

"Let's Hang Out with the Tough Guys...the Sea Monkeys"

Grade Level: 6-12

Subject Area: Biology, Aquaculture, Agriculture, Chemistry **Time**: 24 hours total for hatching *Day 1:* 50 minute class period to decapsulate eggs and prepare for hatching *Day 2:* 50 minute period to observe

hatched brine shrimp

Student Performance Standards (Sunshine State Standards):

11.03 Identify and describe the basic structures and external anatomy of crustaceans (LA.910.1.6.1, 2, 3, 4, 5; LA.910.2.2.2).

11.10 List and describe the major factors in the growth of aquatic fauna and flora (LA.910.1.6.1, 2, 3, 4, 5; LA.910.2.2.2; SC.7.L.17.1, 2, 3).

12.01 Recognize and observe safety practices necessary in carrying out aquaculture activities (LA.910.1.6.1, 2, 3, 4, 5).

12.03 Safely operate aquaculture machinery and equipment (LA.910.1.6.1, 2, 3, 4, 5).

14.01 Identify factors to consider in determining whether to grow an aquaculture species (LA.910.1.6.1, 2, 3, 4, 5; LA.910.5.2.3, 4, 5; LA.910.2.2.2; SC.7.L.17.3).

14.02 Identify/describe facilities used in a grow out operation (LA.910.1.6.1, 2, 3, 4, 5; LA.910.6.4.1).

14.04 Determine the purpose and functions of a hatchery (LA.910.1.6.1, 2, 3, 4, 5; LA.910.6.4.1).

Objectives: Students will be able to:

1. Propagate and rear aquatic species.

2. Construct qualitative and quantitative observations on different developmental stage and behavior of aquatic species.

Abstract:

Brine shrimp live and are harvested primarily from natural salt lakes and solar salt operations around the world; the original and still popular commercial source for brine shrimp is the Great Salt Lake in Utah, and the San Francisco Bay in California. Adult brine shrimp are also called 'sea monkeys' and are used by hobbyists for their personal entertainment or to feed some of their pet fish. The following lesson plan provides a summary on how to decapsulate and hatch brine shrimp cysts since it is an essential tool to know in aquaculture practice, and also necessary to conduct a myriad of experiments that call for brine shrimp as model in many disciplines. Brine shrimp can be used to observe, control, and manipulate the life cycle of a species under laboratory conditions, over a very short period of time (less than 2 weeks). They can be an excellent model for exploring the life cycle of an arthropod in the classroom as well as a useful tool to teach the scientific method through an inquiry-based experimental approach.

Interest Approach:

"Let's hang out the tough guys...the sea monkeys"

- 1. Temperatures: 6-35°C/43-95°F
- 2. High salinity: 70-240 parts per thousand (ppt)
 - a. the ocean is just 35 ppt
 - b. sea monkeys have the most efficient systems to regulate salt (chloride, sulphate, and carbonate waters)
- 3. Adults are 2-3 millimeters and the eggs are 200-300 microns in diameter (very small)
- 4. The very young (embryos) ca live most of their lives inside a capsule in a dry state and without much oxygen
- 5. Larvae molts ~15 times after hatching
- 6. Artemia live in harsh environments to avoid predators since they are very nutrition and delicious to other bug eating animals
- 7. And they're not very good-looking



Student Materials:

- 1. Air pump
- 2. Air tubing
- 3. Salt
- 4. Measuring spoon
- 5. Thermometer
- 6. Cone like container
- 7. Brine shrimp eggs

Teacher Materials:

Material

Store

8. Beaker of bleach

11. Magnifying glass

10. Gloves

12. Funnel

13. Filter

14. Scale

9. Vial of NaOH (sodium hydroxide)

Estimated Cost

Air pump	PetsMart, Carolina Biological	\$10 and up
One liter bottles or similar containers (one/group of four)	Any grocey store	\$1.50 and up
Artemia eggs ~30 grams/class, 3-5 grams/group	Online (www.aquacave.com for example)	\$14.95 for 4 oz.
Marine salt (for aquariums)	Online, PetsMart	\$2.95 and up
Dechlorinated tap water/fresh water (one gallon/group)	NA	NA
Clorox bleach	Any grocery store	\$5 and up
NaOH	Carolina Biological	\$7 and up
100-150 μL mesh filter (one/group of four)	Online	\$2 and up
Digital scales that measure to grams	NA	NA

Student Instructions:

Decapsulation Instructions:

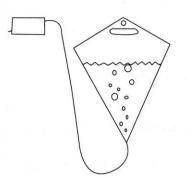
- 1. Put on gloves because you will be working with caustic materials.
- 2. Measure out 150 mL of bleach and add to hydrated brine shrimp container.
- 3. Measure out 5 mL of NaOH and add to brine shrimp container.
- 4. Mark the time.
- 5. Place aeration into solution.

