

HANDLING CULTURES UPON ARRIVAL

Read the enclosed strain information sheets(s). Recommended media and growth temperatures are included. Please see <https://ncma.bigelow.org/algal-recipes> for media recipes used at NCMA. Whenever filtered seawater is used to make marine media, we use our local Gulf of Maine seawater with a salinity of about 32-33 ppt prior to autoclaving.

1. **Examine cultures with a microscope**, preferably a compound microscope so that the health of the cells can be assessed.
2. **Allow cultures to equilibrate** at their recommended growing temperature. They should be exposed to some light immediately.
3. **Subculture**. Between 5ml to one half of the biomass from each shipping vessel should be removed and subcultured into fresh medium within a day. Do not dilute the culture too much at first, but rather use a 2:1 to 5:1 dilution of fresh medium to received algal culture. Use medium light to start unless otherwise indicated.
4. **Keep some of the received culture in the original shipping vessel and medium** in case the new water or medium is not suitable to sustain growth.
5. **Monitor your cultures closely** until you determine the growth rate. They may recover quickly and start growing or decline before you are aware

DO NOT place in refrigerator.

DO NOT leave unattended.

We DO NOT recommend 24 hour continuous light exposure. Phytoplankton are photosynthetic organisms and adapted to the natural rhythm of day and night. Some organisms will tolerate continuous light, but we maintain ALL our strains on 13:11 light dark cycle. We use cool white fluorescent light bulbs and our cultures are exposed to light intensities between 40μEinstiens to 120μEinstiens according to need. Upon receipt of strains from the NCMA, you will have to discover empirically if your strain of interest will tolerate continuous light.

We do not grow any of our smaller cultures (less than 100mL) using any means of agitation.

▶▶▶▶▶ Tip: *Aliquot the subcultures into a few vessels that can be exposed to various light levels.* ◀◀◀◀◀

Cultures are shipped when they are healthy and robust. No cell counts are performed. Specific cell densities cannot be guaranteed. There is always cell death in transit. Some strains travel well, and arrive in good condition. Most likely, they can be used as soon as fresh growth is detected. Other strains do not travel well and can take more time to recover before they can be used. Our goal is to have cultures arrive alive and in a condition that there will be enough live cells to use as a starter inoculum.

REPLACEMENT POLICY

If both tubes did not survive shipment with our shipping company, we will re-ship a starter culture for the price of shipping and handling only (the culture will be free). However, we must be notified in writing that it did not survive within two (2) weeks of delivery. Cultures will be re-shipped only once. We will only reship if BOTH tubes are dead on arrival. Kindly note that this policy is **VOID** when the customer/recipient uses their own shipping company, the order is shipped via ground service, the cultures are moved to a location other than its shipped location and/or there is a delay in the shipment (the order is held in customs, etc.)

Before making arrangement for a reshipment, we need to have the following questions answered so that we may have more success at you receiving your strains alive. Please send us an e-mail with your order number referenced, the strain numbers listed, and your answers to the questions below:

- a. Transit & Shipping: Is it possible that the cultures overheated or froze in transit or while sitting on a loading dock? Are you aware of any delay in delivery of the strain?
- b. Please tell us what you did with the cultures(s) upon arrival.
- c. Did you examine them with a microscope when you received them?
- d. When did you subculture them?
- e. Please tell us what medium, temperature and light conditions you are using to grow the culture. Please provide us with the specifics of the media you used, including salinity.