**Exploring Cell Membrane Structure using Beetroot**

**Aim:** To investigate the chemical nature of the cell membrane using beetroot.

**Background**

Cell membranes are composed of a phospholipid bilayer, with integral, transmembrane and peripheral proteins associated with it. The cell membrane is selectively permeable. This permeability can be altered by disrupting the membrane components: phospholipids are disrupted by alcohol; proteins are disrupted by high temperatures.

Beetroot cells contain a red pigment, called betalain, within their vacuoles. Disruption of cell membranes increases cell permeability, leading to loss of betalain into the surrounding medium. The greater the disruption to cell membranes, the more intensely red the surrounding medium will become. This colour intensity can be monitored using colorimetry and can be used to estimate the relative proportions of phospholipids and proteins in the cell membrane.

**Materials**

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| Beetroot | 4 boiling tubes + rack |
| Cork borer | Water bath at 60°C |
| Cutting board / white tile | 20% ethanol |
| Knife / scalpel | Distilled water |
| Tongs / forceps | Measuring cylinder |
| Ruler | Colorimeter |
| Large beaker of distilled water | Cuvettes |
| Blue roll | Stopwatch  |
| 3 cm3 plastic pipettes |  |

**Method**

1. Use a cork borer to extract 3 bores of beetroot of equal diameter. Use the knife and ruler to trim the bores to the same length.
2. The process of cutting the beetroot will disrupt cells and cause pigment to seep. This pigment must be removed. Rinse the beetroot bores in water, blot dry with blue roll and place in a beaker of water for 2 hours.
3. After 2 hours, rinse the beetroot again, blot dry, and replace the water in the beaker.
4. Continue to steep the beetroot bores overnight, replacing the water in the beaker several times.
5. After 24 hours, rinse the beetroot bores once more. The water should run quite clear. Blot the bores dry.
6. Set up the test tubes as outlined in the table. Allow liquid contents to reach the specified temperature prior to adding the beetroot bore.

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| **Test tube** | **Temperature (°C)** | **Volume of distilled water (cm3)** | **Volume of 20% ethanol (cm3)** | **Number of bores to add** |
| 1 | 20 | 20 | - | 0 |
| 2 | 20 | 20 | - | 1 |
| 3 | 60 | 20 | - | 1 |
| 4 | 20 | - | 20 | 1 |

1. Allow the bores to incubate for 30 minutes.
2. Remove the bores from the incubating medium and discard. Retain the incubating medium.
3. Set the colorimeter to a blue filter (~465nm) and zero using the liquid in test tube 1. Add approximately 3cm3 of the liquid to a cuvette, using a plastic pipette.
4. Measure the absorbance of each of the remaining liquids and record on the table below.

The higher the absorbance, the higher the pigment concentration resulting from a more permeable membrane. This would reflect a greater level of membrane disruption due to the conditions applied.

**Adaptations of the practical**

* Investigating the loss of betalain from beetroot cells over a range of temperatures
* Investigating the loss of betalain from beetroot cells over a range of ethanol concentrations (consider adaptations to the suggested risk assessment in this case).
* Red cabbage could also be used as an alternative to beetroot. Generally more affordable but more challenging to create “bores” of equal diameter and length.

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| **Condition** | **Absorbance** |
| 20**°**C, water |  |
| 60**°**C, water |  |
| 20**°**C, ethanol |  |