters A, B, C ....
  - Calculate Simpson index D, Simpson index of diversity and Simpson reciprocal index.

To calculate Simpson's index for a particular place:

- Identify the habitat to be studied.
- The number of individuals sampled for each species must be recorded.

To analyze the distribution and abundance of organisms in an area of study, there are different sampling methods.

Note that, sampling only one quadrat would not give reliable estimate of the diversity of the ground flora in the wood.

## a. Random sampling method

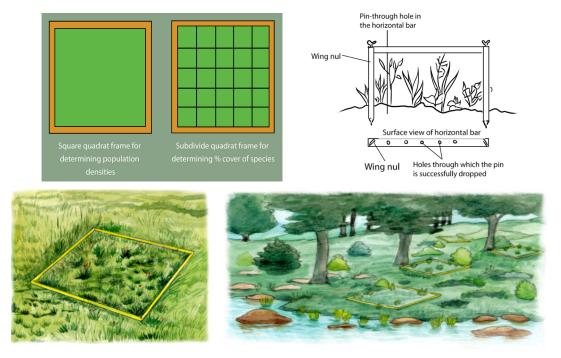
A random sampling method is a sampling method where samples are taken from different positions within a habitat and those positions are chosen randomly.

## b. Quadrat sampling method

A quadrat is a square area that is marked using a pre-made square of plastic, or stakes and string and it can range in size. Different species and their numbers within the quadrat are counted. Counting is repeated many times in different places in the habitat to get an accurate representation of biodiversity.

## c. Frame quadrats

Frame quadrats are small plot used to isolate a standard unit of area for the study of the distribution of an item over a large area. While originally rectangular, modern quadrats can be rectangular, circular, and /or irregular. The quadrat is suitable for sampling plants, slow-moving animals such as millipedes and insect and some aquatic organisms.



*Figure 1.4: Different frame quadrats* 

#### d. Transect sampling

Transect sampling is done using a transect line, which is usually a rope or measuring tape that has been marked at set intervals, such as every meter. The line is unrolled within the habitat. At every interval, the type and number of species along the line are recorded. A measured line is laid across the area in the direction of the environmental gradient. The species touching the line can be recorded along the whole length of the line (continuous sampling) or at specific points along the line (systematic sampling).

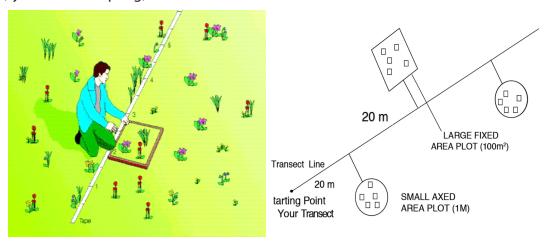
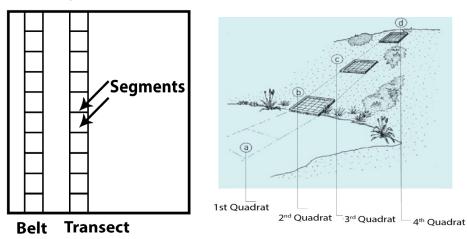


Figure 1.5: Line transect

#### e. Belt transects method

Belt transects method is the same as the line transects but widens the sampling area. The samples are taken and the abundance, percentage cover in a defined area determined. Samples can be taken within the belt.



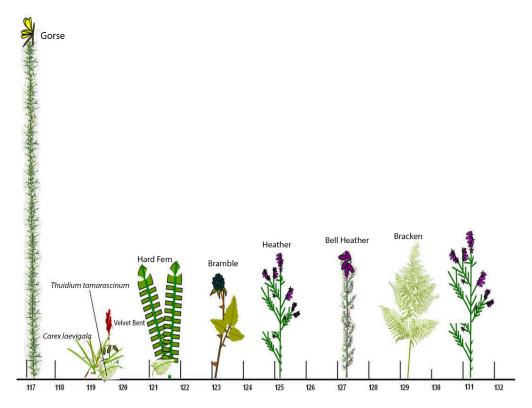


Figure 1.6: Belt transect

## f. Netting

Netting is a sampling method where fine mesh nets are used to capture different organisms that include insects, birds and bats. The technique is also used for

sampling small aquatic organisms like daphnia, and water boatman.



Figure 1.7: Netting method of sampling.

#### g. Capture -recapture technique

This method is useful for sampling non-fixed population and is suitable for animal such as fishes, birds, lizards and insects. A sample of the population to be studied is first captured and each individual is marked with a spot for identification. These are then released and given enough time to mix up with the rest of the members in the habitat. After a certain period of time, another sample is taken.

During the mark-release-recapture technique, the total population can be estimated

by the use of the formula:  $\frac{n1xn2}{n3}$ , where

n1 is a number caught and marked in first sample,

n2 is a number caught in second sample

n3 is a number in the second sample that had been marked.

To understand this application, let us use the following examples:

1. A team of students used a sweep net to sample brown grasshoppers and each collect insect was marked with a very small spot of non-toxic waterproof paint and then they were released in the field. The next day, a second large sample was conducted and data were recorded as follows: number of caught and marked in first sample (n1) = 247, number of caught in second sample (n2) = 269, and the number in the second sample that had been marked (n3) = 16. What is the number of estimated population?

#### Solution

The estimated number = 
$$\frac{n1xn2}{n3} = \frac{247x269}{16} = \frac{66443}{16} = 4152$$
 grasshoppers

2. A student collected 16 butterflies which he marked and released. For a second time he collected 18 butterflies among which 12 were already marked from the first sampling. Estimate the population size of butterflies in that area.

#### Solution

The estimated number 
$$=\frac{n1xn2}{n3} = \frac{16x18}{12} = \frac{288}{12} = 24$$
 butterflies

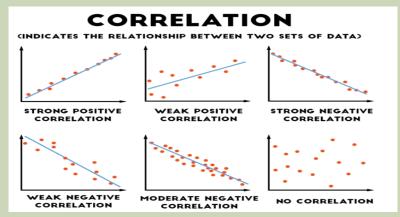
#### Self-assessment 1.4

- 1. Explain the advantages of the random sampling techniques.
- 2. Use suitable methods, such as frame quadrats, line transects, and belt transects, to assess the distribution and abundance of insect species in a school garden. Record your data and use the Simpson index ofdiversity (D) to calculate the diversity of collected insects.
- 3. Suggest the benefits of using the following sampling techniques:
  - a. Ouadrats
  - b. Transect
  - c. Mark-capture-recapture
- 4. State the conditions in which quadrats, transect and mark recapture are suitable sampling methods.

#### 1.5. Pearson's linear correlation

## **Activity 1.5**

Some of the following figures indicate a positive, negative or non-correlation.



- 1. What do you understand by the term correlation?
- 2. Categorize the graphs given as positive, negative or weak or no correlation
- 3. In which conditions results can indicate a positive correlation?
- 4. Conclude about your results when there is no correlation.

To decide if there is an association between collected data, a correlation coefficient is calculated and plot scatter graph drawn in order to make a judgment. The strongest correlation is present for studied items when all the points lie on a straight line. In this case, there is linear correlation, and the correlation coefficient equals

1. If a given variable X increases so does another variable Y, the relationship is a positive correlation. If a variable X increases while the variable Y decreases, then the relationship is a negative correlation. A correlation coefficient of 0 means there is no correlation at all. These correlation coefficients are ways to test a relationship observed and recorded to see if the variables are correlated and, if so, to find the strength of that correlation.

#### a. Pearson's correlation coefficient

Pearson's correlation coefficient can only be used where there might be a linear correlation and when there are collected quantitative data as measurements (for example, length, height, depth, and light intensity, mass) or counts (for example number of plant species in quadrats). The data must be normally distributed.

$$r = \frac{\sum xy - n\overline{x}\overline{y}}{ns_x s_y}$$

#### Where:

r is the correlation coefficient

x is the number of species in a quadrat

y is the number of species in the same quadrat

n is the number of readings (From1 to n)

x is the mean number of species

 $\overline{y}$  is the mean number of species

s<sub>v</sub> is the standard deviation for x

 $s_y$  is the standard deviation for y

#### Self-assessment 1.5

Use Pearson's linear correlation to analyze the relationships between the distribution and abundance of species and abiotic or biotic factors.

#### **End of unit assessment 1**

#### Section A: Answer as true or false

- 1. Abiotic factors are the non-living physical aspects of the environment.
- 2. Capture –recapture is a method used to integrate the numbers of mobile animals in a particular place.
- 3. A correlation coefficient of 0 means that there is no correlation at all.

- 4. A sample is a portion, piece, or segment that is representative of a whole area of study.
- 5. In the Simpson's index, N represents the total number of organisms of a particular species

#### Section B: Long and short answer based questions

- 1. What do you understand by the term biodiversity?
- 2. What do you think would happen to plants if there were no insects?
- 3. Suggest different ways to conserve our forests.
- 4. A student has randomly collected 5 types of species at the following frequencies.

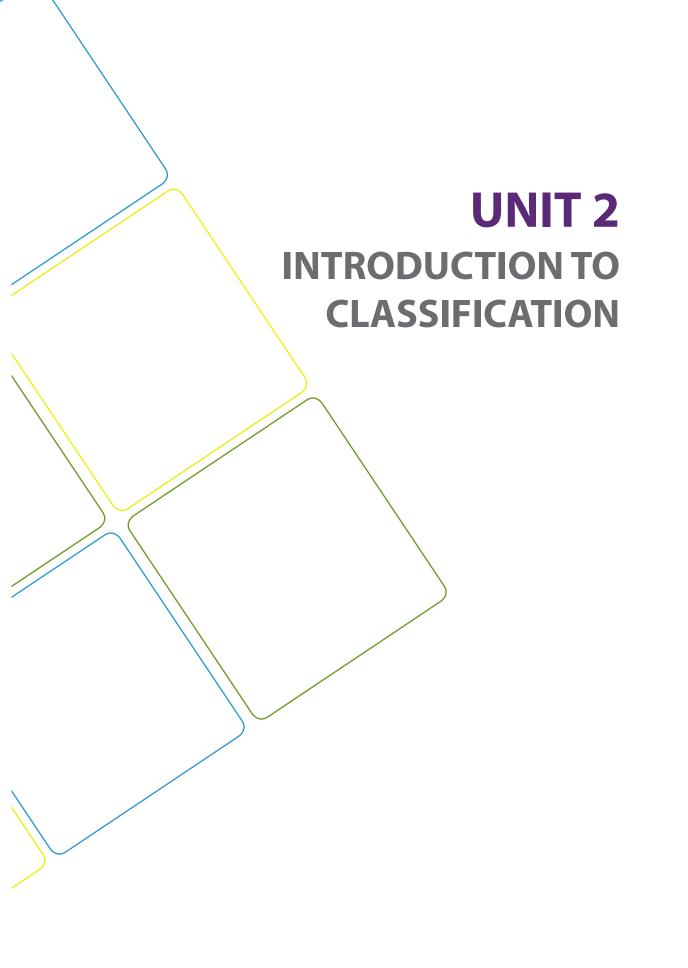
Species A	A	В	C	D	E
Frequency (n)	2	6	3	4	2

Calculate the Simpson's diversity index of this community.

5. A team of students conducted the capture- recapture sampling method of tilapia from lake Muhazi at different times of the day as recorded in the data below:

Time/Hours	12:00	15:00	18:00	21:00	00:00	03:00	06:00	09:00
Number of fish	24	12	8	2	1	4	6	24

- a. Plot the graph for the date provided and describe the shape of the graph.
- b. From the graph, determine the appropriate time to have the most catch.
- 6. What do you understand by term endangered species?
- 7. Describe how diversity is threatened by climate change and human activities.



## **UNIT 2: INTRODUCTION TO CLASSIFICATION**

#### **Key Unit Competence**

Apply the basic knowledge of classification to group living organisms into the three domains.

#### **Learning objectives**

- Describe the classification of species into the taxonomic hierarchy of domain, kingdom, phylum, class, order, family, genus and species.
- Outline the characteristic features of the three domains Archaea, Bacteria and Eukarya.
- Draw and label the structure of a typical bacterial cell.
- Identify common bacterial diseases in plants and animals.
- Outline the characteristic features of the kingdoms Protoctista, Fungi, Plantae and Animalia.
- Explain why viruses are not included in the three domain classification.
- Outline how viruses are classified limited to type of nucleic acid and their host.
- Describe the role of bacteria in the production of dairy products.
- Describe methods of preventing common bacterial diseases.
- Construct a dichotomous key for a group of organisms.
- Recognize that microorganisms can survive in hot springs

## Introductory activity

Collect different fruits such as oranges, lemons, avocado, green paper, red paper, bananas, mangoes and tomatoes.

- 1. Observe each of the above fruits and group them based on their external features.
- 2. Based on groups made, which fruits are most closely related?

For more than 3.5 billion years, life on earth has been constantly changing. Natural selection and other processes have led to a staggering diversity of organisms. A tropical rain forest, for example, may support thousands of species per meter square. Recall that a species is a population of organisms that share similar characteristics and breed with another to produce fertile offspring. Biologists have identified and named about 1.5 million species so far, and they estimate that between 2 and 100 million additional species have yet to be discovered.

## 2.1. Taxonomic hierarchy

## **Activity 2.1**

You are provided with cards written on a list of words such as continent, district, country, cell, province, sector, village and family.

- 1. Arrange the above words in increasing size
- 2. What is your opinion about the people of the same family and those in the whole country?
- 3. Compare your arrangement above with 8 groups of the biological taxonomic hierarchy.

Taxonomy is the study and practice of classification, which involves placing organisms in a series of taxonomic units, or taxa (singular: taxon). In biological classification, these taxa form a hierarchy. Each kind of organism is assigned to its own species, and similar species are grouped into a genus (plural: genera). Similar genera are grouped into a family, families into an order, orders into a class, classes into a phylum (plural: phyla) and phyla into a kingdom. The domain is at the top of this hierarchical system.

The hierarchy classification starts from the largest group, the **domain.** The eight levels of classification are known as **taxa** (taxon in singular), these include: Domain, Kingdom, phylum, class, order, family, genus and species. As one moves down the taxonomic hierarchy, it follows that the number of individuals decreases but the number of common features increases. For example, there are numerous individuals in the domain Eukarya, with very few features in common.

#### **Binomial nomenclature**

When precision is not required one generally reverts to common names. The trouble is that an organism may be known by different common names, and sometimes the same name may be given to two quite different organisms because common names are not internationally recognized and they change from one region to another one, or from one country to another one. To solve this problem, the **binomial system** also known as **scientific name** was introduced and it was pioneered by the Swedish naturalist **Carl Linnaeus** (1707-1778).

In this system, each organism is given two Latin names: a **generic** name beginning with a capital letter and a specific name beginning with a lower case letter based on the physical characteristics of studied species. The **scientific** name is in italic when printed otherwise it is underlined, when hand written.

For example, many cats belong to the genus Felis but there are many species of cats: A wild cat is *Felis sylvestris while* a domestic cat is *Felis domesticus*. These names are in italic because this book was written by the use of computer. Hierarchy taxonomy of human, earthworm and hibiscus plant are given in the table 2.1.

Table 2.1 Taxonomic classification of human being, earthworm and hibiscus

Taxa	Human	Earthworm	Hibiscus
Domain	Eukarya	Eukarya	Eukarya
Kingdom	Animalia	Animalia	Plantae
Phylum	Chordata	Annelida	Angiospermae
Class	Mammalia	Oligochaeta	Dicotyledonae
Order	Primata	Terricolae	Malvales
Family	Hominidae	Lumbricidae	Malvaceae
Genus	Homo	Lumbricus	Hibiscus
Species	H. sapiens	L. terrestris	H. rosa sinesis

Scientific names present more advantages than common names.

- They are necessary whenever precise identification is required, and they enable scientists to communicate accurately with each other.
- They are used worldwide and have the merit that every biologist knows exactly which organism is being referred to.

Currently, with DNA technology, it is possible to investigate relationships based on genes or DNA structure. As this new technology comes to greater use, it is possible to find that some species had to be reclassified into different taxa.

#### Self-assessment 2.1

- 1. An African bush elephant belongs to order Proboscidae and family Elephantae. Its scientific name is *Loxodonta africana*.
  - a. Make a table indicating the hierarchy classification of African bush elephant
  - b. Use the examples from table 2.1 to define the term "taxon"
- 2. Classify each of the following organisms under the following kingdom, phylum and class taxa: honey bee, cockroach, maize, and spider.
- 3. Describe the system of naming species that Linnaeus developed.

## 2.2. Three domains: Archaea, Bacteria and Eukarya.

## **Activity 2.2.**

Using text books and other sources identify the characteristics of each of the three biological domains

Three domains are used by biologists to divide organisms into three large groups based on their cell structure. The domain is the highest taxon in the hierarchy. The prokaryotes are divided between the domains **Bacteria** and **Archaea**, while all the eukaryotes are placed into the domain **Eukarya**.

#### a. Domain Bacteria

Domain bacteria include prokaryotic organisms as their cells have no true nucleus. They are all microscopic that vary in size between 0.2 to 10 micrometres. The characteristic features of bacteria are:

- Cells with no true nucleus
- DNA exists in circular chromosome and does not have histone proteins associated with it
- No membrane-bound organelles (mitochondria, endoplasmic reticulum, Golgi body, chloroplasts)
- Contain mesosomes as infolding of membrane and acts as sites for respiration.
- Ribosomes (70 S) are smaller than in eukaryotic cells
- Cell wall is always present and contains peptidoglycans in place of cellulose
- Cells divide by binary fission
- Usually exist as single cells or colonies

#### b. Domain Archaea

This contains bacteria that live in extreme environments where few other organisms can survive. They are classified according to the environments they live in;

- Methanogenic bacteria that live in habitats deprived of oxygen and give off methane as a product of metabolism for example those that live in the guts of ruminant animals
- Halophilic bacteria live only in salty conditions
- Thermoacidophilic bacteria tolerate extreme acid and temperature that exceed boiling point of water and a pH below 2.

## c. Domain Eukarya

All the organisms classified into this domain have cells with nuclei and membranebound organelles. Their characteristic features are:

- Cells with a nucleus and membrane-bound organelles
- linear DNA associated with histones arranged within a chromosome in the nucleus
- Ribosomes (80S) in the cytosol are larger than in prokaryotes, while chloroplasts and mitochondria have 70S ribosomes, like those in prokaryotes.
- Chloroplast and mitochondrial DNA is circular as in prokaryotes suggesting an evolutionary relationship between prokaryotes and eukaryotes
- A great diversity of forms: unicellular, colonial and multicellular organisms
- Cell division is by mitosis
- Many different ways of reproduction including asexually and sexually.

#### Self-assessment 2.2

- 1. What are the three domains of living things?
- 2. Describe the ways in which a domain differs from a kingdom?
- 3. It is confirmed that: "Some bacteria can survive in extreme temperatures such as hot springs". Justify this statement.
- 4. How is the information about evolutionary or phylogenetic relationships useful in classification of the living things?

## 2.3. Five kingdoms of organisms

#### Activity 2.3.

- Collect organisms from a habitat near your school including a housefly, spider, frog, gecko, bean/maize plant, moulds/mushroom, spirogyra (algae) and a hen. If there are small rapidly moving land animals such as insects, anaesthetise them by placing them in an ether/ethanol bottle for few seconds. Preserve the collected specimens for future use
  - a. Examine each organism, using a hand lens.
  - b. Make a table of the features observed and identify the kingdom to which each organism belongs.
- 2. Which feature do all animals (except sponges) have that distinguishes them from plants and fungi?

There are different ways of classifying the living world into kingdoms but the most common and recommended is the five kingdom classification.

According to Kent (2000) the kingdoms are:

- Kingdom Monera or prokaryote
- Kingdom Protoctista
- Kingdom Fungi or kingdom mycota
- Kingdom Plantae
- Kingdom Animalia

## 2.3.1. Kingdom Protoctista

This kingdom is made up of a very diverse range of eukaryotic organisms, which includes those that are often called protozoans and algae. Any eukaryote that is not a fungus, plant or animal is classified as a protoctist. The characteristic features of protoctists are listed according to the different phyla due to their diverse range:

- Rhizopods that have pseudopodia for locomotion. Example, amoeba
- Flagellates which are heterotrophic organisms with at least one flagellum for locomotion. Example, trypanosoma.

- Sporozoans which are mainly parasitic organisms that reproduces by multiple fission. Example plasmodium.
- Ciliates which are organisms with cilia. Example paramecium
- Euglenoid flagellates which are organisms with flagella but with a biochemistry quite distinct from that of flagellates. Example Euglena
- Oomocytes which are similar to fungi except that they have cell wall with cellulose. Example Phytopthora infestans; potato blight
- Green algae which are photosynthetic organisms with chlorophyll pigments similar to the ones of plants. Example chlorella
- Red aglae which are photosynthetic organisms with organelles with red pigment as well as chlorophyll. Example, chondrus
- Brown algae which are photosynthetic organisms with organelles which contain brown pigments as well as chlorophy. Example Fucus, sea weed

Living things such as paramecium (a), amoeba (b), euglena (c) and plasmodia belong to the kingdom Protoctista.

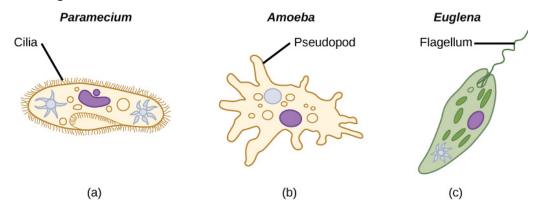


Figure 2.1 Structures of some of the examples of organisms in protoctista showing some common features: (a) paramecium (b) Amoeba (c) Euglena.

## 2.3.2. Kingdom Fungi

Fungi have some similarities with plants, but none of them is able to photosynthesise. They are all heterotrophic, obtaining energy and carbon from dead and decaying matter or by feeding as parasites on living organisms. There is a vast range in size from the microscopic yeasts to what may be the world's largest organisms. Other characteristic features of fungi are:

- Heterotrophic nutrition they use organic compounds made by other organisms as their source of energy and source of molecules for metabolism
- Reproduce asexually by means of spores and sexually by conjugation
- Simple body form, which may be unicellular or made up of long threads called hyphae (with or without cross walls).
- Large fungi such as mushrooms produce large compacted masses of hyphae known as fruiting bodies to release spores
- Cells have cell walls made of chitin or other substances



Figure 2.2 Forms of fungi

## 2.3.3. Kingdom Plantae

Plants are all multicellular photosynthetic organisms. They have complex bodies that are often highly branched both above and below the ground. Characteristic features of plants are:

- Multicellular eukaryotes with cells that are differentiated to form tissues and organs.
- Few specialized cells
- Cells have large and often permanent vacuoles for support with cell walls made of cellulose
- Most plants store carbohydrates as starch or sucrose

## 2.3.4. Kingdom Animalia

Animals (Fig 2.3) are multicellular organisms that are all heterotrophic with different methods of obtaining their food. Organisms in this kingdom have other additional features.

- Different types of specialized cells
- Cells do not have chloroplasts and cannot photosynthesize (although some, such as coral polyps have photosynthetic protoctists living within their tissues)
- Cell vacuoles are small and temporary (for example lysosomes and food vacuoles)
- Cells do not have cell walls
- Communication is by the nervous system

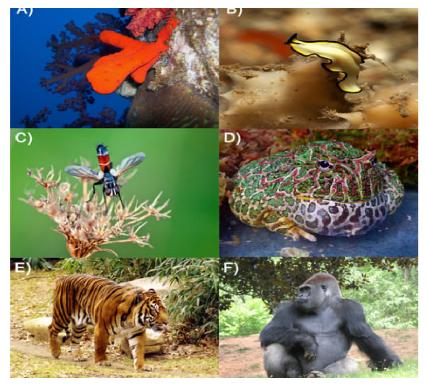


Figure 2.3. A variety of animals (A) Sponge, (B) Flatworm (C) Housefly (D) Frog (E) Tiger (F) Gorilla.

## 2.3.5. Kingdom Monera

Organisms in this kingdom have single cells that do not have a nucleus. They are prokaryotic. They are the smallest and simplest organisms. Examples are bacteria which form a diverse group with members that range widely in size and shape. Some of them stick together to form chains or clusters while others are single cells.

The figure below (Figure 2.4) shows a typical structure of a bacterial cell which contains all the main features of prokaryotes

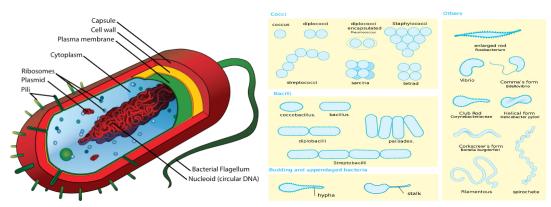


Figure 2.4 (a) Structure of a typical bacterial and (b) bacterial features

### Self-assessment 2.3

- 1. The kingdom protoctista contains groups which do not appear to show an evolutionary relationship. On this basis, is the five kingdom classification a natural or artificial classification?
- 2. What are the three methods that protists use to obtain food?
- 3. Identify three characteristics of protists
- 4. The following is a list of organisms belonging to various kingdoms: housefly (*Musca domestica*), maize (*Zea mays*), Frog (*Rana spp*), Bat and Eagle.
  - a. Classify these organisms into their kingdoms
  - b. Name any two organisms that are not closely related and give a reason.
- 5. How are fungi different from members of kingdom plantae?

# 2.4. Economic importance of bacteria

## **Activity 2.4**

When an animal dies in a forest, it decays after a certain period of time. Once a farmer grows beans in the soil with such dead animal decay, beans grow well.

- 1. What cause the dead animal to decay?
- 2. Why the beans have grown well?

Bacteria are economically important because they are essential in many beneficial biological and industrial processes. There exist some examples of bacteria that are pathogens as they cause disease and spoilage of food..

### 2.4.1. Useful bacteria

# a. Biotechnology

Bacteria are used in biotechnology and industry. They are used to manufacture products such as ethanol, acetone, organic acid, enzymes, and perfumes. In the chemical industry, bacteria are most important in the production of pharmaceuticals. For example, *E. coli* is used for commercial preparation of riboflavin and vitamin K.

# b. Genetic engineering

Bacteria are used in genetic engineering through the manipulation of genes, also called recombinant DNA technology. In this case, bacterial cells are transformed and used in production of commercially important products for example, production of human insulin used in treatment of diabetes.

# c. Decomposition

In addition, bacteria are important in decomposition of dead organisms and animal wastes such as feces to form organic matter. This process improves soil fertility and plays an important role in mineral recycling in an ecosystem.

### d. Fibre retting

Some bacteria including *Clostridium butyricum* are used to separate fibres in a process called retting. In this process, fibres are formed to make ropes and sacks.

### e. Nitrogen fixation

Some other bacteria are used to fix nitrogen in form of nitrates into the soil. For example, *Rhizobium* bacteria which live in root nodules of leguminous plants. Such bacteria help in improvement of soil fertility.

### f. Digestion

Some bacteria living in the gut of ruminant animals such as cattle, horses and other herbivores secrete cellulase, an enzyme that helps in the digestion of cellulose of plant cell walls. Another example is Escherichia coli that live in the human large intestine which synthesizes vitamin B and releases it for human use.

### g. Biological control

Some bacteria are used as biological agents in biological pest control such as Bacillus thuringiensis (also called BT) instead of pesticides. Because of their specificity to the host, these bacteria are regarded as environmentally friendly, with little effect on humans, wildlife, pollinators, or other beneficial insects.

### Self-assessment 2.4

Bacteria are both useful and harmful to humans". Discuss the validity of the statement.

# 2.5. Common bacterial diseases in plants and animals

# **Activity 2.5**

Suppose there is cholera outbreak in your village and the executive secretary invited you to sensitize people about preventive measures against cholera. Prepare a brief presentation for this purpose.

The bacteria that cause diseases are harmful to humans and other animals and are referred to as pathogenic bacteria. The body is a home to many millions of bacteria both useful and harmful to humans.

A bacterial disease is caused by entry of bacteria into a host where they can grow, flourish then causing harm to the host. Bacterial diseases include cholera, tuberculosis (TB), typhoid fever, pneumonia, tetanus, and diphtheria, and bacterial meningitis, tooth decay in humans and anthrax in cattle.

Table 2.2. Common bacterial diseases in humans

Disease	Cause	Mode of transmission	Signs and symptoms	Prevention and control
Cholera	Vibrion cholerae	Contaminated food and drinks water borne.	Diarrhea Dehydration	Washing hands before and after meals.
		Spread by vectors such house flies from human faeces.	Vomiting	Drinking clean water, proper disposal of household waste, and faecal matter.
Dysentery	Shigella bacterium	Waterborne	Diarrhea Abdominal cramps	Washing hands before and after meals,
			·	Drinking clean water , proper disposal of household waste and faecal matter.
Typhoid Fever	Salmonella typhi	Waterborne and contaminated food	Headache, insomnia, fatigue, continuous fever	Washing hands before and after meals.
			sometimes	Drinking clean proper disposal of household waste, and fecal matter.
	Three	Through the	High fever	Vaccination.
4. Meningitis	forms: bacterial,	air from a sick person	Loss of appetite,	
	fungal	to healthy	Strong headache,	
	and viral meningitis	person through coughing and	Stiff neck,	
	meningidis	sneezing.	Nausea and vomiting,	
			Confusion convulsion.	

### 2.5.1. Common Bacterial Diseases in Plants

The table 2.3 common bacterial diseases in plants

Name of disease	Cause	Signs and symptoms
Common scab	Streptomyces scabies	Scabs or patches on surface of potato tubers.
Bacterial blight of coffee	Pseudomonas sp.	Dark swellings or lesions and scotching of leaves.
Black rot of cotton	Thielaviopsis basicola	Rotting of the whole plant
Blight of beans	Xanthomonas compestris	Scotching of leaves

### Self-assessment 2.5

Mr. Green lives in one of the slums in a certain city. He prepares and sells chapattis on street. He is usually very clean, but one morning, he is late for work so he does not bother to wash his hands after visiting the toilet. That day he prepares 400 chapattis all of which are sold. Few hours later, his customer Sandra suffered from a disease with the following signs and symptoms: severe diarrhea, excessive loss of water leading to dehydration, and vomiting. Five dayslater, all his customers were rushed and admitted in hospital due to the same problem.

- 1. Suggest the disease that Mr. Green's customers were suffering from and what caused the disease
  - 2. Name three ways this disease might be spread around city.
  - 3. After reading this scenario, what message do you have for people who are like Mr. Green?
  - 4. Suppose you were the health officer for the area in town with such a problem. What steps would you take to prevent the disease from spreading further?
  - 5. House flies are described as vectors. Describe how houseflies transmit diseases to humans.

## 2.6. Structure and classification of Viruses

# **Activity 2.6**

Visit the internet and conduct a research to explain the reasons why viruses are not classified in any of the five kingdoms of living organisms.

Viruses are microorganisms whose structure is only visible with electron microscopes. Viruses are acellular and lack cellular structure. Viruses have none of the features that we traditionally use for classification. They are particles made of proteins and nucleic

acids that are found in all cellular organisms, but show metabolism only once inside the host cell.

When they infect cells, they use biochemical machinery and proteins of the host cell to copy their nucleic acids and to make proteins coats often leading to destruction of the host cells. The energy for these processes is provided by the ATP from the host cell.

### 2.6.1. Structure of a virus

A typical virus consists of DNA or RNA within a protective protein coat called **capsid**. The shape of the capsid may vary from one type of virus to another, as shown in Figure 2.5 below.

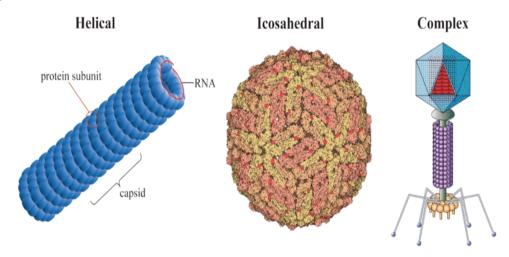


Figure 2.5 Simplified diagrams of different shapes of viruses

Some viruses have an envelope of phospholipids and proteins. The envelope is made from portions of the host's cell membrane. It surrounds the capsid and helps protect the virus from the host's immune system. The envelope may also have receptor molecules that can bind with host cells and facilitate the virus to infect the cells.

### 2.6.2. Characteristics of viruses

An individual virus is called a **virion**. It is a tiny particle much smaller than a prokaryotic cell. Because viruses do not consist of cells, they also lack cell membranes, cytoplasm, ribosomes, and other cell organelles. Without these structures, they are unable to make proteins or even reproduce on their own. Instead, they must depend on a host cell to synthesize their proteins and to make copies of themselves. Viruses infect and live inside the cells of living organisms. They are also regarded as parasites since they depend entirely on living cells for their survival. Although viruses are not classified as living things, they share two important traits with living things: They have genetic material, and they can evolve.

### 2.6.3. Classification of viruses

Viruses can be classified according to:

- Type of nucleic acid molecules of DNA or RNA, forming the core of the capsid:
   Most animal viruses contain RNA while plant viruses contain DNA
- Type of host cell: plant or animal viruses as they are specific to their hosts
- Presence or absence of the envelope: Plant viruses' bacteriophage are nonenveloped while animal viruses like HIV and influenza virus are enveloped.

### 2.6.4. Viruses and human disease

When viruses infect cells of their host, they cause disease. Examples of diseases caused by viruses include HIV/AIDS, influenza (flu), chicken pox, and the common cold. The human immunodeficiency viruses that causes AIDS is a retrovirus. Other viral diseases include rabies, measles, diarrheal diseases, hepatitis A, B and C, polio, and cold sores. One-way virus cause disease is by causing host cells to burst open and die. Viruses may also cause disease without killing host cells. They may cause illness by disrupting homeostasis in host cells.

Some viruses live in a dormant state inside the body. The virus that causes chicken pox may infect a young child and causes the short-term disease chicken pox. Then the virus may remain latent in nerve cells within the body for decades. The virus may re-emerge later in life as the disease called shingles, where the virus causes painful skin rashes with blisters. Some viruses can cause cancer. Examples include the human papillomavirus (HPV) causing cancer of the cervix in females. Hepatitis B virus causes cancer of the liver. A viral cancer is likely to develop only after a person has been infected with a virus for many years.

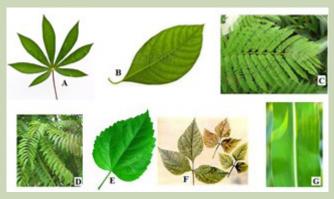
#### Self-assessment 2.6

- 1. What is meant by the term virus?
- 2. State the main components of a virus.
- 3. Describe the two ways how viruses cause an infection.
- 4. Differentiate between a bacteriophage and a retrovirus?
- 5. Do you think viruses should be considered as a form of life? Give reasons for your answer.

# 2.7. Dichotomous key of identification of organism

## **Activity 2.7.1**

The figure below represents different types of plant leaves. Make a classification of these plants based on the external structure of the leaves.



The dichotomous key is also referred to as biological identification key. It is made up of a series of contrasting statements called **leads** indicated by the numbers 1, 2, 3... where each lead deals with a particular observable characteristic. The characteristics used in keys should be readily **observable morphological features** which may be either **qualitative**, such as shape of abdomen, nature of legs, or **quantitative**, such as number of antennae, number of pairs of legs and length of the antennae in case of arthropods. It is essential to note that **size** and **color** are often less considered as both can be influenced by the environment, the season, the age or state of the organism at the time of identification.

# 2.7.1. Guidelines used in construction of a dichotomous key:

The following guidelines must be considered while constructing a dichotomous key.

- Use morphological characteristics which are observable as much as possible such as leaf venation, nature of margin, apex, lamina and nature or length of the petiole (leaf stalk).
- Start with a major characteristic that divide the organism or the specimen into two large groups such as the type of a leaf.
- Select a single characteristic at a time and identify it using a number for example: simple leaf......go to 2, compound leaf.....go to 5. This means that in 2 you will deal with only simple leaves and 5 only compound leaves.
- Use similar forms of words for two contrasting statements for example at 2: leaf with parallel venation ......go to G and leaf with network venation .....go to 3.
- The first statement should always be positive.
- Avoid generalizations or overlapping variations, be specific and precise to the point.

## **Example**

- Collect leaves from the following plants: cassava, avocado, jacaranda, cassia, hibiscus bean, maize or paspalum grass,
- Label different leaves collected as, A, B, C, D, E, F and G
- Observe and familiarize with the specimens before starting the experiment to minimize errors during the identification process
- Make a table summarising the specimens and steps followed to identify each of them.
- Construct a dichotomous key based on the observable features (characteristics) and table of steps followed.

## Solution: The dichotomous key of specimens A, B, C, D, E, F and G.

1.	a. Simple leaves	go to 2
	b. Compound leaves	go to 5
2.	a. Parallel venation	G
	b. Network venation	go to 3
3.	a.Simple digitate	A
	b. Non simple digitate	go to 4
4.	a. Leaf with serrated margin	E
	b. Leaf with smooth margin	В
5.	a.Leaf with three leaflets (compound trifoliate)	F
	b. Leaves with more than three leaflets	go to 6
6.	a. Pinnate leaf	D
	b. Bipinnate leaf	С

### 2.7.2. Common features used for identification of animals

Animals are classified based on the following features:

- Locomotory structures such as legs, wings and fins
- Antennae (presence, nature and number)
- Presence or absence of eye and eye type
- Number of body parts for example insects have three body parts
- Body segments (nature and number)
- Body surface structures such as fur, hair, feathers and scales
- Feeding structures such as mouth parts in arthropods for example in insects
- Type of skeleton present such as endoskeleton, exoskeleton and hydrostatic

# 2.7.3. Common features used for identification of plants

Plants can be classified basing on the following features:

- The leaf structure such as nature of apex, margin, venation, lamina and petiole
- The flower structure including inflorescence type, flower shape and number of floral parts
- The type of stem (woody, fleshy and herbaceous), shape (rectangular, cylindrical) and texture of the stem (smooth, spiny and thorny) ...
- The type of root system, tap root, storage root, fibrous roots...

### **Precaution**

- Care must be taken while collecting and handling some organisms because some are poisonous, have thorns and others are able to sting
- Collection of specimen should be done a day or few days before the experiment depending on nature of the experiment
- Avoid and try to minimize where possible, uprooting, cutting down or plucking and pruning of plants as this may threaten the biodiversity as well as result into environmental degradation

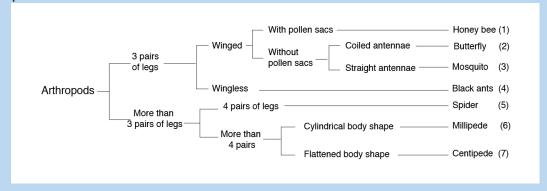
## **Activity 2.7.2**

Construct and interpret a dichotomous key of arthropods listed below.

- Collect the following litter arthropods: honey bee, spider, millipede, butterfly, sugar ant, centipede and mosquito and label each specimen as A, B, C, D, E, F and G respectively
- Observe and familiarize yourself with the specimens before starting the experiment.
- Use sharply contrasting external features of collected specimens /diagrams to construct a dichotomous key.

#### Self-assessment 2.7

Read and interpret the dichotomous tree below and use it to answer the following questions.



- 1. Specify the phylum of kingdom animalia represented by the above dichotomous tree?
  - Give one observable reason to support your answer.
- 2. According to this dichotomous tree, which characteristic feature was used to classify different insects?
- 3. Which observable characteristic feature distinguishes between a spider and a mosquito?
- 4. How does a millipede differ from a centipede?
- 5. To which classes do a millipede and a centipede belong?
- 6. Which class of arthropods is not represented on the dichotomous tree?

### **End of unit assessment 2**

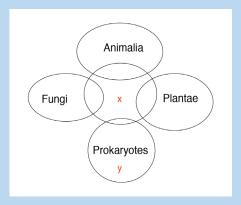
- 1. Which one of the following living organisms belongs to domain bacteria?
  - a. Euglena
  - b. Vibrio cholerae
  - c. Paramecium
  - d. moulds
- 2. The group of classification where organisms resemble one another and are capable of interbreeding together to produce viable offspring is known as:
  - a. Species
  - b. kingdom
  - c. Genus
  - d. Phylum

- 3. Which one of the following is not a kingdom of living organisms?
  - a. Monera
  - b. Animalia
  - c. Annelida
  - d. Protoctista
- 4. Which one of the following is a characteristic feature common to fish, reptiles and birds but absent in mammals?
  - a. Possession of scales
  - b. Has no limbs
  - c. Possession of feathers
  - d. Undergo internal fertilization
- 5. Which one of the following statements about fish is not correct?
  - a. Fish live both in water and on land and undergo external fertilization.
  - b. Most fish have bones while others are cartilaginous
  - c. Most fish have streamlined body, lateral line and swim bladder.
  - d. Gills are organs for gaseous exchange in fish
- 6 Which one of the following is not a characteristic of all insects?
  - a. They have three body parts namely head, thorax and abdomen.
  - b. They have three pairs of jointed legs attached on segment of the thorax.
  - c. They have four pairs of jointed legs
  - d. They have a pair of antennae attached on the head.
- 7. The following are characteristics of all mammals except;
  - a. They have mammary glands to secrete milk feed their young ones.
  - b. Their skin is covered with hair.
  - c. Undergo internal fertilization and internal development of the embryo.
  - d. They have a pair of wings made up feathers.
- 8. The point where the leaf joins the stem is called;
  - a. Apex
  - b. Margin
  - c. Leaf base
  - d. Lamina
  - e. Length of petiole.
- 9. Which of the following is less considered while identifying feature to construct a dichotomous key of leaves?

- a. Nature of margin
- b. Nature of apex
- c. Size and color of leaf
- 10. The following are characteristics of arachnids except;
  - a. Four pairs of jointed legs
  - b. Two body parts
  - c. Three body parts
  - d. Do not have wings
- 11. Match the structures with the organisms which possess them.

Structures	Organisms
Antennae	Fungus
Flagella	Snail
Spores	Housefly
Coiled shell	Euglena
Pseudopodia	Amoeba
Cilia	Paramecium

12. A group of S4 students drew a Venn diagram below to summarize the five kingdoms into which organisms are classified. Study the diagram and answer the questions that follow:



- a. Which kingdoms are represented by the letters x and y?
- b. State one characteristic that organisms of x may share with:
  - i. Prokaryotes
  - ii. Fungi
  - iii. Plantae

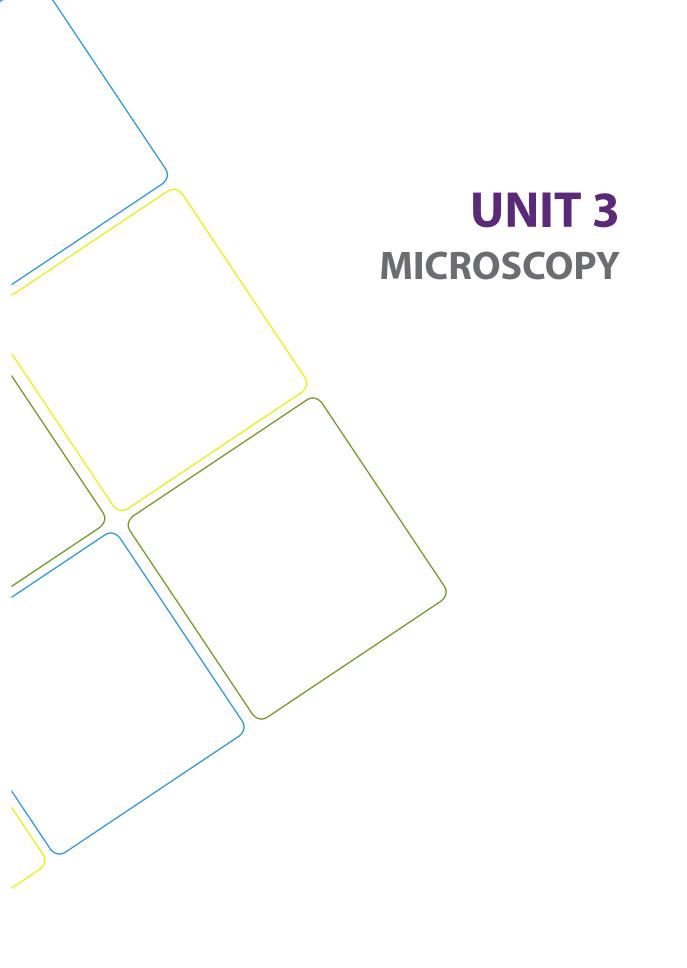
13. Complete the table to summarize the characteristics of each class of phylum Arthropoda.

Characteristic	Insecta	Arachnida	Crustacea	Chilopoda	Diplopoda
Number of body parts					
Number of limbs or legs					
Gaseous exchange structure					
Number of antenna					
Number of wings					
Number and nature of eyes					

- 14. What is the significance of classification of living organisms?
- 15. The binomial system of naming a blue monkey, Cercopithecus mitis, is provided below;

Complete the table by filling the missing words.

kingdom	
	Chordata
Class	
Order	
family	Hominidae
	mitis



## **UNIT 3: MICROSCOPY**

## **Key Unit Competence**

Distinguish between the types of microscopes and their principal uses.

### **Learning objectives**

By the end of this unit, I should be able to:

- Describe the main features and functions of the components of a compound light microscope.
- Manipulate a compound light microscope to observe prepared slides.
- Show perseverance when using light microscopes.
- Pay attention when using a compound light microscope to avoid damage of the lenses, mirrors and slides.
- State that magnification is the increase in the apparent size of the object.
- State that resolution is the ability of the microscope to show two objects as separate.
- Appreciate the importance of magnifying instruments in Biology.
- Use of a microscope to determine the relationship between actual size of the specimen and the image.
- Calculate the approximate size of different biological structures using an appropriate unit of measurement
- State the advantages and disadvantages of using an electron microscope.
- State the principles and limitations of TEM (Transmission Electron Microscopy).
- State the advantages and disadvantages of using SEM (Scanning Electron Microscopy).
- Compare light and electron microscopes
- Acknowledge the use of electron microscopes in modern science with reference to electron micrographs.
- Observe and draw biological specimens under a light microscope.
- Prepare temporary slides for observation under light microscopes using different objective lenses
- Appreciate the importance of magnifying instruments in Biology

# **Introductory activity**

Point out scientific activities that require the use of microscope in our daily lives.

A microscope is used to produce a magnified image of an object or specimen. Anton Van Leeuwenhoek (1632-1723) was the first to invent a microscope powerful enough to explore the world of microbes. His discoveries stimulated an explosion of interest in scientific use of microscopes. Since the 18th century, many new types have been invented of which the most commonly used today are the compound light microscope and the electron microscope.1 (Kent, 2000, p. 58)).

# 3.1. Compound Light Microscope

## **Activity 3.1.1**

Some of the living things including protoctista and fungi have small size to be observed by naked eyes. Discuss the ways used by biologists to observe and identify different parts of these living organisms.

The optical microscope, often referred to as light microscope is a type of microscope which uses visible light and a system of lenses to magnify images of small samples.

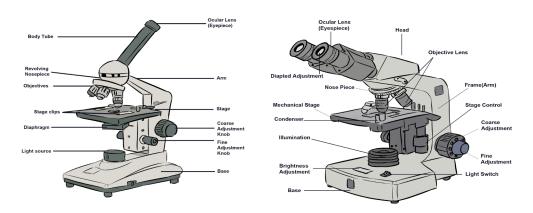


Fig.3.1. Monocular light microscope (1) and Binocular light microscope (2) both have two lens systems; the objective lens and the ocular or eye piece lens that are used in combination to view an image

The different parts of light microscope are described below:

- Base: supports and stabilizes the microscope on the table or any other working place
- Light source: It is made by lamp or mirror which provides light for viewing the slide.
- **Stage:** is a platform used to hold the specimen in position during observation.
- **Stage clips:** are pliers used to fix and hold tightly the slide on stage.
- **Arm:** supports the body tube of microscope
- Body tube: maintains the proper distance between the objective and ocular lenses
- Arm: used for holding when carrying the microscope and it holds the body tube which bears the lenses.
- Coarse focus adjustment: moves stage up and down a large amount for coarse focus
- **Fine focus adjustment:** moves stage up and down a tiny amount for fine focus
- **Objective lenses:** focuses and magnifies light coming through the slide

- **Revolving nosepiece:** rotates to allow use of different power objectives
- **Slide:** is a transparent pane on which a specimen is placed.
- **Eye piece/ocular lens:** magnifies image produced by objective lens
- Condenser: It will gather the light from the illuminator and focus it on the specimen lying on the stage. The function of the condenser is to focus the light rays from the light source onto the specimen.
- Iris diaphragm lever: This allows the amount of light passing through the condenser to be regulated to see the object.

### **Activity 3.1.2**

## Using the light microscope

## a. To observe under low power and low magnification, proceed as follows:

- Objects (specimens) to be observed under the microscope are first placed on a glass slide and covered with a cover slip.
- Place the specimen on the stage of your microscope; in other words, arrange
  it so that the specimen is exactly at the center of the hole at the stage.
- Fix the slide in place with two clips.
- Rotate the nosepiece so that small objective lens is immediately above the specimen.
- Set the angle of the reflector mirror so that light is directed up through the microscope.
- Look down the microscope through the eye piece. Adjust the iris diaphragm so that the field of vision is bright and not dazzling.
- Turn the course adjustment knob until the tip of the objective lens is close to the slide.
- Now look down the microscope again. Slowly turn the course adjustment knob in the other direction, so the tube gradually moves upwards. The specimen on the slide should eventually come into view.
- Use the course and fine adjustment knobs to focus the object as sharply as possible.
- If necessary readjust, the iris diaphragm so the specimen is correctly illuminated. You will get a much better image if you don't have too much light coming through the microscope.

# b. To observe under high power at a greater magnification, proceed as follows:

- Rotate the nosepiece so that the large objective lens (with higher magnifying power) is immediately above the specimen. The nosepiece should click into position, as before.
- If the specimen is not in focus, focus it with fine adjustment knob. Be careful
  that the tip of the objective lens does not touch the slide.
- Readjust the illumination if necessary.

Microscope uses transmitted light for observation. However, microscope uses specific light characteristics for specific samples, such as transparent specimens and samples that do not pass light. All parts of a microscope work together, the light from the illuminator passes through the aperture, through the slide, and through the objective lens, where the image of the specimen is magnified. Then the magnified image continues up through the body tube of the microscope to the eyepiece, which further magnifies the image the viewer can see.

Light from the source is focused on the specimen by the **condenser** lens. It then enters the **objective** lens, where it is magnified to produce a **real image**. The real image is magnified again by the **ocular** lens to produce a **virtual image** that is seen by the eye.

### Care of the compound microscope

The microscope is an expensive instrument that must be given proper care. Always general instructions have to be respected when using a microscope. These include:

- Carry the microscope with both hands, one hand under the base, and the other on the arm.
- When getting ready to put the microscope away, always return it to the low power or scanning power setting.
- When setting the microscope on a table, always keep it away from the edge.
- It is generally better to clear your lab table of items that are not being used.
- Never clean lenses with anything other than lens paper, don't use towels and other paper tissues because they scratch the lens.
- Inform the instructor or the biology lab technician if there is any microscope damage or irregularity in its operation as soon as possible. Do not return a faulty microscope without first informing the instructor or lab technician.
- You are responsible for the microscope while using it treat it with care!

### Self-assessment 3.1

1. Complete the table below:

Parts of microscope	Functions	
Base		
	rotates to allow use of different power objectives	
Coarse focus adjustment		
	focuses and magnifies light coming through the slide	
Eye piece / ocular lens:		

- 2. What is the importance of a light microscope?
- 3. Suggest a reason why it is not advisable to clean the objective and eye piece lens with a wet cloth or towel?

# 3.2. Magnification and resolution of a compound light microscope.

# **Activity 3.2.1**

Work out the following equivalent measurements:

- 1. 1 millimetre (mm) =..... metre (m)
- 2. 1micrometre (μm) =.....mmetre (m)
- 3. 1 nanometre (nm) =....metre (m)
- 4. 1 metre (m) = ......mm = .........  $\mu$ m = ......nm,
- 5. 1 kilometre (km) = .....m

## a. Magnification

Magnification refers to increase in the apparent size of the object, while resolution of a microscope is the ability to show two close objects as separate. The maximum magnification of an ordinary light microscope is about x1500. Magnification must be written on the right side and below the biological drawing and it does not have units. The size of the image is measured in mm but converted into micrometres or nanometres to work out the actual size. It is calculated as follows:

$$Magnification = \frac{\text{measured size}}{\text{actual size}} = \frac{\text{size of image}}{\text{actual size of object}}$$

## **Example**

Calculate the magnification if the actual size is  $5\mu m$  and the measured image of the specimen has the size of 40mm.

### **Answer:**

- Make the size of the image and the actual size in the same units by converting mm in  $\mu$ m. This is done by multiplying 40mm by 1000 so that 40mm = 40000  $\mu$ m
- Magnification =  $\frac{\text{measured size}}{\text{actual size}} = \frac{\text{size of image}}{\text{actual size of object}} = \frac{40000 \ \mu\text{m}}{5 \mu\text{m}} = (40000 \ \mu\text{m})/5 \mu\text{m}$

Note that the magnification of the specimen under a light microscope is calculated by multiplying the magnification of the objective used to that of the eyepiece. For example: 10x (objective) 10x (eyepiece) = x100.

### b. Microscopic observation

### **Activity 3.2.2**

Using prepared slides of microorganisms such as a bacterium, amoeba, and paramecium.

Observe, draw and label the visible parts under a light microscope. Avail these materials before you start: Petri-dishes, plate covers, pencil, transparent tape, microscope, agar powder, and permanent slide of bacteria, amoeba, and paramecium, Bunsen burner or any other source of heat.

### **Procedure**

- Prepare agar medium by boiling a mixture of 10g of agar powder with 50ml of water
- Label a control and exposed petri dishes in which you pour prepared agar medium.
- Cool both plates for 20 minutes until the medium hardens.
- Tape closed the cover of the control plate and removes the cover of the exposed plate.
- Leave both plates for 5 minutes, and do not touch or breathe on the agar.
   After five minutes, tape closed the lid of the exposed plate and store both plates upside down in a warm place and draw your observations
- Repeat the observation by using mounted slides of amoeba and paramecium and make a comparison between bacteria, amoeba and paramecium: what is your conclusion?

For this experiment, light microscope allows to observe organisms of small size including bacteria, amoeba and paramecium. Some other parts of macroscopic organisms such as cells and tissues of plants and animals or some parts of these living organisms such as stems and roots can also be observed under light microscope. Some specimens can be observed directly after collection and preparation. However, some of the details might not be clearly observed because specimens are not colored. Also, some material distorts when you try to cut the specimen into thin sections. To overcome this challenge, slides can be prepared in advance by the use of the following steps:

- Staining: colored stains are chemicals that bind to chemicals on or in the specimens. This allow the specimen to be seen. Some stains bind to specific cell structures. For example, acetic orcein stains DNA dark red, while gentian violet stains bacterial cell walls.
- Sectioning: specimens are embedded in wax, where thin sections are then
  cut without distorting the structure of the specimen. This is particularly useful
  for making sections of soft tissue, such as brain. Safety measures might be
  taken. Make sure that hands are washed with soap and warm water after the

experiment. Use a disinfectant to wipe down all surfaces where bacteria may have been deposited for example. Be sure that some substances and animals might be harmful to the life.

## **Activity 3.2.3**

### Preparing of temporary slides and observation under light microscope

Make temporary preparation of slides of epidermis of onions young stems by fixing, staining and mounting. Observe under low and high power of a light microscope.

### **Preparation and procedures**

- Add a drop of water at the center of the microscopic slide to flatten the membrane
- Pull of a thin membrane from the onion layer and lay it at the center of the microscopic slide
- Add a drop of iodine solution or methylene blue on the onion membrane
- Gently lay a microscopic cover slip on the membrane and press it down gently using a needle to remove air bubbles.
- Touch a blotting paper on one side of the slide to drain excess iodine/water solution,
- Place the slide on the microscope stage under low power to observe.
- Adjust focus for clarity to observe.
- Make another slide without adding the stain to see the difference between a stained slide and a non- stained slide.
- Draw and label the observed parts of each of the two slides and compare a drawing of a stained slide and that of a non-stained slid.

# c. Measuring cells

Cells and organelles can be measured with a microscope by means of an eyepiece called graticule. This is a transparent scale, usually having 100 divisions (Figure 3.4, A). The eyepiece graticule is placed in the microscope eyepiece so that it can be seen at the same time as the object to be measured (Figure 3.4, B). At this figure (Figure 3.4, B), the cell lies between 40 and 60 on the scale, so that it measures 20 eyepiece units in diameter (60 - 40 = 20).

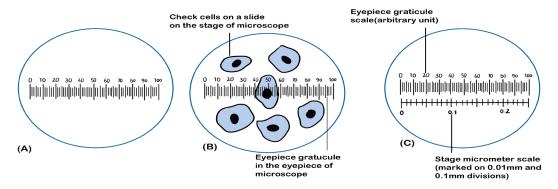


Fig. 3.2. Measuring the length of cell using a graticule in a light microscope

To calibrate the eyepiece graticule scale, a miniature transparent ruler called a stage micrometer scale is placed on the microscope stage and is brought into focus. This scale may be fixed onto a glass slide or printed on a transparent film. It commonly has subdivisions of 0.1 and 0.01 mm. The images of the two scales can then be superimposed (Figure 3.4, C). If in the eyepiece graticule, 100 units measure 0.25 mm, the value of each eyepiece unit equals  $\frac{0.25}{1.00} = 0.0025$  mm.

By converting mm to  $\mu$ m, the value of eyepiece equals  $\frac{(0.25 \times 1000)}{100} = 2.5 \, \mu$ m. The diameter of the cell shown superimposed (Figure 3.4, B) measures 20 eyepiece units. Its actual diameter equals  $20 \times 2.5 \, \mu$ m = 50  $\mu$ m. This diameter is greater than that of many human cells because the cell is a flattened epithelial cell.

Use the following instructions to measure the length of one cell

- Measure the distance in millimetre from the start of one cell to the end of 10 cells
- Divide by 10 to find the length of one cell in the specimen.
- Convert this length in millimetre to micrometer by multiplying by 1000.
- Find the actual length of a cell by dividing this length by the magnification of thespecimen got from the product of eye piece and objective lens used.

### Self-assessment 3.2.

- 1. Calculate the magnification of an image with 50mm, and the object measuring 5µm. in length.
- 2. If a nucleus measures 100mm on a micrograph, with a magnification of X10 000, what is the actual size of the nucleus?

# 3.3 Electron microscopes

## **Activity 3.3**

Suggest the form and source of energy used by electron microscope. How does this differ from that used by a compound microscope?

An electron microscopes use a beam of accelerated electrons as a source of illumination.

Electron beams have a much smaller wave length than light rays and therefore have greater resolving powers and can produce higher effective magnifications than light microscopes. There are two types of electron microscopes;

- Transmission electron microscope (TEM)
- Scanning electron microscope (SEM)

Electron microscopes are used to study the details of internal structures (the ultrastructures) of cells. Most modern TEMs can distinguish objects as small as 0.2nm. This means that they can produce clear images magnified up to 250,000 times.

## Formation of an image by the TEM:

- Extremely thin samples of the specimen are needed and are cut by using diamond or glass knives as they are supported in resin block to prevent them from collapsing
- The section is then impregnated with a heavy-metal stain
- As the beam passes through the specimen, electrons are absorbed by the heavily stained parts but passes readily through the lightly stained parts.
- Electro magnets bend the electron beam to focus an image onto the florescent screen or photographic film to form an electron micrograph

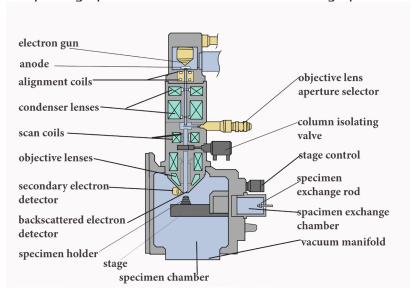


Fig. 3.3. A transmission electron microscope showing how a beam of electrons is emitted and processed to form an electron micrograph on a florescent screen

# Scanning electron microscope (SEM)

The SEM is used to produce 3D images of surfaces of the specimens. Electrons are reflected from the surface of the specimen stained with a heavy metal. This enables the SEM to produce images of all specimens, cells, tissues, or even organisms

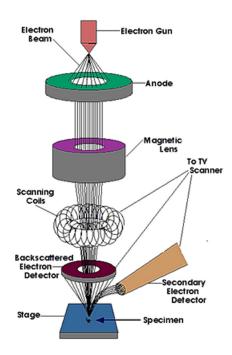


Fig. 3.4. Image of Scanning Electron Microscope (SEM)

### a. Advantages of the electron microscope over light microscope

Electron microscope has a higher resolution and istherefore able of a higher effective magnification estimated at up to 250,000 million times compared to the light microscope which can show a useful magnification only up to 1000-2000 times. This is because a light microscope uses a beam of light with a longer wave length while Electron microscopes use a beam of electrons that have a short wave length.

# b. Disadvantages of electron microscope

Despite the advantages, electron microscope presents a number of setbacks and limitations.

- They are extremely expensive and the maintenance costs are high.
- Sample preparation is often much more technical requiring special training.
- Samples must be dead, exposed to high radiation and are placed in a vacuum so that it is impossible to observe living specimens
- It is not possible to observe colors because electrons do not possess a color. The image is only black-white, even if sometimes the image is colored artificially to give a better visual impression.
- They require more training and experience in identifying artifacts that may have been introduced during the sample preparation process.

# c. Comparison between light and electron microscopes

Light and electron microscope presents the following similarities and differences. The following are some of the similarities:

- Both light and electron microscopes form larger (magnified) and more detailed (highly resolved) images of small objects or small areas of larger objects
- Both light and electron microscopes are used in biology study, research and medical sciences particularly histology, material sciences such as metallurgy and other aspects of science.
- Specimens must be carefully prepared using techniques appropriate for both the equipment and the sample including slicing, staining, and mounting.

Despite the similarities, light and electron microscope presents differences such as these summarized in the following table:

Table 3.1. Differences between light and electron microscopic

Criteria	Light microscopes	Electron microscopes	
Size	Smaller and lighter, so are easier to move and set-up	Large and heavy, so are not easier to move and set-up	
Cost	Less expensive	Very expensive	
Radiation type	Use light (approximate wavelength: 400-700 nm)	Use beams of electrons (approximate equivalent wavelength: 1 nm).	
Control of image formation	Light via glass lenses	Beams of electrons can be focused using electromagnets due to negative charges of electrons.	
Resolution	Lower resolution than electron microscopes (200nm)	Higher resolution - good for measuring sizes of smaller features (1nm)	
Magnification	Lower magnification than electron microscopes (x2 000)	Higher magnification - so several micrographs may be needed per specimen (x1 000 000)	
Depth: 2d or 3d images	Image plane approximate flat (2d) but, as above, can adjust focus through specimen.	2d only in tem. SEM shows surface images, hence a depth info that seems like 3d	
Thickness of specimen	Specimen must be thin but can adjust focus to different positions (heights) within thin specimen on glass slide	Very thin sections in tem and images surfaces in SEM	
Color images	Images including the range of wavelengths (colors) are provided by the light source. Colors seen are often due to stains rather than the actual colors present in nature	Produce grey scale black and white images. False-color electron micrographs are common	

Preparation of specimens	Simpler preparation and staining is still required	Harsher preparation procedures including use of corrosive chemicals that may cause artefacts in the resulting micrographs
Living cells and tissues	Can watch living processes take place such as microscopic pond life in action, and even cell division	Not possible to view any living material due to vacuum inside electron microscope
Image formation	Images can be viewed directly	Require use of a fluorescent screen, photographic plate or electronic display because electrons cannot be observed directly by the human eye.
Usage limitations	Living or dry specimens can be viewed directly	Living specimens cannot be viewed because electron microscopes require a vacuum in the tube - otherwise the electrons would be absorbed by air molecules

### Self-assessment 3.3

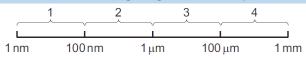
- 1. How is magnification varied in;
  - a. A light microscope
  - b. An electron microscope?
- 2. Why is the resolving power of an electron microscope such better than that of a light microscope?
- 3. Make a comparison between light and electron microscope, highlighting the advantages and disadvantages for each type of microscope.

 $Summarise the similarities and differences between light and electron \, microscopes$ 

### **End of unit assessment 3**

# Section A. Multiple choice questions

1. Which ranges can be viewed using a light microscope?

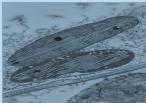


- a. 4 only
- b. 1 and 2 only
- c. 2 and 3 only
- d. 3 and 4 only
- 2. The figure below shows a mitochondrion drawn from an electron micrograph. Study it carefully and answer the following questions.



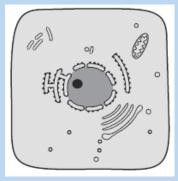
If the length of the mitochondrion line X Y is 3000 nm. What is the magnification of the drawing of the mitochondrion?

- a. ×100
- b. ×1000
- c. ×10 000
- d. ×100 000
- 3. A light microscope is used to observe two membranes that are 200 nm apart. How far apart are the membranes when the objective lens is changed from low power (×40) to high power (×400)?
  - a. 2 µm
  - b. 20 μm
  - c. 200 nm
  - d. 2000 nm
- 4. The electronmicrograph below is that of a chloroplast.

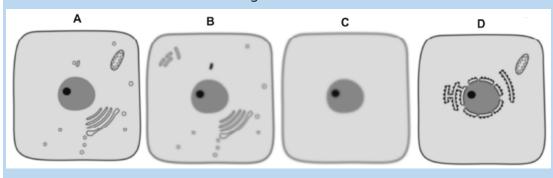


The length of the chloroplast along the line shown is 80 mm. The actual length of the chloroplast is 10  $\mu$ m. What is the magnification of the chloroplast?

- a.  $\times 8 \times 102$
- b.  $\times 8 \times 103$
- c.  $\times 8 \times 104$
- d.  $\times 8 \times 106$
- 5. The following diagram below is drawn from an electron micrograph of an animal cell.

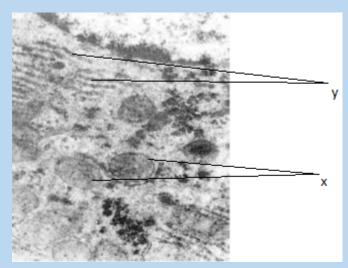


Which represents the same cell, seen under a light microscope at  $\times 400$  magnification?



### **Section B:**

6.



The micrograph above shows a transverse section of part of an animal cell.

- a. Identify organelles labelled X and Y
- b. The maximum actual diameter of Y is 2um. Calculate the magnification of this organelle in the electron micrograph.
- c. Determine with two reasons whether the cell is prokaryotic or eukaryotic.
- d. From evidence in the electron micrograph, deduce two substances that were being synthesised by large quantities by this cell.
- e. The dark granules in the cell are glycogen. Explain the conclusion that you draw from this information



## **UNIT 4: CELL STRUCTURE AND SPECIALIZATION**

## **Key Unit Competence**

Describe the structure and function of cells in an organism.

### **Learning objectives**

By the end of this unit, I should be able to:

- Identify plant and animal cell structures visible under a light microscope.
- State functions of cell structures as seen under an electron microscope.
- Describe the nature of artefacts.
- State the importance of freeze fracturing for examining membrane structure.
- Explain how cell organelles can be isolated by cell fractionation.
- List the functions of cell membranes.
- Describe the fluid mosaic structure of cell membranes.
- Explain the role of the different components of a cell membrane.
- Explain cell specialization as the differentiation of a cell or process to do a particular function.
- Interpret charts and micrographs to relate the structure of specialized cells to their functions.
- Prepare, observe and draw diagrams for specimens on temporary slides for:
   Wandering Jew, in plants and cheek cells under a light microscope.
- Distinguish between ultra-structures of plant cells and animal cells.
- Compare ultra-structures of prokaryotic and eukaryotic cells
- Show resilience and be aware of artefacts when preparing temporary slides.
- Appreciate the importance of cell specialization in multicellular organisms.

# **Introductory Activity**

Using addition resources to your textbook available in your school such as the books from the school library and search further information from the internet:

- 1. Differentiate between prokaryotic and eukaryotic cells.
- 2. By using charts for the two cells, identify different organelles of eukaryotic cell that may perform functions similar to those of a prokaryotic cell.

# 4.1. Ultra-structure of a cell

### **Activities 4.1**

- 1. Observe the chart given for Ultra structure of a cell and identify parts that are easily recognizable when compared with a photomicrograph form a light microscope.
- 2. Identify the mitochondria and ribosomes and state their roles in the life of the cell.

Cytology is the study of the structure and function of cells. A Cell is the basic unit of

life. All living organisms are made up of cells.

Living organisms are classified into:

- Unicellular organisms are made of only one cell, such as bacteria,
- Multicellular organisms are animals and plants composed of many cells. In multicellular organisms, cells divide and then undergo differentiation or specialisation for specific functions.

## Cell theory.

The cell theory states that all living organisms are made up of cells, and cells are the basic unit of structure function in all living organisms.

The main principles of cell theory are based on the following ideas.

