

# LABORATORY

# BIOLOGY

## SEMESTER I (2023/2024)

- Food Test : Test For Carbohydrate, Protein and Lipid
- Cells Structure And Function
- Transport Across Membrane
- Inheritance
- Pollen Germination





**UNISZA SCIENCE AND MEDICINE FOUNDATION CENTRE,  
UNIVERSITI SULTAN ZAINAL ABIDIN,  
GONG BADAQ, TERENGGANU**

## **PAA 10104: BIOLOGY I**

### **Format of Laboratory Report**

1. **Title** : State the title of the experiment.
2. **Objective** : State the objective of the experiment.
3. **Apparatus** : State the instruments used.
4. **Theory** : Explain briefly the theory.
5. **Procedures** : Describe the steps and methods to perform the experiment.
6. **Data/Results** : Present the data/results appropriately.
7. **Discussion** : Explain your results and discuss.
8. **Conclusions** : Indicate what is measured, the uncertainties and the sources of uncertainties
9. **Reference** : List the references either from books and journals.



**PUSAT ASASI SAINS DAN PERUBATAN UNISZA,  
UNIVERSITISULTAN ZAINAL ABIDIN, GONG BADAQ,  
TERENGGANU**

**LABORATORY REPORT  
PAA 10104: BIOLOGY I**

**TITLE OF EXPERIMENT:**

<b>Name</b>	:	
<b>Student ID</b>	:	
<b>Lecture's Group</b>	:	
<b>Date of Experiment</b>	:	
<b>Date of Submission</b>	:	
<b>Lecturer's name</b>	:	
<b>Group Members</b>	:	1) 2) 3) 4) 5) 6) 7)

## **LEARNING OUTCOMES**

Students are required to conduct biology practical in order to get a better insight into the concept of biology introduced during lectures.

Upon completion of these practical, students should be able to:

- Practice the correct techniques of handling apparatus.
- Plan, understand and carry out the practical as instructed.
- Observe, measure and record data consistency, accuracy and units of the physical quantities.
- Define, analyze data and information in order to evaluate and deduce conclusion from the practical.
- Discuss data and create conclusions from biological data.
- Develop solution to biological problems.
- Measure, record and analyze data as well as to determine the uncertainties (error).
- Determine the limitations to the accuracy of observations and measurements.

## LIST OF PRACTICALS

### BIOLOGY I (Semester I)

<b>Practical</b>	<b>Title</b>	<b>Page</b>
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3.	Transport Across Membrane	14-17
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5.	Pollen Germination	23-24

## EXPERIMENT 1 : FOOD TESTS

### Learning outcome

At the end of this practical, the students are able to:

1. Identify the category of different types of food that contain carbohydrates, proteins and lipid.
2. Identify the usage of each test to differentiate the types of macromolecules.
3. Describe the various forms and classes of macromolecules.

### Objective

To determine the presence of macromolecules in the sample substances.

### Hypothesis

Experiment A:

The stock samples with the presence of reducing sugar will change the colour after tested with Benedict solution. The stock samples with the presence of starch will change the colour after tested with Lugol's solution.

Experiment B:

The stock samples with the presence of protein will change the colour after tested with Biuret test.

Experiment C:

The stock samples with the presence of lipid will form the grease spot after tested with Grease spot test. The stock samples with the presence of lipid will stain red with Sudan red test.

### Introduction

The foods that we eat daily consist of macromolecules, such as carbohydrates, proteins, lipids, minerals and vitamins. Carbohydrates  $C_n(H_2O)_n$  are organic compounds which can be divided into four main chemical groups of monosaccharides, disaccharides, oligosaccharides and polysaccharides. In general, carbohydrates are important for energy storage (starch and glycogen) and as structural components (cellulose and chitin).

Proteins molecules are organic compound found in all living organisms, which play a significant role in growth and maintenance of the body. It improves a person health by influencing the immune system through production of antibodies in the body. Enzymes and hormones that are also a class of protein are vital for metabolism, digestion and other important processes.

Lipids are organic compounds that can supply energy as much as four times the amount of energy in carbohydrates or proteins. The main biological function of lipids includes energy storage, structural components of plasma membrane and as signalling molecules.

### Apparatus

Water bath, test tube, test tube clamp, test tube rack, graduated cylinder beaker, filter paper, parafilm

### Materials

- Benedict's solution

- Carbohydrate - stock samples of glucose, maltose, lactose, sucrose, starch, fructose, orange juice, soft drinks, vegetable oil and soybean drink
- Protein & Lipid – stock samples of distilled water, vegetable oil, soft drink, soybean drink
- Lugol's solution
- Biuret's solution
- Sudan IV solution

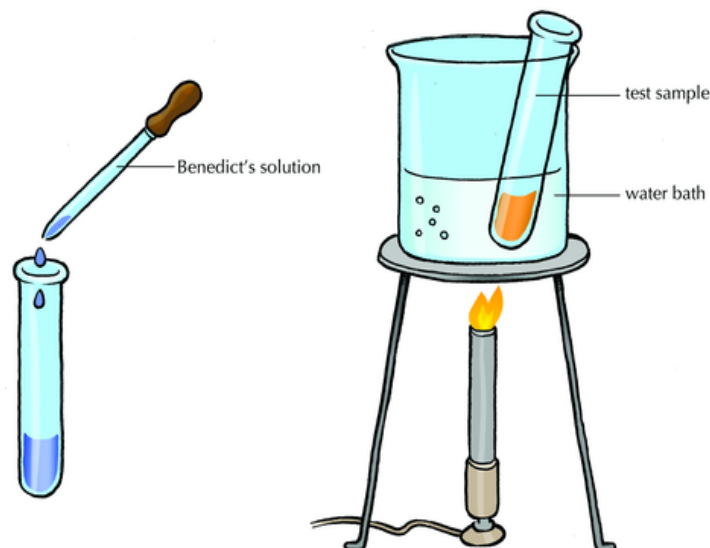
## A. Test for carbohydrates

### (a) Testing for carbohydrates (sugar) using Benedict solution

Benedict's solution composed of copper sulphate, sodium carbonate and sodium citrate is used to test for the presence of reducing sugar. Alkaline solutions of copper are reduced by reducing sugars that have a free aldehyde or ketone group by forming coloured cuprous oxide.

