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OSMOSIS: A DIFFERENT APPROACH

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Introduction

Traditional experiments on osmosis usually include a study of the glucose-water experiment but lack a connection to a living system. This is a fine experiment but fails to give students a true picture of what is occurring in a living system.

The following experiments describe a method of examining osmosis and some factors that can have an effect on it in a living blood cell. Depending on the teacher's dedication, it may be their own blood, taken by a doctor, or blood from a cow or similar animal. The latter is easily obtained from a butcher or packing plant.

Part I
Osmosis and Hemolysis

From a 1% solution of NaC1 prepare the following saline solutions:

0.85%
0.70%
0.60%
0.50%
0.40%
0.30%
0.20%
0.10%
Final volume
to be five (5) ml.

Students can make their own solutions by dilution or the instructor may want to make these up ahead of time to conserve time and ease confusion. If the latter option is exercised, a volume suitable for the class should be made as each student will require five ml of each solution.

Place five ml of each solution in a small test tube (13 × 100 mm works best) and add one drop of blood to each. Quickly invert to mix and allow to stand for thirty minutes. At the end of this time centrifuge for two minutes in a low speed centrifuge (a clinical type works well). This should sediment any remaining intact erythrocytes. Determine the concentration at which initial and complete hemolysis occurs. Hemolysis occurs when the first perceptable pink color remains after centrifugation. To make this easier to detect a colorimeter could be used and

absorbance plotted against concentration. This adds a new dimension to the experiment but is not a necessity. Table 1 gives sample data for this part of the experiment.

Table 1
Hemolysis in Sodium Chloride

NaCl Concentration	Hemolysis
0.85%	*None*
0.70%	*None*
0.60%	*Initial*
0.50%	*Increasing hemolysis*
0.40%	*" "*
0.30%	*Complete hemolysis*
0.20%	*" "*
0.10%	*" "*

Part II
Hemolysis by a Penetrating Substance

This part of the experiment examines the ability of various substances to penetrate the cell membrane of a red blood cell. Five ml of the solutions shown in Table 2 are added to small test tubes. A drop of blood is then added to each tube and the tubes inverted to mix the blood and the solution. At the end of 15 minutes, centrifuge. The tubes should then be examined to determine the extent of hemolysis. The same criteria should be applied here as in Part I. Table 2 gives sample data for this part of the experiment.

Table 2
Effect of Penetrating Substances

Solution	Concentration	Hemolysis
NaCl	*0.85%*	*None*
UREA	*1.80%*	*Complete in 20 sec.*
NH_4Cl	*0.8%*	*Complete in 310 sec.*
Dist. H_2O		*Complete in 110 sec.*

Part III
Effect of Carbon Chain Length

In the final experiment, the effect of the size of a molecule can be examined. Here molecular size is relative to the number of carbon atoms in its structure. The reagents used and their concentrations are found in Table 3. Add five ml of each to a separate test tube along with a drop of blood. Invert the tubes and mix. After 10 minutes, centrifuge and observe the degree of hemolysis. It is also interesting to note the color of both the liquid and the sediment. Again, the criteria for hemolysis is the same as in Part I.

Table 3
Effect of Carbon Chain Length

Substance	Concentration	Hemolysis
Ethanol	*0.3 M*	*Complete in 28 sec.*
Propanol	*0.3 M*	*Complete in 10 sec.*
Butanol	*0.3 M*	*Instantaneously*

Discussion

Once interested, students may be guided to answer questions such as:

How much salt is in a red blood cell?
How does the size of a molecule effect osmosis and why?
Is there a difference between the use of organic and inorganic salts?
What is the effect of increasing the carbon chain length?
How much of a chemical imbalance can a cell tolerate?
Does the type of chemical substance used effect the answer to the previous question?
Do electrolytes cause osmosis to occur more rapidly?
What can you say about the selectivity of the membrane based on the materials used here?

Summary

As long as all reagents are of good quality and solutions accurately made, this series of experiments can provide consistent results and generate discussion of the data collected. If little advanced information is given to the students on the expected outcome of the experiments, the discussion should prove thought provoking. I have listed only a few of the possible questions to be explored in an exercise of this type. I am sure that teachers will develop others, not to mention those posed by students.

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