



Experiment B-2 Diffusion in Biology



Objectives

- To study the effect of different concentration gradients of NaCl ions on their diffusion rate by measuring their conductivity.

Modules and Sensors

- PC + NeuLog application
- USB-200 module 
- NUL-215 Conductivity logger sensor 

Equipment and Accessories

▪ Utility stand	1
▪ Right angle clamp	1
▪ Extension clamp	1
▪ Pasteur pipette	4
▪ 50 ml beaker	4
▪ 250 ml beaker	1
▪ Sample container	1
▪ Black marker	1
▪ 10 cm of Dialysis tubing	1
▪ Wash bottle	1
▪ Plastic container	1
▪ Clip	2

- The items above are included in the NeuLog Utilities accessories kit, UTL-KIT.

Materials

- | |
|---------------------------------------|
| ▪ 2 g of NaCl |
| ▪ 350 ml of Distilled water |
| ▪ Distilled water for the wash bottle |

Introduction

Diffusion is a physical process in which a substance (gas, liquid, or solid) spontaneously moves toward its surrounding area. The movement of individual particles is unpredictable, but we can predict the movement of groups of particles. If the particles are not distributed evenly then there are at least two regions, one with a higher concentration and one with a lower concentration. This uneven distribution creates a **concentration gradient**. Because there are more particles in the higher concentration region, more particles move randomly from there to the lower concentration region. If particles are not added to or removed from the system, a state of equilibrium is ultimately reached.

The rate of the diffusion of particles might be affected by the following parameters: particles at a higher temperature will have more energy and will move faster than particles at a lower temperature. Smaller particles will move more rapidly than larger particles. The charge (positive or negative) on the particle could also affect the rate of diffusion according to the nature of the material they are moving through.

There are two ways in which molecules move through the cell membrane: passive and active transport. Active transport requires energy, while passive transport does not. Diffusion is part of the passive transport process. The cell membrane allows small and uncharged molecules as water (H₂O), Oxygen (O₂) and Carbon dioxide (CO₂) to pass through it easily.

In this experiment, you will use a dialysis tubing to simulate the membrane in a cell and use three different concentrations of a salt solution. You will place each solution in the dialysis tubing and then place the tubing in a beaker full of distilled water. The salt, NaCl, produces ions when it dissolves in water. Since ions are electrically charged, water solutions containing ions conduct electricity. The ions will diffuse through the dialysis tubing and a conductivity sensor will be used to monitor the conductivity of the water in the beaker. Conductivity will be measured as a function of time and the salt concentration. From your measurement you will obtain the rate of diffusion.

Procedure

Experiment setup

1. Assemble a system like the one in the picture bellow.



2. Make sure you have four 50 ml beakers. One with 50 ml of distilled water and the other three empty.
3. Label the beakers with "DW" (distilled water), "1% NaCl", "5% NaCl" and "10% NaCl". These will be the salt solutions you will test.



4. For the 10% solution, pour 2 g (about half a tea spoon) of NaCl into the corresponding beaker. Pour distilled water into the beaker up to the 20 ml mark. Shake the beaker until the solution is clear.
5. For the 5% solution, pour 10 ml of the 10% solution into the corresponding beaker and add 10 ml of distilled water.
6. For the 1% solution, take 2 ml from the 5 % solution with a Pasteur pipette and pour them into the corresponding beaker. Add distilled water up to the 10 ml mark.
7. Attach the sensor's probe to the utility stand using the extension clamp.
8. Wash the sensor's probe with the wash bottle (which should be filled with distilled water) and the plastic container.
9. Insert the sensor's probe into a 250 ml beaker filled with 100 ml of distilled water.
10. Make sure that the electrodes (in the shape of a circle with a dot inside) are covered with water.



11. Wet the dialysis tubing using the wash bottle (work on a clean table). Each time, you will pour a different salt solution into the dialysis tubing. The salt ions will diffuse into the surrounding water.
12. Fold one end of the dialysis tubing and close it with the plastic clip.
13. Open the dialysis tubing on the other side.