#### Procedures

1. Label all the 10 test tubes with marker for every stock sample (maltose, sucrose, glucose, starch, fructose, lactose, soft drink, orange juice, vegetable oil & soy bean drink).
2. Pipette 3 ml of the correct stock solutions into the test tube. Use different pipette for different stock solution.
3. Add 3 ml of Benedict's solution to each test tube. Agitate the mixture by shaking the tubes from side to side. Record the initial colour.
4. Carefully heat the test tubes by suspending them in a hot water bath at about 90°C for five minutes.
5. Remove the test tubes after 5 minutes.
6. Record the final colour after heating and conclusions regarding the presence/absence of reducing sugar.

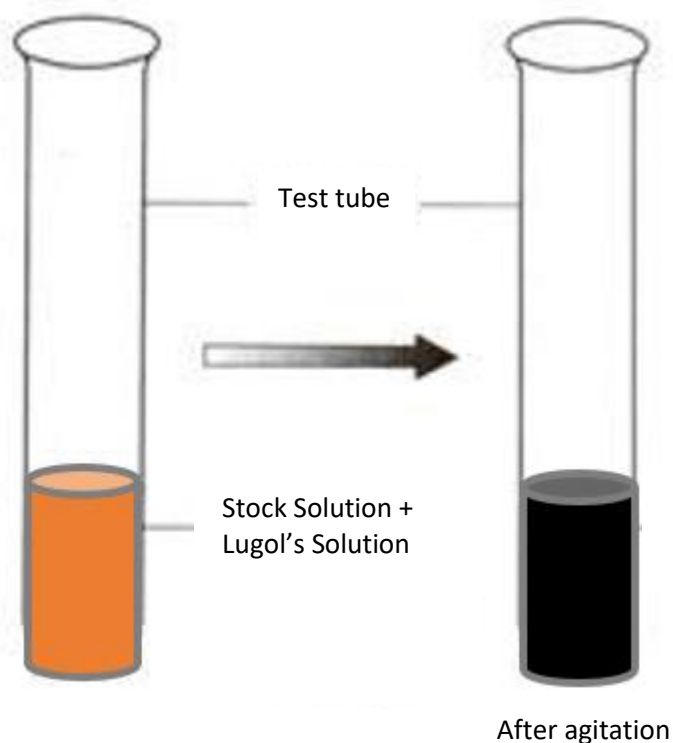


### (b) Testing for carbohydrates (starch) using Lugol's solution

Lugol's solution which contains iodine, potassium iodide mixed with distilled water is a brown solution that is used as an indicator test for the presence of starch. Starches include the plant starches amylose and amylopectin and glycogen in animal cells.

#### Procedures

1. Label all the 6 test tubes with marker for every stock sample (maltose, sucrose, glucose, starch, fructose & lactose).
2. Pipette 3 ml of the correct stock solutions into the test tube. Use different pipette for different stock solution.
3. Record the initial colour.
4. Add 9 drops of Lugol's solution to each test tube. Agitate the mixture by shaking the tubes from side to side. Record the colour after adding the Lugol's solution.



### B. Test for protein

#### (a) Biuret test for protein

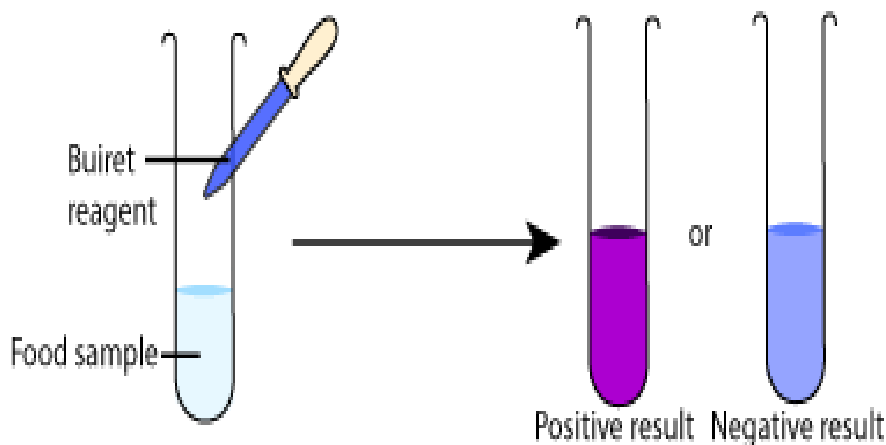
The Biuret test is a chemical test used to identify the presence of protein. Biuret's solution is blue in colour and will change to pink-purple when it reacts with peptide bond.

#### Procedures

1. Label all the 3 test tubes with marker for every stock sample (vegetable oil, soft drink & soy bean drink).
2. Pipette 1 ml of the correct stock solutions into the test tube. Use different pipette for different stock solution.



3. Add 2 ml of Biuret's solution to each test tube. Agitate the mixture by shaking the tubes from side to side.
4. Record the colour of mixture after 2 minutes.