- All known living organisms are made up of one or more cells,
- All cells come from pre-existing cells by division
- Cells contain the hereditary information that is passed from cell to cell during cell division.
- Metabolism takes place in cells
- Given suitable conditions, cells are capable of independent existence

When a cell is viewed under light microscope, the most obvious features observed are the very large nucleus and a clear cytoplasm surrounded by a cell membrane. **However,** under electron microscope, it is possible to identify a range of organelles in plant and animal cells. **Ultrastructure** is the detailed of cell as revealed by the electron microscope.

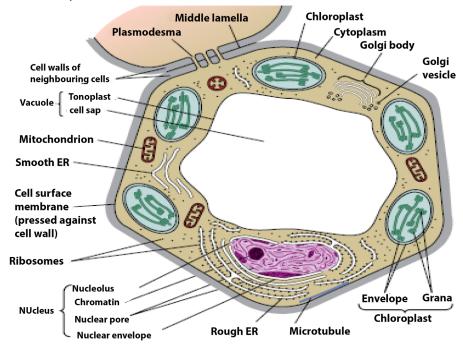


Figure 4-1: Ultrastructure of a typical plant cell (**Adapted from** Cambridge International AS and A Level Biology Course Book Fourth Edition)

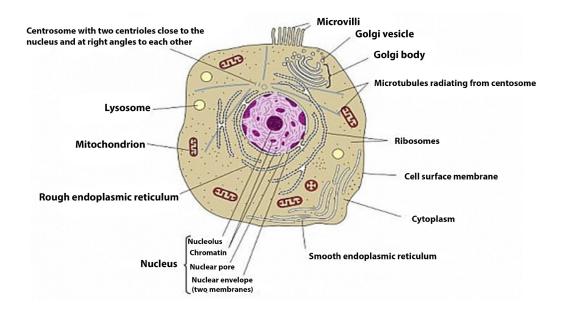


Figure 4.2 **A generalized animal and cells showing structures visible under electron microscope Adapted from** Cambridge International AS and A Level Biology Course Book Fourth Edition

## Similarities between animal cell and plant cell

- Both have a cell membrane, a cytoplasm and a nucleus.
- Both animal and plant cells have mitochondria, Golgi apparatus, Reticulum endoplasmic, lysosome, big ribosomes (80S), peroxisome, microtubules.

Table 4.1: The differences between animal and plant cell

Feature	Animal Cell	Plant Cell
Shape	Ovoid or spherical	Polygon
Plastids	Absent Present	
Centrioles	Present	Absent
Vacuole	Small or absent	Big with a tonoplast
Cell wall	Absent	Present
Microvilli	Present	Absent
Plasmodesmata	Absent	Present

### Self-assessment 4.1

- 1. What structures do both animal and plant cells have in common?
- 2. State any five principles of the cell theory.
- 3. Give the major difference between a plant and animal cell. Which organelles does this difference relate to?

## 4.2. Prokaryotic cells

### **Activities 4.2**

Under microscope, observe mounted slides of bacteria, and plant cells. Draw and label the parts that are common in both plant and bacterial specimens

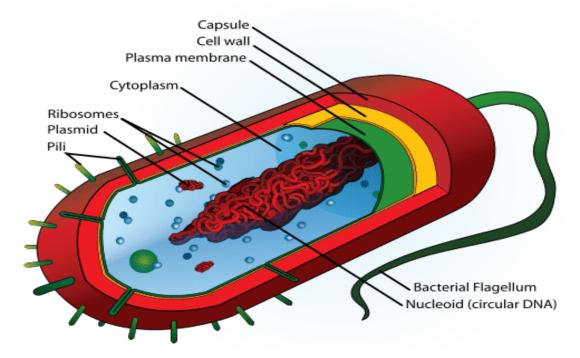


Figure 4.3: Structure of a typical prokaryotic cell. © Mariana Ruiz Villarreal, ladyofHats.

A typical bacterial cell has a cell surface membrane enclosing the cytoplasm that contains enzymes, ribosomes and food granules. The membrane is surrounded by the cell wall and this may in turn be enclosed in a capsule. A bacterial cell lacks high level of organization compared to animal or plant cell. It has no Golgi apparatus or endoplasmic reticulum. The genetic material is a single strand of DNA usually coiled up into the center of the cell to form a nucleoid. This nucleoid has no double membraned nuclear envelope so is often described as an 'ill-defined nucleus'.

- Some bacterial cells contain plasmids with additional DNA.
- Respiration generally takes place in mesosomes which is an in-folding of the cell surface membrane but lack mitochondria
- Photosynthesizing bacterial cells such as cyanobacteria (blue green algae) have a special form of chlorophyll but it is not enclosed in a double membraned chloroplast

## Comparison between prokaryotic and eukaryotic cells

Table 4.2 Comparison between prokaryotic and eukaryotic cells

Criterion	Prokaryote	Eukaryotic Plant cell	Eukaryotic Animal cell
Cell membrane	Present	Present	Present
Cell wall	Present	Present	Absent
Nuclear envelope	Absent	Present	Present
Chromosome	Circular	Threadlike	Threadlike
Mitochondria	Absent	Present	Present
Chloroplast	Absent	Present	Absent
Endoplasmic	Absent	Present	Present
reticulum			
Golgi body	Absent	Present	Present
Ribosomes	Small (70S)	Big (80S)	Big (80S)
Vacuole	Absent	Big	Absent
Lysosomes	Absent	Present	Present
Centrioles	Absent	Absent	Always present

### Self-assessment 4.2

Organisms such as bacteria are known as prokaryotes.

- 1. Which structure in a bacterial cell resembles a nucleus?
- 2. How does it differ from the nucleus of eukaryotic cells?

# 4.3. Cell organelles

### **Activities 4.3**

By using iodine solution, methylene blue, a piece of onion leaf, a scalpel, forceps, light microscope, slides and cover slips, clean cotton wool bud, and onion bulbs. Observe cells from onion epidermis under light microscope.

# Observation of a plant cell

- Add a drop of diluted iodine solution on the slide.
- Remove a transparent layer of onion epidermis from the inner side that you will mount on the slide and add iodine solution.
- Cover your preparation with a cover-slip and mount it on the stage.
- Observe the preparation under the low power and thereafter under high magnification.

Why did you use iodine solution in this experiment?

What main parts of a plant cell are easily observed from a light microscope?

# Observe animal cells from mouth cheek epithelium

- By using a clean cotton wool bud, wipe over inside of your cheek.
- Smear cells over surface of a clean grass microscope slide containing a drop of methylene blue stain
- Carefully put the cover-slip on the preparation and mount it on the stage to observe.

Draw both plant and animal cell and label the cell wall, nucleus and vacuole.

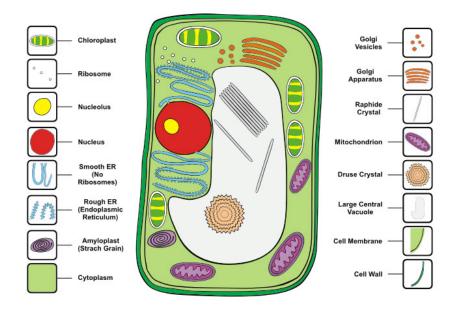


Figure 4-4: Cell organelles © E.M.Armastrong 2001

### 4.3.1. Nucleus

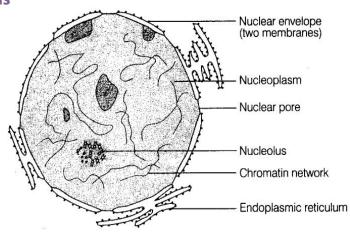


Figure 4.5 Ultra-structure of a nucleus

The cell nucleus contains nearly all the cell's DNA with the coded instructions for making proteins and other important molecules. The nucleus is surrounded by a double nuclear envelope, which allow materials to move into and out of the nucleus through nuclear pores. The granules found in the nucleus are called **chromatin** which consist of DNA bound to protein. When a cell divides, the chromatin condenses into chromosomes containing the genetic information. The nucleus contains a dense spherical structure called **nucleolus** in which assembly of ribosomes occurs.

# 4.3.2. Endoplasmic reticulum (ER)

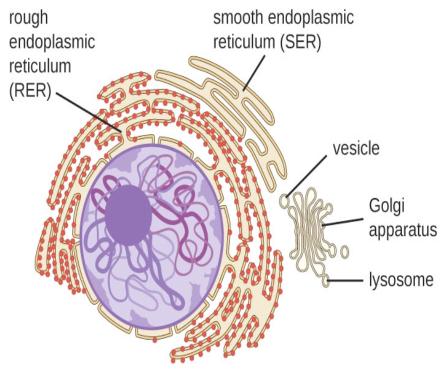


Figure 4.6. Endoplasmic reticulum. (open educational resources, Rice University)

The ER consists of a series of flattened membrane-bound sacs called **cisternae.** The rough ER is surrounded with ribosomes. The rough ER transports proteins made on attached ribosomes. The smooth ER is made of tubular cavities lacks ribosomes, and it involves in synthesis of lipids that the cell needs. The number and distribution of the ER relates to the functions of the cell; glandular cells are seen to have several RER for synthesis of hormones and enzymes. Examples include liver cells, plasma cells, and pancreatic cells.

# 4.3.3. Golgi apparatus

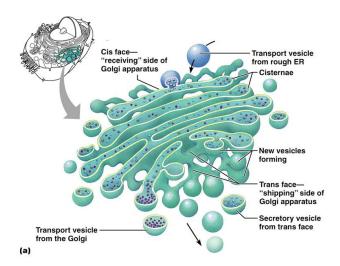


Figure 4.7. Golgi apparatus. © 2008 Pearsoneducation

The Golgi apparatus is a stack of membrane-bound, flattened sacs, which receives proteins from the ER and modifies them. It may add sugar molecules to them to form glycoproteins or lipids to form glycolipids. The Golgi apparatus then packages the modified substances into vesicles that can be transported to their final destinations throughout the cell or outside of the cell by exocytosis.

#### 4.3.4. Mitochondria

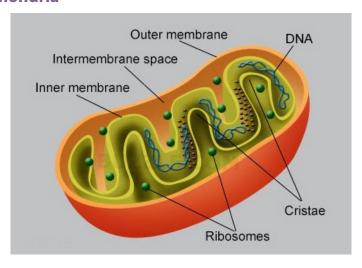


Figure 4.8: Mitochondrion. © Buzzle.com

Mitochondrion have two membranes separated by a fluid-filled intermembrane space. The inner membrane is highly folded to form **cristae** that plays a big role in aerobic respiration. The central part of the mitochondrion is called **matrix**.

The mitochondria are the site where **Adenosine triphosphate (ATP)** is produced during aerobic respiration.

# 4.3.5. Chloroplasts

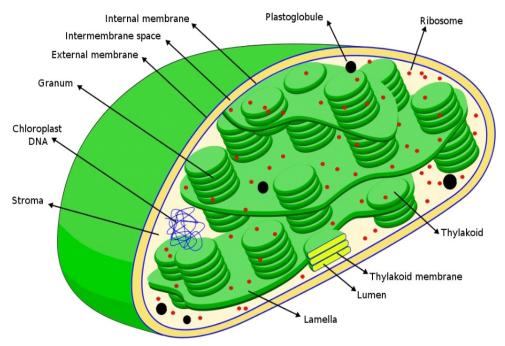


Figure 4.9: Chloroplast

Chloroplasts are the site of **photosynthesis** in plant cells. These are found in plant cells and in cells of some protoctists. They also have two membranes separated by a fluid-filled space, circular DNA as in mitochondria. The inner membrane is continuous, with **thylakoids**. A stalk of thylakoids is called a **granum (plural: grana)**. Chlorophyll molecules are present on the thylakoid membranes.

## 4.3.6. Lysosomes

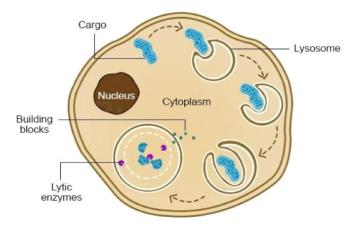


Figure 4.10: Lysosome.

These are spherical sacs surrounded by a single membrane. They contain powerful **digestive enzymes.** Their role is to break down materials such as worn out cell organelles, and destroy foreign microorganisms that enter the body. In acrosome, lysosomes help the sperm to penetrate the egg by breaking down the material surrounding the egg. Lysosomes are also involved in autolysis, breakdown of dead tissues or harmful objects inside the cell. Therefore, lysosomes are referred to as 'suicide bags'

### 4.3.7. Ribosomes

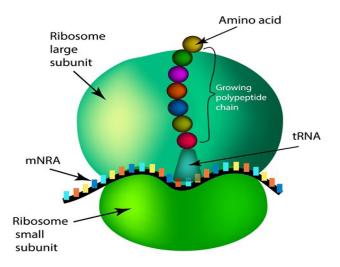


Figure 4-11. Structure of a Ribosome. © Timonina / shutterstock.com

Ribosomes appear as dark granules in the cytoplasm and are not surrounded by a membrane. They have the same size as those found attached to the rough endoplasmic reticulum- about 20nm in diameter and known 80S. Free ribosomes make proteins that are as enzymes or in other forms in the cytoplasm. Ribosomes are made in a region of the nucleus called the nucleolus.

#### 4.3.8. Centrioles

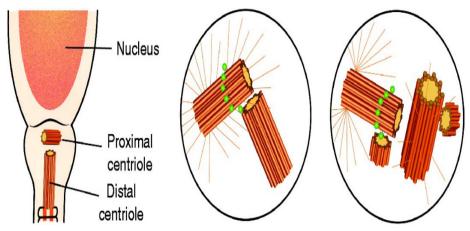


Figure 4.12: Centrioles.(Adapted from: <u>Heide Schatten</u> and <u>Qing-Yuan Sun1</u>)

Centrioles are small tubes of protein fibers called microtubules which have many roles including moving chromosomes during nuclear division. Animal cells have structures called centrioles which consist of two groups of nine triple microtubules. Centrioles form an anchor point for microtubules during cell division.

### 4.3.9. Vacuole

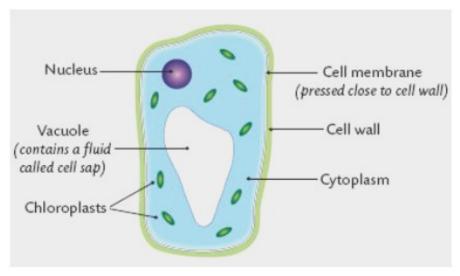


Figure 4.13: Vacuoles

A vacuole is a saclike structure that stores materials such as water, salts, proteins, and carbohydrates. In many plant cells there is a single and large central vacuole filled with liquid. The pressure in the cells of central vacuole makes it possible for plants to support heavy structures like leaves and flowers. Some animals and unicellular organisms contain **contractile vacuoles** which contract to pump excess water out of the cell.

#### Self-assessment 4.3

- 1. Explain why muscle cells contain several mitochondria compared to fat storage cells
- 2. What kind of information is contained in chromosomes?
- 3. Describe the functions of the endoplasmic reticulum, Golgi apparatus, chloroplasts, mitochondria and nucleus in the cell.
- 4. Consider the 3D structures which would be visible in ultrastructure of a plant cell.
  - a. Identify their parts and label them
  - b. State one function for each part

#### 4.4. Membrane structure

## **Activity 4.4**

Learners mix a portion of cooking vegetable oil with water and shake the mixture vigorously and leave it to settle. Note the way water and oil are distributed within the mixture and suggest a possible explanation for your observation.

Cell membranes cover surfaces of every cell. Some organelles in cytoplasm are enveloped by membranes. The cell membranes ultrastructure is not easily visible under a light microscope but is studied by electron microscopes, freeze structuring and other modern techniques which reveal complex structures

A detailed study of a cell membrane reveals that it is 7-8nm wide and is made of a phospholipid bilayer.

- Lipid component makes up 45% protein and 10% carbohydrate. Most of the lipids are phospholipids
- Each molecule of phospholipid consists of a hydrophobic tail of two fatty acids and a hydrophilic phosphate head. They arrange themselves in phospholipids bilayer with their tails pointing inward away from the water both inside and outside the cell

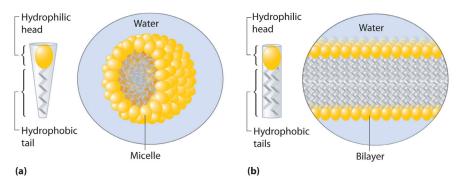


Figure 4.14. Behavior of phospholipids in water. Being amphipathic, the arrangement is determined by the way the molecules interact with water.

In 1972, Jonathan singer and Garth Nicolson proposed the fluid mosaic model of the cell membrane structure. This was done after realizing that membranes must have a complex structure to carry out a variety of activities. In their model;

- Individual protein molecules shift and move on a fluid bilayer of phospholipids;
   some spanning the width of the membrane (intrinsic proteins), others confined to the outer or inner surface (extrinsic protein)
- Protein molecules are variable in structure and function but they all contribute to the mechanical strength of membranes

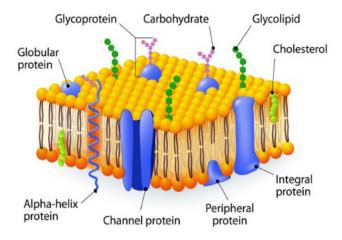


Figure 4.15 An illustration of the fluid mosaic model of the cell surface membrane

The membrane is referred to as;

- A fluid because it appears to have the properties of a fluid rather than a solid as the major constituent, lipids and proteins move about the structure
- Mosaic because protein and lipid components form a pattern of parches model

# 4.4.1. Properties of the cell membrane

- It is mainly made of lipids, proteins and carbohydrates.
- It is semi-permeable or partially permeable to allow some substances to pass

- through but prevents others to cross depending on their size, charges and polarity.
- It is positively charged outside and negatively charged inside and has a hydrophilic pole and a hydrophobic pole
- It is a bilayered sensitive and flexible. It has inorganic ions and its proteins and lipids may be mobile and contains different types of enzymes and coenzymes.
- It is perforated of pores and recognizes chemicals messengers including hormones and neurotransmitters.

# 4.4.2. Roles of different components of cell membrane

#### a. Cholesterol

- Gives the membranes of some eukaryotic cells the mechanical stability.
- It fits between fatty acid tails and helps make the barrier more complete, so substances like water molecules and ions cannot pass easily and directly through the membrane.

## b. Channel proteins

- Allow the movement of some substances across the membrane.
- Large molecules like glucose enter and leave the cell using these protein channels.

## c. Carrier proteins

- Actively move some substances across the cell membrane. For example, magnesium and other mineral ions are actively pumped into the roots hair cells from the surrounding soil.
- Nitrate ions are actively transported into xylem vessels of plants

# d. Receptor sites

- Allow hormones to bind with the cell so that a cell response can be carried out.
- Glycoproteins and glycolipids may be involved in cells signaling and they allow the immune system to recognize foreign objects to the cells.
- Some hormone receptors are glycoprotein and some are glycolipid.

# e. Enzymes and coenzymes

- Some reactions including metabolic processes in photosynthesis take place in membranes of chloroplasts.
- Some stages of respiration take place in membranes of mitochondria, where Enzymes and coenzymes may be bound to these membranes.
- The more membrane there is, the more enzymes and coenzymes it can hold and this helps to explain why mitochondrial inner membranes are folded to form cristae, and why chloroplasts contain many stacks of membranes called thylakoids.

### 4.4.3. Functions of a cell surface membrane

 The cell membrane acts as a selective barrier at the surface of the cell, and controls the exchange between the cell and its environment.

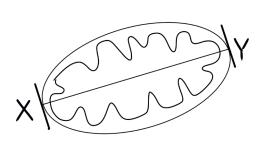
- Glycoproteins and glycolipids are involved in the cell protection, the process by which cell adhesions are brought about and in the cell recognition.
- Receptor sites for hormones and neurotransmitters
- Transmission of nerve impulses
- Insulation of nerves to improve transmission speeds.

#### Internal membranes:

- Act as reaction surfaces
- Act as an intra cellular transport system
- Providing separate intra cellular compartment, isolating different chemical reactions as in organelles.

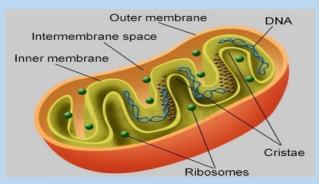
#### Self-assessment 4.4

- 1. What is meant by the fluid mosaic model of the cell membrane?
- 2. State at least three properties of the cell membrane.
- 3. Describe at least 4 types of the proteins in the cell membrane and their roles.
- 4. What is a partially permeable membrane?



- 5. What do the words hydrophilic and hydrophobic mean?
- 6. The diagram below shows the structure of a cell membrane. Study it carefully and answer the following questions.
  - a. Name parts labelled A, B, C and D and give the function of the part B.
  - b. What types of molecule are likely to be involved in?
    - i. Cell signaling and recognition
    - ii. Allowing small charged molecules to pass through the cell membrane
    - iii. Site metabolic reactions
- 7. What is the difference between rough and smooth endoplasmic reticulum?
- 8. Describe the role of cytoskeleton

9. The photograph in the figure below shows an organelle of the living cell.

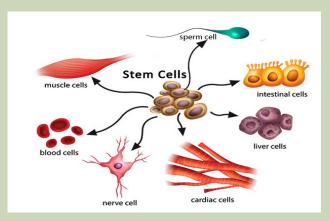


- a. Name this organelle.
- b. What is the function of this organelle?
- c. In which ways is this organelle similar to a chloroplast?

# 4.5. Specialized cells

# **Activity 4.5**

By using the diagrams below, relate the structure of specialized cells to their functions.



Differentiation refers to the changes occurring in cells of a multicellular organism so that each different type of cell becomes specialized to perform a specific function. In animals, the first type of cells in the developing embryo is stem cells. These are unspecialized cells that go on to form all the different types of cells in adult. Cell can differentiate in many ways, with changes to the shape of the cell, the number of particular organelles and the content of the cell.

# 4.5.1. Specialized animal cells and their functions

#### 4.5.1.1. Red blood cells

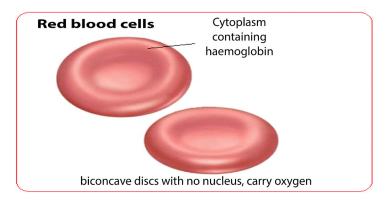


Figure 4.16: Red blood cell. https://www.quora.com/What-is-an-example-of-a-red-blood-cells-diagram

All blood cells are produced from undifferentiated stem cells in the bone marrow but the cells destined to become **erythrocytes** (red blood cells) lose their nucleus, mitochondria, Golgi apparatus and rough endoplasmic reticulum. They are packed full of the protein called haemoglobin. The shape of this cells change so that they become biconcave discs, and they are then able to transport Oxygen in the body.

### 4.5.1.2. Sperm cell

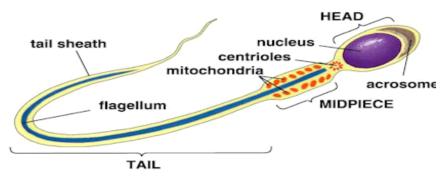


Figure 4.17: Sperm cell. https://www.pinterest.com/pin

Sperm cells are specialized to fertilize the egg. Its specialization involves many changes in shape and organelles content.

**By shape:** the sperm cells are very small, long and thin to help them to move easily, and they have a flagellum which helps them to move up the uterine tract towards the egg.

**By organelles content:** sperm cells contain numerous mitochondria which generate much energy for their movement. Their acrosome has specialized lysosomes containing many enzymes that are released on the outside of the egg. These enzymes lyse the wall of the egg, and facilitate the sperm nucleus to penetrate easily. In content, the sperm cell nucleus contains the half number of chromosomes of the germ cell in order to fulfil its role as a gamete of fertilizing the egg.

**Did you know:** As a sperm fuses with an ovum to form a zygote which grows into an individual, in the same way: a man maries a woman to form a couple which will produce children and form a family.

#### 4.5.1.3. Nerve cells

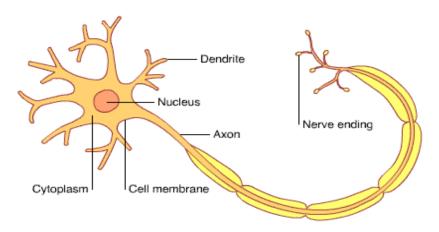


Figure 4.18: Structure of a neuron. © 2018 elephant tube .me.

Nerve cells also known as neurons are specialized cells to carry nervous impulses in the body. These signals between neurons occur via specialized connections called synapses. Specialized animal cells have different functions. Some of them are summarized in the following table.

Table 4.3: Specialized animal cells and their functions.

specialized cell	specialization	function
Erythrocytes	Lack the nucleus, mitochondria, Golgi apparatus, and rough ER. have biconcave shape possess hemoglobin	Transport of Oxygen in the body
Sperm	flagellum acrosome with enzymes half number of chromosomes	Reproduction; fuse with of the egg to for a zygote
Nerve cell	acrosome, axon, node of Ranvier, Myelin sheaths, and dendrite	Conduction of nervous information.
Epithelial cells	Having different size and shape	Protection, Secretion, absorption and movement
Smooth muscle cells	Lack striation and located in blood vessels, cavities.	Involuntary contraction

Pigment cells	contain pigment proteins which determine color, Melanin for the skin colour, iodopsin and rhodopsin in the retina	Determine the colour
Flame cells	They contain excretory structures. They are specialized excretory cells found in the simplest freshwater invertebrates, including flatworms.	Osmoregulation and excretion.
Nematocysts	Specialized stinging cells in Coelenterates for paralyzing the prey.	Their venomous coiled thread (bladder) can be projected in self-defense or to capture prey.

# 4.5.2. Specialized plant cells and their functions

# 4.5.2.1. Root hair cells

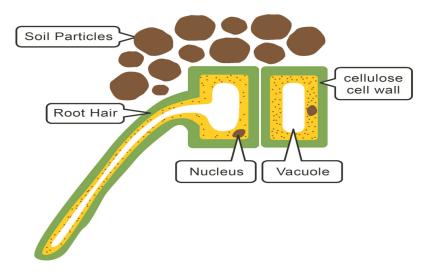


Figure 4.19: Root hair cell

**The root hair cells** have hair-like projection from their surface out into the soil. This increase the surface area of root available to absorb water and minerals from the soil.

#### 4.5.2.2. Palisade cells

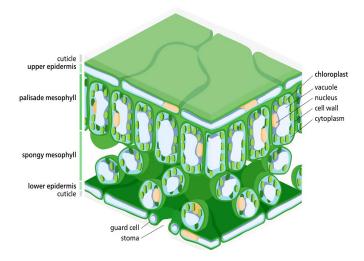


Figure 4.20: A section of the leaf showing a variety of specialized cells. Main focus is on palide and spongy mesophyll. {Adapted from: Higaki, T.; Rasmussen, H.P.; Carpenter, W.J. (1984).}.

**Palisade cells** are in leaves, right below the upper epidermis. They are vertically elongated, a different shape from the spongy mesophyll cells beneath them in the leaf. Their large numbers of chloroplasts allow them have several chloroplasts used in photosynthesis.

# Parenchyma cells

Parenchyma is composed of relatively simple and undifferentiated parenchyma cells. They function in storage, photosynthesis. In most plants, metabolic activity such as cell division, respiration, and photosynthesis occurs in these cells because they retain their active cytoplasm.

#### 4.5.2.3. Guard cells

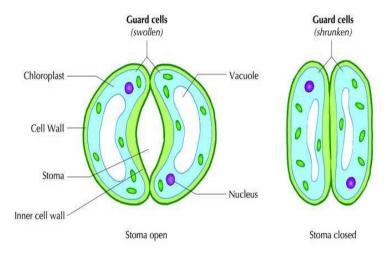


Figure 4.21: Open and closed stoma showing the shape of the guard cells.© 2003-2018 Shutterstock, Inc.

**Guard cells** are cells surrounding each stoma. Guard cells are specialized cells in the epidermis of leaves, stems and other organs that are used to control gas exchange. They are produced in pairs with a gap between them that forms a stomatapore. Guard cells have the following feature:

- Un even thick walls
- Possess chloroplasts; they are the epidermal cell that have chloroplasts an adaptive feature in controlling pore opening.

#### Self-assessment 4.5

- 1. Explain why differentiation to produce erythrocytes involves a change in shape.
- 2. Red blood cells cannot divide as they have no nucleus. State two other biological processes that red blood cells cannot carry out.
- 3. Describe how the following are specialized for their roles:
  - a. Neutrophil
  - b. Sperm cell
  - c. Root hair cell
- 4. Explain why photosynthesis is carried out in palisade mesophyll more than in spongy mesophyll.
- 5. In what kinds of organisms is cell specialization pronounced characteristic?
- 6. Discuss the advantages of cell specialization in living things

#### **End of unit assessment 4**

## **Section A. Multiple choice questions**

- 1. Which organelle converts the chemical energy in food into a form that cells can use?
  - a. Chromosome
  - b. Chloroplast
  - c. Nucleus
  - d. Mitochondrion
- 2. The cell membranes are constructed mainly of:
  - a. Carbohydrate gates
  - b. Protein pumps
  - c. Lipid bilayer
  - d. Free-moving proteins
- 3. In many cells, the structure that controls the cell's activities is the:
  - a. Nucleus
  - b. Nucleolus
  - c. Cell membrane
  - d. Organelle
- 4. Despite differences in size and shape, all cells have cytoplasm and a
  - a. Cell wall
  - b. Cell membrane
  - c. Mitochondria
  - d. Nucleus
- 5. If a cell of an organism contains a nucleus, the organism is a (an)
  - a. Plant
  - b. Eukaryote
  - c. Animal
  - d. Prokaryote

6. Match each part of the cell (left column) to corresponding statement (right column):

Nucleus controls movement of substances in and out of the cell

Mitochondrion where photosynthesis takes place

Chloroplast where aerobic respiration takes place

Smooth ER controls the activity of the cell

Ribosomes where lipids including steroids are made

## **Section B: Questions with short answers**

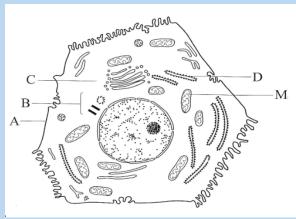
1. How does a cell membrane differ from a cell wall?

- 2. Name the structures that animal and plant cells have in common, those found in only plant cells, and those found only in animal cells.
- 3. List:
  - a. Three organelles each lacking a boundary membrane
  - b. Three organelles each bounded by a single membrane
  - c. Three organelles each bounded by two membranes (an envelope)
- 4. Identify each cell structure or organelle from its description below.
  - a. Manufactures lysosomes and ribosomes
  - b. Site of protein synthesis
  - c. Can bud off vesicles which form the Golgi body
  - d. Can transport newly synthesized protein round the cell
  - e. Manufactures ATP in animal and plant cells
  - f. Controls the activity of the cell, because it contains the DNA
  - g. Carries out photosynthesis
  - h. Can act as a starting point for the growth of spindle microtubules during cell division
  - i. Contains chromatin
  - j. Partially permeable barrier only about 7 nm thick
  - k. Organelle about 25 nm in diameter
  - I. Which two organelles other than the nucleus contain their own DNA?

# **Section C: Essay questions**

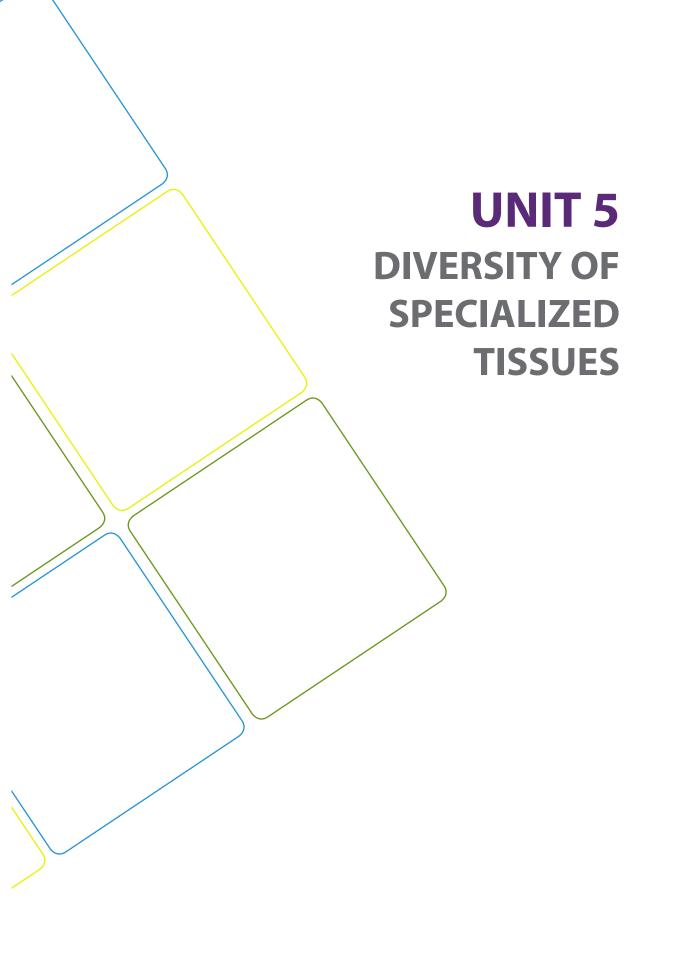
- 1. Describe the structure and function of the cell membrane and cell wall.
- 2. Describe the basic structure of the cell membrane.
- 3. Explain two common characteristics of chloroplasts and mitochondria. Consider both function and membrane structure.

4. The diagram below shows the structure of a liver cell as seen using an electron microscope.



- a. Name the parts labelled A, B, C and D.
- b. The magnification of the diagram above is x12 000. Calculate the actual length of the mitochondrion labelled M, giving your answer in  $\mu m$ . Show your working.
- c. Explain the advantage to have a division of labor between different cells in the body.





# **UNIT 5: DIVERSITY OF SPECIALIZED TISSUES**

### **Key Unit Competence**

Describe different specialized plant and animal cells and adaptation of tissues.

## Learning objectives

By the end of this unit, I should be able to:

- Define a tissue as a group of cells with similar structure working together for a function.
- Name the main types of animal and plant tissues.
- Define an organ as a structure made up of a group of tissues with related functions working together to perform bodily functions.
- Explain how epithelial tissues are adapted to perform a diversity of functions in the body.
- State the advantages and disadvantages of being unicellular.
- Observe and draw plant and animal tissues as seen under a light microscope.
- Interpret photomicrographs of plant and animal tissues
- Acknowledge the relationship between levels of organization
- Recognize the efficiency shown by multicellular organisms to explore more modes of life that are not available to single celled organisms that show little or no specialization

# **Introductory activity**

Read the following passage and use it to answer the following questions:

In an anthill, there are different groups of termites such as a queen, workers and soldiers. Each group has a specific role to play in the colony. The structure termites of each group is related to their role for example soldiers that protect the colony have mouth parts shaped like a pair of scissors building and a slightly larger abdomen for storing water. The queen is the largest of all and has a role of laying eggs. Workers have mouth parts for cutting and chewing food or soil particles. Some members of workers are in charge of caring for the young while others find food and defend the colony or remove dead members. Their specialization and division of labor bring about efficiency in the colony.

- 1. Specify the message addressed by the above paragraph.
- 2. Explain how is the structure of termites related to their functions?
- 3. What is the significance of specialized tissues in multicellular organisms like plants and animals?

The study of tissues is known as **Histology**. A **tissue** is a group of associated, similarly structured cells that perform specialized functions for the survival of the organism. In histology, **differentiation** is the process by which structures become modified and specialized to perform specific functions. Differentiation is also known as **'specialization**'. In animals, the first type of cells in the developing embryo is stem cells. These are unspecialized cells that go on to form all the different types of cells in adult.

# 5.1. Specialized plant tissues

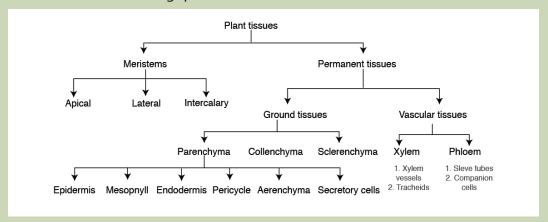
## **Activity 5.1.1**

- Remove an epidermis layer from the ventral side of an onion leaf.
- Mount it on the slide containing a drop of iodine solution
- Observe your preparation under a light microscope
- Draw, label and describe your findings.
- From your discussion:
  - 1. What is a tissue?
  - 2. What is the role of epidermis in onion?

#### 5.1.1. Plant tissues

## **Activity 5.1.2**

The following figure represents the flow chart of subdivisions of plant tissues. Use it to answer the following questions.



- 1. How do meristems differ from permanent tissues?
- 2. Plant tissues are classified into ground tissues and vascular tissues as shown in the figure above. What is meant by the term vascular tissues?
- 3. How is the structure of the xylem and phloem vessels related to their function?
- 4. From the flow diagram above, identify three types of ground tissues.

- 5. Write down short notes on each of the following types of meristems.
  - a. Apical meristems.
  - b. Lateral meristems
  - c. Intercalary meristems

Plant tissues can be divided into two main groups, Meristematic tissues (apical, lateral, and intercal arymeristems) and Permanent tissues (ground tissues and vascular tissues).

#### 5.1.2. Meristem tissues

**Meristem tissue** is a group of cells which retain the ability to divide by mitosis. Meristematic tissues are specialized to carry out specific functions such as reproduction, growth, photosynthesis and replacement of old or damage tissues. The cells making a meristem tissue are small, have a central large nucleus and dense cytoplasm, thin-walled, with no or small vacuole, and no specialized features. The cells are rectangular and closely packed with no intercellular air spaces.

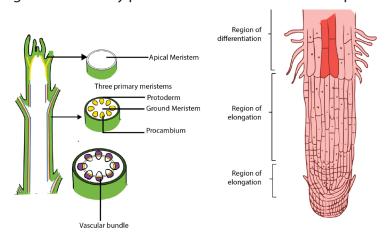


Figure 5.1: Structure of meristematic cells

# Types of meristematic tissues

Meristematic tissues are subdivided into apical meristems, lateral meristems (cambium) and intercalary meristems

## a. Apical meristems

They are located in the root and shoot apex (at the growing points of roots and stems). They are responsible for primary growth, leading to the increase of primary plant body.

### b. Lateral Meristems (cambium)

Lateral meristems are in lateral parts of the plant, where they are responsible for

secondary growth. The cambium gives rise to secondary vascular tissues (secondary xylem and secondary phloem) in dicotyledonous plants.

# c. Intercalary meristems

These are found in the region of permanent tissues like at nodes of monocotyledonous plants (e.g. sugar cane). It allows growth in length to occur between internodes.

#### **Functions of meristematic tissues**

- The main function of meristematic tissue is to produce new cells by mitosis.
   The cells elongate and differentiate to form new cells for primary growth of shoot and root.
- Vascular cambium produces new cells to increase the diameter of stems and roots during secondary growth.
- Cork cambium called (phellogen) produces the outer cork layer called phellem which consists of suberized cells. The cork layer reduces water evaporation from the plant and protects the plant against the entry of pathogens.
- The intercalary meristems allow growth and increase in length in regions other than the tips.

### 5.1.3. Permanent tissues

Permanent tissues consist of two groups of tissues such as: ground and vascular tissues.

#### 5.1.4. Ground tissues

The ground or fundamental tissues are plant tissues which function in storage, metabolism and support. There are three types of ground tissues: **parenchyma**, **collenchyma** and **sclerenchyma** tissues.

# 5.1.5. Parenchyma tissues

Parenchyma is a soft plant tissue made up of thin-walled cells that forms the greater part of leaves, stem pith, roots, and fruit pulp. They are the main sites for physiological and biochemical processes in the plants including photosynthesis, protein synthesis and storage of starch and mineral ions. Parenchyma tissues can be found in epidermis, mesophyll, endodermis, pericycle, aerenchyma and secretory cells.

#### Characteristics

- Parenchyma tissues consist of large living cells, with relatively thin wall containing cellulose, pectin and hemicellulose.
- Parenchyma tissues consist of cells, usually having a large central vacuole.
   They are often partially separated from each other.
- Spongy cells present intercellular spaces that intervene in gaseous exchange and transpiration through stoma. They are usually stuffed with plastids.
- Parenchyma tissues consist of cells with polygonal and spherical shapes in

the leaf. They form the mesophyll, and are located between upper and lower epidermises. They are responsible for photosynthesis.

## **Functions of parenchyma tissues**

- In the leaves, parenchyma tissues form the mesophyll and are sites for photosynthesis, gaseous exchange and transpiration.
- They store food substances such as starch, proteins and lipids
- They can be modified to form specialized cells to carry out other function in epidermis, endodermis, pericycle, parenchyma, and secretory cells.

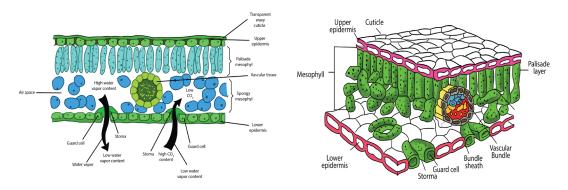


Figure 5.2: Diagram showing parenchyma tissue in the leaf

## Adaptations of parenchyma for its function

- Parenchyma tissues are made of unspecialized cells with variety of functions:
- Parenchyma can become specialized to carry out specific functions e.g. mesophyll has cells with many chloroplasts, and aerenchyma which has air spaces. All of these adaptations help in photosynthesis and gas exchange.
- They have isodiametric cells and function as packing tissue and storage tissue.
- Cells are loosely packed with many large intercellular spaces. This permits diffusion of gases.
- They have thin cellulose cell wall which is permeable so that it permits passage of materials.
- The walls are transparent and permit entry of light in photosynthesis cells.
- Large cells with large vacuoles provides space for storage of substances, where the entry of water causes vacuole to expand and cells become turgid
- Leucoplasts act as storage of starch while chromoplasts present in some cells
   e.g. in petals attract insects for pollination.

# 5.1.6. Collenchyma tissues

Their cells are elongated with irregularly thickened cell walls that provide structural support, particularly in growing shoots and leaves. Their thick cell walls are composed of cellulose and pectin. These cells are often found under the epidermis, or the outer layer of cells in young stems and in leaf veins.

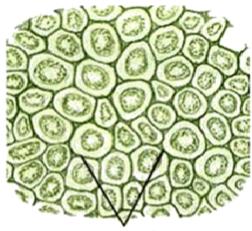


Figure 5.3 Illustration of collenchyma

# 5.1.7. Sclerenchyma tissues

Sclerenchyma is found in hard parts of the plant body. They are very common in roots, stems, leaves and petioles. They may be present in patches, groups or layers. The cells of the sclerenchyma are dead, they are elongated, narrow, and thick walled and lignified. They are pointed at both ends where it gives strength, rigidity and flexibility to the plant body. They consist of fibres and sclereids. Fibres are long, narrow, thick and liquefied cells usually tapering at both ends. Sclereids cells are normally short with very thick walls, irregular and not tapering at the ends.

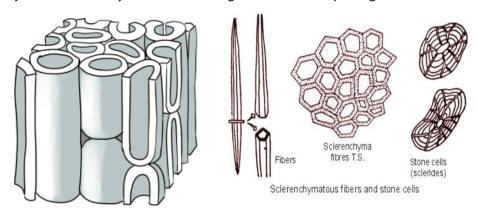


Figure 5.4. Sclerenchyma tissues structures

Table 5.1: Comparison between collenchyma and sclerenchyma tissues

Collenchyma	Sclerenchyma	
Made of living cells	Made of dead cells	
Tapering ends do not overlap	Tapering ends overlap and interlock	
Ensure mechanical support and flexibility	Ensure mechanical support only	
Cell wall is not lignified.	Cell wall is lignified.	

#### 5.1.8. Vascular tissues

The vascular tissue system consists of two kinds of conducting tissues: **the xylem** responsible for conduction of water and dissolved mineral nutrients, and the **phloem** responsible for conduction of elaborated food.

## a. Xylem

The xylem tissues are made of **dead cells** which have the cell walls removed at the end of the cells, forming tubes through which the water and dissolved mineral ions can flow. Xylem vessels are involved in the movement of water through a plant from its roots to its leaves via the stem. During this process water is absorbed from the soil through root hair cells, moves by osmosis from root cell to root cell until it reaches the xylem, and finally it is transported through the xylem vessels up the stem and then to the leaves.

Xylem vessels are hollow tubes or lumen with a thick strengthened cellulose cell wall. The hollow tubes act like pipes allowing water and dissolved minerals to flow through them. They develop from cylindrical cells arranged end to end, in which the cytoplasm dies and the cell walls between adjoining cells breaks down leaving a dead empty tube. The cell walls in xylem vessels contain a substance called lignin which strengthens the cells and gives structural support.

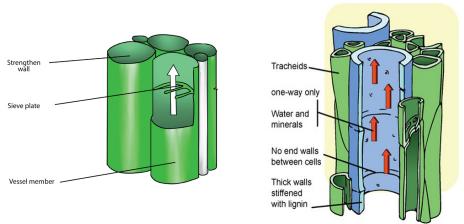


Figure 5.5: Xylem vessel

#### b. Phloem

Phloem vessels are involved in translocation of elaborated substances. Dissolved sugars, produced during photosynthesis, and other soluble food molecules are moved from the leaves to growing tissues such as the tips of the roots and shoots and storage tissues such as in the roots. In contrast to xylem, phloem consists of columns of living cells. The cell walls of these cells do not completely break down, but instead form small holes at the ends of the cell. The ends of the cell are referred to as **sieve plates**. The connection of phloem cells effectively forms a tube which allows dissolved sugars to be transported.

Phloem tubes carry food substances like sugar and amino acids produced in leaves during photosynthesis to every part of the plant. The movement of food substances through the plant is called **translocation**.

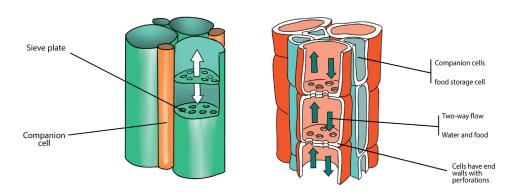


Figure 5.6: Phloem tube cells showing sieve plates that are reinforced cell walls between. Modified from: https://dr282zn36sxxg.cloudfront.net/datastreams/f-

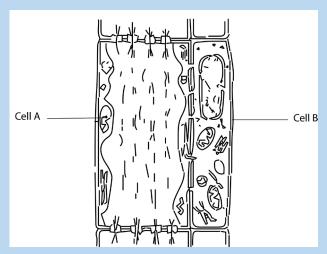
Table 5.2: Comparison between Xylem and Phloem tissues

	Xylem	Phloem
Diagrams	Serengthen with with the series of the serie	Sieve plate  Companion cell
Transport	Water and mineral from the roots to the shoots and leaves	Sugar and amino acids produced in leaves during photosynthesis to every part of the plant.
Process	Transpiration	Translocation
Structure	Cylindrical cells arranged end to end, in which the cytoplasm dies and the cell walls between adjoining cells breaks down leaving a dead empty tube with strengthened cell walls.	Phloem tubes are made up of columns of living cylindrical cells. The cell walls between adjoining cells develop holes like a sieve allowing transport through the tube.
Components	Dead cells and Fibers	Living cells and companion cells
Direction of flow	Upwards	Up and downwards
Permeability	Impermeable	Permeable
Cytoplasm	None	Cytoplasm lining

Cell wall thickness	Thick	Thin
Cell wall materials	Formed by Lignin	Formed by Cellulose

### Self-assessment 5.1

- 1. State where in a flowering plant you would find:
  - a. Lateral meristem
  - b. Intercalary meristem
  - c. Apical meristem
- 2. Give characteristics of meristematic cells.
- 3. What do you understand by the following terms?
  - a. Differentiation
  - b. Cambium
  - c. Wood
  - d. Meristem
- 4. Differentiate between Collenchyma and sclerenchyma
- 5. State the main structures (components) that make up a xylem and phloem tissues.
- 6. Explain how the structure of Parenchyma and Xylem tissues are suitable to their functions.
- 7. The diagram below shows a longitudinal section of two cells of phloem tissue in a plant stem.



- a. Name the cells labelled A and B on the diagram.
- b. State the function of phloem in a plant.

### 5.2. Animal tissues

## **Activity 5.2**

Conduct a research by using different sources of information to find out the structures and the main functions of the following four groups of animal tissues: epithelial, connective, muscular and nervous tissues.

There are four basic types of animal tissues such as epithelial **tissue**, **muscle tissue**, **nervous tissue**, **and connective tissue**.

# 5.2.1. Epithelial tissue

Epithelial tissue consists of closely packed cells arranged in single or multilayered sheets. It is made up of layers of tightly packed cells that form the external surfaces of the body and cover the outer and the inner surfaces of the organs. Some are specialized to form glandular tissues (glands). The epithelium lining the inside of the heart, blood vessels and lymph vessels is referred to as **endothelium**. Two criteria for classifying epithelia are: **the number of cell layers and the shape of cells on the free surface.** The following are the types of epithelium tissues:

## a. Simple cuboidal epithelium

This is a tissue with cells that are cubical in shape. Cuboidal cells are specialized for secretion and they make up the epithelia of kidney tubules and many glands including salivary glands, and thyroid gland.



Figure 5.7 a) Simple cuboidal epithelium.

# b. Simple squamous epithelium

It is thin, leaky and functions in the exchange of material by diffusion. This type of epithelium lines blood vessels and the air sacs of lungs, where diffusion of nutrients and gases is critical.



Figure 5.8 b) Simple squamous epithelium.

### c. Simple columnar epithelium

These are columnar in shape with free surface containing extensions of micro villi. It lines the intestines. This epithelium secretes digestive juices for the final stages of digestion and absorbs nutrients to blood stream.

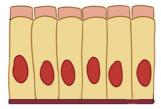


Figure 5.9 c): Simple columnar epithelium.

# d. Pseudo-stratified ciliated columnar epithelium

It forms a mucous membrane that lines the nasal passages of many vertebrates. The beating cilia move the film of mucus along the surface.

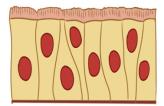


Figure 5.10 d): Pseudo-stratified ciliated columnar epithelium.

# e. Stratified squamous epithelium

It regenerates rapidly by cell division near the basal lamina. The new cells are pushed outward to replace cells that are sloughed off. This epithelium is commonly found on surfaces subject to abrasion, such as the outer skin and lining of the esophagus, anus, and vagina.

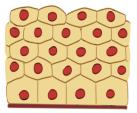


Figure 5.11 e) Stratified squamous epithelium.

# f. Transitional epithelium

In this type of stratified epithelium, the surface cells change their shape from round to squamous. Transitional epithelium lines urinary bladder. When the bladder is empty, the surface cells are rounded. As the bladder fills urine, these cells become flattened. Transitional epithelium enables the bladder to fill and stretch without tearing the lining.

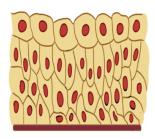


Figure 5.12 f) Transitional epithelium.

## g. Stratified columnar epithelium

It is a rare type of the epithelial tissue composed of column shaped cells arranged in multiple layers. They are found in the conjunctiva or the eye, in parts of the pharynx, anus, uterus, the male urethra and vas deferens.

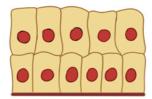
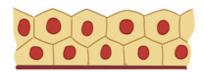


Figure 5.13 g) Stratified columnar epithelium.

## h. Stratified cuboidal epithelium

It is a type of epithelial tissue composed of multiple layers of cube-shaped cells. Only the most superficial layer is made up of cuboidal cells and the other layers can be cells of other type. It has several locations in the body including sweat gland ducts, egg-producing vesicles and ovaries.



*Figure 5.14 h): Stratified cuboidale pithelium.* 

# **5.2.2.** Main characteristics of epithelial tissues

# a. Polarity

All epithelia have a **free surface** and a lower attached **basal surface** that differ in structure and function. For this reason, epithelium is described as showing polarity.

# b. Supported by connective tissue

All epithelia are supported by connective tissue. For instance, deep to the basal lamina is **reticular lamina**, an extracellular material containing collagen protein fiber which forms the basement membrane. The basement membrane reinforces the epithelium and helps it to resist stretching and tearing.

**c.** They are avascular; epithelia have no blood vessel in them. Nutrients and gases are supplied by blood through the connective tissue by simple diffusion

# d. Regeneration

Epithelium have a high regenerative capacity and can reproduce rapidly as long as they receive adequate nutrition.

## **Functions of epithelium**

- Epithelium forms a protective layer: The epithelium of the skin protects the body from mechanical damage, entry of pathogens, ultraviolet rays and dehydration. Epithelium lining the respiratory air passages secretes mucus which traps inhaled dust particles and microbes.
- The ciliated epithelium cells have cilia that propel the mucus and trapped particles to the throat.
- Glandular tissues secrete the digestive enzymes, hormones, mucus, sweat and sebum.
- Acts as a barrier and regulates movement of substances across kidney
- Some epithelial cells can divide mitotically producing new cells to replace damaged or dead cells.
- Some epithelial cells such as taste buds and retina cells are specialized to form sensory receptors.

### 5.2.3. Muscular tissues

Muscle tissues consist of elongated cells held together by connective tissue. Muscle cells are highly specialized in that they are able to shorten to a half or even a third of their resting length by the process of contraction. The contraction is caused by two types of fibrous proteins: **myosin** and **actin**.

Muscles in the body provide the necessary force for the motion and they convert chemical energy into kinetic or mechanical energy. There are three types of muscle tissue:

- Smooth muscle which is found in the inner linings of organs;
- Skeletal or striated muscle, which is attached to bone and helps in movement of the body;
- Cardiac muscle which is found only in the heart.

Smooth and cardiac muscles are involuntary muscles whereas skeletal muscles are called voluntary muscles because they are under voluntary (conscious) control.

#### a. Smooth Muscle

**Smooth muscle** is also called unstriated, unstriped, involuntary or visceral muscle. It is found in the walls of the hollow internal organs such as blood vessels, intestinal tract, urinary bladder, and uterus. Smooth muscles have the following features;

- It is under control of the autonomic nervous system; they cannot be controlled consciously, so they are also called involuntarily muscle. They do not have striations.
- Smooth muscle cells contract slowly and rhythmically

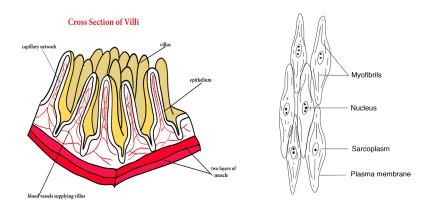


Figure 5.15: Smooth muscle cells.

### b. Cardiac tissue

**Cardiac tissue** (figure 5.10 a) is found in the walls of the heart and it is under control of the autonomic nervous system. Cardiac muscle has the flowing basic features.

- It contracts and relaxes continuously.
- It is branched and connected to other cardiac muscle fibers through intercalateddiscs(Figure 5.16 b), which are reinforced membranes that hold the cells together during contractions. These interconnections or intercalated discs between the fibers ensure a rapid and uniform spread of excitation throughout the wall of the heart which in turn ensures a synchronous contraction.
- They are myogenic (their contraction originate from within the heart itself).

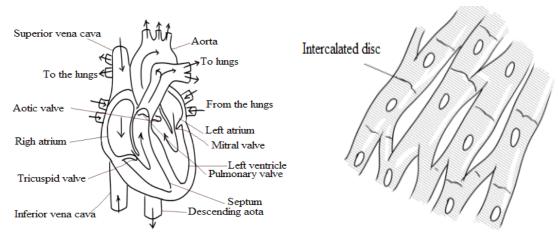


Figure 5.16: Cardiac muscle tissue(a) and intercalated disk (b)

### c. Skeletal Muscle

Skeletal muscle is also called striated, striped, or voluntary. They are attached to bone, and are responsible for body movements and body posture. There are approximately 639 skeletal muscles in the human body.

#### Characteristics of skeletal muscles:

- They are under control of voluntary nervous system
- They are attached to bone and this is the reason why they are called skeletal muscles.
- They are made of elongated and cylindrical muscle fibres
- They appear under microscope to have alternate light and dark bonds and this
  is why they are called striated muscles.
- Their muscle fibres are multinucleated (many nuclei per cell)
- These muscle cells also contain light and dark stripes called striations

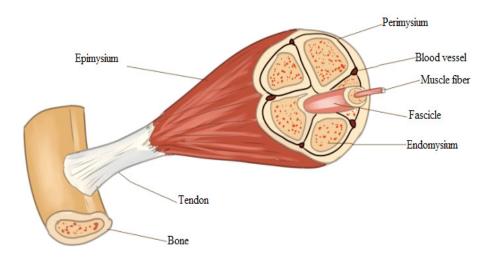


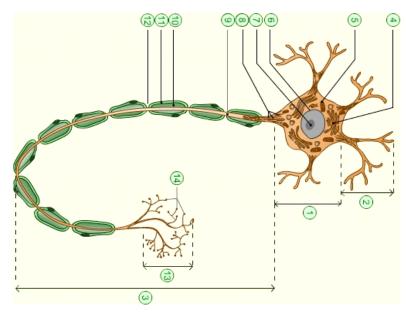
Figure 5.17: Skeletal muscle structure.

### **General functions of muscle**

The main function of muscle is its contribution to motion, where body movements such as walking, breathing, and speaking, as well as movements associated with digestion and the flow of fluids take place. Muscles contribute to the heat production, maintenance of posture and body support and communication through facial expression, writing and speech.

### 5.2.4. Nervous tissue

Nervous tissue is composed principally of densely packed cells called the nerve cells (neurons) that together form the nervous system including the brain and spinal cord. Neurons are specialized for transmitting electrical nerve impulses.



- 1. Cell body or soma,
- 2. Dendrites,
- 3. Axon or fibre,
- 4. Endoplasmic Reticulum,
- 5. Mitochondrion,
- 6. Nuclear membrane,
- 7. Nucleus,
- 8, 9 and 12. Node of Ranvier,
- 10. Cylindraxa / axoplasm,
- 11. Myelin sheath,
- 13. Terminal branches,
- 14. Synaptic knobs.

Figure 5.18: Typical neuron structure

A typical neuron has three main parts: Cell body, dentrites and axon.

## a. The cell body or soma

- It is the main part from which, extensions derive (Axon and Dendron).
- It is made of a great spherical nucleus, granular cytoplasm and controls all nerve cell activities.
- **b. Dendrites (Dendron when single):** small branches attached to the cell body and receive nerve impulse from other neurons

## c. Axon or cylindrax:

- It is the thinner nerve fibre that carries messages away from the cell body and can be as long as 1 m. In some neurones, the axons have a fatty myelin sheath formed by Schwann cells which wrap themselves around the axon to increase the speed of impulse transmission.

#### 5.2.5. Connective tissues

**Connective tissue** is made up of many different types of cells that are all involved in structure and support of the body. **Bone, blood, fat,** and **cartilage** are all connective tissues. Connective tissues can be densely packed together, as bone cells are or loosely packed, as adipose tissue (fat cells) are. A connective tissue is made up of a variety of cells embedded in a large amount of intracellular substance called matrix and fibers which are non-living products of the cells.

## a. Common functions of connecting tissues:

- Connective tissues protect and support the body and internal organs.
- They act as connecting systems, binding all other tissues together.
- They also form surrounding sheaths to separate the various organs.

#### b. Cells of connective tissue

The specialized cells of the various connective tissues produce the extracellular matrix. The names of the cells end with suffixes that identify the cell functions as blasts, cytes, or clasts. Blasts create the matrix, cytes maintain it, and clasts break it down for remodeling. For example: **Fibroblasts** are cells that form fibrous connective tissue and **fibrocytes** maintain it, **chondroblasts** form cartilage and **chondrocytes** maintain it, and osteoblasts form bone, osteocytes maintain it, and osteoclasts break it down

Adipose, or fat cells, also called Adipocytes, contain large amounts of lipid. The lipid pushes the rest of the cell contents to the periphery, so that each cell appears to contain a large and centrally located lipid droplet with a thin layer of cytoplasm around it. Adipose cells are rare in some connective tissue types like cartilage but they are abundant in others like loose connective tissue, and they are predominant in adipose tissue.

Mast cells are commonly found beneath membranes in loose connective tissue and along small blood vessels of organs. They contain chemicals such as heparin, histamine and proteolytic enzymes. These substances are released in response to injury such as trauma and infection and play important roles in inflammation.

White blood cells continuously move from blood vessels into connective tissues. The rate of movement increases dramatically in response to injury or infection. In addition, accumulations of lymphocytes, a type of white blood cell, are common in some connective tissues, such as in the connective tissue beneath the epithelial lining of certain parts of the digestive system.

**Macrophages** are found in some connective tissue types. They are derived from monocytes, a white blood cell type. Macrophages are either **fixed** and do not move through the connective tissue in which they are found or are wandering macrophages and move by amoeboid movement through the connective tissue. Macrophages phagocyte foreign or injured cells, and they play a major role in providing protection against infections.

Note that there are three structural major components of the extracellular matrix of connective tissue such as fluid, ground substance consisting of non-fibrous protein and other molecules and protein fibers. The structure of the matrix gives connective tissue types most of their functional characteristics, such as the ability of bones and cartilage to bear weight, tendons and ligaments to withstand tension, and dermis of the skin to withstand punctures, abrasions, and other abuses.

#### c. Fiber connective tissues

Another type of connective tissues consists of fibers. Fibers are of different types including:

– Connective tissue fibers: which are made of protein and are of three kinds: collagenous, elastic and reticular fibers.

- Collagenous fibers: These provide strength combined with flexibility. They are made up of collagen, probably the most abundant protein in the animal kingdom.
- Elastic fibers: These are easily stretched but are also resilient, snapping back to their original length when tension is released. Shaped as long threads, elastic fibers are made of a protein called elastin.
- **Reticular fibers:** These are thin collagen fibers coated with glycoprotein. They are very short, thin fibers that branch to form a network and appear different microscopically from other collagen fibers. Reticular fibers are not as strong as most collagen fibers, but networks of reticular fibers fill space between tissues and organs.

#### d. Loose connective tissue

This is also called areola connective tissue and is the most widespread connective tissue in all animal tissues. It binds epithelial tissues to underlying tissues and functions as packing material, holding organs in place. Loose Connective tissue has the following main components;

- Fibers: collagen, elastic and reticular.
- Cells; fibroblasts and macrophages. Fibroblasts secrete the protein ingredients of the extracellular fibers. Macrophages are cells that roam the maze of fibers, engulfing both foreign particles and the debris of dead cells by phagocytosis.

### e. Fibrous connective tissue

Fibrous Connective tissue is dense with collagenous fibers. The fibers form parallel bundles, which maximize non-elastic strength. Fibrous Connective tissue is found in tendons, which attach muscles to bones, and ligaments, which connect bones at joint.

## f. Adipose tissue

A dipose tissue is a specialized form of loose connective tissue that stores fats in a diposecells distributed throughout its matrix. Adipose tissue consists of adipocytes, or fat cells, which contain large amounts of lipid. Unlike other connective tissue types, adipose tissue is composed of large cells and a small amount of extracellular matrix that consists of loosely arranged collagen and reticular fibers with some scattered elastic fibers. Blood vessels form a network in the extracellular matrix. The fat cells are usually arranged in clusters or lobules separated from one another by loose connective tissue. Adipose tissue functions as:

- An insulator against heat loss
- A protective tissue to delicate internal organs
- A site of energy storage in the form of fat.

# g. Bone and Cartilage tissue

Cartilage has an abundance of collagenous fibers embedded in a rubbery matrix made of a protein-carbohydrate complex called **chondroitin sulfate**. Cartilage is composed of specialized cells, called **chondrocytes**, surrounded by a gelatinous matrix of collagen, a tough protein. The cartilage surface is covered by a membrane known as the **perichondrium**. There are three types of cartilage (hyaline cartilage, yellow elastic and white fibrous cartilage.)

- Hyaline cartilage is semi-transparent and is often stained light blue or pink in tissue sections. It is extremely very strong but very flexible and elastic. Hyaline cartilage occurs in the trachea, larynx, tip of the nose, connection between the ribs and the breastbone; and at the ends of bones where they form joints. It also forms much of the fetal skeleton.
- Elastic cartilage is similar to hyaline cartilage, but in addition to the collagenous fibers. The matrix of the elastic cartilage also contains an abundant network of branched elastic fibers. This type of cartilage is found in the lobe of the ears, the epiglottis and in the parts of the larynx. They provide flexibility and elastic support.
- Fibro-cartilage(White fibrous cartilage) is an extremely tough tissue. It is found as discs between the vertebrae, bones, anterior joint between the two halves of pelvic girdle and at points where tendons inserted on bones near hyaline cartilage. It resists compression and absorbs shock in some joints.

#### **Bone tissue**

This is a firmer and denser material that has the following features:

- Hard and compact
- Has many collagen fibres
- Its matrix has inorganic salts such is calcium carbonate and calcium phosphate
- Has few cells located in the lacunae in the matrix
- Has osteoblasts as mature and non-dividing cells
- Have a harversian canal
- Consists of irregular cylinder with layer of matrix call lamellae

The following are the main functions of bone tissue:

- Structural support of the body
- Protection of internal organs, heart and lungs.
- Attachment of the muscles to effect movement.
- Production of blood cells

#### h. Blood tissue

Blood is a flowing made up of particles suspended in a fluid composed of fluid called plasma, and several kinds of cells. Within the blood plasma, there are erythrocytes (red blood cells), leukocytes (white blood cells), thrombocytes (platelets) and other substances. Blood performs the following important functions:

## **Transport**

 Blood transports absorbed substances such as glucose, amino acids, mineral ions and vitamins from the small intestine.

- Blood transports the respiratory gases (Oxygen and Carbon dioxide).
- Blood transports the excretory wastes such as urea, uric acid to excretory organs to be removed out of the body.
- Blood transports hormones e.g. insulin from pancreas to the liver where it is stored.

### **Homeostasis**

Na<sup>+</sup> affects the water potential of the blood and regulates the diffusion of water between blood and tissues. Hydrogen carbonates help to maintain the pH of the blood.

#### **Protection**

- Leucocytes such as neutrophils and macrophages engulf pathogens e.g. **bacteria**
- B-lymphocytes produce antibodies to destroy pathogens or to neutralize
- T-lymphocytes destroy infected cells.
- Platelets, fibringen and prothrombin play an important role in blood clotting to reduce blood loss and the entry of pathogens.

#### Self assessment 5.2

You are provided with photomicrographs or slides of different plant and animal tissue. Study them carefully and answer questions that follow.

Identify the different tissues provided and where they are located.

One of the images is a blood smear. Draw a well labeled diagram of this tissue

# 5.3. Levels of organization: cell, tissue, organ and system

# **Activity 5.3**

Visit a classroom block, administration block or any building in school which is constructed with bricks and use it to answer the following questions.

- 1. What is the smallest unit or component of the classroom block?
- 2. How are bricks arranged?
- 3. Do you think the brick has other smaller particles in it?
- 4. How many bricks does a classroom block have?
- 5. How are walls, classrooms, washrooms and other apartments of the block formed?
- 6. Arrange the following in their ascending order of size (from the smallest to the largest); whole block, wall, a brick, a room, course (a line of bricks).
- 7. Relate the above arrangement of a building to levels of organization in multicellular organisms, beginning with a cell and ending with an organism

The human body is organized into structural and functional levels of increasing complexity. Each higher level incorporates the structures and functions of the previous level. The simplest is the cells, organized into tissues, organs, and organ systems. All of the levels of organization of the human body are represented in the following figure.

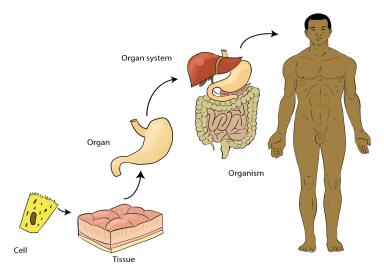


Figure 5.19: Levels of organization of the human body

### 5.3.1. Cells

The smallest structural and functional living units of living things are **cells**. There are many different types of human cells, though they all have certain similarities. Each type of cell is made of chemicals and carries out specific chemical reactions.

### **5.3.2. Tissues**

A **tissue** is a group of cells with similar structure and function. There are four groups of tissues (**Epithelial tissues**, **Connective tissues**, **Muscle tissues**, **Nerve tissue**)

# 5.3.3. Organs

An **organ** is a group of tissues precisely arranged so as to accomplish specific functions. Examples of organs are the kidneys, individual bones, the liver, lungs, and stomach. The kidneys contain several kinds of epithelial or surface tissues, for their work of absorption. The stomach is lined with epithelial tissue that secretes gastric juice for digestion. Smooth muscle tissue in the wall of the stomach contracts to mix food with gastric juice and propel it to the small intestine. Nerve tissue carries impulses that increase or decrease the contractions of the stomach.

# 5.3.4. Organ systems

An **organ system** is a group of organs that all contribute to a particular function. Examples are the urinary system, digestive system, and respiratory system. For example, the urinary system consists of the kidneys, ureters, urinary bladder, and

urethra. These organs all contribute to the formation and elimination of urine. The Human body has 11 organ systems: circulatory, digestive, endocrine, and excretory (urinary), the lymphatic, integumentary, muscular, nervous, reproductive, respiratory, and skeletal systems.

