# Spawning & Larviculture of Bivalve Mollusks

| Grade Level: | Subject Area: | Time: |
|---|---|---|
| 9-12 | Aquaculture, Biology, Reproduction, Anatomy | *Preparation:* 30 minutes to prepare<br>*Activity:* 50 minute lecture (may require 1 ½ class periods)<br>*Clean-up:* NA |

**SPS (SSS):**

*06.04* Employ correct terminologies for animal species and conditions (e.g. sex, age, etc.) (LA.A.1.4.1-4; LA.A.2.4.4; LA.B.1.4.1-3; LA.B.2.4.1-3; LA.C.1.4.1; LA.C.2.4.1).
*11.09* Develop an information file in aquaculture species (LA.A.1.4, 2.4; LA.B.1.4, 2.4; LA.C.1.4, 2.4, 3.4; LA.D.2.4; SC.D.1.4; SC.F.1.4, 2.4; SC.G.1.4, 2.4).
*11.10* List and describe the major factors in the growth of aquatic fauna and flora (LA.A.1.4, 2.4; LA.B.1.4, 2.4; LA.C.1.4, 2.4, 3.4; LA.D.2.4; SC.D.1.4, SC.F.1.4, 2.4; SC.G.1.4, 2.4).
*13.02* Explain how changes in water affect aquatic life (LA.A.1.4, 2.4; LA.B.2.4; LA.C.1.4, 2.4; LA.D.2.4; SC.F.1.4; SC.G.1.4).
*13.03* Explain, monitor, and maintain freshwater/saltwater quality standards for the production of desirable species (LA.A.1.4, 2.4; LA.B.2.4; LA.C.1.4, 2.4; LA.D.2.4; MA>B.1.4; MA.E.1.4, 2.4, 3.4; SC.E.2.4; SC.F.2.4; SC.G.1.4).
*14.01* Identify factors to consider in determining whether to grow an aquaculture species (LA.A.1.4, 2.4; LA.B.2.4; LA.C.1.4, 2.4; MA.A.1.4, 2.4, 5.4; MA.B.1.4, 3.4; MA.D.2.4; DD.N.2.4; SS.D.2.4, 3.4).
*14.02* Identify/describe facilities used in a grow out operation (LA.A.1.4, 2.4; LA.B.2.4; LA.C.1.4, 2.4; MA.A.5.4; SC.G.1.4; SS.B.2.4).
*14.04* Determine the purpose and functions of a hatchery (LA.A.1.4, 2.4; LA.B.1.4, 2.4; LA.C.1.4, 2.4; MA.A.1.4, 5.4; MA.E.1.4, 2.4; SC.F.1.4, 2.4; SC.G.1.4, 2.4; SS.B.2.4; SS.D.1.4, 2.4).
*14.05* Identify and describe the sexual reproductive process and methods of reproducing aquaculture organisms (LA.A.1.4, 2.4; LA.B.2.4; LA.C.1.4, 2.4; SC.F.1.4, 2.4; SC.G.1.4, 2.4; SC.H.1.4, 2.4, 3.4).
*14.06* Identify and describe the spawning facilities used in aquaculture (LA.A.1.4, 2.4; LA.B.2.4; LA.C.1.4, 2.4; MA.A.1.4, 2.4, 3.4, 4.4, 5.4; MA.C.1.4; SC.F.1.4, 2.4; SC.G.1.4, 2.4; SC.H.1.4, 2.4, 3.4).
*15.01* Identify the types of growing systems and important factors in their selection, design, and use (LA.A.1.4, 2.4; LA.B.2.4; LA.C.1.4, 2.4; MA.A.1.4, 2.4, 3.4, 4.4, 5.4; MA.B.1.4, 2.4,

3.4, 4.4; MA.C.3.4; MA.D.1.4, 2.4; MA.E.2.4; SC.G.1.4, 2.4; SC.H.3.4).

**Objectives**:

1. Students will be able to describe reproductive characteristics in bivalve mollusks.
2. Students will be able to describe the environmental conditions that induce spawning in bivalve mollusks.
3. Students will be able to describe the process of spawning for bivalve mollusks.