### C. Test for lipid

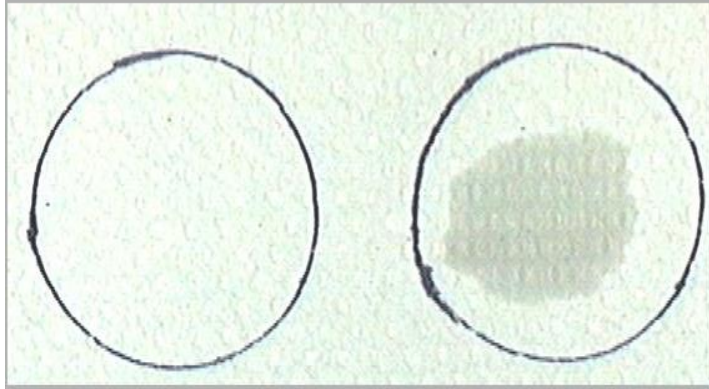
The presence of lipids in a sample substance can be tested by grease spot test and Sudan red test. The grease spot test is used to test for the presence of lipid by making a paper to be translucent. As for Sudan red test, the dye of Sudan red that is lipid soluble will return the sample into red.

#### (a) Grease Spot Test

##### Procedures

1. Obtain a piece of filter paper. Use a pencil to label 2 samples areas, namely distilled water & vegetable oil.
2. Place a drop of distilled water and a drop of vegetable oil on an area. Set aside to dry for 10 minutes.
3. Describe the appearance of each spot on the paper after 10 minutes. Record your observation.

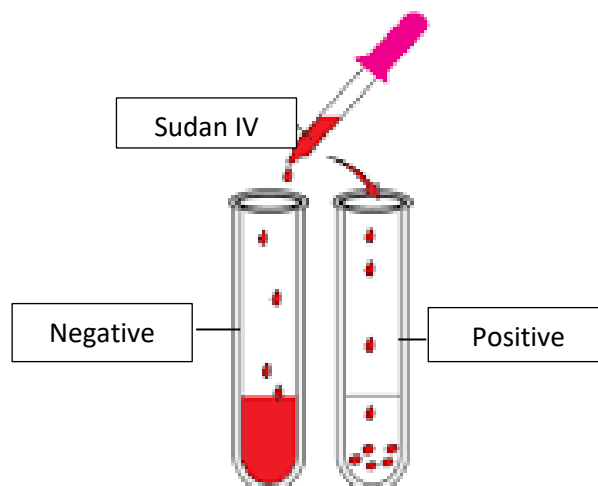
## Grease spot test (Left negative, right positive)



### (b) Sudan IV Test

#### Procedures

1. Add 3 ml of distilled water to 2 test tubes followed by 3 ml of each stock sample (vegetable oil & soft drink).
2. Add 9 drops of Sudan IV to each test tube. Agitate the mixture by shaking the test tube.
3. Add another 2 ml of distilled water to each test tube.
4. Record your observation.



## EXPERIMENT 2 : CELL STRUCTURE AND FUNCTION

### Learning Outcome

At the end of this practical, the students are able to:

1. Identify the cell structures and their components.
2. Describe the function of cells components.
3. Compare the structures of prokaryotic cells and eukaryotic cells.
4. Illustrate the structures of plant and animal cells.

### Objective

To observe the absence of a nucleus in a prokaryotic cell as well as eukaryotic cell using compound microscope.

### Hypothesis

Experiment A:

The plant cells have nucleus, plasma membrane, cytoplasm, cell wall and chloroplast.

Experiment B:

The animal cells have a nucleus, plasma membrane and cytoplasm.

Experiment C:

The prokaryotic cells do not have nucleus but have a nucleoid.

### Introduction

All living organisms can be sorted into one of 2 groups depending on the fundamental structure of their cells. These 2 groups are the prokaryotes and the eukaryotes. Prokaryotes are organisms made up of cells that lack a cell nucleus or any membrane-encased organelles. Eukaryotes are organisms made up of cells that possess a membrane-bound nucleus (that holds genetic material) as well as membrane-bound organelles.

Prokaryotes are evolutionarily ancient. They were here first and for billions of years were the only form of life. Even with the evolution of more complex eukaryotic cells, prokaryotes are very successful. All bacteria and bacteria-like Archaea are prokaryotic organisms. Eukaryotic cells are more complex, evolving from a prokaryotic-like predecessor. Most of the living things that we are typically familiar with are composed of eukaryotic cells; animals, plants, fungi and protists. Eukaryotic organisms can either be single-celled or multi-celled.

### A. Preparing and Studying Slides of Plant Cells

#### Apparatus

Light microscope, scalpel, forceps, a glass dropper, a mounting needle, filter paper

