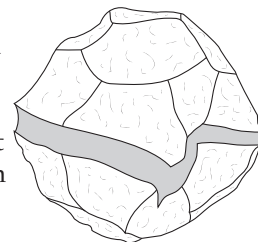


# Culturing *Peridinium* and Other Dinoflagellates

## Background

*Peridinium* is a freshwater dinoflagellate found in lakes, ponds and pools. These unicellular protists are frequently used in cell biology research areas of nuclear structure and function, circadian rhythms and endosymbiosis. They are circular-oval shaped and range in size from 0.01 to 0.1 mm in diameter. *Peridinium* range in color from green to yellow-brown. Few species of *Peridinium* are purely photosynthetic. Instead, most species are mixotrophic, supplementing autotrophy with heterotrophy.

Dinoflagellates, freshwater and marine, are environmentally significant as they are the primary food source for many aquatic organisms. However, they can also be problematic as parasites to certain fish or other protists. In the spring and summer months, species will reproduce in mass quantities causing the water to appear gold or red. During “red tides,” dinoflagellates release a neurotoxin that affects muscle function in susceptible organisms. Humans that consume fish or shellfish that contain the neurotoxin may suffer from ciguatera or PSP, paralytic shellfish poisoning, both serious conditions that are not typically fatal.



## Culturing/Media

Upon arrival of *Peridinium*, loosen the lid and aerate with a pipet to replenish any oxygen lost during shipping. *Peridinium* is ready for use immediately and will show best results if used within 2–3 days. However, with minimal care, the culture should last two weeks, but will degrade over time. Keep *Peridinium* at room temperature, 16–22 °C, under a full spectrum fluorescent light with a 16-hour light and 8-hour dark cycle. The light source should be 18–24" above the culture. If the dinoflagellates are bioluminescent, then use a regimented 12-hour light and 12-hour dark cycle to elicit the natural circadian rhythm of the protist.

Maintain autotrophic *Peridinium* cultures in clean jars or containers with Pringsheim's Soil-Water. See below for preparation instructions for Pringsheim's Soil-Water. Algae culture media is also available from Flinn Scientific. If light is limited to induce and observe heterotrophy, use Hay Infusion media to introduce bacteria to the water and provide a food source for the *Peridinium*. The directions for preparing Hay Infused medium are below. *Paramecium* medium is also available from Flinn Scientific.

## Pringsheim's Soil-Water

Select a rich garden soil that has not been recently fertilized. Add 300 g of the soil to a heat-resistant gallon jar or 2000-mL beaker. Fill  $\frac{3}{4}$  full with distilled or deionized water and then add one gram of calcium carbonate ( $\text{CaCO}_3$ ). Loosely plug or cap the jar and heat the mixture on a hot plate until it steams. Allow the mixture to steam for about one hour and then cool to room temperature. Repeat this procedure for three consecutive days. *Note:* To culture diatoms, add 10–30 mg of sodium metasilicate ( $\text{Na}_2\text{SiO}_3 \cdot 9\text{H}_2\text{O}$ ) to every liter of medium.

## Hay Infusion

Boil 10 g of chopped Timothy hay (other types of hay will also work) in one liter of spring water for about 30 minutes. Filter the boiled hay mixture through several layers of cheesecloth. Allow the solution to cool and then add 2 drops of 1 M NaOH and a pinch of black soil. Pour 200 mL of the liquid medium into each culture dish (stacking culture dishes works best) and let sit for 24 hours uncovered.

Either boil two wheat seeds in a few mL of water for 15 minutes or dry lettuce leaves slowly in an oven until crisp. Crush the lettuce with a mortar and pestle. Store extra dried lettuce in a tightly sealed container for future use. Add the wheat seeds or 1–2 g of crushed dried lettuce to each dish.

### Tips

- Large culture jars should be cleaned with soap and water before using. The bottom half of clear plastic two-liter bottles also work well for housing dinoflagellate cultures.
- Do not house cultures in windows for light. The extreme temperatures may cause the cultures to perish.

### Disposal

Peridinium cultures may be disposed of according to Flinn Suggested Biological Waste Disposal Method Type I. Please consult your current *Flinn Scientific Catalog/Reference Manual* for proper disposal procedures.

**Materials for *Culturing Peridinium and Other Dinoflagellates* are available from Flinn Scientific, Inc.**

Catalog No.	Description
LM1251	Peridinium, Dinoflagellate
FB1820	Algae Culture Media
FB0540	Timothy Hay, 100 g
FB0541	Wheat Seed, 100 g
FB0570	Cheesecloth, 4 sq. yards
FB0514	Paramecium Medium, 946 mL
ML1381	Microscope Slides, Glass
ML1385	Coverslips, 22 × 22 mm
MS5040	High School Compound Microscope

Consult your *Flinn Scientific Catalog/Reference Manual* for current prices.