Table 5.3: Major organ systems of the human body

Systems	Functions	Organs, Tissues, and Structures Involved
Cardiovascular	Transporting nutrients, oxygen, and other substances to the body cells, and carbon dioxide, wastes, and other substance away from cells	Heart, blood, blood vessels
Lymphatic	Defense against infection and disease, transfer of lymph between tissues and the blood stream	Lymph, lymph nodes, lymph vessels, tonsils, adenoids, thymus, and spleen
Digestive	Food transformation and absorption of nutrients, minerals, vitamins, and water	Salivary glands, oesophagus, stomach, liver, gallbladder, pancreas, small intestine, large intestine
Endocrine	Communication within the body with hormones; directing long-term change over other organ systems to maintain homeostasis	Among many, the pituitary gland, pineal gland, thyroid, parathyroid gland, adrenal glands, testes, and ovaries
Nervous	Receiving, transferring and processing information; directing short-term change over other organ systems inorder to maintain homeostasis	Brain, spinal cord, nerves, and sense organs (eyes, ears, tongue, skin, nose)
Reproductive	Production of sex cells (gametes) and sex hormones; production of offspring	Fallopian tubes, uterus, vagina, ovaries, testes, vas deferens, seminal vesicles, prostate, and penis
Respiratory	Release of air to sites where gas exchange can occur between blood and air (lungs)	Mouth, nose, pharynx, larynx, trachea, bronchi, lungs, and diaphragm
Urinary	Control of pH, removal of excess water, salts, and waste products from blood and body.	Kidneys, ureter, urinary bladder, and urethra
Skeletal Support and protection of soft tissues of body; mineral storage, production of blood cells; movement at joints		Bones, cartilage, ligaments

### Self-assessment 5.3

- 1. Answer by true or false
  - a. Organic chemicals are often very complex and always contain the element carbon only.
  - b. A tissue is a group of cells with similar structure and function.
  - c. Integumentary organ system plays the role in protection of the human body from injury and fluid loss.
  - d. An organ system is a group of organs that all contribute to a particular function.
- 2. Explain why the cell as level of organization of human body is said to be:
  - a. Basic unit of human body
  - b. Structural unit of human body
  - c. Functional unit of human body

# 5.4. Advantages and disadvantages of being Unicellular or Multicellular

### Activity 5.4

Discuss the advantages and disadvantages of an organism being unicellular or Multicellular

# 5.4.1. Advantages of unicellular organisms

- Unicellular organisms need fewer nutrients and can survive in unfavorable conditions.
- Some of the organisms can generate energy through photosynthesis.
- Sometimes different bacteria work together to work to their advantages.
- Unicellular organisms can multiply quickly and have less energy/resource demands.

# 5.4.2. Disadvantages of unicellular organisms

Unicellular organisms only have one cell that is used to function their entire being. This is a disadvantage compared to multicellular organisms, which have many cells and function more easily and properly.

# 5.4.3. Advantages of a multicellular state of an organism

- Multicellular organism usually has a wider range of functions because of the aggregation of different types of cells.
- Multicellular organisms have many more necessities and can only survive in certain conditions.
- Multicellular organisms such as animals are unable to make their own food so they survive by eating living things such as vegetables, fruits, and meat. They

can also eat things that are produced by other living things such as eggs, milk, and honey.

#### Self-assessment 5.4

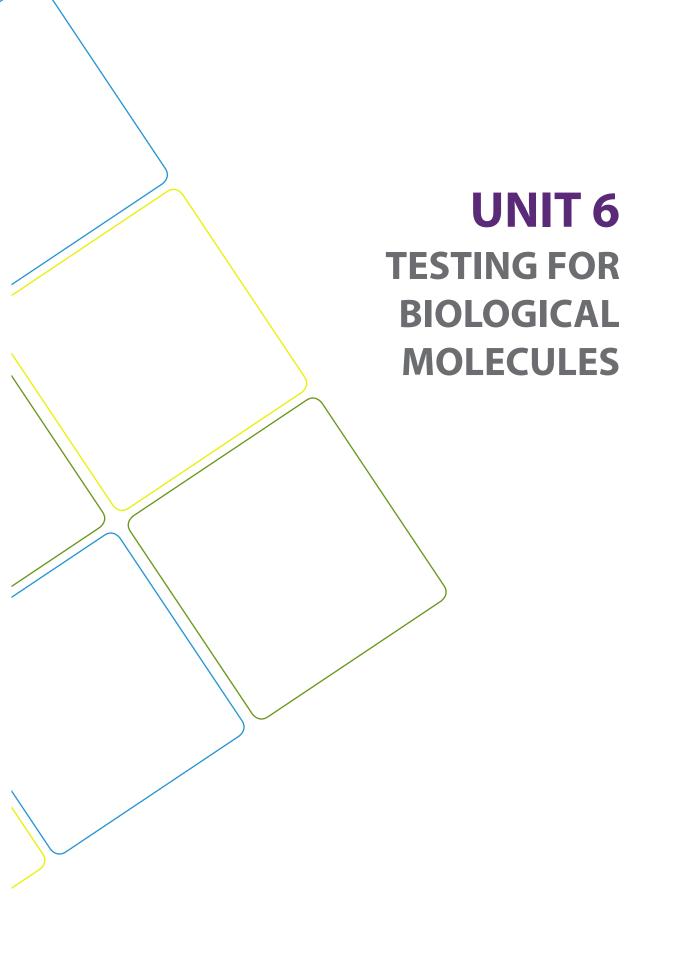
- 1. Give the advantages and disadvantages of being Unicellular organisms.
- 2. Describe how unicellular organisms perform their functions.

#### **End of unit assessment 5**

- 1. Which type of tissue forms glands?
  - a. Epithelial
  - b. Connective
  - c. Nervous
  - d. Muscles
- 2. What are the four types of animal tissues?
  - a. Epithelial, squamous, muscular, connective
  - b. Epithelial, connective, muscular, cardiac
  - c. Connective, muscular, epithelial, nervous
  - d. Cuboidal, ciliated, glandular, columnar
- 3. Which type of the tissues form glands
  - a. Epithelial
  - b. Connective
  - c. Nervous
  - d. Muscle
- 4. Describe how epithelial tissues have adapted to their functions
- 5. Describe the three main functions of the blood
- 6. Complete the following table by filling in the examples of the respective tissues:

Categories of Tissues according to their functions	Examples of tissues
Growth tissues	
Protective tissues	
Storage tissues	
Support tissues	
Conducting tissues	
Secretory tissues	





## **UNIT 6: TESTING FOR BIOLOGICAL MOLECULES**

## **Key unit competence**

Test for biological molecules in a variety of contexts, such as identifying the contents of mixtures of molecules and to follow the activity of digestive enzymes

## **Learning objectives**

By the end of this unit, I should be able to:

- Write out procedures in the identification of biological molecules
- Explain the importance of the reagents used in the identification of biological molecules.
- Carry out tests for the identification of biological molecules
- Compare reducing and non-reducing sugars
- Appreciate the importance of identification of food values in the food industry and in processing and packaging.
- Show resilience making observations on color changes during food tests

### **Introductory activity**

You are given solutions containing different food stuffs including maize flour, vegetable cooking oil, and egg white sugar cane liquid and passion fruit. Using prior knowledge of biological molecules to suggest the type of biological molecule in each one of them. Suggest the chemical tests used to identify each of the molecules.

# 6.1. Test for carbohydrates

## **Activity 6.1**

Materials required:

Starch powder, Irish potatoes juice, prepared porridge, Iodine solution, beakers, droppers, source of heat and test tubes

#### a. Test for starch

#### **Procedure**

- Mix 1g of starch powder with 100ml of water
- Boil the mixture while stirring; then cool the solution
- Boil the mixture while stirring; then cool the solution
- Put 2ml of starch solution in a test tube labeled 1, 2ml of Irish potato juice in a test tube labeled 2 and 2ml of prepared porridge in a test tube labeled 3
- In each test tube put 2 drops of lodine solution and shake
- Record your observation and draw a conclusion

## b. Test for reducing sugar

### Requirements

Glucose powder, beaker and test tube, Benedict solution, Bunsen burner, droppers

### **Procedures**

- In the beaker mix 1cm3 of water and 1g of glucose powder.
- Pour the prepared solution of glucose in a test tube and
- Add 2ml of benedict's solution and heat
- Record your observation.

Biological molecules are grouped into organic molecules including carbohydrates, proteins, lipids, nucleic acids and vitamins. They also contain inorganic molecules such as minerals and water. The first four organic molecules are called macromolecules because they are required in organism in large quantity. Carbohydrates including starch, reducing and non-reducing sugars appear in this category and are the main energy producers in the organisms. Others, including lipids and proteins are needed for building organisms while vitamins protect the organisms against diseases. We need to ensure that what we take from diet have all required biological molecules.

#### a. Test for starch.

Carbohydrates such as starch are tested by mixing a sample with 2-4 drops of iodine or Lugol's solution. If the sample contains starch the solution will turn from a yellowbrown color to a dark purple/dark blue (Figure 6.1). The color change is due to a chemical reaction between the large carbohydrate molecule and the iodine ions. If the sample does not contain starch the solution remains yellow-brown.

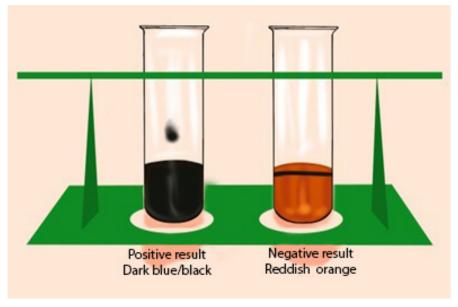


Figure 6.1: Color change during the test for starch

## b. Testing for reducing and non-reducing sugar

The presence of reducing sugar can be tested by using benedict reagent. Benedict solution has copper ions that have a light blue color. When this solution is heated in the presence of simple reducing sugars such as glucose, the blue color of copper ions changes from a light green color to rusty orange-brown color (Figure 6.2).

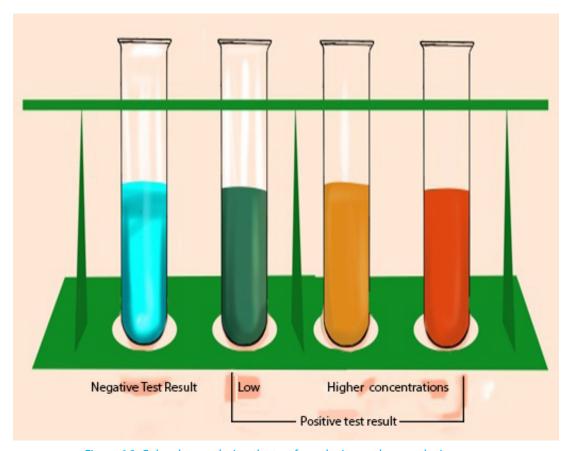


Figure 6.2: Color change during the test for reducing and non-reducing sugar

If the color of Benedict reagent persists, the sugar tested is not a reducing sugar. Note that there is no special reagent to test for non-reducing sugar, but by the addition of HCl, non-reducing sugars can be hydrolyzed to reducing sugars. To test the presence of reducing sugars, a solution of sodium hydroxide is needed to neutralize the acidity because Benedict reagent works better in neutral solution

#### Self-assessment 6.1

A student prepared carbohydrate solution labeled C1. Perform the following experiment to confirm whether C1 is starch, reducing sugar, or non-reducing sugar.

Experiment	Observation	Conclusion
<ol> <li>In a test tube 1:</li> <li>Pour 4 drops of the solution C1</li> <li>Add 2-3 drops of lodine solution</li> <li>Note down your observation and conclusion in the following columns</li> </ol>		
<ul> <li>2. In another test tube 2:</li> <li>Pour 4 drops of the solution C1</li> <li>Add an equal volume of Benedict solution and boil for 1 min</li> <li>Note down your observation and conclusion in the following columns</li> </ul>		
<ul> <li>3. Take another test tube 3</li> <li>Pour 3 drops of the solution C1</li> <li>Add 4-5 drops of dilute hydrochloric acid and boil for 1min.</li> <li>Cool in water</li> <li>Add some drops of sodium hydroxide until the solution turns red litmus paper to blue color.</li> <li>Add 4 drops of Benedict solution, then boil</li> <li>Note down your observation and conclusion in the following columns</li> </ul>		

- a. Is this solution a carbohydrate?
- b. Specify the role of Hydrochloric acid and sodium hydroxide used at stage 3 in the table above.

# 6.2. Test for proteins

# **Activity 6.2**

# **Materials required**

Milk, eggs, soybeans, test tubes, beakers, mortar for crushing beans, 1% NaOH or 1% KOH solution, 0.1M of CuSO4 solution and Millon's reagent.

### **Procedure**

Extract the white fluid from an egg

- Prepare an extra of soya bean and 10ml of fresh milk
- Put 2ml of albumen solution in a test tube labelled A1 and 2ml in A2.
- Put 2ml of milk solution in a test tube labelled B1 and 2ml in B2
- Put 2ml of soya bean solution in a test tube labelled C1 and 2ml in C2
- Put 1ml of KOH or NaOH solution in each of the test tubes A1, B1, and C1. Shake the mixture and add 1ml of CuSO4 solution in each (A1, B1, and C1) test tube
- Put 1ml of Millon's reagent in each of test tubes (A2, B2, and C2). Shake the mixture and thereafter boil the three test tubes (A2, B2, and C2).

The Biuret reagent is used to test for the presence of proteins. It contains copper ions with blue characteristic color. During the copper ions react with protein molecules and causes the biuret solution to turn from a light blue color to purple if proteins are present.

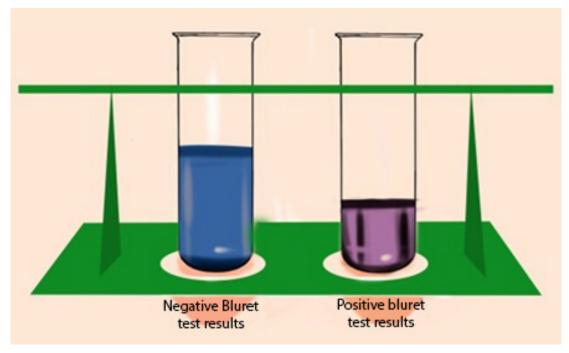


Figure 6.3: Color change during the test for proteins

The test can also be done by using Millon's reagent, which in the presence of proteins, the Millon reagent changes from colorless to pink.

#### Self-assessment 6.2

1. You are provided with the sample of the substance M and A. Carry out the following experiments and complete the table below.

Food substance	Reagent	procedure	Conclusion
M	Millon's	<ul> <li>Take 2cm <sup>3</sup> of test solution M</li> <li>Add 2 cm<sup>3</sup> of Millon's reagent</li> <li>Boil for 1-2 minutes</li> <li>Complete the following column with your observation</li> </ul>	
	Biuret's	<ul> <li>Take 2 cm³ of test solution M</li> <li>Add 2 cm³ of Biuret's</li> <li>Shake and complete the following column with your observation</li> </ul>	

- 2. Carry out the same experiment using the substance A and compare your findings with M.
- 3. Which of the substance A and M contain proteins?

# 6.3. Test for lipids

## **Activity 6.3**

Materials required - Olive oil, test tubes, ethanol, water, sudan III solution

## **Procedure:**

Use olive oil to carry out the following experiments

Add 2 cm<sup>3</sup> of olive oil in the test tube:

- Add 5 cm³ of ethanol followed by 5 drops of water.
- Shake the mixture and record your observation.
- To another test tube containing 2 cm<sup>3</sup> of olive oil:
- add 5 drops of Sudan III solution
- Shake thoroughly and examine the mixture in the test tube after few minute and record your observations

The presence of lipids can be determined by using Sudan III indicators, which are fat-loving molecules that are colored. During the test for a solution containing lipids, two results are likely to be found: there is either the separation of layers indicating the levels of water and lipid, or the dye migrates toward one of the layers. If the mixture is composed of water, the conclusion is that the lipids are not present. In this case, the Sudan III indicator will form small micelles/droplets and disperse throughout the solution. A positive result indicates the lipid layers sitting on top of the water layer with a red-orange color. When using ethanol for testing lipids the presence of the color changes from colorless to milky (emulsion test).

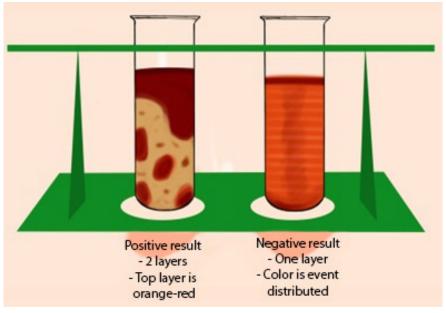


Figure 6.4: Color change during the test for lipids

#### Self-assessment 6.3

You are provided with a solution X. Use Sudan III indicator to test the presence of lipids in the solution X.

# 6.4. Test for vitamin C (Ascorbic Acid).

# **Activity 6.4**

Squeeze the orange fruits to extract the juice and carry out the following test.

Experiment	Observation	conclusion
<ul> <li>To 3cm³of DCPIP add drops of juice extracted from orange</li> <li>Note the observation and draw a conclusion</li> </ul>		
<ul> <li>To 2cm³of water add 2cm³of DCPIP</li> <li>Note the observation and draw a conclusion</li> </ul>		

Which of the two solutions give a positive solution for DCPIP?

Vitamin C is tested by using DCPIP (Dichlophenol Indophenol). Its positive (presence of vitamin C) test decolorizes DCPIP, while the negative (absence of vitamin C) test is indicated by the persistence blue color of DCPIP.

### Self-assessment 6.4

In this experiment you are to press a tomato fruit (s) to get juice out of it. Use the juice to carry out the test for vitamin C Draw a table of results that includes the procedure, observation and conclusion.

### **End of unit assessment 6**

- 1. Biological molecules are divided into:
  - a. Organic molecules and inorganic molecules
  - b. Carbohydrates and starch
  - c. Lipids, carbohydrates and water
  - d. Carbohydrates, food and potatoes
- 2. Name the reagents that are used to test for the following food substances
  - a. Lipids
  - b. Starch
  - c. Reducing sugar
- 3. You are provided with the following specimen:

Specimen A: Sorghum

Specimen B: Irish potatoes

Specimen C: Oranges

Specimen D: Sunflower seeds

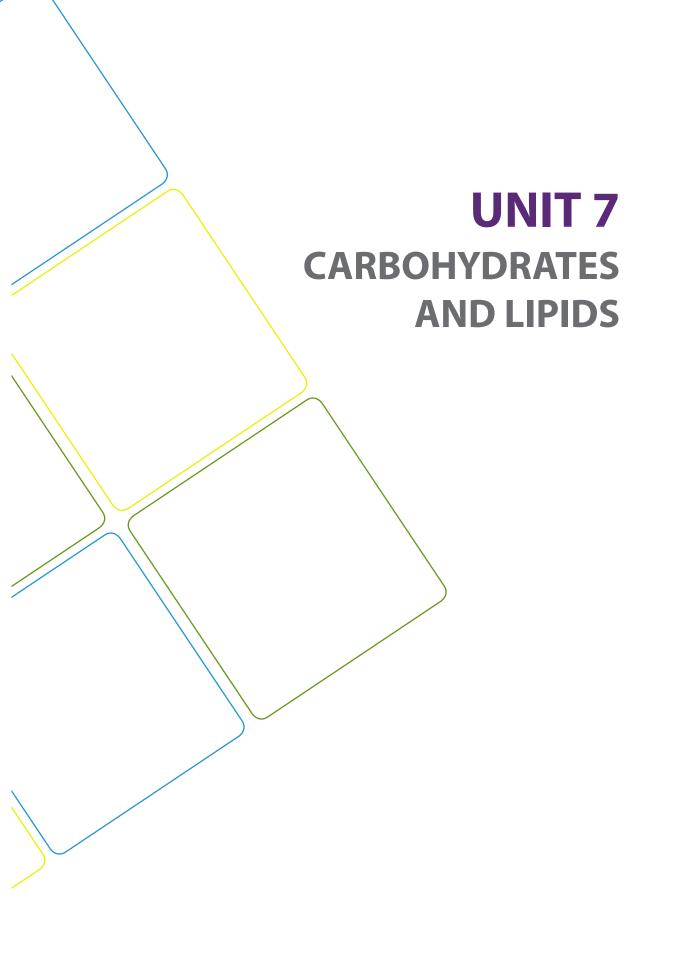
- a. Carry out chemical tests to determine the composition of the above seed to tell whether they are composed of proteins, fats, starch or vitamin C.
- b. Draw the table of used reagent, procedure and observation in (a)
- 4. Some drops of fresh pineapple juice are added drop by drop to DCPIP solution. The deep blue color of the DCPIP quickly fades.
  - a. Explain why the blue colour disappeared?
  - b. What is the importance of this food substance to the human body?
- 5. The result of food tests on unknown sample are shown below. Copy and complete the table to show the conclusions which could be drawn from these tests.

Food test	Result	Conclusion
Sample mixed with iodine in potassium iodide	Blue-black color	
Sample boiled with Benedict's solution	Blue color	
Sample treated with dilute acid, neutralized and then tested with Benedict's solution	precipitate	
Sample tested using Biuret solution	Blue ring on the surface and on shaking purple solution	

6. This is a practical question to be conducted using provided materials and reagents to determine the food nutrients in each solution: You are provided with the following solutions, A (sucrose 0.5%), B (1%starch), C (dilute hydrochloric acid) and D (sodium hydroxide) and 6 test tubes labeled 1 to 6. Use the reagents provided to determine the chemical nature of the substance present in the solutions. Indicate your observations and conclusions in the table below:

Step	Tests	Observation	Deductions (Conclusion)
1	To 1 cm <sup>3</sup> of solution <b>A</b> in a test tube 1 add 2 drops of iodine solution.		
2	To about 1cm <sup>3</sup> of solution <b>A</b> in test tube 2 add an equal volume of Benedict's reagent solution and boil.		
3	To about 1cm³ of solution <b>A</b> in test tube 3 add 4-5drops of solution <b>C</b> and boil. Cool under cold tap water and add an equal volume of solution <b>D</b> then 2 drops Benedict's reagent and boil		

- a. Why was it necessary to boil solutions A and B with solution C in test (3) and (6)?
- b. Why was solution D added to test tubes 3 and 6?



## **UNIT 7: CARBOHYDRATES AND LIPIDS**

## **Key Unit Competences**

Explain the important roles of carbohydrates and lipids in the provision and storage of energy and for a variety of other functions.

### **Learning objectives**

By the end of this unit, I should be able to:

- State the roles of carbohydrates and lipids.
- Recall the elements that make up carbohydrates and lipids.
- Explain the proportion of hydrogen in carbohydrates and lipids and relate this to the amount of energy released when oxidized.
- Define the terms monomer, polymer, macromolecule, monosaccharide, disaccharide and polysaccharide.
- Describe the ring forms of  $\alpha$ -glucose and  $\beta$ -glucose structure.
- Explain the formation of glycosidic bonds.
- Describe the structure of phospholipids and relate to their functions in living organisms.
- Describe the molecular structure and formation of triglycerides and phospholipids, and give their functions in living organisms.
- Demonstrate that phospholipids have a hydrophilic head and hydrophobic tails using a heterogeneous mixture made up of water and cooking oil.
- Interpret the charts and illustrations of molecular structure and the formation of maltose and triglycerides.
- Demonstrate through a process of combustion that sugars and lipids are biological fuel
- Differentiate between starch and cellulose.
- Appreciate the importance of carbohydrates and lipids in organisms.
- Be aware of the other roles of lipids in the formation of soap and with carbohydrates and syrups in medicine

# Introductory activity

- 1. In the previous unit (test for biological molecules), we tested carbohydrates, starch, reducing sugar, lipids, proteins, and vitamins. Where do you classify monosaccharide, disaccharides and polysaccharides in the above tested biochemical compounds?
- 2. Sometimes people say that fatty persons do not feel cold. What could be the reasons?

## 7.1. Classes of monomers

### **Activity 7.1**

Visit the internet and conduct a research the following:

- 1. The description of the term monomer
- 2. Where can we find monomers?
- 3. The biological importance of monomers?

A monomer is a molecule that can combine with others of the same kind to form a polymer. A polymer is a large molecule or macromolecule composed of many repeated sub-units (monomers). Because of their broad range of properties, both synthetic and natural polymers play essential and ubiquitous roles in everyday life. Polymers make up many of the materials in living organisms including proteins, cellulose, and nucleic acids. Glucose molecules for example, are monomers that combine to form the polymer cellulose. The examples of monomers are summarized in the table 7.1.

Table 7.1: Biological molecules and their monomers

Biological molecules	Monomers
Carbohydrates(Polysaccharides)	Monosaccharide
Nucleic Acid	Nucleotides
Proteins	Amino acids

Carbohydrates comprise a large group of organic compounds which contain carbon, hydrogen and oxygen. The word carbohydrate suggests that these organic compounds are hydrates of carbon. Their general formula is  $C_{\nu}(H_{\nu}O)_{\nu}$ . In carbohydrates the ration hydrogen-oxygen is usually 2:1. Carbohydrates are divided into three groups including the monosaccharide (single sugars), disaccharides (double sugars) and polysaccharides (many sugars). The most common monosaccharide of carbohydrates is glucose with molecular formula C<sub>6</sub>H<sub>12</sub>O<sub>6</sub>.

#### Self-assessment 7.1

- 1. What are some examples of polymers and monomers?
- 2. How are monomers, polymers and macromolecules related?

# 7.2. Ring form of $\alpha$ -glucose and $\beta$ -glucose

Monosaccharides are group of sweet and soluble

# Activity7.2

- 1. Based on the knowledge acquired during the lesson of monomers and further information from books and internet:
  - a. What are the examples of monosaccharide?
  - b. Give the molecular formula of each of the monosaccharide stated above
  - c. Use the books to illustrate the structural formula of each of the monosaccharide stated above

crystalline molecules of relatively low molecular mass. They are named with the suffix –ose. The general formula of a monosaccharide is (CH $_2$ O) n, with n the number of carbon atoms. The simplest monosaccharide has n=3 and it is a triose sugar. When n = 5, this is a pentose sugar, and when n = 6, this is a hexose sugar. The two common pentose sugars are ribose and deoxyribose, while the most known hexose is glucose. Its molecular formula is  $C_6H_{12}O_6$ . It is the most important simple sugar in human metabolism called simple sugar or monosaccharide because it is one of the smallest units which has the characteristics of this class of carbohydrates.

Monosaccharides can exist as isomers. The isomer is defined as each of two or more compounds with the same formula but a different arrangement of atoms in the molecule and different properties. For example, glucose, fructose and galactose share the same molecular formula which is  $C_6H_{12}O_6$ . However, they differ by their structural formulae as follow:

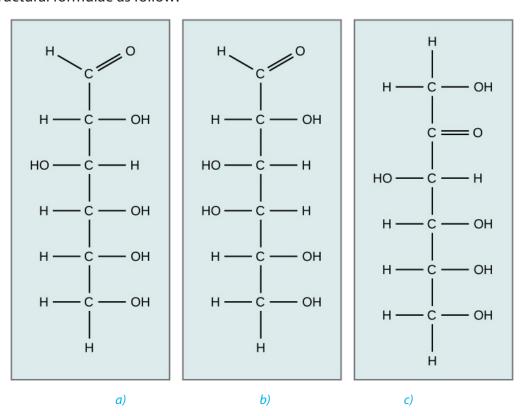


Figure 7.1: Structural formula of glucose(a), galactose(b) and fructose(c)

One important aspect of the structure of pentoses and hexoses is that the chain of carbon atoms is long enough to close up on itself and form a more stable ring structure. This can be illustrated using glucose as an example. When glucose forms a ring, carbon atom number 1 joins to the oxygen on carbon atom number 5 (Figure 7.2).

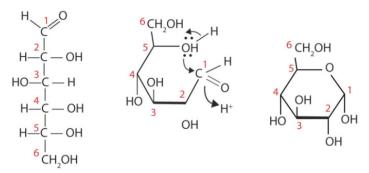


Figure 7.2: Ring formation in the molecule of glucose

All hexoses sugars can exist as straight-chain structures but they tend to form ring structures. Glucose, fructose, galactose can exist in ring structures (Figure 7.3).

Figure 7.3: Ring structures of glucose, galactose and fructose

Ring monosaccharides are said to be alpha ( $\alpha$ ) if the -OH group located on carbon 1 is below the ring and beta ( $\beta$ ) when the -OH group is above the ring. The molecule of glucose for example can exist as alpha and beta glucose denoted by  $\alpha$ -glucose and  $\beta$ -glucose (Figure 7.4)

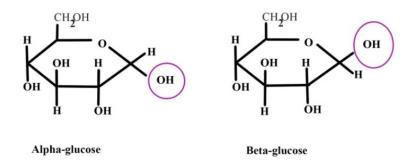


Figure 7.4: Ring form of  $\alpha$ -glucose and  $\beta$ -glucose

#### Self-assessment 7.2

- 1. How do we call the monosaccharide with 3, 5 and 6 carbon atoms?
- 2. Differentiate between  $\alpha$  and  $\beta$  glucose
- 3. What are the properties of glucose?

# 7.3. Formation and breakdown of glycosidic bonds

### **Activity 7.3**

- 1. Monomers are joined to form polymers, use a point as a monomer to illustrate how a polymer can be formed
- 2. How do you call joining structures between atoms?
- 3. Use books or other sources to show how monosaccharide form a disaccharide.

#### 7.3.1. Monosaccharides

Monosaccharides may combine together in pairs to give a disaccharide (double-sugar). The union involves the loss of a single molecule of water and is therefore a condensation reaction. The bond which is formed is called a glycosidic bond. It is usually formed between carbon atom1of one monosaccharide and carbon atom 4 of the other, hence it is called a -1, 4- glycosidic bond. Any two monosaccharides may be linked together to form a disaccharide of which maltose, sucrose and lactose are the most common.

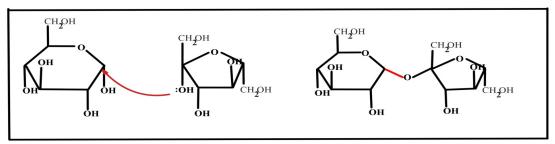


Figure 7.5: Formation of glycosidic bond

The addition of water under suitable conditions is necessary if the disaccharide is to be split into its constituent monosaccharide. This is called hydrolysis water-breakdown or more accurately, breakdown by water.

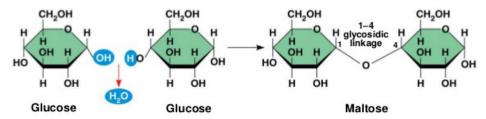
#### 7.3.2. Disaccharides

These are carbohydrates made of two monosaccharides. They include maltose (glucose + glucose), sucrose or saccharose (glucose +fructose), and lactose (glucose+ galactose). The maltose is the sugar from the germinating seeds, sucrose or saccharose is the common table sugar obtained from sugarcane, while lactose is the sugar from the milk. In addition, sucrose is a non-reducing sugar.

Table 7.2: Types of disaccharides and their monomers

Disaccharides	Monomers	
Maltose(malt sugar)	Glucose + glucose	
Sucrose(cane sugar)	Glucose + Fructose	
Lactose(milk sugar)	Glucose + galactose	

In maltose ring, the two ring of glucose are bonded by the -1, 4-glycosidic bond while in sucrose the glucose and fructose are bonded by -1, 2-glycosidic bond.



*Figure 7.6: Illustration of the formation of maltose* 

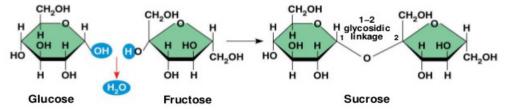


Figure 7.7: Illustration of the formation of sucrose

All the disaccharides are non-reducing sugar, except maltose which behaves in the same as a reducing sugar with benedict's solution. All monosaccharides and disaccharides have the following characteristics: sweet taste, soluble in water and lower molecular mass.

#### Self-assessment 7.3

- 1. Write the molecular structure of sucrose
- 2. How is the glycosidic link is formed
- 3. Sucrose is formed when two monosaccharide are assembled together:
  - a. Name those two monosaccharides.
  - b. Using the ring form of these monosaccharide named above to explain and show sucrose formation?

# 7.4. Polysaccharides: starch, glycogen and cellulose

## **Activity 7.4**

- 1. Based on the meaning of monosaccharide, what is a polysaccharide?
- 2. Classify the following compound into polysaccharide, monosaccharide and disaccharide
  - a. Glucose, fructose and galactose
  - b. Lactose, sucrose, and maltose
  - c. Starch, cellulose and glycogen
- 3. Use glucose to form any polysaccharide of your choice

In the same way that two monosaccharides may combine in pairs to give a disaccharide, many monosaccharides may combine by condensation reactions to form a polysaccharide. The number of monosaccharides that combine is variable

and the chain produced may be branched or unbranched. Polysaccharide are many but the most known are starch, glycogen and cellulose.

#### a. Starch

Starch is made up of two components: amylose and amylopectin. Amylose is a linear unbranched polymer of 200 to 1500  $\alpha$ -glucose units in a repeated sequence of  $\alpha$ -1, 4-glucosidic bonds. The amylose chain coils into helix held by hydrogen bonds formed between hydroxyl groups. A more compact shape is formed. The amylose helices are entangled in the branches of amylopectin to form a complex compact three dimensional starch molecule.

Amylopectin is a branched polymer of 200 to 200,000  $\alpha$ -glucose units per starch molecule. The linear chains of  $\alpha$ -glucose units are held together by  $\alpha$ -1, 4-glucosidic bonds. Branches occur at intervals of approximately 25 to 30 where  $\alpha$ -1, 6-glucosidic bonds occur. Starch grains are found in chloroplast, potato tubers, cereals and legumes. Starch is insoluble in cold water. It is digested by salivary amylase and pancreatic amylase into maltose and the latter is hydrolyzed by maltase enzyme to form glucose. Therefore, diabetic people should avoid tubers since they are rich in starch which in turn gives glucose (Figure 7.8).

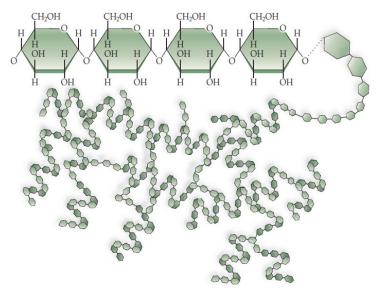


Figure 7.8: Structures of starch molecule. The long chains of alpha glucose molecules are coiled into a helix with most of the hydroxyl groups pointing inwards. © Mader, S. S., et al (2010). Biology 10th Edition.

# b. Glycogen

Glycogen is often called animal starch because it is a major polysaccharide storage material in animals and fungi. The brain and other tissues require constant supply of blood glucose for survival. Some tissues particularly the liver and skeletal muscles store glycogen in the form that can be rapidly mobilized to form glucose. Like starch, glycogen is made up of  $\alpha$ -glucose and exists as granules. It is similar to amylopectin in structure but it has shorter chains (10-20 glucose unit) and is more highly branched.

Figure 7.9: Part of a glycogen showing a straight chain section of alpha glucose joined with 1,4 glycosidic bonds and a branch formed by 1,6 glycosidic bond

#### c. Cellulose

Cellulose is the structural polysaccharide in plant cell wall. It is found in vegetables and fruits but it cannot be hydrolyzed by enzymes in the human digestive system. Cellulose is composed of long unbranched chains of up to 10,000  $\beta$ -glucose units linked by  $\beta$ -1,4-glucosidic bonds. Each  $\beta$ -glucose unit is related to the next by a rotation of 180°C with OH groups projecting outwards on either side of the chain.

Cellulose chains run parallel to one another. Unlike amylopectin and glycogen molecules, there are no side chains (no branch) in the cellulose. This allows the linear chains to lie close together. Many H-bonds are formed between the OH groups of adjacent chains. The chains group together to form microfibrils arranged in larger bundles of macrofibrils. The fibrils give the plant cell their high tensile strength and rigidity. The layers of fibrils are permeable to water and solutes.

Figure 7.10: The structure of a cellulose molecule.

Cellulose is formed from ß - glucose units linked by 1,4 glycosidic bonds. The hydroxyl groups alternate on either side of the molecule forming straight chains giving cellulose a fibrous structure. Cellulose are strengthened further by hydrogen bonds that link adjacent chains.

#### d. Chitin

Chitin is one of naturally occurring Polymers. It forms a structural component of many animals such as exoskeleton in arthropods. Chitin is a polymer of glucose although in its structure a molecule of amino acid is added to each glucose. The digestion of chitin yields simple sugars and ammonia.

#### Self-Assessment 7.4

- 1. What type of reaction is involved in the formation of glucose from starch?
- 2. Use the type of reaction above to form glucose from sucrose molecule
- 3. What are the 2 main components of starch? Give the difference between them

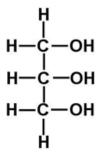
## 7.5. Lipids

### **Activity 7.5**

Use textbooks and/or internet to

- 1. List the monomers that are present in lipids
- 2. Locate where can we find lipids?
- 3. Discuss the reasons why animals like pig do not like hot weather.

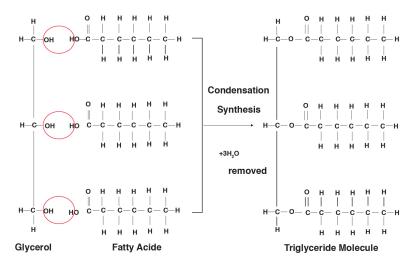
Lipids are a broad group of naturally occurring molecules which include fats, waxes, sterols, fat soluble vitamins (such as vitamins A, D, E and K), monoglycerides, diglycerides, Phospholipids and others. Lipids are grouped into fats which are solid at room temperature and oils which are liquid at room temperature. Lipids are made by carbon, hydrogen and oxygen, but the amount of oxygen in lipids is much smaller than in carbohydrates. Lipids are made by two components namely glycerol and fatty acids. The chemical formula for glycerol is  $C_3H_8O_3$  with structural formula as shown in the figure 7.11



7.11: The Chemical structure of glycerol

In all lipids glycerol do not show any variation while fatty acids vary. Therefore, the nature of lipid depends on the fatty acid it contains. There are two types of fatty acids: unsaturated fatty acid characterized by the chain of hydrocarbon containing one or more double and triple bonds; and saturated fatty acid characterized by the chain of hydrocarbon without any double or triple bond.

Lipids are formed when glycerol combines with one, two, or three fatty acids to form monoglyceride, diglyceride or Triglyceride. A bond is formed between the carboxyl (-COOH) group of a fatty acid and one of the hydroxyl (-OH) groups of the glycerol. This is a condensation process and water is lost. The resulting bond is known as ester link, and the type of reaction is called esterification.



7.12: The illustration of the formation of a lipid

Lipids are of different types as it is summarized in the following table (Table 7.3)

Table 7.3: Types of lipids, their structure, main role and features

Lipid	Structure	Main role	Other features
Triglyceride	Glycerol plus fatty acids	Compact energy store, insoluble in water so doesn't affect water potential.	Stored as fat, which also has thermal insulation and protective properties.
Phospholipid	Glycerol plus two fatty acids and a phosphate group	Forms a molecule that is part hydrophobic, part hydrophilic ideal for basis of cell surface membranes	Phosphate groups may have carbohydrate parts attached: These Carbohydrates are involved in cell signaling.
Cholesterol	Four carbon- based ring structures joined together	Manufacture of Vitamin D Component of mammalian cell membranes Strengthening the membranes at high body temperatures; acts a s thermostat	Used to form steroid hormones

Other lipids include:

### a. Waxes

Waxes are similar to triglycerides but contain fatty acids bonded to long chain alcohol rather than to glycerol. Waxes form the cuticle that protects the leaves and surfaces of insects against the loss of water

#### b. Steroids

A steroid is an organic compound with four rings of carbon and hydrogen atoms with various side chains. Steroids have several functions. It is a component of most animal hormones like estrogen, testosterone.

## **General functions of lipids**

Lipids perform a number of functions within living organism:

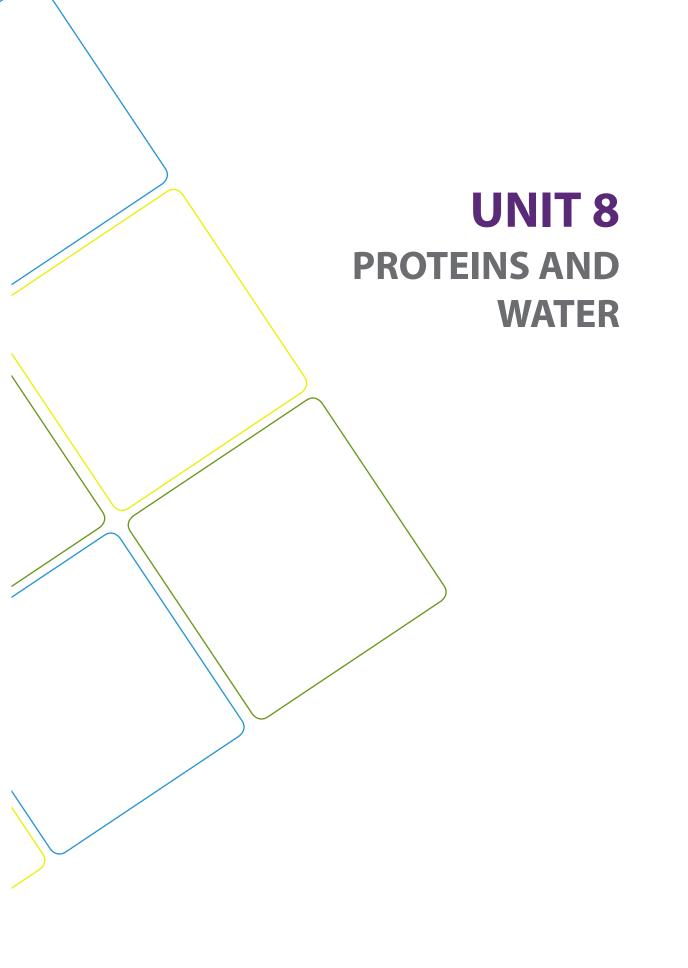
- Lipids are source of energy: due to the presence of C-H bond, lipids can generate more ATP compared to the carbohydrates of the same mass
- Lipids are storage of energy in adipose cells forming adipose tissue in fat of organism
- Lipids act as insulators of the organism. For example, they reduce heat loss. Lipids also are electrical insulators around the nerve cells, the Myelin sheath
- Lipids have a role of protection, in the cuticle of plant leaves against drying, in exposed organ like hand and knees
- Synthesis of hormones such as steroid hormones (most of sex hormones) are made by lipids
- Lipids are used in production of soap by saponification reaction

#### Self-assessment 7.5

- 1. Name the small units found in lipids
- 2. Differentiate between fats and oils

#### **End of unit assessment 7**

- 1. Write the formula of a monosaccharide with 3 atoms of carbon
- 2. Compare the structure of fat(triglycerides)and the phospholipids
- 3. Give two examples of how carbohydrates are used in the body.
- 4. The formula for a hexose is C<sub>6</sub>H<sub>12</sub>O<sub>6</sub> or (CH<sub>2</sub>O)<sub>6</sub>. What would be the formula of?
  - a. Triose
  - b. Pentose
- What type of chemical reaction would be involved in the formation of glucose from starch or glycogen?
- 6. Distinguish between:
  - a. Alpha glucose and beta glucose
  - b. Glycogen and cellulose
  - c. Amylopectin and amylose



## **UNIT 8: PROTEINS AND WATER**

### **Key Unit Competence**

Describe how protein structure is related to function and the role of water as a special molecule with extrardinary properties that make life possible.

## Learning objectives

By the end of this unit, I should be able to:

- Describe the structure of an amino acid and the formation and breakage of a peptide bond.
- Describe the primary, secondary, tertiary and quaternary structure of proteins.
- Describe the molecular structure of hemoglobin as an example of a globular protein.
- Describe the functions with an emphasis on iron in the hemoglobin molecule.
- Explain the effect of heat, pH and chemicals on protein structure.
- Explain how hydrogen bonding occurs between water molecules and relate the properties of water to its roles in living organisms.
- Devise an experiment to investigate the effect of temperature, pH and chemicals on the structure of protein.
- Relate the structure of globular and fibrous proteins to their functions.
- Investigate the effect of lowering temperature on water.
- Distinguish between collagen molecules and collagen fibres
- Appreciate the importance of globular and fibrous proteins in biological processes such as the transport of gases and providing support for tissues.
- Express that protein structure is central to many aspects of biology, such as enzymes, antibodies and muscle contraction.
- Acknowledge that water is a special molecule with extraordinary properties that make life possible on this planet.

# **Introductory activity**

Conduct a research by using different sources of information and answer the following questions

- 1. What is a protein?
- 2. What do you understand by universal solvent in living organisms?

## 8.1. Proteins

Proteins are organic compounds of large molecular mass. For example, the hemoglobin has a molecular mass of 64500. In addition to carbon, hydrogen and oxygen, proteins always contain nitrogen, usually sulphur and sometimes phosphorus. Proteins are polymers of amino acids and they are not truly soluble in water, but form colloidal suspensions.

### 8.1.1. Amino acids

Amino acids are group of over a hundred chemicals of which around 20 commonly occur in proteins. They always contain a basic group, the amine group (-NH<sub>2</sub>) and a carboxylic acid group(-COOH) together with -R group or side chain (Figure 8.1). All the amino acid differs one to another by the structure of their side chain.

Figure 8.1: The generalized structure of amino acid. R is a variable group that changes from one amino acid to another and so determines the type of amino acid

Amino acids are divided into two categories: essential amino acid and non-essential amino acid. Essential amino acids are those amino acids which cannot be synthesized by the body. Non –essential amino acids are synthesized by the organism. All 20 amino acids can be found in diet from plant and animal tissues.

Figure 8.2 a: Table of categories of amino acids that make up proteins

### Amphoteric nature of an amino acid

When an amino acid is exposed to basic solution, it is deprotonated (release of a proton H+) to became negative carboxylate COO -while in acid solution it is protonated (gains of a proton H+) to became ammonium positive ion -NH3 +(Figure 8.1.3.a and Figure 8.1.3.b).

Figure 8.3 a: The amino acid in a basic solution

$$H_3$$
 $\stackrel{\uparrow}{N}$ 
 $\stackrel{\downarrow}{=}$ 
 $\stackrel{\downarrow}{=}$ 

Figure 8.4 b: The amino acid in acid solution

At a physiological pH, usually around 7, the amino acid exists as **ZWITTERION** (from German means hermaphrodite) it is a molecule with two different charges (positive and negative) at the same time (Figure 8.1.4).

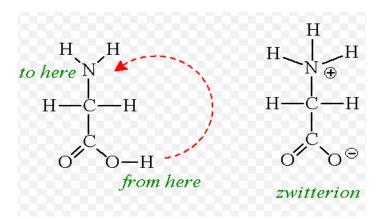


Figure 8.5: Formation of zwitterion in amino acid

## 8.1.2. Formation and breakage of peptide bond

The formation of peptide bond follows the same pattern as the formation of glycosidic bond in carbohydrates and ester bond in fats. A condensation reaction occurs between the amino group of one amino acid and the carboxyl group of another, to form a dipeptide (fig 8.5).

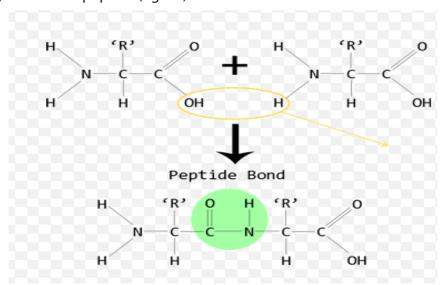


Figure 8.6: The formation of the dipeptide by condensation reaction resulting in a peptide bond

A peptide bond is formed between two amino acids to form a dipeptide molecule. If three amino acids are assembled together they form a tripeptide while four amino acids form a tetrapeptide and so on. A long chain of amino acid it is called a polypeptide. The polypeptide chain or oligopeptide comprise more than 50 amino acids joined together by peptide bonds.

During digestion, proteins are hydrolyzed to give amino acids that can be diffused across the wall of intestine into blood stream. In hydrolysis the peptide bond breaks down by the addition of a water molecule (Figure 8.5).

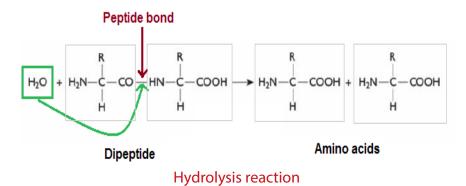


Figure 8.7: Breakage of a peptide bond by addition of water.

#### Self-assessment 8.1

- 1. Explain what are essential amino acids?
- 2. Describe the formation of a peptide bond?
- 3. At physiological pH, the amino acid exists as zwitterions. What is a zwitterion?
- 4. Alanine is an amino acid with -CH<sub>3</sub> as a side chain. Write its structural formulae.
- 5. Most plants lack one or more of the essential amino acids needed by the body. Explain how a vegetarian can obtain the essential amino acids.

# 8.2. Structure and denaturation of proteins

# **Activity 8.2**

- 1. From the books make a research on proteins and answer to the following questions:
  - a. What are different structures of proteins?
  - b. Differentiate globular proteins and fibrous proteins.
- 2. Take a plastic rope cord, create the nodes bulk on it and suppose that those are monomers of a long chain of polymer (the whole cord). Heat it using a Bunsen burner or another source of fire. Discuss the change that takes place.

# 8.2.1. Structure of proteins

The long chain of polypeptide can take different forms according to its molecular weight and the types of bond that hold together atoms and molecules.

# a. Primary structure of proteins

Primary structure of a protein is the sequence of amino acid that is made up of the polypeptide chain or chains.

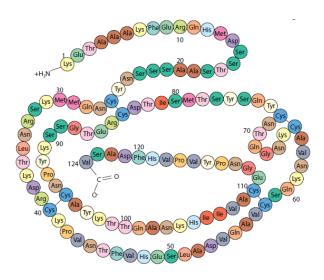


Fig 8.8: Primary structure of protein: example ribonuclease.

Ribonuclease is an enzyme found in pancreatic juice, which hydrolyses (digests) RNA. Notice that at one end of the amino acid chain there is an -NH3+ group, while at the other end there is a -COO- group. These are known as the amino and carboxyl ends, or the N and C terminals, respectively. (Adapted from Cambridge International AS and A Level Biology Course Book Fourth Edition)

## b. Secondary structure of proteins

The regular arrangement of amino acids in primary structure can induce the interaction of the back bone of the polypeptide chain (side chain) by hydrogen bonds. Those side chains are coiled and folded in the patterns that contribute to the protein's overall shape. One such secondary structure is α-helix and sometime β-pleated sheet (Figure 8.8).

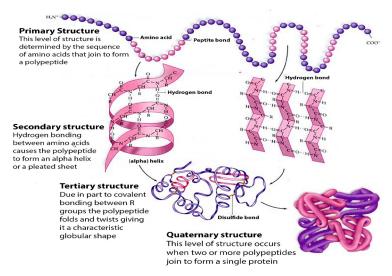


Figure 8.9: A alpha(a) helix and beta (B)pleated sheet of the secondary structure of proteins. The hydrogen bond at different locations of the molecule determines the shape of the structure.© www.slideshare.net

## c. Tertiary structure of proteins

In addition to hydrogen and peptide bond in primary and secondary structure, the tertiary structure of protein has other types that include:

- Hydrophobic interaction
- lonic bond between positively and negatively charged r groups.
- Disulfide bridges (-s-s)

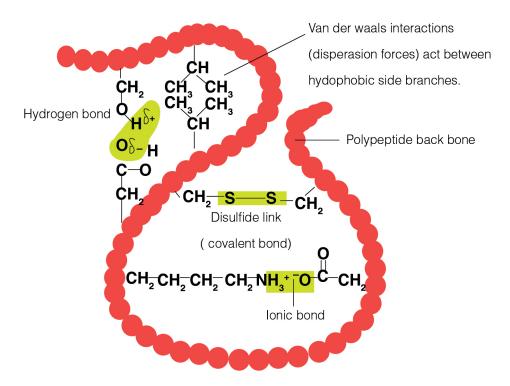


Figure 8.10: Tertiary structure of proteins. (Source Wikipedia)

## d. Quaternary structure of proteins

Quaternary structure involves more than one polypeptide chain chemically bonded to each other. The quaternary structure refers to the way in which these polypeptide chains are arranged in the protein. Examples, Hemoglobin that is composed of:

- Four polypeptide subunits, two  $\alpha$  chains and two  $\beta$  chains. Both  $\alpha$  and  $\beta$  subunits primarily are  $\alpha$  helical secondary structure polypeptide chain with 140 amino acids.
- Haeme composed of iron that binds with oxygen.

Collagen: this is a fibrous protein consisting of three helical polypeptides that are supercoiled to form a rope like structure of great strength

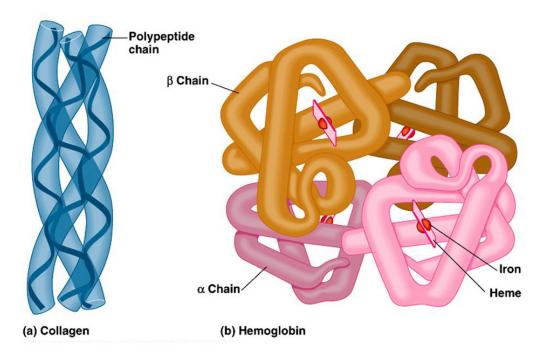


Fig 8.11: (a) Quaternary protein structure of Collagen, (b) Structure of haemoglobin.©Campbell, N.A.,(2008). Biology. 8th Ed.

# **Globular protein**

These are polypeptide chains that are tightly folded to form a spherical shape. Many globular proteins are folded so that their hydrophobic groups are on the inside of the molecule and the hydrophilic groups face outwards making these proteins soluble in water.

Properties of globular protiens:

- They are spherical in shape
- Physiologically active
- Soluble in water.
- May contain prosthetic group for example the iron (haeme)
- Examples include hemoglobin and enzymes.

#### 8.2.2. Protein denaturation

**Protein denaturation** is a process by which protein changes shape due to breakage of bonds holding the polypeptide chains. Protein denaturation may be temporary or permanent.

The agent of denaturation can be caused by;

- Extremely high temperatures beyond optimum,

- changes in pH,
- Ultra Violet (UV) rays,
- High salt concentration and heavy metals.

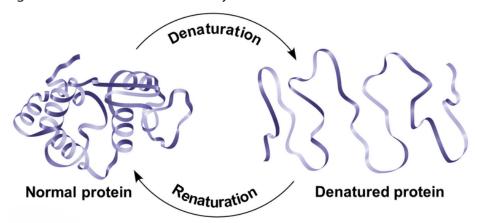


Figure 8.12: Denaturation and renaturation of a protein. ©Jackson, B.R. (2008). Biology. 8th Ed.

# 8.2.3. Functions of proteins.

- Proteins such as lipase, pepsin and protease act as enzymes as they play a crucial role in biochemical reaction where they act as catalysts.
- Proteins play an important role in coordination and sensitivity (hormones and pigments).
- Proteins have a transport functions. Example: Haemoglobin transport oxygen
- Proteins in the cell membrane facilitate the transport of substance across the cell membrane.
- Proteins provide a mechanical support and strength.
- Proteins such as myosin and actin are involved in movement.
- Proteins play the role of defense of the organisms. Example: Antibodies are proteins

#### Self-assessment 8.2

1. Describe the protein structures

# **8.3. Water**

# **Activity 8.3**

- 1. What is the medium of reaction in the organisms?
- 2. If two people are boiling the same quantity of cooking oil and water, which one could evaporate first? Explain your choice.

Living organisms contain between 60% and 90% of water, the remaining being the dry mass. The function of water is defined by its physical and chemical properties that differ from those of most liquids and make it effective in supporting life.

# 8.3.1. Biological significance of the physical properties of water

Property of water	Significance for living organism
Water is a liquid at room	Provides a fluid environment inside the cells and aquatic
temperature	environment for organisms to live in.
Universal solvent	The chemical reaction inside the cell happens in aqueous solution. Water is the main transport medium in organisms
Water has high surface tension	Water forms a surface film at an air water interface. This allows some aquatic organisms such as pond skaters to land on the surface of ponds and move over it.
Ice is less dense than liquid water	Ice forms on the surface the body of water and insulates the water below, allowing aquatic life to survive)
Water has adhesion forces	Along with low viscosity adhesion forces help capillarity so that for example, water can move upward through narrow channels in the soil against gravity.
High specific heat capacity	Water being a major component of internal fluids, organisms resist temperature changes and so remain relatively stable.
High latent heat of vaporization	Heat is lost from surface when water evaporates from it. This is used as cooling mechanism; sweating in animals and transpiration in plants
High latent heat of fusion	Cell content and aquatic habitats are slow to freeze in cold weather.
Water is colorless and transparent	Transmission of sunlight helps aquatic plants to photosynthesize.
Water is denser than air	Acts as a habitat for large organisms. It helps supports and disperses reproductive structures such as larvae and large floating fruits like coconuts
Water is difficult to compress	Water is an important structural agent acting as a hydrostastic skeleton in invertebrates (worms) and turgid cells in plants
Water takes part in many chemical reactions.	Water is a major raw material for photosynthesis and also take part in many digestive reactions in breaking down food molecules by hydrolysis
Water has high tensile strength	Continuous columns of water are pulled up the xylem to the top of the plant during transpiration
Water combines with many organic molecules to form hydrated molecules	Most organic molecules occur in a hydrated form in a cell. If water is removed, their physical and chemical properties are affected; the use and storage of food.

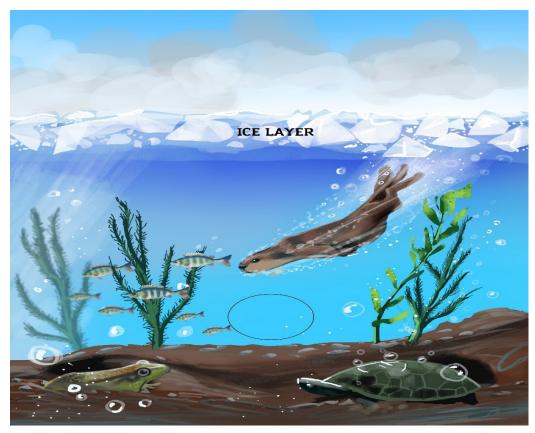


Figure 8.13: The unique behavior of water below and above 4 degrees Celcius that helps aquatic organisms to survive in winter. Songer, M. T. (2010). Biology 10th Edition.

#### **Functions of water**

- Turgidity of plant cell which increases their size is due to the availability of water.
- The transport of substances (minerals, nutrients in plant and animals) that are dissolved in water.
- Excretion of waste product
- Support for hydrostatic skeleton.
- Temperature regulation in plant and animals
- Seed germination by breaking down the seed coat
- Medium for biochemical reaction.

#### Self-assessment 8.3

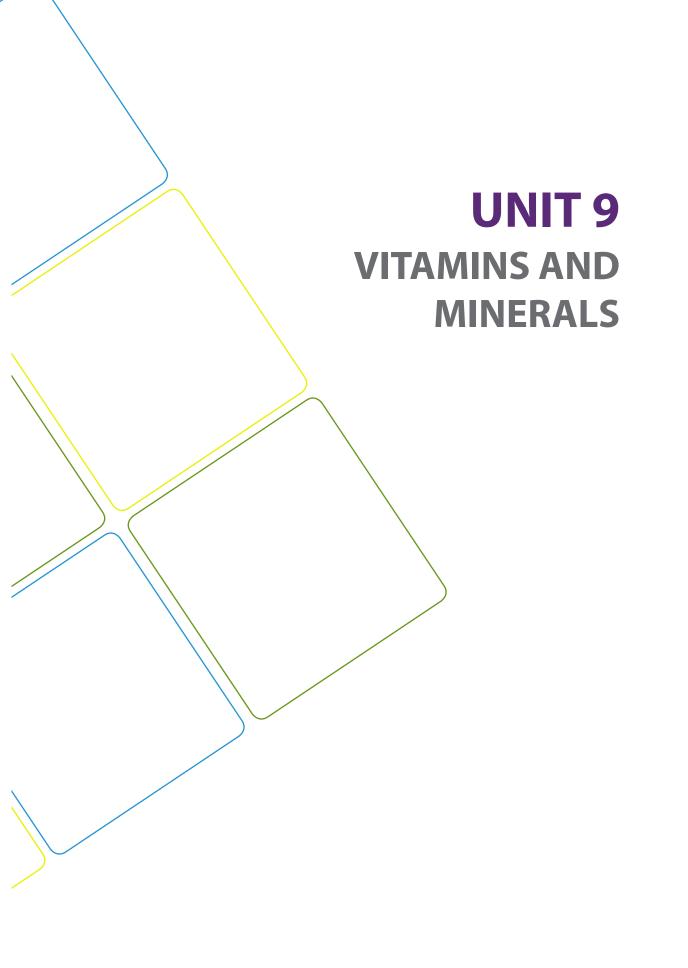
- 1. State the functions of water in animals
- 2. What do you understand by heat capacity?
- 3. Relate the high heat capacity of water to its biology functions.
- 4. Describe and explain how aquatic organisms live below frozen water bodies

#### **End of unit assessment 8**

- 1. Certain drugs can break the covalent bond between two sulfur atoms of non-adjacent amino acids. Which level of protein that can be affected most if the drug is mixed with primary, secondary, tertiary and quaternary structure of proteins?
- 2. Complete the following statements by appropriate terms:
  - a. The formation of large molecules from small repeating units is called .....reaction.
  - b. A carbohydrate(polysaccharide)that is formed by the plant as a reserve food supply and made up of only glucose molecules covalently bonded together is........
- 3. State the property of water that allows each of the following to take place. In each case, explain its importance:
  - a. The cooling of skin during sweating
  - b. The transport of glucose and ions in a mammal
  - c. Much smaller temperature fluctuations in lakes and oceans than in terrestrial (land-based) habitats.
- 4. Construct a three column table and relate the following terms with arrows to indicate the correct match.

Phosphodiester linkages Monosaccharide Polypeptides
Peptide bonds Nucleotides Triacylglycerol
Glycosidic linkages Amino acids Polynucleotides
Ester linkages Fatty acids Polysaccharides

5. Explain what happens during protein denaturation?



# **UNIT 9: VITAMINS AND MINERALS**

## **Key Unit Competence**

Discuss the roles of minerals and vitamins in diet

## **Learning objectives**

At the end of this unit you be able to:

- State the mineral requirements for bodily functions.
- Identify the symptoms of mineral and vitamin deficiency.
- Outline the need for consumption of minerals and vitamins in small amounts.
- Organize a list of foods that are good sources of vitamins and mineral salts.
- Recognize the signs and symptoms of scurvy, night blindness, goiter, and anaemia.
- Differentiate between water soluble and lipid soluble vitamins.
- Analyze one's eating habits and suggest improvements
- Appreciate the importance of a balanced diet in relation to health and economic prosperity.
- Advocate for healthy feeding methods.

# **Introductory activity**

- 1. From the different food stuffs in our community, make a list of food stuffs that are good sources of minerals and vitamins.
- 2. Using text books and other resources, make a list of e vitamins and mineral deficiency diseases

# 9.1. Mineral nutrients in humans

# **Activity 9.1**

Use textbooks and internet to list mineral nutrients found in human diet

Mineral nutrients are sometimes called mineral salts or just minerals. Mineral salts are essential nutrients that our body needs. They are called essential not because they are more important than other substances in our body but because our bodies can not produce them. They include the inorganic substances found in daily diet. They are dissolved in body fluids.

They are found in human body as ions (cations and anions). Organic food like proteins, carbohydrates and fats provide the body with carbon, hydrogen, oxygen, nitrogen, sulfur and phosphorus. But there are several more elements that the body needs and occur as salts in the food we eat. They constitute about 1% of an organism by weight. Even though they are required in a very small amount, they are nonetheless essential for body processes.

Some mineral nutrients are required by animals, plants, a few by both. Humans require a number of minerals for the good functions of their bodies. Those are: Calcium (Ca<sup>2+</sup>), phosphorus (H<sub>2</sub>PO-<sub>4</sub>), Nitrogen(NO-<sub>3</sub>), Sulfur (SO<sup>2-</sup><sub>4</sub>) Potassium (K+), Sodium (Na+), Iron (Fe<sup>2+</sup> or Fe<sup>3+</sup>), Magnesium (Mg<sup>2+</sup>), Iodine (I-), Chloride (Cl-), Manganese (Mn<sup>2+</sup>), Fluoride (F-), zinc (Zn<sup>2+</sup>), Cobalt (Co<sup>2+</sup>), Chromium (Cr<sup>2+</sup> or Cr<sup>3+</sup>) and molybdenum (MoO-<sub>4</sub>)

#### Self-assessment 9.1

- 1. Outline ten mineral nutrients required in human diet.
- 2. Answer by true or false and justify your answer: "Minerals are called essential nutrients because they are more important than others".

## 9.2. Classification of mineral nutrients

# **Activity 9.2**

lodized table salt is advised to prevent goiter. In 100g of table salt there is 99% of NaCl, and only 1% of iodine. Refer to the notes below to find the reason behind this ratio.

The classification of minerals is based upon their requirement rather than on their relative importance. Mineral nutrients are needed in a precise small amount. The five major minerals needed in human body include calcium ( $Ca^{2+}$ ), phosphorus ( $H_2PO_4^-$ ), potassium ( $K^+$ ), sodium ( $Na^+$ ) and magnesium ( $Mg^{2+}$ ). Mineral nutrients are grouped into two groups: the macronutrients or major elements and the micronutrients or trace elements.

**Macronutrients** or **major elements** are minerals needed by humans in a relative large amount (greater than 200 mg/day). Their examples include nitrogen(NO<sub>3</sub>-), phosphorus (H<sub>2</sub>PO $_4$ ), sulfur(SO $_4$ ), calcium (Ca<sup>2+</sup>), sodium (Na<sup>+</sup>), chlorine (Cl<sup>-</sup>), magnesium (Mg<sup>2+</sup>), and iron (Fe<sup>2+</sup> or Fe<sup>3+</sup>).**Micronutrients** or **trace elements** are those which are needed in minute amount (a few parts per million). Examples include manganese (Mn<sup>2+</sup>), iodine (l<sup>-</sup>), zinc (Zn<sup>2+</sup>), molybdenum(MoO $_4$ )and fluorine (F<sup>-</sup>).

#### Self-assessment 9.2

- 1. Categorize mineral nutrients according to their amount in human body.
- 2. Distinguish the two categories of mineral nutrients needed by the human body.
- 3. From the minerals listed here, identify the five major minerals in the human body: Sulfur (S), Potassium (K), Sodium (Na), Iron (Fe), Magnesium (Mg), Iodine (I), Chloride (CI), zinc (Zn), Cobalt (Co), chromium, Calcium (Ca), phosphorus (P), Nitrogen (N).

# 9.3. Sources, functions and deficiency symptoms of mineral nutrients in humans

#### **Activity 9.3**

Use your textbook to answer the following questions:

Hereunder is a variety of food stuffs: Banana, cassava, wholegrain, oranges, pumpkin, potato, beans, water melon, green leafy vegetables, poultry, eggs, liver, and milk. Choose the food stuffs which are good sources of minerals.

Human body requires mineral nutrients to survive and to carry out daily functions and processes. Minerals keep humans healthy and have key roles in several body functions. Humans receive minerals by eating plants that absorb minerals from the soil and by eating meat and other products from animals, which graze on plants. The deficiency of mineral nutrients results into body functional disorders and diseases. Most are found in the blood and cytoplasm of cells, where they assist basic functions. For example, calcium and potassium regulate nerve and muscle activity



Figure 9.1: Examples of some foods rich in minerals

Table 9. 1 Minerals required in humans and their sources

Mineral	Major dietary sources	Some major functions	Mineral deficiency diseases and their symptoms.
Iron (Fe)	Meats, eggs, legumes, whole grains, green vegetables	Component of hemo- globin and of electron carriers in energy metabolism; enzyme cofactor	Iron-deficiency anemia, weakness, impaired immunity
Calcium (Ca)	Milk, soy milk, green leafy vegetables, sardines	Needed for nerve and muscle action; builds bone and teeth; helps blood clot	Retarded growth, possibly loss of bone mass and bone deformation called rickets.