14. Use a Pasteur pipette to fill the tubing with 8 ml of the 1 % salt solution.
15. Fold the other side of the dialysis tubing and close it with the paper clip (do not let air to get inside).
16. Wash the outside of the tubing with the wash bottle to rinse any remnants of salt water.

Sensor setup

17. Connect the USB-200 module  to the PC.
18. Check that the conductivity sensor  is connected to the USB-200 module.

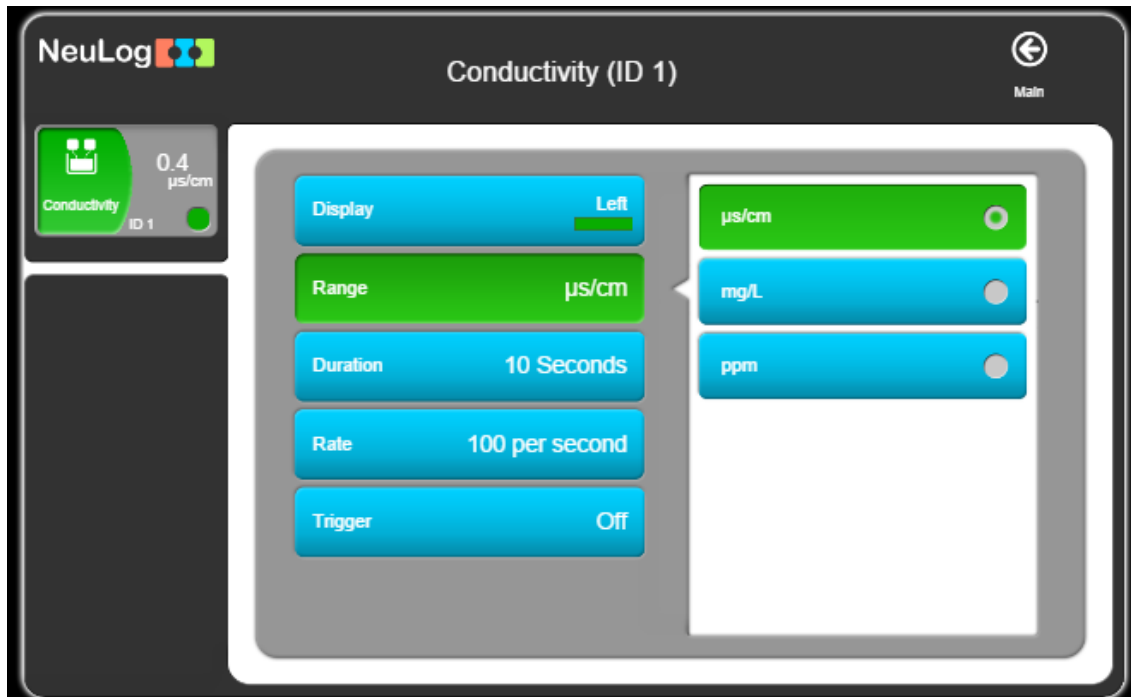
Note:



The following application functions are explained in short. It is recommended to practice the NeuLog application functions (as described in the user manual) beforehand.