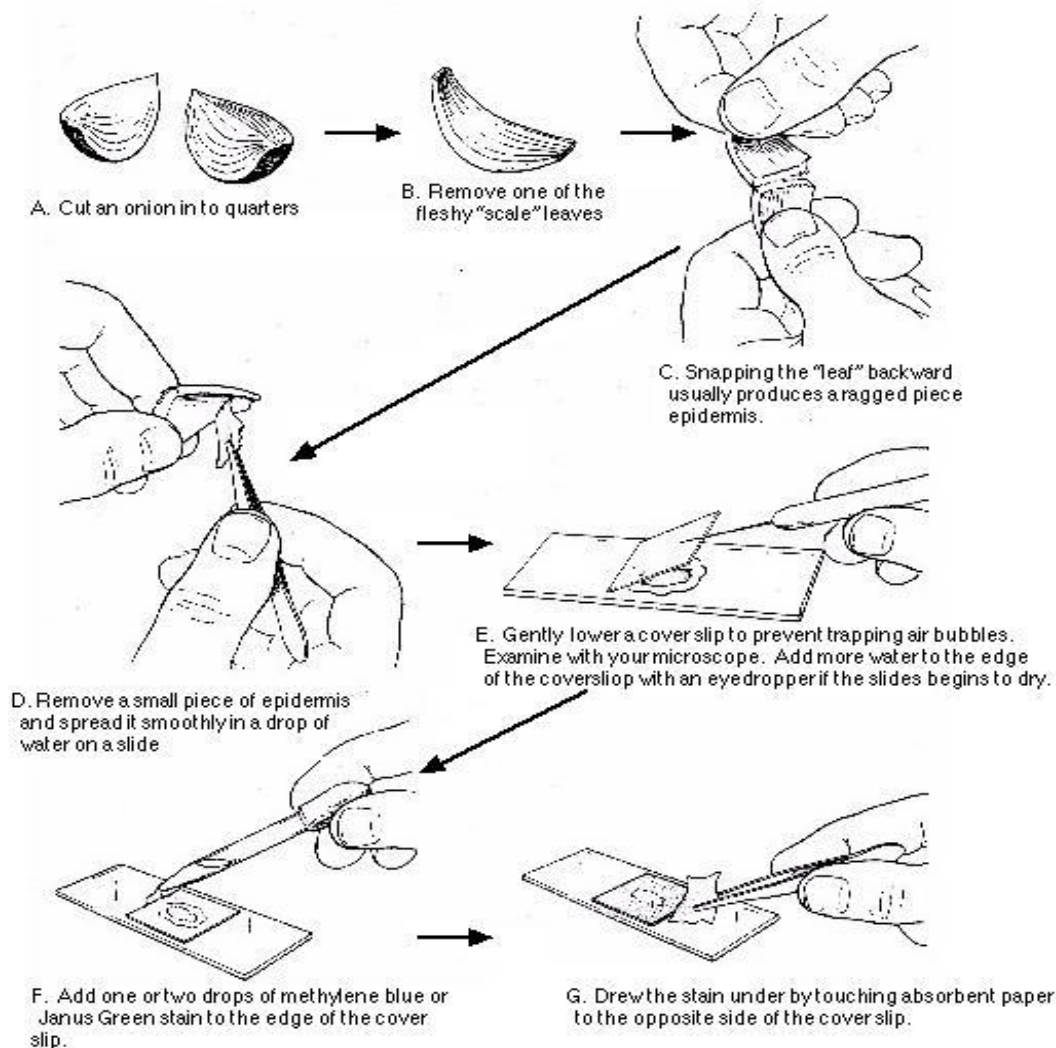
#### Materials

An *Allium* sp. bulb, iodine solution, distilled water, a glass slide, cover slip

#### Procedures

1. A piece of *Allium* sp. scale (1 cm x 1 cm) is cut from *Allium* sp. bulb using a scalpel.
2. The transparent epidermis from the inner surface of the *Allium* sp. scale is gently peeled off using a pair of forceps.
3. The epidermis is then mounted without being folded on a slide in a drop of water.

4. The specimen is covered with a cover slip at an angle of 45°. With the help of a mounting needle, the cover slip is lowered slowly to ensure that air bubbles are not trapped under the cover slip. This specimen slide is called a wet mount. (Note: air bubble that is trapped under the cover slip can be removed by tapping the cover slip gently with the sharp end of a pencil.)
5. The prepared slide is examined under the light microscope, first under low power, then under high power.
6. A drop of dilute iodine solution is placed onto one side of the cover slip. A filter paper is held at the opposite end of the cover slip to draw the iodine solution across the slide to stain the cells. (Note: dilute iodine solution is prepared by dissolving 1.5 g of potassium iodide in 25 ml of water. 1 g iodine is added to the potassium iodide solution.)
7. Excess iodine is absorbed using another filter paper.
8. The slide is observed again under the light microscope.
9. The structure of the epidermal cells is drawn and its components are labelled accordingly. The magnification used is also recorded.



## B. Preparing and Studying Slides of Animals Cells

### Apparatus

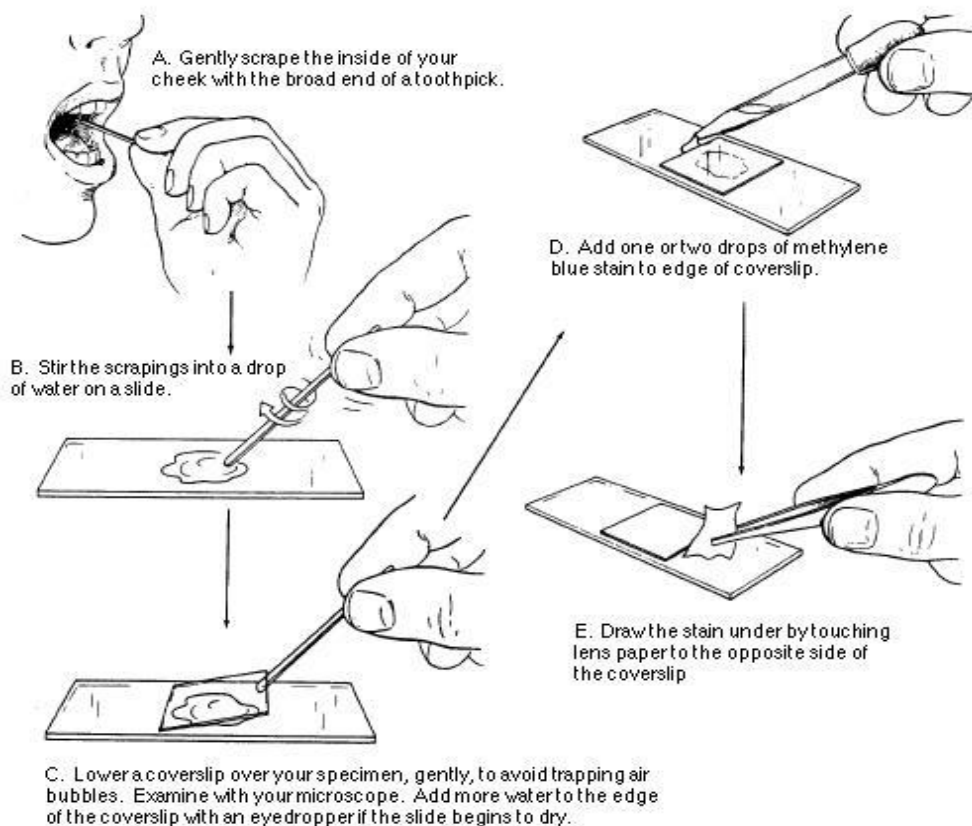
Light microscope, glass slide, cover slip, scalpel, forceps, a glass dropper, a mounting needle, filter paper, toothpicks.

