**EFFECT OF ALCOHOL ON CELL MEMBRANES**

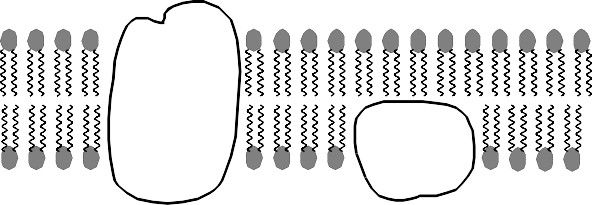
## LAB CELL 1

**INTRODUCTION**

A eukaryotic cell, a cell with a nucleus, not only has a plasma membrane as its external boundary, but it also has a variety of membranes that divide the internal space into discrete compartments, separating processes and cell components. The complex and varied design of cell membranes gives them the remarkable properties that allow them to serve the variety of specific functions required by different types of cells. One of the most significant properties of membranes is selective permeability. This permits some molecules and ions to pass freely through the membrane, but excludes others from doing so. Understanding the structure and properties of cell membranes is important to understand how cells function.

Cell Membrane

Hydrophillic (water-soluble) Exterior



Lipid Bilayer

Protein

Protein

Hydrophobic (oily) Interior

Although the details of cell membrane composition and structure vary according to its specific function, all of them are composed of mainly lipids and proteins in a structure known as a lipid bilayer. This is a fluid structure that allows for the flexibility required for cell growth and movement in addition to the selective permeability that is so central to membrane function.

One component of plant cells that usually contains water is the *vacuole*, which is surrounded by a membrane known as the *tonoplast*. In beet plants, the vacuole also contains a water-soluble red pigment, *betacyanin*, which gives the beet its characteristic color. If the tonoplast is damaged, however, the contents of the vacuole will spill out into the surrounding environment. In the case of beets, when the membrane is damaged, the

## References:

*Biology with Calculators*, Vernier Software & Technology, 2000.

Kirby, J., Mortellaro, J., Prockup, J. “Effects of Temperature and Solvents on the Cell Membrane, SOTM LAB B14.” <http://library.marist.edu/sotm/html/b14.html> (29 September 2002).

red pigment will leak out into the surrounding environment. The intensity of color in the environment should be proportional to the amount of cellular damage sustained by the beet.

This experiment allows you to test the effect of three different alcohols (methanol, ethanol, and 1-propanol) on cell membranes. Ethanol is found in alcoholic beverages. Methanol, sometimes referred to as wood alcohol, can cause blindness and death.

Propanol is fatal if consumed. One possible reason why alcohols are so dangerous to living organisms is that they might damage cellular membranes, therefore resulting in the death of the entire cell.

Methanol, ethanol, and 1-propanol are very similar alcohols, differing only by the number of carbon and hydrogen atoms within the molecule. Methanol, CH3OH, is the smallest; ethanol, CH3CH2OH, is intermediate in size; and 1-propanol, CH3CH2CH2OH, is the largest of the three molecules.

The alcohol solutions used in this experiment are clear and colorless. If the beet pigment leaks into the solution, it will turn the solution red. The intensity of color in the solution should be proportional to the amount of cellular damage sustained by the beet. As the concentration of pigment in the solution increases, it absorbs more light. In this experiment, the absorbance of light at 460 nm will be used to monitor the extent of cell membrane damage.

# PURPOSE

The purpose of this experiment is to study the effect of different alcohols at various concentrations on the integrity of cell membranes by visible spectroscopy.

# EQUIPMENT/MATERIALS

|  |  |
| --- | --- |
| 5 test tubes | marker |
| test tube rack | 10 mL graduated cylinders |
| cuvettes | Kimwipes |
| Spectronic 20 Genesys | cork borer with 4-mm inside diameter |
| Beets (canned or fresh) | wash bottle with deionized water |
| ruler | gloves |
| scalpel or razor blade | apron |
| small dowel rods | |

***FOR ENTIRE CLASS:***

Alcohol solutions (5 mL/group): 0%, 10%, 20%, 30%, 40% methanol

0%, 10%, 20%, 30%, 40% ethanol

0%, 10%, 20%, 30%, 40% 1-propanol

# SAFETY

* Always wear an apron and goggles in the lab.
* Beets will stain hands and clothing. Gloves should be worn, in addition to aprons, to prevent staining.

# PROCEDURE

1. Cut 5 uniform cylinders of beet using a cork borer with a 4-mm inside diameter. Line up the pieces, cut off the ends and make another cut to obtain 15-mm long pieces (all pieces must be the same size).
2. Rinse each piece for 2 minutes in a large beaker of tap water to remove the excess red dye that leaked during the cutting procedure.
3. Number each of your five test tubes 1 – 5.
4. Using the table below as a guide, pour 5 mL of alcohol solution into each of the test tubes. Your teacher will tell you which alcohol your group will be testing (methanol, ethanol, or 1-propanol). Record the type of alcohol tested on your data sheet.

|  |  |
| --- | --- |
| **Test Tube Number** | **Alcohol %** |
| 1 | 0 |
| 2 | 10 |
| 3 | 20 |
| 4 | 30 |
| 5 | 40 |

1. Add one piece of beet to each of the 5 test tubes and begin timing for 10 minutes. During the 10 minutes, use the procedure shown by your instructor to gently mix the contents of each of your six test tubes *every minute*. **Note:** During this time your teacher may also have you zero the Spec 20. See steps 7 - 12.
2. When the 10 minutes is up, carefully pour the solution from each of the five test tubes into five different cuvettes until each is about ¾ full. **Be carefull to keep track of which test tube was used to fill each of the cuvettes!**
3. Ensure that the spectrophotometer has warmed up for at least 20 minutes. Use the same instrument for the entire experiment.
4. Set the spectrophotometer to 460 nm.
5. Fill a cuvette about 3/4 full with deionized water. This is the "blank cuvette".
6. Place the blank cuvette into the spectrophotometer with the triangle on the cuvette facing the front of the instrument.

**Note:** Before inserting a cuvette into the spectrophotometer, wipe it clean and dry with a kimwipe, and make sure that the solution is free of bubbles. Do not touch the clear sides of the cuvette.

1. Press 0 ABS 100%T.
2. Remove the blank cuvette from the instrument.
3. Place the cuvette containing the solution from test tube “1” into the spectrophotometer. Make sure that the triangle on the cuvette is facing the front of the instrument. **Do not press 0 ABS 100%T.**
4. Record the absorbance of the solution from test tube “1” in the Data Table.
5. Repeat steps 13 and 14, for the solutions from test tubes 2 – 5.

# PROCESSING THE DATA

Using a spreadsheet or graph paper, make a graph of absorbance vs. alcohol concentration. Absorbance should be plotted on the y-axis, and alcohol concentration (%) on the x-axis.

**DATA SHEET** Name

Name Period Class Date

# EFFECTS OF ALCOHOL ON CELL MEMBRANES

**AlcoholsTested**

**DATA TABLE**

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| **Test Tube** **Number** | **% Alcohol** |  | **Absorbance** |  |
| **Methanol** | **Ethanol** | **1-Propanol** |
| **1** | **0** |  |  |  |
| **2** | **10** |  |  |  |
| **3** | **20** |  |  |  |
| **4** | **30** |  |  |  |
| **5** | **40** |  |  |  |

**QUESTIONS**

1. What trend do you see in cellular damage based on the data from the single alcohol you tested?
2. What trend do you see in cellular damage based on the data from all three alcohols? What factors may be responsible for this?
3. Why were significantly higher concentrations of alcohol not tested?