AQA Biology A-Level

Required Practical 4

Investigation into the effect of a named variable on the permeability of cell-surface membranes.
Cell-surface membranes are made up of a phospholipid bilayer which makes them selectively permeable. This permeability can be changed by different variables, such as temperature and concentration of solvents, like ethanol.

The permeability of a membrane can be measured by using beetroot cells, which contain a purple pigment called betalain. When the cell-surface membrane has a higher permeability, more pigment leaks out of cells. The permeability can therefore be measured by the amount of pigment leaked from beetroot cells into an aqueous solution using a colorimeter.

**Equipment list**

- Beetroot
- Scalpel
- Forceps
- Cutting board
- Ruler
- Tongs
- Distilled water
- Boiling tubes
- Boiling tube rack
- Colorimeter
- Cuvettes
- Filter paper
- Timer
- Water bath
- Thermometer
- Ethanol

**Method**

1. Cut beetroot into 6-10 identical cubes using a scalpel.
2. Wipe/rinse to clean off any pigment released as a result.
3. If investigating temperature: place each of the cubes of beetroot in an equal volume of distilled water (5-15ml).
4. Place each test tube in a water bath at a range of temperatures (30-80°C).
5. **If investigating concentration of solvents:** create a dilution series of ethanol using distilled water. Ethanol concentrations should range from 0-100% ethanol.

6. Leave the samples for 20 minutes - the pigment will leak out of the beetroot.

7. Set the colorimeter to a blue filter and zero using a cuvette with distilled water.

8. **Filter** each sample into a cuvette using filter paper.

9. Measure the **absorbance** for each solution. A higher absorbance indicates higher pigment concentration, and hence a more permeable membrane.

### Risk Assessment

<table>
<thead>
<tr>
<th>Hazard</th>
<th>Risk</th>
<th>Safety Precaution</th>
<th>In emergency</th>
<th>Risk Level</th>
</tr>
</thead>
<tbody>
<tr>
<td>Scalpel</td>
<td>Cuts from sharp object</td>
<td>Cut away from fingers; use forceps to hold sample whilst cutting, keep scalpel away from the edge of the desk</td>
<td>Elevate cuts; apply pressure; seek medical assistance</td>
<td>Low</td>
</tr>
<tr>
<td>Broken glass</td>
<td>Cuts from sharp object</td>
<td>Take care when handling glassware; keep away from edge of desk</td>
<td>Elevate cuts; apply pressure; do not remove glass from wound; seek medical assistance</td>
<td>Low</td>
</tr>
<tr>
<td>Ethanol</td>
<td>Irritant/flammable</td>
<td>Wear eye protection; keep away from naked flames</td>
<td>Wash eyes and skin with cold water</td>
<td>Low</td>
</tr>
<tr>
<td>Hot liquids</td>
<td>Scalding</td>
<td>Handle with care; use tongs to remove boiling tubes from water bath; wear eye protection, keep away from the edge of the desk</td>
<td>Run burn under cold water; seek medical assistance</td>
<td>Low</td>
</tr>
</tbody>
</table>
Graph
- Plot a graph of absorbance against ethanol concentration/temperature.

Conclusion

**Temperature**
- As the temperature increases, the permeability of the cell-surface membrane also increases. This is because the proteins in the membrane denature as the heat damages the bonds in their tertiary structure. This creates gaps in the membrane, so it is easier for molecules to pass through it.
- At low temperatures, phospholipids have little energy and are packed closely together to make the membrane rigid. This causes a decrease in permeability and restricts molecules from crossing the membrane.

NB: At very low temperatures, ice crystals can form which pierce the cell membrane and increase the permeability.

**Concentration of ethanol**
- Ethanol causes the cell-surface membrane to rupture, releasing the betalain pigment from the cell. Higher concentrations of ethanol will cause more disruption to the membrane and more gaps will form. Thus, as concentration of ethanol increases, the permeability of the cell-surface membrane also increases.