**Abstract**:

In the simplest molluskan reproductive systems, two gonads sit next to the coelom that surrounds the heart and shed ova or sperm into the coleom, from which the nephridia extract them and emit them into the mantle cavity. Mollusks that use such a system remain of one sex all their lives and rely on external fertilization. Some mollusks use internal fertilization and / or are hermaphrodites, where an individual can function as both sexes; both of these methods require more complex reproductive systems.

The most basic molluskan larva is a trochophore which is planktonic and feeds on floating food particles by using the two bands of cilia around its "equator" to sweep food into the mouth, which uses more cilia to drive them into the stomach, which uses further cilia to expel undigested remains through the anus. New tissue grows in the bands of mesoderm in the interior, so that the apical tuft and anus are pushed further apart as the animal grows. The trochophore stage is often succeeded by a veliger stage in which the prototroch, the "equatorial" band of cilia nearest the apical tuft, develops into the velum ("veil"), a pair of cilia-bearing lobes with which the larva swims. Eventually the larva sinks to the seafloor and metamorphoses into the adult form. In some species the newborn larvae are already veligers, and other species have direct development, in which a miniature adult emerges from the egg.

Bivalve reproduction takes place externally when eggs and sperm from the **gonads** are released into the water, and a female may spawn millions of eggs in her lifetime. Once fertilized, tiny, free-swimming larvae are formed. This planktonic stage usually lasts a few days, and the larvae settle and attach themselves to a hard surface, where they then begin to form a shell.

**Interest Approach**:

In small groups have students try to come up with definitions for the following words:

hermaphroditic
protandrous

oocytes
synchronous spawning
trochophore
veliger
metamorphosis

Share a few of the class definitions then explain that the lecture will cover these words.

**Student Materials**:
1. Paper
2. Pencil

**Teacher Materials**:

| *Material* | *Store* | *Estimated Cost* |
|---|---|---|
| *Bivalve Reproduction* lecture notes | NA | NA |
| *Bivalve Reproduction* figures | NA | NA |

**Student Instructions:**
1. Actively participate in a lecture regarding bivalve reproduction.

**Teacher Instructions:**

*Preparations*:
This activity is an overview of bivalve reproduction and is intended to be used within a classroom lecture framework. Lecture notes and figures for this lesson are in the SUPPORT MATERIALS section.

*Activity*:
1. Lecture on *Bivalve Reproduction*

*Post work/Clean-up*:
There is no clean-up for this lesson.

NOTE: As an alternate activity, have students collect adult clams or oysters from the wild during the appropriate season (spring/fall). Dissect some of the specimens to determine if they are in reproductive condition to spawn.

**Anticipated Results:**

Students will become knowledgeable of the reproductive biology of bivalve mollusks, spawning and larval development, and setting and nursery culture. The hard clam, known throughout New England as the northern quahog, (*Mercenaria mercenaria*) will be used as a model.

**Support Materials:**

1. *Bivalve Reproduction* lecture notes
2. *Bivalve Reproduction* figures
3. Online Publications: Southern Regional Aquaculture Center — www.msstate.edu/dept/srac/publicat.htm
4. *Biology and Culture of the Northern Quahog* (*Mercenaria mercenaria*) (SRAC 433)
5. *The Cultivation of American Oysters* (SRAC 432)
6. Helm, M.M. and N. Bourne. 2004. *The Hatchery Culture of Bivalves: a Practical Manual*. FAO Technical Paper No. 471. http://www.fao.org/docrep/007/y5720e/y5720e00.htm
7. Adams, C. *et al.* 1991. *Investing in Commercial Hard Clam Culture: A Comprehensive Guide to the South Atlantic States*. Florida Sea Grant Report No. 104 (SGR-104). Gainesville, FL. 128 pp.
8. Huner, J.V. and E.E. Brown (Eds.). 1985. *Crustaceans and Mollusk Aquaculture in the United States*. AVI Publishing Company, Inc., Westport, Connecticut. 476 pp.
9. Castagna, M. and J. Kreuter. 1981. *Manual for Growing the Hard Clam Mercenaria.* Sea Grant Program, Virginia Institute of Marine Science, Gloucester Point, VA. 107 pp.
10. Landau, M. 1992. *Introduction to Aquaculture*. John Wiley and Sons, New York. 440 pp.
11. Manzi, J. and M. Castagna. (Eds.). 1989. *Clam Mariculture in North America.* Elsevier Press, New York, New York. 461 pp.
12. Manzi, J. and J.M. Whetstone. 1983. *Intensive Hard Clam Mariculture: A Primer for South Carolina Watermen*. South Carolina Sea Grant Consortium Publication No. 91-01. 20 pp.
13. Quayle, D.B. and G.F. Newkirk. 1989. *Farming Bivalve Mollusks: Methods for Study and Development*. Advances in World Aquaculture, Volume 1. World Aquaculture Society, Baton Rouge, LA. 294 pp.
14. Sarkis, S. 2008. *Installation and Operation of a Modular Bivalve Hatchery*. FAO Fisheries Technical Paper No. 492. Rome, Italy. 173 pp.
15. Vaughan, D., L. Creswell, and M. Pardee. 1990. *A Manual for Farming the Hard-Shell Clam in Florida*. Florida Dept. of Agriculture and Consumer Services, Tallahassee, FL. 42 pp.

