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?

   Basic 1: Identify the DV and IV

   IV is the concentration of ethanol. DV is the membrane permeability to the red pigment.

2. 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.

   Good 11. Explain how variables are connected with the manipulation and measurements

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

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

   Basic 3. Predict the results

   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.

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.

   Excellent 28. Discuss the limitations and strengths of the alternative designs (e.g. within subject and between subject 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 particular concentration of ethanol.

   Design (2) avoids the above problem. But the beetroot tissues put into different concentrations of ethanol are not the same - their cells may contain different amounts of red pigment or they have different shape and size. It makes the comparison between different treatments unfair.

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.

   Excellent 22. Explain why some important CVs need to be controlled

   The shape and size of beetroot affect its surface area in contact with the ethanol. A larger surface area will speed up the action of ethanol on the membrane, making the comparison between different treatments unfair.

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.

   Good 17. Explain why a specific step is conducted.

   It is better to cut the beetroot into smaller discs. It increases the surface area and makes the actions of the ethanol on the beetroot faster. This would shorten the time needed for the experiment and produce more obvious results for comparison between the treatments. (25)

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?

   Good 12. Identify important CVs
   Excellent 22. Explain why some important CVs need to be controlled

   The time that the beetroot is immersed in the ethanol. The longer the immersion time, more membrane is damaged and thus more pigment is leaked out.

   The temperature of the ethanol. A higher temperature will make the red pigment diffuse faster and enhance the actions of the ethanol on the membrane.

   The source of the beetroot. Different beetroot or the beetroot tissues from different locations of a beetroot may have varying amounts of red pigment in the vacuoles. This variation can be reduced by obtaining beetroot dices from the same beetroot and **randomly** put several beetroot discs into each concentration of ethanol.

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.

   Excellent 22. Explain why some important CVs need to be controlled
   Good 17. Explain why a specific step is conducted.

   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** affect the action of ethanol on the membrane.

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).

   Excellent 27. Identify the major, significant assumptions of the design

   The red pigment observed in the ethanol is all leaked from the vacuoles of the beetroot after their membrane is damaged by ethanol.

   The membrane permeability of beetroot is proportional to the amount of pigment leaked out from vacuoles.

10. Your teacher suggest you to replicate the experiment if time is allowed. Explain the underlying reason(s).

    Excellent 24. Explain why some procedures can reduce measurement errors (e.g. repeated measurement for reducing random errors; calibration for reducing systematic errors; involving one person to observe the results to reduce subjectivity)

    Replication of the experiment means that the whole experiments are repeated for several times. As such, the result for each concentration of ethanol come from the average of several setups. It will reduce the errors resulting from the variations between treatments e.g. beetroot size and shape, source of beetroot, volume and temperature of ethanol, time of immersion, etc.

Item	competence	Basic/good/excellent
1	1	B
2	11	G
3	3	B
4	28	E
5	22	E
6	17	G
7	22	E
8	22	E
9	27	E
10	24	E

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.