19. Run the NeuLog application and check that the conductivity sensor is identified.




Settings

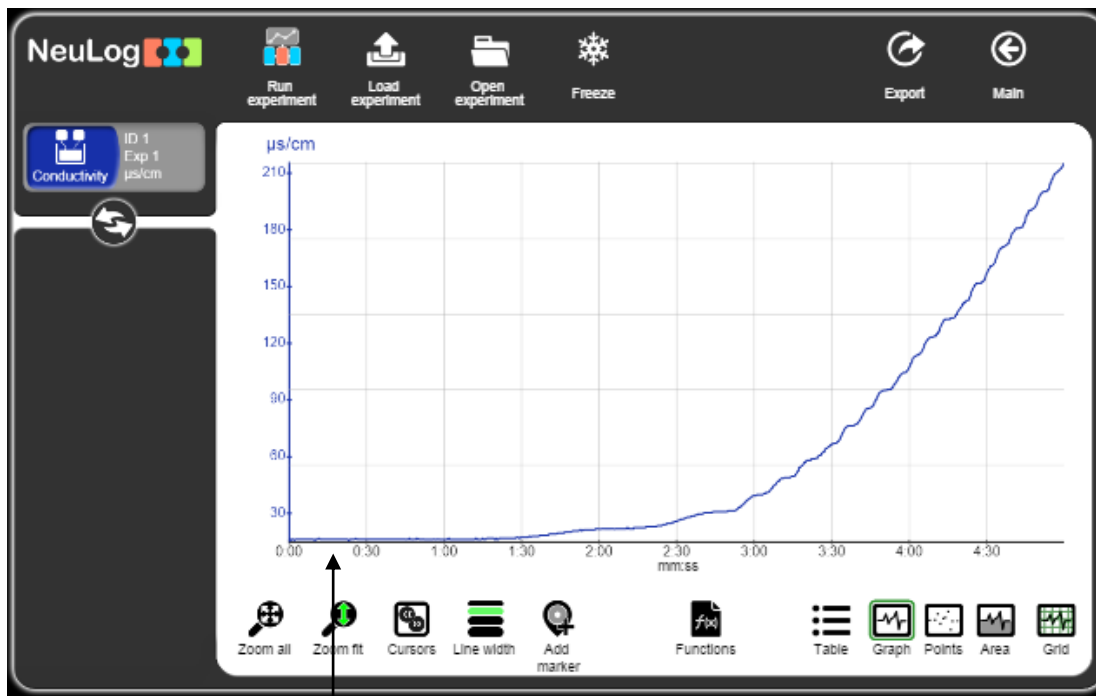
20. Click on the **Sensor's Module** box.
21. Select the $\mu\text{s}/\text{cm}$ button to change the sensor's mode.



22. Click on the  icon to go back to the graph.
23. Click on the **Run Experiment** icon  and set the:
Experiment duration to 5 minutes
Sampling rate to 5 per second




Testing and measurements

24. Insert the dialysis tubing into the 250 ml beaker. Make sure the tubing is completely covered by the water.
25. The distance between the sensor's probe and the dialysis tubing should be the same during all the measurements.
26. Click on the **Record** icon  to start the measurement.
27. Click on the **Arrows** icon  in order to see the sensor's values during the measurement.
28. Click on the **Zoom Fit** icon .
29. Your graph should be similar to the following:

