

THE EFFECT OF TEMPERATURE ON THE HATCHING SUCCESS OF BRINE SHRIMPS

Purpose

- To investigate the effect of temperature on the hatching success of brine shrimps.

SAFETY

Write a risk assessment detailing any safety precautions. Include consideration of any ethical issues arising from the use of living organisms. Discuss this with your teacher before starting.



Wash your hands thoroughly after handling the organisms and before you leave the laboratory.

Brine shrimps

Brine shrimps are small, salt water crustaceans; the adults are about 8 mm in length. They are relatively easy to keep in the laboratory and will produce dormant egg cysts that hatch to produce young shrimp larvae.

YOU NEED

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| <ul style="list-style-type: none"> Brine shrimp egg cysts 2 g sea salt for each treatment 100 cm³ de-chlorinated water for each treatment 40 cm³ beaker of salt water 100 cm³ beakers (one for each temperature to be tested) Water baths or incubators (one for each temperature to be investigated) | <ul style="list-style-type: none"> Stirring rod Magnifying glass Pair of forceps Fine glass pipette Bright light Access to refrigerator Sheet of graph paper 3 cm × 4 cm |
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To find out more about brine shrimps, visit the British Ecological Society website, which you can access by going to the weblinks for this activity.

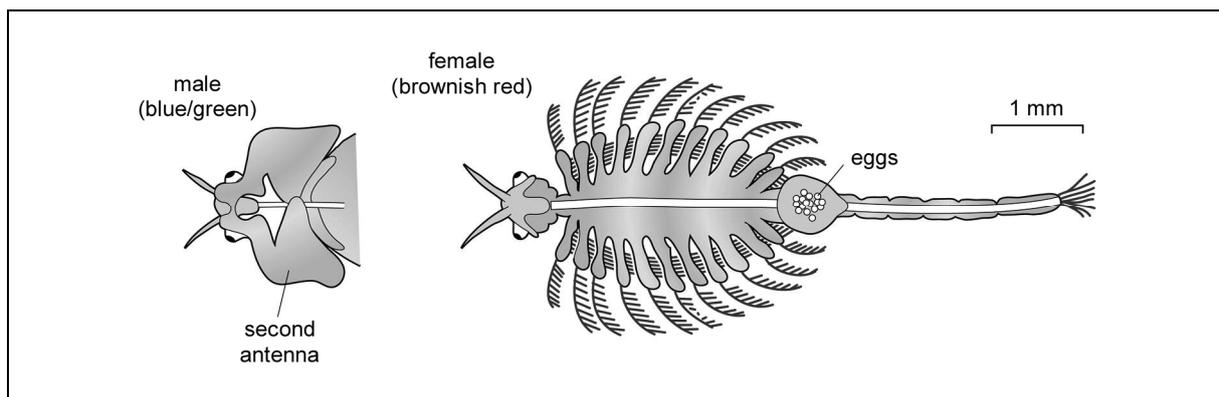


Figure 1 Drawings to show the features of a brine shrimp.

Planning and experimental design

Read through the procedure provided below and assess its suitability for investigating the effect of temperature on the hatching of brine shrimps. Make sure you think about the following points: variables, suitability of the apparatus and method to provide accurate and precise results, and any sources of error. Write a risk assessment as detailed in the safety notes above.