**Explanation of Concepts:**
Characteristics of bivalve reproduction
Environmental conditions for bivalve reproduction
Spawning, larviculture, metamorphosis, and post-set nursery

## *Support Materials*

### *Bivalve Reproduction*
### *Lecture Notes*

I. General Characteristics
   a. Bivalve mollusks, such as clams, oysters and scallops have unique reproductive biology, in that they are all **hermaphroditic**
      i. They are both male and female during their lifetime
      ii. Clams and oysters are **protandrous** (begin as males and later develop into females)
      iii. In contrast, many fish such as grouper are **protogynous** (females that later develop into males)
      iv. Scallops are even more unique, they are **simultaneous hermaphrodites**
      v. Reproductive organs
         1. Oocytes (eggs) and sperm are located overlying the visceral mass (see bivalve anatomy)
         2. Often ranging in color from pale yellow or white to orange, oocytes appear slightly granular
         3. Sperm are a more creamy texture
         4. As the gametes develop they can completely fill the body cavity within the two valves (shells), particularly in ripe females; scientist quantify this development as the **condition index**.
   b. Bivalve mollusks are **broadcast spawners**, a common term used to describe external fertilization
      i. Males and females release their gametes through their excurrent siphon
      ii. The eggs are fertilized in the open water
      iii. It is important that the eggs and sperm are released at the same time to ensure fertilization before currents carry the gametes in different directions — hence, the term **synchronous spawning**.

II. Determining Factors and Environmental Conditions

a. Bivalves become conditioned to spawn at a time that will optimize the survival of the planktonic larvae, primarily the availability of food — microscopic, unicellular, planktonic plants called phytoplankton

b. In temperate areas
   i. New England and the mid-Atlantic region
   ii. Phytoplankton standing crop gradually increases in the spring as the water warms
   iii. At higher latitudes temperature is the primary factor for phytoplankton growth
   iv. Bivalves typically spawn in the late spring, and again in the fall after they have regenerated their gonads

c. In tropical climates (lower latitudes)
   i. Seasons are distinguished by precipitation rather than temperature (wet/dry season rather than winter/summer)
   ii. Runoff of nutrients from the land plays the primary role in stimulating phytoplankton growth and bivalve reproductive condition

d. In Florida
   i. Bivalves respond to both of these factors (being a subtropical climate)
   ii. Along the east coast of Florida the spawning period for clams extends, generally, from early March through mid-May and again in October through November

e. Most large hatcheries have "conditioning tanks" which hold the broodstock at a constant, cool temperature (22°C)

f. In order to bring the clams into spawning condition, significant amounts of algal feed are required
   i. Usually only large hatcheries with year-round operations are equipped to handle the algal feed requirements

g. Small hatcheries typically collect broodstock from the environment when conditions are right, operating their larviculture system for a few months a year.

