Experimental question

How does the concentration of ethanol affect the membrane permeability of beetroot?

Design

The cells of beetroot have red pigment in the vacuoles. When the membranes of the vacuole and the cell membrane are damaged by ethanol, a kind of alcohol, pigment will leak out. With this information, design an experiment to answer the experimental question.

1. In this experiment, we are investigating how a factor (**independent variable, IV**) affects another factor (**dependent variable, DV**). What are the DV and IV of this experiment?

IV is the concentration of ethanol. DV is the membrane permeability. (#1)

Explain how the membrane permeability of the beetroot cells can be measured based on the above information. Suggest an accurate and reliable method for the measurement.

The membrane permeability can be measured by the amount of red pigment leaked out of the vacuoles. The amount of red pigment can be estimated from the intensity of red colour as judged by eyes or more accurately by colorimeter. (#11)

3. What do you predict the results when beetroot tissues are put into different concentrations of ethanol?

The higher the concentration of the ethanol, the more the membrane is damaged. More red pigment will leak out from the vacuoles, producing darker red colour at higher ethanol concentrations. (#3)

4. Will you (1) put the **same** beetroot into different concentrations of ethanol **one after one**, or, (2) put **different** beetroot into different concentrations of ethanol? Discuss the strengths and drawbacks of each design.

Design (1) has the problem that the membrane of beetroot has been damaged by the previous treatment of the ethanol and some red pigment has leaked out. It thus cannot show the effect of a concentration of ethanol. Design (2) avoids this problem, but the beetroot put into different concentrations of ethanol may be different, e.g. cells containing different amount of red pigment. It makes the comparison between different treatments unfair. (#28)

5. Your teacher stresses that the beetroot has to be cut into same size and shape in each concentration of ethanol. Explain why it is needed.

The shape and size of beetroot affect its surface area in contact with the ethanol. The one with larger surface area will have faster leakage of pigment. It makes the comparison between different treatments unfair. (#22)

6. One student proposes putting a 3 cm cylinder of beetroot into each concentration of ethanol. Another student thinks the cylinder should be cut into three 1-cm discs to be put into each concentration of ethanol. Which one do you think is better? Explain why.

It is better to cut the beetroot into smaller discs. It increases the surface area and speeds up the leakage of pigment. This would shorten the time needed for the experiment and produce more obvious results for comparison between the treatments. (#17)

7. Apart from ethanol and the shape and size of beetroot, are there other factors that may affect the leakage of pigment from beetroot cells? Explain your answers. How can these factors be controlled?

The longer the beetroot is immersed in the ethanol, the more the membrane is damaged and the more pigment is leaked out. It can be controlled by keeping the time of immersion the same for each treatment. (#22)

There may be varying amounts of red pigment in the vacuoles of different beetroot cells. The cells with higher pigment concentration will leak out more pigment. This variation can be reduced by using the same beetroot, and by **randomly** putting the beetroot discs into different concentrations of ethanol. (#22)

8. Do you think the **volume** of ethanol (not the concentration) bathing the beetroot needs to be kept the same for each treatment? Explain your answer.

The **volume** of ethanol needs to be kept the same for each treatment because it will affect the colour intensity observed. A larger volume, however, would **not** make greater damage to the membrane and more leakage of pigment. (#22)

9. What is the **major assumption** underlying the whole experimental design? (An assumption is something we think it is true, though we cannot be sure. A major assumption is the one that the experiment cannot make any conclusion without assuming it to be true).

Membrane permeability of beetroot is proportional to the amount of pigment leaked out from vacuoles. (#27)

Procedure to be handed out to students after completing the design.

Materials

Item Amount 1. Ethanol (15%, 30%, 50%) each 1 tube 2. Beetroot 1 pc 3. Razor blade 1 pc 4. Plastic chopping board 1 pc 5. Test-tube 4 pcs 6. Test-tube rack 1 pc 7. Stopper 4 pcs 8. Measuring cylinder (10 ml) 1 pc 9. Labels 4 pcs 10. Forceps 1 pc 11. Cork borer 1 pc 12. Beaker (250 ml) 1 pc 13. White paper 1 pc On side bench: (for the whole class) 13. Distilled water (in wash bottle)

Procedure

(Write down the steps necessary for performing the experiment. All quantities, e.g. volume of liquid used, amount of materials, time period for treatment, time taken in conducting the experiment, etc., are required to state clearly.)

- 1. Transfer 10 ml of distilled water, 15%, 30% and 50% of ethanol into each of 4 test tubes respectively. Label the tubes.
- 2. Using the same beetroot, prepare 4 cylinders of beetroot, each of 2 cm long, using cork borer and razor. Cut the cylinder of beetroot into four discs of the same thickness.
- 3. Rinse the cylinders of beetroot in running tap water until no pigment comes out from the damaged cells.
- 4. Randomly put 4 discs of beetroot into each of the test tubes prepared in step 1. Stopper the tubes.
- 5. Leave the tubes for 20 minutes. For every 5 minutes, shake the tube gently for a few seconds.
- 6. Take the cylinders of beetroot out from the test tubes using a pair of forceps.
- 7. Place the test tubes in front of a white paper. Record and compare the intensity of red colour of the solutions in the tubes with naked eyes.