Phosphorus (P)	Meat, poultry, pumpkin seeds, sunflower seeds, water melon, whole grains	Component of bones, teeth, lipids, cell membrane, and nucleotides.	Phosphorus deficiency results in a form of Bone malformation known as rickets	
Sodium (Na)	Table salt, most processed foods	Needed for muscle and nerve function; helps maintain salt-water balance in body fluids, and assists active trans- port of certain material across the cell	cramps, reduced appetite	
Potassium (K)	Meats, soy, beans, orange juice, tomato, potatoes, bananas	Assists active transport of certain material across the cell membrane Needed for muscle and nerve function; helps maintain salt-water balance in body fluids	Muscular weakness, paralysis, nausea, heart failure	
Sulfur (S)	Whole grains, meats, seafood, eggs	Necessary component of amino acids many proteins and some coenzymes, e.g. acetyl coenzyme A	Symptoms of protein deficiency	
Nitrogen (N)		Is a component of amino acids, proteins, coenzymes, vitamins	Digestion problem, skin disorders, defective bone growth.	
Chlorine (CI)	Table salt, most processed foods	Helps maintain water and pH balance; helps to form stomach acid (HCI)	Muscle cramps, reduced appetite	
Fluorine (F)	Drinking water, tea, seafood	Component of certain digestive enzymes, component of teeth and bones	Tooth decay	
Magnesium (Mg)	Whole grains, green leafy vegetables, nuts, seeds	Needed to form several enzymes	Nervous system disturbances	

Copper (Cu)	Seafood, nuts, legumes, organ meats,	Enzyme cofactor in iron metabolism, melanin synthesis, electron transport, maintains the immune system stronger.	Growth failure, scaly skin inflammation, reproductive failure, impaired immunity.
lodine	Seafood, dairy products, iodized salt	Component of thyroid hormones	Cretinism, Goiter (enlarged thyroid)
Cobalt	Meats and dairy products	Component of vitamin B12	Deficiency of vitamin B12
Molybdenum	Beans, lentils, peas, grain products and nuts are the richest sources of molybdenum.	Promotes normal cell function, functions as a cofactor, used in production of red blood cells, enables the body to use nitrogen.	Tachycardia, tachypnea, headache, nausea, vomiting, and coma.
Manganese	Nuts, grains, vegetables, fruits, tea	Enzyme cofactor, growth factor in bone development.	bone and cartilage deformation
Zinc	oysters, beef, lamb, spinach, pumpkin seeds, squash seeds, nuts, dark, pork chocolate, beans, chicken and mushrooms.	It acts like a powerful antioxidant, is needed for proper cell division, to convert vitamin A into its active form in order to maintain proper vision, it is also needed for proper immune system functioning and wound healing.	Skin rashes and acne, thinning hair, nerve dysfunction, weak immunity, and nutrient malabsorption.

#### Self-assessment 9.3

1. Match the mineral nutrients with its function

a. lodine 1. make bones hard

b. Fluorine 2. maintains the immune system stronger

c. Phosphorus 3. component of hemoglobin

d. Iron 4. prevents tooth decay

5. used in synthesis of thyroid hormone (thyroxin) e. Copper

2. In a tabular form, identify the major dietary sources, the functions in human bodies and the deficiency diseases of the following minerals: Ca, I, P, zinc, and Cu

- 3. Choose the best answer.
  - i. They are the minerals we need a lot in every day diet. How are they called?
    - a. Macronutrents
    - b. Micronutrients
    - c. Giant minerals
    - d. Monster minerals
    - ii. This mineral helps to build up strong teeth and bones. How is it called?
    - a. Calcium
    - b. Iron
    - c. Zinc
    - d. potassium
    - iii. What are foods that are natural good source of iron?
    - a. Roast Beef
    - b. Macaroni and cheese
    - c. Baked beans
    - d. Water melon
    - iv. Select 2 that are natural good sources of calcium?
    - a. Milk and cheese
    - b. Whole-wheat bread
    - c. Iceburg lettuce
    - d. Scanned salmon
  - v. The mineral that helps in oxygen transport to lungs is?
    - a. calcium
    - b. iron
    - c. zinc
    - d. potassium
  - vi. Which foods are good sources of zinc?
    - a. Milk and cheese
    - b. Lamb and pork
    - c. Macaroni and cheese
    - d. Peanuts and lentils
- vii. Bananas are great source of this mineral, which helps our muscles and nervous system to maintain your right water levels. What is it called?
  - a. Calcium
  - b. Iron
  - c. Zinc
  - d. Potassium

viii. Which mineral is important and needed by our body to fight off infection?

- a. Calcium
- b. Iron
- c. 7inc
- d. Potassium
- ix. Which of the following mineral are needed in large amount every day?
  - a. Zinc
  - b. Iron
  - c. Calcium
  - d selenium
  - x. Which is the type of mineral that keep your nervous system health?
    - a. Calcium
    - h Iron
    - c. Zinc
    - d. Potassium
- 4. From the diseases listed below, what are those caused by the deficiency of minerals?

Goiter, malaria, diabetes, rickets, beriberi, scaly skin, night blindness, anemia, impaired immunity, diarrhea

# 9.4. Vitamins and the classification of vitamins

# **Activity 9.4**

Two students with different complains went to consult a medical doctor.

Student A says to the doctor that whenever he/she bleeds whenever she /he brushes teeth.

Student B doesn't see well objects around him/her,

The results from the doctor showed that they all have lack some vitamins.

- 1) What kind of vitamins that each student needs to take?
- 2) Use your student textbook to explain your answer

Like minerals, vitamins are also essential for the human body. They are required for metabolism, protection health and growth. Vitamins also assist in formation of

hormones, blood cells and genetic material. Vitamins are directly absorbed from the small intestine into the blood stream. Water -soluble vitamins are absorbed in the ileum while fat-soluble vitamins are absorbed in jejunum. Features shared by all vitamins:

- They are not digested or broken down for energy
- They are not synthesized into the body structures
- Most are rapidly destroyed by heat.
- They are essential for good human health and needed in a very small amount
- They are required for chemical reactions in cells, working in association with enzymes.

There are thirteen vitamins required by human body. They are classified by their solubility, whether they dissolve in water or in fats. Water-soluble vitamins including vitamins C and B complex, and fat-soluble vitamins including vitamins A, D, E and K (Table 9.2). Excess water- soluble vitamins are simply excreted in urine, while fatsoluble vitamins are stored in body fatty tissues to be used later if there is deficient in diet. Excess intakes of these vitamins are stored in fatty tissues of the body, where they can build up to toxic levels, especially if they are taken improperly in supplements.

Table 9.2 Water-soluble and fat-soluble vitamins

Water-soluble vitamins		Fat-soluble vitamins		
Vitamin	Name	Vitamin	Name	
B1	Thiamine	Α	Retinol	
B2	Riboflavin	D	Calciferol	
B3	Niacin (nicotinic acid)	Е	Tocopherol	
B5	Pantothenic acid	К	Phylloquinone	
B6	Pyridoxine			
B7	Biotin			
B9	Folic acid (folacin)			
B12	Cyanocobalamine			
С	Ascorbic acid			

#### Self-assessment 9.4

- 1. How many vitamins does the human body needs to function properly?
- 2. Describe the classification of vitamins.

# 9.5. Sources, functions and symptoms of vitamin deficiency

## **Activity 9.5**

Here is a number of foodstuffs rich in vitamins.



Figure 9.2 Food rich in vitamins

From the list of provided food stuffs (Banana, cassava, wholegrain, oranges, pumpkin, potato, beans, water melon, green leafy vegetables, and milk). Can you give some foods that are good sources of vitamins?

Some vitamins, including some vitamin B complex and Vitamin K are produced by bacteria that normally live in the intestines, where they help to digest food. Vitamin D is synthesized in the skin when it is exposed to UV radiation in sunlight.

Vitamins and their derivative are coenzymes; note that a coenzyme is an organic molecule that combines temporaly with enzymes making them more efficient. For example, Niacin or vitamin B3 is an essential component of coenzymes NAD and NADP involved in lipid metabolism. It inhibits production of cholesterol and catabolism of triglyceride. Thiamin or vitamin B1 is a coenzyme for many different enzymes that break complex molecules such as carbohydrates to produce ATP.

Thiamin deficiency results into Beriberi anemia and stunted growth in children. Vitamin K is an essential coenzyme for synthesis of several blood clotting factors. Several vitamins, including vitamins C and E, act as antioxidants. An antioxidant is a compound that neutralizes chemicals called free radicals. Free radicals are produced naturally during cellular activities and may cause some types of cancer. Neutralizing free radicals makes them harmless.

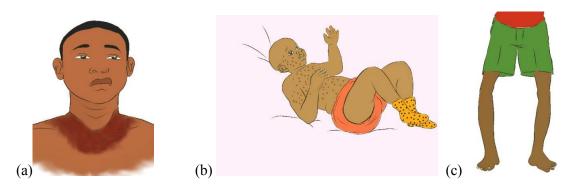
The table: 9.3. The major dietary sources, functions and possible symptoms of vitamin deficiency

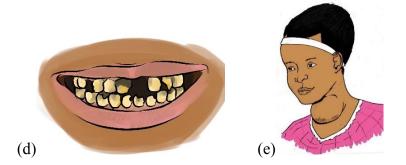
Vitamins	Major dietary sources	Function in our lives	Possible symptoms of deficiency
Vitamin B1 (thiamin)	Whole-grain products, eggs, peas, fish, yeast, beans, peanuts, meats	Helps break down macronutrients; essential for proper functioning of nerves	Beriberi: anemia, nerve disorders (such as confusion, paralysis, atrophy of limbs, hallucinations), and stunted growth in children.
Vitamin B2(riboflavin)	Milk, liver, green leafy vegetables, beef, eggs, Whole- grain products peanuts, yeast soybeans, lamb.	Helps the body process amino acids and fats; acts as antioxidants	Hair loss, insomnia, skin lesions such as cracks at corners of mouth, dermatitis and blurred vision. Cataracts, lesion of intestinal mucosa, and one type of anemia.
vitamin B9 Folic acid (folate or folacin)	Liver, green leafy vegetables, citrus fruits, legumes, and fortified bread.	Needed for normal production of red blood cells and white blood cells. Also is needed in DNA and RNA synthesis.	Anemia, gastrointestinal problems.
Vitamin B3 Niacin or (nicotinic acid)	Meat, liver, fish, Whole-grain products, nuts, peas, beans.	Essential coenzyme in lipid metabolism.	Pellagra characterized by dermatitis, diarrhea and psychological disturbances.
Vitamin B5 (Pantothenic acid)	Kidney, liver, sunflower seeds, broccoli, avocado, edible mushrooms, cereals.	A coenzyme in cellular respiration	Neuromotor disorders, fatigue and muscle cramps

Vitamin B6 (Pyridoxine)	Liver, kidney, fish	Essential coenzyme for normal amino acids metabolism	Dermatitis of the eye, nose and mouth. Retarded growth, kwashiorkor symptoms and nausea.
Vitamin B7 (Biotin)	Yeast, liver, egg yolk, kidneys	Enables synthesis of fatty acids; helps store energy; keeps level of blood sugar stable	Mental depression, muscular pain, dermatitis, fatigue, nausea.
B12 (Cyanocobalamin)	shellfish eggs		Pernicious anemia, memory loss, weakness, personality and mood changes
C (Ascorbic acid)	Tomatoes, citrus fruits, green vegetables,	Needed to make many biological chemicals; acts as antioxidant, aids in detoxification; improves iron absorption	Scurvy (degeneration of skin, teeth, gum, blood vessels), weakness, delayed wound healing, impaired immunity.
Vitamin A (Retinol)	Carrots, spinach, milk, eggs, liver	Needed for good vision, reproduction, and fetal development	Night blindness, dry scaling skin, inability to gain weight. Increased incidence of ear, respiratory, urinary and digestive infections.
Vitamin D (Calciferol)	Liver, fish oils, dairy products, action of sunlight on the skin.	Essential in absorption as well as utilization of calcium and phosphorus, therefore important in formation of teeth and bones	Defective utilization of Calcium and phosphorus by bones leads to rickets in children and osteomalacia in adult.

Vitamin E (Tocopherol)	seed oils, nuts, green leafy vegetables, whole grains, fish	Acts as antioxidant to inactivate free radicals. Is involved in formation of DNA, RNA and red blood cells. It promotes wound healing	Hemolytic anemia,
Vitamin K (Phylloquinone)	Cabbage, liver, green vegetables	Coenzyme essential for synthesis of clotting factors by liver including prothombin.	Delayed clotting time resulting in excessive bleeding.

Many vitamin supplements are available in the market. However, it is always advisable to obtain them from their natural sources by eating food rich in vitamins daily. Possible symptoms of vitamins deficiency are shown by the following pictures:





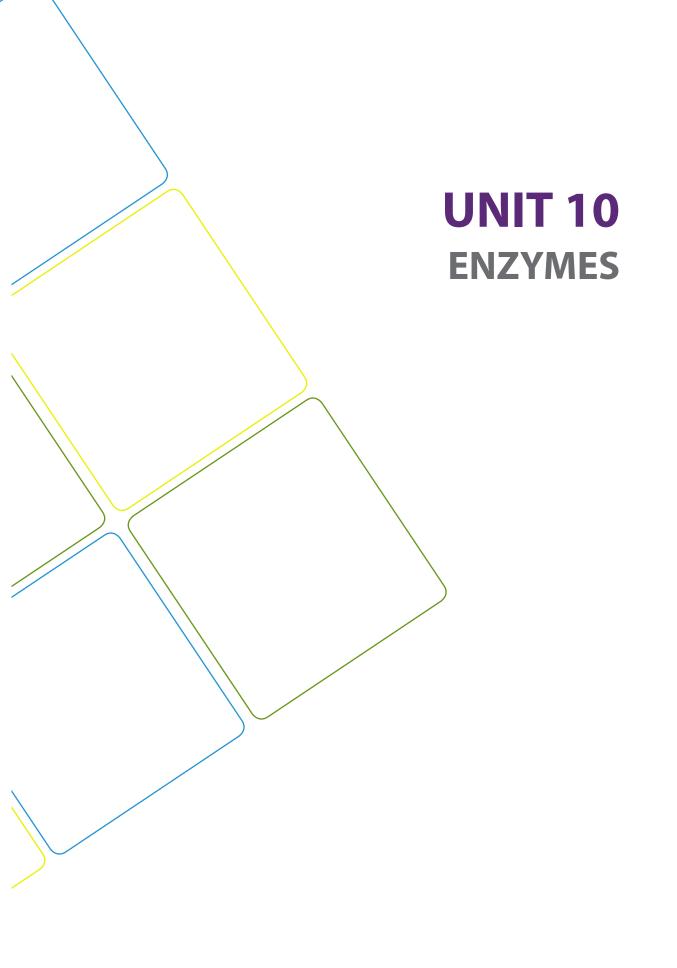
Figures 9. 3 (a) Pellagra caused by niacin (Vitamin B3), (b) Beriberi caused by the lack of thiamin (vitamin B1), (c) **Rickets** due to the vitamin D or calcium deficiency, **(d)Scurvy** due to the lack of vitamin C (ascorbic acid), (e) Goiter caused by iodine deficiency.

#### End of unit assessment 9

- 1. Choose a mineral which is an electrolyte and is found in almost every food. It helps to lower blood pressure.
  - a. Zinc
  - b. Potassium
  - c. Calcium
  - d. Iron
- 2. choose a mineral which helps to make our blood vessels, tendons, and nerves strong.
  - a. Iron
  - b. Magnesium
  - c. Chromium
  - d. Copper
- 3. The following vitamins are part of Niacin and Thiamin minerals
  - a. Vitamins B1
  - b. Vitamins B2
  - c. Vitamins B3
  - d. a and b
- 4. Vitamin C is required for the production and maintenance of:
  - a. Collagen
  - b. Hormone
  - c. Ascorbic Acid
  - d. Red Blood Cells
- 5. Vitamin C deficiency is called:
  - a. Scurvy
  - b. Cold
  - c. Cancer
  - d. Rickets
- 6. Which of the following is a function of Vitamin A in the body?
  - a. Vision, bone and body growth
  - b. Immune defenses, maintenance of body linings and skin
  - c. Normal cell development and reproduction
  - d. All of the above
- 7. Common food sources of Vitamin A are:
  - a. Milk, eggs, butter, cheese, cream, and liver
  - b. White sugar, honey, and sugar cane
  - c. Broccoli, apricots, Cantaloupe, Carrots, Sweet potato, Spinach
  - d. Both a and c

- 8. Which of the following is a function of Vitamin B-12?
  - a. Influences the cells that build bone tissue
  - b. Is essential to the formation of bone
  - c. Helps to maintain acid-base balance
  - d. Maintains the sheaths that surround and protect nerve fibers
- 9. Vitamin B-12 deficiency caused by lack of intrinsic factor is called:
  - a. Pernicious anemia
  - b. Poor circulation of the red blood cells
  - c. Beriberi
  - d. None of the above
- What groups of people need additional Vitamin K? 10.
  - a. Premature newborns
  - b. People who do not have enough bile to absorb fat
  - c. Both A and B
  - d. None of the above answers
- 11. A common function of Thiamin, Riboflavin and Niacin is that:
  - a. They all are used in synthesis of blood clotting proteins
  - b. They all work as a part of a coenzyme used in energy metabolism
  - c. They all help to strengthen blood vessel walls
  - d. They are used to stabilize cell membranes
- The vitamin Folate works together with \_\_\_\_\_ to produce new red blood cells.
  - a. Vitamin D
  - b. Vitamin A
  - c. Vitamin B-12
  - d. None of the above
- 13. Which of the following is a function of Vitamin B-12?
  - a. Red blood cell formation
  - b. Myelin sheath that protects nerve biers
  - c. Vision
  - d. Both A and B
- Vitamin C helps in maintenance and repair of collagen which: 14.
  - a. Forms the base for all connective tissue in the body
  - b. Aids in digestive processes
  - c. Promotes good eyesight
  - d. Prevents PMS symptoms

- Which of the following is not a function of Vitamin D? 15.
  - a. Acts like a hormone
  - b. Stimulates maturation of cells
  - c. Maintains calcium cells
  - d. Builds tissue
- Some food sources of Vitamin D are: 16.
  - a. Fruits and vegetables
  - b. Salmon and egg yolks
  - c. Butter and fortified milk
  - d. Both B and C.
- Humans obtain vitamins from natural sources such as vegetables, fruits, 17. meat, fish and dairy products. What are the two vitamins that are not provided by fruits and vegetables?
- What would you advise someone starting to have symptoms of? 18.
  - a. Scurvy
  - b. Rickets
  - c. Teeth decay
  - d. Heart failure
  - e. Pernicious anemia



# **UNIT 10: ENZYMES**

# **Key Unit Competence**

Describe the mode of action and factors affecting enzymes and their importance for the existence of life

## Learning objectives

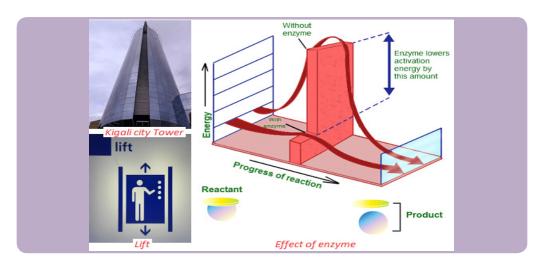
At the end of this unit you be able to:

- Define the term enzyme.
- Explain the criteria of naming enzymes.
- State that enzymes function inside cells and outside cells.
- Explain that enzymes are globular proteins that catalyze metabolic reactions.
- Describe the mode of action of enzymes in terms of the lock and key and the induced fit hypotheses.
- Explain factors affecting enzyme activity.
- Define enzyme technology and its role in industry.
- Investigate the progress of an enzyme-catalyzed reaction by measuring rates of formation of products.
- Investigate the effects of temperature, pH, enzyme and substrate concentration, and inhibitors on enzyme activity.
- Interpret graphs of the effects of reversible and irreversible inhibitors on the rate of enzyme activity.
- Investigate the effect of immobilizing an enzyme in alginate as compared with its activity when free in solution.
- Use a computer to plot graphs of the rate of enzyme controlled reaction. Calculate  $Q_{10}$  of an enzyme controlled reaction.
- Acknowledge that enzymes are essential in speeding up reactions that would be too slow to sustain life.
- Appreciate the importance of planning and carrying out experiments under controlled conditions.
- Understand the roles of enzymes in industry and medicine.

# **Introductory activity**

Discuss in pair the following questions and share with another pair your findings.

- 1. What do you understand by the term enzyme?
- 2. Two individuals want to reach the last floor of Kigali city tower. One climbs up using the ladder but another one uses a lift. What advantage the lift gives over the ladder?
- 3. Why is it easy to digest hot foods than cold ones?



# 10.1. Criteria for naming enzymes

## **Activity 10.1**

You are provided with three groups of enzymes:

	Group A	Group B	Group C
Enzymes	Maltase and lactase	Dehydrogenase and oxidase	Pepsin and renin

Make a research to find out:

- a. specific role of each of the six enzymes mentioned above
- b. criterion followed to name enzymes of group A, B and C respectively

Enzymes are biological catalysts produced by a living organism to control the rate of specific biochemical reactions by lowering the activation energy of reactants

First of all, individual enzymes are named by adding -ase to the name of the substrate with which they react. The enzyme that controls urea decomposition is called urease; those that control protein hydrolyses are known as proteases.

A second way of naming enzymes refers to the enzyme commission number (EC number) which is a numerical classification scheme for enzymes based on the chemical reactions they catalyze. In a system of enzyme nomenclature, every EC number is associated with a recommended name for the respective enzyme catalyzing a specific reaction. They include:

- Oxidoreductases: catalyze redox reactions by the transfer of hydrogen, oxygen or electrons from one molecule to another. Example: Oxidase catalyzes the addition of oxygen to hydrogen to form water.
  - Glucose + oxygen oxidase gluconic acid +water
- **Hydrolase:** catalyzes the hydrolysis of a substrate by the addition of water. Sucrose + water <u>Hydrolase</u> glucose+ fructose
- **Ligases:** catalyze reactions in which new chemical bonds are formed and use ATP as energy source.
  - Amino acid + tRNA <u>Ligase</u> amino acid-tRNA complex.

- Transferases: catalyze group transfer reactions. The transfer occurs from one molecule that will be the donor to another molecule that will be the acceptor. Most of the time, the donor is a cofactor that is charged with the group about to be transferred. Example: Hexokinase used in glycolysis.
- **Lyases:** catalyze reactions where functional groups are added to break double bonds in molecules or the reverse where double bonds are formed by the removal of functional groups. For example: Fructose bisphosphate aldolase used in converting fructose 1, 6-bisphospate to G3P and DHAP by cutting C-C bond.
- **Isomerases:** catalyze reactions that transfer functional groups within a molecule so that isomeric forms are produced. These enzymes allow for structural or geometric changes within a compound. Sometime the inter converstion is carried out by an intramolecular oxidoreduction. In this case, one molecule is both the hydrogen acceptor and donor, so there's no oxidized product. The lack of an oxidized product is the reason this enzyme falls under this classification. The subclasses are created under this category by the type of isomerism. For example: phosphoglucose isomerase for converting glucose 6-phosphate to fructose 6-phosphate by moving chemical group inside the same substrate.

A third way of naming enzymes is by their specific names e.g. trypsin and pepsin are proteases. Pepsin, trypsin, and some other enzymes possess, in addition, the peculiar property known as autocatalysis, which permits them to cause their own formation from an inert precursor called zymogen.

## Self-assessment 10.1

- 1. How to name enzymes?
- 2. What is the role of peptidase?

# 10.2. Characteristics of enzymes

# **Activity 10.2**

Requirement: Three test tubes, match box, about 1g of liver, 1g of sands, 1% H<sub>2</sub>O<sub>2</sub> and MnO<sub>2</sub> powder.

#### **Procedure:**

- Label three test tubes A, B and C respectively.
- Put about 0.1 g of MnO<sub>3</sub> powder in test tube A and 1g of liver in tube B and 0.1g of sand in tube C.
- Pour 5 ml of H<sub>2</sub>O<sub>2</sub> (hydrogen peroxide) in each tube. What do you observe?
- Place a glowing splint in the mouth parts of each test tube. What do you observe?

#### **Questions**

- 1. Explain your observations.
- 2. Write down the chemical equation of the reaction taking place in tube A and B
- 3. Carry out your further research to find out the characteristics of enzymes

Enzymes speed up the rate of metabolic reactions by allowing the reaction to go through a more stable transition state than would normally be the case. As a result, the rate of reaction is increased. In many chemical reactions, the substrate will not be converted to a product unless it is temporarily given some extra energy referred to as activation energy (the minimum energy required the make a reaction take place).

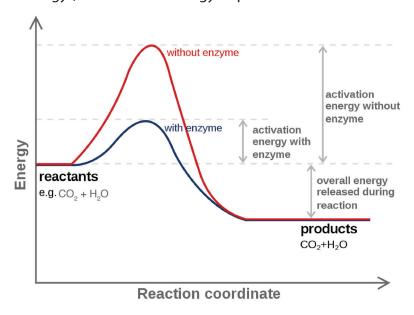


Figure 10.1: Energy profile diagrams for a metabolic reaction

Enzymes speed up the rate of biochemical reactions in the cell but remain unchanged at the end of the reactions. An enzyme has no effect on the relative energy content of products versus reactant. Chemical reactions catalyzed by enzymes are usually reversible e.g. enzyme carbonic anhydrase catalyses both synthesis and breakdown of carbonic acid.

An enzyme provides a reaction surface and a hydrophilic environment for a reaction to take place. This is normally a hollow or cleft in the enzyme which is called the active site, but it is normally hydrophobic in nature rather than hydrophilic.

A very small amount of enzymes is needed to react with a large amount of substrate. The turnover number of an enzyme is the number or reactions an enzyme molecule can catalyse in one second. Enzymes have a high turnover number e.g. the turnover number of catalase is 200,000 i.e. one molecule of enzyme catalase can catalyse the breakdown of about 200,000 molecules of hydrogen peroxide per second into water and oxygen at body temperature.

A cofactor is the best general term to describe the non-protein substances required by an enzyme to function properly. This term covers both organic molecules and metal ions. A co-enzyme is an organic molecule that acts as a cofactor. A prosthetic group is a cofactor that is covalently bound to the enzyme.

#### Self-Assessment 10.2

- 1. State any four properties of enzymes.
- 2. Enzymes have generally high turnover number. What is the significance of the high turnover of enzymes?

# 10.3. Mode of action of enzymes

# **Activity 10.3**

There are two main hypotheses that explain the mode of action of an enzyme on its substrate: the lock and key hypothesis and the induced-fit hypothesis. Carry out a research to find the relevance of each.

Enzymes do not change but substrates are converted into products. A substrate is a molecule upon which an enzyme acts. In the case of a single substrate, the substrate binds with the enzyme active site to form an enzyme-substrate complex. Thereafter the substrate is transformed into one or more products, which are then released from the active site. This process is summarized as follows:

 $E + S \rightarrow ES \rightarrow EP \rightarrow E + P$ 

Whereby: E = enzyme, S = substrate(s), ES = Complex Enzyme-Substrate and P = product(s). There are two main hypotheses explaining the mechanism of enzyme action:

# a. The lock and key hypothesis by Emil Fischer

In this hypothesis the substrate is the key and enzyme is the lock. The active site is exactly complementary to the shape of the substrate as shown below.

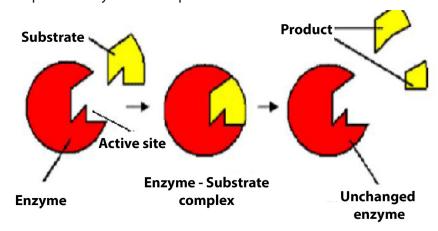


Figure 10.2: Lock and key hypothesis

# b. The induced-fit hypothesis by Daniel Koshland

The induced-fit hypothesis is a modified version of the lock and key hypothesis and is more widely accepted hypothesis. In this hypothesis, the active site is flexible and is not fully complementary with the shape of the substrate. An enzyme collides with

the substrate molecule and binds to the active site. This induces a slight change in the shape of the enzyme making the substrate the fit more precisely. This reduces the potential energy of the substrate and allows the reaction to occur. The products formed move away from the active site and regains its original configuration ready for the next reaction to take place.

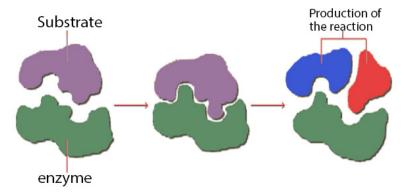


Figure 10.3: Induced fit hypothesis model. Entrance of the substrate into active site cause a change in the configuration of the active site of the enzyme finally allowing the reaction to occur.

#### Self-Assessment 10.3

The key and lock hypothesis is a model that explain the mode of action of an enzyme on the substrate. In the same context, analyse the diagram below and then answer question that follow.



- 1. What does the lock represent?
- 2. What does the key represent?
- 3. Where is the active site?
- 4. Suggest another diagram that can better represent the induced fit hypothesis. Write short notes to explain its functioning.

# 10.4 Factors affecting enzyme action

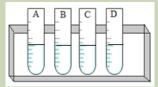
## **Activity 10.4**

#### You will need

Eight test tubes containing 2 cm<sup>3</sup> starch solution, amylase solution, cold water (ice) water bath, iodine solution, HCl solution, and droppers

#### **Procedure:**

1. Label your test tubes A-D as follows:



- 2. Add 1 cm3 of starch solution to each test tube
- 3. Keep tube A and B in cold (ice) and tube C and D in the water bath at 35°C for 5 minutes.
- 4. Add 1 cm3 of 1M HCl on test tubes B and D, then shake the mixture to stir.
- 5. Add 1 cm3 of amylase solution on each test tube. Shake and therefore keep A and B in cold and C and D in water bath for 10 minutes.
- 6. Take a sample from each tube and mix it with one drop of iodine. Use a different tile for each test tube. Record and interpret your observation and then draw a conclusion.

Enzymes activities can be limited by a number of factors such as the temperature, the pH, the concentration of the substrate or the enzyme itself and the presence of inhibitors.

# i. Temperature

At zero temperature, the enzyme cannot work because it is inactivated. At low temperatures, an enzyme-controlled reaction occurs very slowly. The molecules in solution move slowly and take a longer time to bind to active sites. Increasing temperature increases the kinetic energy of the reactants. As the reactant molecules move faster, they increase the number of collisions of molecules to form enzyme-substrate complex.

At optimum temperature, the rate of reaction is at maximum. The enzyme is in active state. The optimum temperature varies with different enzymes. The optimum temperature for enzymes in the human body is about 37°c. When the temperature exceeds the optimum level, the enzyme is denatured.

The effect is irreversible. However, some species are thermophilic that is they work better at high temperatures; others are thermophobic, that is they work better at low temperatures. For example, some thermophilic algae and bacteria can survive in hot springs of 60°c.

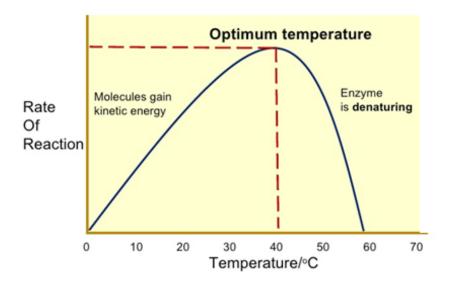


Figure 10.4: Effect of heat on the enzyme controlled reaction

The rate doubles for each  $10^{\circ}\text{C}$  rise in temperature between  $0^{\circ}\text{C}$  and  $40^{\circ}\text{C}$  (figure 10-5). The temperature coefficient  $Q_{10}$  is the number which indicates the effect of rising the temperature by  $10^{\circ}\text{C}$  on the enzyme-controlled reaction. The  $Q_{10}$  is defined as the increase in the rate of a reaction or a physiological process for a  $10^{\circ}\text{C}$  rise in temperature. It is calculated as the ratio between rate of reaction occurring at (X + 10) °C and the rate of reaction at X °C. The  $Q_{10}$  at a given temperature x can be calculated from:

$$Q_{10} = \frac{\text{Rate of reaction at } (x+10)^{0}\text{C}}{\text{Rate of reaction at } x^{0}\text{C}}$$

## **Worked out example**

The rate of an enzyme-controlled reaction has been recorded at different temperatures as follows:

Temperature / °C	0.0	10.0	20.0	30.0	40.0	50.0	60.0	70.0
Rate / mgs-1	0.01	0.1	0.2	0.4	0.8	0.4	0.2	0.02

Calculate the Q<sub>10</sub> of that reaction at 30 °C

#### Solution

Q10 at 30 °C = 
$$\frac{\text{Rate of reaction at (x+10)}^{\circ}\text{C}}{\text{Rate of reaction at x}^{\circ}\text{C}} = \frac{\text{Rate at (30+10)}^{\circ}\text{C}}{\text{Rate at 30}^{\circ}\text{C}} = \frac{\text{Rate at 40}^{\circ}\text{C}}{\text{Rate at 30}^{\circ}\text{C}} = \frac{0.8}{0.4} = x2$$

This means that the rate of the reaction doubles if the temperature is raised from 30°c to 40°c

Be aware that not all enzymes have an optimum temperature of 40°c. Some bacteria and algae living in hot springs (e.g. Amashyuza in Rusizi) are able to tolerate very high temperatures. Enzymes from such organisms are proving useful in various industrial applications because they do not denature up to 70°c

## ii. The pH

Most enzymes are effective only within a narrow pH range. The optimum pH is the pH at which the maximum rate of reaction occurs. Below or above the optimum pH the H<sup>+</sup> or OH<sup>-</sup> ions react with functional groups of amino acids in the enzyme which loses its tertiary structure and become natured.

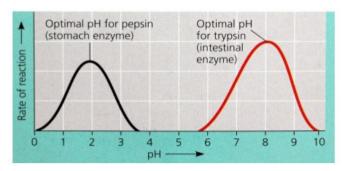


Figure 10.5: Optimum pH depends on nature of enzyme

Different enzymes have different pH optima (look in the table).

**Optimum pH** Enzyme Pepsin and rennin 2.0 Salivary amylase 6.8 Trypsin 7.8 Lipase 9.0

Table 10.1. Optimum pH of some digestive enzymes

# iii. Enzyme concentration

The rate of an enzyme-catalyzed reaction is directly proportional to the concentration of the enzyme if substrates are present in excess concentration and no other factors are limiting.

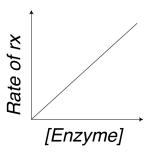


Figure 10.6: Effect of enzyme concentration on the rate of reaction

#### iv. Substrate concentration

At low substrate concentration, the rate of an enzyme reaction increases with increasing substrate concentration. The active site of an enzyme molecule can only bind with a certain number of substrate molecules at a given time. At high substrate concentration, there is saturation of active sites and the velocity of the reaction reaches the maximum rate.

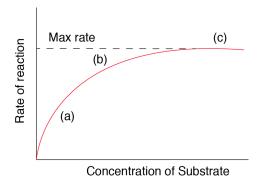


Figure 10.7: Effect of the concentration of substrate on the rate of reaction

#### b. Inhibitors

The inhibitors are chemicals or substances that prevent the action of an enzyme. An inhibitor binds to an enzyme and then decreases or stops its activity. There are three types of inhibitors:

i. Competitive inhibitors are molecules that have the similar shape as the substrate. At high concentration, they compete with the substrate for the active site of the enzyme e.g. O<sub>2</sub> competes with CO<sub>2</sub> in RuBP-carboxylase.

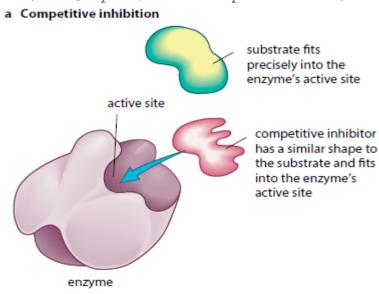


Figure 10.8: Competitive inhibition. An inhibitor has a shape resembling that of the substrate and so relatively compete with each other for the active site depending on the concentration of either substrate or inhibitor

ii. Non-competitive inhibitors are molecules that can be fixed to the other part of enzyme (not to the active site) so that they change the shape of active site,

due to this the substrate cannot bind to the active site of the enzyme.

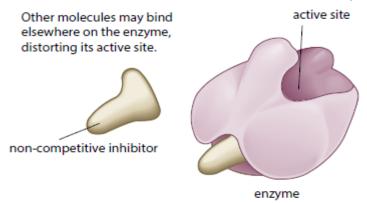


Figure 10.9: Non-competitive inhibition. Anon competitive inhibitor attaches to the site other than the active site and changes the shape of the active site eventually preventing the formation of enzyme substrate complex.

## iii. End product inhibitor, Allosteric inhibitor or Allostery.

This is a chain enzymatic metabolic pathway where the final end product acts as an allosteric reversible inhibitor for the first, the second or the third step in the metabolic pathway. The shape of an allosteric enzyme is altered by the binding of the end product to an allosteric site. This decreases enzymatic activity. By acting as allosteric inhibitors of enzymes in an earlier metabolic pathway, the metabolites can help to regulate metabolism according to the needs of organisms. This is an example of negative feedback.

This often happens when few enzymes are working on a large number of substrate e.g. ATP is an end-product inhibitor of the enzyme PFK (**P**hospho**f**ructo**k**inase) in glycolysis during cell respiration. The end-product inhibitor leads to a negative feedback.

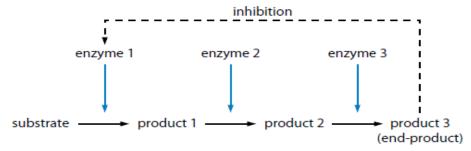


Figure 10.10: Model of end – product inhibition.

The products of enzyme-catalysed reactions are often involved in the feedback control of those enzymes. Glucose-1-phosphate is the product formed from this enzyme-catalysed reaction. As its concentration increases, it increasingly inhibits the enzyme.

## Importance of reversible and irreversible inhibition

The nerve gas **DIPF** (**Dil**sopropyl **P**hospho **F**luoridate) is an irreversible inhibitor. It binds permanently with enzyme acetylcholisterase, altering its shape. The enzyme cannot bind with and break down its substrate acetylcholine (neurotransmitter). Acetylcholine molecules accumulate in the synaptic cleft. Nerve impulses cannot be stopped causing continuous muscle contraction. This leads to convulsions, paralysis and eventually death.

Many pesticides such as organophosphate pesticides act as irreversible enzyme inhibitors. Exposure to pesticides can produce harmful effects to the nervous and muscular systems of humans. Heavy metal ions such as Pb<sup>2+</sup>, Hg<sup>2+</sup>, Ag<sup>+</sup>, As<sup>+</sup> and iodine-containing compounds which combine permanently with sulphydryl groups in the active site or other parts of the enzyme cause inactivation of enzyme. This usually disrupts disulphide bridges and cause denaturation of the enzyme.

#### Self-Assessment 10.4

- 1. What is  $Q_{10}$  of an enzyme controlled reaction?
- 2. You are provided with the table below of the rate of an enzyme controlled reaction.

Temperature / °C	0.0	10.0	20.0	30.0	40.0	50.0	60.0	70.0
Rate / mgs <sup>-1</sup>	0.01	0.1	0.2	0.4	0.8	0.4	0.2	0.02

Calculate the value of Q10 at:

- a. 0° c
- b. 10° c
- c. 50° c
- 3. Explain why thermophile bacteria and algae are useful in some industrial processes
- 4. The diagram below represents a metabolic pathway controlled by enzymes.

$$V \xrightarrow{e_1} W \xrightarrow{e_2} X \xrightarrow{e_3} Y \xrightarrow{e_4} Z$$

- V is a substrate
- W, X and Y are intermediate compounds
- Z is a product
- $e_1$ ,  $e_2$ ,  $e_3$ , and  $e_4$  are enzymes
- a. Name the type of control mechanism which regulates production of compound Z
- b. Explain how an excess of compound Z will inhibit its further production.

# 10.5. Importance of enzymes in living organisms

# Activity10.5

Discuss and present your ideas about the need for different enzymes in living organisms.

Without enzymes, most of the biochemical reactions in living cells at body temperature would occur very slowly or not at all. Enzyme can only catalyze reactions in which the substrate shape fits that of its active site

There are thousands of metabolic reactions that place in the body that require enzymes to speed up their rate of reaction, or will never happen. Enzymes are very specific, so nearly each of these chemical reactions has its own enzyme to increase its rate of reaction. In addition, the organism has several areas that differ from one another by the pH. Therefore, the acid medium requires enzymes that work at low pH while other media are alkaline and require enzymes that work at high pH. In addition to digestion, enzymes are known to catalyze about 4,000 other chemical reactions in your body. For example, enzymes are needed to copy genetic material before your cells divide.

Enzymes are also needed to generate energy molecules called ATP, move fluid and nutrients around the insides of cells and pump waste material out of cells. Most enzymes work best at normal body temperature about at 37° c -- and in an alkaline environment. As such, high fever and over-acidity reduce the effectiveness of most enzymes. Some enzymes need co-factors or co-enzymes to work properly.

Self-Assessment 10.5			
1. Fill the blank with a	ppropriate terms:		
Enzymes are biological produced by			
cells. Enzymes reduce the amount of			
	_ energy required for reaction	ns to occur. They consist of	
globular	with	structure.	
2. Answer the following	g questions:		
a. What is the main	role of enzymes?		
b. What would happ	oen if there are no enzymes ir	n the cell?	

# 10.6. Enzymes technology

# Learning activity 10.6

Enzymes are needed in everyday life. At school you can use salivary amylase to hydrolyse starch. There is industrial technique used to get large amounts of enzyme amylase.

Read through the notes below and answer the following questions below:

- a. State the different processes in which enzyme technology is applied
- b. What is the role of thermophilic bacteria in this process?
- c. How is the effectiveness of an enzyme improved for used in industry?

The market for enzymes is prosperous. The demand keeps on increasing as new applications of enzymes are discovered. Enzymes have been used in cheese-making, in leather industries, and making washing powders.

Microbial cells are still the most sources of industrial enzymes because microorganisms naturally produce enzymes inside their cells known as intracellular enzymes. When microorganisms secrete their enzymes for an action outside their cells, the enzymes are called extracellular enzymes. Microorganisms may have specific genes introduced into their DNA by genetic engineering so that they produce enzymes naturally made by other organisms.

Once enzymes are produced by the microorganisms they are isolated by centrifugation in order to remove the large cell fragments. The enzyme is precipitated from solution by a salt such as  $(NH_4)_2SO_4$  or an alcohol such as  $CH_3$ -CHOH-CH $_3$ . Thereafter the enzyme can be purified by the process known as electrophoresis or column chromatography. The enzyme stability is a key factor in the industrial use of enzymes. The stability of an enzyme is its ability to retain its tertiary structure under a wide range of conditions.

As many industrial processes require high temperatures and extreme pH, it is recommended to use bacteria such as Bacillus subtilis which withstand harsh conditions such as high temperature. Those thermophilic bacteria produce thermostable enzymes that do not denature at high temperature because their optimum temperature between 65 - 75°c.

Some useful enzymes are not thermostable. Such enzymes should be improved by the technique called immobilization i.e. the enzyme is attached to or located within an unreactive support such as nylon that protects it from denaturation.

#### Self-Assessment 10.6

- 1. What is the role of alcohol or ammonium sulphate during the extraction of enzymes?
- 2. Why is thermostability of enzymes so important for many industrial processes?

#### **End of unit assessment 10**

- 1.
- a. What is the meaning of the following terms related to enzyme activity?
  - i. Catalyst
  - ii. Activation energy
  - iii. Lock and key hypothesis
  - iv. Q<sub>10</sub>
- b. Why are there hundreds of different enzymes in a cell?
- c. How do enzymes reduce the activation energy of a reaction?
- 2. Enzyme activity is affected by a number of factors.
  - a. Explain why enzymes work faster at relatively high temperatures
  - b. Describe what happens to the enzyme structure if the temperature is raised above the optimum temperature.
  - c. How are enzymes affected by pH?
  - d. Why do different enzymes have a different optimum pH?
  - e. What is the difference between a reversible and irreversible enzyme inhibitor?
- 3. Some bacteria and algae can survive in boiling water of hot springs. Enzymes from those organisms are used in industrial processes. Why are those enzymes useful?
- 4. The following set of data shows the effect of temperature on the completion time of an enzyme reaction.

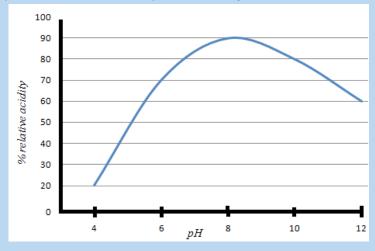
Temperature / °c	0.0	15	25	35	45	55	65
Rate of reaction /	0.00	0.07	0.12	0.25	0.50	0.28	0.00
min-1							

- a. Plot the data on a graph
- b. What is the optimum temperature of this reaction?
- c. Describe the shape of the graph between 10 and 40° c
- d. Calculate the rate of increase between 20 and 30°c.

5. The table below shows the rate of an enzyme reaction at a range of temperature:

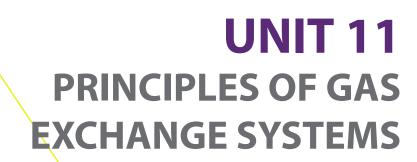
Temperature / °c	Mass lost by reactants /mg	Rate of reaction= loss in mass: temperature
10	5	
20	10	
30	40	
40	80	
50	20	

- a. Fill that table with the values of the rate of reaction and plot a graph of rate at different temperatures (use x-axis for temperature).
- b. Calculate Q<sub>10</sub> at 30°c.
- c. Explain what happen between 20 and 30°c, and between 40 and 50°c.
- 6. The graph below shows the activity of a commercial enzyme alcalase at different pH value. Alcalase is a protease enzyme.



- a. What are the compounds digested by this enzyme?
- b. Describe the change in enzyme activity with pH.
- c. How does this curve compare to the pH curve of a human digestive enzyme such as pepsin?
- 7. Outline how a specific enzyme can be produced from bacteria.





# **UNIT 11: PRINCIPLES OF GAS EXCHANGE SYSTEMS**

## **Key Unit Competence**

Explain the principles of gaseous exchange systems

# Learning objectives

At the end of this unit learners will be able to:

- Explain the relationship between size and surface area to volume ratio.
- Describe how different respiratory surfaces are modified to speed up the diffusion process.
- State the characteristics of gaseous exchange surfaces.
- Describe the effects of tar and carcinogens in tobacco smoke on gas exchange system with reference to lung cancer and Chronic Obstructive Pulmonary Disease (COPD).
- Describe the short-term effects of nicotine and carbon monoxide on the cardiovascular system.
- Observe prepared slides of gaseous exchange surfaces and identify their characteristics.
- Dissect fish gills and observe the surface area for gas exchange.
- Observe mammal's lungs and state their adaptation for gaseous exchange.
- Use internet to make research and deduce the findings
- Appreciate the evolution of gaseous exchange surfaces from simple to complex.

# **Introductory activity**

Kalisa and Uwase wanted to rear tilapia at their home. They bought a nice transparent plastic box. They filled it with 1.5L of clean mineral water, put in some pieces of meat and plant leaves. They finally introduced a living tilapia in the box and covered. After two days they were happy to see their fish swimming. But on the third day, they become sad after finding it dead and yet the food was still in water.

What could have caused the death of the fish?

# 11.1. Relationship between size and surface area to volume

# **Activity 11.1**

- 1. Use Manila paper, scissors, and graduate ruler to create three cubes: 3cm x 3cm, 2cm x 2cm, 1cm x 1cm
  - a. Calculate the surface area, the volume, and the surface area to volume ratio of each cube. What do you conclude from these ratios?
  - b. Compare the surface area to the volume of a spherical alveolus having a radius of 0.001m and that of another animal with a radius of 0.000001m.
- 2. What do you understand by surface area to volume ratio?

The surface area to volume ratio is the relationship between the surface area and the volume of an object. Small or thin objects have a large surface area compared to the volume. For example, the surface area of a sphere is calculated by  $A=4\pi r^2$  and its Volume by  $V=\frac{4\pi r^3}{3}$ .

As the length or radius of the sphere increases, the increase in the surface area is squared (X<sup>2</sup>) and the increase in the volume is cubed (X<sup>3</sup>). The surface area to the volume ratio gets smaller as the cell or animal gets larger. Thus, if the cell grows beyond a certain limit, not enough material will be able to cross the membrane fast enough to accommodate the increased cellular volume.

As a cell grows, its surface area to volume ration decreases. At some point in its growth its surface area to volume ratio becomes so small that its surface area is too small to supply its raw materials to its volume. The cell will reach a size at which substances cannot enter or leave the cell in sufficient time to sustain life. At this point the cell cannot get larger. The volume of the cell will also be so large that the diffusion rate will be too low to distribute necessary substances throughout the cell within a reasonable time. This brings about the need of having a mechanism of ventilation that speeds up the rate of gaseous exchange.

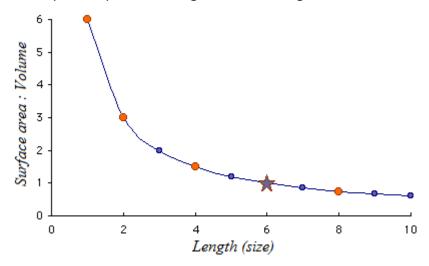


Figure 11.1: How Surface area to Volume ratio varies with the size.

The rate of oxygen consumption by an animal gives a relatively accurate indication of the rate of its metabolic activity. The need of oxygen varies with the activity, the size of the organism, and their health. In general, small mammals need more oxygen than large mammals because:

- Small mammals have a big respiratory surface area to the volume ratio
- Small mammals are too motile than large mammals. Therefore, they need to produce more energy through aerobic respiration
- Small mammals reproduce more rapidly than large mammals.

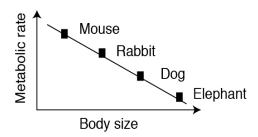


Figure 11.2: Relationship between metabolic rate and size of mammals

A running man needs a double volume of oxygen than a sleeping man and a pregnant woman needs more oxygen than a normal woman.

#### **Self-Assessment 11.1**

Determine the surface area to volume ratio of a sphere having a diameter of 4 mm

# 11.2. Characteristics of gas exchange surfaces

The following are the characteristic features of gaseous exchange surfaces:

**Large surface area:** they should have a large surface area to allow adequate and fast gaseous exchange in order to provide enough oxygen to cells and to get rid of the carbon dioxide that is released.

**Rich supply of blood:** in animals with a transport system, the respiratory surface areas found in the lungs and gills have rich supply in blood capillaries to quickly transport gases to and from the cells. Gases diffuse into the blood and are carried to and from the body cells.

**Thin surface or thin wall:** respiratory surfaces should have thin walls or thin surface area to maximize the diffusion. The alveoli in the lungs have thin squamous epithelium that enables gases to diffuse quickly between the alveoli and blood.

According to Fick's law, the rate of diffusion is proportional to:

# Surface area X Difference in concentration

#### Thickness of membrane

**Moist surfaces area:** to enable gases to dissolve and pass through the solution.

High diffusion deficit / concentration gradient: respiratory surface areas should have a high diffusion deficit / concentration gradient to ensure faster diffusion of respiratory gases.

**Protection against injury and dry out**: lungs and gills are protected by the bones and cartilage and mucus protects them from drying out.

#### Self-Assessment 11.2

- 1. State the features common to all respiratory surfaces in living organisms
- 2. Explain how the following features of a respiratory surface helps gaseous exchange:
- 3. Short diffusion distance
  - a. Protection
  - b. A rich blood supply
  - c. Protection

# 11.3. Modifications of gaseous exchange surfaces to speed up the rate of gaseous exchange in different organims

# **Activity 11.3**

Use appropriate laboratory equipment to extract gills in fish to show the gill filaments. Draw and label to show the parts observed.

#### a. Insects

The spiracles are openings of small tubes running into the insect's trachea system that terminates into small fluid-filled tracheoles in which the gases are dissolved. The fluid is drawn into the muscle tissue during physical exercise, and this increases the surface area of air in contact with the cells.

Ventilation movements of the body during exercise may help this diffusion. The spiracles can be closed by valves and may be surrounded by tiny hairs. The later help to keep humidity around the opening to ensure that there is a lower concentration gradient of water vapour, and so less is lost from the insect by evaporation.

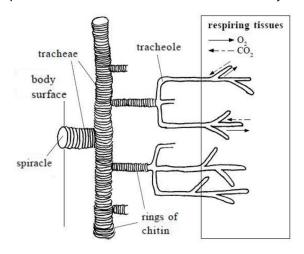


Figure 11.3: Structure of the trachea system showing the tracheoles that are fluid-filled and make contact with individual tissues to exchange gases

# b. Fish and tadpoles

Fish and young amphibians (tadpoles) use gills for the gaseous exchange.

Gills have numerous folds that give them a very large surface area.

- The rows of gill filaments have many protrusions called gill lamellae. These filaments help in the exchange of respiratory gases
- They also have an efficient transport system within the lamellae which maintains the concentration gradient across the lamellae. The arrangement of water flowing passes the gills in the opposite direction to the blood (called counter-current flow) means that they can extract oxygen at 3 times the rate a human can.

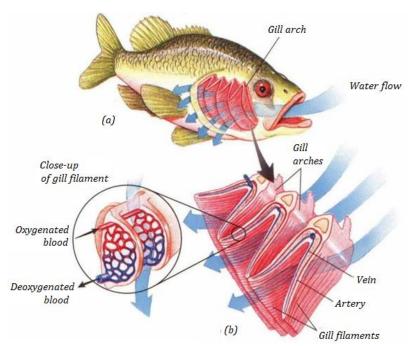


Figure 11.4 Gaseous exchange in bonny fish showing how water and blood flow in different direction over the gills; an adaptation to maintain a high diffusion gradient.

# c. Amphibians, Reptiles, Birds and Mammals

These have **alveoli** in their lungs. Air reaches the alveoli via a system of tubes (trachea, splitting into two bronchi - one for each lung - and numerous bronchioles):

- Numerous alveoli air sacs, providing a massive surface area over which gases can diffuse
- Have a short diffusion distance between the alveolus and the blood because the lining of the lung and the capillary as they are only one cell thick.
- The blood supply is extensive, which means that oxygen is carried away to the cells as soon as it has diffused into the blood.
- Ventilation movements also maintain the concentration gradients because air

is regularly moving in and out of the lungs due to changes in volume and pressure

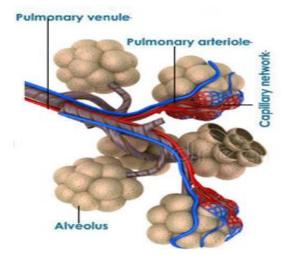


Figure 11.5: Gas exchange surface area displaying alveoli

# Activity11.3

You will need: Lungs of a sheep or pig, newspaper, plastic sheets, dissecting board, sharp scalpel, dissecting needles, scissors, dissecting tray, latex gloves, CPR mouth piece, soap to wash hands and surfaces.





#### **Procedure**

- Place the dissecting board on the newspaper and lay the lungs on the board.
- Use a scalpel to cut the lungs in half in longitudinal section.
- Identify the trachea, right lung, left lung, cartilage rings, bronchus, larynx, alveoli, and bronchiole. You can use a magnifying hand lens to observe structures in the lungs.
- Inflate the lungs by blowing through the CPR (cardio-Pulmonary Resuscitation) mouth piece to see how the lungs expand.
- Feel the slippery inside of trachea, press the lung with your finger and look at cartilaginous rings.
- Remember to wash your hand s with soap as you finish your experiment.
  - 1. Explain what it feels like as you press the lungs with your fingers.

- 2. Look at cartilaginous rings. What function do they serve?
- 3. (a) List four features of respiratory surfaces you can identify from the specimen.
  - (b) Examine the lung and explain how the lungs are suited for efficient gaseous exchange.

Table 11.1: Parts of the human gas exchange system and their respective functions

Part	Functions
Nasal	The hairs in the nostrils trap dust and other small particles.
passages	The mucus that lines the nasal passages traps germs.
Pharynx	Warms and moistens the air entering the lungs as the air passes over blood vessels.
Epiglottis	Stops food and liquids from going into the trachea during swallowing.
Trachea	Provide an open passage for air to enter and leave the lungs.
and	The mucus that lines the inside walls traps dust and germs.
bronchi	Move mucus, which contains dust and germs, to the pharynx, using hair-like structures (cilia) that line the inside walls.
Alveoli	Enable the exchange of gases between the blood passing through the lungs and the air in the lungs
Cartilage	Cartilage is a rigid, but flexible supporting material. Its incomplete rings support the smooth muscle of these tubes, keeping them in an open position. It prevents the trachea from collapsing when the air inside them is lowered when breathing in.
Smooth muscle	As bronchioles are not supported by cartilage, contraction of the rings of smooth muscle around them causes the bronchioles to constrict. In this way, the flow of air to and from alveoli can be restricted and therefore controlled.
Elastic fibres	They are flexible fibres that recoil if stretched when the smooth muscle contracts. As the smooth muscle relaxes, the elastic fibres spring back to their original positions and the bronchiole dilate. They therefore control the air-flow to and from the alveoli.
Goblet cells	They produce the mucus that forms the thin layer over the whole inner surface of these structures. Mucus acts as barrier, preventing pathogens from entering the cells.

Ciliated epithelium

It is a thin layer of epithelial cells that have hair-like organelles called cilia on one surface. These cilia move in a synchronised manner and so transport the dirt-laden mucus which surrounds them upwards towards the pharynx.

#### Self-Assessment 11.3

- 1. List the adaptations of the gills for gaseous exchange
- 2. List the structures through which air passes on its way from the nose to the alveoli.
- 3. Give two reasons why mammals need lungs, rather than exchanging gases through the skin.

# 11.4. Smoking and related risks

# **Activity 11.4**

In groups, make research to find out main health risks related to smoking. Analyse the photographs below and answer questions that follow.



Between the lungs of individuals, A and B, which one is most likely that of the cigarette smoker?

Read the notes below to identify at least three risks related to smoking cigarette.

Cigarette smoking harms nearly every organ of the body, causes many diseases, and reduces the health of smokers in general (Figure 11.6). Quitting smoking lowers the risk for smoking-related diseases and can increase the longevity. Inhaling cigarette smoke is called passive smoking and presents a health hazard to people nearby who inhale it. Of the thousands of chemicals in tobacco smoke three important ones are:

- **Carbon monoxide (CO)**, a poisonous gas form incomplete combustion carbon. CO in tobacco smoke combines easily, but irreversibly, with haemoglobin to form carboxy haemoglobin and therefore reduces oxygen carrying capacity of the blood. This can lead to hypotension and heart failure.
- **Nicotine**, a poisonous alkaloid drug that is addictive. Nicotine in tobacco smoke stimulates the production of the hormone adrenaline by adrenal gland, leading to an increase in the heart rate and raised blood pressure. Nicotine also makes the red blood cell stickier and this leads to high risk of thrombosis and hence of the strokes.
- Tar- is a sticky and brown substance. It appears in tobacco spoke minute droplets.

Tar in tobacco smoke is a mixture of chemicals that enter the respiratory tract. It is an irritant and causes inflammation of the mucous membranes lining the trachea. bronchi and bronchioles, resulting in producing more mucus. Tar also thickens the epithelium and paralyses the cilia on its surface. As a result, cilia cannot remove the mucus secreted by epithelium lining.

# a. Short-term effects of smoking

- Tar causes constriction of finer bronchioles by increasing resistance to the flow
- Tar paralyses the cilia which remove dirt and bacteria; the accumulation of extra material in the air passage can restrict air flow.
- Smoke acts as an irritant; this causes secretion of excess mucus from goblet cells and excess fluid into the airways, making it more difficult for the air to pass through them.
- Mucus accumulating in the alveoli limits the air that they can contain and lengthens the diffusion pathway.
- Coughing of many smokers, way of trying to remove the build-up of mucus from the lungs, can cause damage to the airways and alveoli; scar tissue builds up which again reduces air movement and rates of diffusion
- Infections arise because the cilia no longer remove mucus and pathogens
- Allergens such as pollen also accumulate, leading to further inflammation of the airways, reduced air-flow in and out of the lungs, and possible asthma attacks.

# b. The long-term effects of smoking

- Bronchitis: Bronchitis is inflammation of the lining of the air passages and may be acute or chronic.
- Emphysema: One in five smokers develop the crippling lung disease called emphysema i.e. condition of gradual breakdown of the thin wall of the alveoli leading to sensation of breathlessness as the gaseous exchange reduces.
- Lung cancer: Lung cancer usually starts in the epithelium of the bronchioles and then spreads throughout the lungs as dividing cells cease to respond to the normal signals around them and form unspecialized masses of cells called

tumours. The tar is carcinogen i.e. contains chemicals which cause cancer. The irritation causes thickening of the epithelium by extra cell division and this may trigger the cancer. Almost all people who die from lung cancer are smokers.

# **Self-Assessment 11.4**

Analyze the photograph and share ideas with your group members.



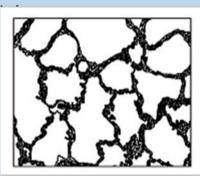
- 1. Between the baby and the parent who will suffer more the effects of tobacco? Give reasons
- 2. Discuss any side effects of smoking.
- 3. Design a sign post to advocate against smoking.

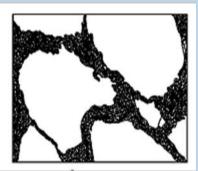
#### End of unit assessment 11

1. Match the terms in Column A with the correct definition in Column B.

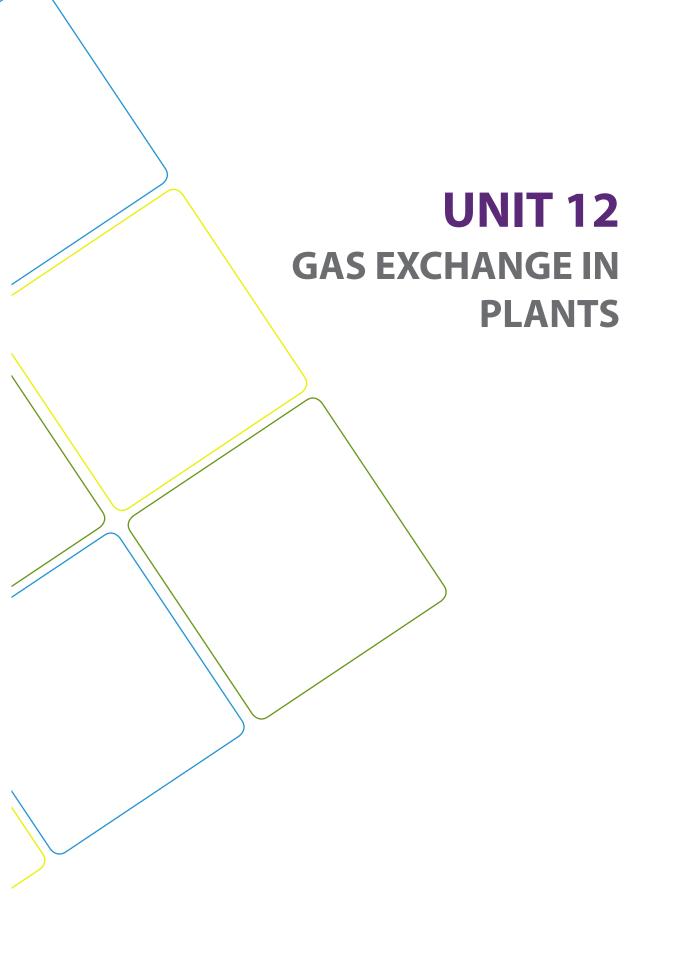
Column A	Column B
(1) Gaseous exchange	A. A slimy substance to keep surfaces moist.
(2) Stoma	B. Taking air into the lungs.
(3) Inhalation	C. The process where food is made in the plant.
(4) Photosynthesis	D. The exchange of gases in living organisms.
(5) Mucus	E. Opening found in leaves.

- 2. Describe how the human lungs serve as good gaseous exchange organs.
- 3. Emphysema is a disease of the lungs. People who smoke cigarettes are more likely to suffer from emphysema. The diagrams show lung tissue from a healthy person and lung tissue from a person with emphysema.





- a. Identify on the figure above by using E (for emphysema) and we (without emphysema) and give a reason for your choice.
- b. Explain how emphysema reduces the amount of oxygen which diffuses into the blood.
- c. What are the features that make the gill of fish an efficient respiratory organ?
- d. Compare respiratory system of fish and with that of a mammal.
- e. Why do people who smoke have high chances of developing lung cancer?
- f. Design a simple model that shows the structure and functioning of gas exchange system in mammals.



# **UNIT 12: GAS EXCHANGE IN PLANTS**

## **Key Unit Competence**

Describe structures of gaseous exchange organs in plants

# **Learning objectives**

By the end of this unit I should be able to:

- Describe the structure of the stoma.
- Explain how stomata, lenticels and breathing roots are adapted to their function.
- Explain the theories of opening and closure of stomata stating limitations of
- Relate the differences between the structures of aquatic and terrestrial leaves to a habitat.
- Draw and label a diagram of stoma as observed under alight microscope.
- Compare gaseous exchange structures of aquatic and terrestrial plants
- Relate the structure and function of aquatic and terrestrial plants
- Defend the relationship between structure and function in aquatic and terrestrial plants

# Introductory activity

Suggest the different parts of a plant that are used in gaseous exchange

# 12.1. Structure of stoma

# **Activity 12.1**

# Requirements

Light microscope, glass slide, cover slip, Commelina zebrine leaves, razor blade, forceps, Pasteur dropper and iodine solution.





#### **Procedure**

- Identify Commelina zebrina or commelina tradescantia plant nearby the school. You can also use any other monocotyledonous plant with succulent leaves.
- Remove a leaf from a plant. Then peel off gently the lower epidermis. It must be thin enough to allow light to pass through
- Smear the epidermis on a slide containing one drop of dilute iodine solution.
- Put on a cover slip and then observe under the lower and medium magnification.
- Repeat the observation in morning hours and in the afternoon hours.

#### **Questions**

- 1. Why should the sample used in the preparation must be transparent?
- 2. Draw and label structures observed under light microscope

Stomata (stoma in singular) are microscopic pores in epidermis of the leaves and stems of terrestrial plants. They function in gas exchange between plant and the atmosphere and in transpiration.

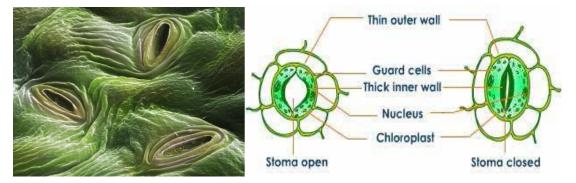
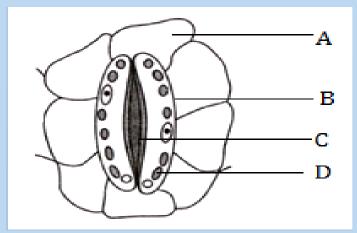


Figure 12.1. Scanned Electron Micrograph (left) and structure of stomata (right).

Each stoma is bordered by two saucer shaped cells called guard cells, which are specialized epidermis cells whose movements control the size of the aperture (pore). Unlike other epidermis cells, guard cells have kidney shape and have many chloroplasts. Their inner cell wall is thick and less elastic while the outer cell wall is thin and more elastic. Guard cells shrink when the plant has too little water. This closes the stomata. When the plant has enough water, the guard cells swell up again. This opens the stomata. In this way, the guard cells enable gaseous exchange. Oxygen in the atmosphere diffuses through the stomata into the air spaces between the cells of the spongy mesophyll tissue while carbon dioxide diffuses through the stomata out to the atmosphere.

#### Self - assessment 12.1

Analyze the diagram below and answer to the following questions



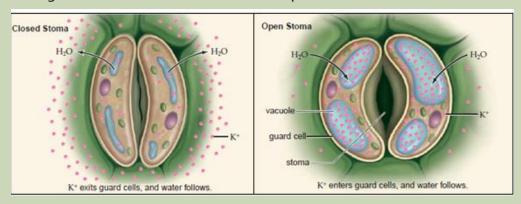
- 1. What title fits better to this diagram:
  - a. an open stoma
  - b. a closed stoma
  - c. of a guard cell and neighbouring cells
  - d. of a stoma and neighbouring cells
- 2. The part labelled C is:
  - a. Vacuole
  - b. Thick inner cell wall
  - c. Chloroplast
  - d. Thin outer cell wall
- 3. The part which better represents the neighbouring cell is:
  - a. Part A
  - b. Part B
  - c. Part C
  - d. Part D
- 4. If the guard cells become more turgid, what is more likely to happen?
  - a. The cells A will swell
  - b. The pore will increase its diameter
  - c. The number of structures C will decrease
  - d. The structure B will stretch in ward
- 5. Which of the following statements is false about that diagram?
  - a. The stoma is closed
  - b. The inner cell wall of guard is thicker than the outer cell wall

- c. There are many chloroplasts in neighbouring cells
- d. The guard cells have many chloroplasts.

# 12.2. Theories used to explain the mechanism of opening and closure of stomata

#### Activity12.2

The diagram below shows a closed and an open stoma.



Carefully analyse the diagrams above and brainstorm your observation. Illustrate how stomata open and close

Many theories have been proposed regarding opening and closing of stomata. The four important theories of stomatal movement are the following:

- Theory of photosynthesis in guard cells
- Theory of starch sugar inter-conversion
- Theory of glycolate metabolism and
- Theory of active potassium pump.

The combined outcome of the four theories shows that in general stomata open during the day (light) and close during the night (dark). But how does this happen?

In light, guard cells are stimulated. They absorb  $K^+$  ions from the neighbouring cells.  $K^+$  ions make the guard cells more permeable to  $CO_2$ . As the guard cells perform photosynthesis, the concentration of  $CO_2$  falls and the pH rises. Elaborated starch therefore splits into malate. The high concentration of malate and the rise of pH in guard cells develop a decrease in water potential. Hence, the guard cells withdraw water from the neighbouring cells and extend backward leaving an open pore in between whereby water is lost by evaporation.

During the night, there is no light to stimulate neither the absorption of  $K^+$  ions nor the photosynthesis. Guard cells undergo cell respiration using photosynthetic products as source of energy (carbohydrates: malate, glucose). Therefore, the concentration of malate decreases making the guard cells hypotonic than neighbouring cells. As guard-cells lose their water content, they shrive and the pore in between closes. Stomatal transpiration ceases.