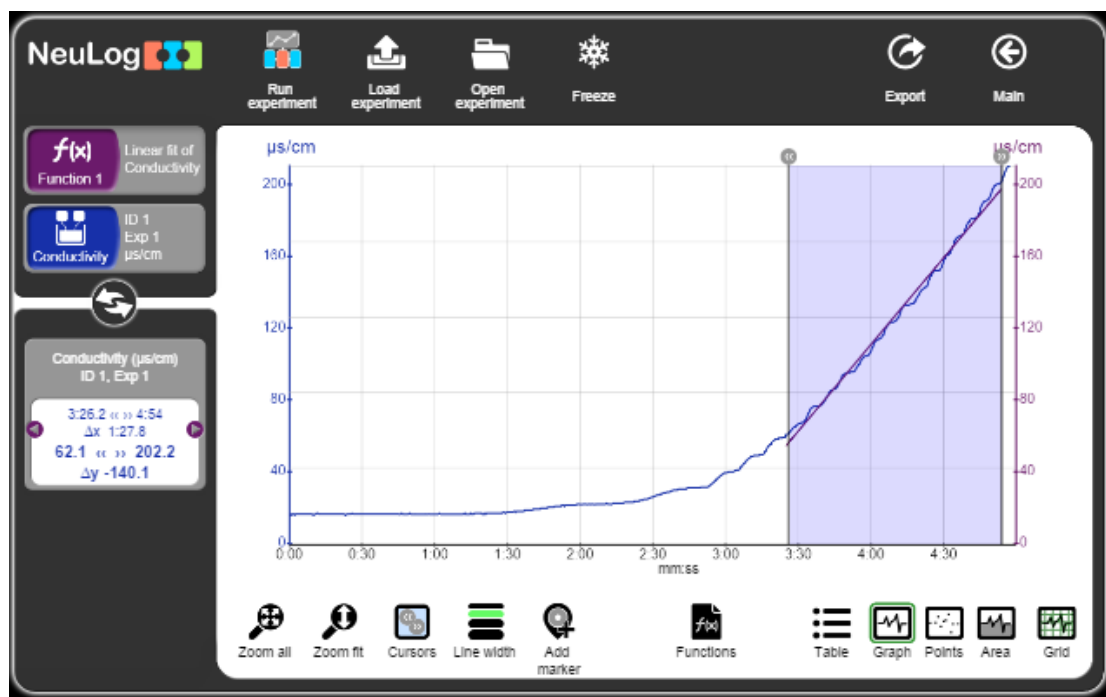


The dialysis tubing was placed in the beaker

30. Click on the **Export** Icon  and then on the **Save value table (.CSV)** button to save your graph.

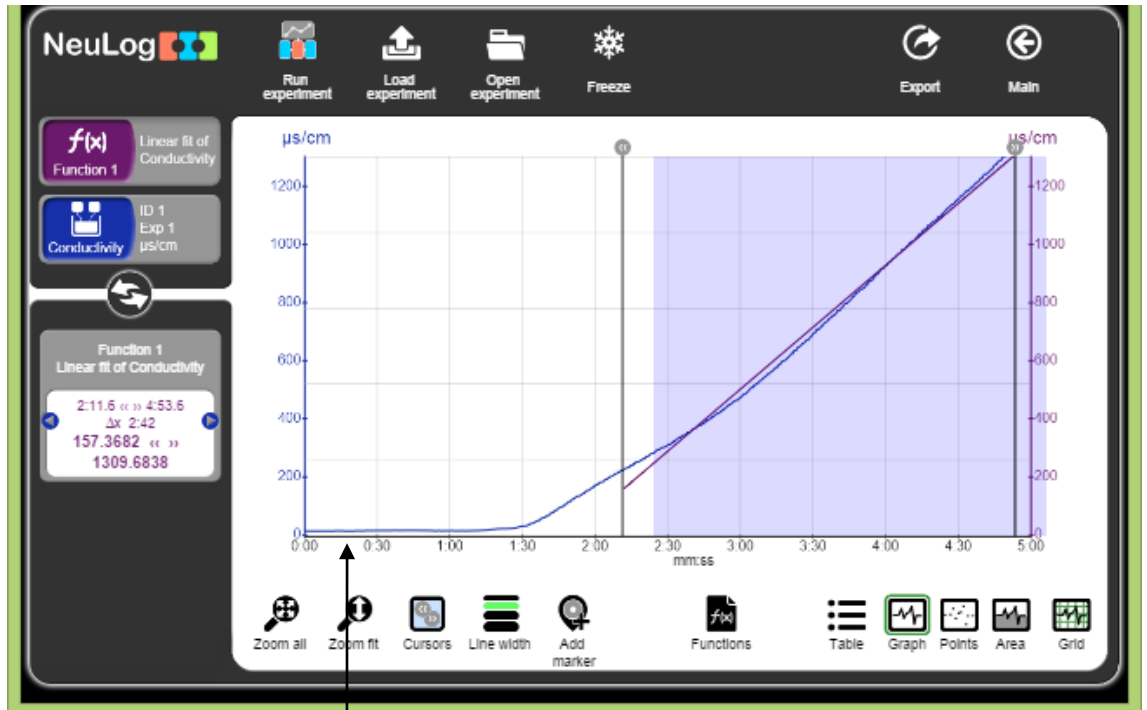
31. Click on the  icon to go back to the graph.
32. Carefully open the plastic clip on one of its sides, spill all the liquid out of the tubing and replace the water in the 250 ml beaker with 100 ml of distilled water.
33. Repeat the experiment, using this time the 5% solution.
34. Repeat again the experiment, using the 10% solution.
35. In order to determine the rate of diffusion in each graph, click on the **Cursors** icon  and select a part of the graph as in the picture below.
36. Click on the **Functions** icon .
37. Click on the **Functions** button on the left of the screen and then click on the **Linear fit of A** button.