III. Spawning

a. When broodstock clams are in spawning condition, they should be transferred to a spawning table or trays
   i. Tables are usually long and narrow
   ii. Less than 6 inches deep
   iii. Painted black to facilitate observation of the eggs and sperm
      1. Glass or clear plastic trays can be placed on a black surface (or paper) with the same result
   iv. Place the clams on a table or in trays and add cool seawater (20°C.)
      1. If algal food is available, it should be added to induce the clams to extend their siphons and begin feeding

b. After about 20 minutes, and after the clams have their siphons extended indicating that they are filtering, the water temperature is gradually raised to around 30°C over a 20 minute period
    i. This technique is known as "thermal shock"
    ii. It provides a critical stimulus for synchronous spawning
c. Typically, male clams will begin to spawn prior to females, ejecting a constant stream of sperm into the water
d. Females will respond to the presence of sperm and will release eggs in a more sporadic fashion
e. If the clams do not begin to spawn within 30 minutes to an hour, the water temperature should be lowered again to 20°C, and the process repeated
    i. Interestingly, clams tend to spawn on rising temperature cycles during the spring, and falling temperature cycles in the fall
f. If clams fail to spawn after two or three heating/cooling cycles, sperm can be extracted from gonads of a dissected male and put in suspension with seawater
    i. The sperm water mixture is then pipetted into the spawning water, or in proximity to the incurrent siphon of a feeding female (Figure 3B)
    ii. The sperm ingested provides a strong stimulus for spawning, and if the females are in spawning condition, this technique routinely results in females releasing eggs and subsequent fertilization
g. Spawning males should be removed quickly from the spawning waters to avoid excessive amounts of sperm
    i. Results in several sperm penetrating a single egg, resulting in "polyspermy" and eggs that do not properly develop
    ii. Just the slightest amount of sperm is sufficient to fertilize all of the eggs
h. Once females begin to spawn they should be isolated
i. Fertilized eggs should be collected on a 20 µm (micrometers or microns) screen
    ii. Thoroughly rinsed to remove excess sperm
    iii. Fertilized eggs are almost neutrally buoyant and require only slight aeration to keep them suspended in the water column

IV. Larviculture
a. The resulting fertilized eggs, now called zygotes, cleave within 45 minutes
    i. The zygote develops into the **trochophore** stage
        1. Multi-celled stage
        2. Motile, non-shelled, ciliated mass that spins around randomly in the water column
b. After about 24 hours, trochophores develop into a D-shaped **veliger**

    i. Shelled, free-swimming larval form
    ii. Straight-hinged
    iii. Still measuring approximately 60 μm and with a rudimentary shell
    iv. Extending out from between the two valves is the **velum** or **velar lobes**, a mass bordered by cilia (looking much like eyelashes) that create a sweeping motion used for swimming and for collecting food particles

  c. The new veligers are stocked in larval rearing tanks at a density of about 10 larvae/ml
    i. For the next several days they will be moved daily to a new clean tank filled with filtered seawater
    ii. Veligers are drained onto fine mesh screens of specific size to retain them
    iii. They are counted, observed under a microscope to detect abnormalities, disease, or mortalities, and then returned to a new tank filled with seawater that has been filtered through a 5 μm filter and disinfected with ultraviolet (UV) light
    iv. Micrcoalgal feed is added to the tank to achieve the appropriate algal cell density based on the number and size of the veligers (usually ranging between 10,000 and 40,000 algal cells/ml)
    v. Larvae actively feed on the phytoplankton for about 8 – 10 days at 28°C

  d. As the veliger reaches this final stage, called the **pediveliger** stage
    i. As the veliger grows to almost 200 μm, the umbo, or raised beak of the shell becomes prominent (it comes to look more like a clam), and the muscular foot which it will use to burrow into the sediment begins to form
    ii. It extends its foot in search of appropriate bottom sediments suitable for **metamorphosis**, also called **settlement** or **set**
      1. If a substrate is detected, the larva will lose its ciliated velar lobes, and its free-swimming lifestyle, burrowing into the sediment to assume its relatively stationary, benthic, adult form
      2. At this stage, a hard clam is about the size of the period at the end of this sentence.