- 6. Observe color change for 3-8 minutes; brine shrimp eggs will change color from dark brown to gray to orange, this is the "shell" or capsule being dissolved off the brine shrimp egg membrane.
- 7. Rinse the decapsulated eggs with fresh water, through a fine mesh filter.
- 8. Continue to run the fresh water over the fine mesh filter until bleach smell can no longer be detected.
- 9. Rinse from filter with fresh water into hatching container.
- 10. Prepare decapsulated eggs for hatching.

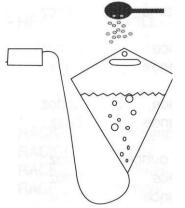
Hatching Instructions:

DAY 1

- 1. Place aeration tubing into hatching container.
- 2. Add one liter of spring water to hatching container with the already decapsulated brine shrimp eggs.

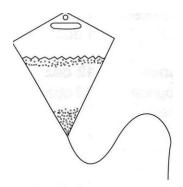


3. Measure out 25 grams of marine salt and add to hatching container.



4. Eggs will hatch in 24-36 hours depending on water temperature.

- DAY 2
 - 1. After eggs hatch, turn off aeration. Wait for brine shrimp to settle to the bottom of the hatching container.



- 2. Take pipette and pipette-out several mL of hatched brine shrimp.
- 3. Place in Petri dish and observe brine shrimp under dissecting scope.
- 4. If the brine shrimp are fed yeast, they can be observed over the next few days, as the newly hatched brine shrimp become adults.

Teacher Instructions:

Preparations:

- 1. Hydrate the brine shrimp eggs for each group 1 hour before class is to begin by hydrating one pound of brine shrimp eggs with one gallon of fresh water under aeration- this becomes your stock solution of hydrated brine shrimp eggs. This will be enough hydrated brine shrimp eggs for 30 groups (4 students per group or 120 students total). It is essential for eggs to be hydrated before the start of decapsulation. Then separate out hydrated eggs into smaller one-liter containers for each participating group.
- 2. Prepare stock NaoH solution by adding 1.5 oz of sodium hydroxide to 1 quart of water.
- 3. Place all student materials at their stations.

Activity:

1. Observe students preparing decapsulation of brine shrimp. Say a word about safety to students since they will be working with bleach and sodium hydroxide. Sodium hydroxide is a strong base and can be caustic.

Post work/Clean-up:

1. After brine shrimp have hatched and activity is concluded, the remaining live brine shrimp can be given to aquarium fish to eat. They make a tasty and nutritious snack for most aquarium fish. If you don't have access to a fish tank, then place in a local pond and many animals will begin to prey upon.

Anticipated Results:

The activity should result in a successful performance of the total decapsulation process as well as observation of hatched brine shrimp. The students should observe

the behavior and record their observations. The following are optimal conditions under which brine shrimp should be hatched:

Optimal conditions recommended for hatching of brine shrimp cysts include:

1. Constant temperature between 25-28 °C.

2. Strong illumination of 2000 lux.

3. Salinity of 15-35 parts per thousand and oxygen levels above 2 parts per million. 4. Decapsulating brine shrimp eggs should be considered. Decapsulation consists of removing the chorion, or shell, of the artemia cysts and leaving only the embryo surrounded by its thin membrane. Decapsulated artemia cysts offer several advantages compared to the normal packed cysts; most prominent decapsulated artemia cysts have higher energy content, and the soft eggs are easier to digest.

The brine shrimp is used extensively for experiments from simple description of crustacean life cycle, physiological response to varying environmental conditions, to assessment of effects that microgravity and cosmic rays have on developing organisms; the latter experiments easily planned by students in K-12, and under sponsorship from NASA, was conducted by astronauts in space orbit. A simple search in the internet entering key words such as brine shrimp modules, experiments - provide an exhaustive list for activities that can easily be adopted to any type of classroom setting. For example, use brine shrimp as a model to understand effects environmental changes influence on an organism throughout its life. Specifically, some examples include:

1. The effect temperature, varying salt tolerances, and light intensity have on hatching rates, and survival of the nauplii.

2. The effect feeding different types of foods (e.g., yeast and yeast + Liquid HUFA) have on adult reproduction, and their offspring. HUFA's, or highly unsaturated fatty acids, are an essential nutrient mostly for the rapidly developing young of both terrestrial and aquatic species. Brine shrimp are an excellent substitute to more traditional large animal systems to quickly demonstrate the effects that such nutrients may have on the health of organisms.

Support Materials:

1. Teacher background with references

Explanation of Concepts:

Science concepts demonstrated include using observational skills, making experimental measurements, and following procedures. Students are also learning a critical aquaculture skill: the procedure behind raising an organism from egg to adult. See teacher background for more of the science concepts covered.



Teacher Background with References

Brine Shrimp Decapsulation and Hatching

Brine shrimp, or Sea Monkeys, are Crustaceans and their scientific name is *Artemia salina*; or simply referred to as Artemia. Crustaceans are arthropods so their sister groups are the arachnids or spiders, centipede/millipedes, and the insects. Crustaceans are a highly valuable group of aquaculture species comprised of such animals like crabs, lobsters, and shrimps; although they do not crawl instead attach themselves to rocks, the barnacles are also considered crustaceans. Most crustaceans are raised for direct human consumption like marine and freshwater shrimps; but the brine shrimp (*Artemia* spp.) and more recently copepods are popular for feeding the very small larval and juveniles of other animals destined also for aquaculture purposes (like finfish and even other but larger, edible shrimps and lobsters).