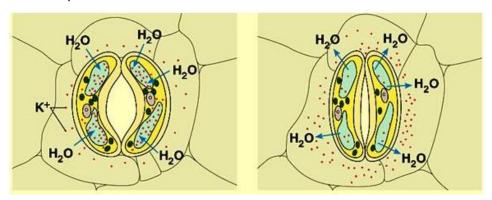


Figure 12.2: Ionic mechanism of opening and closing of stomata

Plant physiologists are certain that stomatal aperture varies as a result of changes in the turgidity of the guard cells. But they are less certain about how these changes are brought about, though the following observations have been made:

- Most stomata open during the day and close at night
- Some stomata show a circadian (daily) rhythm of opening and closing even when kept in constant conditions
- Stomata generally close when a plant suffers water stress, for example, when transpiration exceeds water absorption
- The stomata of some desert plants close during the day and open at night to reduce transpiration

Plants can therefore vary the stomatal aperture. This allows a compromise between the need to conserve water and the need to exchange gases for photosynthesis.

The compensation point is the point when the rate of photosynthesis is equal to the rate of respiration. This means that the  $\mathrm{CO}_2$  released from respiration is equivalent to that which is taken up during photosynthesis. The compensation point is reached as light intensity increases. If the light intensity is increased beyond the compensation point, the rate of photosynthesis increases proportionally until the point of light saturation is reached, beyond which the rate of photosynthesis is no longer affected by light intensity.

For a plant under water stress, its need to conserve water is greater than its need to obtain carbon dioxide for photosynthesis. Under these conditions a plant secretes abscisic acid (ABA). This is a chemical messenger which causes stomata to close. It is thought that ABA triggers a metabolic pump which actively secretes potassium ions out of guard cells, causing the cells to lose water and become flaccid.

#### Self-assessment 12.2

- 1. According to the ionic theory of opening and closing stoma, what is the role of potassium ions in the guard cell?
- 2. What would happen to guard cells if the concentration of malate doubled?
- 3. What is meant by compensation point?

# 12.3. Structural adaptations and function of stomata, lenticels and breathing roots.

## **Activity 12.3**

Observe the adaptations of these plants for gas exchange.







(a) Mangrove

(b) Breathing roots

(c) Pneumatophores

- 1. How is each of these plants adapted for gas exchange?
- 2. Read through the notes that follow and describe any two adaptations for gas exchange

The exchange of atmospheric gases is essential to photosynthesis and cell respiration. In plants, the gas exchange takes place through stomata, breathing roots, lenticels and cuticles. Most stomata are on the lower epidermis of the leaves on plants. Unlike other plant epidermal cells, the guard cells contain chlorophyll to carry out photosynthesis. This allows the cells to expand or contract to open or close the stomata.

Guard cells swells, through the process of osmosis, to allow opening of the stomata for CO<sub>2</sub> to enter and excess O<sub>2</sub> and H<sub>2</sub>O to leave, and they shrink in order to force the stomata shut either partially or completely to prevent dehydration. The number of stomata on the epidermal surface depends on the ecology of plants. Usually, plants on wet climate have fast growth and a high concentration of stomata. Plants on dry weather have lower rates of photosynthesis, lower growth and lower concentrations of stomata.

Xerophytic plants or xerophytes are plants that inhabit arid regions (desert). They have the following adaptations:

- Stomata sunken in grooves and reduced in number
- Ability to fix CO<sub>2</sub> at night, so the stomata are closed during the day.

- Epidermis infolded to reduce the surface area
- Leaves reduced to scales or thorns to reduce the surface area for transpiration
   Hydrophytes or water plants are plants that grow submerged or partially submerged
   in water. To thrive in this environment, hydrophytes have the following features
  - developed stomata on large upper surface of their leaves (rather than underside) making gas exchange more efficient.
  - large air space to facilitate evaporation from the mesophyll.
  - little or no lignified supporting tissues on the submerged parts.
  - poorly developed transport tissue,
  - stems and leaves have little or no lower cuticle but large continuous air spaces, forming reservoir of oxygen and CO<sub>2</sub> which also provides buoyancy to the plant tissues when submerged.



Figure 12.3: Water lily is a best example of hydrophytes

A halophyte is a plant that grows in water of high salinity and they come into contact with saline water through its roots or by salt spray, such as in saline semi-deserts, mangrove swamps, and marshes. Halophytes are adapted in the flowing ways:

- Store water in succulent tissues which have high concentration of salt. They
  can thus take up water from the sea water by osmosis.
- Extensive air spaces throughout the stem and roots making air available to all cells, and giving buoyancy to the stem and leaves at highest tides.
- They develop breathing roots called pneumatophores which grow upward and protrude out of the ground. e.g. mangrove tress.

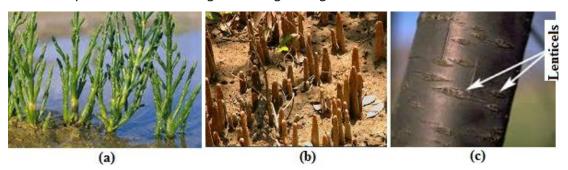


Figure 12.4: Adaptations for gas exchange in plants: (a) Salicornia europaea in a highly saline environment (b) Pneumatophores above the ground (c) Lenticels on stems

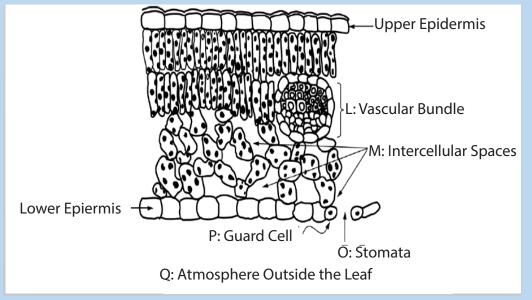
#### Self-assessment 12.3

What features are common to plants living in desert and saline soils?

#### End of unit assessment 12

Section A: Objective questions

- 1. You are provided with the diagram below. Analyze it and then choose the correct answer. Transpiration in the leaf depends on the transport of potassium ions into:
  - a. Into O
  - b. Into P
  - c. From M to L
  - d. From M to O
  - e. From P to L.

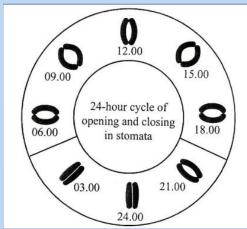


- 2. The theory that says than during the light time, potassium pumps open and this brings about diffusion of CO2 from the atmosphere to the guard cells for photosynthesis is called:
  - a. Theory of photosynthesis in guard cells
  - b. Theory of starch sugar inter-conversion
  - c. Theory of glycolate metabolism
  - d. Theory of active Potassium Pump.

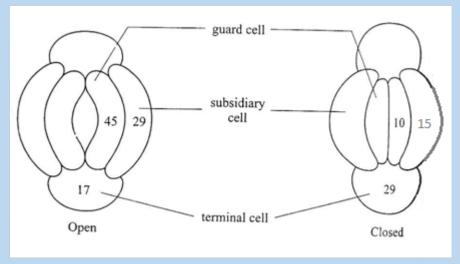
- 3. What is the main difference between the guard cells and the other epidermal cells?
  - a. Guard cells have chloroplast while the remaining epidermal cells have no chloroplast
  - b. Guard cells have oval shape while other cells have cubic shape
  - c. Guard cells are beneath the spongy mesophyll
  - d. Guard cells are covered by a transparent cuticle
- 4. Water lily is:
  - a. Xerophytes
  - b. Halophyte
  - c. Hydrophyte
  - d. Heleophyte
- 5. Mangroves are plants adapted to estuaries or marine region with high salinity. What statement does not describe the adaptations of mangroves?
  - a. The presence of lenticels that help in gas exchange and evaporation
  - b. Presence of large number of stomata on the upper side of the leaves
  - c. The presence of pneumatophores which are breathing roots
  - d. Presence of succulent tissues that have high concentration of salt

#### **Section B**

- 6. Explain how gaseous exchange occurs in the leaf.
- 7. How does gaseous exchange occur in woody stems?
- 8. Describe how roots get oxygen.
- 9. a. Draw a labelled diagram of a stoma
  - b. Draw arrows on the diagram to show how gaseous exchange occurs.
- 10. The drawing shows a 24-hour cycle for the opening and closing of stomata from the same plant.



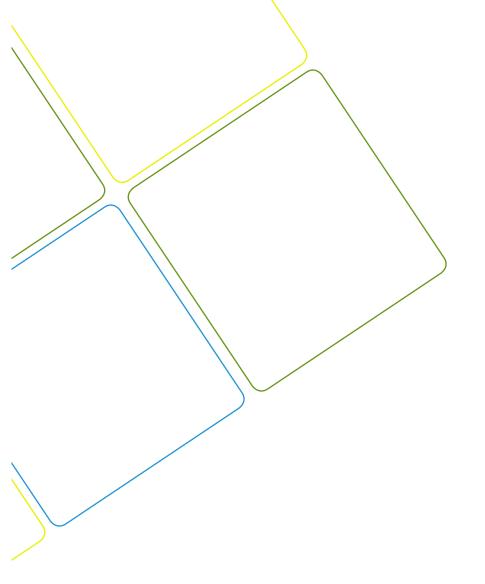
- a. Explain how this cycle of opening and closing of stomata is advantageous to the plant.
- b. The diagram shows the potassium (K<sup>+</sup>) concentrations in the cells around open and closed stomata in commelina. The concentrations are in arbitrary units.



- i. Explain how the movement of K<sup>+</sup> ions accounts for the opening of stomata.
- ii. Explain how K<sup>+</sup> ions are moved against a concentration gradient.



# GROWTH AND DEVELOPMENT IN PLANTS AND ANIMALS



# **UNIT 13: GROWTH AND DEVELOPMENT IN PLANTS** AND ANIMALS.

# **Key Unit Competence**

Account for the processes of growth and development in plants and animals

# **Learning Objectives**

By the end of this unit, I should be able to:

- Describe dormancy as a state of inactivity to absolute minimum due to the morphological and physiological state of a plant structure
- Explain how dormancy is maintained and broken
- State the conditions required for germination
- Outline the role of enzymes in the process of germination
- State types of plant growth hormones and their functions
- Identify the hypocotyl and coleoptile in a germinating seed
- Describe the stages and types of germination
- Recognize that a meristem is a growing point of the plant and the main meristematic regions of a tree
- Describe current views about photoperiodic control of flowering
- Observe structures of endospermic and non-endospermic seeds
- Demonstrate how fruit and seed dispersal takes place
- Demonstrate hypogeal and epigeal germination
- Carry out an investigation to distinguish between primary and secondary growth
- Appreciate the importance of fruit and seed dormancy and germination in the life cycle of plants
- Carry out an experiment on the development of eggs at different temperatures
- Describe the process of metamorphosis in arthropods and amphibians
- Distinguish the various stages of development in frogs
- Analyse complete and incomplete metamorphosis
- Compare growth patterns in arthropods and vertebrates
- Appreciate the demands of the terrestrial environment to the adaptation of amphibians

# **Introductory activity**

Suggest morphological differences by which different organisms develop and grow to maturity.

# 13.1. Fruit, seed and bud dormancy

# **Activity 13.1**

- Put dry bean seeds and maize grains in boiling water for 10 minutes.
- Use nails to make a longitudinal section of the bean and maize.
- Compare the two sections.
- From your point of view, do you think that all plants' organs are always active? Justify your point of view.
- Produce a picture showing how plant organ (seed and bud) behaves in relation to oxygen, temperature and water.

A seed is a plant organ that develops from the fertilized ovule. As the seed embryo develops from the zygote, the seed makes a food reserve of macronutrients including carbohydrates, proteins and lipids. The amount of reserved food type depends on the plant species. Legumes like peanuts and soybeans store proteins as well as fats while beans store more starch and proteins. A seed consists of a plant embryo surrounded by the food supply in cotyledon or endosperm and a protective coat called seed-coat or testa. The plant embryo is composed of the radicle or embryonic root, the hypocotyl which is the embryonic stem and the plumule, the embryonic leaves.

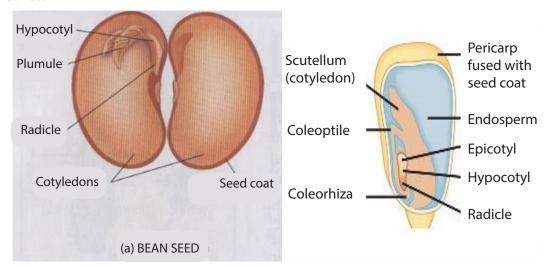


Figure 13. 1: (a) Dicot seed structure (b) Monocot seed structure

Dormancy is a period of cells' inactivity due to a very low metabolism to prevent growth when environmental conditions are unfavorable. The dormancy can be for fruit, bud or seed.

# a. Bud dormancy

Bud dormancy is a suspension of most physiological activities and growth that can be reactivated. It may be a response to environmental conditions such as seasonality or extreme heat, drought, or cold. The exit from bud dormancy is marked by the resumed growth of the bud. Bud dormancy may proceed to dormancy of the whole plant.

# b. Seed dormancy

Seeds exhibit dormancy to avoid growth during unfavorable conditions. During the last stages of its maturation, the seed dehydrates until it gets dry. The embryo which is surrounded by a food supply (cotyledons, endosperm or both), enters dormancy. Most seeds are enough durable that can last a year or two until conditions are favorable for germinating. However, the length of time a dormant seed remain viable and capable of germinating varies from a few days to years depending on the plant species and environmental conditions. This may justify the reason why after a bushfire or other environmental disruption, vegetation reappears as their seeds have accumulated and stayed for longer in the soil. Many plants are easily grown from seeds. Although its embryo is alive, a dormant seed cannot germinate until it is exposed to certain environmental conditions to break the dormancy. Thus, it germinates and starts to grow.

# **Conditions needed for seed germination**

Germination is the process by which a seedling develops from a seed embryo. There are requirements for germination to be successful. Environmental conditions such as water, oxygen, and temperature trigger seed germination. For a seed to germinate, it must be found in place where all these conditions are available. If one condition is not available, germination becomes impossible. These conditions include:

#### a. Water

For a seed to germinate, it requires water. Mature seeds are very dry and must absorb water by imbibition to germinate. Water softens the seed coat for embryo sprouting and provides a medium for reactions during hydrolysis. Enzymes breakdown macronutrients such as starch, proteins and fats stored in the cotyledons and the endosperm to simple sugars such as glucose, amino acids which provides energy for the embryo to grow.

# b. Oxygen

This is needed for needed for cellular respiration by oxidizing glucose to liberate ATP to provide energy for embryo development.

# c. Temperature

Seeds germinate only if exposed to their optimum temperature varying between 5 to 40 °c depending on the seed species. Enzymes are sensitive to temperature because enzymes need a favorable temperature to work efficiently. Burying seeds too deeply in the soil prevents them from germinating as they are cut off from temperature and air.

#### Self-assessment 13.1

- 1. Define the term dormancy?
- 2. Explain how dormancy is maintained and broken?
- 3. Suggest the advantage of dormancy in plants?
- 4. What are conditions needed for seed germination?
- 5. Explain the role of enzymes during the process of seed germination.
- 6. Draw and label endospermic and non-endospermic seeds

# 13.2. Types and stages of germination.

# Learning activity 13.2

Conduct the experiment below to compare the two types of germination.

## Requirements

Two bean seeds or soybean seeds, two sorghum or maize grains, water and two plastic devices/containers

#### **Procedure**

- Fill each container with soil
- Put two grains of maize in the container labeled A and two seeds of bean in the container labeled B
- Pour some water in each container simply to moisture the soil.
- Put both containers A and B in a secured place.
- Record your observations from day five to day twenty one with emphasis to developmental changes.

In conducive environment, a seed can germinate. Germination involves three main stages: imbibition, radicle sprouting and plumule sprouting.

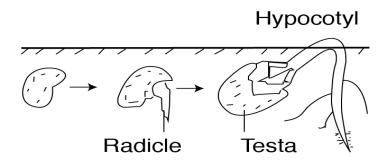


Figure 13. 2: Stages of seed germination\

The dormant seed contains very little water, when placed in moist conditions, it absorbs water by osmosis. The intake of water by a dry material is called imbibition. As the seed absorbs water and oxygen, it swells, the embryo grows and the seed-coat cracks, and the radicle also called primary root emerges downward. This is seed coat rupture and radicle sprouting. The primary root is the first organ to appear during embryo development. The plumule cells develop and the first leaf forms upwards in the process known as plumule sprouting.

A germinating seed shows the primary root from which secondary roots start to develop and the primary leaf which is surrounded by a protective sheath called coleoptile. The Hypocotyl is the stem below the plumule while the epicotyl is the stem above the plumule.

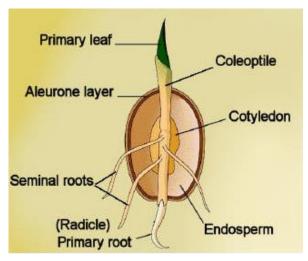


Figure 13.3: Coleoptile

# 13.2.1. Types of seed germination

There are two types of seed germination: hypogeal and epigeal seed germination.

# a. Epigeal germination

Epigeal germination is typical to non-endospermic seeds like beans, soybeans and coffee seeds. In this type of germination, the cotyledons come above the soil surface into air and light, due to rapid growth and elongation of hypocotyl. The cotyledons are green and make food to be used by stem during growing season. They finally dry off and seedlings become independent. The example of epigeal germination is like the one found in bean seeds.

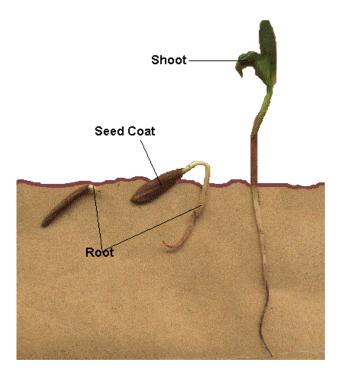


Figure 13.4: Epigeal germination

# b. Hypogeal germination

In this type of germination, the cotyledons remain in soil or just above the surface. Here the epicotyl elongates, pushing plumule upwards. Cotyledons do not turn green and gradually dry up and fall off. An example of this type of germination is found in pea, mango, and groundnut seeds.

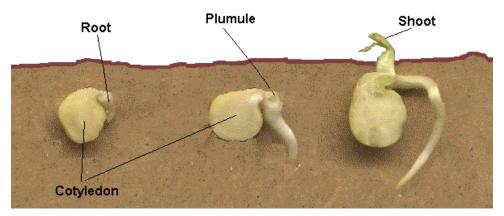


Figure 13.5: Hypogeal germination

#### Self-assessment 13.2

- 1. Use diagrams to demonstrate and distinguish the types of seed germination.
- 2. Describe the three stages of seed germination.

# 13.3. Primary and secondary growth

### Learning activity 13.3

Move around the school garden or anywhere in your surroundings, then collect two plant species in which one is a monocotyledon and another is a dicotyledon.

- 1. By examining their physical characteristics, describe similarities and differences between collected plants.
- 2. Predict the reason why all plants grow in length.
- 3. From your observation, suggest/ predict the reasons justifying why monocotyledons do not grow in width while dicotyledons do.

Growth is a permanent increase in the size of an organism or of some parts of it. It is brought by cell division and the assimilation of new material within the cells which result from the division and the cell expansion which follows. Cell expansion is particularly noticeable in plants, where rapid enlargement can occur as a result of water taken up by osmosis. If the organism achieves its mature size, it starts development, the formation of new structures or organs to perform specific functions. That is the production of reproductive organs and locomotive or protective organs. It is controlled by cell differentiation. Growth is either primary or secondary.

### a. Primary growth

Primary growth consists of the increase in length and formation of primary plant organs including roots, stem, leaves, flowers, and fruits. It occurs in most herbaceous plants. The roots elongate to penetrate the soil, and shoots elongate to reach the sunlight. Primary growth is controlled by apical meristems. Meristematic cells divide by mitosis. Some daughter cells absorb water and nutrients. As the cell takes in water the cell walls stretches, the cell elongates and slightly enlarges. After this growth by elongation, cells differentiate and specialize for specific functions. The cells formed from apical meristems do not expand laterally and this limits their size. The herbaceous plants exhibiting such growth tend to be short lived. They are called annual plants because most of them do not live for more than one growing season (a year) after which they enter dormancy and survive as seeds.

# b. Secondary growth

It consists of getting wider or thicker, and occurs in roots and stems of perennial woody plants, all trees and shrubs. Woody plants grow taller than herbaceous plants and they live longer, more than two years some plants while some others may live for and over 30 years.

Secondary growth is controlled by lateral meristems: vascular cambium and cork cambium. Vascular cambium is located between primary phloem and primary xylem. Vascular cambium cells divide by mitosis. Some cells remain meristematic while other cells expand sideways and differentiate. They form the secondary phloem outwards and the secondary xylem inwards. As the secondary growth continues,

the outermost phloem cells die. Their death causes no matter to the whole plant as dead cells are continually replaced by new ones.

The secondary xylem cells are strengthened by the accumulation of lignin and cellulose. They form a wood also called secondary xylem providing the mechanical support to very taller trees. The cork cambium situated between the epidermis and secondary phloem produces the cork cells by mitosis. The cork cells are pushed toward the epidermis and accumulate a waxy substance called suberin making the cork waterproof. The cork gradually replaces the epidermis. Like the epidermis, the cork protects the plant from dehydration and infection. The expansion of the internal tissues results into the continual cork shedding. Because of meristematic cells that continuously divide, perennial plants have unlimited growth.

#### Self-assessment 13.3

- 1. In the table list the differences between primary growth and secondary growth.
- 2. Describe briefly what the wood is and its major function.
- 3. What is the name given to a substance that makes the cork to resist plant dehydration?
- 4. Identify the importance of apical and lateral meristems in plant growth.

# 13.4. Determination of growth

# **Activity 13.4**

In the learning activity 13.1 you have grown two types of seeds (bean and maize). Based on that experiment do the following to investigate the primary growth in a seedling.

- Use a centimeter ruler, measure the height of each plant once each five days.
- Use a thread and a centimeter ruler to measure the width.
- Record your measurement in a tabular form as shown below.

Plant type	Plant height				
	Day 5	Day 10	Day 15	Day 20	
Bean					
Maize					

- 1. On the basis of the period of time indicated in the above table, count the number of leaves. What do you notice?
- 2. Among the two given plants, predict the one with an increased volume of protoplasm and dry mass.

The growth rate of an organism is measured by recording the variation in length, in width and in mass through a period of time. The aspects that can allow the measurement of the growth of a plant are the following:

- The increase in the dry mass
- The increase in the volume of protoplasm
- The increase in the length
- The increase in the thickness
- The ability to reproduce.

Experiments show that the growth rate is faster in young plants and starts to decrease as the plant gets older.

#### Self-assessment

- 1. A sign is hammered into a tree 2 m from the tree's base. If the tree is 10 m tall and elongates 1m each year, how high will the sign be after 10 years? A mark is hammered into a tree 2 m from the tree's base. If the tree is 10 m tall and elongates 1m each year, how high will the mark be after 10 years
- 2. What features would enable you to conclude that this organism has grown?
- 3. Would you expect a tropical tree to have distinct growth rings? Why or why
- 4. If a complete ring of bark is removed around a tree trunk (a process called girdling), the tree usually dies. Explain why?

# 13.5. Phytohormones

# **Activity 13.5**

The immature avocado fruits often fall down without being mature as well as sunflower plant which grows faster when young flourish in the opposite direction to the sun. Have you ever think and wonder about this? From your point of view, brainstorm the cause of the situations described above. Use internet and textbooks, to outline roles played by cytokinines, Gibberellin and ethylene plant hormones.

Plant growth is influenced by both external and internal factors. External factors include;

- light,
- moisture
- Temperature, while internal factors include phytohormones or plant growth

Internal factors are chemical substances that are produced in the plants or artificially synthetized for regulating plant growth. Those substances are capable of accelerating, inhibiting or modifying growth in plants. If two hormones work together to reinforce an effect, they are synergist but if the presence of one

hormone prevents the action of another they are antagonist.

The plant hormones include five major groups:

#### a. Auxins

It is produced in growing regions of plant such as shoots, tips, and young leaves, and developing fruits. The most known auxin is Indol Acetic Acid (IAA). Artificially produced auxins are widely used to:

- Promote the cell elongation in the region behind the apex of the stem
- Promote root formation on stem and leaf cuttings
- Increase number of fruit
- Prevent dropping of fruit
- Prevent sprouting of stored potatoes and onions

### b. Gibberellin or gibberellic acid (GA)

Produced in all parts of plants, especially in immature seeds. Gibberellin has different functions:

- It promotes the parthenocarpy (formation of fruits without fertilization).
- It breaks down the bud dormancy
- It promotes the seed germination.

### c. Cytokinins

They are produced in developing roots, fruits and seeds, cytokinin and have the following roles:

- Promote cell division
- Promote the growth of lateral buds
- Promote the growth of fruits
- Are used to delay aging and death (senescence).
- Work with gibberellins to break down the bud dormancy and to promote the seed germination

# d. Abscissic acid (ABA)

It is produced in leaves:

- Promotes the abscission i.e. falling of some organs of the plants
- Promotes the bud and seed dormancy
- Inhibits the stem growth during the stress
- Promotes stomatal closure
- Inhibits other hormones blocking thus the growth.

# e. Ethene or ethylene (C<sub>2</sub>H<sub>4</sub>)

It is produced in fruits, flowers, leaves and roots and:

- Promotes ripening of fruit
- Promotes flowering in mangoes and pineapples
- Promotes abscission (detachment of leaves).

### **Commercial application of synthetic phytohormones**

Artificial auxins are widely used. For example, 2,4-D or 2,4-dichlorophenoxyacetic acid and MCPA 4-chloro-2-methylphenoxyacetic acid are used as weeds killer (selective herbicides). Synthetic auxins used in right concentration, cause excessive growth and very rapid metabolism of broad-leaved dicotyledons herbaceous plants that are weeds in cereals. As weeds grow faster, they soon die increasing thus the cereals crop yield.

Napthaleneacetic acid (NAA) or rooting powder is another kind of auxin sprayed on stem cuttings for stimulating the development of adventitious root. This is very important in vegetative propagation of plants of economic value. Synthetic gibberellin helps to increase fruit crop yield because when sprayed on non-fertilized flowers, they promote parthenocarpy (fruit formation without fertilization). It is mostly used in production of seedless grapes such as seedless tomatoes or citrus. Synthetic cytokinin is sprayed on cut flowers, fruits and vegetables to keep them fresh and extend shelf-life.

#### Self-assessment 13.5

- 1. What are the plants hormones?
- 2. Describe the role played by each plant hormone.
- 3. Explain why some plants develop lateral shoots when the apex is cutoff.

# 13.6. Plant movements and photoperiodism

#### 13.6.1. Plant movements

# **Activity 13.6.1**

Take the container with the bean seedling in the classroom nearby the window and observe the changes within one week.

Like animals, plants move as response to changes in their environment (internal or external changes). Plant movements are grouped into two categories: tropisms and nastic movements.

# 1. Tropism

A tropism is a movement of parts of a plant in response to external stimulus. The movement is always a growth movement. External stimuli cause changes in the direction of the plant's growth, such as bending, turning or curving. Tropic responses are described as positive or negative depending on whether growth is towards or away from the source of the stimulus respectively. According to the type of stimulus, tropisms are classified as follow:

**Table 13. 1: Classification of tropisms** 

Stimulus	Tropism
Light	Phototropism
Gravity	Gravitropism or geotropism
Chemical	Chemotropism
Water	Hydrotropism
Touch	Thigmotropism

#### a. Phototropism

Phototropism is a directional growth depending on the direction of the light source. Growth towards a light source is a positive phototropism, while growth away from light is called negative phototropism. It is believed that light destroys auxin where it strikes the stem, causing an imbalance in which the side of the stem that receives less light has more auxin. This causes the plant to have elongated cells on the farthest side from the light. Because more auxin is present, the cells on the darker side are able to elongate more than the cells on the lighted side, causing the plant to bend toward the light.

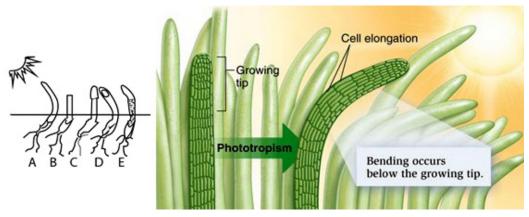


Figure 13. 6: Phototropism

Phototropism is caused by unequal distribution of auxins in the plant stem. There is more auxin on the side of the stem away from the light. This results in cell elongation, but only on that side. As the cells grow, the stem bends toward light.

## b. Gravitropism

When a seed germinates, the young root turns downward regardless of the way in which the seed is planted. This bending, known as positive geotropism, enables a plant to anchor itself in the soil. The young stem, which turns upward away from the earth, is said to be negatively geotropic. The gravity causes auxins to concentrate on the down side than on the upper side. High concentration of auxins on the down side of the stem promotes elongation of its cells. Therefore, the stem grows and bends upward. But high concentration of auxins on the down side of the stem inhibits cell elongation; therefore, the root grows and bends downward, that is why the root grows shorter than the stem.

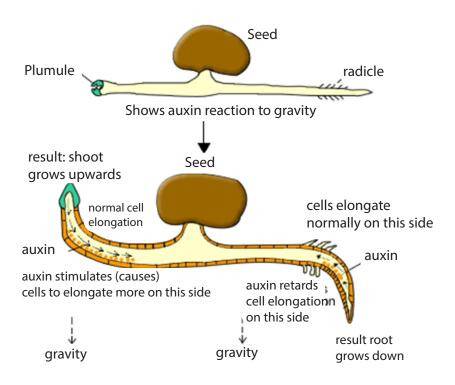


Figure 13.7: Gravitropism

## Use of clinostat in tropism

A clinostat is a piece of laboratory equipment with a turntable that allows a plant placed on it to be exposed to a stimulus such as light or gravity equally on all sides. As the clinostat is turning, all parts of the plant receive the stimulus equally and therefore the plant will not bend but will grow straight away. In otherwise the clinostat is used to minimize the effect of the direction of a stimulus.

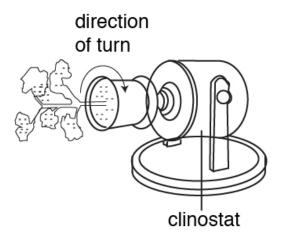


Figure 13.8: Clinostat

### c. Chemotropism

Chemotropism is a growth of a plant or plants parts, navigated by chemical stimulus from outside of the organism. An example of chemotropic movement can be seen during the growth of the pollen tube, where growth is always towards the ovules. Fertilization of flowers by pollen is achieved because the ovary releases chemicals that produce a positive chemotropic response from the developing pollen tube. It is different from chemotaxis. The major difference being that chemotropism is related to growth, while chemotaxis is related to locomotion. For example, the movement of antherozoids (sperm) in ferns, swim toward the chemicals produced by archegonia.

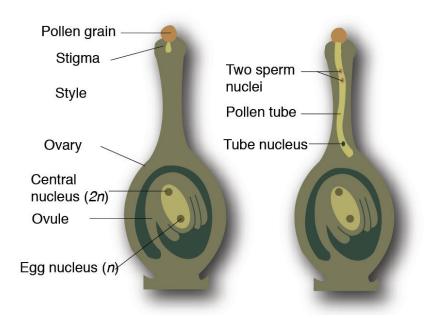


Figure 13. 9: Pollen tube grows towards the ovule

## d. Thigmotropism

Thigmotropism is a movement in which an organism grows in response to touch or contact stimuli. Usually thigmotropism occurs when plants grow around a surface, such as a wall, pot, or trellis. Climbing plants, such as vines, develop tendrils that coil around supporting objects. Touched cells produce auxin and transport it to untouched cells. Some untouched cells will then elongate faster so cell growth bends around the object. Some seedlings also inhibit triple response, caused by pulses of ethylene which cause the stem to thicken (grow slower and stronger) and curve to start growing horizontally.

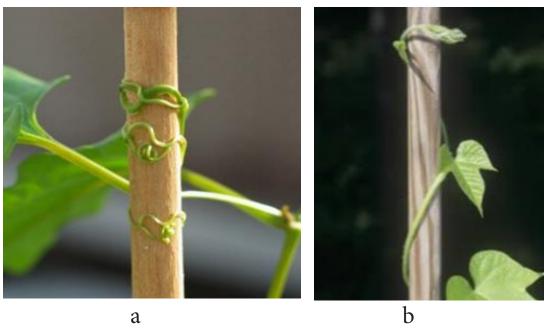


Figure 13.10: Climbing plants grow around the support using tendrils (a) or the whole stem (b)

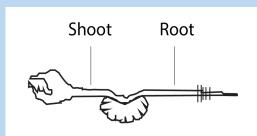
#### 2. Nastic movement

Nastic movements are non-directional responses to stimuli such as temperature, humidity, light and irradiance. An example of such a response is the opening and closing of flowers known as photonastic response and the opening and closing of carnivorous plants known as thigmonastic response. The folding and unfolding of some sensitive plants like Mimosa pudica when touched, is a nastic response which protects them from insect damage or water loss during winds. They are named with the suffix "-nasty" and have prefixes that depend on the stimuli:

- Photonasty (response to light),
- nyctinasty (movements at night or in the dark),
- chemonasty (response to chemicals or nutrients),
- hydronasty(response to water),
- thermonasty (response to temperature).

#### Self-assessment 13.6

1. A bean seedling has been placed on horizontal position as shown by the diagram below.



- a. Illustrate the expected shape of the shoot and that of the root after a week.
- b. Suggest names (of what) for the expectations in the above experiment.
- c. Based on the above experiment, draw a diagram illustrating the shape and size of cells of both upward and downward side of the root.
- d. Suggest a technique that can be used to minimize the effect of the stimulus in this experiment.
- 2. a. What is the difference between 'antagonistic' and 'synergistic' when referring to plant growth substances?
  - b. What are the two plant growth substances that act antagonistically and which act synergistically?
- 3. Copy and complete the following table

Plant growth substance	Site of synthesis	Effect in plant
Auxin		
Gibberellin		
Cytokinin		
Abscisic acid		
Ethylene		

# **Activity 13.6.2**

Most of plants grow toward the sunlight direction. The few which have been observed respond to external stimuli like touching and temperature.

- 1. From your experience, brainstorm what will happen to the plant when:
  - it is exposed to the direction of the sunlight

- its growing part is exposed to a physical material like a stone
- a living organism touches on it (Mimosa pudica)
- 2. Use internet and textbooks to describe why some plants flourish during long daylight while others do not?
- 3. What names can be given to the above processes.

# 13.6.2. Photoperiodism

The light provides energy that plants need to make its own food. The duration of daylight affects the plant growth and plant development.

Photoperiodism is a plant physiological response to relative lengths of daylight and darkness. Photoperiodism affects many plant processes, including the formation of storage organs, flowering and bud dormancy.

Plants monitor changes in day length with a bluish, light-sensitive pigment called phytochrome. The alternation of darkness and light triggers the phytochrome to change from one chemical form to another. By detecting the type and the amount of phytochrome present, plants determine the length of darkness and light each day.

One of the effects of photoperiodism is that plants produce fruits at different times and are classified into three categories:

- Short-day plants (SDP),
- Long-day plant (LDP)
- Day–neutral plants (DNP).

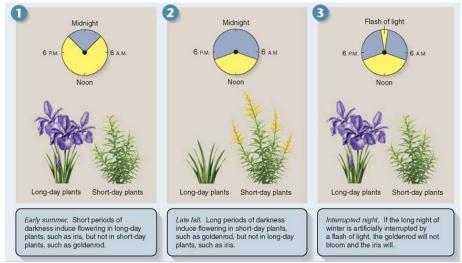


Fig 13.11: Photoperiodism and flowering

 Short-day plants (SDP): they only flower when the days are short and the night are longer than a certain length. Examples strawberry, blueberry, goldenrods, cocklebur and soybeans, tobacco are short day plants for flowering.

- Long-day plant (LDP): they produce flower when the period of daylight exceeds a critical minimum length. Radishes, asters, apple trees, squash trees, and beets.
- Day-neutral plants (DNP) are not dependent on day length for flowering. They produce flower regardless of the length of the daylight. Day neutral plants for flowering include tomatoes, roses, corn, cucumber, carrot, cotton and beans. They can produce fruit throughout the entire growing season.

#### Self –assessment 13.6.2

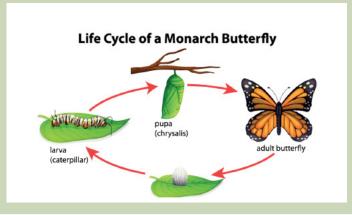
- 1. What role does phytochrome play in photoperiodism?
- 2. What is the difference between a short-day plant and a long-day plant?
- 3. What could happen if a short-day plant is grown in the long days of summer?

# 13.7. Metamorphosis and growth patterns in insects and amphibians

### **Activity 13.7**

From a pond or swamp, collect frog eggs together with water. Keep some eggs in warm conditions at room temperature between 2 to 30 days.

- 1. Visit them regularly and note the observation.
- 2. Use the diagram below and relate your observation from question (1) to the one given in the diagram.



# 1. Metamorphosis and growth patterns in animals

Metamorphosis is a process consisting of changes in body form of a young organism before it reaches its adult size and becomes sexually mature, for example, the change from tadpole to frog or from caterpillar to butterfly.

# a. Metamorphosis and growth patterns in insects

All insects develop and grow by metamorphosis. Some insects show incomplete metamorphosis. In incomplete metamorphosis, an immature nymph is hatched from the egg that looks like the adult, but it is smaller, and its wings and reproductive organs are undeveloped. It molts several times with each molt, the wings become larger and more fully formed. The final molt transforms the nymph into an adult that can reproduce and fly. Insects with incomplete metamorphosis are known as heterometaboles e.g. weevil, cockroach (*Periplaneta americana*), Grasshopper; mayflies; dragonflies and termites.

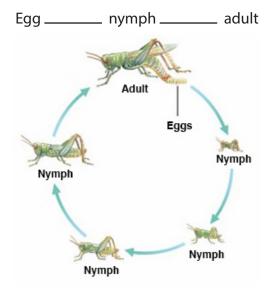


Figure 13.12: Incomplete metamorphosis

## **Complete metamorphosis**

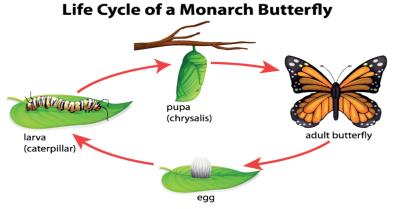


Figure 13.13: Complete metamorphosis

In complete metamorphosis eggs hatch into larvae which are morphologically, physiologically and behaviorally different from adult (wormlike larva) or caterpillar. The caterpillar molts several times, when it reaches its full size, it prepares the pupa, or chrysalis, a hard, sometimes thorny and oval structure. Inside the pupa, the larval tissues break down and group of cells called imaginal disks develop into wings and other tissues of the adult: imago stage, the pupa becomes sexually mature.

Examples: Butterflies, moths, mosquitoes, beetles, bees, housefly.

Egg \_\_\_\_\_\_ larva (caterpillar) \_\_\_\_\_ pupa \_\_\_\_\_ imago (adult)

### Importance of metamorphosis

In life cycle based on complete metamorphosis, the larval and adult stages often fulfill different functions, live in different habitat and eat different foods. Example: mosquitoes.

Metamorphosis also enhances insect survival by helping insects survive harsh period. Example: butterflies (caterpillars feed on leaves, but adult butterflies feed on nectar from flowers).

# b. Metamorphosis and growth patterns in amphibians

Many amphibian species like the frog breed in water and their eggs are fertilized externally. The fertilized eggs hatch into swimming, tailed larvae called tadpoles. Tadpoles, which usually live in water, look somewhat like small fish. A tadpole has an oval body, gills for breathing, and a long, muscular tail with fins along the upper and lower edges for swimming.

Then the tadpole grows legs—the hind legs appear first—and resorbs its tail. It loses its gills and grows lungs, and the structure of the heart, digestive system, and skeleton changes. The horny beak and other mouthparts adapted for eating algae disappear and are replaced by the long, sticky, projectile tongue that helps adult frogs catch insects. Frogs reach reproductive age anywhere from several months to several years after metamorphosing.

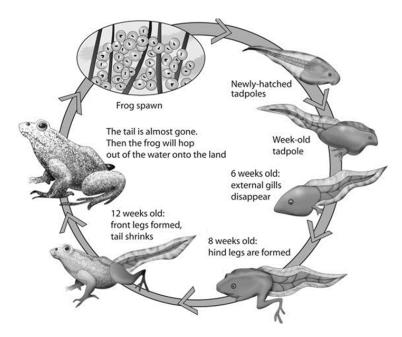


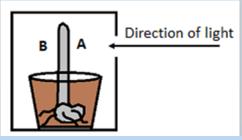
Figure 13.14 Metamorphosis in frogs

#### Self-assessment 13.7

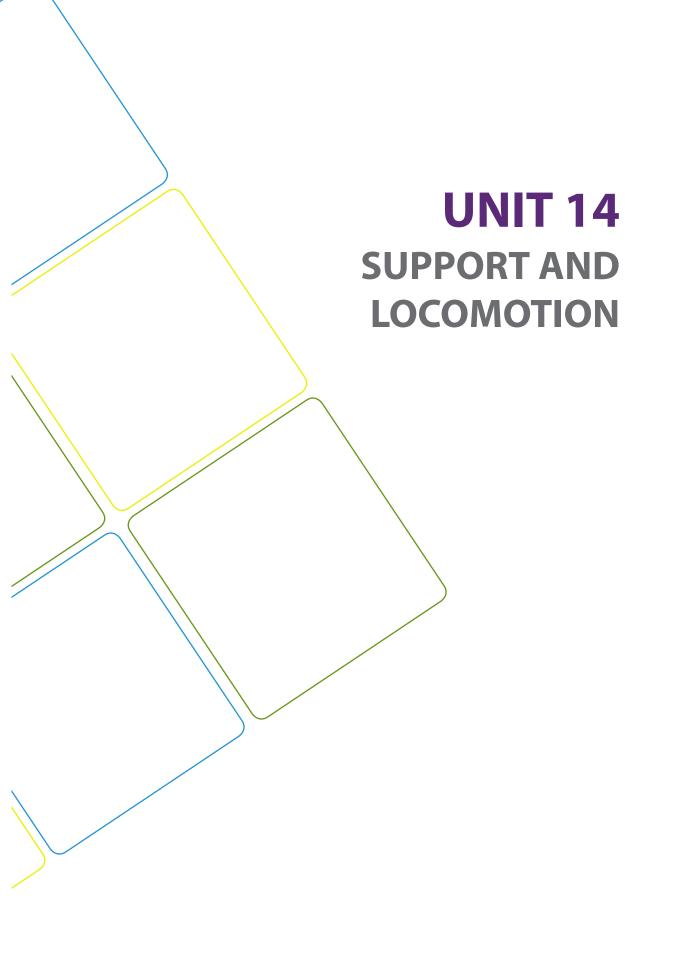
- 1. What is metamorphosis?
- 2. Describe the changes that occur during metamorphosis in frog.
- 3. Discus reasons why complete metamorphosis may have greater adaptive value for an insect than incomplete metamorphosis.
- 4. Compare metamorphosis of a butterfly and that of a grasshopper.

#### **End of unit assessment 13**

- 1. What do you understand by:
  - a. Dormancy
  - b. Gravitropism
  - c. Chemonasty
  - d. Phytochrome
  - e. Short-day plants
- 2. a. What factors can allow to measure the growth of a plant?
  - b. State any four external factors that can affect the growth of a plant.
- 3. a. State any three characteristics of the phytohormones.
  - b. What is phytohormone (s) responsible for:
    - i. The falling of some plant organs during the stress
    - ii. The fruit ripening.
    - iii. Development of the lateral buds
    - iv. Stem growth and parthenocarpy
  - c. What is meant by parthenocarpy? Give one example of a plant that shows this phenomenon
- 4. A seedling has been grown in an opaque box receiving the light from a single direction as shown by the diagram below.



- a. What will happen on the coleoptile (seedling) as it grows?
- b. Suggest a name to the phenomenon investigated in this experiment.



## **UNIT 14: SUPPORT AND LOCOMOTION**

#### **Key Unit Competence**

Explain and demonstrate modes of locomotion in protists, insects, fish, amphibians, birds and mammals

### Learning objectives

By the end of this unit, I should be able to:

- Explain non-muscular movement in amoeba or paramecium.
- Describe support and movement on land.
- Describe skeletal modification in birds.
- Explain how movements and support of fish are brought about in water.
- Explain how support structures are related to the environment of the animal.
- Observe locomotion of animals and identify reasons for their movement.
- Demonstrate the arrangement of muscles in fish.
- Dissect a fish to observe its swim bladder.
- Observe and explain the relationship between muscles, joints and musculoskeletal attachments in fish, birds, amphibians and mammals.
- Compare the flight of birds and insects.
- Compare the jumping movement of grasshoppers and toads/frogs.
- Appreciate the need for locomotion in animals.
- Recognize that the types of locomotion of animals depends on their habitat.

# **Introductory activity**

Animals have muscles and different types of skeleton.

What might happen if a large animal such as a cow does not have a skeleton. How that animal would look like? What will happen to animal without skeleton or muscles? Can you then think about the role of skeleton and muscles in living organisms?

# 14.1. Locomotion and its requirements

# **Activity 14.1**

From your experience and knowledge from books and the internet:

- 1. Give details about the concept of locomotion
- 2. How do different animals move?
- 3. Explain why animals need to move from one place to another?

Living organisms particularly animals need to move from one place to another. This is known as locomotion which should not be confused with movement which occurs in plants. Movement is the displacement of part of an organism. Therefore, movement is a characteristic of all living things.

Locomotion in animals is brought about by the action of muscles on a skeleton.A skeleton is a rigid framework that maintains shape and supports the internal organs and provides attachment for muscles, while a muscle is a soft tissue formed by muscle cells which found in most animals. Each muscle cell contains actin and myosin proteins that produce a contraction that changes both the length and the shape of the cell. Thus, muscles function to produce force and motion. In animals without muscles such as sea sponges, locomotion is brought by mesohyl or cells which act as actual muscles.

Depending on the type of animals, three types of skeletons are distinguished.

## 1. Hydrostatic skeleton

It is mostly seen in invertebrates and earthworm. These consist of fluid filled body cavity surrounded by antagonistic sets of muscles. Movement results from compressive contraction action of the contraction of these muscles on this fluid.

#### 2. Exoskeleton

It characterizes the arthropod insects. It is a hard cuticle made of chitin which lies outside the muscles. It sheds during molting when the organism outgrows it. It does not grow because it is a dead material.

### 3. Endoskeleton

It is seen in vertebrates where the bones and cartilages are found within the interior of the body on which muscles are attached. It is a living tissue and it grows with the rest of the body.

For efficient locomotion, exoskeleton and endoskeleton provide a system of levers to which muscles are attached.

# Types of locomotion

The locomotion can be either terrestrial, aerial or aquatic (swimming). In most animals, the locomotion is by running, climbing, crawling, swimming, jumping, gliding, hoping, and flying with aid of limbs or appendages. For animals without limbs such as snake, its locomotion is by forming its body into zig - zag, gripping the ground with its undersides and pushing itself forward. For ducks, their movement in water is by floating. Some invertebrates like roundworms, flatworms, squids, octopus, and jellyfish without special organs of locomotion are propelled by the muscular contractions.

# **Advantages of locomotion**

Based on the types of locomotion mentioned above, an animal is capable to:

- Escape danger such as fire or predator
- Look for food, water and shelter

- Reproduce
- Avoid competition with other animals of the same or different species
- Avoid overcrowding which enables offspring to move to another place
- Avoid unfavourable condition

#### Self-assessment 14.1

- 1. What is locomotion?
- 2. What are the requirements for locomotion?
- 3. Given the following animals: Frog, dragonfly, squid, spider, antelope, kangaroo, fish, grasshopper, bee, duck, worm, zebra, snake, and cow. Identify those which fly, crawl, hop, and or run/walk.
- 4. Discuss why locomotion is very important in animals?

# 14.2. Support and locomotion in non-muscular organisms

### Learning activity 14.2

From a culture of paramecium:

- 1. Use a microscope to observe the locomotion in Amoeba and Paramecium
- 2. From what you have observed identify Amoeba, Paramecium, Euglena and in Trypanosoma moves in relation to their structures / diagrams below

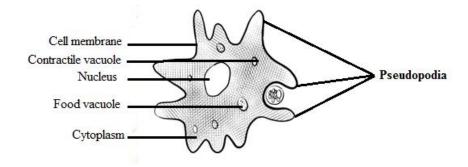


3. Discuss how is locomation performed in those organisms.

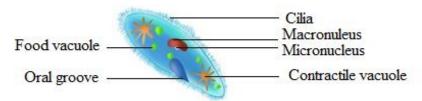
Non-muscular locomotion is identified in living organisms that belong into protoctista kingdom. Depending to individuals belonging to protoctista, locomotion is either amoeboid, ciliary, flagellated or euglenoid type.

Amoeba moves by amoeboid locomotion i.e. by putting out pseudopodia. Locomotion is not maintained in any particular direction for long. Amoeba is constantly changing shapes as it changes direction. Amoeboid locomotion is brought about by cytoplasmic streaming and between a gel and sol state. These cytoplasmic streaming requires Ca<sup>2+</sup> ions and ATP. Amoeboid locomotion is common to all rhizopodes including Amoeba and white blood cells of the vertebrates.

#### a. Amoeba



#### b. Paramecium



#### c. Euglena

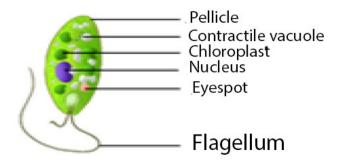


Figure 14.1: Labelled diagrams of non-muscular organisms (a, b, c)

Paramecium moves by means of cilia and Euglena move by the use of flagella. Cilia and flagella have similar structure except that cilia are short and many. Both cilia and flagella consist of fine tubes composed of an extension of plasma membrane. Euglenas have an intricate cell membrane called a pellicle. The latter is folded into ribbon-like ridges and each ridge is supported by microtubules. The pellicle is tough and flexible, letting euglenas crawl through mud when there is not enough water for them to swim.

During cilia or flagellum locomotion, tubules slide past each other in a movement similar to that of actin and myosin filaments in skeletal muscles. Hence Ca<sup>2+</sup> ions and ATP are also required in the ciliary locomotion.

#### Self-assessment 14.2

- 1. Describe the type of locomotion found in:
  - a. Amoeba
  - b. Paramecium caudatum
  - c. Trypanosoma gambiense
  - d. Trypanoma vaginalis
  - e. Giardia intestinalis
- 2. How do cilia differ from flagellum?
- 3. Produce picture showing the locomotion of amoeba

# 14.3. Support and locomotion in fish

## **Activity 14.3**

1. Observe the freshly collected fish or the figure, to label fins and lateral line.



- 2. Dissect a fresh fish or observe the above given diagram. Redraw and show the swim bladder and the arrangement of muscles
- 3. If you have a live fish, put it in water and observe its locomotion.
- 4. From what you have observed, draw and label the external and internal features that contribute to fish locomotion.

Fish like other aquatic animals are adapted to such habitat in terms of locomotion due to its structural adaptive features particularly skeleton which gives shape as well as muscles arrangement and swim-bladder.

# Adaptive features of fish for locomotion in water

The streamlined body shape of the fish reduces friction between water and fish. The body of fish is mostly covered by scales which overlap one another and point backwards and lie close to the body. The scales are covered by mucus which reduces the drag.

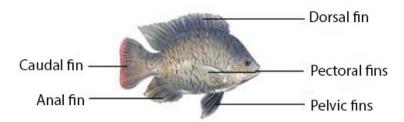


Figure 14.2: External adaptations of fish for locomotion

Tail or caudal fin has a large surface area, which increases the amount of water that is displaced as it provides much of the push during swimming. Paired pectoral and pelvic fins bring about downward and upward movement. With pectoral fins, the control of direction of a fish in water is possible whereas the pelvic fins bring about the balance, preventing diving and rolling. There are also unpaired dorsal and anal fins for stabilizing the fish and thus preventing it from rolling or yawing.

Fish is also adapted to locomotion in water by its strong tail muscles and highly flexible vertebrate column which enables the tail to move from side to side against water. In addition, inflexible head and neck maintain forward thrush.

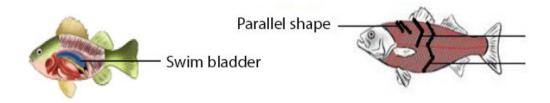


Figure 14.3: Internal adaptations of fish for swimming

Internally, a fish is adapted to swimming by swim bladder and muscles. Air or gas-filled sac called swim-bladder, outgrowth of the pharynx, helps a fish to change its buoyancy as it alters the gas pressure in the bladder. So that, it floats at any depth in water without using its muscles. Swim-bladder also helps fishes to maintain a density that is equal to that of the surrounding water. Muscles or myotomes / myomeres (segments or sheets of muscles separated from its neighbor by a sheet of connective tissue) enable fishes to move in water owing the shapes of muscles that are located on either side of vertebral column.

Myotomes contribute to the mechanism of swimming by its arrangements. They may be parallel, V-shape, or W-shape arranged in bundles or blocks that are separated by myosepta.

Although there are such arrangements, the myoseptal organization and orientation of fibres is complex. In bony fish, myomeres are V-shaped with new myomeres added posteriorly. With those myotomes, a fish swim by passing a wave of contracting muscle from anterior to posterior. Muscles near the head of the fish contract first and contraction proceeds posteriorly down the length of the fish to the caudal fin. Thus,

a fish moves forward from the contraction and relaxation (antagonistic) of myotome on either side of the body.

Undulatory swimming of the fish is also powered by the segmental body musculature of the myotomes. Myotome and myosepta orient more perpendicularly to midline to push aside. Therefore, the fish can bend laterally. With contraction muscle fibres shorten by half their length while maintaining volume. Without myosepta, but simply a series of interconnected muscle fibres, then the wave would be much dampened.

#### Self-assessment 14.3

- 1. How does swim bladder help the fish in locomotion?
- 2. Illustrate how the arrangements of fish myotomes contribute to fish locomotion.?
- 3. What does it make a fish to move in undulatory propulsion?
- 4. What are the anatomical structures that give rise to the direction of a fish and preventing diving and rolling?

# 14.4. Support and locomotion in terrestrial animals

## **Activity 14.4**

- 1. Through internet observe and think about how locomotion in dogs, chicken, frog and earthworm brought about.
- 2. Make a diagram showing how support and movement of different animals such as dogs, chicken, frog and worm is brought about on land.
- 3. Show by using diagrams the relationship between muscles, joints and musculo-skeletal attachment in mammals, birds, frog and earthworm.

All animals living on land move due to the musculoskeletal system. The rigid nature of bone also gives a structure for muscles to pull, by their contraction, to create a movement as they act as levers. The synovial joints also allow certain movements.

The support and movement differ from specimen to another. Thus, animals can walk and run on land for moving from one place to another. This is possible by their endoskeleton and its muscles. By its muscles, flexor (a muscle whose contraction bends a limb or other part of the body) and extensor (a muscle whose contraction extends or straightens a limb or other part of the body or any or a muscle that increases the angle between members of a limb, as by straightening the elbow or knee or bending the wrist or spine backward); contractions of those muscles cause the limbs act as levers for them which result to the foot being pressed downwards and backwards against the ground. For example, flexor and extensor work as illustrated below:

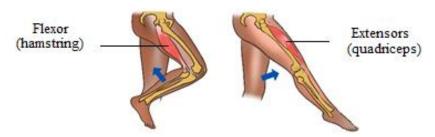


Figure 14.4: Flexor and extensor

### a. Locomotion in quadruped animals e.g. dog and frogs

When a dog walks, its vertebral column remains rigid, and the forward movement is achieved by the activity of the hind limbs. When its extensor muscle contracts, each hind limb, acting as a lever, extends and exerts a backward force against the ground, thrusting the animal forward and slightly upwards. When the flexor contracts, the limb is lifted clear of the ground and pulled forward. Only one limb is raised at any one time, the other three providing a tripod of support which balances the rest of the body.

Beginning with the left forelimb in a stationary dog, the sequence of leg movement is as follows when it walks forward: left forelimb-right hind limb-right forelimb-left hind limb and so on. Such walking in quadrupedal animal is also identified in frogs when they can walk on land.

### b. Running of the dog

As a dog begins to run, it loses its quadrupedal movement which means, it develops a type of movement where the forelimbs move together, followed by the hind limbs.

# c. Walking in bipedal animals e.g. humans

Humans are bipedal, they walk on two legs. When standing upright, the weight is balanced over the two legs. When a stride is taken by the right leg, the heel is raised first by the contraction of the calf muscles. As this occurs, the weight of the body is brought over the left foot which is still in contact with the ground and acting as the prop for the rest of the body.

When the right leg extends the heel is the first part of the foot to touch the ground. The weight off the body is gradually transferred from the left side to a position over the right heel and then the body continues to move forward, over the right toes, backward pressure against the ground generally being exerted through the right big toe. Like human does, a bird also can walk on ground through the movement of contractions of its leg muscles particularly flexor and extensor.

# d. Crawling of earthworm

Earthworms are organisms having hydroskeleton with soft-bodied animals due to fluid secreted within the body and surrounded by the muscles of the body wall. They are capable to move by aid of their muscles. These muscles are not attached to any structures and thus can pull against each other. The combined effect of muscle contraction and fluid pressure serves to maintain the shape and form of the animal. Generally, there are two muscle layers, longitudinal in which muscle fibres are arranged parallel to the long axis from one end of a segment to another and circular with muscle fibres arranged in concentric circles to the circumference of the worm. When those muscles act antagonistically against each other, locomotion is achieved. The fluid which acts as pressurisable hydrostatic skeleton contained in body cavity or coelom presses against the muscles which in turn are able to contract against the fluid. Earthworm movement is also helped by bristles like setae or called chaetae (hair like structures on ventral surfaces) which anchor the worms to the substrate.

Contraction of the circular muscles makes the worm thinner, but because liquid is essentially incompressible (and so maintains a constant volume) and the increase in pressure forces the liquid outwards, stretching the worm, so the worm becomes longer and thinner. Contraction of the longitudinal fibres shortens the worm, former the coelomic liquid out to the sides and making the worm fatter. If the body is segmented, then such pressure is localized and only certain segments will move or change shape.

#### Self-assessment 14.4

- 1. What are the main muscles that contribute to locomotion in mammals, amphibians and birds?
- 2. Draw an earthworm and illustrate the muscles that contribute to its locomotion.
- 3. What type of skeleton system found in mammals, birds, amphibians and annelids?
- 4. . Illustrate how flexor and extensor muscles contribute to lifting up a leg in human being.

# 14.5. Flight through air by birds and insects

# **Learning activity 14.5**

Make a research on internet as well as books and do the following:

- 1. Observe pictures below (Figure 14.6) related to birds and make a description of skeletal modification in birds. Illustrate the skeletal modification in birds
- 2. Draw a bird and show by using arrow the structures that enable a bird to fly
- 3. How will the external features of birds will behave when flying in high or low atmospheric pressure
- 4. Make a table illustrating how does flight of birds and insects differ and similar
- 5. Observe and compare the flight of birds and insects

Bird can fly either by flapping their wings or gliding by spreading its wings. Like in animals moving on land, locomotion by flying in birds is brought about by the action of flexor and extensor muscles as well some other structures given diagram below like pectoralis major, pectoralis minor and keel of sternum.

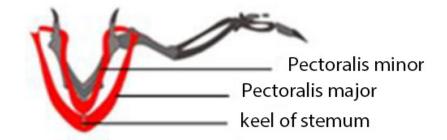


Figure 14.5: Adaptive features for bird to flight

Based on the above diagram, wings move down by the contraction of pectoralis major and then move up under the contraction of pectoralis minor.

## Adaptive features of birds for movement in air

A number of features enable birds to aerial locomotion. Those features include body shape, modified limbs, and modification in internal organs particularly bones. The body of bird is highly streamlined and covered with light feathers that overlap backwards thus reducing air resistance during flight. Those light features that increase the surface area of wings without increasing weight.

Differently from quadrupedal animals, birds are adapted to flying by modification of fore limbs into wings. Such modification goes with a well-developed or large keel sternum that provides large surface area for the attachment of the flight muscles namely major and minor pectoral muscles which give the power to flap the wings in flight. Also, birds have hollowed bones making the body light and vertebrae of trunk are fused. As flight requires much energy, birds are adapted to that by an efficient breathing system with air sacs attached to the lungs necessary to provide oxygen for respiration and to remove the resulting carbon dioxide.

Other adaptations include a high metabolic rate for providing the high amount of energy required, an efficient circulatory system necessary for transporting both the nutrients and respiratory gases at speed related with the body needs, a high red blood cell count for efficient oxygen transport, and a keen eye sight to enable them to judge distances correctly especially on landing.

#### Self-assessment 14.5

- 1. What are the muscles that enable the flight in birds?
- 2. Describe how bird skeleton contributes to its flight?
- 3. Describe how birds are adapted to flying.

# 14.6. Hopping locomotion in grasshoppers and toads

### **Activity 14.6**

Use a collecting net to catch a grasshopper and toad from school compound. Put them down on cemented ground for observing them very carefully when they make a jump and then answer to the following:

- 1. Identify and describe anatomic structures that enable grasshoppers to jump
- 2. Illustrate how legs' muscles behave when they are resting and or jumping

Skeletal muscles such as extensor and flexor that occur in pairs are often antagonistic. With such antagonistic behaviour, when one contracts the other relaxes to produce controlled movement in the opposite directions.

### a. Locomotion of grasshopper

Insects have a skeleton which is on the outside of the body called an exoskeleton. They can walk on the land but they are mostly adapted to hopping owing to their muscles which are inside the hard shell as well as skeleton system. The muscles which make them capable to move are flexors and extensors which are antagonists, attached to internal surface of exoskeleton and the rear or back legs of a grasshopper which are long and muscular, adapted for hopping. Additionally, there are two main muscles inside are the extensor tibiae muscle which contracts to extends the leg, and the flexor tibiae muscle which contracts to flex the leg as illustrated in figure below. Those muscles pull on tendons which are attached to the tibia on either side of the joint pivot.

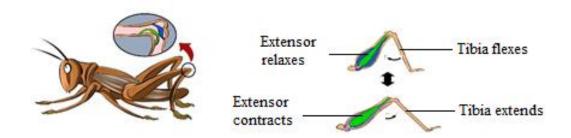


Figure 14.6: Muscles for jumping in grasshopper

The back legs are much longer than the others for helping in hopping. With those long legs, grasshopper is capable to make high jumping distance. As illustrated above, flexor muscles bend a joint whereby extensor ones straighten it. The flexor muscle contracts and the lower leg is pulled towards the body. Thus, the hind leg is folded in a Z shape and ready for jumping. Being in resting or sitting position,

the extensor muscle contracts which enable then the legs jerk or move very quickly backwards propelling the grasshopper.

### b. Locomotion in toads and frogs

On land, frogs and toads move by hopping (going from place to place).

- When a frog is at rest, the hind legs are folded up in the shape of a letter Z.
- When it hops, the legs are quickly straightened out, lifting the animal of the ground.
- The fore-limbs are used as shock absorbers on landing and they also prop up (to give support) the front end of the body when the animal is at rest.

They also hope but do not travel as high as far as a frog does at each hop.

#### Self-assessment 14.6

- 1. What are the muscles that contribute to high jumping in a grasshopper?
- 2. How do muscles (flexor and extensor) behave when toads and grasshopper are resting?
- 3. Draw a leg of grasshopper and the one of toad when are jumping

#### **End of unit assessment 14**

- 1. Describe ways of locomotion in Amoeba, Paramecium, Euglena and in Trypanosom
- 2. Produce a cartoon showing different adaptive features of fish for aquatic locomotion
- 3. Describe how different fish fins contribute to locomotion and balance
- 4. Describe how the movements and support of fish in water do they occur? are brought in water
- 5. Show by diagrams the relationship between muscles, joints and musculoskeletal attachment in mammals, birds, frog and earthworm.
- 6. Describe how flexor and extensor muscles work to enable the locomotion on land, water and in air
- 7. What are the features that enable aerial locomotion in animals?
- 8. Describe how a grasshopper and toad is adapted to jumping





# **UNIT 15: CLASSIFICATION AND PATTERNS OF DISEASE**

### **Key Unit Competence**

Describe the social factors that affect good health and apply knowledge gained in familiar and unfamiliar contexts.

### **Learning objectives**

- By the end of this unit, I should be able to:
- Explain what is meant by health and disease.
- Identify different categories of disease and give an example of each.
- Explain the theory of the disease and the causes, sources, transmission, symptoms and controls of the disease.
- Discuss how global patterns of disease are studied.
- Analyze and interpret records from a given hospital to identify diseases as endemic, epidemic or pandemic.
- Apply knowledge gained to classify common diseases.
- Appreciate the importance of germ theory of disease by showing that the death rate related to infections is greater than those caused by accidents.

## **Introductory activity**

- a. Suggest measures to be taken for addressing issues related to eating without washing hands.
- b. Discuss on different communicable diseases got from eating without washing hands.

# 15.1. Germ theory of diseases

# **Activity 15.1**

Discuss the following questions

- a. What are the causes of death?
- b. Why it is difficult to eradicate malaria in Rwanda?

The germ theory states that many diseases are caused by the presence and actions of specific microorganisms within the body. In 1677, Antoni van Leeuwenhoek was the first to observe microorganisms in the droplets of water. But he did not make the connection with disease. Later, Spallanzani and Louis Pasteur observed germs in the blood of people suffering from disease. They suggested that the germs were an effect of the disease rather than the cause. The observations and actions of Ignaz Semmelweis, Joseph Lister and John Snow were a great contribution to the acceptance of germ theory. However, the laboratory works of Louis Pasteur in the 1860s and Robert Koch in the following decades, provided the scientific proof for

germ theory. Their works opened the door to research related to the identification of disease-causing germs and potential life-saving treatments.

#### 15.1.1. The work of Louis Pasteur and Semmelweiss

The work of Eduard Jenner and Ignas Semmelweiss showed that infectious diseases maybe caused by an infectious agent or germ. This was accepted as the germ theory of disease for a very long time. The work of Robert Koch and Louis Pasteur led to a wide acceptance of the germ theory

Pasteur made a number of important steps forward. He indicated that fermentation is the result of the action of microorganisms (Yeast) on sugar. Huge number of people at the time believed that living things could arise spontaneously from non-living things and this theory is known as **spontaneous generation.** To reject this theory, he showed that if broth is boiled in a sealed container, it would stay clear, but once he added material which had been exposed to the air, microorganisms grew in the broth. Finally, he designed a series of experiments using swan necked flasks which showed once and for all that any microorganisms which appear in boiled broth come from the air not arise spontaneously from nonliving organism such as broth.

In 1845-1846, Pasteur found a way of avoiding the disease of silkworms by observing the infected eggs under microscope and thereby saving the silk industry. This was the first clear evidence of microorganisms causing disease. Pasteur even developed vaccines against a number of these diseases.

# 15.1.2. Summary of the contributions of Louis Pasteur in microbiology and medicine

The contribution of Louis Pasteur in microbiology and medicine also include:

- The fight against spontaneous generation theory
- The technique of sterile culturing of microorganisms
- The technique of fermentation and conservation of drinks. This technique is known as Pasteurization.
- The technique of antiseptic surgery to prevent contamination of wounds during the surgical operations in hospitals.

Louis Pasteur developed the germ theory of disease which postulates that all contagious and infectious diseases must be caused by pathogenic microorganisms.

# 15.1.3. The Germ Theory and Koch's Postulates

Diseases can be spread by air, water, food, and human as well as animal vectors. In, an english physician called John Snow (1854) and a German microbiologist called Robert Koch (1884) found a relationship between polluted water and disease. Robert Koch, has isolated the bacterium Vibrio cholera, the cause of cholera from Elbe River water to provide the relationship.

Koch went on to formulate an established set of procedures to isolate and identify the causative agent of a particular microbial disease. The following four steps, which are still used today, are known as Koch's Postulates:

**Postulate 1**: A specific organism must always be observed in association with the disease.

**Postulate 2**: The organism must be isolated from an infected host and grown in pure culture in the laboratory.

**Postulate 3:** When the organism from the pure culture is inoculated into a susceptible host organism, it must cause the disease.

**Postulate 4**: The infectious organism must be re-isolated from the diseased organism and grown in pure culture.

#### Self-assessment 15.1

- 1. What are the Koch's Postulates?
- 2. Explain the theory of spontaneous generation.

### 15.2. Classification of diseases

### **Activity 15.2**

Use the Knowledge gained to answer to the following questions:

- 1. Propose two infectious diseases and for each disease, give:
  - a. their causal agents
  - b. causal agents' type
  - c. their symptoms
  - d. their methods of prevention
  - e. Treatment
- 2. Does being healthy means just the absence of the disease? Explain.

The following are the meaning of disease, signs and symptoms:

- Disease is the disruption of normal body function.
- Signs are indications of a disease that can be observed by examining the patient.
- Symptoms are indications of disease perceive only by the patient.

