The Effect of Alcohol on Biological Membranes

The primary objective of this experiment is to determine the stress that various alcohols have on biological membranes. Membranes within cells are composed mainly of lipids and proteins and often serve to help maintain order within a cell by containing cellular materials. Different membranes have a variety of specific functions.

One type of membrane-bound vacuole found in plant cells, the tonoplast, is quite large and usually contains water. In beet plants, this membrane-bound vacuole also contains a watersoluble red pigment, betacyanin, which gives the beet its characteristic color. Since the pigment is water soluble and not lipid soluble, it remains in the vacuole when the cells are healthy. If the integrity of a membrane is disrupted, however, the contents of the vacuole will spill out into the surrounding environment. This usually means the cell is dead.

In this experiment, you will test the effect of three different alcohols (methanol, ethanol, and 1-propanol) on 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 they are so dangerous to living organisms is that they damage cellular membranes. Methanol, ethanol, and 1-propanol are very similar alcohols, differing by the number of carbon and hydrogen atoms within the hydrophobic part of the molecule. Methanol, CH₃OH, is the smallest, ethanol, CH₃CH₂OH, is intermediate in size, and 1-propanol, CH₃CH₂CH₂OH, is the largest of the three molecules. It is the hydrophobic part of an amphipatic molecule that interact with the phospholipids bilayer. In fact, phospholipids, which have long hydrocarbon chains attached to a smaller hydrophilic head group, are generally soluble in nonpolar solvents such as alcohols, so the relevant part of an alcohol molecule is the nonpolar hydrocarbon segment. In solution, the hydrophobic part of an alcohol will inevitably interact with the polar heads of the phospholipids. The bigger the hydrophobic part, the more damage it causes.

The alcohol solutions used in this experiment are clear and colorless. If beet membranes are damaged, the 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. As the concentration of pigment in the solution increases, it absorbs more light.

To measure the color intensity, you will be using a Colorimeter or Spectrometer. In this device, blue light from the LED light source will pass through the solution and strike a photocell. The alcohol solutions used in this experiment are clear. If the beet pigment leaks into the solution, it will color the solution red. A higher concentration of colored solution absorbs more light and transmits less light than a solution of lower concentration. The device monitors the light received by the photocell as either an absorbance or a percent transmittance value.

You are to prepare five solutions of differing alcohol concentrations (0%, 10%, 20%, 30%, and 40%) for each of the three alcohols. A small piece of beet is placed in each solution. After ten minutes, each alcohol solution is transferred to a cuvette that is placed into the Colorimeter or Spectrometer. The amount of light that penetrates the solution and strikes the photocell is used to compute the absorbance of each solution. The absorbance is directly related to the amount of red pigment in the solution. By plotting the percent alcohol vs. the amount of pigment (that is, the absorbance), you can assess the amount of damage various alcohols cause to cell membranes.

OBJECTIVES

In this experiment, you will

- Use a Colorimeter or Spectrometer to measure the color intensity of beet pigment in alcohol solutions.
- Test the effect of three different alcohols on membranes.
- Test the effect of different alcohol concentrations on membranes.