V. Metamorphosis
  a. When the clam larvae reach the pediveliger stage, they are removed from the larviculture tank and transferred to a **setting trough**
    i. The setting trough is shallow (about 30 cm deep by 60 cm wide by 240 cm long, although the exact dimensions and shape can vary) with a false bottom (about 2.5 cm above the bottom)
    ii. Covered in 115 μm mesh screen

iii. Below the screen is a 1” PVC pipe, perforated, running the length of the trough
iv. The pipe is connected to an airlift pump that pumps the water from underneath the screen back to the water surface, creating a downwelling effect
    1. This downward current places the pediveligers in contact with a substrate and facilitates metamorphosis

b. Algal food is added to the setting tray to sustain the larvae for the next 24 – 48 hours as they reabsorb their velar lobes and settle to the bottom
c. The newly set clams remain in the trough for two to three weeks until they will be retained on a 210 µm screen during sieving
d. Other bivalve mollusks (those that do not bury into the sediment) are usually provided a hard substrate for setting
    i. Oysters, for example, will attach to the side of a fiberglass tank and have to be physically removed
        1. Typically, the culturist will provide oyster shell or concrete-coated fiberglass as a substrate
        2. the best approach is to use ground oyster shell (called micro-cultch) so that the newly set oyster (called spat) will be individually attached
    ii. Similarly, mussels, scallops, ark clams, and others will attach to surfaces using fine threads, called **byssal threads** or **byssus**
        1. Culturists often use plastic “onion bags”, polypropylene rope, or some other fibrous material as an attachment surface for these species

VI. Post-set Nursery

a. After the clams have set, they are usually maintained onshore or in land-based systems for a few months before field planting
    i. Land-based nurseries afford the greatest degree of control and protection, however, they are more expensive than field-based nurseries due to higher equipment and utility costs, requirements for producing algal feeds, and access to waterfront property
b. Historically, shallow raceways or trays, with or without substrate, have been used to nursery hard clams, but this method usually employs flow-through systems that use naturally occurring algal feeds rather than cultured algae.

**Figure 3A.** An adult male hard clam releasing sperm into the water.

**Figure 3B.** Stripping sperm from male gonads to add to spawning water.

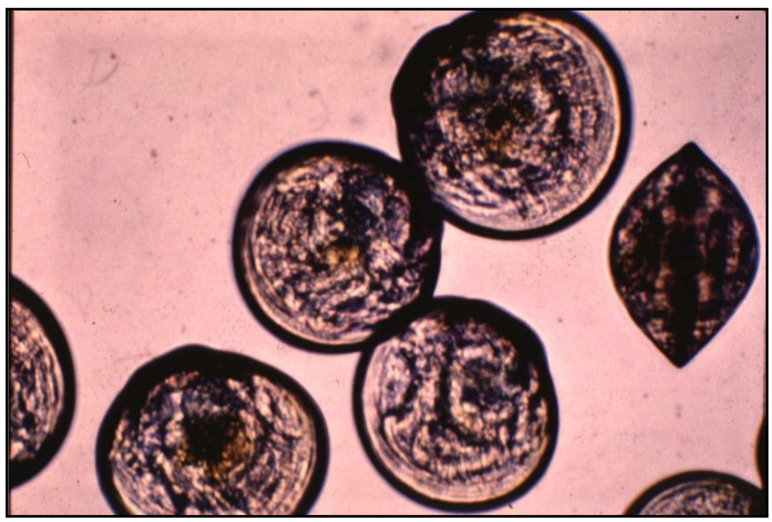
***Bivalve Reproduction***
***Figures (part 2)***

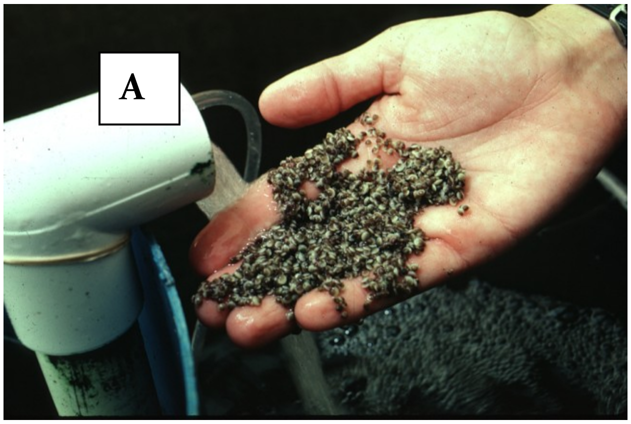
**Figure 6.** Larviculture tank during a daily draindown. Clam larvae are captured on a 54 μm mesh screen, the tank is cleaned, refilled with filtered, UV sterilized water, and then the larvae are returned to the tank. After the draindown the larvae are fed microalgae (phytoplankton).

