How to prepare a brine shrimp culture in a hatching cone

Purpose

This standard operating procedure (SOP) describes methods for preparing brine shrimp cultures to feed young stickleback (*Gasterosteus aculeatus*).

Policy

These parameters must be regularly checked and documented to ensure in compliance with CCAC. Failure to abide by the SOP may result in disciplinary action against the individual(s).

Responsibility

Student, technical personnel, investigator who have successfully completed the ethic training requirements of the Canadian Council on Animal Care (CCAC) and National Institutional Animal User Training (NIAUT) Program.

Materials

- Hatching cone
- Brine shrimp eggs
- Clean, dechlorinated saltwater
- Air pump and tubing
- Warm water bath, such as a 3/4-filled aquarium, containing water heated to 27-29 degrees C OR environment chamber heated to 25-29 degrees C
- Aquarium heaters for water bath
- A lamp giving off bright light, day and night
- Scrub pad to clean hatching cone

Procedure

1. Scrub the hatching cone to remove any bacterial slime from the sides.
2. Fill the cone with salt water, between 10-12 ppt (Note: this low salinity is ideal for hatching brine shrimp, but a higher concentration, 28-32 ppt, is necessary to keep the shrimp alive over 24h, as they will die at lower concentrations). Use a higher salt concentration when rotating cultures. Adjust salinity by adding salt or water, as necessary.

3. Insert the air line tubing into the cone and make sure the straw at the end of the air line is inserted into the hole at the bottom of the cone. The water should be bubbling vigorously.

4. Place the bright light above the jar and keep on always.

5. Eggs should hatch within 24 hr.

6. Separate the live brine shrimp from the eggs. When hatching is complete remove the air line from the cone and leave alone for about 15 minutes. The empty shells, if present, will float to the top. Unhatched eggs will sink to the bottom. The live brine shrimp (bright orange) will be hovering in the middle.

You may have to repeat the egg separation step a few times. It is important that we separate as many eggs as possible as they will block the digestive systems of the baby fish, which will cause mortality.