The normal functioning of the body is disturbed, when the body is infected. Many types of diseases are broadly divided into two categories: Infectious diseases and non-infectious diseases

#### 15.2.1. Infectious diseases

Infectious diseases are caused by microorganisms known as **pathogens** which may include viruses, bacteria, fungi and protozoa. Those diseases are called **communicable diseases**. as they can be transmitted from one person to another. They include cholera, malaria, typhoid, HIV and AIDS...Malaria is one of the most

dangerous infectious diseases, endemic in Latin America, Africa and South-East Asia. Some infectious diseases can also be from animals to humans.

The following are some technical terms used when discussing about infectious disease.

- Aetiology: The study of the cause of disease.
- **Epidemiology:** The study of all the factors that contribute to the appearance of a particular disease
- **Causative agent:** The organism which causes the disease
- Vector: An organism which carries the causative agent of the disease from one person to another or from infected animal to human.
- Incubation period: The period of time between the original infection and the appearance of signs and symptoms.
- **Infective period:** The time during which a person is capable of passing the disease on to another person.
- Carrier: The person who has been infected but develop no signs and symptom, the carrier can pass the disease on to another person
- **Prevention:** Measures taken to prevent diseases.
- Treatment: Measures taken to cure diseases Antibody: Is a protein produced by the body's immune system when it detects harmful substances called antigen.
- Antigen: Is any substance that causes your immune system to produce antibodies against it.
- **Host:** A host can be anything living organism ion which pathogens can survive
- **Hygiene:** Practices that help to maintain health and prevent the spread of diseases
- Immunity: Is the ability of the body to resist to infections.

# a. Important advances in the work against infectious disease

Table 15.1: The chronology of some of the most important advances in the work against infectious disease

Year	Scientist	Development	Significance
1794	Eduard Jenner	Demonstration that vaccination with cowpox protected against smallpox	The first vaccine

1847	Ignaz Semmelweiss	Showed that childbirth fever could be prevented by hygiene.	Evidence of contagion, use of control group
1861	Louis Pasteur	Showed that living organisms. do not arise spontaneously.	Evidence for contagion and against miasma theory and spontaneous generation of germs
1865	Joseph Lister	Used antiseptics to reduce deaths after surgery	An important practical application of Pasteur's ideas to save lives
1876	Robert Koch	Showed that Bacillus causes anthrax.	Very first direct evidence of a microbe causing a disease.
1879	Louis Pasteur	Discovered how to weaken chicken cholera so that it could be used as a vaccine without causing infection	First technique for production of a vaccine by deliberately weakening a bacterium. Idea of preventative medical care.
1882	Robert Koch	Discovered the TB bacillus	Further confirmation of the bacterial cause of disease

1883	Robert Koch	Discovered the cholera bacillus	Confirmation of Snow's work by discovery of the causative agent
1928	Alexander Fleming	Discovered penicillin, the first antibiotic  R H S CH <sub>3</sub> CH <sub>3</sub> OH	The start of the introduction of bacteria-specific medicines which have saved millions of lives
1921	Calmette & Guerin	Produced BCG vaccine against TB	Application of Pasteur's approach to a vaccine against a disease which killed many people

### b. Some groups of communicable diseases

- Bacterial diseases: these are diseases caused by bacteria. They include cholera, typhoid, tetanus, tuberculosis, etc.
- Viral diseases: these are diseases caused by viruses. They include AIDS, polio, measles, Ebola, etc.
- Protozoan diseases: these are diseases caused by protozoa. They include malaria, sleeping sickness, trichomoniasis, etc.
- Fungal diseases: these are diseases caused by fungi. They include candidiasis, athlete's foot, ring worms, etc.
- Worm diseases: these are diseases caused by worms. They include elephantiasis, bilharzias, etc.
- **Sexually transmitted diseases:** these are diseases transmitted through sexual contact. They include HIV-AIDS, syphilis, gonorrhea, etc.

#### c. Transmission of infectious diseases

Pathogens can spread when you have direct contact with an infected person. For example, if you have contact with the person's blood, body fluids or open wounds. Pathogens can also be spread through contaminated food, water or air. Infected animals can spread pathogens to people.

The following conditions lead to the spread of an infectious disease:

- A pathogen which causes the disease.
- A source which is an infected organism.

Mode of transmission a pathogen must be able to enter the body of the new host to cause an infection. Infectious diseases follow a pattern of development from the time of infection. The pattern of development has five distinct periods, as described in table below:

Table 15.2: The pattern of development's distinct periods for infectious diseases

Period	Description
Incubation period	The pathogen grows and multiplies in the host's body.  There are no symptoms yet.
Prodromal period	The host is usually tired, lacks appetite and begins to feel ill
Clinical symptoms' Period	The host shows typical symptoms of the particular disease, for example, spots and fever.
Convalescence period	The host usually recovers from the disease and the symptoms disappear
Complications 'Period	Some diseases cause further complications. For example, a pregnant women who has a Measles may give birth to a deformed baby.

## d. Epidemiology

The study of patterns of disease and of the various factors that affect the spread of disease is called **epidemiology.** 

Epidemiologists try to discover the factors that cause a disease and develop methods to prevent its spread. The main clue they use come from data about the number of people in a particular area affected by specific diseases, and the number of death The data are commonly expressed as incidence or morbidity and mortality rates.

The incidence rate is the number of new cases of disease in a given population occurring during a specific period (a week, month or a year). It is calculated as:

Incidence = 
$$\frac{\text{Number of cases of a given disease}}{\text{Number of individuals in the population}}$$
 x 100

Example: among 900 students at a given school, 50 suffer from malaria and 4 deaths were recorded. Calculate the morbidity or the incidence of malaria at that school.

Solution: Incidence = 
$$\frac{50}{900}$$
 x 100 = 5.5%

To find how many cases of a disease are new, this calculation requires information about the prevalence rate. This is the total number of individuals infected in a population at any one time.

The mortality rate of a disease may be estimated for a whole population irrespective of whether they have the disease or not.

Mortality rate = 
$$\frac{\text{Number of death due to a given disease}}{\text{Number of individuals in the population}} \times 100$$

It may be calculated by using only those people who have disease.

Mortality rate = 
$$\frac{\text{Number of death due to a given disease}}{\text{Number of population with tha same disease}} \times 100$$

To make fair comparisons between different populations with the same disease, epidemiological information is usually adjusted. For example, the mortality rate among those with a particular disease is usually expressed as a percentage or ratio per year. Hence, if in one year 7500 people in a given area die as a result of AIDS and the total number of population infected was 30 000, the mortality rate would be 25% for the rate.

Epidemiological studies are used to identify whether a disease is endemic, epidemic, or pandemic:

- Endemic disease is a disease that is always present in a people e.g. malaria in tropical Africa.
- Epidemic disease is a disease that spreads rapidly, suddenly, and unexpectedly to affect many people. e.g. cholera in refugees' camp. Pandemic disease is a disease that affects people over very large area, such as a continent or even the whole world e.g. AIDS and TB are pandemic at present

#### 15.2.2. Non-infectious diseases

These diseases are also called non-communicable diseases. They cannot be transmitted from one person to other examples: albinism, kwashiorkor, cancer, diabetes, etc

Categories of diseases	Examples of diseases	Comments
Inherited disease	Haemophilia, cystic fibrosis, sickle-cell anaemia	A disease caused by a genetic fault that may be passed from parents to offspring.
Degenerative disease	Arthritis	A gradual decline in function often associated with ageing.

Table 15.3: Six groups of non-communicable diseases

Social disease	Alcoholism	Drug dependence, often induced by social pressure and social behaviour.
Mental illness	Anorexia, schizophrenia	Disorder in mind which may or may not have a physical or chemical cause.
Eating disorder	Anorexia, obesity	A disease caused by under eating or overeating
Deficiency disease	Scurvy, rickets	A disease caused by a poor diet lacking one or more essential nutrients.

#### Lesson self-assessment 15.2

- 1. Answer by true or false
  - a. Epidemic disease is a disease that is always present in population.
  - b. The diseases that transmitted among people by pathogens are called transmissible diseases
  - c. The study of patterns of disease and of the various factors that affect the spread of disease is called epidemiology.
  - d. Cholera is infectious disease
  - e. Malaria is non-infectious disease whose vector is mosquito.
- 2. Distinguish between morbidity and mortality

### 15.3. Common infectious diseases

# **Activity 15.3**

Choose in the following list the infectious diseases and explain why:

Cholera, typhoid, Alcoholism, tetanus, tuberculosis, AIDS, Haemophilia, polio, measles, Ebola, malaria, Anorexia, obesity, sleeping sickness, trichomoniasis, sickle-cell anaemia, candidiasis, athlete's foot, ring worms, elephantiasis, bilharzias, syphilis, gonorrhea, cystic fibrosis, Arthritis, Anorexia, schizophrenia.

#### 15.3.1. Measles

Measles is a contagious acute viral disease with symptoms that include a bright red rash of small spots that spread to cover the whole body. Small white spots, known as Koplik's spots, appear in the mouth on the inside of the cheeks a few days before the rash appears and can be used in diagnosis.

Table 15.4: The features of measles.

Pathogen	Morbilli virus
Method of transmission	Droplets
Global distribution	Africa, South-East Asia, Indian subcontinent, Middle East
Incubation period	8 to 14 days
Site of action of pathogen	Respiratory tract (trachea and bronchi)
Clinical features	Fever and rash

#### Failure to eradicate measles

- Incubation period is short hence it is difficult to identify and isolate before they become infectious
- It is transmitted through a carrier mother to healthy children hence it is hard to eradicate.
- It targets young children who like playing together. This makes ease the spread of the disease.

### 15.3.2. Typhoid

### a. Causal agent of typhoid

Typhoid is waterborne disease caused by Salmonella typhus, a Gram negative bacterium. The bacteria are derived from the feces of a patient. It has high infectivity as low dosage of organisms is only needed for typhoid to spread. Common sources of typhoid infection are contaminated water, milk and food.

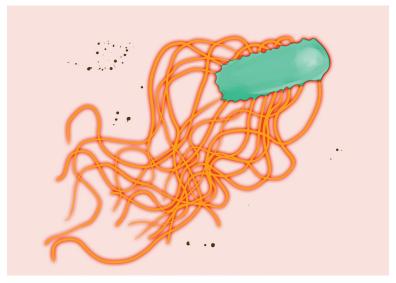


Figure 15.1: Salmonella typhus

### b. Development of typhoid illness

The incubation period is of ten days. After this time, the germs enter the bloodstream and the patient develops the following symptoms: Headache, Muscular pains, Fever reaching its peak after about 1 week, faint rash may appear, diarrhea in the second week, mental confusion, etc. The third week shows the peak of the illness and the patient may die if not treated.

### c. Treatment and prevention of typhoid

The disease had a 20% fatality rate before the use of antibiotics. Chloramphenicol and Ampicillin are effective and reduce fatality rate from 1 up 5%. Today, Ciprofloxin is used as another antibiotic. The two most important preventive measures are; proper treatment of sewage and disinfection of water supplies. Hygienic measures in the food trade and at home, and control of flies, which can transfer fecal material to food. Vaccine is formed by a polysaccharide from the capsule of the bacteria.

#### 15.3.3. Cholera.

Cholera is a good example of a waterborne disease. It is endemic in parts of Asia, particularly India. The organism which causes cholera is a comma shaped motile bacterium called Vibrio cholerae.

### a. Transmission and symptoms of cholera



Figure 15.2: Vibrio cholerae

The main source of infection is water contaminated by feces with Vibrios. It is estimated that only about one infected person in 50 develops the disease, the rest being carriers. Drinking contaminated water, or washing food or utensils in it, is the most common means of transmission. Direct contamination of food with feces as a result of poor hygiene is also possible, house flies being the main vector in this last case.

### b. Signs and symptoms of cholera

Vibrio cholerae multiply in the intestine, releasing a powerful toxin which results in violent inflammation of the intestine and production of the watery diarrhea.

The main sign of the disease is severe diarrhea due to irritation of the bowel by toxins from the Vibrios. The liquid of the feces is so profuse and cloudy like "rice water".

Abdominal pain and vomiting are also common. Dehydration is rapid and quickly results in death unless rehydration treatment is given. Fever is absent; in fact, the skin feels deathly cold and often damp.

Table 15.5: The features of cholera.

Pathogen	Vibrio cholera
Methods of transmission	food-borne, water-borne
Global distribution	Asia, Africa, Latin America
Incubation period	two hours to five days
Site of action of pathogen	wall of small intestine
Clinical features	severe diarrhoea ('rice water'), loss of water and salts, dehydration, weakness
Method of diagnosis	microscopic analysis of faeces

#### c. Treatment of cholera

The primary cause of death from cholera is dehydration i.e. loss of water with its minerals salts. For that, it is obligatory to rehydrate with oral serum which contain mineral salts and sugar.

The loss fluid may be replaced by administration of a drip food into a vein.

Various antibiotics, such as **tetracycline's** and **chloramphenicol**, are used to treat cholera. Chloramphenicol is effective against tetracycline-resistant Vibrios.

#### d. Prevention of cholera

- Use clean drinking water,
- Proper treatment of sewage and sanitation
- High standards of public and personal hygiene, particularly in relation to food (such as washing hands after defecation)
- Health education
- Vaccination is recommended for people visiting areas where cholera is endemic and for those living in such areas. But the vaccine lasts few months.
- Isolation of patients and hygienic disposal of feces and vomit from patients.

#### e. Failure to eradicate cholera

- Vaccination is not very effective
- It is a waterborne disease i.e. transmitted through contaminated water
- Poor sanitation condition in camps.

### 15.3.4. Tuberculosis (T.B)

TB spreads when infected people with the active form of the illness cough or sneeze and the bacteria are carried in the air in tiny droplets of liquid.

### a. Causal agent of tuberculosis

Tuberculosis is caused by bacterium called Mycobacterium tuberculosis, first discovered by Robert Koch in 1882. It is sometimes referred to as the tubercle bacillus, bacilli being rod-shaped bacteria. The common form is pulmonary T.B which infects the lungs, although other organs may be affected.

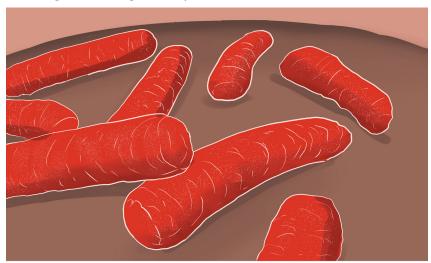


Figure 15.3: Mycobacterium tuberculosis

Two strains of the bacterium may cause the disease, the human and the bovine forms. The latter can be present in cattle and can enter the milk of cows. It is very resistant and can remain alive for long time in milk products as well as in durst.

Pathogen	Mycobacterium tuberculosis; Mycobacterium bovis	
Methods of transmission	airborne droplets (M. tuberculosis); via undercooked meat and unpasteurized milk (M.bovis)	
Global distribution	Worldwide	
Incubation period	few weeks or up to several years	
Site of action of pathogen	primary infection in lungs; secondary infections in lymph nodes, bones and gut	
Clinical features	racking cough, coughing blood, chest pain, shortness of breath, fever, sweating, weight loss	
Methods of diagnosis	microscopic examination of sputum for bacteria, chest X-ray	

Table 15.6: The features of TB.

#### b. Transmission of tuberculosis

Tuberculosis is mainly airborne disease. The infection is done through the droplets from the patient. It is much less infectious as it requires prolonged contact between people, poor ventilation and overcrowded living conditions. In addition, TB is an opportunistic infection, striking many people with a depressed immunity.

### c. Signs and symptoms of tuberculosis

The disease is frequently characterized by vague symptoms such as: loss of appetite; loss of weight; excessive sweating; coughing, appearance of blood in the sputum, pains on the chest, shortness of breath (case of lung tuberculosis).

### d. Treatment and prevention of tuberculosis

Vaccine against the disease has been developed by Albert Calmette and Camileguérin). Antibiotics such as **rifampicin**, isoniazid and **streptomycin** are used to treat tuberculosis.

#### e. Failure to eradicate tuberculosis

- Patients can carry pathogen and infection without showing symptoms. Therefore, they are difficult to identify due to a long period of incubation
- Germs of tuberculosis can survive longer in the house dust
- The disease is related to poverty where many people share the same room and have malnutrition.
- The disease is associated with AIDS that reduced the body immunity
- Long period of treatment (6-8 moths), hence patients give up when not yet fully healed. The pathogens then form endospores that resists to medicines.
- The disease is also spread through milk from infected animals. Tuberculosis is an airborne disease i.e. spread in air

#### 15.3.5. Malaria.

#### a. Causal agent

Malaria is caused by four species of plasmodium: Plasmodium falciparum, P. vivax, P. ovale, and P. Malariae. The parasite is transmitted by the bite of female mosquitoes (the vector) belonging to the genus Anopheles.

### b. Symptoms

Malaria is characterized by severe chills, fever, sweating, fatigue and great thirst. Victims die of anemia, kidney failure or brain damage.

#### c. Occurrence of malaria

The disease now occurs in tropical and subtropical regions of the world, and its distribution is limited by conditions of the development of the mosquito vector such as temperature and altitude.

#### Malaria is **endemic** in tropics because:

- Tropical climate provides the best breeding and living conditions for the Anopheles mosquito which transmits malaria

- The Anopheles cycle requires areas of stagnant water, common within tropics
- In the tropical areas, there is presence of bushes or abundant vegetation which makes suitable habitat for mosquitoes
- Plasmodium needs temperature in excess of 20°C for it to complete its cycle within the mosquito.

Table 15.7: The features of malaria.

Pathogen	P. falciparum, P. vivax, P. ovale, P. malariae
Modes of transmission	insect vector: female Anopheles mosquito
Global distribution	Tropics and subtropics
Incubation period	from a week to a year
Site of action of pathogen	liver, red blood cells, brain
Clinical features	fever, anaemia, nausea, headaches, muscle pain, shivering, sweating, enlarged spleen
Method of diagnosis	microscopic examination of blood dip stick test for malaria antigens in blood

### d. Eradication and prevention of malaria

- Drainage of stagnant water: The larval stages of the mosquito live in stagnant water, so drainage removes breeding sites.
- Destruction of the adult mosquitoes by spraying insecticide.
- Clean bushes nearest houses where mosquitoes lay eggs
- Sleeping under mosquito net during the night

# e. Failure to eradicate malaria

- There is no effective vaccine against malaria
- The pathogens are transmitted by mosquitoes which are difficult to eradicate.
- The plasmodium has become resistant to different anti-malarial drugs
- Ignorance of some people toward the disease and how it is spread.

# 15.3.6. Smallpox

# a. Cause of smallpox

Smallpox was a horrible viral disease caused by Variola virus (DNA virus), a pox virus. It was a highly infectious disease transmitted by direct contact and it affects the respiratory passage.

# b. Signs and Symptoms of smallpox



Figure 15.4: Child suffering from Smallpox

The following are Signs and Symptoms of smallpox:

 Obvious symptoms of the disease were red sports on the face, trunk, and extremities that change to pea-sized blisters and became filled with pus. High fever and generalized aching.

#### c. Modes of transmission

This disease spread through droplet infection (contagion possible via wounds in skin, clothing, bedding and dressing)

#### d. Prevention and treatment

Large populations were vaccinated through Ring vaccination and people with the disease were isolated.

The eradication of Smallpox was successful because:

- The smallpox virus is stable. so the same vaccine could be used
- The smallpox virus does not linger in the body after infection, nor does it infect other animals, so it cannot remain hidden anywhere.
- The vaccine used was highly effective and easy to administer by scratching technique.
- It was easy to identify people with the disease.

Table 15.8: The features of Smallpox

Pathogen	Variola virus (DNA virus), a pox virus.
Method of transmission	Droplet infection( contagion possible via wounds in skin, clothing, bedding and dressing)
Clinical features	Fever, vomiting, mouth sores, High fever and generalized aching.
Diagnosis method	Based on symptoms

#### 15.3.7. Tinea

Tinea is a skin infection due to a fungus. Often, there are several patches of ringworm on the skin at once. Tinea is also known as Ringworm.

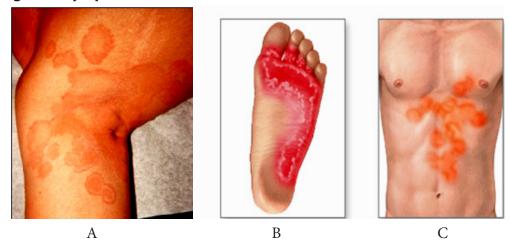
#### a. Cause of Tinea

- Tinea is caused by a tiny fungus known as **dermatophyte**. These tiny organisms normally live on the superficial skin surface, and when the opportunity is right, they can induce an **infection**.
- The disease can also be acquired by person-to-person transfer usually via direct skin contact with an infected individual. Animal-to-human transmission is also common.
- Ringworm commonly occurs on pets (dogs, cats) and the fungus can be acquired while petting or grooming an animal.
- Ringworm can also be acquired from other animals such as horses, pigs, ferrets and cows.
- The fungus can also be spread by touching inanimate objects like personal care products, bed linen, combs, athletic gear, or hair brushes contaminated by an affected person.

Individuals at high risk of acquiring ringworm include those who:

- Sweat excessively, as sweat can produce a humid wet environment where the pathogenic fungi can thrive.
- Wear tight, constrictive clothing with poor aeration.
- Have a weakened immune system
- Live in crowded, **humid** conditions.
- Participate in close contact sports like soccer, rugby.

### b. Sign and symptoms of Tinea



Figures 15.5:Ringworm on human skin

The following are Sign and symptoms of Tinea:

- Enlarging raised red rings with a central area of clearing (ringworm).
- The edge of the **rash** appears elevated and is scaly to touch.
- Sometimes the skin surrounding the rash may be dry and flaky.
- There will be hair loss in areas of the infection.

### c. Diagnosis

Superficial scrapes of skin examined under a **microscope** may indicate the presence of a **fungus**. Diagnostic method called KOH Test is used, where the skin scrapings are placed on a slide and immersed on a drop of potassium hydroxide solution to dissolve the keratin on the skin scrapings thus leaving fungal elements such as hyphae, septate or yeast cells viewable. If the skin scrapings are negative and a fungus is still suspected, the scrapings are sent for culture. Because the fungus grows slowly, the culture results do take several days to become positive.

#### d. Prevention

Basic prevention measures include:

- Serious washing of hands after handling animals, soil, and plants.
- Avoiding touching characteristic lesions on other people.
- Put on loose-fitting clothes.
- Promoting good hygiene when participating in sports that involve physical contact with other people.

#### e. Treatment

Application of topical antifungals creams to the skin. In extensive or difficult cases, systemic treatment with oral medication may be required. Among the available prescription drugs are tolnaftate, terbinafine, naftifine, itraconazole.

#### 15.3.8. Hookworm

Hookworm is a humans' intestinal parasite. The adult worms and their larvae can cause intestinal disease in which they live.

#### a. Cause of hookworm disease

The hookworm is caused by two main species of hookworm infecting humans which are: *Ancylostoma duodenale and Necatora mericanus* 

#### b. Method of transmission

If an infected person defecates near bushes, in a garden, or field, or if the feces from an infected person are used as fertilizer, eggs are deposited on soil. They can then mature and hatch, releasing larvae (immature worms). The larvae mature into a form that can penetrate the skin of humans. Hookworm infection is transmitted primarily by walking barefoot on contaminated soil.

### c. Signs and symptoms of hookworm

- Itching and a localized rash are often the first signs of infection. These symptoms occur when the larvae penetrate the skin.
- A person with a light infection may have no symptoms but a person with a heavy infection may experience abdominal pain, diarrhea, loss of appetite, weight loss, fatigue and anemia (pale skin etc.) and protein deficiency caused by blood loss, constipation, congestive heart failure, excessive coughing during larvae migration, stomach or chest pain, vomiting, weight loss.
- The physical and cognitive growth of children can be affected. There is a decreased rate of growth and mental development in children (caused by protein and iron deficiency)

### d. Diagnostic method

Taking a stool sample and using a microscope to look for the presence of hookworm eggs.

#### e. Prevention of hookworm

Avoid walking barefoot in areas where hookworm is common and where there may be fecal contamination of the soil.

Avoid skin-to-soil contact and ingesting such soil.

Avoid defecating outdoors or using human feces as fertilizer, and by effective sewage disposal systems.

#### f. Treatment for hookworm

Hookworm infections are generally treated for 1-3 days with medication prescribed by your health care provider. The drugs are effective and appear to have few side effects. Iron supplements may be prescribed if you have anemia.

#### g. Preventive treatment

- In developing countries, groups at higher risk for soil-transmitted infections

- are often treated without a prior stool examination. Treating in this way is called preventive treatment.
- School-age children are often treated through school-health programs and preschool children and pregnant women at visits to health clinics.

### h. Life cycle of Hookworm

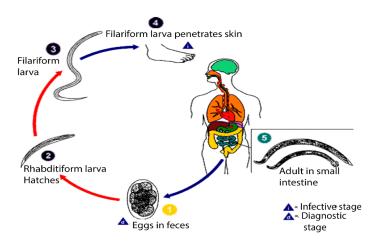


Figure 15.6: Life cycle of Hookworm

#### Self-assessment 15.3

- 1. Which of the following diseases is transmitted by an insect vector?
  - a. Cholera
  - b. HIV/AIDS
  - c. Malaria
  - d. TB
- 2. What are the ways in which cholera is transmitted from person to person?
- 3. Explain why there is such a high risk of cholera following natural disasters such as earthquakes, hurricanes, typhoons and floods.
- 4. Explain why there is a high death rate from TB in countries with a high proportion of the population who are HIV-positive.
- 5. TB is an opportunistic infection. Why?
- 6. Describe how malaria is transmitted.

# 15.4. Health and community: criteria for good housing

### **Activity 15.4**

Housing quality is associated with morbidity from different factors. State any three factors

Housing refers to houses or buildings, accommodation of people. It is an important determinant of health, and substandard housing is a major public health issue.



Figure 15.7: Grass thatched houses, examples of poor housing quality nowadays eradicated from countrywide land.

The public health community is aware of the importance of social determinants of health (including housing) in recent years. Yet defining the role of public health practitioners in influencing housing conditions has been challenging. Responsibility for social determinants of health is seen as lying primarily outside the scope of public health. The quality and accessibility of housing is, however, a particularly appropriate area for public health involvement.

An evolving body of scientific evidence demonstrates solid relations between housing and health. The public health community is developing, testing, and implementing effective interventions that yield health benefits through improved housing quality.

### Criteria for good housing

- Good housing must be well equipped.
- Good housing must be well localized
- Water and electricity
- Big size and ventilated





Figure 15.8: Model village well equipped, with water and electricity



Figure 15.9: Modern house, example of standard housing quality nowadays for promotion of good health

An increasing body of evidence has associated housing quality with morbidity from infectious diseases, chronic illnesses, injuries, poor nutrition, and mental disorders.

#### Self-assessment 15.4

What measures are taken by Rwanda government to ensure high quality of housing conditions?

### 15.5. Public health services

### **Activity 15.5**

- 1. Suppose that you are one of Rwandans who have food industry in our country, and you expect your production to be inspected. Outline the main requirements for good production to be inspected?
- 2. Clean water is good for health. Discuss the ways you would use to obtain clean water at home.

# 15.5.1. Food inspection requirement

### a. Food inspection services

Food inspections services help to let you have a complete check of your running production in factories or across the country. An inspector performs a random selection checking on quantity, packing, labeling, dimension, weight and visual aspects. Inspection allows to spot inconsistencies in your production lots before they leave the factory: you can react timely and avoid costly rework, sorting or recalls.

# 15.5.2. Need for control of housing conditions

Living conditions affect people's lives, be it at home or the workplace. Without good living conditions, people's health and work will be affected. Nowadays, the quality of housing accompanied with good housing facilities is now improving.

#### 15.5.3. Need for control of clean water.

For being healthy, only clean water must be used. Different materials are used to clean water. They include "Sûr–eau", heating or by using water purification etc. The steps of water purification are: storage reservoir, aeration, filtration, disinfection, reduction of chlorine concentration, covered service reservoir, distribution.

### 15.5.4. Need for control of hygiene

Many diseases can be prevented from having a damaging effect on the body by the action of natural defenses, antibiotics or other medicines. However, there are many steps that can be taken by individuals, and by the community as a whole, to fight microbes even before they enter the body and cause disease. These steps concern personal hygiene (cleanliness) and sanitation (public cleanliness involving community efforts in disease prevention), both of which help to prevent disease.

Note that: The World of Health Organization defines health as a state of complete physical, mental and social well-being and not merely the absence of disease and infirmity. The responsibility for good health lies in the hands of each individual in the community and the nation.

### a. Personal hygiene

- Hands frequently touch many things which may carry pathogens. They must be always washed using a soap before preparing food, eating and after a visit to a toilet.
- It is essential to bath frequently because sweat and oil secretions on the skin enable bacteria and fungi to breed easily. This helps to prevent skin infections.
- Bath towels and sponges should not be shared Combs and hairbrushes should not be shared.
- Hair should be washed frequently to avoid lice and mites. These can spread typhus fever.
- Teeth should be cleaned at least twice a day, preferably after each meal because the spaces between teeth where food particles are trapped provide excellent breeding grounds for bacteria.
- Clothes would be clean and changed frequently.
- Shoes should be worn to prevent cuts and infection by hookworms.
- Finger nails and toe nails must be always kept short and clean so that they do not provide breeding places for germs.

Exercise, recreation and rest are other factors that are important in promoting health.

#### i. Exercise

Exercise makes the muscles strong so that they can support the body better.

It helps to get rid of excretory materials and to improve digestion.

It quickens blood circulation and improves the action of the glands and nerves.

### ii. Recreation

- Gardening, playing games and reading can remove any dullness and mental tiredness resulting from everyday work.

### iii. Sleeping is the best form of rest.

- Adults need about eight hours of sleep a day. A great deal of repair of wornout tissues in the body and the building up of new ones takes place when body rests.
- Other good personal habits include avoiding smoking, alcoholic drinks and other drugs.

#### Lesson self-assessment 15.5

- 1. In which ways a personal cleanliness may be achieved.
- 2. How to promote a hygienic food preparation?
- 3. Discuss about good personal Hygiene.

### **End of unit assessment 15**

- 1. Answer to the following questions:
  - a. What does the germ theory of disease mean?
  - b. State any four causes of diseases in our life.
- 2. State any TWO diseases caused by:
  - a. Bacteria
  - b. Protozoa
  - c. Microscopic fungi
- 3. Match the following scientists with their scientific contributions:

Scientists	Scientific contributions
Alexander Fleming	A. Shows that living organisms only grow from other Living organisms. They do not arise spontaneously
	B. Discovers penicillin, the first antibiotic
Robert Koch	R H S CH <sub>3</sub> CH <sub>3</sub> O O O O O O O O O O O O
Louis Pasteur	C. Isolated a bacillus and showed that it caused anthrax
	D. Discovers how to weaken chicken cholera so that it could be used as a vaccine without causing infection
	E. Discovers the cholera bacillus

- 4. List the reasons why smallpox is easier to eradicate than AIDS.
- 5. Suggest reason why Malaria is endemic disease in tropics.
- 6. Describe the biological factors that make malaria a difficult disease to control.
- 7. Describe the precautions that people can take to avoid catching malaria.
- 8. Explain what is meant by ring vaccination.



### **UNIT 16: ASEXUAL REPRODUCTION IN PLANTS**

### **Key Unit Competence**

Account for various methods of asexual reproduction as means of increasing crop yield.

### **Learning Objectives**

By the end of this unit, I should be able to:

- Describe the various methods of asexual reproduction: fragmentation, budding, and spore formation.
- Demonstrate asexual reproduction mechanisms in lower organisms.
- Discuss the advantages and disadvantages of asexual reproduction.
- Describe the characteristics of vegetative reproductive parts in a flowering plant.
- Apply principles of artificial propagation in growing varieties of plants that are economically important.
- Apply the knowledge acquired to produce economically important plants.
- Appreciate the use of artificial propagation in increasing crop yields.

### **Introductory activity**

The kingdom Plantae comprises about 260,000 known species including flowering and non-flowering plants. All plants have a general organization which includes vegetative and reproductive organs. Plants reproduce through different ways: Use the books and other source of information to

- 1. Write on how lower organisms such unicellular plant and another like cassava, sugar cane and apple reproduce.
- 2. Describe the techniques used by people to grow Irish potatoes, cassava and bananas.
- 3. Describe each of the following methods of asexual reproduction: fragmentation, budding and spore formation.

# 16.1. Asexual reproduction

# **Activity 16.1**

Using textbooks or internet and search on asexual reproduction.

# **Asexual Reproduction**

This is a type of reproduction without production of gametes. It usually results in the production of identical offspring; the genetic variation arises as a result of random mutation among the individuals.

#### Self-assessment 16.1

Write an essay on asexual reproduction.

# 16.2. Types of asexual reproduction

### Activity 16.2.1

Discuss on asexual reproduction in lower organisms and write a report as an outof-class activity.

There are five common modes of asexual reproduction: fission, budding, vegetative reproduction, spore formation and fragmentation.

#### 16.2.1. Fission

An important form of fission is binary fission. In binary fission, the parent organism is replaced by two daughter organisms, because it divides in two, i. e: Unicellular Fungi. Another type of fission is multiple fission that occurs in many algae. The nucleus of the parent cell divides several times by mitosis, producing several nuclei. The cytoplasm then separates, creating multiple daughter cells.

### 16.2.2. **Budding**

Some cells split via budding resulting in a 'mother' and 'daughter' cell. The offspring organism is smaller than the parent. Budding is also known on a multicellular level.

# 16.2.3. Vegetative reproduction

Vegetative reproduction is a type of asexual reproduction found in plants where new individuals are formed without the production of seeds or spores. Examples of vegetative reproduction include the formation of miniaturized plants called plantlets on specialized leaves (for example in kalanchoe) and some produce new plants out of rhizomes or stolon (for example in strawberry). Other plants reproduce by forming bulbs or tubers (for example tulip bulbs and dahlia tubers). Some plants produce adventitious shoots and suckers that form along their lateral roots. Vegetative reproduction may form a clonal colony where all the individuals are clones.

# 16.2.4. Spore formation

Many multicellular organisms form spores during their life cycle in a process called sporogenesis. On the other hand, plants and many algae undergo sporic meiosis where meiosis leads to the formation of haploid spores rather than gametes. These spores grow into multicellular individuals called gametophytes. Fungi and some algae can also utilize true asexual spore formation, which involves mitosis giving rise to reproductive cells called mitospores that develop into a new organism after dispersal.

#### Activity16.2.2

Demonstration of asexual reproduction by fragmentation in algae

### **Requirements:**

Glass beakers of 500ml, Scalpel, Forceps, Pins, Spatula, Weighing balance, Labels, Artificial, fertilizers, Clear river water and Spirogyra (algae).

#### **Procedure**

- 1. Label five beakers of the same size as A, B, C, D and E. Pour water in each beaker. Weigh several measures of artificial fertilizers of 1 g each.
- 2. Transfer 1g of fertilizer to beaker A, then 2g to beaker B, 3g to beaker C, 4g to beaker D and 5g of fertilizers to beaker E. Note the concentration of fertilizers is increasing from A- E.
- 3. Using forceps pick spirogyra and put it on a tile. Add several drops of water to avoid drying. Tease off a piece of spirogyra using a pin. Cut that piece into 5 fragments of the same length and transfer each piece into the beaker.
- 4. Stand the beakers in a place where they can receive adequate sunlight for the seven days. On the next day, start to examine the fragments in each beaker every day and record any observable changes such the increase in size of the spirogyra.

Draw a table as this shown here and record your observation.

Beaker Rate of growth Reason

A
B
C
D
E

**Table 16.1: Rate of growth** 

# 16.2.5. Fragmentation

Fragmentation is a form of asexual reproduction where a new organism grows from a fragment of the parent. The fragment develops into a mature and fully grown individual. Fragmentation is seen in many organisms such as fungiplants and gemma in liverworts. Most lichens, which are a symbiotic union of a fungus and photosynthetic algae, reproduce through fragmentation to ensure that new individuals contain both symbionts. These fragments can take the form of soredia, dust-like particles consisting of fungal hyphen wrapped around photobiont cells.



Figure 16.1: Bryophyllum, a plant that reproduces asexually via new shoots from the leaves.

#### Self-assessment 16.2

- 1. Write a short essay on asexual reproduction.
- 2. What is fragmentation? give one example of plant which reproduces by fragmentation

# 16.3. Advantages and disadvantages of asexual reproduction

# 16.3.1. Advantages of asexual reproduction.

### **Activity 16.3**

Discuss on asexual reproduction in lower organisms and higher plants, and outlining advantages and disadvantages.

Since asexual reproduction does not involve the process of gamete formation. It can be completed much more quickly, allowing the individual to spread its genetic material in a shorter period of time. It is also an advantage for small populations. as there may not be an adequate number of plants to pollinate one another to reproduce. If the parent plants are successful, it is a good idea to make clones of the plant that can be successful as well.

# 16.3.2. Disadvantages of asexual reproduction.

Asexual reproduction does not have genetic diversity. With sexual reproduction, mixing a gene pool can produce diversity. The organism becomes less adapted to certain environmental changes when there is less variation produced in offspring. The entire communities of the genetically identical organisms might not survive such changes. Asexual reproduction usually leads to struggle for existence as well as  $overcrowding. The \, main \, distinction \, of a sexual \, reproduction \, from \, sexual \, reproduction \, distinction \, of a sexual \, reproduction \, distinction \, distinction \, of a sexual \, reproduction \, distinction \, distincti$ is that there is no need for two parents as well as special cells to reproduce. It means that special the mechanisms of combining sex cells for fertilization is no longer required. This form of reproduction process is simply mitosis that copy a parent organism.

#### Self-assessment 16.3

Explain the advantages and disadvantages of asexual reproduction.

# 16.4. Vegetative and artificial propagation in flowering plants

### **Activity 16.4**

Demonstration of asexual reproduction in plants by cuttings

### Requirements

Growth medium or moist soil, sweet potatoes vines, elephant grass, sugarcane or cassava stems, secateurs/sharp knife and rooting hormone.

#### **Procedure**

- 1. Collect clean and healthy stems from cassava, sugarcane or potato plants.
- 2. Using a secateurs/sharp knife, cut the stem of either cassava, sugarcane or sweet potato stems into fragments of suitable sizes.
- 3. Place them in either suitable medium of growth with rooting hormone if available or plant them in moist soil in the school garden.
- 4. Leave the set up for about 13 days, and then observe the development of roots and leaves at nodes.

Draw and record what you will observe after 13 days on the development of roots and leaves at nodes.

Artificial vegetative propagation is the deliberate production of new plants from parts of old plants. This can be done by following three methods: Cutting, layering, and grafting.

### a. Cutting

This is a simple procedure in which part of the plant called a 'cutting'is removed from the parent plant by cutting and placed in a suitable medium for grow. In this method, one-year-old stem of root is cut from a distance of 20 to 30 cm and buried in the moist soil in natural position. After sometime, roots develop from this cutting and it grows into a new plant. This method is commonly used in rose and sugar cane. Care is taken that nodes which were lower in parent plant (morphologically) are put in the soil, while the morphologically higher nodes are kept up. Adventitious roots are given off at the lower nodes.

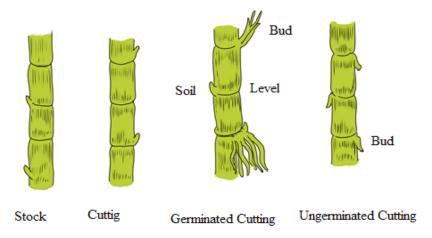


Figure 16.2 Cutting technique

### b. Layering

This method of vegetative propagation is used in those plants with soft branches such as jasmine plant. In this method, a branch of the plant which is near to the ground is pulled towards the ground and covered with moist soil leaving the tip of this branch above the ground. After sometime, roots develop from that part of the branch which was buried in the soil. This branch is then cut of along with the roots from the parent plant and develops into a new plant. This method of asexual reproduction is also used in the production of plants such as Bougainvillea, guava, strawberries, lemon, China rose etc.

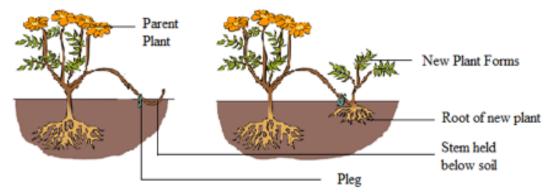


Figure 16.3: Vegetative propagation by layering.

### c. Grafting

In this method of vegetative propagation, the stems of two different plants are joined together so as to produce a new plant containing the characters of both plants. The plant of which the root system is taken is called 'stock', while the other plant of which the shoot is selected is known as 'scion' or 'graft'. These two stems i.e. the stock and the scion are fitted together by making slanting cuts into them and bound tightly with a piece of cloth and then covering it with a polythene sheet.

The diameter of the stock and scion chosen for grafting should be equal. Scion gets the mineral and water from the soil through the stock and develops branches and produce fruits. This method of propagation is used in mango, apple, banana, pear, grape, pineapple and peach.

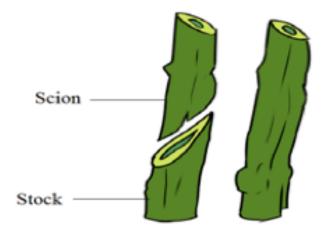


Figure 16.4: Vegetative propagation by grafting.

### Vegetative reproductive parts in flowering plants

The reproductive part of the plant is a flower. The union of male and female gametes to form a zygote is called fertilization. The transfer of pollen grains from the anther to the stigma of the same flower or the different flower is called pollination. Plants reproduce asexually in a variety of ways. The vegetative reproductive parts in flowering plant are stem, branches, and leaves and they have the following characteristics:

#### **Characteristics of Stem:**

Stem develops from the plumule of embryo. It is generally the ascending part of the plant axis. It bears a terminal bud for growth in length and is differentiated into nodes and internodes. The stem nodes possess dissimilar appendages called leaves. The young stem is green and capable of performing photosynthesis. In the mature state, stem bears flowers and fruits. Leaves and stem branches develop exogenously. Stem exposes leaves, flowers and fruits to their most suitable position in the aerial environment for optimum function. Stems usually show positively phototropic, negatively geotropic and negatively hydrotropic.

#### **Characteristics of Leaf**

- It is dissimilar lateral flattened outgrowth of the stem,
- The leaf is exogenous in origin
- It is borne on the stem in the region of a node,
- An axillary bud is often present in the axil of the leaf.
- Leaf has limited growth. An apical bud or a regular growing point is absent,
- The leaf base may possess two lateral outgrowths called stipules,

- A leaf is differentiated into three parts: leaf base, petiole and lamina.
- The lamina possesses prominent vascular strands called veins,
- It is green and specialized to perform photosynthesis,
- Leaf bears abundant stomata for exchange of gases and it is the major seat of transpiration.

#### **Characteristics of branches**

A branch is a woody structural member connected to but not part of the central trunk of a tree. Large branches are known as boughs and small branches are known as twigs. Due to a broad range of species of trees, branches and twigs can be found in many different shapes and sizes.

#### Self-assessment 16.4

- 1. Discuss on the methods of artificial vegetative propagation.
- 2. Cassava produces flowers, fruits and seeds. Why people prefer to grow cassava by cutting rather than using seed?
- 3. Describe the characteristics of vegetative reproductive parts in a flowering plant

# 16. 5. Application of artificial propagation in production of improved varieties of plants

# **Activity 16.5**

Use textbooks and other sources of information to discuss on application of artificial propagation in production of improved varieties of plants.

Artificial vegetative propagation is usually used in agriculture for the propagation (or reproduction) of those plants which produce either very few seeds or do not produce viable seeds. Such plants include: Banana, Pineapple, Orange, Grape, Rose, etc.

Reasons for preferring vegetative rather than sexual means of reproduction vary, but commonly include greater ease and speed of propagation of certain plants. Another major attraction is that the resulting plants are clones of the parent plants.

The methods of vegetative reproduction range from rooting cuttings to grafting and artificial propagation by laboratory tissue culture. In horticulture, a "cutting" is a piece that has been cut off from a mother plant and then caused to grow into a whole plant. A popular use of grafting is to produce fruit trees, sometimes with more than one variety of the same fruit species growing from the same stem. Rootstocks for fruit trees are either seedlings or propagated by layering.



Figure 16.5: Mass propagation of eucalyptus seedlings

#### Self-assessment 16.5

Explain the application of artificial propagation in production of improved varieties of plants.

#### **End of unit assessment16**

### A. Multiple choice questions: choose the best answers.

- 1. In vegetative propagation, cuttings are mainly taken from
  - a. Leaves of parent plant
  - b. Roots or stems of parent plant
  - c. Shoots of parent plant
  - d. Buds of parent plant
- 2. Artificial methods of vegetative propagation includes
  - a. Cloning
  - b. Grafting
  - c. Cuttings
  - d. Both b and c
- 3. Example of plant in which vegetative propagation is occurred by leaves is called
  - a. Cannabis
  - b. Chrysanthemum
  - c. Bryophyllum
  - d. Brassica
- 4. Which of the following is NOT an advantage of asexual reproduction?
  - a. Rapid reproduction.

- b. High genetic diversity.
- c. No need for a mate.
- d. Low resource investment in offspring.

### **B.Questions with short and long answers**

- 1. Name the plants which are grown by grafting method.
- 2. What do you understand by grafting?
- 3. Show how vegetative propagation takes place in potatoes?
- 4. Explain the method by which the sugarcane and rose are propagated.
- 5. Give the names of the different methods of artificial vegetative reproduction.
- 6. Explain the term vegetative reproduction and give one example of plant which reproduces by using this type of asexual reproduction.





### **UNIT 17: SEXUAL REPRODUCTION IN PLANTS**

### **Key Unit Competence**

Describe sexual reproduction in plants.

### **Learning objectives**

By the end of this unit, I should be able to:

- Explain the meaning of the term alternation of generations.
- Describe the types and structure of flowers.
- Describe pollination and fertilization in flowering plants.
- Explaining the events that takes place in a flower after fertilization.
- Describe the types and structure of seeds and fruits.
- Discuss the modes of dispersal of fruits and seeds.
- Observe and draw pollen grains.
- Draw and interpret floral formulae and diagrams.
- Relate the floral structures to the mode of pollination.
- Draw and label structures of fruits and seeds.
- Appreciate the role of pollinating agents in flowering plants.

### **Introductory activity**

- 1. Observe the following pictures and suggest what is going on.
- 2. How are the pictures below related to reproduction in flowering plants?





# 17.1. Alternation of generations in bryophytes and pteridophytes

### **Activity 17.1**

Using different resources to compare the life cycles of mosses and ferns.

**The life cycle of an organism** is the progressive sequence of changes which an organism goes through from the moment of fertilization to death. During its life cycle, the organism produces new generations of individuals which repeat continuously the process. New generations are produced by **reproduction**, which may be **sexual** or **asexual**. The life cycle involves the **mitosis** and **meiosis**. This unit concerns how **meiosis** can affect the life cycle of living organisms.

The life cycle is seen in seaweeds, mosses, ferns and their relatives. Their life cycles start with the sexually mature adult plants. Since they produce gametes, they are called the gametophytes (gamete plants). These are haploid, as they produce eggs and sperms. The egg and sperm fuse to produce a diploid zygote, but this does not develop directly into a new gametophyte. Instead it grows (by mitotic divisions) into another plant which is guite distinct from the gametophyte called **sporophyte** (spore plant). The function of sporophyte is to produce spores. In bryophytes, the sporophyte depends on gametophyte for nourishment. As spores are formed by meiosis, they are haploid. When the later are dispersed by wind on suitable soil, they germinate and grow by mitosis into gametophytes, which then repeat the sequence of life cycle. Ferns and mosses consist of two distinct plants: haploid gametophyte and diploid sporophyte, which alternate each other within the life cycle. This phenomenon is known as alternation of generation.

So, alternation of generation is a phenomenon in the plant life cycle in which a diploid stage a sporophyte alternates with a haploid stage of gametophyte.

The importance of alternation of generation to organisms:

- Spores produced can survive hash conditions and only germinate when conditions are favorable.
- It ensures rapid multiplication of the plant species as spores are usually produced in vast numbers.
- Interdependence between the gametophyte and sporophyte generations ensures that both generations exist at any given time. This prevents extinction of the plant species

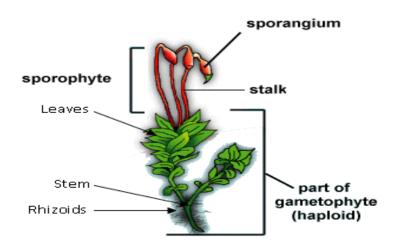
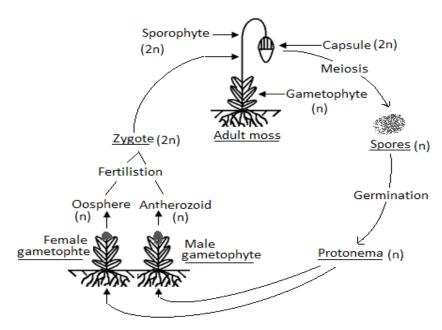


Figure 17.1: Moss plant.

The life cycle in mosses involves alternation of generations between the **diploid** (2n) sporophyte and haploid (n) gametophyte. In bryophytes, the gametophyte is a **dominant stage** of the life cycle, while the sporophyte is dependent on gametophyte for supplying water and nutrients. Gametes are formed in special reproductive organs at the tips of gametophytes. Sperms are produced in **antheridia** (**singular: antheridium**), and eggs in a**rchegonia** (**singular archegonium**). Some species produce both sperms and eggs on the same plant, whereas others produce sperms and eggs on separate plants.

During fertilization which requires water, the sperms released from antheridia fuse with the eggs and form a diploid (2n) zygote. The zygote grows into the sporophyte. Sporophyte is a long stalk ending in **a capsule** in which haploid **(n) spores** are formed by meiosis. When spores become mature enough, the capsule bursts and spores are scattered by wind. Under suitable conditions, spores then germinate by forming underground filaments called **protonemata (singular: protonema)**. Small buds produced by protonemata give rise to new gametophyte plants which can start the cycle again.



*Figure 17.2: Alternation of generations in bryophytes (mosses).* 

**Checkpoint:** why bryophytes grow in habitats where water is available constantly: For fertilization to occur, the sperm of bryophyte must swim to an egg. Without water, this movement is impossible. Because of this dependence on water for reproduction, bryophytes must live in habitats where water is available at least part of the year.

# 17.1.1. Alternation of generations in pteridophytes (ferns)

Pteridophytes also exhibit alternation of generations. Ferns are formed of true roots, stem and leaves (fronds) with vascular tissues. They have **lignified** tissues. The horizontal underground stem is called **rhizome** which bears adventitious roots. The leafy plant is a **sporophyte**. Mature leaves commonly called **fronds** bear yellow or orange masses of sporangia which are grouped into the structures called **sori** (singular: sorus) on their lower side.

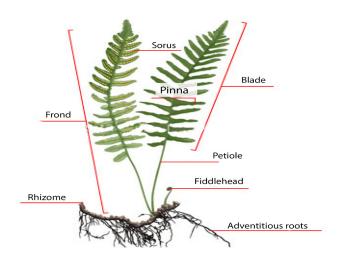


Figure 17.3. Diagram of ferns.

Ferns have a life cycle in which the diploid (2n) sporophyte is the dominant generation. Sporophyte is large (some fern trees can have 7 m of height), and differentiated into leaf, stem and roots with vascular tissues, while the haploid gametophyte (**prothallus**) is very simple with few millimeters. In ferns, the sporophyte produces haploid spores by meiosis. This is done on the underside of the leaf called **frond** in **sporangia** (singular: sporangium). Sporangia are grouped into **sori** (singular: sorus).

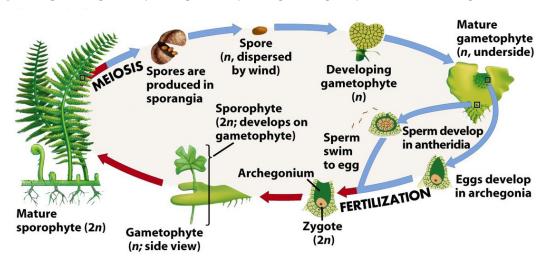


Figure 17.4: Alternation of generations in pteridophytes (ferns)

When spores are mature enough, the sporangia burst and spores are released on ground. If conditions are favorable, spores germinate and grow into the haploid heart-shaped gametophytes (n), which grows independently of sporophyte. Antheridia and archegonia found underside of gametophyte produce sperms and eggs respectively. During fertilization, sperms swim towards eggs and fuse together to form diploid zygotes, which grow and develop into new sporophytes. As sporophyte grows, the gametophyte dries and dies.

### 17.1.2. Differences between the mosses and ferns

Table 17.1: Comparison between moss and fern.

Mosses	Ferns
Have no vascular system	Have vascular system
No leaves, stems and roots	True leaves (fronds), stems, and roots
Gametophyte is dominant generation	Sporophyte is dominant generation
Spores in sporangia do not form clusters	Sporangia form clusters called sori
Transfer of sperm from antheridia to the archegonia occurs by rain-splash.	Sperms swim in water from the antheridia to an egg cell at the base of the archegonium

#### Self-Assessment 17.1

- 1. Explain the meaning of the term alternation of generation
- 2. Why is water essential in the life cycle of a bryophyte?
- 3. What are the archegonium and antheridium?
- 4. Why are these structures important in the life cycle of a moss plant?
- 5. What is the dominant stage of the fern life cycle?
- 6. Explain the relationship between gametophyte and sporophyte phases of the fern.
- 7. Compare gametophyte and sporophyte stages of the plant cycle. Which is haploid? Which is diploid?
- 8. How do bryophytes reproduce asexually?

# 17.2. Types and structures of flowers

#### **Activities 17.2**

Collect different forms of flowers from the school compound or around the school, such as hibiscus, morning glory, sweet potato, or maize flower:

- 1. Observe and describe the structures of collected flowers.
- 2. How do collected flowers differ externally?
- 3. Cut one of the flowers into two halves, draw and label one half of the flower.

## 17.2.1. Structure of a typical complete flower

A flower is a reproductive organ of a plant, which produces fruits and seeds.

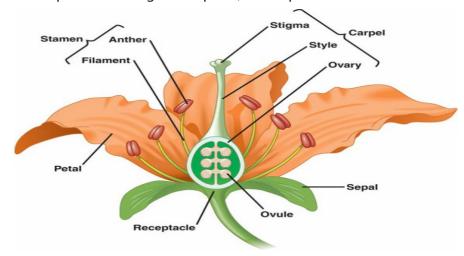


Figure 17.5: Diagram of a complete flower.

A typical hermaphrodite or bisexual flower contains the following parts:

- **Pedicel:** it is the stalk which attaches the flower on the main floral axis.
- Receptacle: it is the swollen part at the end of the stalk where other parts of the flower are attached.
- The calyx: it is the set of sepals, generally having green colour. They protect the internal parts of the flower. In some plants, the sepals are coloured and are called petaloids.
- The corolla: it is the set of petals, with different colours and nectar glands that produce sugary substances which participate in attraction of pollinating agents. In some plants, the petals are green and are called **sepaloids**. Both calyx and corolla are collectively called **perianth**. They form a floral envelope or accessory organs as they do not participate directly in reproduction, or in formation of fruits and seeds, they all insure the protection of internal parts of the flower.
- Androecium: is the male reproductive organs of the flower. It consists of many stamens. A stamen consists of: the filament which supports anther, and anther which contains the pollen grains or male gametes.
- **Gynoecium/pistil:** is the female reproductive organ. It consists of many carpels, and each carpel is made of: stigma (plural: stigmata), style and ovary with ovules.
- **a.** The stigma: receive pollen grains from anther during pollination.
- **b. Style:** maintains the stigma in a good position to receive pollen grains.
- c. Ovary: a sac where ovules are produced. Ovules become seeds after fertilisation.

## 17.2.2. Types of flowers

- 1. According to absence of some reproductive parts of the flower, we can distinguish:
  - **a.** Unisexual flower: is a flower that consists of one type of reproductive organ. This can be: **staminate**: unisexual male (with androecium only), or **carpellate**: unisexual female (with gynoecium only). E.g. flower of papaya.
  - **b.** Bisexual or hermaphrodite flower: a flower with both male and female reproductive organs (androecium and gynoecium). E.g. flowers of beans.

**Dioecious** plants are plants that have male flowers and female flowers on separate plants (e.g. papaya/pawpaw) while **monoecious** plants are plants that have both male and female flowers on the same plant (e.g. maize).

- 2. According to the position of ovary in the point of insertion of calyx, corolla and stamen, we can distinguish:
- a. A flower with inferior ovary: it is when the ovary is located below the point of insertion of calyx, corolla and stamens.
- **b.** A flower with superior ovary: it is when the ovary is located over the point of insertion of calyx, corolla and stamens.
- c. The semi-infer or semi-super flower: when ovary is neither inferior nor superior but in the middle of receptacle which is hollowed.
  - When sepals are joined together, the flowers are called gamosepal, and where are not joined together, the flower is called dialysepal.
  - When petals are joined together, the flowers are called gamopetal, and when are not joined together, the flower is called dialypetal. When they are absent, the flower is called apetal.
- 3. According to the shape and symmetry of the flower, we can distinguish:
  - **i. Zygomorphic or irregular flower:** a flower with a bilateral symmetry. The flower cannot be divided into two similar halves. E.g. flowers of beans, cassia.
  - ii. Actinomorphic or regular flower: a flower with a radial symmetry. The flower can be divided into two or more planes to produce similar halves. E.g. flowers of coffee, orange.

**Dichogany**: it is when male and female organs of the flower mature at different times. We can distinguish:

- 1. Protandry: when stamens mature before pistil.
- **2. Protogyny**: when pistil matures before stamen.

**Inflorescence** is when two or more flowers are borne on a common stalk.

# 17.2.3. Representation of the number and characteristics of a flower

There are two ways by which we can present the number and characteristics of different parts of the flower. These ways include: floral diagram and floral formula.

#### Floral formula

The floral formula indicates the number and characteristics of different floral organs. It varies from one flower to another. By convention, there are standard symbols that are used to represent different parts of the flower and their characteristics:

K: for calyx, K5: calyx with five free sepals, K (5): calyx with five fused sepals.

C: for corolla, C5: corolla with five free petals, C (5): corolla with five fused petals.

P: for perianth, P4: four free tepals, P (4): four fused tepals, P2+2: four tepals in two whorls of free each.

A: for Androecium or stamen, A5: androecium with five free stamens, A (5): five fused stamens, A5+5: ten stamens in two whorls of five each, A0: stamens absent, A ∞: stamens indefinite in number, A (9) +1: androecium of 10 stamens nine fused together and one free.

G: for pistil or gynoecium, G2: two free carpels, G (2): two fused carpels, G0: carpels absent, G (2): Bicarpellary, syncarpous semi-inferior ovary.

Representation of the symmetry of flower: zygomorphic or irregularflower: Actinomorphic or regular flower.

Representation of sex of the flower:  $\varnothing$ : staminate flower,  $\wp$ : pistillate flower,  $\wp$ : bisexual flower

The floral formula is specific to each species of plant. Examples:

## Write a floral formula of coffee having

- The bilateral symmetry
- Hermaphrodite flower
- 1 calyx with 5 fused sepals
- 1 corolla with 5 petals
- 5 fused stamens
- 1 pistil with 2 carpels each one with infer ovary of 2 chambers
- (5S) + (5P) +(5A) + 2C-2 or K (5) + C (5) + A (5) + G-2(2)

## Write a floral formula of Irish potato having

- 5 free sepals
- 5 free petals
- 5 free stamens
- 2 fused carpels with 2 chambers having many ovules
- 1 infer ovary
- Bisexual flower
- Radial symmetry.
- K(5) + C(5) + A5 + G-2(2) ∞

#### Self-assessment 17.2

- 1. What are the male and female structures of a flower?
- 2. What is the advantage for a plant to have many flowers together in a single structure?
- 3. Where does the female gametophyte develop?
- 4. Describe the flower and how it is involved in reproduction.

# 17.3. Pollination and double fertilization in flowering plants

## **Activity 17.3**

Use various resources to identify different pollinating agents and describe the process of double fertilization in flowering plants.

Pollination is transfer of pollen grains from anther to the stigma.

**Types of pollination**: there are two types of pollination such as: self-pollination and cross-pollination.

- **i. Self-pollination**: it is the transfer of pollen grains from anther to the stigma of the same flower, or of different flowers but of the same plants. It involves one plant. E.g. flowers of maize and beans.
- **ii. Cross-pollination:** it is the transfer of pollen grains from anther to the stigma of the flower of another plant. It involves two plants. E.g. flowers of pawpaw.

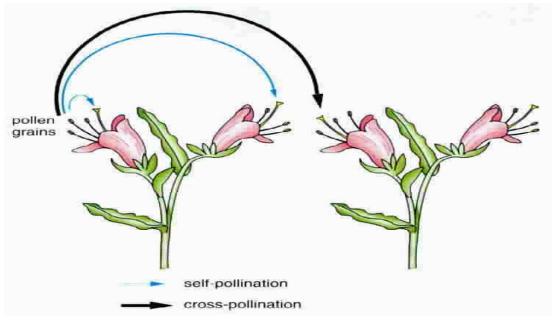


Figure 17.6:Self and cross-pollination

## 17.3.1. Main Pollinating agents

Flower structure is closely related with the way they are pollinated. This means that flowers are adapted to specific agents or mode of pollination. The common agents of pollination are: insects (entomophily), wind (anemophily), water (hydrophily), humans (anthropophily), and birds (ornithophily).

### Characteristics of insect-pollinated flowers: (entomophilous flowers):

- Flowers produce the nectar to attract pollinators.
- Flowers have a large brightly coloured corolla to attract pollinators.
- Production of scents to attract pollinators.
- The surface of the stigma should be sticky to hold pollen grains.
- Pollen grains are sticky and rough enough to remain on the surface of stigma.

### Characteristics of wind-pollinated flowers: (anemophilous flowers)

- The flowers have large stigma to hold pollen grains.
- The surface of the stigma should be sticky to hold pollen grains.
- Pollen grains are rough enough to remain on the surface of stigma.
- The flowers are or are not brightly-colored.
- They have or do not have scent.
- They do or do not secrete nectar.
- They produce large quantities of pollen grains, as much of them never reach the stigmas.

# 17.3.2. Double fertilization and events after fertilization in flowering plants

Double fertilization is a complex fertilization mechanism of flowering plants (angiosperms). This process involves the joining of a female gametophyte (megagametophyte, also called the embryo sac) with two male gametes (sperms).

# Development of pollen grains and plant ovules.

The pollen grains are produced in the anthers while the ovules are produced in the ovary.

## **Pollen grains**

Each anther has four pollen sacs which contain many diploid microspore mother cells that undergo **meiosis** to form four **microspores** each. At first, the four microspores remain together as tetrads. The nucleus of each microspore then divides by mitosis, forming a generative nucleus and a tube or vegetative nucleus. At this point, the content of the pollen grain may be considered as the male gametophyte. A twolayered wall forms around each pollen grain. The outer wall, the exine is thick and sculptured. The inner wall, the intine is thin and smooth. There are many pores or apertures in the wall through which a pollen tube may emerge.

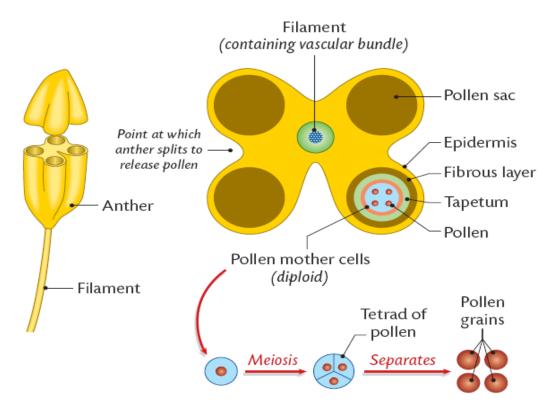


Figure 17.7: Development of pollen grains

### Plant ovule

Each ovule is attached to the ovary wall by a short stalk called **funicle.** 

The main tissue in the ovule is the **nucellus** which is enclosed and protected by the **integuments**. At one end of the ovule, there is a small pore called micropyle. A single diploid megaspore mother cell in the nucellus undergoes meiosis, producing four **megaspores**. Three of the four megaspores degenerate, while the remaining cell, called the embryo sac, grows to many times its original size. The nucleus of the **embryo sac** divides mitotically three times, resulting in eight haploid nuclei which are arranged in groups of four nuclei at the two poles. At this point, the contents of the embryo sac may be regarded at the female gametophyte.

One nucleus from each pole migrates to the centre of the embryo sac. These two **nuclei** are called polar nuclei, and they fuse to form a single diploid nucleus. Meanwhile, cell walls form around the remaining six nuclei and they form the **synergids**, antipodals and the **egg** (ovum). Only the egg functions as the female gamete.

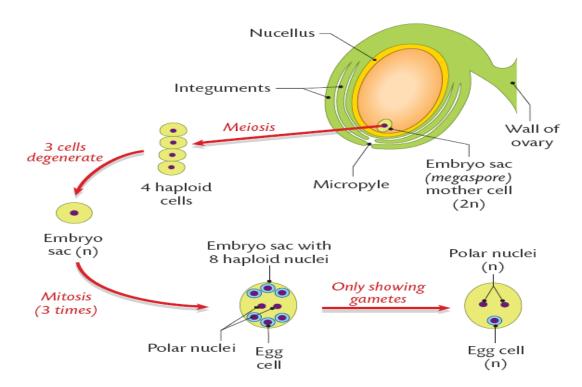


Figure 17.8: Development of plant female gamete

In summary, the pollen grain: contains two haploid nuclei: one called **generative nucleus**, and the other the **tube nucleus**.

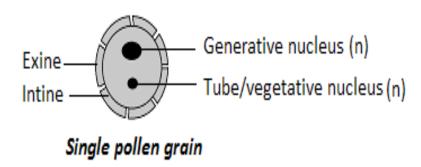


Figure 17.9: A pollen grain

On the other hand, the ovule or embryonic sac contains eight nuclei:

- Three antipodal nuclei/cells at one end
- Two polar nuclei/cells in the middle of ovule
- Two synergids (non-functional nuclei)
- One big egg cell.

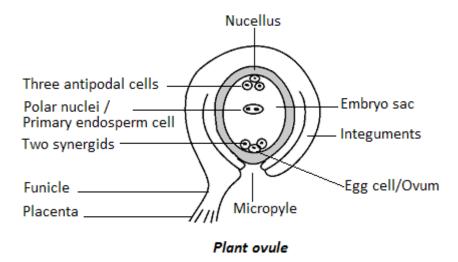
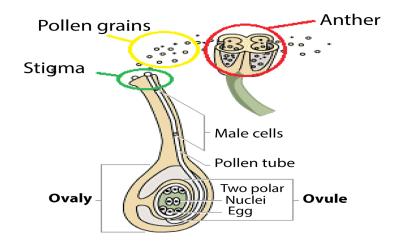


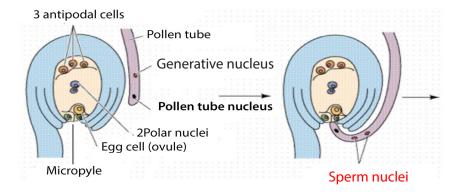
Figure 17.10: Structure of ovule

**The process of double fertilization:** It begins when a **pollen grain** adheres to the stigma of the **carpel**, the female reproductive structure of a flower. The pollen grain then takes in moisture and begins to **germinate**, forming a **pollen tube** that extends down toward the **ovary** through the style.

The growth of the pollen tube is controlled by the **pollen tube nucleus**. In the pollen tube, the generative nucleus divides mitotically into two haploid nuclei which are the **male gamete nuclei**. These follow behind the tube nucleus as the pollen tube grows down the style towards the ovule. The tip of the pollen tube then enters the ovary and penetrates through the **micropyle** opening, releasing the two sperms in the megagametophyte or ovule.

The tube nucleus degenerates, leaving a clear passage for the entry of male nuclei. *One nucleus fertilizes the egg* cell to form a **diploid zygote (2N)**, which will grow into a new plant embryo; the other fuses with polar nuclei to form a **triploid nucleus (3N)**, which will grow into a food-rich tissue known as **endosperm**, which nourishes the seedling as it grows.





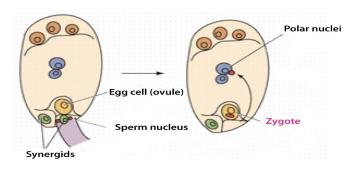


Figure 17. 11: The process of double fertilization.

This process is described as double fertilisation and is typical of angiosperms. If there is more than one ovule in the ovary, each must be fertilised by separate pollen grain and hence the fruit will have many seeds genetically different from each other.

#### a. Events in a flower after fertilization

After fertilisation, the calyx, corolla, stamens and style may wither gradually and fall off, but in some flowers the calyx may persist. The ovule forms the seed, the two integuments of the ovule will form the seed coat, and the ovary will develop into fruit, with the ovary wall forming the pericarp (fruit wall). The diploid zygote undergoes cell division to form the embryo, the triploid primary endosperm nucleus develops into endosperm, a store used by the developing embryo. This persists in endospermic seeds of monocotyledons. The micropyle persists as a small hole in the seed coat through which water is absorbed during germination.