**Figure 7.** Clam larvae collected on a sieve during a transfer (see Figure 6).

**Figure 8.** Pediveliger clams are transferred from the larviculture tanks to setting raceways. The bottom of the raceway is covered with 150 μm mesh screen, and water is recirculated in the tank to create a downward current (downweller).

**Figure 9.** Ten-day old hard clam veligers with prominent umbo, soon to progress to the pediveliger stage.





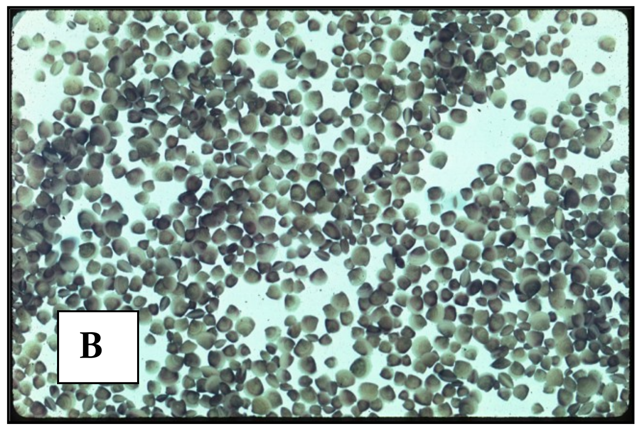



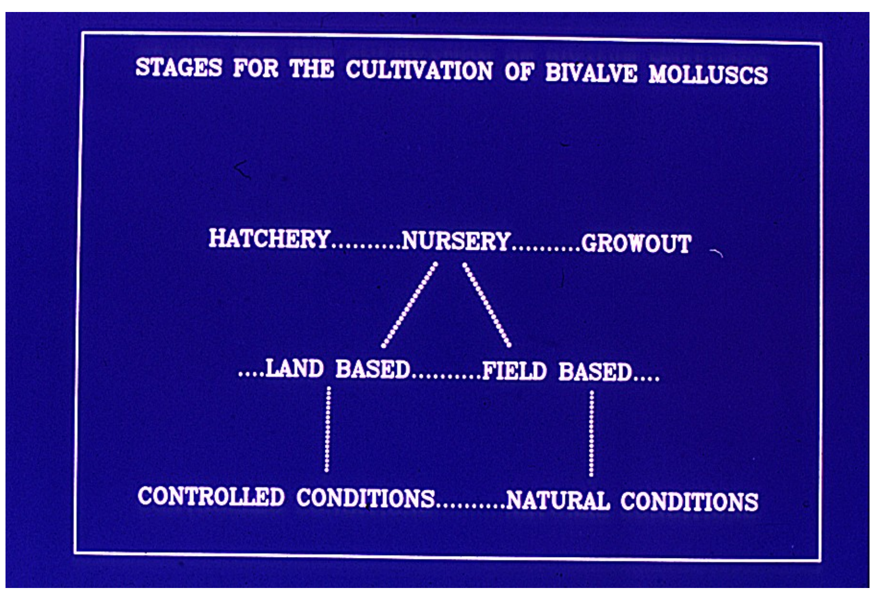


**Figure 10A, B, and C.** Post-set hard clams raised in aland-based nursery and fed cultured algae.

***Bivalve Reproduction***
***Figures (part 3)***

**Figure 11.** Clam nursery raceways using downweller trays. Algal food is added to the raceway and circulated through the downwellers.

**Figure 12.** Stages for cultivating bivalve molluscs from highly controlled hatchery conditions through growout under natural conditions. Variation in production strategies occurs during the nursery phase (land-based vs. field-based), with many operators combining both types of systems.



