

## Rotifer Production (as a First Feed Item) for Intensive Finfish Larviculture

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Live feeds are an integral component in the cultivation of most marine finfish species during larval stages. The first live feed that has demonstrated acceptability for most marine species, and which can typically be raised on a commercial scale, is the rotifer *Brachionous* spp. While approximately two thousand species of rotifers have been identified, most culturists rely upon the marine rotifers *Brachionus plicatilis* (L-strain) with a size range of 200 to 360  $\mu\text{m}$  and *B. rotundiformis* (s-strain) with a size range of 150 to 220  $\mu\text{m}$  (see figure 1). Rotifers are favored due to their size, their ability to be raised in mass cultures, and the fact that their nutrient composition can be quickly improved through the use of specialized enrichment diets.

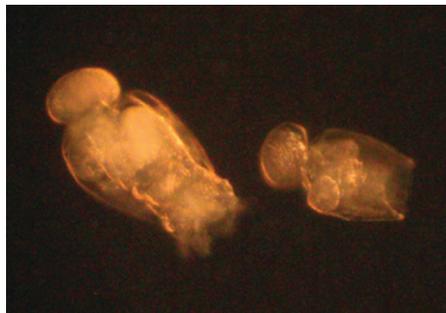


Figure 1: Rotifers

As the initial food source provided for larval marine finfish, rotifers must be clean and nutritious. Rotifer cultures can represent a significant potential vector for the introduction of unwanted pathogens into the fish culture tanks. Therefore, it is important to maintain hygienic production protocols to minimize this risk.

There are two general methods for culturing rotifers: 1) the batch method, where a given volume of water is added or exchanged each day and the culture is restarted at regular intervals (see figure 2); or 2) the continuous method, which often employs recirculation-based technology to increase the density of rotifers cultured while minimizing the need to restart cultures (see figure 3).



Figure 2: Recirculating rotifer production system.

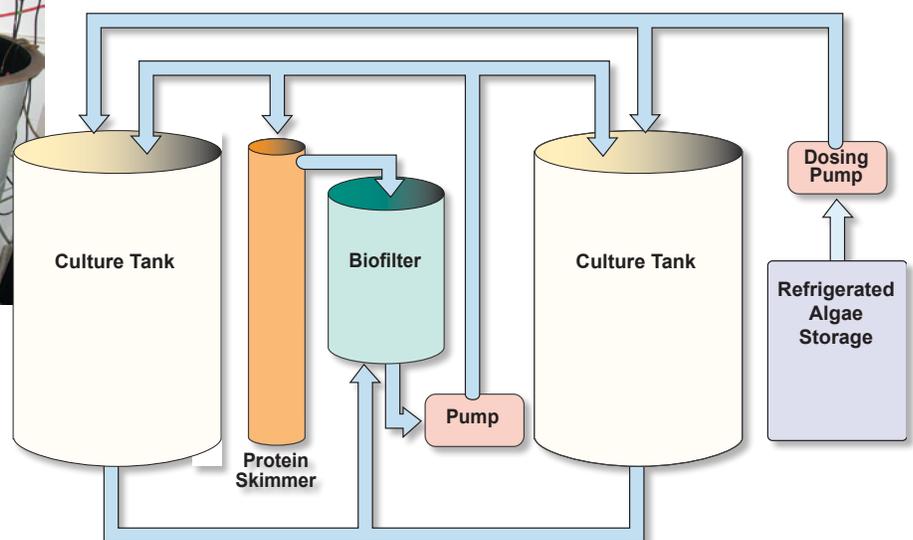




Figure 3: Typical batch culture tank with floc trap pulled out of tank.

## General Culture Requirements

**Temperature:** 22-28°C

**pH:** 7-8.5

**Dissolved Oxygen:** >4ppm

**Salinity:** 10-35 ppt

**Total Ammonia Nitrogen:** ≤5ppm

Stock cultures of rotifers can be purchased from numerous aquaculture supply companies, either as live animals or as desiccated cysts. When purchasing stock rotifers to initiate larviculture, it is important to purchase the correct rotifer size strain for the species to be raised; otherwise larvae may be physically unable to ingest their prey. When starting a new culture, initial stocking densities should be  $\geq 200$  rotifers/ml of culture water. Lower stocking densities will result in delayed start-up time and may help promote the growth of unwanted contaminants. When starting with live animals, take care to minimize changes in water quality when transferring the rotifers from the shipping bags to the grow-out vessel to minimize shock. Specifically, temperature, pH and salinity should be measured and adjusted as necessary. Adding about 10 percent of the shipping water volume to the new culture can help minimize transfer shock. Under ideal conditions, it is not uncommon for a rotifer population to grow by 50 percent each day although a longer-term average might be closer to 20 percent to 30 percent daily growth.

