WJEC Wales Biology A Level

SP 1.3c: Investigation into the permeability of cell membranes using beetroot

Practical notes

Introduction

Cell membranes are described as '**fluid**' because **phospholipid** molecules can move within or between monolayers. **Temperature** affects the fluidity and resulting permeability of cell membranes. This can be observed using beetroot cells.

Beetroot cells contain **belatin**, a **red-purple** pigment within vacuoles. As cell membranes become more permeable, belatin leaks out and dyes the surrounding solution. A **colorimeter** is used to measure the **% absorbance** or **% transmission** of light through the solution which provides an indication of membrane permeability.

Equipment

- Beetroot cylinders
- Distilled water
- 6× boiling tubes
- 6× bungs
- Boiling tube rack
- 5 cm^3 syringe
- Scalpel
- Ruler
- Forceps
- Stopwatch
- Water baths (20, 30, 40, 50, 60 and 70°C)
- Colorimeter (with green filter)
- Cuvettes

Risk assessment

Hazard	Risk	Precaution	Emergency
Broken glass	Cuts	Keep glassware away from the edge of the desk	Dispose of broken glassware carefully; elevate cuts and apply pressure; do not remove glass from cuts; seek medical assistance
Scalpel	Cuts	Direction of cut away from the body; do not attempt to change blade; keep scalpel away from the edge of the desk	Elevate cuts and apply pressure; wash minor cuts in cold water; seek medical assistance

Boiling water	Scalding	Handle boiling water with care; use tongs to transfer boiling tubes; wear safety goggles	Run burn under cold water; seek medical assistance

Method

1. Using the scalpel and ruler, prepare six beetroot cylinders **50 mm** long
2. Gently rinse each cylinder with distilled water to wash away any pigment leakage
3. Dry each beetroot cylinder by rolling it over a paper towel **three times**
4. Prepare six boiling tubes with **5 cm^3 distilled water**
5. Label each boiling tube with a temperature (20, 30, 40, 50, 60 and 70°C) and place in the corresponding **water bath** for **5 minutes**. *This allows time for the distilled water to equilibrate to the temperature of the water bath.*
6. Using **forceps**, place a beetroot cylinder in each boiling tube. *Add a bung to prevent the evaporation of water.* Set a **stopwatch** for **15 minutes**.
7. After 15 minutes, swirl the contents of the boiling tubes and remove each beetroot cylinder
8. Transfer the contents of each boiling tube to a **cuvette**
9. **Zero** the **colorimeter** (with a **green filter**) using **distilled water**
10. One at a time, place each cuvette into the colorimeter and measure the **% absorbance** of the solution. *Insert the cuvette so that the two clear walls align with the path of light.* Record the results (see below)
11. **Repeat** the experiment a further **two** times
12. Plot a **graph** of **mean % absorbance** against **temperature**

Variables

Independent variable

The variable that is **changed**
i.e. temperature

Dependent variable

The variable being **measured** whose value depends on the independent variable
i.e. % absorbance

Controlled variables

The variables that are kept **constant** during the experiment:

- Surface-area-to-volume ratio of the beetroot cylinder
  Controlled using a ruler to ensure the cylinders are the same length
- Length of time left in the water bath
  Controlled using a stopwatch to time 15 minutes
- Volume of water initially on the surface of the beetroot cylinders
  Controlled by rolling over a paper towel three full times
- Volume of water in each boiling tube
  5 cm^3 syringe used to measure 5 cm^3 of distilled water
- Same type and age of beetroot
  Cylinders should be from the same beetroot or the same type of beetroot

Results

Temperature (°C)	% absorbance			Mean % absorbance
	Repeat 1	Repeat 2	Repeat 3	
20				
30				
40				
50				
60				
70				

Conclusion

As **temperature increases**, the **% absorbance increases** and the **% transmission decreases**.

High temperatures increase the **fluidity** of cell membranes as phospholipid molecules have more **kinetic energy**. Proteins within the bilayer are also **denatured** at high temperatures. Cell membranes become **more permeable** and **belatin leaks out**, dyeing the surrounding solution. More light is absorbed by the solution and less light transmitted through it.