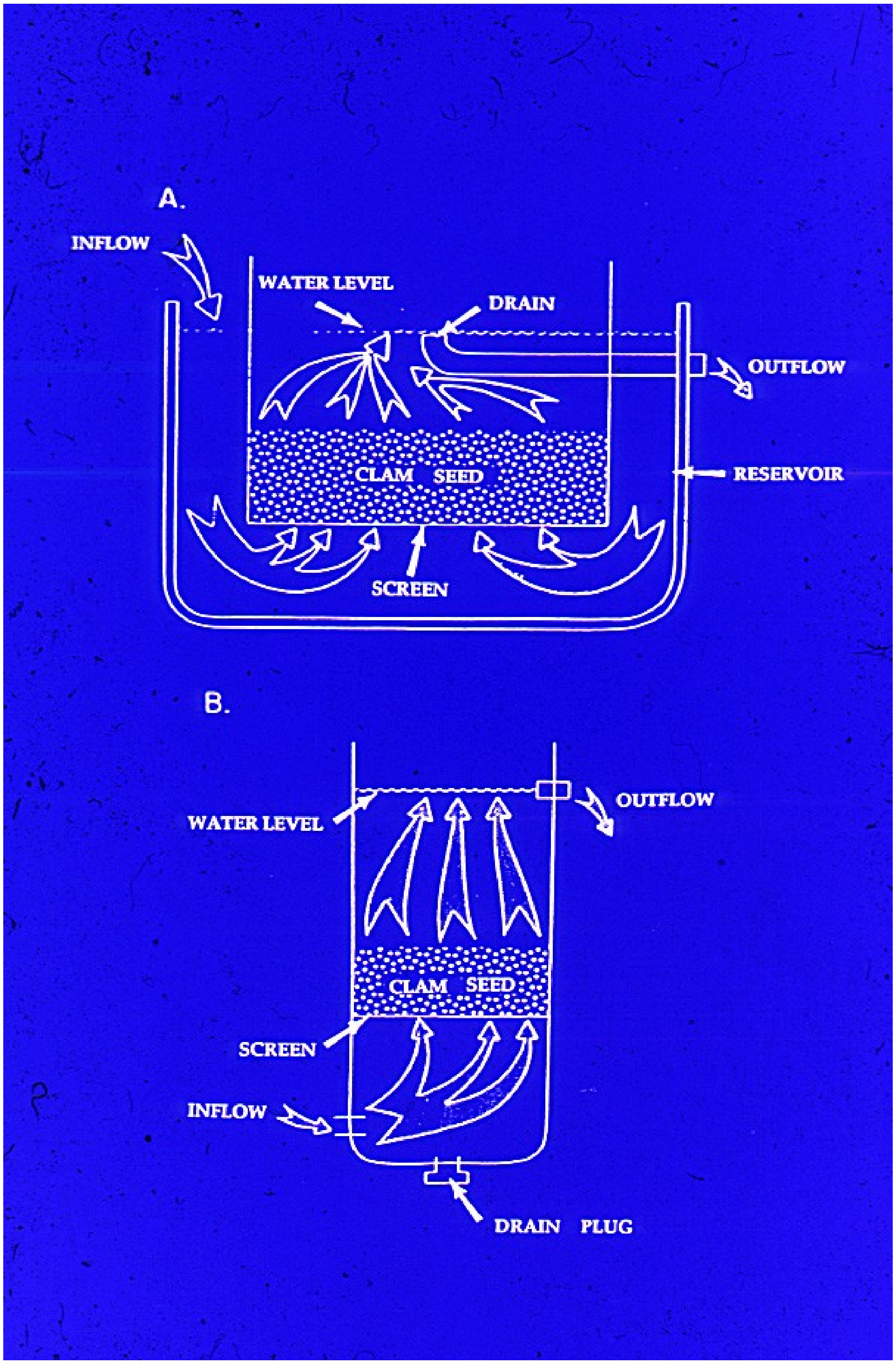



**Figure 13.** Schematic diagram of an upweller.

**Figure 14.** A floating upwelling system (FLUPSYS) is a standard for field-based nursery systems in the United States.