### Materials

Animal cells (cheek cells), methylene blue solution, distilled water,

### Procedures

1. The blunt end of a clean toothpick is used to gently scrape the inner lining of the cheek.
2. The scrapping is mounted in a drop of water on a clean glass slide.
3. The specimen is then covered with a cover slip.
4. A drop of methylene blue solution is placed onto one side of the cover slip. A filter paper is then placed onto one side of the cover slip. A filter paper is the placed at the opposite side of the cover slip to draw the methylene blue solution across the specimen. (Note: methylene blue solution is prepared by dissolving 1 g of methylene blue and 0.6 g of sodium chloride in 100 ml of distilled water)
5. The slide is observed under the low-power objective lens of the light microscope and then the high-power objective lens.
6. The cheek cells are drawn and the cell structures label accordingly. The magnification used is also recorded.



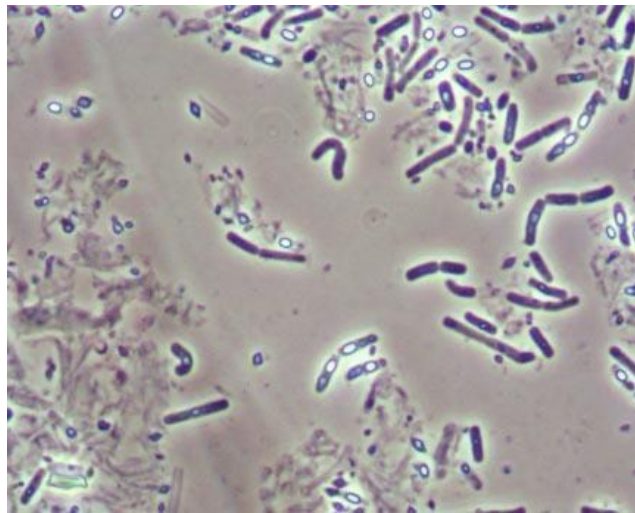
## C. Preparing and Studying Slides of Prokaryotic Cells

### Materials

Plain yogurt with active cultures

### Procedures

1. Clean your slides and cover slips from dust and other particles.
2. Place a very small portion of plain yogurt onto the slide and add one drop of water. Place the coverslip on top.
3. Under low-power objective lens, then to the high-power objective lens of the light microscope find a section where the yogurt is a thin layer; this is a suitable area to observe the bacteria.
4. Switch to high power (100X) for a better view of the bacteria, using immersion oil.
5. Draw and label accordingly on structure of the cells that you have viewed under the light microscope. The magnification used is also recorded.



## EXPERIMENT 3: TRANSPORT ACROSS MEMBRANE

### Learning Outcome

At the end of this practical the students are able to:

1. Define the osmotic pressure of a cell.
2. Describe the transport across membrane cell process.
3. Explain the haemolysis process in a red blood cell.

### Objectives

1. To determine the osmotic pressure of a cell.
2. To determine the osmotic pressure in the atmosphere from the graph.
3. To determine the concentration of sodium chloride solution that causes haemolysis.
4. To determine the concentration of sodium chloride that is isotonic to red blood cells.

### Hypothesis

Experiment A:

If the potato strips are placed in the hypotonic solution, its size will increase. If the potato strips are placed in the isotonic solution, its size will be the same. If the potato strips are placed in the hypertonic solution, its size will decrease.

Experiment B:

If the blood cells are placed in the hypotonic solution, its shape will be haemolysed. If the blood cells are placed in the isotonic solution, its shape will be the same. If the blood cells are placed in the hypertonic solution, its shape will be crenated.

### Introduction

The cell membrane is a selective permeable structure because only selected materials can pass through it. Water molecules can easily pass through the membrane and the movement of water is called osmosis. The direction of movement of water molecules is determined by the concentration of the solutes of both sides of the membrane. The water potential inside and outside of the cell is said to be isotonic, that is the movement of water molecules in both direction is at the same rate. The vacuolar membrane is also a selective structure and the condition in the vacuole is isotonic to the cell environment.

In a hypertonic environment, water molecules will move out of the cell and the cell shrinks. The shrinking of cell is due to the hypertonic environment outside the plant and animals cells. The shrinking of plant cell is called plasmolysis while the shrinking of animal cell is called crenation.

When a plant cell is in a hypotonic environment, it will expand but the increase in size is restricted by the cell wall (turgid). On the other hand, animal cells which are in the hypotonic environment will expand and burst and this is called lysis or haemolysis.

### A. Osmotic pressure

#### Apparatus

Boiling tube, beaker, petri dish, measuring cylinder (25 ml), forceps, pipette (10 ml), cork borer, electronic balance.

## Materials

Fresh potato tuber, sucrose solutions 1.0M (40 ml per student), distilled water, labelling paper, graph paper, razor blade, tile, ruler, filter paper.

## Procedures and Observation

1. Prepare 20 ml of sucrose solution with different molarities using the dilution method. The molarities required are 0.2M, 0.3M, 0.4M and 0.5M. The volume of the sucrose solution (1.0M) and the distilled water must be recorded into the table below. Label every tube.