One of the major criteria for classifying crustaceans is that many have freeswimming, nauplius larvae (plural: nauplii), characterized with three pairs of appendages and usually a single eye. Aside from the human edible shrimps and lobsters, the brine shrimp or Artemia is probably the best known of convenient replacements for the natural diet of many fish larvae and juveniles used in aquaculture today. The use of brine shrimp was considered a breakthrough in larval feeding technology and permitted the mass culture of many valuable species of freshwater and marine fish species (e.g., flatfishes, sea bream, sea basses, carps). Brine shrimp are also commonly used to feed many types of ornamental fish, as well as the postlarvae of edible shrimps, crabs, and lobsters; brine shrimp has also been identified as a critical component in the diet for the 'conditioning' or maturation of these animal groups.

Brine shrimp live and are harvested primarily from natural salt lakes and solar salt operations around the world; the original and still popular commercial source for brine shrimp is the Great Salt Lake in Utah, and the San Francisco Bay in California. Although some brine shrimp are harvested as adult individuals (e.g., used by hobbyists for their personal entertainment or to feed some of their pet fish), the vast majority of the production is in the form of cysts. Hundreds of tons of brine shrimp cysts are harvested annually, packed, and utilized by the aquaculture industry worldwide. Brine shrimp females can produce either live nauplii or form cysts, depending on their surrounding environment. Such reproductive adaptation permits survival of the species during periods of extreme situations like high salinity, extreme temperatures, and drought or desiccation. Brine shrimp cysts are true embryos (not eggs in the strict meaning of the word); embryos that are in a state of physiological dormancy or 'diapause'. In the wild, the cysts float at the surface of the water, where they are harvested either in the open water or along the shoreline as they are driven ashore by winds and waves. Dry artemia cysts are commercialized more commonly as brine shrimp eggs. The packed cyst may be stored for months or even years, and will remain in dormancy as long as they are kept dry. The dormant embryo is encased by a membrane and an outer shell, or chorion, that is very hard and protects the embryo from the harsh outside conditions. Dry, brine shrimp eggs are small (around 200 microns in diameter) and when viewed under a microscope have a large dimple on their side. Changes in water temperature and salinity usually cause the brine shrimp eggs to rehydrate and continue their development again; as the cysts hydrate they become spherical. After the cyst hydrates for some 20 hours (at 25-28 °C), its ruptures and the embryo appears surrounded by a membrane. Shortly thereafter the embryo hatches releasing a free-swimming nauplius. The nauplius larvae molt a couple of times, while they are feeding on their own yolk reserves. The naupllii at this stage of development are richest in energy, therefore, are the most widely used form of artemia in aquaculture. Brine shrimp are suspension feeders, and feed continuously from the water column. Brine shrimp can be fed, yeast, algae, or bacteria. Since brine shrimp are raised primarily to feed other aquaculture species, they can also be enriched with lipids, proteins, and other nutrients beneficial to the animal that ingest them. The nauplli can also serve as a carrier of chemicals to treat or prevent diseases on those who eat them. After 2 weeks of culturing the brine shrimp may be about 5 mm in length and can also be harvested. Before reaching adulthood, the nauplii molt some 15 times and reach a size of approximately 8-10 mm. Adult male brine shrimp are easily recognized having a well developed mandible on their heads and the female having a distinct brood pouch towards the end of the main body. Female brine shrimp can live up to 3-4 months and every 4-6 days release some 300 nauplii or cysts.

Year-round availability of brine shrimp eggs is necessary to supply aquaculture hatchery and nursery operations worldwide. As mentioned before, most of the world supply of brine shrimp is harvested from the wild. Most brine shrimp cysts are purchased, and their production in aquaculture operations is restricted to hatching and harvesting the nauplii. The hatching and harvesting of brine shrimp cysts is a relatively simple operation but several minimal considerations and critical factors should be followed to obtain the large quantities of nauplii needed in commercial aquaculture operations (see separate instructions, in lesson plan).

References

Manual on the Production and Use of Live Food for Aquaculture, URL: ftp://ftp.fao.org/docrep/fao/003/w3732e/w3732e00.pdf

About brine shrimp, URL: http://saltlakebrineshrimp.com/about-brine-shrimp/

Use of Artemia (Biology, Tank and Pond Production), URL: http://www.aquaculture.ugent.be/coursmat/online%20courses/faoman/1st_page.htm

Decapsulating brine shrimp eggs, URL: http://edis.ifas.ufl.edu/FA023

The Culture and Use of Brine Shrimp, as Food For Hatchery-Raised Larval Prawns, Shrimps, and Fish-FAO, URL: http://www.fao.org/docrep/field/003/AB906E/AB906E00.HTM

The Brine Shrimp Project, URL: http://www.ncsu.edu/sciencejunction/terminal/lessons/brine.html