Results for the 1% solution:



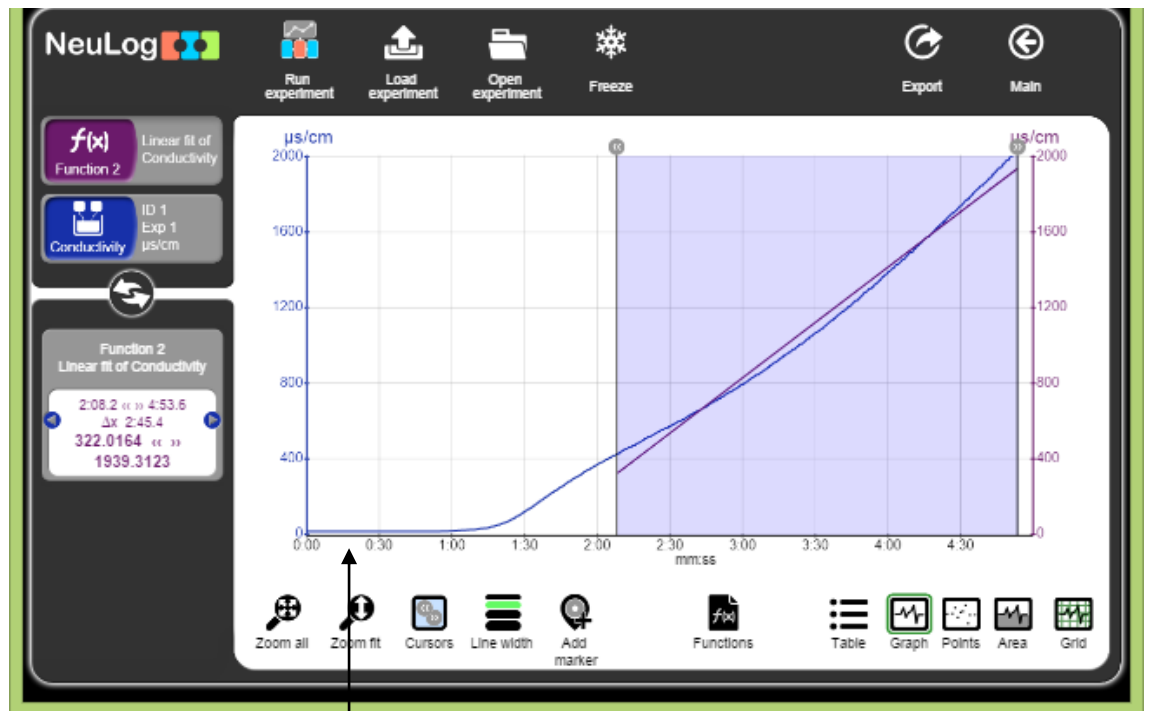
Function 1
Linear fit of Conductivity
 $1.6109X-275.8082$

Results for the 5% solution:



The dialysis tubing was placed in the beaker

Function 1
Linear fit of Conductivity
 $7.1131X-778.7104$

Results for the 10% solution:

The dialysis tubing was placed in the beaker

Function 2
Linear fit of Conductivity
 $9.7781X - 931.5348$

The graphs begin with a slow increase of conductivity, then the curve becomes linear; the slope is the rate of diffusion in each graph.

We can see that as the concentration of the NaCl was higher, the diffusion rate increased. That is because the diffusion rate is affected by the concentration gradient, the greater the difference between two areas, the greater will be the rate of diffusion. When the gradient is zero, there will be no net diffusion.

Summary questions

1. Give an example of another factor that could affect the diffusion rate and was not mentioned.
2. How do you think the graph would look like if we would have continued the measurements for more than 5 minutes? Explain.
3. Can Na^+ and Cl^- ions get in and out through the cell membrane by diffusion? Explain.