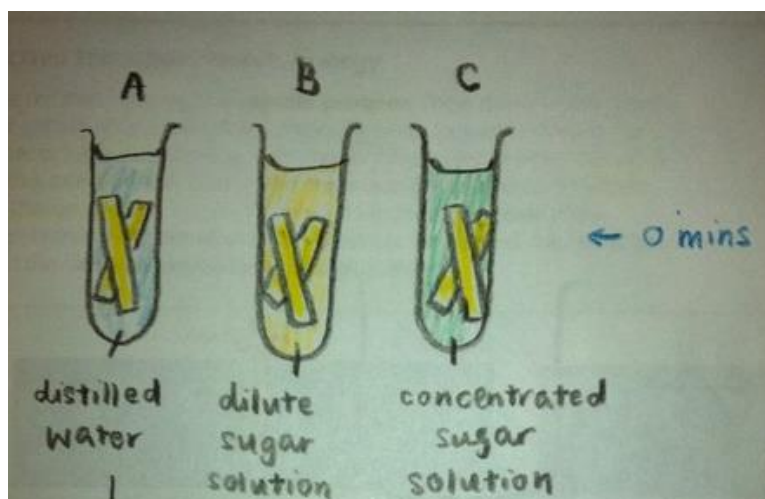
Molarity	0.1M	0.2M	0.3M	0.4M	0.5M
Volume of 1.0 M sucrose (1ml)					
Volume of distilled water (ml)					

2. Prepare 15 pieces of potato strips using cork borer. The length of each strip should approximately be 4 – 6 cm.
3. Take 3 strips of potato, record the initial weight of each strip and put them into the tube. Repeat with the other concentration of sucrose solutions.
4. After 30 minutes, remove the three strips from each boiling tube, wipe and immediately record the final weight of each strip in the same table.
5. Determine the average changes in weight (%) of potato strips.
6. Using a graph paper:
  - (i) Draw a standard graph of osmotic pressure against the molarity of sucrose solution to determine the osmotic pressure of the potato strip.

Molarity (M)	0.05	0.10	0.15	0.2	0.25	0.3	0.35	0.4	0.45	0.5	0.55
Osmotic pressure (atm)	1.3	2.6	4.0	5.3	6.7	8.1	9.6	11.0	12.6	14.3	16.0

- (ii) Draw a graph to show the changes in weight (%) of the potato strips against the molarities of the sucrose solutions.
7. From the graphs, determine
    - (i) The osmotic concentration of the potato tissue in isotonic sucrose solution.
    - (ii) The osmotic pressure in atmosphere unit (atm) for isotonic sucrose solution.





## B. Haemolysis

### Apparatus

6 test tubes, pipette 10 ml, glass rod, compound microscope, slides, cover slips, sterilized lancet, cotton, dropper

### Materials

40 ml of 1.0% NaCl solution, distilled water, ethanol, oil immersion.

### Precaution

- i) Wear gloves and do not use the same lancet twice.
- ii) Dispose all used gloves and lancets.
- iii) Avoid touching the test tubes containing the blood sample without gloves.

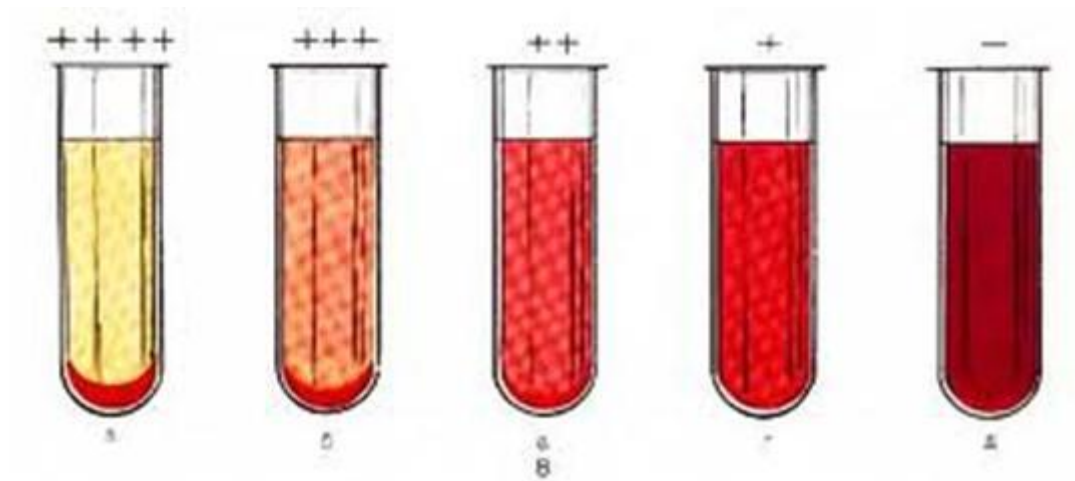
### Procedures and Observation

1. Label 6 test tubes from A to F.
2. Prepare NaCl solution of different concentration from the stock solution of 1 % NaCl as shown in the table below.

	A	B	C	D	E	F
Concentration of NaCl (%)	1	0.8	0.6	0.4	0.2	0
Volume of 1 % NaCl (mL)	5	4	3	2	1	0
Volume of distilled water (mL)	0	1	2	3	4	5
<b>Total volume (ml)</b>	<b>5</b>	<b>5</b>	<b>5</b>	<b>5</b>	<b>5</b>	<b>5</b>

3. Clean your hands using ethanol. Use a sterilized lancet to prick one of the fingers. Dispose the lancet.
4. Add 2 drops of blood into each test tube.
5. Invert each tube slowly and leave for 5 minutes at room temperature,
6. Examine the colour of the solution in each tube.
7. Transfer a drop of the solution from each tube onto a slide.
8. Place a cover slip onto the slide and examine under 100x magnification.

9. Record your observation in the table.



**Results**

Test tubes	Concentration of NaCl (%)	Concentration of solution (clear/cloudy)	Appearance of erythrocyte
A			
B			
C			
D			
E			
F			

**Questions**

1. (a) Does the erythrocyte become haemolysed if the solution is still clear?  
 (b) How do you explain this situation?
2. (a) What is the concentration of NaCl for haemolysis to occur?  
 (b) Explain how do you get your answer in 2(a).
3. Erythrocytes can show various forms of haemolysis in a hypotonic solution. Give evidence to support the above statement by comparing your result with your friend's result.

