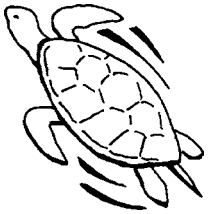


Lab Background and Procedures: Brine Shrimp Tolerance for Fluctuating Environmental Conditions

Introduction

Brine shrimp are crustaceans, like lobsters, crabs, and crayfish. A hard exoskeleton supports their bodies and protects them from injury. Brine shrimp are found throughout the world and are adapted to live in harsh, changing environments. These are not exactly the same environmental conditions as the red shrimp that we've been studying inhabit. However, conditions for both shrimp can vary widely in "salinity" (salt concentration) and temperature.

In this lab, you will test the tolerance of brine shrimp to variations in environmental conditions. The measure of tolerance that you will use is the hatching rate of brine shrimp eggs. The environmental condition that you will vary is salinity.

Brine shrimp eggs are initially housed within a structure called a "cyst," which is similar to an eggshell. Brine shrimp cysts can dry out and the egg inside them will remain viable (able to hatch) for many years. As the egg develops, the cyst bursts. For the first few hours after the cyst bursts, the embryo hangs beneath the cyst in the "umbrella" stage. The embryo continues to develop and will emerge as a free-swimming larva.

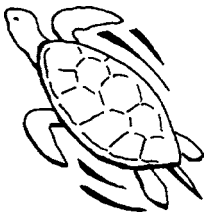
Brine shrimp tend to be brownish-orange in color in this first larval stage. After about 12 hours, they molt (shed their exoskeleton) and begin feeding on tiny algae, bacteria, or nonliving debris. They continue to feed and grow, molting 15 times before reaching adulthood in about eight days.

Adult brine shrimp are generally around 8 mm long. Adult males can be identified by large claspers near their heads, and females by the brood pouch, where the cysts develop, just below the last of their legs. Their lifespan is typically several months.

Materials

Lab Period One

- Brine shrimp cysts
- 1/8 tsp measure
- Four test tubes (150 ml) with stoppers
- Labels or labeling pens for the test tubes
- Test tube rack
- Sea salt or noniodized salt
- Graduated cylinder (more than 100 ml)
- Triple beam or electronic balance
- Distilled water or tap water that has been aged 24 hours in an open container to dechlorinate it



Lab Periods Two and Three

- Hand lens or dissecting microscope
- Sampling pipette (transparent 1 ml or larger)
- Four petri dishes

Lab Period One Procedure

1) On the lab worksheet, write the question or problem investigated by this experiment and your hypothesis.

2) Using salt and the distilled or aged tap water, mix solutions of the following concentrations:
   1 percent salt solution
   5 percent salt solution
   10 percent salt solution
   20 percent salt solution

   The formula for creating solutions of a specific concentration is:
   X grams of NaCl (salt) per 100 ml H_2O (X=target percent concentration)

   Measure 100 ml of water into the graduated cylinder and pour it into a test tube. Weigh the salt and add it to the test tube. Place the stopper on the test tube and mix until the salt is dissolved. Place the test tube in the rack and label it. Mix all of the solutions before moving on to the next step.

3) Unstop all of the test tubes and add 1/8 teaspoon of brine shrimp cysts to each test tube. Leave the tubes open, unless you are moving your test tubes and rack to another part of the room.

4) Leave your test tube rack out of direct sunlight, preferably in a dim part of the classroom. All lab groups should leave their test tube racks in the same location in the classroom.

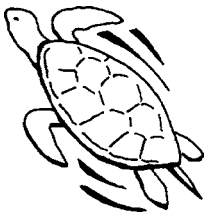
5) Answer question #3 on the lab worksheet.

Lab Period Two Procedure

1) Observe your test tubes. Write your observations on the lab worksheet (question #4, first row in the table)

2) Working with one test tube at a time, make a count of the brine shrimp in a sample volume of water. If the shrimp are not evenly dispersed throughout the water, gently swirl the test tube. Use a pipette to measure 1 ml of water and shrimp (if any have hatched) from the test tube and into a clean petri dish.

3) Using the hand lens or dissecting microscope, count the shrimp in that sample of water and record your findings on the lab worksheet (question #4, first row in the table).



4) Place the shrimp and water back into the test tube they came from.

5) Continue until you have sampled and counted shrimp from each of your four test tubes and recorded your observations on the lab worksheet (question #5, first row of the table). Replace your test tubes and rack to the location where you kept them previously.

6) Answer question #6 on the lab worksheet.

Lab Period Three Procedure

1) Observe your test tubes. Write your observations on the lab worksheet (question #4, second row of the table).

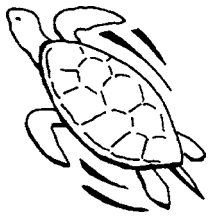
2) Working with one test tube at a time, make a count of the brine shrimp in a sample volume of water. If the shrimp are not evenly dispersed throughout the water, gently swirl the test tube. Use a pipette to measure 1 ml of water and shrimp (if any have hatched) from the test tube and into a clean petri dish.

3) Using the hand lens or dissecting microscope, count the shrimp in that sample of water and record your findings on the lab worksheet (question #5, second row of the table).

4) Replace the shrimp and water back into the test tube they came from.

5) Continue until you have sampled and counted shrimp from each of your four test tubes.

6) Answer questions #6-10 on the lab worksheet.



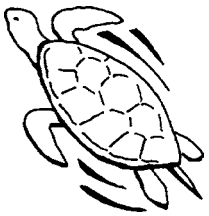
Lab Worksheet: Brine Shrimp Tolerance for Fluctuating Environmental Conditions

Lab Period One Date:

1) State the problem or question this experiment investigates.

2) State your hypothesis.

3) Why is it important for all lab groups to leave their test tubes in the same location? Use the concept of "variable" to explain your response.



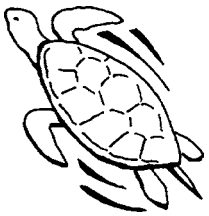
Lab Period Two (Date:) **and Three** (Date:)

4) Test tube observations

	1% solution	5% solution	10% solution	20% solution
Lab Period Two				
Lab Period Three				

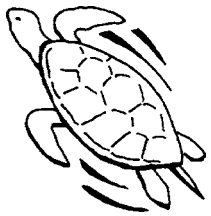
5) Shrimp count per 1 ml water

	1% solution	5% solution	10% solution	20% solution
Lab Period Two				
Lab Period Three				



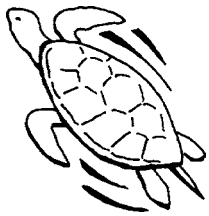
6) Did your group encounter any difficulties counting the brine shrimp in the samples? If so, how were you able to resolve them?

7) Summarize the results of your experiment.



8) Based on the results of your experiment, which solution was the best in which to hatch brine shrimp eggs? Explain your answer.

9) Design another experiment to measure the tolerance of brine shrimp to a different environmental variable such as light or temperature. Describe it here.



10) What types of field observations (made at the ponds, not in a laboratory experiment) could you conduct to test the habitat preferences of the shrimp that live in the anchialine ponds on Maui?