

OSMOTIC PRESSURE INFLUENCE ON THE CELL (ERYTHROCYTE) VOLUME

INTRODUCTION

Every cell of the body is bathed in a watery fluid that contains a mixture of molecules that are essential to its survival. This fluid may be the plasma of blood or the tissue fluid in the interstitial spaces. In either case, these molecules, whether water, nutrients, gases, or ions, pass in and out of the cell through the plasma membrane. Some molecules, usually of small size, are able to diffuse passively through the cell membrane from areas of high concentration to low concentration. Organic molecules, such as glucose and amino acids and certain ions move through the plasma membrane either with or against a concentration gradient by active transport, which requires energy expenditure by the cell. Movement of the molecules occurs if the membrane is viable and undisturbed. The integrity of plasma membranes throughout the body is due largely to the fact that body fluids are isotonic. In this experiment a study will be made of osmosis and the effects of hypertonic, isotonic and hypotonic solutions on the plasma membrane of red blood cells (RBC).

Water molecules because of their small size move freely through cell membranes into and out of the cell. Water acts as a vehicle which enables ions and other molecules to move through the membrane. This movement of water molecules through a semipermeable membrane, from the side where their concentration is higher to the side where the concentration is lower is called osmosis.

Although water molecules are always moving both ways through the membrane, the predominant direction of flow is determined by the concentration of solutes on each side of the membrane. The force that would be required to restrain the flow of water is called effective osmotic pressure.

Solutions that contain the same concentration of solutes are said to be isotonic solutions. RBC immersed in such solution gain and lose water molecules at the same rate, establishing an osmotic equilibrium. If a solution contains a higher concentration of solutes than is present in cells, water leaves the cells faster than it enters causing the cells to shrink and develop cupped (crenate) edges. This shrinkage is called plasmolysis or crenation. Such solutions of high solute concentration are called hypertonic solutions and are said to have a higher osmotic potential than isotonic solutions. A solution that has a lower solute concentration than is present in cells is said to be a hypotonic solution. In such solution water flows rapidly into the cells, causing them to swell and burst (lysis) as the plasma membrane disintegrates. Lysis of RBC is called osmotic hemolysis.

To observe the effect of different types of solution on RBC, these will be added to various concentrations of solutions (hypo, iso and hypertonic solutions). The effect of the solutions on the cells will be determined macroscopically and microscopically.

MATERIALS: test tubes, stand for test tubes, pipettes, microscope, NaCl solutions 3g‰, 9g‰, 15g‰, materials for venous puncture- cotton, syringe, needle, garrot, slides, cover glasses.

PROCEDURE: In three test tubes are introduced:

2 ml of NaCl - 3 g‰ (hypotonic solution)

2 ml of NaCl - 9 g‰ (isotonic solution)

2 ml of NaCl - 15 g‰ (hypertonic solution)

A drop of blood is introduced in every test tube and they are let in stand for 1 hour. After that the changes are noted macroscopically and microscopically. For microscopically examination put a drop of blood from every test tube on a slide. Cover it with a cover glass and examine under the microscope using a dry objective (20X or 40X).

INTERPRETATION:

In the first test tube (hypotonic sol.), the supernatant is colored in red because water enters the cells, which swell until they burst and the content (hemoglobin) is released into the solution. There is no deposit

of RBC at the bottom of test tubes as RBC were hemolysed- osmotic hemolysis. Under the microscope, RBCs appear larger with the outline round.

In the second test tube (isotonic solution) one can be noted that there is a deposit of blood at the bottom of test tube and the supernatant is clear (colorless). The size of RBC is not changed when they are examined under the microscope, the outline is round.

In the third test tube (hypertonic solution), the supernatant is also clear and there is a deposit of blood at the bottom of test tube. Under the microscope the RBCs appear smaller and the outline is crenated because the water leaves the cells.

Examine each slide under high-dry objective and record your results in your copy books.

HYPOOSMOTIC SOLUTION



**CELLS SWELL AND
EVENTUALLY BURST**



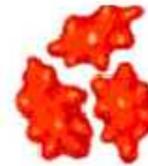
ISOSMOTIC SOLUTION



NORMAL CELLS



HYPEROSMOTIC SOLUTION



SHRIVELED CELLS

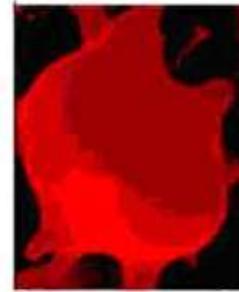


Figure 2