## **EXPERIMENT 4 : INHERITANCE**

### **Learning Outcome**

At the end of this practical, the students are able to:

1. Define terminologies used in genetic inheritance.
2. Characterize inherited human genetic.
3. Differentiate between phenotype and genotype definition.
4. Explain the ABO blood group inheritance.

### **Objectives**

1. To observe inheritance of certain traits/characteristics among students in the class.
2. To learn on how to do ABO blood group test and its principle.

### **Hypothesis**

Experiment A:

The inheritance characteristics found in human will determine the traits whether it is dominant or recessive alleles.

Experiment B:

Both A and B alleles are dominant over O alleles.

### **Introduction**

Physical traits are observable characteristics determined by specific segments of DNA called genes. Multiple genes are grouped together to form chromosomes, which reside in the nucleus of the cell. Every cell (except eggs and sperm) in an individual's body contains two copies of each gene. This is due to the fact that both mother and father contribute a copy at the time of conception. This original genetic material is copied each time a cell divides so that all cells contain the same DNA. Genes store the information needed for the cell to assemble proteins, which eventually yield specific physical traits.

Most genes have two or more variations, called alleles. For example, the gene for hairline shape has two alleles – widow's peak or straight. An individual may inherit two identical or two different alleles from their parents. When two different alleles are present they interact in specific ways. For the traits included in this activity, the alleles interact in what is called a dominant or a recessive manner. The traits due to dominant alleles are always observed, even when a recessive allele is present. Traits due to recessive alleles are only observed when two recessive alleles are present.

A number of human characteristics are determined by single gene. These characteristics include the shape of nose, earlobe, the ability of tongue rolling, the presence of dimple, left-handed, pigmentation of iris and the ability to taste Phenylthiocarbamide (PTC). A single gene also determines disease such as juvenile retinoblastoma and Huntington's chorea.

### **A. Inheritance characteristics found in human**

#### **Procedures and Observation**

1. Below are some inherited characteristics found in human:

- (i) Shape of nose

- (ii) Straight nose (E\_) is dominant to curved nose (ee)  
Earlobe  
Free earlobe (P\_) is dominant to attached earlobe (pp)
- (iii) Tongue rolling  
Ability of tongue rolling into "U" shape (C\_) is dominant to inability of tongue rolling into "U" shape (cc)
- (iv) Dimple  
Individual with dimple are genotypically (D\_) dominant compared to those without dimple (dd).
- (v) Left-handed  
The right-handed characteristic (H\_) is dominant to left-handed (hh).
- (vi) Hitch hiker thumb  
The ability to bend thumb at 60° angle or more are genotypically (tt) recessive compared to normal thumb-bending (T\_).

2. Based on the above characteristics,

- (i) Determine your genotypes.
- (ii) Calculate the percentage of each genotype for the class.
- (iii) Mark your genotype by circling it in Table 1 in the observation section, then determine your genotype number. Calculate the class genotype frequencies.

**Results :**

Table 1.

Genotype					No. of genotype	Frequency of genotype	Percentage of genotype
E_	P	C_	D_	H_	T_	1	
				tt	2		
			hh	T_	3		
			tt	4			
		dd	H_	T_	5		
			tt	6			
			hh	T_	7		
			tt	8			
	cc	D_	H_	T_	9		
			tt	10			
			hh	T_	11		
			tt	12			
		dd	H_	T_	13		
			tt	14			
			hh	T_	15		
			tt	16			

ee	pp	C_	D_	H_	T_	17			
					tt	18			
				hh	T_	19			
				tt	20				
			dd	H_	T_	21			
					tt	22			
			hh	T_	23				
				tt	24				
		cc	D_	H_	T_	25			
					tt	26			
					hh	T_	27		
					tt	28			
	dd			H_	T_	29			
					tt	30			
		hh	T_	31					
			tt	32					
	P_	C_	D_	H_	T_	33			
					tt	34			
					hh	T_	35		
						tt	36		
				dd	H_	T_	37		
						tt	38		
				hh	T_	39			
					tt	40			
			cc	D_	H_	T_	41		
						tt	42		
						hh	T_	43	
						tt	44		
		dd			H_	T_	45		
						tt	46		
			hh	T_	47				
				tt	48				
		pp	C_	D_	H_	T_	49		
						tt	50		
						hh	T_	51	
					tt		52		
				dd	H_	T_	53		
						tt	54		
				hh	T_	55			
					tt	56			
	cc		D_	H_	T_	57			
					tt	58			
				hh	T_	59			
				tt	60				

				H <sub>-</sub>	T <sub>-</sub>	61		
			dd		tt	62		
				hh	T <sub>-</sub>	63		
					tt	64		

### Questions:

1. A student inherited a recessive phenotype of a particular characteristic. However, both of the parents are phenotypically dominant for the characteristics. Explain.
2. Individuals with certain heterozygous characteristics are usually called a carrier. What does a carrier mean?
3. What factor determines whether a particular phenotype occurs more frequent than others in certain population?

### B. ABO Blood Group Inheritance

ABO blood groups in human are examples of multiple alleles of a single gene and also codominant alleles. Type A groups are determined by the presence of antigen A found on the surface of red blood cells (erythrocytes), while the blood plasma contains B antibody which agglutinates type B blood. Individuals with type B blood have antigen B and antibody A which agglutinates type A blood. Individuals with type AB blood have both antigen A and antigen B but without antibodies A or B. Finally, individuals with type O blood have antibody A and antibody B but without any antigen.

Table 2 shows individual characteristics for all ABO blood groups.