Procedure

- 1 Decide on a range of temperatures from 5 °C to 35 °C to be tested.
- 2 Place 2 g of sea salt into a 100 cm³ beaker.
- 3 Add 100 cm³ of de-chlorinated water and stir until the salt completely dissolves.
- 4 Label the beaker with the temperature at which it will be incubated.
- 5 Place a tiny pinch of egg cysts onto a large sheet of white paper.
- 6 Wet the piece of graph paper using a few drops of salt water. Dab the paper onto the white sheet to pick up approximately 40 eggs. This will look like a tiny shake of pepper. Use a magnifying glass to count the eggs. Cut the graph paper so that there are exactly 40 eggs.
- 7 Put the paper with the 40 eggs into the beaker (eggs-side down). After 3 minutes, use a pair of forceps to gently remove the paper, making sure that all the egg cysts have washed off into the water.
- 8 Repeat steps 2 to 7 for all the temperatures that are to be investigated.
- 9 If possible, replicate the treatments.
- 10 Incubate the beakers at the appropriate temperatures. In the ICT Support there is a datalogging sheet on monitoring temperature in a long-term investigation. Set an alarm to remind you to check your brine shrimps at the same time on three or four successive days.
- 11 The next day count the number of hatched larvae in each of the beakers. To do this, place a bright light next to the beaker. Any larvae will swim towards the light. Using a fine glass pipette, reversed with the tip inserted into the teat, catch the hatched brine shrimps and place them in a small beaker of salt water. Repeat the counting daily. Brine shrimps are very delicate and care must be taken when handling them. Finally, release the young brine shrimps into a salt water aquarium.
- 12 Record the number of larvae that have successfully hatched at each temperature.
- 13 Write-up your experiment making sure your report includes:
 - a discussion of any safety precautions taken
 - comments on the ethical issues arising from the use of living organisms
 - results presented in the most appropriate way
 - an explanation of any patterns in the data using evidence from the data and your own biological knowledge
 - details of any statistical analysis of the results
 - comments on how valid your conclusion is
 - comments on how you ensured that the results obtained in this experiment were accurate and precise
 - suggestions for how you could have improved the quality of your results.

Once you have completed your investigation you could use the Developing Practical Skills Self-evaluation Sheet to reflect on what you have done in this practical.

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SAFETY

Review the students' risk assessments and discuss any safety and ethical considerations.

Ensure students wash their hands thoroughly after handling the organisms and before they leave the laboratory.



Notes on the procedure

If students are given basic information on maintaining brine shrimps, this activity could be planned before they are given the Student Sheet which provides a detailed procedure. The Student Sheet only requires students to assess the procedure, and particularly focuses on safety and ethical issues arising from the use of living organisms. The need to keep conditions other than temperature constant should be appreciated. Brine shrimps hatch in salt water that is 2 to 5% salt (optimum 2.8%) and pH 8.5. Oxygen must be present. Light is an added (but not essential) factor for hatching so the brine shrimps in the refrigerator might be even less successful than expected. Datalogging could be used to check that the conditions within each of the treatments are maintained at a constant level.

This experiment could be completed by small groups of students. If each group completed the range of temperatures then the error between replicates could be investigated. Brine shrimps will hatch in 24 hours at temperatures between 28 and 30 °C, with an optimum of 28 °C.

The practical procedure and technical notes are based on an investigation that appears in the British Ecological Society publication *Brine Shrimp Ecology* by Michael Dockery and Stephen Tomkins. This book contains detailed information on the care and breeding of brine shrimps, and can be downloaded free from the British Ecological Society website.

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General note

This practical takes place over several days. The students set up the beakers with eggs to hatch on the first day. On subsequent days they count the number that have hatched. For detailed guidance on the care and breeding of brine shrimps see the British Ecological Society publication *Brine Shrimp Ecology*. This can be downloaded free from the British Ecological Society website.

Requirements per student or group of students	Notes
Brine shrimp egg cysts	Available from pet shops and Blades Biological. There are approximately 24 000 egg cysts per gram so only tiny quantities are required. Brine shrimps will breed and produce cysts that can be collected. They adhere to the sides of an aquarium tank.
100 cm ³ beakers (one for each temperature to be tested)	
100 cm ³ de-chlorinated water for each beaker	Tap water can be de-chlorinated by leaving it to stand for 48 hours.
2 g sea salt for each beaker	Students could be supplied with the salt water already prepared.
Stirring rod	
Access to refrigerator	
Water baths or incubators (one for each temperature to be investigated)	A range of temperatures between 5 and 35 °C is recommended. The optimum for hatching is 28 °C.
Sheet of graph paper 3 cm × 4 cm	
Magnifying glass	
Pair of forceps	
Bright light	A lamp or light from one side, for counting larvae on the second day.
Fine glass pipette	For counting larvae on the second day.
Small beaker of salt water	For counting larvae on the second day.
Large beaker or tank of salt water	To hold the brine shrimps after hatching.