**Floral Part** Fate after fertilization sepals, petals & stamens All wither and drop off fruit ovary ovary wall Fruit, Pericarp ovules Seeds Outer integuments Seed coat (Testa) Inner integumets Tagmen **Endosperm and Embryo** Embryo sac

Table 17.2: Floral parts and their fate after fertilization

### Self-assessment 17.3

- 1. Are angiosperms typically wind or animal pollinated? How does this process occur?
- 2. What is meant by the term endosperm?
- 3. What is the importance of brightly coloured petals to the plant?
- 4. What is double fertilization?
- 5. What happens to the antipodal cells and synergids cells after fertilization?

# 17.4. Structures and types of fruits and seeds

## **Activity 17.4**

Observe slides containing micrographs of different fruits and seeds. According to their characteristics:

- a. Differentiate fruits.
- b. Draw and show a structure of seed as seen on microscope

Below are some examples of fruits:



Figure 17. 12: Different fruits

A fruit is a structure formed from the ovary of a flower, usually after the ovules have been fertilized. It is normally produced only after fertilization of ovules has taken place, but in many plants, largely cultivated varieties such as seedless citrus fruits, grapes, bananas, and cucumbers, fruit matures without fertilization, a process known as parthenocarpy. Ovules within fertilized ovaries develop to produce seeds. In unfertilized varieties, seeds fail to develop, and the ovules remain with their original size.

A fruit consists of two main parts; **pericarp** (fruit wall) and the **seed.** The pericarp has three layers: epicarp or exocarp (outermost), mesocarpe (middle) and endocarp (inner).

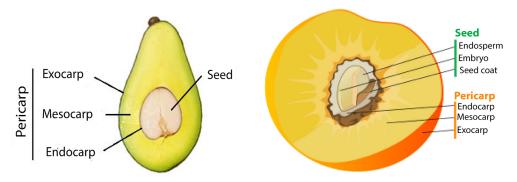


Figure 17. 13: Structure of a fruit

The fruit can have a dry pericarp or fleshy pericarp. The fruits with fleshy pericarp include: berry and drupe. Drupe is a fleshy fruit with only one seed,

E. g. avocado.



Figure 17. 14: Structure of a drupe

Berry is a fleshy fruit having many seeds inside of it. E.g. tomatoes, orange, and pawpaw.



Figure 17. 15: Structure of berry

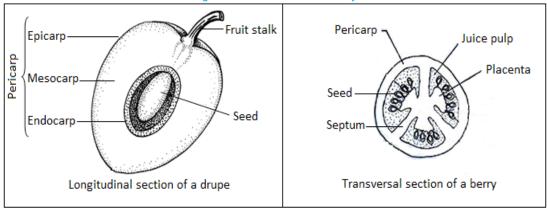


Figure 17. 16: Structures of drupe and berry

The fruits with dry pericarp include **indehiscent fruit or dehiscent fruit**. Indehiscent fruits do not open. Seeds remain inside of the fruits. E.g. fruits of coconuts. Dehiscent fruits open and release seeds. They include: dehiscent fruits with one carpel, and those with many carpels. Dehiscent fruits with one carpel include; those which open along one side, e.g. follicle; and those which open along both sides, e.g. legume (beans). Fruits of eucalyptus are examples of dehiscent fruits with many carpels.

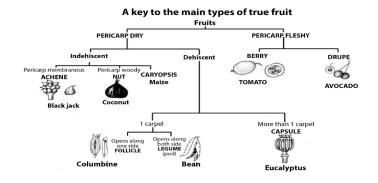


Figure 17. 17: Types of fruits

The major function of a fruit is the protection of developing seeds. In many plants, the fruit also aids in seed distribution (dispersal).

#### Food value

Fruits are eaten raw or cooked, dried, canned, or preserved. Carbohydrates, including starches and sugars constitute the principal nutritional material of fruits. Citrus fruits, tomatoes, and strawberries are primary sources of **vitamin C**, and most fruits contain considerable quantities of vitamin A and vitamin B. In general, fruits contain little protein or fat. Exceptions are avocados, nuts, and olives, which contain large quantities of fat, and grains and legumes, which contain considerable protein.

A **seed** is an embryonic plant enclosed in a protective outer covering. The formation of the seed is part of the process of reproduction in seed plants, the spermatophytes, including the gymnosperm and angiosperm plants. Seeds are the product of the ripened ovule, after fertilization by pollen and some growth within the mother plant. The embryo is developed from the zygote and the seed coat from the integuments of the ovule.

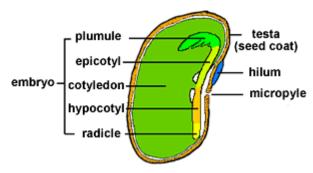


Figure 17. 18: Structure of a seed

The main components of the embryo are: seed made up of a seed coat (testa), one or two cotyledons and an embryonic axis. The embryonic axis is made up of a plumule, an epicotyl, a hypocotyl and a radical. A seed which has one seed-leaf is described as monocotyledonous and one which has two, as dicotyledonous. Maize is monocotyledonous seed while bean is a dicotyledonous seed.

- The cotyledons: the seed leaves attached to the embryonic axis. There may be one (Monocotyledons), or two (Dicotyledons). The cotyledons are also the source of nutrients in the non-endospermic dicotyledons, in this case, they replace the endosperm, and they are thick and leathery. In endospermic seeds, the cotyledons are thin and papery.
- The epicotyl: the embryonic axis above the point of attachment of the cotyledon(s).
- The plumule: It is located at the tip of the epicotyl and has a feathery appearance due to the presence of young leaf primordial at the apex. It will become the shoot upon germination.
- The hypocotyl: the embryonic axis below the point of attachment of the cotyledon(s) connecting the epicotyl and the radicle, being the stem-root transition zone.
- The radicle: the basal tip of the hypocotyl and it grows into the primary root.

Monocotyledonous plants have two additional structures in the form of sheaths. The plumule is covered with a **coleoptile** that forms the first leaf while the radicle is covered with a **coleorhiza** that connects to the primary root and adventitious roots form from the sides. Here the hypocotyl is a rudimentary axis between radicle and plumule.

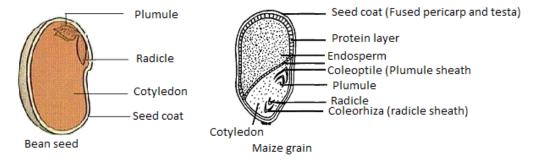


Figure 17. 19: Structure of dicotyledonous and monocotyledonous seeds

#### Self-Assessment 17.4

- 1. Describe the structure of a drupe
- 2. Differentiate between a drupe and a berry
- 3. What would happen to the fruit if ovules in the flower did not develop?
- 4. Compare the typical structure of seeds that are dispersed by animals to those dispersed by wind and water.

# 17.5. Fruits and seeds dispersal with their adaptations

## **Activity 17.5**

Use books and internet and to answer to the following questions:

- 1. Suggest the ways of fruits and seeds dispersal.
- 2. Explain adaptation of fruits dispersed by animals.

**Dispersal of fruits** and **seeds** is the scattering of fruits and seeds from their mother plants. They are four methods of seeds and fruits dispersal such as: (1) Dispersal by **Wind** (2) Dispersal by **Water** (3) Dispersal by Animals and (4) **Mechanical Dispersal.** Seeds dispersed by wind or water are typically lightweight, allowing them to be carried in air or to float on the surface of water. The wind carries also small seeds that have wing-like structure. Seeds dispersed by animals are typically contained in sweet and nutritious flesh fruits. They can be carried externally on their feet, fur, feathers, or beaks. Those seeds with hooks or sticky substances rely on the chance that they will attach themselves to a passing animal. Other seeds are eaten by animals and passed out in the faeces. These seeds will germinate where the faeces will be deposited.

With mechanical dispersal: all dehiscent fruits scatter the seeds when they burst. This dehiscence is accompanied by the expression of great force in many fruits so

that seeds are jerked at a considerable distance away from the mother plant. Such fruits are called explosive fruits.

The dispersal of seeds is important for the survival of the plant species because:

- It minimises overcrowding of plants growing around the parent plant that could then result in too much competition for nutrients and light;
- It allows the plant species to colonise new habitats which can offer suitable conditions.

### Self-assessment 17.5

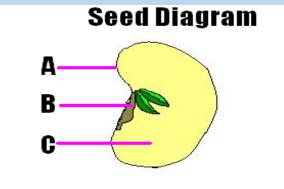
- 1. Why is it adaptive for some seeds to remain dormant before they germinate?
- 2. The seeds of a bishop pine germinate only after they have undergone a forest fire. Evaluate the significance of this structural adaptation.
- 3. Evaluate the importance of seed dispersal.

#### **End of unit assessment 17**

- 1. Answer by true or false
  - a. Mosses have life cycle that depends on water for reproduction.
  - b. In ferns, the gametophyte depends on sporophyte.
  - c. In mosses, the sporophyte dominates over the gametophyte.
  - d. Seeds that are dispersed by animals are not contained in a flesh-sweet tissue.
  - e. During pollination, pollen grains move from stigma to anthers.
- 2. Choose the letter that best answers the question or complete the statement.
  - a. Which of the following is not part of a flower?
    - i. Stamens
    - ii. Petals
    - iii. Carpels
    - iv. Stem
    - v. Sepals
  - b. Which is the structure of a flower that includes all parts listed below?
    - i. Stigma
    - ii. Carpel
    - iii. Ovary
    - iv. Style
    - v. Ovule

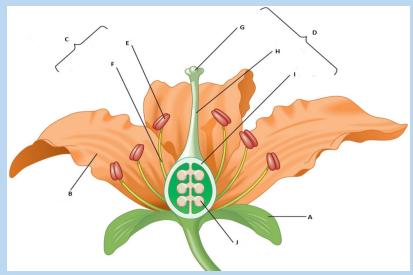
- c. The thickened ovary wall of a plant may join with other parts of the flower to become the
  - i. Cotyledon
  - ii. Fruit
  - iii. Endosperm
  - iv. Seed
- d. In angiosperms, the structures that produce the male gametophyte are called the
  - i. Pollen tubes
  - ii. Stigma
  - iii. Anthers
  - iv. Sepals
  - e. The small and multicellular structures by which liverworts reproduce asexually are
    - i. Archegonia
    - ii. Gemmae
    - iii. Protonema
    - iv. Rhizoids
  - f. In angiosperms, the mature seed is surrounded by a
    - i. Flower
    - ii. Fruit
    - iii. Cotyledon
    - iv. Cone
  - g. The leaves of ferns are called
    - i. Spores
    - ii. Fronds
    - iii. Sori
    - iv. Rhizomes
  - h. The most recognizable stage of a moss is the
    - i. Gametophyte
    - ii. Archegonium
    - iii. Protonema
    - iv. Sporophyte
- 3. Which are more likely to be dispersed by animals- the seeds of angiosperms or the spores of a fern? Explain your answer.
- 4. Pollination is a process that occurs only in seed plants. What is the process in seedless plants is that is equivalent to pollination?

- 5. What is the dominant stage of the ferns life cycle? Explain the relationship between gametophyte and sporophyte phases of the fern
- 6. Why is water essential in the life cycle of a bryophyte?
- 7. What is the characteristic responsible for the small size of bryophytes? Explain.
- 8. Briefly explain why a seed may remain dormant even when the environmental conditions are favorable for germination.
- 9. Describe the relationship between the gametophyte and sporophyte in mosses.
- 10. During the life cycle of a moss, what are the environmental conditions necessary for fertilization to occur?
- 11. Describe the dominant stage in the life cycle of a fern.
- 12. Propose a hypothesis to explain why angiosperms have become the dominant type of plant on the earth.
- 13. Moss plants are small. Ferns can grow as tall as small tree. Explain why. How does your answer illustrate a major characteristic of the plant kingdom?
- 14. Study the structure of the seed bellow

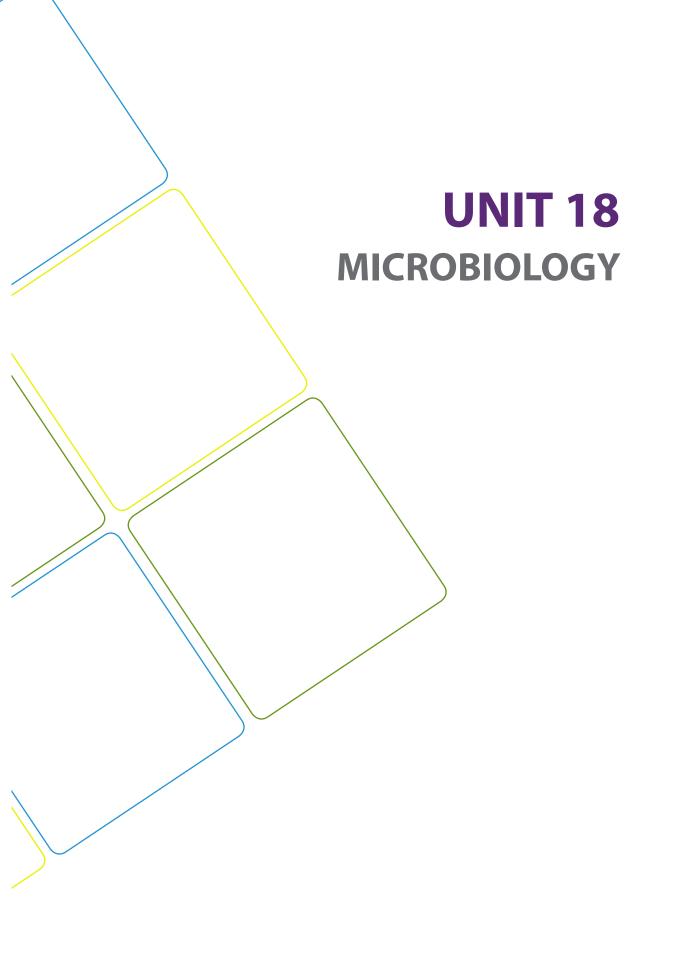


- a. Name the parts A, B and C
- b. What is the importance of the part C for a growing seedling?
- 15. Many flowers have bright patterns of coloration that directly surround the reproductive structures. Evaluate the importance of those bright-colored patterns to plants.
- 16. What is the function of endosperm?
- 17. Some plants form flowers that produce stamens but no carpels. Could fruit form on one of these flowers? Explain your answer.
- 18. Distinguish between pollination and fertilization.

19. Give names corresponding to the following to the letters: from A to J. Explain the function of the parts represented by: B, G, and E.



- 20. Explain why the relationship between bees and flowers is described as mutually beneficial.
- 21. What is the main advantage of cross-pollination?
- 22. Why are the stamens of wind-pollinated plants and insect-pollinated plants different?
- 23. Differentiate wind-pollinated flowers from insect-pollinated flowers.
- 24. Give one example of a plant that uses each of the following dispersal mechanism:
  - a. An explosive device which works by being inflated with water.
  - b. A winged seed lifted by air currents
  - c. A buoyant seed carried by sea currents
  - d. A gluey substance which sticks the seed to an animal.



## **UNIT 18: MICROBIOLOGY**

## **Key Unit Competence**

Describe the structure and characteristics of viruses, bacteria, and fungal and nonfungal moulds.

### Learning objectives

By the end of this unit, I will be able to:

- Describe the basic structure of viruses.
- Explain how a retrovirus reproduces.
- Identify the effects of viruses (e.g. AIDS, influenza, measles, feline leukemia, some human cancers) and prokaryotes (e.g. tuberculosis, bubonic plaque, cholera) on organisms.
- Describe how plant viruses can be transmitted.
- Explain how and why archaebacteria are thought to have been the first forms of life.
- Describe the structure and life cycles of Escherichia coli
- Relate the structures and functions of Prokaryotes
- Describe the structure of fungal and non-fungal moulds and explain how they reproduce
- Appreciate the importance of microorganisms in life.

## **Introductory activity**

A student left fresh milk in a cup exposed to the air. After 6 hours, he/she found that milk changed its state from fresh milk to stale milk. Why do you think this happened?

Mukamukiza prepared food for dinner. Some of the food was immediately put in tightly covered flask while the remaining food was left in the saucepan covered with banana leaves. In the evening, food in the flask was warm and safe while food in the saucepan has deteriorated. What is the cause of the food spoilage in the saucepan?

# 18.1. Introduction to microbiology.

# Activity 18.1.1

Discuss on the term microbiology and on the groups of microorganisms.

The term "microbiology" comes Greek words: 'micros' which means small, 'bios' which means life and 'logos' which means science. Microbiology is the study of microorganisms which are too small organisms to be seen with the unaided eye and require a microscope to be seen. They are also referred to as microbes. They include bacteria, fungi, algae, protozoa and viruses, they are useful to humans and they play a vital role in decay and recycling of nutrients in the environment. Some of them cause diseases

Micro-organisms are everywhere: in the air, water soil, on plants, on rock surfaces in very hot and cold places (ice). Before the invention of the microscope, microbes were unknown and thousands of people died in devastating epidemics because, vaccines and antibiotics were not available to fight against infectious diseases.

Nowadays, microorganisms can be grown in the laboratory and studied.

### a. The Prokaryotes

Prokaryotes can be categorized by their mode of nutrition and how they obtain energy and the carbon used to build the organic molecules that make up cells.

Organisms that obtain energy from light are called **phototrophs** and those that obtain energy from chemicals are called **chemotrophs**. Organisms that need only inorganic compounds such as CO<sub>2</sub> as a carbon source are called **autotrophs**. **Heterotrophs** require at least one organic nutrient such as glucose to make other organic compounds. Prokaryotes usually range in size from 1 to 5 micrometers making them much smaller than most eukaryotic cells.

## b. Classification of prokaryotes

Traditionally, bacteria have been classified based on their structure, physiology, molecular composition rather than on their evolutionary relationships. The bacteria that we generally refer to as germs are classified in the domain **Eubacteria**. More frequently, members of this kingdom are simply called **bacteria**. The other type of bacteria is known as archaebacteria. These bacteria, which are more ancient than the Eubacteria, are classified in the domain Archaebacteria. Taxonomists used to classify all prokaryotes in kingdom Monera, yet they slightly differ in characteristics.

#### 18.1.2. Archaebacteria and Eubacteria

## Activity 18.1.2

Discuss on the characteristics of given examples of both archaebacteria and Eubacteria.

#### a. Archaebacteria

Taxonomists treat archaebacteria as a separate kingdom because they are so different from other bacteria. Archaebacteria have unusual lipids in their cell membranes. Their cell wall is characterized by the absence of peptidoglycans, a protein carbohydrate compound found in the cell walls of Eubacteria. Archaebacteria were first discovered in extreme environmental conditions such as swamps, salt lakes, hot springs. Examples include:

## 1. Methanogens

- They have unique method of harvesting energy by converting H2 and CO<sub>2</sub> in methane.

 Methanogens can live only in anaerobic condition, such as the bottom of a swamp, and in sewage where they are the source of marsh gas, because oxygen is a poison to them.

### 2. Extreme halophiles

- These are salt-loving archaebacteria living in environment with very high salt concentration such as the Dead Sea. High salt concentration would kill most bacteria.
- These organisms use salt to generate ATP.

## 3. Thermoacidophiles

- This third group of archaebacteria lives in extremely acidic environments that have extremely high temperature such as hot springs. Thermoacidophiles live at 110°C and at a pH of 2.
- Thermoacidophiles live near volcanic vents on land or near hydrothermal

## How and why Archaebacteria are thought to have been the first forms of life?

The Archaebacteria comprise a group of single-celled microorganisms that, like bacteria, are prokaryotes that have no cell nucleus or any other organelles within their cells. They are known to have an independent evolutionary history and have numerous differences in their biochemistry compared to other forms of life.

Archaebacteria are now classified as in separate domain in the three-domain system by Carl Woese who introduced three main branches of evolutionary descent currently known as the Archaea, Eukarya and Bacteria. Classifying Archaea remains difficult, since many of them have never been studied in the laboratory and have only been detected by analysis of their nucleic acids.

#### b. Eubacteria

They occur in many shapes and sizes and have distinct biochemical and genetic characteristics. Eubacteria that are rod-shaped are called **bacilli**, sphere-shaped are called **cocci** (sing. Coccus) and spiral-shaped are called **spirilla** (sing. Spirillum).

- 1. The bacilli: bacteria with rod-shape. Ex: Clostridium tetani, Bacillus subtilis
- 2. Vibrios: comma-shaped with a single flagellum. eq: Vibrio cholera
- 3. The cocci: group of bacteria with spherical shape such as Streptococci. Cocci that occur in chains are Staphylococci which are grapelike clusters of cocci and Diplococci which is sphere shaped that are grouped two by two.
- **4. The spirilla**: bacteria with spiral shape. e.g.: Spirillum volutans.

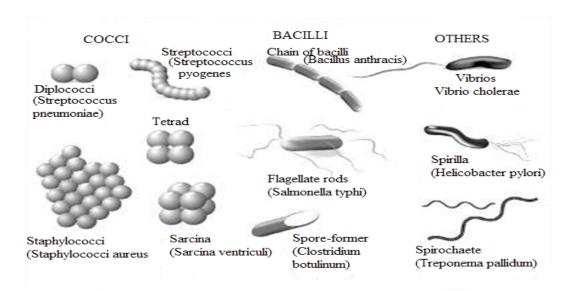


Figure 18.1: Shapes of bacteria cells

### 18.1.3. Gram stain

Bacteria have a peptidoglycan or **murein** cell wall that maintains cell shape, provides protection and prevents the cell from lysis. Based on the composition of the cell wall, bacteria can be classified as **Gram-positive** and **Gram-negative**. During the process of Gram staining, some bacteria without a lipid layer along with their peptidoglycan cell wall take the gram stain and appear violet (purple) and are therefore called gram positive. Example streptococcus and staphylococcus. Bacteria having a lipid layer along with their peptidoglycan cell wall do not take up the gram stain and are therefore called gram negative.

Example: Escherichia coli, Azotobacter, Salmonella.

#### Self-assessment 18.1

- 1. Describe the characteristics of the two domains of prokaryotes.
- 2. What factors can be used to identify prokaryotes?
- 3. How do bacteria maintain equilibrium in the environment?
- 4. Identify the parts of a prokaryote.
- 5. Describe briefly how some prokaryotes obtain their energy.

# 18.2. The structure and life cycle of Escherichia coli

## Activity 18.2.1

Using text books, videos or computer aided materials to describe the cycle life of E. coli.

E. coli reproduce asexually by undergoing binary fission. This type of reproduction begins with the replication of DNA molecule. Then, the copies of the genetic material attach themselves to the cell membrane. When the bacterium's size has doubled from its original size, the cell membrane starts pinching inward and a cell wall is produced between the two DNA molecules. Finally, the cell wall divides the cell into two daughter cells.

*E. coli* can also go through another process of reproduction known as conjugation. Conjugation is a reproduction process which involves the transfer of genetic material by the sex pili between two bacteria. This is not a sexual reproduction because there is no combination of gametes. The process of conjugation starts once the *E. coli, called a donor*, has finished to replicate its genetic material in form of a plasmid. The enzyme of the donor can now send signals to show that it is ready to mate. Once a mate is found, the donor attaches itself to the sex pilus of its mate. By doing so, the donor transfers the plasmid.

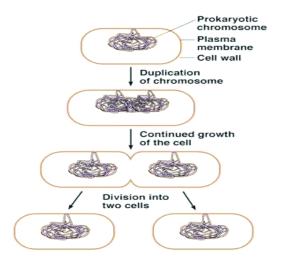


Figure 18.2: Binary fission in E. coli

# 18.2. E. coli and food poisoning

### **Activity 18.2.2**

Using textbooks to brainstorm the process of food poisoning, evolution of harmful strain of *E. coli* and food preservation

*E. coli* is a rod-shaped bacterium measuring about 2.5μm by 0.5μm. It is mainly found in guts of vertebrates. It is chemoheterotrophic, capable of thriving on a variety of the organic molecules. Its presence in water indicates contamination by faeces. *E. coli* reproduces asexually by binary fission. It can also take part in a primitive form of sexual activity called conjugation where genetic material is passed in one direction from bacterium to another through a pilus. Although conjugation does not in itself produce new offspring, after the process has finished, the bacteria reproduce asexually, passing on their new genetic make-up to their offspring.

## 18.2.1. Evolution of harmful strain of bacteria

E. coli was thought to be a relatively harmless resident of the human gut which might linked to the occasional upset stomach and mild diarrhoea. When massive colonies of mutualistic bacteria are present in the gut, including most strains of E. coli, they help to keep harmful bacteria away from starving them of food. They also help make vitamin K. But in 1982, it became clear that a new strain of E. coli had evolved into a much more troublesome organism. The strain had acquired a gene that enabled it to produce a powerful toxin which damages the intestinal wall, causing severe diarrhoea and internal bleeding.

This may lead to internal serious dehydration in young children and elderly people, and may result into death. In majority of the cases, infections of pathogenic strain of E. coli are not fatal and the disease clears without treatment.

### 18.2.2. Sources of infection

Touching a source of contamination and not washing hands before handling food may be sufficient to cause the infection.

In 1996, there was an outbreak which led to 20 deaths in Scotland due to contaminated meat. In the same period, another one was traced due to apple juice poisoning. Contaminated person can pass the bacteria on vegetables, and other foods. We must practice good habits of dealing and handling food to minimise cases of contamination. It is therefore, important to practice good hygiene. It is also essential to store and package food. It might be vital to pasteurise all fresh fruit juices just as milk is required to be pasteurised.

## 18.2.3. Food storage and packaging

The optimum storage conditions differ; raw meat and poultry are kept at around  $0^{\circ}$ c, meat products at 1°c - 40°c. Canned foods and many vegetables in dry conditions at 10°c - 150°c, and dried foods such as flour are stored, in air tight containers at 10°c 150°c. For long term storage, meat and fish are vacuum-sealed or can be vacuum. packed in laminated plastic containers. For pasteurisation, food and drinks such as milk are heated to a temperature that kills disease causing microorganisms. Example: Mycobacterium tuberculosis.

#### Self-assessment 18.2

- 1. Suggest the process by which *E. coli* reproduces.
- 2. What is the probable source of the gene that transforms harmless E. coli into pathogenic *E. coli*?
- 3. At what temperature is *E. coli* in meat killed?
- 4. How is food poisoned?
- 5. How can you minimise food and drink poisoning?

## 18.3. The structure and life cycle of viruses

### Activity18.3.1

Using textbooks, chart or videos to describe the structure, life cycle and effects of viruses.

The term "virus" was first used in the 1890s to describe agents smaller than bacteria that cause diseases. The existence of viruses was established in 1892, when, Russian scientist, **Dmitry Ivanovsky** discovered later microscopic particles known as the **tobacco mosaic virus** 

There are at least 3,600 types of virus. Hundreds of which are known to cause diseases in animals, bacteria, and plants. Viruses consist of an inner core of either **ribonucleic acid (RNA) or deoxyribonucleic acid (DNA)** plus a protein protective coat called capsid made of protein or of protein combined with lipid or carbohydrate components. An entire virus particle is called vibrios?

The core confers infectivity, and the capsid provides specificity to the virus. In some virions, the capsid is further enveloped by a fatty membrane. The later may cause virion inactivation by exposure to fat solvents such as ether and chloroform.

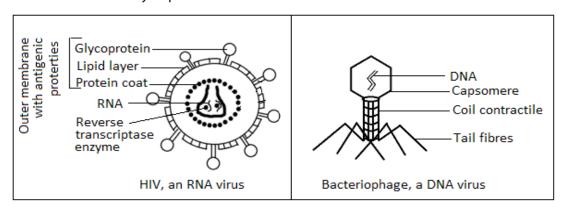


Figure 18.3: Structure of Virus

#### 18.3.1. Characteristics of viruses

- Viruses are complex biochemical molecules having the following characteristics:
- Viruses are not visible under light microscope because they are very small than bacteria.
- They possess a single type of nucleic acid either DNA or RNA enclosed in a protein coat.
- They can reproduce and grow inside the host cell.
- They have no cell and no cell organelles.
- They are obligate parasite i.e. cannot survive outside a host cell.
- They do not feed, respire and excrete.

## **18.3.2. Virus types**

DNA and RNA viruses differ in the way they use the host cell's mechanisms to produce new viruses.

For example, a DNA virus may act in one of the two ways:

The virus may directly produce RNA that is used to make more viral proteins or it may join with the host cell's DNA to direct the synthesis of new viruses.

RNA viruses replicate differently from DNA viruses. Upon entering the host cell, a viral RNA is released into the host cell's cytoplasm. There, it uses the host cell's ribosomes.

Some RNA viruses known as retroviruses contain an enzyme called reverse transcriptase in addition to RNA. Reverse transcriptase uses RNA as a template to make DNA. The DNA then makes an RNA transcript of itself. This RNA is then translated into proteins that become part of new viruses. Reverse transcriptase is so named because it reverses the normal process of transcription, in which DNA serves as a template for producing RNA.

## 18.3.3. Viral replication

Because viruses are not cells, they can replicate only by invading a host cell and using the enzymes and organelles of the host cell to make more viruses. Because they depend on host cells for replication, viruses are called obligate intracellular parasites. Outside the host cell, a virus is a lifeless particle with no control over its movements. It is spread by wind, water, in food, or via blood or other body secretions.

## 18.3.4. Life cycle of Bacteriophage

Bacteriophage is a virus that infects bacteria. Bacteriophage is composed of an icosahedral head that contains a nucleic acid. Beneath the head is a contractile tail that includes a collar and a sheath.

The contractile tail helps to inject the nucleic acid into the host cell. The tail rests on a base plate from which tail fibers emerge. These fibers assist the virus to attach to a host cell.

Viruses replicate by using either the **lytic cycle** or the **lysogenic cycle**:

# a. The lytic cycle

## **Activity 18.3.2**

Describe the sequence of events that occur during a lytic infection.

During the lytic cycle, a virus invades a host cell, produces new viruses, destroys the host cell, and releases newly formed viruses. Viruses that undergo the lytic cycle are called **virulent** because they cause disease. The lytic cycle consists of five phases:

 The Bacteriophage first attaches to susceptible bacterium by attaching its tail fibers to a receptor site. Receptor sites are specific sites that viruses recognize

- and attach to on the host cell's surface. If the Bacteriophage does not find a receptor site, it cannot infect the cell.
- Next the Bacteriophage releases an enzyme that weakens a spot in the cell wall of the host. Then the phage presses its sheath against the cell and injects its DNA into the host cell through the weak spot in the cell wall. The Bacteriophage leaves its capsid outside.
- The virus then takes control of the host's protein synthesizing mechanisms, transcribing mRNA from the viral DNA. The resulting Bacteriophage mRNA is translated on ribosomes and proteins that are synthesized form B a capsid. So the viral DNA is also replicated during this phase.
- Every replicated viral DNA is enclosed in the newly created viral capsid. The assembly of new virus particles usually occurs in the cytoplasm.
- During the last phase of the lytic cycle, one of the enzymes that are produced by the Bacteriophage genome causes the host cell to disintegrate, releasing new Bacteriophage. The cell disintegration is called lysis. In case of the enveloped viruses, the newly formed viruses move to the cell surface and force their way through the cell membrane.

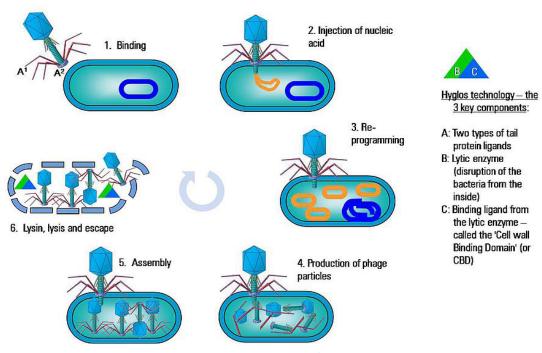


Figure 18.4: lytic Cycle of a Bacteriophage

The **first step** in the replication of the phage in its host cell is called **adsorption** or binding. The Bacteriophage adheres to the receptor site by means of its tail fibres. Following adsorption, the **phage injects its DNA** into the bacterial cell.

The tail sheath contracts and the nucleic acid or the core is driven through the wall to the membrane. This process is called **penetration** and it may be both mechanical and enzymatic.

Immediately after **injection** of the viral DNA there is **transcription** and translation of a section of the phage DNA to make a set of proteins that are needed to replicate the phage DNA and proteins that make up the capsid and the various components of the tail.

After making all viral parts, the **assembly** process follows. While the viruses are assembling, produced lysozymes are used to break down the cell wall peptidoglycans of the host bacteria. This is known as lysis and then mature viruses are released and spread to nearby cells for new infection.

### b. The lysogenic cycle.

### **Activity 18.3.3**

Using textbooks to describe what happens to the host cell infected by a temperate virus.

Some viruses can infect a cell without causing its immediate destruction. Such viruses stay in their host cell for an extended period of time: days, months or years in a lysogenic cycle. A virus that replicates through lysogenic cycle and does not kill the host cell immediately is called a temperate virus.

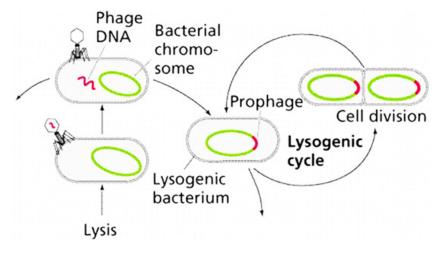


Figure 18.5. The lysogenic cycle

Retroviruses, such as HIV, have RNA that is transcribed into DNA by the viral **enzyme** *Reverse transcriptase* upon entry into the cell. (The ability of retroviruses to copy RNA into DNA earned them their name because this process is the reverse of the usual transfer of genetic information, from DNA to RNA). The DNA form of the retrovirus genome is then integrated into the cellular DNA and is referred to as the provirus. The viral genome is replicated every time the host cell replicates its DNA and is thus passed on to daughter cells.

## 18.3.5. Some common viral diseases

Table 18.1: Some common viral diseases

Name of disease	Cause	Signs and symptoms
Influenza	Myxovirus (DNA virus)	Sudden fever with headache, sore throat and muscular aches.
Common cold	Large variety of viruses, most common are rhinovirus (RNA virus)	Nasal and bronchial irritation, resulting in sneezing and coughing.
		Usually affects upper respiratory passages.
Measles	A paramyxovirus (RNA virus)	Occurs mainly in children. Sore throat, runny nose, watery eyes, cough and fever.
Mumps	A paramyxovirus (RNA virus)	Occurs mainly in children. Fever, followed by swelling of the parotid (salivary) glands on one or both sides lasting about 10 days. Testes, ovaries and pancreas may be affected.
Poliomyelitis (Polio)	Poliovirus (a picornavirus) (RNA virus)	Fever, headache and feeling of stiffness in neck and other muscles.
		Nerve cells and muscles are destroyed causing paralysis and muscle wasting. Most cases of paralysis occur in children aged 4 – 12 years, but adults may also be affected.
Yellow fever	An arbovirus, that is arthropod-borne virus (RNA virus)	Fever, headache, backache, nausea, tenderness in pit of stomach. Affects lining of blood vessels and liver.
AIDS	HIV virus; a retrovirus (RNA virus)	Loss of appetite, loss of weight, fevers, persistent dry cough,
Hepatitis B	DNA virus	Infects liver, flu-like symptoms, jaundice, nausea and severe loss of appetite.
German measles (Rubella)	Rubella virus	Occurs mainly in older children and adults. Affects respiratory passages, lymph nodes in neck, eyes and skin. Slight fever, body rash which disappears after three days.

## 18.3.6. Virus as living or non-living

### Activity 18.3.4

"Viruses are said to be on the border line of living organisms and non-living things". Discuss on this statement.

Viruses do not belong to any of the five kingdoms into which life is classified. It is difficult to say whether they are living or non-living.

### a. Features that make viruses to look like living things:

- They have the genetic material composed of either DNA or RNA They cause diseases to other living things: All viruses are infectious.
- They evolve as a result of mutation and natural selection.
- They reproduce /multiply only in other living things: they are obligate intracellular parasites

## b. Features that make viruses non-living things:

- They cannot metabolize
- They crystallize in isolation.
- They cannot reproduce outside of host.
- They are not made of cells. This means that they have a relatively simple noncellular organisation.
- They cannot respond to stimuli
- They have one type of nucleic acid, either DNA or RNA. But living cells contain both DNA and RNA.

**Table 18.2: Comparison between viruses and cells** 

Characteristics of life	Virus	Cell
Growth	No	Yes
Homeostasis	No	Yes
Metabolism	No	Yes
Mutation	Yes	Yes
Nucleic acid	DNA or RNA	DNA and RNA
Reproduction	Only within host cell	Independently by cell division
Structure	Nucleic acid, protein coat and an envelope in some species	Cytoplasm, cell membrane, cytoskeleton and in eukaryotic cells, organelles.

#### Self-assessment 18.3

- 1. What are the parts of a virus?
- 2. Describe the two ways by which viruses cause infection.
- 3. Distinguish between Bacteriophage and a prophage.
- 4. What is meant by retrovirus?
- 5. What are the strengths and weaknesses of the tobacco mosaic virus hypothesis?
- 6. Which characteristic feature is common to all viruses?
- 7. How is a capsid protein important to the functioning of a virus?
- 8. What is the best way to protect humans against most viral diseases?

### **18.4.** Moulds

## **Activity 18.4**

Using text books or computer aided materials to describe the life cycle of bread mould.

Moulds pervade our world, living wherever moisture is present. Some are of great benefit to humans, providing antibiotics, acting as decomposers so that nutrients can be recycled, or taking part industrial processes. Other moulds cause diseases which lead to serious damage.

Moulds have cells arranged in long thread-like filaments, the hyphae, that form a mass called **Mycelium**. Moulds are usually considered as fungi, but mould may also be formed by filamentous bacteria, slime moulds, and water moulds. Therefore, there are two main types of moulds: fungal moulds and non-fungal moulds

# 18.4.1. Fungal moulds

All fungi that produce mycelia can be called moulds, but the term is usually used for an organism in which the mycelium forms the main body of the fungus. In the black bread mould Rhizopus and the pin mould Mucor, the mycelium consists of a tangled mass of hyphae with many nuclei. These hyphae are called coenocytic because the fungal tissue is not separated by cell walls.

Fungal hyphae have an outer cell wall made of chitin and inner lumen which contains the cytoplasm and organelles. A cell surface membrane surrounds the cytoplasm and sticks tightly to the cell wall.

Rhizopus and Mucor are **Saprotrophic**, obtaining their nutrients from dead organic material. Rhizopus nigricans and Mucor mucedo can live on bread but some species of Rhizopus feed on living plants, and Mucor commonly grows on rotting fruits and vegetables, in the soil or on dung.

Rhizopus and Mucor secrete hydrolytic enzymes onto their food source and digest

the food outside the organism and then absorb the soluble digestion products and assimilate them.

### a. Life cycle of Rhizopus and Mucor.

Rhizopus and Mucor belong to the fungal phylum **Zygomycota**. The phylum got its name because its members produce two kinds of spores: Sexual zygospores as well asexual sporangiospores.

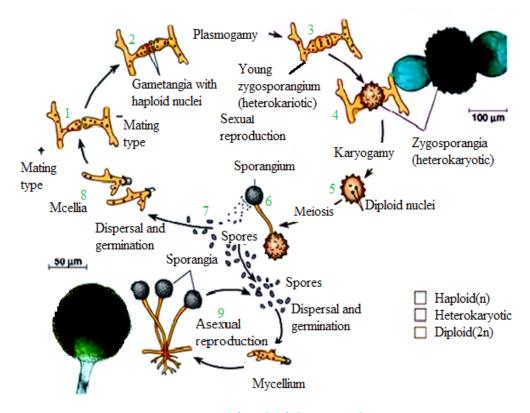


Figure 18.6. Life cycle of Rhizopus and Mucor

The asexual sporangiospores formed by mitosis, develop in **sporangium** at the tip of hyphae. When sporangium busts, the spores are released.

In most species of *Mucor*, the sporangium dissolves then water enters the spore mass, and the spores are dispersed by the raindrop or are transported by the insects. In most Rhizopus species, the sporangium wall fractures and dry spores are released by the wind.

The sexual reproduction involves **conjugation**. Usually the hyphae from mycelia of different mating types meet and interconnect via outgrowths. The interconnecting walls break down and their cytoplasm containing haploid nuclei mix, then the diploid zygote formed by the fusion of two nuclei develops a thick, rough, black coat and becomes a dormant zygospores. Meiosis probably occurs at the time of germination; the **zygospore** cracks open to liberate several haploids spores which can give rise to asexual sporangia and mycelia of either mating strain.

#### b. Use of moulds

Even if species of *Rhizopus* and *Mucor* are responsible for the spoilage of food, they are also useful as follow:

- They are used to make the human foods. For example, *Mucor* is used with soya beans to make a cheese called sufu, in eastern Asia. In Indonesia, R. *oligosporus* and R. *oryzae* are used to produce a food called tempeh from boiled skinless soya beans.
- The fungal moulds belonging to the Zygomycota are used to make anaesthetics, birth control pills, meat tenderisers, and the yellow colouring agents used in margarines and butter substitutes.

## 18.4.2. Non-fungal moulds

The following are different groups of non-fungal moulds:

- **a. Bacterial moulds**: including those of *Streptomyces griseous*, which secretes the antibiotic **streptomycin**
- **b. Slime moulds:** These are a peculiar group of organisms that resemble fungi in appearance and lifestyle, but are more closely to protoctists such as Amoeba in their cellular organization, reproduction, and life cycles. There are two types of Slime moulds:
  - **1. Plasmodial slime moulds** which have the following characteristics:
  - They have no connection with the parasitic protoctists belonging to the genus
     Plasmodium which causes malaria.
  - They exist as thin, streaming masses of colourful protoplasm that creep along moist, rotting logs and leaves.
  - They move in an amoeboid fashion, engulfing food particles by Phagocytosis.
  - A single mould may extend for many centimetres, but it is not multicellular.
  - They are made up of a continuous mass of cytoplasm with many nuclei called coenocytic mass.
  - **2. Cellular slime moulds** (also called Acrasiomycotae) which have the following characteristics.
  - They have a unicellular feeding stage resembling an amoeba, with each cell functioning individually.
  - When food is scarce, the individual cells group into a mass resembling that of Plasmodial slime moulds.
  - The individual cells of Cellular slime moulds retain their identity and have separate cell surface membranes

## c. Water moulds (Oomycota)

 Although water moulds and fungi are closed related and have a similar structure, water moulds are generally regarded as a separate and more ancient group belonging to the protoctists.

- Water moulds include rusts and mildews which consist of coenocytic masses of hyphae similar to fungi, for example Plasmodial slime moulds,
- Most water moulds have cell wall made of cellulose, while the cell wall of the true fungi is made of chitin.
- Some of the most devastating plant diseases are caused by water moulds. For example, the Phytophthora infestans causes potato blight, and Pythium which is a relatively unexpected parasite attacks a great variety of plants causing soft rot.
- Water moulds reproduce asexually by structure called conidia, and by moving spores with flagella, called zoospores.
- They reproduce sexually by producing moving male gametes that fertilizes large immobile egg cells. These egg cells give the group its name Oomycotae (where "Oo" means egg)

### Self-assessment 18.4

- 1. How are the cell walls of fungi similar to exoskeleton of insects?
- 2. Distinguish between hyphae and mycelium.
- 3. What are conditions necessary for fungal spores to germinate?
- 4. Explain the basis of classification of fungi.
- 5. Why do many biologists think that Penicillium evolved from an ascomycete?
- 6. Briefly describe sexual and asexual reproduction in fungi.
- 7. The antibiotic penicillin is a natural secretion of a certain kind of fungusgreen mould called Penicillium, penicillin kills bacteria. Why might a mould species have evolved way of killing bacteria?

# 18.5. Penicillium and Saccharomyces

# **Activity 18.5**

Make a research from the internet or textbooks to find out:

- 1. The structure of Penicillium, and yeast cell.
- 2. How saccharomyces reproduces.
- 3. The explanation of budding.

### 18.5.1. Penicillium and antibiotics

Penicillium is highly known for producing penicillin, the first antibiotic discovered in 1928 by a scientist **Alexander Fleming** when he was culturing some Staphylococcus bacteria during his medical research.

After leaving some Petri dishes for many days, he found a mouldy growth of Penicillium notatum contaminating a corner of one of dishes. Then Fleming realised that Staphylococcus next to the mould has been destroyed.

After studying Staphylococcus closely, Fleming concluded that the *Penicillium* mould was producing a substance that killed the Staphylococcus. He carried on with finding out if the broth of *Penicillium* mould contained penicillin which could destroy pathogenic bacteria.

In 1931, Fleming dropped his research. Howard Florey and Ernst Chain went on to produce purified penicillin. A successful work was reported 1940, and penicillin has been used to treat wounded soldiers in Second World War. In 1945, Fleming, Florey and Chain received the Nobel Prize for the discovery of penicillin.

#### a. The structure of Penicillium

*Penicillium* is **septate**; its hyphae have cross-walls called **septa.** However, the septa are not formed by cell division, and at the centre of septum there is a usually a pore which allows cytoplasm to flow from one compartment to another. Each compartment may contain one or more nuclei. Though Penicillium has septa, is a coenocyte like the non-septate moulds Rhizopus and Mucor.

*Penicillium* is saprotroph, feeding on organic matter in damp soil, leather, bread, and decaying fruit. The mycelia of *Penicillium* species form circular green, yellow, or blue moulds (depending to the species).

Penicillium reproduces asexually by means of spores called conidia formed at the tip of special hyphae called **conidiophores**.

Spores of *Penicillium* are exposed and free to be dispersed as they are mature.

# 18.5.1. Saccharomyces

### a. Definition and characteristics

- Saccharomyces is a genus of yeasts which include all unicellular fungi that reproduce asexually by budding.
- They occur commonly on faeces, in the soil, and on the surfaces of plants and animals.
- The most familiar and industrial important yeast is Saccharomyces cerevisiae.
- The tiny cells of this yeast are very active metabolically. They are usually aerobic but in the absence of oxygen they use anaerobic metabolism, producing carbon dioxide and ethanol (alcohol) as waste products which are industrially useful
- Each cell of Saccharomyces cerevisiae has a single nucleus and is usually egg shaped.
- Cells contain most of organelles of a typical eukaryote.

## b. Structure of yeast

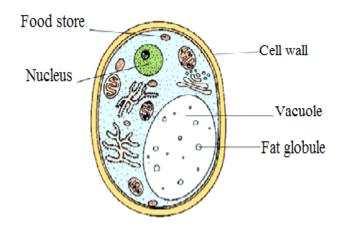


Figure 18.7. Structure of yeast

## c. Mode of reproduction

Saccharomyces cerevisiae can reproduce either asexually or sexually.

In **asexual reproduction**, the single cell divides by budding and separate into two cells. Some buds group together to form colonies; other separate to grow individually into a new yeast.

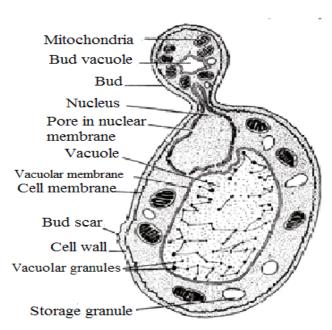


Figure 18.8. Saccharomyces cerevisiae. Sectional view of budding cell.

In sexual reproduction, two cells fuse to form a diploid cell which then forms haploid spores by meiosis

#### Self-assessment 18.5

- 1. Which feature does all yeast have in common?
- 2. How do hyphae of *Penicillium* differ from those of *Mucor*.
- 3. Describe the evidence for penicillin's effectiveness.

### 18.6. Protozoa that cause disease

### **Activity 18.6**

Observe prepared slides of *Entamoeba histolytica*, *Plasmodium* and *Trypanosoma* to compare their structures.

## 18.6.1. Entamoeba histolytica

## a. Characteristics of Entamoeba histolytica

Entamoeba histolytica is a protozoan parasite responsible for a disease called **amoebiasis**. It occurs usually in the large intestine and causes internal inflammation as its name suggests (**histo** which means tissue, **lytic** which means destroying). 50 million people are infected worldwide, mostly in tropical countries in areas of poor sanitation. Inside humans Entamoeba histolytica lives and multiplies as Trophozoites. Trophozoites are oblong and about 15–20 μm in length. In order to infect other humans, they encyst and exit the body.

# b. Life cycle Entamoeba histolytica

Entamoeba histolytica **life cycle** does not require any intermediate host. Mature cysts (spherical, 12–15 μm in diameter) are passed in the feces of an infected human. Another human can get infected by ingesting them in fecally contaminated water and food. If the cysts survive the acidic stomach, they transform back into Trophozoites in the small intestine. Trophozoites migrate to the large intestine where they live and multiply by binary fission. Both cysts and Trophozoites are sometimes present in the feces. Cysts are usually found in firm stool, whereas Trophozoites are found in loose stool. Only cysts can survive longer periods (up too many weeks outside the host) and infect other humans. If trophozoites are ingested, they are killed by the gastric acid of the stomach. Occasionally Trophozoites might be transmitted during sexual intercourse.

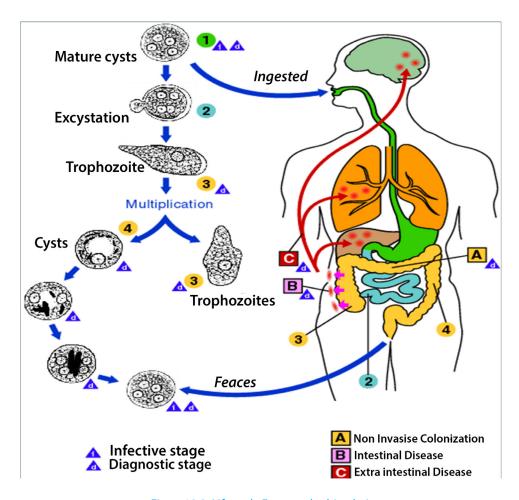


Figure 18.9. Life cycle Entamoeba histolytica

### c. Symptoms

Many Entamoeba histolytica infections are asymptomatic and Trophozoites remain in the intestinal lumen feeding on surrounding nutrients. About 10-20 % of the infections develop into amoebiasis which causes 70 000 deaths each year. Minor **infections** (luminal amoebiasis) can cause **symptoms** that include:

- Gas (flatulence) intermittent
- constipation loose stools
- stomach ache
- Stomach cramping.

Severe infections inflame the mucosa of the large intestine causing amoebic dysentery. The parasites can also penetrate the intestinal wall and travel to organs such as the liver via bloodstream causing extra-intestinal amoebiasis. Symptoms of these more severe infections include: Anemia, Appendicitis (inflammation of the appendix), bloody diarrhea, fatigue, fever, gas (flatulence), genital and skin

lesions, intermittent constipation, liver abscesses (can lead to death, if not treated), malnutrition, painful defecation (passage of the stool), peritonitis (inflammation of the peritoneum which is the thin membrane that lines the abdominal wall), pleura-pulmonary abscesses, stomach ache, stomach cramping, toxic mega-colon (dilated colon), Weight loss.

### d. Prevention

To prevent spreading the infection to others, one should take care of personal hygiene. Always wash your hands with soap and water after using the toilet and before eating or preparing food. Amoebiasis is common in developing countries. Some good practices, when visiting areas of poor sanitation:

- Wash your hands often.
- Avoid eating raw food.
- Avoid eating raw vegetables or fruit that you did not wash and peel.
- Avoid consuming milk or other dairy products that have not been pasteurized.
- Drink only bottled or boiled water or carbonated (bubbly) drinks in cans or bottles.

Natural water can be made safe by filtering it through an "absolute 1 micron or less" filter and dissolving iodine tablets in the filtered water.

### e. Methods of diagnosis

Amoebiasis is **diagnosed** by your health care provider under a microscope by finding cysts and (rarely Trophozoites) from a stool sample. The results are usually said to be negative, if *Entamoeba histolytica* is not found in three different stool samples. But it still does not necessarily mean that you are not infected because the microscopic parasite is hard to find and it might not be present the particular samples. A blood test might also be available but is only recommended, if your health care provider believes that the infection could have spread to other parts of the body. Trophozoites can be identified under a microscope from biopsy samples taken during colonoscopy or surgery.

# 18.6.2. Plasmodium spp.

#### a. Characteristics:

- Plasmodium is the genus of the class of Sporozoa that includes the parasite that causes malaria. Plasmodium is a type of protozoa, a single-celled organism that is able to divide only within a host cell.
- The main types of *Plasmodium spp* are *P.falciparum*, the species that causes falciparum malaria, the most dangerous type of malaria; *P. malariae*, the species that causes quartan malaria; *P. ovale*, a species found primarily in east and central Africa that causes ovale malaria; and *P. vivax*, the species that causes vivax malaria, which tends to be milder than falciparum malaria.

# b. Life cycle of *Plasmodium*

Plasmodium species exhibit three life-cycle stages gametocytes, sporozoites, and

#### merozoites.

Gametocytes within a mosquito develop into sporozoites. The sporozoites are transmitted via the saliva of a feeding mosquito to the human blood stream. From there, they enter liver parenchyma cells, where they divide and form merozoites. Inside the host's liver cell, the *Plasmodium* cell undergoes asexual replication. The products of this replication, called merozoites, are released into the circulatory system. The merozoites invade erythrocytes and become enlarged ring-shaped Trophozoites.

More erythrocytes are invaded, and the cycle is reinitiated. The merozoites are released into the bloodstream and infect red blood cells. Rapid division of the merozoites results in the destruction of the red blood cells, and the newly multiplied merozoites then infect new red blood cells. Some merozoites may develop into gametocytes, which can be ingested by a feeding mosquito, starting the life cycle over again.

The red blood cells destroyed by the merozoites liberate toxins that cause the periodic chill-and-fever cycles that are the typical symptoms of malaria. P. vivax, P. ovale, and P. falciparum repeat this chill-fever cycle every 48 hours (tertian malaria), and P. malariae repeats it every 72 hours (quartan malaria). P. knowlesi has a 24-hour life cycle and thus can cause daily spikes in fever.

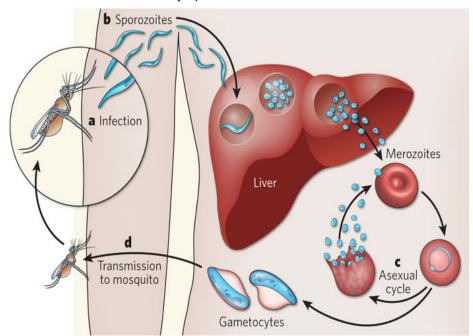


Figure 18.10. Cycle of Plasmodium.

# 18.6.3. Trypanosoma spp.

### a. Characteristics

- Trypanosoma is the genus containing a large number of parasitic species which

- infect wild and domesticated animals and humans in Africa.
- Commonly known as African sleeping sickness, human trypanosomiasis is caused by the species *Trypanosoma brucei* and is transmitted to humans through either a vector or the blood of ingested animals.
- The most common vector of *Trypanosoma brucei* is the tsetse fly, which may spread the parasite to humans and animals through bites.
- Through a process called antigenic variation, some trypanosomes are able to evade the host's immune system by modifying their surface membrane, essentially multiplying with every surface change. *Trypanosoma brucei* gradually infiltrates the host's central nervous system.

### b. Symptoms

Symptoms include: Headache, weakness, and joint pain in the initial stages; anaemia, cardiovascular problems, and kidney disorders as the disease progresses; in its final stages, the disease may lead to extreme exhaustion and fatigue during the day, insomnia at night, coma, and ultimately death.

### c. Occurrence

Human trypanosomiasis affects as many as 66 million people in sub-Saharan Africa. Trypanosomes are also found in the Americas in the form of *Trypanosoma cruzi*, which causes American human trypanosomiasis, or Chagas' disease. This disease is found in humans in two forms: as an amastigote in the cells, and as a **trymastigote** in the blood

### d. Mode of transmission

- The vectors for *Trypanosoma* cruzi include members of the order Hemiptera, such as assassin flies, which ingest the amastigote or trymastigote and carry them to animals or humans.
- The parasites enter the human host through mucus membranes in the nose, eye, or mouth upon release from the insect vectors. Left untreated, Chagas' disease may cause dementia, megacolon, and megaesophagus, and damage to the heart muscle, and may result in death.

## e. Life cycle of Trypanosoma

*Trypanosoma's* cell structure plays a vital role in allowing the cell to morph into three forms (trypomastigote, epimastigote, and amastigote) during its life cycle, depending on where the cell is located in the host's anatomy. The location of the kinetoplast in relation to the nucleus and the flagellum emergence dictate in which stage the trypanosome cell is found.

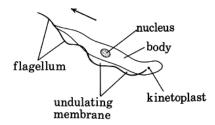


Figure 18.11: Cell Structure of Trypanosoma

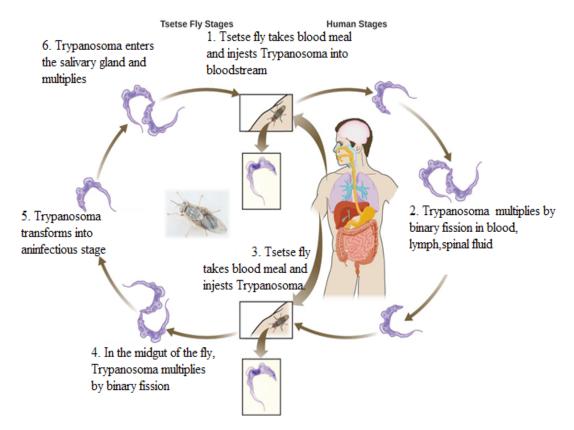


Figure 18.12. Life cycle of Trypanosoma

### **Role of Microbes**

Microorganisms are usually associated with major diseases such as AIDS, uncomfortable infections, or food spoilage.

However, the majority of microorganisms make crucial contributions to the welfare of the world's inhabitants by maintaining balance of living organisms and chemicals in our environment. Therefore, microorganisms are essential for life on earth. They have important beneficial biological functions such as:

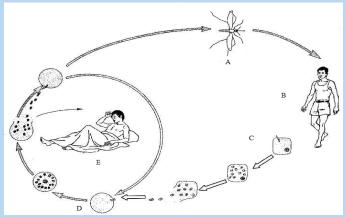
- 1. Photosynthesis: Marine and freshwater microorganisms (Algae and some bacteria) capture energy from sunlight and convert it to food, forming the basis of the food chain in oceans, lakes, and rivers and generates oxygen which is critical for life on Earth.
- 2. Decomposers: Soil microbes break down dead and decaying matter and recycle chemical elements that can be used by other organisms.
- **3. Nitrogen Fixation**: Some bacteria can take nitrogen from air and incorporate it into organic compounds in soil, water, and air.
- 4. Digestion: Human and many other animals have microorganisms in their digestive tract that are essential for digestion and vitamin synthesis. Examples include:

- Cellulose digestion by ruminants (cows, rabbits, etc.)
- Synthesis of Vitamin K (for blood clotting) and Vitamin B (for metabolism) in humans.
- **5. Synthesis of chemical products**: microorganisms have many commercial applications, such as the synthesis of acetone, organic acids, enzymes, alcohols.
- **6. Medicine**: many antibiotics and other drugs are naturally synthesized by microbes e.g. Penicillin is made by a mold.
- **7. Food industry**: many important foods and beverages are made with microbes e.g. vinegar, pickles, alcoholic beverages, green olives, soy sauce, buttermilk, cheese, yogurt, and bread.
- 8. Genetic engineering: recombinant microbes produce important
  - a. Medical and therapeutic products: human growth hormone, insulin, blood clotting factor, recombinant vaccines, monoclonal antibodies, etc.
  - b. Commercial products: cellulose, digestive aids, and drain cleaner.
- **9. Medical Research:** Microbes are well suited for biological and medical research for several reasons:
  - a. Relatively simple and small structures, easy to study
  - b. Genetic material is easily manipulated.
  - c. Can grow a large number of cells very quickly and at low cost.
  - d. Short generation times make them very useful to study genetic changes.

Though only minority of microorganisms is pathogenic (disease-causing), practical knowledge on microbes is necessary for medicine and related health sciences. For example, hospital workers must be able to protect patients from common microbes that are normally harmless but pose a threat to the sick and injured.

### Self-assessment 18.6

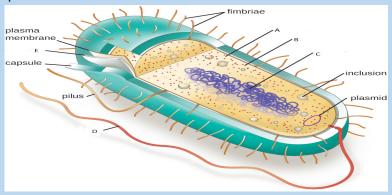
- 1. Name the causative agent of malaria.
- 2. The diagram below shows the life cycle of plasmodium. Analyse it and then answer the questions that follow.



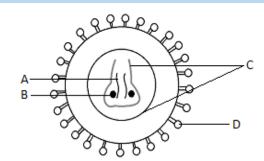
- a. What is the vector of malaria?
- b. Between stages C and D, which one takes place in the red blood cells and which one takes place in the hepatic cell (liver)?
- c. State any two symptoms of malaria displayed in individual in stage E.

### **End of unit assessment18**

- 1. State any TWO diseases caused by:
  - a. Bacteria
  - b. Protozoa
  - c. Microscopic fungi
- 2. What is the main feature of moulds?
- 3. Why viruses are not generally considered to be living things?
- 4. The figure below shows the structure of a bacterial cell seen using an electron microscope.

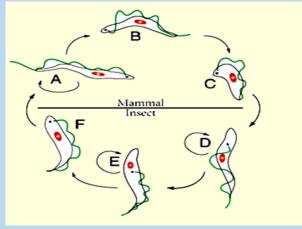


- a. Name the parts labeled A, B, C and D
- b. Describe the roles of parts B, C and E
- 5. The diagram below represents the structure of the human immunodeficiency virus (HIV/AIDS).

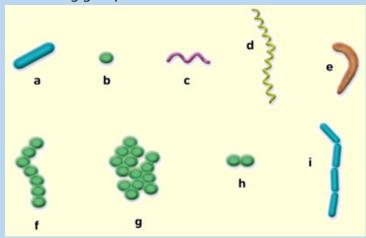


- a. Name A, B, C, and D.
- b. HIV/AIDS is under retroviruses. What is meant by retroviruses?
- c. What type of leucocytes (white blood cells) are destroyed by HIV/AIDS?
- 6. Discuss the methods of reducing the risk of food poisoning by pathogenic bacteria
- 7. Why the hyphae of Mucor is called coenocytic?

8. The figure below shows the life cycle of one of microorganisms.



- a. Which is the name of the microorganism having the life cycle represented on this diagram of?
- b. Name the parts labelled A, B, C, D, E and F
- 9. Identify the following groups of bacteria







## **UNIT 19: CULTURING MICRO-ORGANISMS**

### **Key Unit Competence**

Explain the process of culturing microorganisms and the factors affecting their population growth.

### Learning objectives

- List and describe the roles of microorganisms and their requirements for arowth.
- Explain the role of environmental variables in culturing microorganisms.
- Describe the different types of culture media.
- Draw and interpret the graph of the population growth of bacteria.
- Carry out an experiment to stain bacteria for examination with a light microscope.
- Observe and compare the numbers of bacteria present in fresh and stale milk.
- Distinguish between gram negative and gram positive bacteria.
- Describe the main features of aseptic techniques.
- Explain how pure cultures of pure bacteria can be obtained.
- Describe the methods of inoculation.
- Use sterile techniques to prepare agar plates to culture bacteria and fungi
- Carry out research on why microorganisms are particularly suitable for industrial use.
- Appreciate the importance of culturing microorganisms.
- Show perseverance when inoculating a solid and liquid medium.
- Show concern for taking the basic precautions in the school laboratory when carrying out routine microbiological work.

# Introductory activity.

Use different books and visit internet make a short summary about the culture of microorganisms and suggest why cultures are not incubated at 37° C in a school lab.

# 19.1. Requirements for culturing of microorganisms

# **Activity 19.1**

Use textbooks and other sources of information to discuss the requirements of growth of microorganisms.

Many microorganisms can be grown in the laboratory. This allows scientists to learn a lot about them. We can find out which nutrients they need to survive and which chemicals will kill them. We can also discover which microorganisms can be useful to us and which cause deadly disease.

To find out more about microorganisms, you need to culture them. Culturing microorganisms involves growing very large numbers of them so that you can see the colony as a whole.

To culture microorganisms, you must provide them with everything they need. This usually involves providing a culture medium containing carbohydrates to act as an energy source. A long with this, various mineral ions some supplement of proteins and vitamins are included.

The nutrients are often contained in an agar medium. Agar is a substance which dissolves in hot water and sets to form a jelly. You pour hot agar containing all the necessary nutrients into a Petri dish. Microorganisms are living organisms. Therefore, they have requirements for their growth, maintenance and multiplication. These include:

- Optimum temperature (30-40°C) for enzymes to work better.
- Source of energy such as glucose, maltose, juice.
- Source of other nutrients (minerals such are as potassium, sodium, iron, magnesium and calcium, vitamins, proteins
- Air for aerobic microbes or complete absence of air for anaerobic microorganisms.

The medium for culture of microbes can be the dead organic matters (food, fruits, remaining of organism, juice, milk) or a prepared medium such as Agaragar (universal medium for any germ), Lowenstein medium (selective medium for tuberculosis bacillus). The medium can be wet or dry. Different types of media are used culture microorganisms.

# 19.1.1. Types of media

There are many different types of media described by their components or ingredients.

Universal media: this allow the growth of every type of bacteria e.g. agar-agar

Differential/selective media: are specific to some types of bacteria for example Lowenstein for tuberculosis bacteria. Their ingredients will favour growth of certain types of bacteria.

A pure culture: this contains only one kind of microorganism. The pure cultures are important for scientific method as they are free from other types of microorganisms.

# 19.1.2. Principles of sterile culturing

- Wash hands before touching a sterile Petri-dish
- Open the Petri-dish as little as possible, and replace the lid quickly
- Never cough or sneeze near the dish
- Never touch the infected iiffy with fingers
- When culturing is no longer required, they should be flooded with strong disinfectant

- After cleaning out the nutrient from Petri-dish, they should be washed and disinfected, and then if they are glass, heat sterilize.
- Wash your hands thoroughly after all operation by using soap.
- Never push hands near the mouth during experimental work.

### Safety precautions:

Bacteria grow and reproduce more quickly when they are warm than when they are cold. It would be dangerous to incubate cultures at temperatures close to body temperature (37°C) because doing so might allow the growth of pathogens harmful to health. So the maximum temperature used in school and college labs is 25°C. However, higher temperatures can be used industrially, and these produce faster growth.

### Self-assessment 19.1

- 1. What is meant by the term culturing bacteria?
- 2. What do bacteria need to grow?
- 3. Why do we culture microorganisms in the lab?
- 4. Explain why cultures are not incubated at 37°c in a school laboratory.

### 19.2. Culture media

### **Activity: 19.2**

Describe different types of media used in culturing microorganisms.

A medium is a solid or liquid preparation containing nutrients for the culture of microorganisms. A pure microbial culture undergoes the following steps namely:

- Choice of the culture medium.
- Sterilization of the culture medium.
- A culture with a collection of microbial cells growing on or, in a medium.
- Selection of a pure colony from a collection of microbial cells growing
- Introduction of a microorganism into a suitable growth medium
- Streaking to carrying out a pure culture.

Microorganisms may be cultured in a solid medium or a liquid medium or broth. When there is not a culture with a collection of microbial cells growing on or, in a medium. A source of microorganisms is spread on the surface of an agar to produce individual colonies. Once individual colonies are obtained, this collection of microorganisms can then use to carry out a pure microbial culture.

### a. Solid medium.

Solid media are particularly suitable for bacteria and fungi and are prepared by mixing the liquid nutrient solution with a gelling agent, usually agar, at a concentration of about 1-5%, thus, producing nutrient agar that allows the growth of colonies.

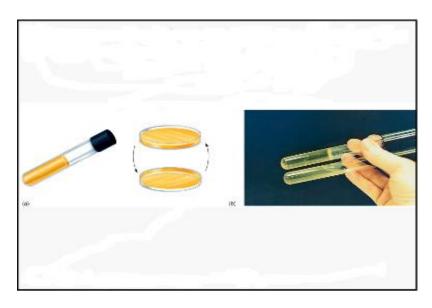


Figure 19.1 Samples of solid media.

## b. Liquid media

The liquid media are water – based solutions that are generally termed as broths, milks and infusions.

Liquid media are often useful for measuring population growth. They may be placed in a test tube, stopped by a plug of cotton wool or a metal cap, or in a glass, screwcrapped bottle such as a universal bottle which holds about 25cm<sup>2</sup> enough for one agar plate.

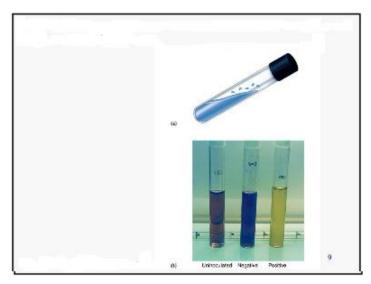


Figure 19.2 Samples liquid media

The medium must be sterilized and after, adding a small quantity of cells to the medium is called inoculation.

#### c. Enrichment media.

An enrichment medium is a medium in which substances are added to meet the requirements of certain microorganisms in preference to others. As a result, certain microorganisms grow better than others.

#### d. A selective medium

It is a medium in which one or more substances are added to favor the grown of specific microorganisms and to inhibit the growth of others. Example, the addition of penicillin to a culture to select for those organisms resisting to it, or the selection of hybridizes cells during the production of monoclonal antibodies.