Blood Group (phenotypes)	Antigen present on erythrocytes	Antibodies present in blood plasma (serum)	Agglutinated blood group
A	A	Anti-B	B
B	B	Anti-A	A
AB	A and B	none	None
O	None	Anti-A and Anti-B	A and B

### Apparatus

Microscope slides, blood lancet (sterilized), blood group test kit, sterile latex surgical gloves.

### Materials

Anti-A and Anti-B serum and Anti-D, tooth picks, alcohol 100%, blood test kit.

### Procedures and Observation

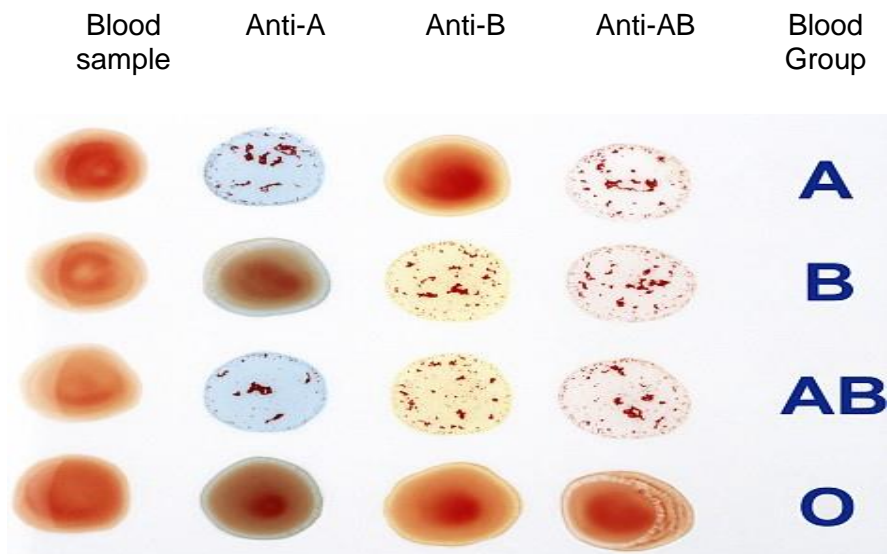
1. Label two clean and dry slides (no 1 and 2).
2. Wash your hands with soap and let them dry.
3. Swing your hand for 10 – 15 seconds.  
(Caution: do not use the same lancet twice or exposed lancet)
4. Apply alcohol to your middle finger. Prick the tip of the middle finger using sterilized lancet.
5. Wipe off the first blood drop.

6. Place the next drop at the centre of slide 1 and 2.
7. Drop an Anti-A serum near the blood on slide 1 and Anti-B serum on slide 2.
8. Mix the blood and serum on slide 1 with toothpick. Use another toothpick for slide 2.
9. You belong to A blood group if agglutination occurs on slide 1 only, B blood group if agglutination is observed on slide 2 only; AB blood group if agglutination occurs on both slides 1 and 2; O blood group if no agglutination is seen on both slides.
10. Calculate the percentage of each blood group in your class. Record your observation in Table 3.

Your blood group:

Table 3: Number of individuals and blood group percentage in the class

Blood Group	Possible Genotypes	Number of individuals	%
A			
B			
AB			
O			



**Questions:**

1. Why do you swing your hand for 10 -15 seconds before pricking the tip of your middle finger?
2. Why can't you use the same lancet twice?
3. Why do you need to wipe off the first blood drop?
4. Why do you need different toothpicks to mix the blood and serum on slide 1 and 2?
5. Can an individual with O blood group donate his blood to an A blood group person?
6. A mother with O blood group gave birth to a baby girl having the same blood group. However, she is not convinced that the baby belongs to her because her husband has AB blood group. She claimed there might be swapping of babies in the nursery. Explain.

## **EXPERIMENT 5 : POLLEN GERMINATION**

### **Learning Outcome**

At the end of this practical, the students are expected to be able to:

1. Describe how the germinating pollen grains occur.
2. Classify the germinated and ungerminated pollens.
3. Describe the general structures and formation of male gametes during pollen germination.

### **Objectives**

1. To observe germinating pollen grains under the microscope.
2. To differentiate germinated and ungerminated pollens.

### **Hypothesis**

Pollen germination will lead to the double fertilization of the ovule.

### **Introduction**

The growth of a pollen tube is a fascinating phenomenon. Pollen grains are morphologically simple, small structures which contain either two nuclei when released from the anther at anthesis. When a viable pollen grain lands on the stigma of a compatible flower, it produces a tube several hundred to several thousand micrometers long in which the pollen nuclei travel to the ovary of the flower. The process of tube formation is a relatively uncomplicated example of growth and development. Pollen germination represents a short, yet very critical event in a series of steps leading to the double fertilization of the ovule.

### **Apparatus**

Compound microscope, glass slides and cover slips, glass rod, dropper, petry dish, graduated measuring cylinder, needles, labelling stickers

### **Materials**

Fresh flower Lily/ Hibiscus, 0.01 g boric acid, 100 mL distilled water, 0.03 g magnesium sulphate, 0.02 g potassium nitrate, 10 g sucrose

### **Procedures and Observation**

1. Prepare the pollen germination medium by dissolving 10 g sucrose, 0.01 g boric acid, 0.03 g magnesium sulphate and 0.02 g potassium nitrate in 100 mL of distilled water.
2. Using a glass rod, stir the solution to mix it well.
3. Using a dropper, take some nutrient solution and put two drops on a clean glass slide.
4. Take a mature flower and dust a few pollen grains from its stamen on to the drop on the slide.
5. After 5 minutes, place the glass slide on the stage of the compound microscope.
6. Observe the slide through the microscope regularly for about half an hour.
7. Observe the pollen grains under a compound microscope using an appropriate magnification power.
8. Draw and label the germinated pollen grains.





### Questions

1. Which of the substance: sucrose or water causes the formation of pollen tubes?
2. What is the purpose of using distilled water?
3. What is the function of sucrose and potassium nitrate in the pollen grain germinating solution?