## Batch Culture

A traditional batch culture system typically follows a 4- to 5-day culture rotation. When using this technique, a tank is inoculated with rotifers on day 1. The rotifers are then fed each day, and the volume of the culture is increased to keep up with rotifer growth. Rotifer densities reached with this method are typically on the order of 500 rotifers/ml. At the end of the cycle, most of the rotifers are harvested and fed to larval fish. However, some of the rotifers are saved for the next tank inoculation. The duration of the cycle can be extended slightly by performing regular water exchanges once a terminal density is reached. Removing 10 percent to 30 percent of the water volume on a daily basis can help keep water quality within desired parameters, however, the culture will eventually need to be restarted due to the accumulation of uneaten feed. “Floc traps,” constructed from commercially available scrubbing pads, can be submerged in the tank to help control protozoans and trap large pieces of suspended matter (see figure 2). These should be cleaned and disinfected daily.

## Continuous Culture

Increasingly, recirculating aquaculture technology has come into favor with aquaculturists wishing to maximize rotifer production per unit volume while decreasing labor inputs. By incorporating biological filters and foam fractionators for fine solids control, it is possible to maintain higher levels of water quality, thus increasing the stability of a culture. Ozone injection and UV sterilization can also be employed to provide additional means to control water quality parameters. These systems generally employ automated feeding systems utilizing commercially available algal pastes, which further decrease the amount of labor involved in rotifer culture.

A typical rotifer recirculating system employs a standpipe screened with 55- $\mu\text{m}$  mesh located within the culture tank. The standpipe allows uneaten feed and ciliated protozoans to pass out of the culture tank while keeping the rotifers inside. This waste stream travels through a biological filter and a foam fractionator prior to being injected back into the culture vessel. Flow rates in this type of system are typically two to five tank turnovers per day through the recirculating system. Daily maintenance for this type of system involves cleaning the screened standpipe and floc traps and purging the settled materials from the bottom of the tank. For long-term maintenance, a daily water exchange of 20 percent to 30 percent of the culture volume (either har-

vested for feed or simply purged to a waste line) should be incorporated. With some practice, rotifer densities in a continuous system should easily approach 1,000/ml, however, densities as high as 10,000/ml have been reported.

## Feeding

Rotifers actively graze the water column feeding on particles approximately 1 to 10  $\mu\text{m}$  in size. There are a number of yeast- or algae-based rations suitable for culturing rotifers that are commercially available. Activated baker's yeast has been used successfully as an inexpensive grow-out diet when fed at approximately 0.5g/million rotifers. It is important to note, however, that such a diet is devoid of beneficial amino and fatty acids and also lacks certain vitamins. As a result, the diet may need to be supplemented in order to achieve maximal culture performance.

Over the past few years, the use of concentrated algal pastes has grown. These products are favored due to their availability, high quality and cost effectiveness.

Recommended daily feeding rates for these algal pastes range from 1 to 1.5 ml algae per million rotifers, depending on specific conditions. It is best to spread the daily ration over numerous feedings in the course of each day. The use of a peristaltic pump connected to a digital timer can facilitate this feeding method. If optimal production is desired, rotifers should not be allowed to go more than a few hours without feed.

## Counting and Observations

Each day, collect a sample from the rotifer culture for observation and enumeration. If the culture is well mixed, the area where the sample is collected should not matter. It is important to note that rotifers are phototactic and have the ability to migrate through the water column, so take care to ensure that the culture is well mixed prior to sampling.

Initially, a sample of live rotifers should be examined under a dissecting microscope. Rotifers should appear very active and full of food. Slow or sluggish rotifers may indicate that water quality parameters are not

ideal, and that corrective action is needed. The overall cleanliness of the culture, in addition to the presence or absence of ciliated protozoan contaminants should be noted. While it is common to have some ciliated protozoans in rotifer cultures, they compete with rotifers for oxygen and food. An overabundance of protozoan contaminants often correlates with excessively high levels of dissolved organic matter, a condition that is not ideal for rotifer culture. High numbers of ciliates can be abated by collecting rotifers in a 55- $\mu\text{m}$  harvest bag, rinsing thoroughly and restarting the culture.

One way to count rotifers is as follows. After observing the live sample, collect a 1 ml sub-sample with a pipette and load a Sedgwick-Rafter slide. Then add one to two drops of formalin or Lugol's solution to immobilize the rotifers. Place the Sedgwick-Rafter slide onto a microscope under 40x magnification and count the total number of rotifers and the number of rotifers with eggs. Samples may need to be diluted prior to loading the Sedgwick-Rafter slide if rotifer densities approach 1000/ml. The percentage of rotifers with eggs is a useful indicator of the health of a population. A rotifer population with  $\geq 30$  percent of individuals with eggs is consistent with a healthy culture. A drop in this percentage to 15 percent to 20 percent is still acceptable, but growth of the culture will be reduced. A decrease to  $< 15$  percent often indicates that conditions in the culture tank are sub-optimal, likely due to feed rate or water quality. In this case, corrective action should be taken immediately.

Another option for making a quick estimate of rotifer density involves collecting a sample from the culture tank with a 1.0 ml pipette with 0.1ml gradations. Count the number of rotifers in 0.1 ml and multiply by 10. All counts should be conducted two to three times for each culture and then used to calculate an average.

Regardless of the culture system or feeding protocol chosen for rotifer production, it is important to remember that rotifers are what they eat. While some feeds may result in satisfactory growth, they may not provide a suitable source of nutrition for developing fish larvae. With few exceptions, it is advantageous to enrich rotifers with specially formulated feed products to boost their nutritional profile prior to feeding them to fish larvae.