### Self-assessment 19.2

- 1. How would you isolate from the soil an organism which could use atmospheric nitrogen as its only source of nitrogen (a nitrogen-fixing bacteria)?
- 2. What is meant by nutrient agar?
- 3. Distinguish between liquid media and solid media.
- 4. Distinguish between enriched media and specific media.

# 19.3. Aseptic technique.

## Activity 19.3.1

Carry out a procedure of culturing fungion a nutrient agar using sterile techniques.

Aseptic technique is using sterilized equipment and solutions and preventing their contamination. Sterilization is the removal or destruction of all living microorganisms, including spores (inactive structures that enable some microorganisms to survive unfavorable periods). Bacterial and fungal spores are abundant in most environments including laboratories. A range of special techniques and apparatus are designed to prevent contamination of nutrients media. Autoclaves are used to sterilize equipment and culture media before experiments and also to sterilize equipment and specimens before disposal.

In addition, after sterilization, a great care is taken during experiments to ensure that there is no infection.

# 19.3.1. Spread plate technique

This is one of the most basic and useful of microbiological techniques. Petri dishes are specially designed as shallow circular dish made of glass or plastic. The shape of the lid allows avoiding contamination, but gas molecules can diffuse between the inside of the dish and the environment through where the base meets the lid. Oxygen can therefore reach the culture and carbon dioxide can escape.

The spread plate technique involves using a sterilized spreader with a smooth surface made of metal or glass to apply a small amount of bacteria suspended in a solution over a plate. The plate needs to be dried at room temperature so that the agar can absorb the bacteria more readily. A successful spread plate will have a countable number of isolated bacterial colonies evenly distributed on the plate.

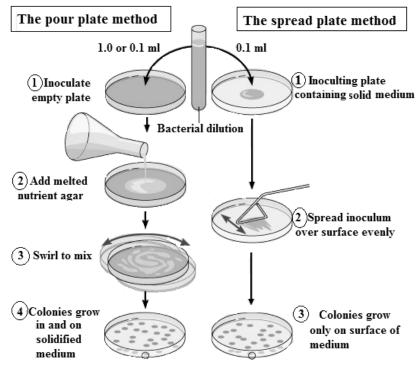


Figure 19.3: Spread plate technique.

### 19.3.2. Methods of inoculation

The introduction of a small number of microorganisms into a nutrient medium is called inoculation. Aseptic technique must be used to avoid contamination. The procedure differs for solid and liquid media.



Figure 19.4: An inoculation loop

### a. Inoculating a solid medium

We use a wire loop. The loop is firstly flamed and after it is then used to lift a thin film of a liquid suspension or a small amount of solid material containing the microorganisms being investigated from the previous culture or any source of microorganisms. The loop is gently stroked across the surface of the medium in a series of sets of streaks.

### b. Inoculating a liquid medium

If the cells to be inoculated are in a liquid, for example water or a broth, a sterile wire loop is used to transfer a sample to the medium, which is often a test medium. The wire loop is simply agitated gently inside the medium. If the cells to be inoculated are in or a solid medium such as soil nutrient agar, a wire loop may be used for transfer to the liquid medium. It can be rubbed on the inside surface of the vessel containing the liquid medium to ensure successful transfer.

### c. Carrying out a pure culture

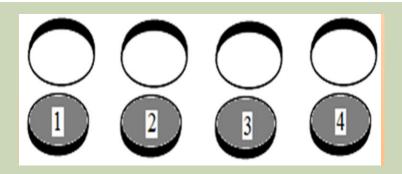
Pure culture technique is a method of culturing microorganisms in which all of the individuals in a culture have descended from a simple individual. The basis of pure technique is the isolation in colonies of individual cells. This is done so as to allow the characterization of specific types of microorganisms.

### d. Incubation on agar-agar.

During incubation, the nutrients are contained in agar medium. Agar is a substance which dissolves in hot water and sets to form a jelly. You pour hot agar containing all necessary nutrients such as carbohydrates, proteins and vitamins into a Petri- dish. Then leave it to cool and set before you add any microorganism. The other way to provide nutrients to grow microorganisms is as a broth in a culture flask. The steps of culturing agar –agar are shown in the following activity.

## **Activity: 19.3.2**

- Boil a mixture of 50 ml of water and 20g of agar-agar powder for 15 minutes as you are stirring
- Pour the jelly mixture into four pre-sterilized glass Petri-dishes. Then allow the broth to coagulate at room temperature.
- Number the dishes; 1, 2, 3 and 4 respectively; on the bases.
- Place a nail scarping from between the teeth onto the jelly in dish 1 and 2, wave the dish 3 on latrine for 1 minute and do not put anything on dish 4.
- Warm the dishes 2 and 4 on the top of water vapour stream for 15 minutes and then cool them (do not open them)
- Then fix the lids tightly to the bases of the four Petri-dishes with clear adhesive tape and place them upside down in an oven/incubator at 37 °C for 3 days.
- Record and interpret your results.



### 19.3.3. Alcoholic fermentation

### Activity 19.3.3

Describe how yeast would be used in alcoholic fermentation.

Yeast releases digestive enzymes which allow the transformation of glucose into ethanol as result of anaerobic fermentation. The presence of bubbles is the evidence that carbon dioxide is released as waste product of the alcoholic fermentation. Making Beer depends on a process called malting. You soak and keep barley grains in water. As germination begins, enzymes break down the starch in the barley grains into a sugary solution. You then extract a solution produced by malting and use it as an energy source for the yeast. The mixture of yeast and sugar solution is then fermented to produce alcohol. Hops are added at this stage to give flavour. The beer is given time to clear and develops its flavour before putting it in bottles or to be sold. Interestingly, alcohol in large quantities is toxic to yeast as well as to people. This is why the alcohol content of wine is rarely more than 14%. Once it gets much higher, it kills all the yeast and stops fermentation.

#### Self-assessment 19.3

From guestions 1-5, circle the letter corresponding to the right answer

- 1. The method of culturing microorganisms in which all of the individuals in a culture have descended from a single individual is called:
  - a. Pure culture technique
  - b. Spread plate technique
  - c. Aseptic technique
  - d. Liquid media method
- 2. Inoculating liquid medium, various instruments are used. Which one of the following is used to transfer the sample to the medium?
  - a. Sterile wire loop
  - b. Inoculating needle.
  - c. Petri-dishes
  - d. None of these.

- 3. Large amounts of alcohol are dangerous to yeast during alcoholic fermentation. Which of the following explains the reason?
  - a. It kills all the yeast and stops fermentation.
  - b. Motivate yeasts
  - c. It kills some few bacteria.
  - d. Temperature affects fermentation.
- 4. The technique of using sterilized equipment and solutions and preventing their contamination is referred to as:
  - a. Pure culture technique
  - b. Spread plate technique
  - c. Aseptic technique
  - d. Petri-dish technique.
- 5. Petri dishes are specially designed as a shallow circular dish made of glass or plastic with a lid. Which one of the following best explains the function of the lid?
  - a. Prevent contamination, but gas molecules can diffuse.
  - b. Spread the bacteria on the plate.
  - c. Allows contamination.
  - d. None of the above.

# 19.4. Population growth of bacteria

# **Activity: 19.4.1**

Use text books and other sources of information to interpret the graphs of bacterial growth.

When bacteria or any other microorganisms are incubated in a suitable culturing medium, they reproduce by binary fissions and the number of individuals increases.

The ordinary growth of population is described as sigmoid curve or S-shaped curve made of 4 main phases:

- The lag phase: period of adaptation of microorganisms to the new habitat (new environment)
- The log or exponential phase: period of high rate of reproduction. Bacteria are sensitive to the limiting factors of the growth or anti-microbial agents
- The stationary phase: Stationary phase of plateau-growth slows down. The population remains constant because the rate of dividing/growth is equal to the rate of death within the population. The maximum number that a habitat can accommodate for a long period is known as the carrying capacity.
- The decline or death phase: period of high rate of death than the rate of

dividing/growth due to the scarcity of food, the abundance of metabolic waste products, presence of antibiotics or any other drugs killing the germs. Figure 19.5 shows the phases explained above.

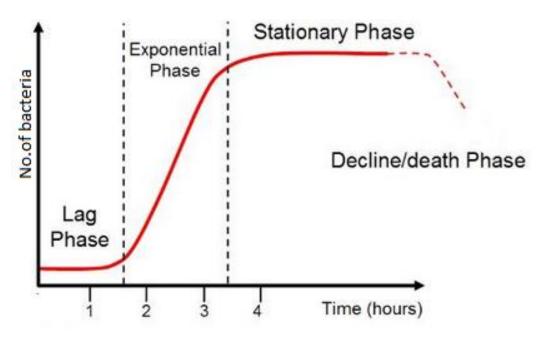


Figure 19. 5: Graph of population growth of bacteria.

# 19.4.1. Measuring population growth of bacteria

The typical growth curve of a population of bacteria is similar to the growth curve expected for yeast, a unicellular fungus or the growth of any population. When measuring the growth of a population of bacteria or yeast, we can carry out direct counting of the numbers of cells or indirectly by measuring some indication of the number of cells such as the coldness of a solution, or production of a gas.

It is usual to inoculate a small sample of the microorganisms in a sterilized nutrient medium and to place the culture in an incubator at the optimum temperature for growth. Other conditions are pH, oxygen concentration and ionic and osmotic balance. Growth can be measured from the time of inoculation. Two types of cell count are possible, namely viable count and total count. The viable count is the total of living cells only and total count is the total number of both living and dead cells and is easier to measure.

### **Activity 19.4.2**

# Investigating the bacterial content of fresh and stale milk.

**Materials required:** Four sterile nutrient agar plates, inoculating loop, Bunsen burner, indelible marker or wax pencil, Fresh pasteurized milk, Stale milk (milk left at room temperature for 24hours) and Incubator set at 35°C

### Procedure:

- Place the inoculating loop in the Bunsen burner flame until the loop is red hot.
- Allow the loop to cool and then dip into a sample of fresh, well shaken milk.
- Lift the lid of the sterile agar plate slightly with the other hand and lightly spread the contents of the inoculating loop over the surface the agar.
- Close the lid of the plate and return the loop to the Bunsen burner flame until red hot.
- Label the base of the plate with an indelible marker or pencil.
- Repeat the process with the second plate and another sample of fresh milk.
- Flame the loop again and after cooling, place it in a sample of stale milk.
- Spread the contents of the loop over the surface of a third plate and then close the lid.
- Label the base of the plate with an indelible marker or pencil.
- Repeat the process with the fourth plate and second sample of stale milk.
- Put the four plates in an incubator at 35°c for about 3 days. They should be placed upside down to prevent condensation falling onto the cultures. After incubation, the two halves of each plate should tape together for safety.
- Record the appearance of the colonies

Give the general comment based on your observations

#### Self-assessment 19.4

1. A culture of yeast, Saccharomyces cerevisiae, had been carried out in the banana juice for 7 days at 30°C. The table below shows the change in number of yeasts within that time:

Time/days	1	2	3	4	5	6	7
No. of yeast	2	2	6	16	20	20	8

- a. Draw a curve showing the growth of the yeast population
- b. What is the role of banana juice in that experiment?
- c. State any two conditions that should be maintained constant during that experiment.
- d. Describe the trend of the graph you have drawn in a
- Design an experiment to test the hypothesis that contact of an agar plate with a finger results in more bacterial growth than exposing the plate to classroom air.

# 19.5. Staining of bacteria

### Activity 19.5

"Staining bacteria for practical purpose is important". Discuss the validity of the statement.

The microorganisms or parts of microorganisms that pick up the stain are clearly / distinctively observed from the rest of the background.

In **simple staining**, all the cells and structures in general stain the same colour. In positive staining, cells structures take in the stain e.g. methylene blue while in negative staining the cells repel the stain and it is taken by the background e.g. Indian ink. **Negative** staining is mostly useful in viewing capsules and such structures that surround the bacteria.

**Differential staining** on the other hand, multiple staining reactions are used to take advantage of the fact that particular types of microorganisms and/or specified structures of microorganism display varied staining reactions that are readily distinguishable by different colours. The stain must be fixed immediately and the dyed specimen is treated in some ways, e.g. by chemicals or heat to tightly bind the stain to the organism or its structures.

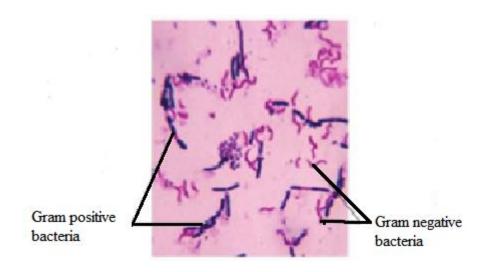


Figure 19.6: Gram positive and Gram negative

# 19.5.1. The purpose of staining bacteria

The purpose of staining bacteria is to see, for example, how thick of a layer of peptidoglycan their cell wall has. In the Gram stain, gram-negative bacteria will stain red or pink because the rinse took out the primary dye and the Safrinin (secondary dye) took over the coloring as the counterstain. In gram-positive bacteria, since it has a thick-layer of peptidoglycan, not all of the Crystal violet color will be rinsed out of the cell wall, so it will be blue or purple. The following are reasons to explain why stained:

- It's for helping classifying and determining what the bacteria are composed of.
- It's very useful tool to help identify bacteria without necessarily killing the cell.
- Gram staining is performed to distinguish between gram positive and negative bacteria.
- To enable the person to visualize its physical features- shape, size, arrangement, etc the bacterial cells are stained with specific dyes or stains

# 19.5.2. Procedure of staining and their corresponding stains.

### **Activity 19.5.1:**

carry out an experiment to stain bacteria for examination under the light microscope

In staining bacteria, we use various staining procedures each having specific set of stains or dyes. Some of them are:

- 1. Gram's Staining Crystal violet, Iodine and Safrinin
- 2. Capsule staining Nigrosin, Safrinin or India Ink, Safrinin
- 3. Spore staining Malachite Green and Safrinin
- 4. PHB staining Sudan black.
- 5. Using decolorizer Alcohol wash

Observe and identify some of the staining methods on figure 19.6 as shown below:

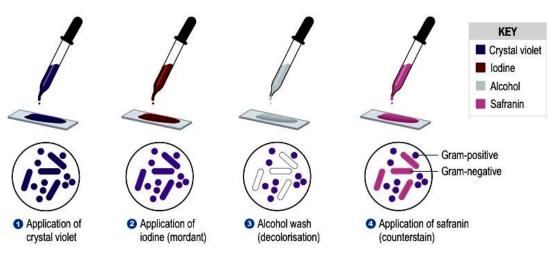


Figure 19.7: different strains of bacteria

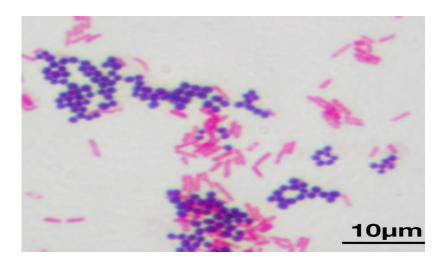


Figure 19.8: Gram positive and Gram negative bacteria.

# 19.5.3. Growing viruses

The culture of viruses is made more difficult than the culture of bacteria or fungi because viruses can only grow and multiply inside living cells. This can be done by infecting whole organisms such as plants or animals but, where possible, cell, tissue cultures are now used. An early technique was to grow certain viruses in chick embryos while the embryo was still growing inside the egg.

### 19.5.4. Tissue Culture of Animal Viruses

Viruses cannot be grown in standard microbiological broths or on agar plates; instead they have to be cultured inside suitable host cells. Note the following facts:

- Tissue culture is a useful method for cultivating clinical samples suspected of harboring a virus. This method helps with the detection, identification, and characterization of viruses in the laboratory.
- Tissue culture of animal viruses involves growing animal cells in flasks using various broth media and then infecting these cells with virus.
- Transfect ion can be carried out using calcium phosphate, by electroporation, or by mixing a cationic lipid with the material to produce liposome's, which fuse with the cell membrane and deposit their content inside.
- Cytopathic effect is a non-lyrics damage that viruses cause to cells. These vary in their manifestation and damaging effect.
- Cell culture is complex process by which cells are grown under controlled conditions, generally outside of their natural environment.

Cell culture is the complex process by which cells are grown under controlled conditions, generally outside of their natural environment. The term "cell culture" is defined as the culturing of cells derived from multi-cellular eukaryotes, especially animal cells. However, there are also cultures of plants, fungi, and microbes, including viruses, bacteria, and protists.

#### Self-assessment 19.5

1. - Match the elements of column A and B in each case.

B

Gram-positive bacteria Stain red or pink

Gram-negative bacteria Stain blue or purple

Cell culture conditions growing cells under controlled

A E

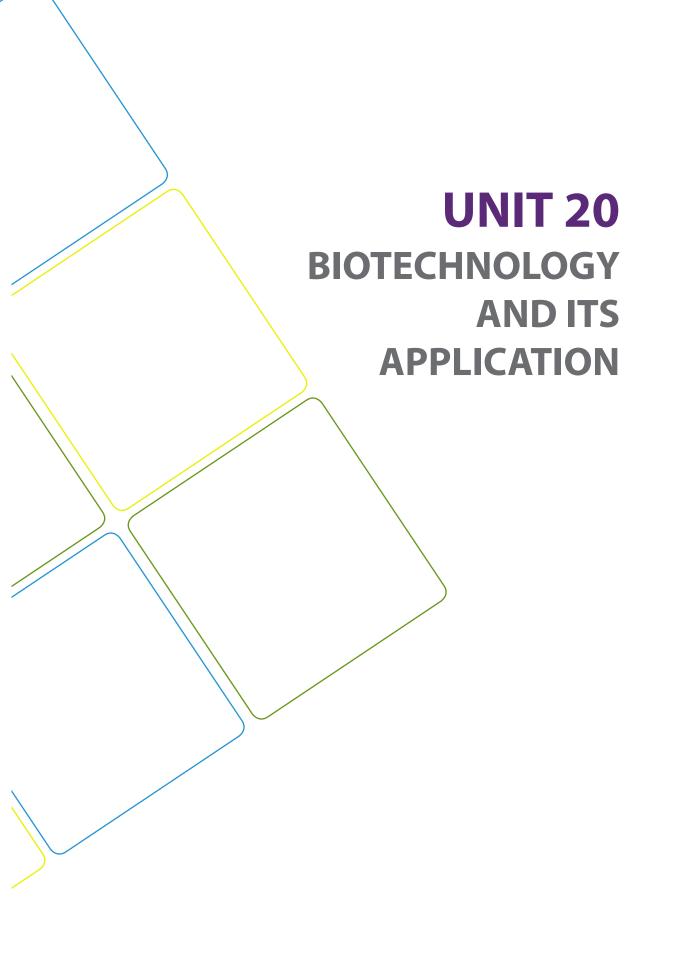
Spore staining Malachite Green and Safrinin

Differential staining multiple staining reactions are used.

- 2. Explain why it is more difficult to culture viruses than culturing bacteria
- 3. How/why are viruses specific to the cells they infect?
- 4. Distinguish between vaccines and antibiotics

#### **End of unit assessment19**

- 1. What are different types of media used in the laboratories for culturing microorganisms?
- 2. Define a pure culture.
- 3. How do biologists differentiate between Gram –positive and Gram –negative bacteria?
- 4. Describe the three methods of preventing bacterial growth in food.
- 5. How does temperature affect the growth of bacteria in culture media?
- 6. Assuming that you have a bacterial infection, would you ask for vaccination against the bacteria? Why or why not?
- 7. How do bacteria maintain the balance in the environment?
- 8. Explain why an infection by Gram-negative bacteria are more difficult to treat than Gram-positive bacteria.
- 9. How would you investigate that temperature affect the bacterial growth?
- 10. Write short notes on each of the following term related to the culture of microorganisms.
  - a. Aseptic techniques.
  - b. Staining bacteria
  - c. Growing viruses
- 11. Explain why microorganisms are particularly suitable for industrial use.



# **UNIT 20: BIOTECHNOLOGY AND ITS APPLICATION**

### **Key Unit Competence**

Explain the biotechnology involved in the production of ethanol, biogas and bread making.

## **Learning Objectives**

By the end of this unit, I should be able to:

- State that bacteria are useful in biotechnology and genetic engineering due to their rapid reproduction rate and their ability to make complex molecules.
- Discuss why bacteria are useful in biotechnology and genetic engineering.
   Focus on: lack of ethical concerns over their manipulation and growth, genetic code shared with all other organisms, and presence of plasmids.
- Show concern for the role of bacteria in genetic engineering.
- Investigate and describe the use of pectinase in fruit juice production and lactase to produce lactose-free milk.
- Describe the role of anaerobic respiration in yeast during bread-making.
- Compare leavened and unleavened bread.
- Appreciate the role of anaerobic respiration in the production of ethanol and in yeast during bread-making.
- Explain how fermenters are used in the production of penicillin.
- Describe the role of the fungus Penicillium in the production of the antibiotic penicillin.
- Interpret and explain graphs showing how the pH and the concentration of penicillin in a culture changes over time when the pH is controlled and not controlled.
- Defend the role played by antibiotics in treatment of bacterial diseases.
- Describe the three stages of biogas production and the role of bioreactors in economically poor rural communities
- Apply the knowledge of bioreactors, using cow dung, agricultural waste and domestic waste to prepare and produce biogas.
- Appreciate the role of biogas production in reducing the environmental degradation.

# **Introductory activity:**

Biotechnology is a broad discipline in which biological processes, organisms, cells or cellular components are exploited to develop new technologies, remember that biotechnology is useful and applied in our daily life activities such as in beverages and food industries, agricultures, medicines. Brainstorm on the role of microorganisms in biotechnology and genetic engineering. Can you think on your own understanding on howbread, juice and beer are made? Why do bacteria become resistant to antibiotics? Make discuss on the biogas production.

# 20.1. Role of bacteria in Biotechnology and genetic engineering

# **Activity 20.1**

Using addition resources to your textbook available in your school such as the books from the school library and search further information from the internet. Discuss the role of bacteria in biotechnology and genetic engineering.

Biotechnology is a broad discipline in which biological processes, organisms, cells or cellular components are exploited to develop new technologies. New tools and products developed by biotechnologists are useful in research, agriculture, industry and the clinic. For example, the use of living cells, bacteria, etc., to make useful products (such as crops that insects are less likely to destroy or new kinds of medicine).

The wide concept of "biotech" or "biotechnology" encompasses a wide range of procedures for modifying living organisms according to human purposes, going back to domestication of animals, cultivation of the plants, and "improvements" to these through breeding programs that employ artificial selection and hybridization. Modern usage also includes genetic engineering as well as cell and tissue culture technologies.

The bacteria have an economic importance which derives from the fact that bacteria are exploited by humans in a number of beneficial ways. Despite the fact that some bacteria play harmful roles, such as causing diseases and spoiling food, the economic importance of bacteria includes both their useful and harmful aspects.

# 20.1.1. Useful Bacteria in Biotechnology

Biotechnology or Industrial microbiology is defined as the use of microorganism such as bacteria, fungi and algae for the manufacturing and services industries. These include:

- Fermentation processes, such as brewing, baking, cheese and butter manufacturing, Bacteria, often Lactobacillus bulgaricusin combination with yeasts and fungi, is used to make yoghurt and cheese have been used for thousands of years in the preparation of fermented foods such as cheese, pickles, soy sauce, sauerkraut, vinegar, and wine.
- In the chemical industry, bacteria are most important in the production of pure chemicals for use as pharmaceuticals or agrochemicals.
- Bacteria are also used in chemical manufacturing such as ethanol, acetone, organic acid, enzymes, and perfumes.
- Bacteria can also be used in the place of pesticides in Biological Pest Control. This commonly uses Bacillus thuringiensis (also called BT), a Gram-positive, soil-dwelling bacterium.
- Saprophytic bacteria help in breaking of complex organic substance to simpler

- forms. Thus, in this process, they help to convert farm refuse, dung and other wastes to manure.
- Number of anti-bacterial and anti-fungal antibiotics such as Hamycin, Polymyxin, and Trichomycin are obtained from fungal mycelia and bacteria (like Streptomyces). Similarly, Bacillus is used for production of antibiotics such as Bacitracin and Gramicidin.
- Different kinds of vitamins are produced from bacteria like Riboflavin from Clostridium butylicum, Vitamin B12 from Bacillus megatherium and Vitamin K and B-complex from Escherichia coli.

## 20.1.2. Useful Bacteria in Genetic engineering

Genetic engineering is the manipulation of genes. It is also called recombinant DNA technology. In genetic engineering, the genetic information for many biological products and biological processes can be introduced into microbes in order to genetically engineer them to produce a substance or conduct a process. The genes can come from any biological source: human, animal, plant or microbes. This opens the possibility for microbial production of foods, fuels, enzymes, hormones, diagnostic agents, medicines, antibiotics, vaccines, antibodies, natural insecticides and fertilizers, and all sorts of substances useful in our civilization and society.

The pieces of DNA (genes) are introduced into a host by means of a carrier (vector) system. The foreign DNA becomes a permanent feature of the host, being replicated and passed on to daughter cells along with the rest of its DNA. Microorganisms especially bacteria play a central role in recombinant DNA technology and genetic engineering. Important tools of biotechnology are **microbial cells** (bacteria, fungi), microbial genes and microbial enzymes.

Bacterial cells are transformed genetically and used in production of commercially important products. For example, bio medical technology bacteria can be bioengineered for the production of therapeutic proteins like: Human Insulin (used against diabetes), Human Growth Hormone (somatotropins used to treat pituitary dwarfism), and others which can be used to fight against viral diseases. Antibiotics are produced in nature by molds such as Penicillium and bacteria such as Streptomyces and Bacillus.

### Self-assessment 20.1

- 1. What is biotechnology?
- 2. What do you understand by genetic engineering?
- 3. Discuss on the role of bacteria in Biotechnology and genetic engineering.

# 20.2. Immobilization of enzymes

## **Activity 20.2**

Carry out research on the action of enzymes with reference to pectinase in fruit juice production and lactase to produce lactose-free milk.

Enzymes catalyze biological reactions in our body, but they can also be used to catalyze industrial reactions outside the body. These enzymes are often bound to a support ('immobilized') and can be used for a wide range of purposes.

# 20.2.1. The advantages of immobilized enzymes

Enzymes have an enormous range of commercial applications for example: in medicine, food technology and industrial processing. Enzymes are expensive. No company wants to have to keep buying them over and over again if it can recycle them in some way. One of the best ways of keeping costs down is to use immobilized enzymes. Using immobilized enzymes means that you can keep and re-use the enzymes, and that the product is enzyme-free. Another advantage of this process is that the immobilized enzymes are more tolerant of temperature changes and pH changes than enzymes in solution. This may be partly because their molecules are held firmly in shape by the alginate in which they are embedded, and so do not denature as easily. It may also be because the parts of the molecules that are embedded in the beads are not fully exposed to the temperature or pH changes.

Using enzymes instead of other molecules in reactions is useful because enzymes catalyze specific reactions and work at much lower temperatures than chemical catalysts.

The molecule that an enzyme acts on is called a substrate. Enzymes can either be mixed freely with the substrate in solution or immobilized to a solid support so they do not mix freely. There are many advantages of immobilization, one of which is that the enzymes can be reused catalyzing the same reaction many times. Binding the enzymes to a surface also makes those more stable and less likely to denature (lose their shape). In addition, there will be no enzyme left in the product at the end, so purification is not necessary.

# 20.2.2. The disadvantages of immobilized enzymes

There are some disadvantages: immobilization requires extra time, equipment and work; there may be a reduction in reaction rates if enzymes cannot mix freely with the substrate; and immobilized enzymes cannot be used if one of the substrates is insoluble.

# 20.2.3. Advantages of Using Immobilized Enzymes

The advantages of using immobilized enzymes are: (i) reuse (ii) continuous use (iii) less labor intensive (iv) saving in capital cost (v) minimum reaction time (vi) less chance of contamination in products, (vii) more stability (viii) improved process control and (ix) high enzyme: substrate ratio. The first immobilized enzymes to be scaled up to pilot plant level and industrial manufacture were immobilized amino acid acylase, penicillin G-acylase and glucose isomerase.

## 20.2.4. Methods of Enzyme Immobilization

There are five different techniques of immobilizing enzymes: (i) adsorption, (ii) covalent bonding, (iii) entrapment, (iv) copolymerization or cross-linking, and (v) encapsulation. For the purpose of immobilization of enzymes carriers i.e. the support materials such as matrix system, a membrane or a solid surface are used.

### i. Adsorption

An enzyme may be immobilized by bonding to either external or internal surface of a carrier or support such as mineral support (aluminum oxide, clay), organic support (starch), and modified sapharose and ion exchange resins. Bonds of low energy are involved e.g. ionic interactions, hydrogen bonds, van der Waals forces, etc. If the enzyme is immobilized externally, the carrier particle size must be very small in order to achieve appreciable surface of bonding. These particles may have diameter ranging from 500 A to about 1 mm. Due to immobilization of enzymes on external surface, no pore diffusion limitations are encountered.

In addition, the enzyme immobilized on an internal surface is protected from abrasion, inhibitory bulk solutions and microbial attack, and a more stable and active enzyme system may be achieved. There are four procedures for immobilization by adsorption: (i)static process (enzyme is immobilized on the carrier simply by allowing the solution containing the enzyme to contact the carrier without stirring (ii)the dynamic batch process (carrier is placed into the enzyme solution and mixed by stirring or agitated continuously in a shaker), (iii) the reactor loading process (carrier is placed into the reactor that will be subsequently employed for processing, then the enzyme solution is transferred to the reactor and carrier is loaded in a dynamic environment by agitating the carrier and enzyme solution), and (iv) the electrode position process (carrier is placed proximal to one of the electrodes in an enzyme bath, the current put on, the enzyme migrates to the carrier and deposited on the surface).

## ii. Covalent bonding

Covalent bond is formed between the chemical groups of enzyme and chemical groups on surface of carrier. Covalent bonding is thus utilized under a broad range of pH, ionic strength and other variable conditions. Immobilization steps are attachment of coupling agent followed by an activation process, or attachment of a functional group and finally attachment of the enzyme. Different types of carriers are used in immobilization such as carbohydrates proteins and amine-bearing carriers, inorganic carriers, etc. Covalent attachment may be directed to a specific group (e.g. amine, hydroxyl, tyrosyl, etc.) on the surface of the enzyme. Hydroxyl and amino groups are the main groups of the enzymes with which it forms bonds, whereas

sulfhydryl group least involved.

There are different methods of covalent bonding such as: (i)diazoation (bonding between the amino group of the support e.g. aminobenzyle cellulose, aminosilanised porous glass, aminoderivatives and a tyrosyl or histidyl group of the enzyme), (ii) formation of peptide bond (bond formation between the amino or carboxyl group of the support and amino or carboxy group of the enzyme), (iii) group activation (use of cyanogen bromide to a support containing glycol group i.e. cellulose, syphadex, sepharose, etc.), and (iv) poly functional reagents (use of a bifunctional or multifunctional reagent e.g. glutaraldehyde which forms bonding between the amino group of the support and amino group of the enzyme). The major problem with covalent bonding is that the enzyme may be inactivated by bringing about changes in conformation when undergoes reactions at active sites. However, this problem can be overcome through immobilization in the presence of enzyme's substrate or a competitive inhibitors or protease. The most common activated polymers are celluloses or polyacrylamides

## iii. Entrapment

Enzymes can be physically entrapped inside a matrix (support) of a water soluble polymer such as polyacrylamide type gels and naturally derived gels e.g. cellulose triacetate, agar, gelatin, carrageenan, alginate, etc. The form and nature of matrix vary. Pore size of matrix should be adjusted to prevent the loss of enzyme from the matrix due to excessive diffusion. There is possibility of leakage of low molecular weight enzymes from the gel. There are several methods for enzyme entrapment: (i)inclusion in gels (enzyme entrapped in gels), (ii) inclusion in fibers (enzyme entrapped in fiber format), and (iii)inclusion in microcapsules (enzymes entrapped in microcapsules formed monomer mixtures such as polyamine and polybasic chloride, polyphenol and polyisocyanate). The entrapment of enzymes has been widely used for sensing application, but not much success has been achieved with industrial process.

# iv. Cross - linking or Co-polymerization

Cross-linking is characterized by covalent bonding between the various molecules of an enzyme via a polyfunctional reagent such as glutaraldehyde, diazonium salt, hexamethylenedisocyanate, and ethylene bismaleimide. The demerit of using polyfunctional reagents is that they can denature the enzyme. This technique is cheap and simple but not often used with pure proteins because it produces very little of immobilized enzyme that has very high intrinsic activity. It is widely used in commercial preparation.

## v. Encapsulation

The encapsulation is the enclosing of a droplet of solution-of enzyme in a semipermeable membrane capsule. The capsule is made up of cellulose nitrate and nylon. The method of encapsulation is cheap and simple but its effectiveness largely depends on the stability of enzyme although the catalyst is very effectively retained within the capsule. This technique is restricted to medical sciences only. In this method a large quantity of enzyme is immobilized but the biggest disadvantage is that only small substrate molecule is utilized with the intact membrane.

# 20.2.5. How are immobilized enzymes used in food?

## i. Immobilization of enzymes use of pectinase in fruit juice production

Pectinases find commercial application in fruit juice, wine, oil, tea, coffee, textile and paper-making industries using a wide variety of carriers and methods. One of the vital applications is the clarification and depectinization of fruit juices. The raw fruit juice obtained after pressing is very turbid viscous and contains a significant amount of colloidal compounds, mainly pectin that causes cloudiness; therefore, clarification of fruit juices involves the removal of juice haze by enzyme hydrolysis with pectolytic enzymes. Although pectinases enhance the clarification of juices, immobilization of these enzymes proves to be beneficial for industrial use. Immobilization of pectinase on celite through adsorption is a simple, cheap and effective method. For the clarification of pineapple juice, excellent results were observed using immobilized polygalacturonase in comparison with free enzyme.

Fruits contain pectin, carbohydrates found in the cell wall that holds the plant cells together. Immobilized pectinase can be used to break down this pectin, loosening the connections between cells. This increases the amount of juice you can get from the fruit, makes the juice runnier and gets rid of the cloudiness that pectin can cause.

## ii. Making lactose-free milk

The enzyme lactase breaks down the sugar lactose, which is found in milk, into the sugars glucose and galactose. Most people produce this enzyme in their bodies, but some people (and most cats) don't, meaning that they are lactose intolerant. Because they can't break down lactose, it builds up in their digestive system where bacteria feed on it, causing digestive problems.

Immobilized lactase can be used to produce lactose-free milk: normal milk is poured down a column containing the immobilized lactase enzymes, which break down the lactose. After the milk has passed through this system, it will only contain the products of the reaction (glucose and galactose), so lactose-intolerant people (and cats) can drink it. The enzyme lactase can be immobilized using alginate beads.

The figures below show one way in which enzymes can be immobilized. The enzyme is mixed with a solution of sodium alginate. Little droplets of this mixture are then added to a solution of calcium chloride. The sodium alginate and calcium chloride instantly react to form jelly, which turns each droplet into a little bead. The jelly bead contains the enzyme.

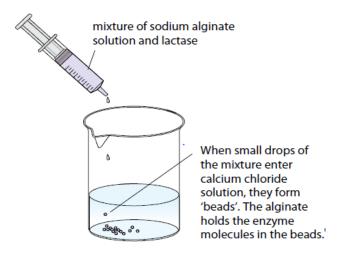


Figure 20.1 Immobilizing enzymes in alginate.

The enzyme is held in the bead, or immobilized. These beads can be packed gently into a column. Milk is then allowed to run through the column of lactase-containing beads. The lactase hydrolyses the lactose in the milk to glucose and galactose. The milk is therefore lactose-free, and can be used to make lactose free dairy products for people who cannot digest lactose. The product continues to trickle down the column, emerging from the bottom, as illustrated in the diagram below, where it can be collected and purified. Not only would you lose the lactase, but also you would have milk contaminated with the enzyme.

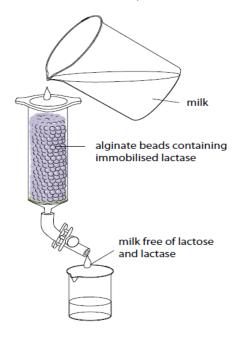


Figure 20.2: Using immobilized enzyme to modify milk.

## iii. Biological washing powders containing enzymes

The biological washing powders contain enzymes like protease and lipase to remove protein stains and fat/grease from clothes. The enzymes break down proteins or fats on the fabric, forming water-soluble substances that can be washed away. Because stains are made of different types of molecules, a range of enzymes are needed to break them down. Proteases break down proteins, so are good for blood, egg, gravy, and other protein stains. Amylases break down starches, and lipases break down fats and grease.

For example: Blood contain the red protein Haemoglobin (Hb). The Proteases in biological washing powder break Hb molecules into smaller molecules, which are not colored and which dissolve in water and can be washed away. This makes the washing powder more effective than detergent alone, especially at lower temperatures. This save energy (no need to boil water), but if the temperature is too high, the enzyme will be denatured.

## iv. Fruit juices

Fruits contain pectin, carbohydrates found in the cell wall that holds the plant together. Immobilized pectinase enzyme can be used to break down this pectin, loosening the connections between cells. This increases the amount of juice you can get from the fruit, makes the juice runnier and gets rid of the cloudiness that pectin can cause.

## 20.2.6. How are immobilized enzymes used in biosensors?

The specificity of enzymes means that they can be used to test for a unique substance, which is exactly what a biosensor does.

# **Glucose test strips**

People with type 1 diabetes lack the hormone insulin, so they have to test their blood sugar levels regularly to ensure they stay within a healthy range. They do this by measuring the amount of glucose in their blood with a glucose test strip. On the test strip is the immobilized enzyme glucose oxidase; when glucose is present, the enzyme catalyzes a reaction that changes glucose into hydrogen peroxide and gluconic acid.

There is also another mediator molecule on the test strip, which catalyses a reaction involving the products of the enzyme reaction. In the early test strips, this second reaction caused a color change, with the color indicating the amount of glucose present. In most modern tests, this second reaction produces electrical current, which can be measured by a meter to give the exact concentration of glucose in the blood.

### Self-assessment 20.2

- 1. Discuss the advantages and disadvantages of immobilized enzymes.
- 2. Write on the use of pectinase in fruit juice production.
- 3. Explain the role of lactase in making lactose-free milk.
- 4. How are immobilized enzymes used in biosensors?

# 20.3. Application of enzyme in technology.

## **Activity 20.3**

Visit a nearby bakery and verify how bread is prepared. Write a short report on the raw materials and procedures used in making bread up to the final product.

# 20.3.1. Enzymes in Brewing

Enzymes increase processing capacity and improve economy in the fruit juice and wine industries. The most commonly used enzymes in these industries are pectinase. Pectinase increases juice yields and accelerate juice clarification. They produce clear and stable single-strength juices, juice concentrates and wines, from not only core-fruits such as apples and pears, but also stone fruits, berries, grapes, citrus-fruits, tropical fruits and vegetables like carrots, beets and green peppers. Future aspects focus on a wider application of enzymes to brew with high amounts of inexpensive raw materials like barley. Barley contains starch that has to be broken down to fermentable sugars before the yeast can make alcohol. Therefore, traditional brewing contains an extra step compared with wine-making, namely malting in which enzymes needed for the degradation of starch into fermentable sugars are produced.

# 20.3.2. Enzymes perform many functions in beverages

The most important field of application for enzymes in the beverage industry is the extraction of fruit juice and vegetable juice. Pectinases, in particular, are employed for apple and pear juice and for juices made from berries and tropical fruits. They break down pectins found in the plant cell walls as supporting substances. This increases the quality of juice extracted and reduces fruit waste. Enzymes can be used in winemaking to increase the preliminary juice extraction and to obtain more high-quality wine. Pectinase not only increase juice yields, but also increase the colour and health-promoting antioxidants in fruit and vegetable juices. They also increase colour extraction and juice volume by reducing fruit and vegetable mash viscosity and improving solid/liquid separation, Pectinase and Amylase enzyme solutions speed up filtration and prevent storage or post-packaging haze formation by depectinizing and reducing starch in raw juices.

## 20.3.3. Medical applications of enzymes

Development of medical applications for enzymes has been at least as extensive as those for industrial applications, reflecting the magnitude of the potential rewards: for example, pancreatic enzymes have been in use since the nineteenth century for the treatment of digestive disorders. The variety of enzymes and their potential therapeutic applications are considerable. At present, the most successful applications are extracellular: purely topical uses, the removal of toxic substances and the treatment of life-threatening disorders within the blood circulation.

# 20.3.4. Applications of enzymes in baking

For decades, enzymes such as malt and fungal alpha-amylases have been used in bread-making. Rapid advances in biotechnology have made a number of exciting new enzymes available for the baking industry. The importance of enzymes is likely to increase as consumers' demand more natural products free of chemical additives. For example, enzymes can be used to replace potassium bromate, a chemical additive that has been banned in a number of countries.

# 20.3.5. Application of enzymes in cheese

The most obvious use of enzyme action in the dairy industry is the coagulation of milk by chymosin. Yet there are many other examples of the involvement of enzymes in determining the quality of milk and milk products that, when the role of the enzyme is properly understood, could be used by the industry to improve the profitability, quality and safety of milk production, and product manufacture. Compared with sectors such as starch hydrolysis, the volume of enzyme use in the dairy sector is low, yet there are many opportunities for specialized applications in product ripening, quality control, preservation and genetic improvements to fermentation cultures.

# 20.3.6. Application of enzymes in yoghurt

Like cheese, yoghurt is produced from milk by the action of lactate producing bacteria, especially *lactobacillus bulgaricus and streptococcus thermophiles*. These bacteria are commonly used in yoghurt starter cultures. Fermentation produces lactate which brings the pH down to about 4.0. Fermentation-by products, including ethanal and methanoic acid, give yoghurt its characteristics flavor. Sometimes fruit pulp, coloring and flavors are added before packaging. Some yoghurt is heat-treated before or after packaging to kill any bacteria, but most yoghurt contain live bacteria.

# 20.3.7. Application of enzymes in breads making.

Bread production involves harvesting the wheat, separating the grain from the husk, crushing the grain to make flour, mixing the flour with water and then finally baking it. The main difference between unleavened and leavened bread is that leavened or risen bread uses leavened dough, and unleavened bread does not. If the leavened bread is desired, then one adds yeast and allowing the bread to sit for a specific amount of time, depending on the type of bread being made.

## **Types of Unleavened Bread**

- 1. Chapatti: Our staple chapatti is widely consumed across India and is a great example of unleavened bread. It is made using atta flour although there are variations that replace atta with wheat, gram, corn flour, or a combination of all three.
- 2. Matzah: Jews only consume matzah during the Jewish Passover, which is unleavened bread. This bread is consumed in remembrance of the Jewish exodus of Egypt, during which the Jews fled in such haste that there was no time to allow their breads to rise up. Matzah is made according to strict interpretations of the Torah using kosher flour whole grain wheat flour.
- 3. Tortilla: Commonly eaten in Mexico and Spain, tortillas are made from corn flour or wheat flour and are similar in appearance to the chapati. Tortillas are flattened and browned over a skillet.
- 4. Pancakes: Pancakes without yeast are considered to be unleavened. Most pancakes are cooked on a griddle and flipped over once the first side has been cooked.

## **Types of Leavened Breads**

Yeast is commonly used to leaven bread and is typically added with sugar or honey to catalyse and activate the yeast in order for the bread to rise. Breads made with yeast is normally allowed to rest for an hour so that it can rise and double in size. It is then punched down and allowed to rise once again before baking. Most types of yeast breads include standard sandwich bread, pizza crust, donuts, and loaf breads and so on.

While yeast is a commonly used leavening agent, it is not the only ingredient that can be used for leavening. Quick breads are any type of breads that are made with an ingredient other than eggs or yeast as a leavening agent. Baking soda and baking powder are common leavening agents and both usually have salt added to the recipe to activate the leavening agent. Quick breads, unlike yeasted breads, are not let to rest before baking. Common types of quick breads include biscuits, muffins, scones, banana bread and cornbread. There are also loaf breads like soda breads which are a type of quick breads. Some donut and pizza recipes are made in the quick bread version.

# Steps involved in bread making

The dough that we make in our bakeries follows all of these 10 steps from start to finish. This ensures we produce the best quality bread without compromising taste, texture, nutrition or our artisan craft. As a home baker, if you follow these 10 steps when making breads at home, you will be on the right path to creating superb loaves.

# 1. Ingredients used to make breads

Using good quality ingredients is crucial to making good bread. The main ingredients

include: bread-flour, dry yeast ('rapid rise'), levain (sourdough), salt, water, sugar, and eggs.

## 2. Mixing

There are two stages to the mixing process: the first is to incorporate ingredients, the second is to develop the structure of the dough, otherwise known as the gluten network. Dough can be kneaded by hand, or mixed in a table top mixer. When using a table top mixer, keep it to the lower speeds to avoid damaging the motor.

## 3. Primary Fermentation

Also referred to as rising, or proofing, this is where the yeast starts to do its work, converting sugars into carbon dioxide, alcohol and organic acids. Every dough has a different primary fermentation time, depending on its formulation. We work with time as well as our senses to determine when the dough is properly fermented.

## 4. Divide and Pre-Shape

When the dough is properly fermented, it is time to divide it to the desired size and give the divided pieces a preshape. A preshape is an intermediate shape a loose suggestion to the dough of where it's headed that will make final shaping easier.

### 5. Bench Rest

After the dough has been preshaped, it needs to rest for a short time before final shaping. Bench rest is typically 15-20 minutes long and during that time, the gluten network, which has been made more elastic through handling, will relax and become more extensible.

# 6. Final Shaping

There are four basic shapes in bread making: the baguette (stick), the boule (round), the bâtard (a football-like shape) and the pan loaf. After shaping, the dough must be set somewhere to rest during its final fermentation. For baguettes and bâtards, we use baker's linen and wooden boards; for boules, we often use wooden proofing baskets. The linen and the baskets help to hold the shape of the dough during the final fermentation.

### 7. Final Fermentation

After shaping, the dough must rest and continue to ferment. The length of the final fermentation varies from dough to dough; it could be anywhere from 15 minutes to 12 or more hours. Again, we work with time and with our "dough sense" to determine when the dough is properly fermented.

## 8. Scoring

Most loaves will be scored, or cut, just before they are baked. Scoring has a decorative function, and it allows the dough to spring properly as the carbon dioxide gas that has accumulated during fermentation expands in the heat of the oven. Scoring is typically done with a razor blade or a small serrated blade.

## 9. Baking

Lean dough (those like baquettes and levain breads made without fats, sugars, eggs, etc.) are typically baked at a very high temperature, around 450-475°F. Enriched breads (brioche, challah, sweet breads) are typically baked around 350-400°F. In most cases, a smaller loaf should be baked at a higher temperature than a larger one, so that it will take on the right amount of color in its baking time. There are a few different ways to determine that a loaf is properly baked by color, by the hollow sound you hear when you knock on the bottom of the loaf, and by internal temperature (at least 190°F for lean breads, 165°F for enriched breads).

## 10. Cooling

Although it is tempting to eat hot bread right of the oven, that's not the best way to really taste its subtle flavors. When bread first comes out of the oven, it is still filled with excess moisture and carbon dioxide. The bread needs time to cool so that the moisture and gas will dissipate. After cooling, the texture, flavor and aroma of the bread will have developed into what they should be and you will have a flavorful, palate-pleasing loaf.

# Self-assessment 20.3

- 1. Explain the application of enzymes in brewing.
- 2. Explain the application of enzymes in cheese and yoghurt.
- 3. Discuss the steps involved in bread making.

# 20.4. Fermentation and fermenters and production of penicillin

# **Activity 20.4**

Use charts, internet, text books and illustrations to explain how fermentation is involved in production of penicillin.

### 20.4.1. Fermentation and fermenters

**Fermentation** is anaerobic breakdown of organic compounds by living cells (microorganisms) that produces ethanol and carbon dioxide or lactate (lactic acid). It occurs in yeast and bacteria, but also in oxygen-starved muscle cells, as in the case of lactic acid. Fermentation is also used more broadly to refer to the bulk growth of microorganisms on a growth medium, often with the goal of producing a specific chemical product. French microbiologist Louis Pasteur is often remembered for his insights into fermentation and its microbial causes. The science of fermentation is known as zymology. To many people, fermentation simply means the production of alcohol: grains and fruits are fermented to produce beer and wine. If a food soured, one might say it was 'off' or fermented. Fermentation react NADH with an

endogenous, organic electron acceptor. Usually this is pyruvate formed from the sugar during the glycolysis step. During fermentation, pyruvate is metabolized to various compounds through several processes:

- a. Ethanol fermentation, alcoholic fermentation, is the production of ethanol and carbon dioxide.
- b. Lactic acid fermentation refers to two means of producing lactic acid:

**Homolactic fermentation** is the production of lactic acid exclusively.

**Heterolactic fermentation** is the production of lactic acid as well as other acids and alcohols.

Sugars are the most common substrate of fermentation, and typical examples of fermentation products are ethanol, lactic acid, Carbon dioxide, and hydrogen gas (H<sub>2</sub>). However, more exotic compounds can be produced by fermentation, such as butyric acid and acetone. Yeast carries out fermentation in the production of ethanol in beers, wines, and other alcoholic drinks, along with the production of large quantities of Carbon dioxide. Fermentation occurs in mammalian muscle during periods of intense exercise where oxygen supply becomes limited, resulting in the creation of lactic acid.

A fermenter also known as **bioreactors** are an apparatus that maintains optimal conditions for culture and growth of microorganisms (on liquid or solid media) to be used in large-scale fermentation and in the commercial production of antibiotics and hormones. The processes that take place in fermenters refers as fermentation which includes aerobic and anaerobic processes.

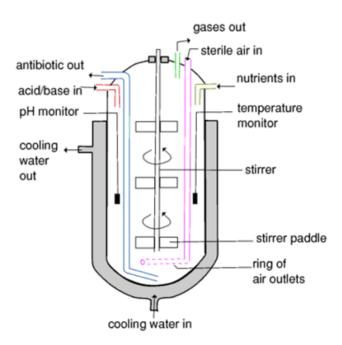


Figure 20.3: Diagram of a fermenter

# 20.4.2. Production of penicillin: Antibiotic

Penicillin, an important part of our anti-microbial armament, had a significant impact on the second half of the twentieth century. Deep-fermentation methods, which were primarily developed for the production of penicillin during the war, gave rise to the development of antibiotics and contributed to the nascent biotechnology industry which appeared in the 1970s.

## **Penicillin production**

In laboratory, it is relatively easy to grow microbes on a small scale in petri dishes. test tubes and flasks, given a suitable nutrient medium, and good environmental conditions. Producing chemicals like penicillin antibiotic from microbes on an industrial scale becomes more complicated as a big number of organisms have to be grown for the venture to be commercially viable. Laboratory procedure should be modified so that it can be used on an industrial scale. This is called scaling up. With scaling up, microorganisms are grown in very large vessels called fermenters or bioreactors. Scaling up to be effective, it requires specialized biologists and engineers to deal with the following problems:

- Avoiding risks of contamination. Only desired organisms must be allowed to grow in the vessel. Others are excluded.
- Big fermenters are built to very strict and specific design.
- Microorganisms should be kept in conditions that allow the optimum production of required substances. This requires installing highly sensitive equipment that maintains PH, temperature and fluid volume within very strict limits.
- To keep nutrients at optimum levels as microbial population grows.
- Removing large amount of heat generated by high levels of microbial activity via a heat exchanger, so that a constant temperature can be maintained.
- Minimizing the build-up of end-products (inhibitors) which may reduce production.
- Monitoring and controlling formation of the foam (unavoidable consequence of carbon dioxide production in a nutrient-rich solution).
- Providing adequate amount of Oxygen to cultures of aerobic organisms by aeration with small bubbles of sterile air which have a large surface area to volume ratio.

Types of culture (of fermentation): there are two main types of culture used in industrial processes such as batch culture and continuous cultures.

In Batch cultures or batch fermentation (closed system), cells are grown in a fixes volume of liquid medium in a closed vessel The conditions are set up and not changed from outside once fermentation starts; for example: no microorganisms, nutrients, or fluid are added or removed from the culture during the incubation period. That is why the process is described as a closed system. The process is stopped once sufficient products have been formed. The contents of fermenter are

then removed, isolated, microorganisms discarded and fermenter is cleaned, and set up for a fresh batch.

Batch cultivation is used to produce secondary metabolites such as penicillin and other antibiotics which are relatively unstable, and not essential for growth of the culture. These secondary metabolites can be extracted economically only when they reach a high concentration in the culture suspension.

In continuous cultures (open system), nutrients are added and cells harvested at a constant rate, so that the volume of suspension is also kept constant. This means that fermenters does not have to be emptied, cleaned and refilled very often. The production is almost continuous. Continuous cultures are very expensive because they need high equipment to maintain constant conditions, and highly skilled staff to operate the equipment.

Table 20.1: Advantages and disadvantages of batch and continuous culture

	advantages	disadvantages
	<ul> <li>Suitable for producing secondary metabolites whose production is not associated with growth. E.g. antibiotics</li> <li>Can use stains which are</li> </ul>	Turnaround time between batches can be prolonged, wasting possible production time  Environment changes in the
Batch culture	<ul> <li>Can use stains which are too unstable for continuous culture.</li> <li>Easier to set up and run than continuous culture.</li> <li>Fermenters are less specialized and may be used for a greater variety of processes, depending on demand.</li> </ul>	Environment changes in the fermenters as the fermentation progresses. Nutrients get
		used up and products build up. Nutrients get used up and products build up. Heat
		out, acid or alkali production, Oxygen consumption increase in a rate as growth progresses. Therefore, conditions
		gradually become unfavorable and growth rate become unfavorable and growth rate gradually declines.

_	Gives more control. Aim
	is to keep environmental
	conditions constant.
	Nutrients are replaced as
	fast as they are used and
	products are removed as
	fast as they are made.

- Productivity is greater because there is no turnaround time (continuous process). Therefore, more cost effective in some situations.
- Greater risk of contamination, although good engineering design can solve this problem.
- Control is more complex.
- When used for brewing gives greater yields but has given flavour problem.

## Continuous culture

- Optimum (maximum or exponential) growth rate is maintained once achieved.
- Small fermenters are need because higher yields are obtained.
- More suitable for production of biomass. Also used for production of ethanol whose synthesis is proportional to rate of growth.
- Demand for labour is more regular

The industrial production of penicillin was generally classified into two processes: Upstream processing and downstream processing. Upstream processing encompasses any technology that leads to the synthesis of a product and includes the exploration, development and production. Downstream processing refers as the extraction and purification of a biotechnological product from fermentation or at the end of culture process. Usually the contents of fermenter are first separated into liquid component and a solid component which contain the cells. This is usually done by filtration or centrifugation. The liquid may contain the desired product in solution or it may be the cells or some products inside the cells that it needs.

Penicillin is produced commercially by growing the fungus *Penicillium chrysogenumin* large stirred fermenters. A solution of essential salts and a nitrogen source are put into the fermenter together with an inoculum of the fungus. All procedures are performed aseptically. The PH of the medium is regulated with ammonium salts

at 6.5 to 7.0. Lactose (a slowly hydrolysed disaccharide) is added to promote cell growth and reproduction and minimize penicillin production. On completion of fermentation (usually 6-7 days) the broth is separated from the fungal mycelium and penicillin extracted. This penicillin can then be modified by chemical procedures to yield a variety of semisynthetic penicillins.

### **Modern Production Methods**

Significant improvements in modern production methods have increased production and decreased cost. Today, commercial producing strains of Penicillium chrysogenum are grown using submerged culture in constantly agitating and aerated 50,000- gallon stainless steel tanks. These industrial strains can now produce 40-50 grams of penicillin per liter of culture with a 90% recovery yield. This is an overwhelming improvement from the earliest Peoria farmer's market strain that only produced 0.15 grams per liter with very low recovery rates. In order to achieve these production rates, modern Penicillium strains display a host of genetic and cellular modifications that result in increased production, including amplification of the penicillin biosynthesis gene cluster, an increased number of peroxisomes, and elevated levels of transporter proteins that secrete newly produced penicillin out of the peroxisomes and the cell.

Temperature and pH are normally controlled in the fermenter. Temperature is kept constant, while pH is held at a value of 5.5 for the first stage of the fermentation and then raised to 6.8 and kept constant for the remainder of the fermentation period.

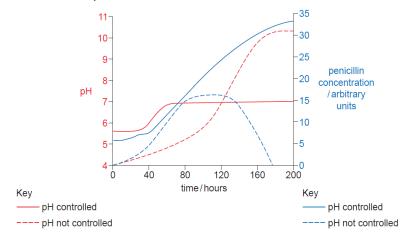


Fig. 20.4: Shows how the pH and the concentration of penicillin in the culture change overtime, when the pH is controlled and when the pH is not controlled.

### Self-assessment 20.4

- 1. What is fermenter?
- 2. Write on upstream processing and downstream processing.
- 3. Write on your own word penicillin.

- 4. Contrast commercial-scale production from laboratory-scale production of penicillin.
- 5. Explain why the continuous culture is described as open system.
- 6. Explain why the batch culture is described as closed system.
- 7. Discuss advantages and disadvantages of batch culture?
- 8. Explain why continuous culture is very expensive.
- 9. What are Advantages and disadvantages of continuous culture?

## 20. 5. Antibiotics

## **Activity 20.5**

Using addition resources to your textbook available in your school such as the books from the school library and search further information from the internet: Brainstorm on the antibiotic resistance and implications of antibiotic use.

Antibiotics are powerful medicines that fight certain infections by either stopping bacteria from reproducing or by destroying them. Before bacteria can multiply and cause symptoms, the body's immune system can usually kill them. The word antibiotic means "against life." Any drug that kills germs in your body is technically an antibiotic.

### How do antibiotics work?

Antibiotics are used to treat bacterial infections. Some are highly specialized and are only effective against certain bacteria. Others, known as broad-spectrum antibiotics, attack a wide range of bacteria, including ones that are beneficial to us.

There are two main ways in which antibiotics target bacteria. They either **prevent** the reproduction of bacteria, or they kill the bacteria, for example by stopping the mechanism responsible for building their cell walls. There are now hundreds of different types of antibiotics, but most of them can be broadly classified into six groups. These are outlined below.

**Penicillin** – widely used to treat a variety of infections, including skin infections, chest infections and urinary tract infections.

**Cephalosporins** – can be used to treat a wide range of infections, but are also effective for treating more serious infections, such as septicaemia and meningitis.

**Aminoglycosides** – tend to only be used to treat very serious illnesses such as septicaemia, as they can cause serious side effects, including hearing loss and kidney damage; they break down quickly inside the digestive system, so they have to be given by injection, but are also used as drops for some ear or eye infections.

**Tetracyclines** – can be used to treat a wide range of infections; commonly used to treat moderate to severe acne and rosacea, which causes flushing of the skin and spots.

**Macrolides** – can be particularly useful for treating lung and chest infections; can also be a useful alternative for people with a penicillin allergy or to treat penicillin-resistant strains of bacteria.

**Fluoroquinolones** – broad-spectrum antibiotics that can be used to treat a wide range of infections. They include: Hypocholesterolemic agents, Lipopeptide, Macrolides, Monobactams, Nitrofurans, Oxazolidinones, Polypeptides, Quinolones, Sulfonamides, Tetracyclines, Lincosamides, Glycopeptides, Immunosuppressive agents, Anti-migraine agents, Anti-bacterials, Antifungals, Penicillins, Aminoglycosides, Ansamycins, Carbapenems, Cephalosporins (1,2, 3, 4, 5 generations), and Fluoroquinolones.

## 20.5.1. Antibiotic resistance

Antibiotic resistance occurs when an antibiotic has lost its ability to effectively control or kill bacterial growth; in other words, the bacteria are "resistant" and continue to multiply in the presence of therapeutic levels of an antibiotic.

## Why do bacteria become resistant to antibiotics?

Antibiotic resistance is a natural phenomenon. When an antibiotic is used, bacteria that can resist that antibiotic have a greater chance of survival than those that are "susceptible." Susceptible bacteria are killed or inhibited by an antibiotic, resulting in a selective pressure for the survival of resistant strains of bacteria.

Some resistance occurs without human action, as bacteria can produce and use antibiotics against other bacteria, leading to a low-level of natural selection for resistance to antibiotics. However, the current higher-levels of antibiotic-resistant bacteria are attributed to the overuse and abuse of antibiotics. In some countries and over the Internet, antibiotics can be purchased without a doctor's prescription. Patients sometimes take antibiotics unnecessarily, to treat viral illnesses like the common cold.

### How do bacteria become resistant?

Some bacteria are naturally resistant to certain types of antibiotics. However, bacteria may also become resistant in two ways: by a genetic mutation or by acquiring resistance from another bacterium.

Mutations, rare spontaneous changes of the bacteria's genetic material, are thought to occur in about one in one million to one in ten million cells. Different genetic mutations yield different types of resistance. Some mutations enable the bacteria to produce potent chemicals (enzymes) that inactivate antibiotics, while other mutations eliminate the cell target that the antibiotic attacks. Still others close up the entry ports that allow antibiotics into the cell, and others manufacture pumping mechanisms that export the antibiotic back outside so it never reaches its target.

Bacteria can acquire antibiotic resistance genes from other bacteria in several ways.

By undergoing a simple mating process called "conjugation," bacteria can transfer genetic material, including genes encoding resistance to antibiotics (found on plasmids and transposons) from one bacterium to another. Viruses are another mechanism for passing resistance traits between bacteria. The resistance traits from one bacterium are packaged into the head portion of the virus. The virus then injects the resistance traits into any new bacteria it attacks. Bacteria also have the ability to acquire naked, "free" DNA from their environment. Any bacteria that acquire resistance genes, whether by spontaneous mutation or genetic exchange with other bacteria, have the ability to resist one or more antibiotics. Because bacteria can collect multiple resistance traits over time, they can become resistant to many different families of antibiotics.

## How does antibiotic resistance spread?

Genetically, antibiotic resistance spreads through bacteria populations both "vertically," when new generations inherit antibiotic resistance genes, and "horizontally," when bacteria share or exchange sections of genetic material with other bacteria. Horizontal gene transfer can even occur between different bacterial species. Environmentally, antibiotic resistance spreads as bacteria themselves move from place to place; bacteria can travel via airplane, water and wind.

People can pass the resistant bacteria to others; for example, by coughing or contact with unwashed hands.

### Can bacteria lose their antibiotic resistance?

Yes, antibiotic resistance traits can be lost, but this reverse process occurs more slowly. If the selective pressure that is applied by the presence of an antibiotic is removed, the bacterial population can potentially revert to a population of bacteria that responds to antibiotics.

# 20.5.2. Implications of antibiotic use

Antibiotics are considered the keystone of modern medicine, but their excessive use continues to generate unwanted side effects. While specialists are making strides to preserve the effectiveness of antibiotics and to slow potential infections through better policy, the overuse of antibiotics continues to have severe health consequences around the world.

### Self-assessment 20.5

- 1. What do you understand by antibiotic resistance?
- 2. Explain how bacteria become resistant.
- 3. Discuss on how bacteria lose their antibiotic resistance.
- 4. Write on implications of antibiotic use.
- 5. Talk on how antibiotic resistance spreads

# 20.6. Biogas production

## **Activity 20.6**

Use diagrams or illustrations and visiting a biogas plants in your region, describe the stages of biogas production and its significance in your area (a simple biogas generator can also be made in schools).

Biogas typically refers to a mixture of different gases produced by the breakdown of organic matter (methanogens or archaebacterial) in the absence of oxygen. Biogas is produced by anaerobic fermentation of organic wastes such as agricultural waste, manure, municipal waste, plant material, sewage, green waste, or food waste. It is a renewable energy source and in many cases exerts a very small carbon footprint.

Biogas is primarily methane ( $CH_4$ ) and carbon dioxide ( $CO_2$ ) and may have small amounts of hydrogen sulphide ( $H_2S$ ), moisture and siloxanes. The gases methane, hydrogen, and carbon monoxide (CO) can be combusted or oxidized with oxygen. This energy released allows biogas to be used as a fuel; it can be used for any heating purpose, such as cooking.

It can also be used in a gas engine to convert the energy in the gas into electricity and heat. Biogas can be compressed, the same way the natural gas is compressed to compressed natural gas (CNG), and used to power motor vehicles. In the UK, for example, biogas is estimated to have the potential to replace around 17% of vehicle fuel. It qualifies for renewable energy subsidies in some parts of the world. Biogas can be cleaned and upgraded to natural gas standards, when it becomes bio methane.

#### **Production**

Biogas is produced as landfill gas (LFG), which is produced by the breakdown of biodegradable wastes inside a landfill due to chemical reactions and microbes, or as digested gas, produced inside an anaerobic digester. A biogas plant is the name often given to an anaerobic digester that treats farm wastes or energy crops. It can be produced using anaerobic digesters (air-tight tanks with different configurations). These plants can be fed with energy crops such as maize silage or biodegradable wastes including sewage sludge and food waste. During the process, the microorganisms transform biomass waste into biogas (mainly methane and carbon dioxide) and digestate (remaining organic matter not transformed into biogas).

The biogas is a renewable energy that can be used for heating, electricity, and many other operations that use a reciprocating internal combustion engine, such as a General Electrical (GE) Jenbacher or Caterpillar gas engines. Other internal combustion engines such as gas turbines are suitable for the conversion of biogas into both electricity and heat. The remaining organic matter that was not transformed into biogas. It can be used as an agricultural fertilizer.

There are two key processes: mesophilic (A mesophyll is an organism that grows best in moderate temperature, neither too hot nor too cold, typically between 20 and 45 °C) and thermophilic (A thermophile is an organism, a type of extremophile, that thrives at relatively high temperatures, between 41 and 122 °C) digestion which is dependent on temperature. The production of biogas involves three stages and three communities of microorganisms namely

- 1. Anaerobic fermentation by eubacteria including lactobacillus, which converts the organic waste into a mixture of organic acids and alcohol, with some Hydrogen, Carbon dioxide, and acetate.
- 2. Acetogenic (acetate-producing) reaction by bacteria acetobacterium which, in addition to acetate, produce hydrogen and Carbon dioxide from the organic acid and alcohol.
- **3. Methanogenic (methane-producing) reactions** by archaebacteria, including Methanobacterium, Metanococcus, and Methanospirillum. The archaebacteria generate methane either:
- By reducing the carbon dioxide:  $CO_2 + 4H_2 \longrightarrow CH_4 + H_2O$ , or
- By converting acetate: CH₃COOH 
  → CH₄ + CO,

## Composition

The composition of biogas varies depending upon the origin of the anaerobic digestion process. Landfill gas typically has methane concentrations around 50%.

	Formula	%	
Methane	CH <sub>4</sub>	50-75	
Carbone dioxide	CO <sub>2</sub>	25-50	
Nitrogen	N <sub>2</sub>	0-10	
Hydrogen	H <sub>2</sub>	0-1	
Hydrogen sulphide	H <sub>2</sub> S	0-3	
Oxygen	O <sub>2</sub>	0-0.5	

Table 20.2: Typical composition of biogas

In some cases, biogas contains siloxanes. They are formed from the anaerobic decomposition of materials commonly found in soaps and detergents. During combustion of biogas containing siloxanes, silicon is released and can combine with free oxygen or other elements in the combustion gas.

# **Applications**

Biogas can be used for electricity production on sewage works, in a combined heat and power (CHP) gas engine, where the waste heat from the engine is conveniently used for heating the digester; cooking; space heating; water heating; and process heating. If compressed, it can replace compressed natural gas for use in vehicles, where it can fuel an internal combustion engine or fuel cells and is a much more effective displacer of carbon dioxide than the normal use in on-site CHP plants.

### Self-assessment 20.6

- 1. What part do acetogenic reactions play in the production of biogas?
- 2. The archaebacteria generate methane either by reducing the carbon dioxide, or by converting acetate: write chemical equations for the two processes.

### **End of unit assessment 20**

## **Multiple choice questions**

- 1. During penicillin production, temperature is maintained at
  - a. room temperature
  - b. 26 °C
  - c. 36 °C
  - d. 46 °C
- 2. In penicillin production, pH of culture medium is maintained between
  - a. 5 and 6
  - b. 4 and 6
  - c. 6 and 7
  - d. 4 and 5
- 3. To produce penicillin, main fermentable source in culture is
  - a. glucose
  - b. lactose
  - c. sulphate
  - d. sugars
- 4. Penicillin production is optimum in
  - a. batch operation systems
  - b. continuous operation systems
  - c. discontinuous operation system
  - d. unique operation system
- 5. What is fermentation?
- 6. The senior four Biology teacher said: "the biogas can contribute to the economic development of Rwanda" defend his idea.
- 7. Explain how are immobilized enzymes made?
- 8. Explain the medical applications of enzymes.
- 9. Explain the importance of using yeast in bread making.
- 10. What Are the Main Ingredients of Bread?

- 11. Summarize the advantages of using immobilized enzymes rather than enzyme solutions.
- 12. Describe the composition of biogas.
- 13. Describe three stages that are involved in production of biogas.

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### **Electronic links**

https://study.com/academy/practice/quiz-worksheet-louis-pasteur-germ-thioryof-disease.html

http://printerfriendly.adam.com/content.aspx?productId=117&pid=1&gid=001439 &c-custid=758

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https://opentextbc.ca/anatomyandphysiology/Epithelial\_Tissue.jpg

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