Title of Lesson - Growing Algae<br>Target Grades - High School Biology

Time Required: 30 minutes of student set up time, 1-3 weeks of growth, $30-45$ minutes of data collection

## Alignment to Standards:

BIO1.LS1:8 Create a model of photosynthesis demonstrating the net flow of matter and energy into a cell. Use the model to explain energy transfer from light energy into stored chemical energy in the product.
BIO.LS2:1 Analyze mathematical and/or computational representations of population data that support explanations of factors that affect population size and carrying capacities of populations within an ecosystem. Examine a representative ecosystem and, based on interdependent relationships present, predict population size effects due to a given disturbance.
BIO1.LS2:2 Create a model tracking carbon atoms between inorganic and organic molecules in an ecosystem. Explain human impacts on climate based on this model.
BIO1.LS2:3 Analyze through research the cycling of matter in our biosphere and explain how biogeochemical cycles are critical for ecosystem function.
BIO1.LS4:3 Identify ecosystem services and assess the role of biodiversity in support of these services. Analyze the role human activities have on disruption of these services.

## Materials Needed

Algae growth medium
Green Algae
Containers to grow algae
Light Source

## Optional materials for testing different variables

Colored film or plastic to filter light
Fertilizer with differing nutrient concentrations
Light bulbs of different wattages
Multiple different algal species
Additional materials will be needed depending on the method chosen for growing and measuring the algae.

## Background Information

Algae are for photosynthetic aquatic organisms. The term "algae" consists of many protists, but also cyanobacteria. They are not plants as they do not have true roots, stems, or leaves. Algae include many single celled organisms, but also large multicellular forms like kelp.

Algae are an important part of the carbon, nitrogen, and oxygen cycles. They are also an important source of food and are the base of many food cycles. They make up about $40 \%$ of global net primary productivity.

Algae require water, nutrients, space, and air in order to grow. Deficiencies in any of these areas can limit the growth of algae.

Algae are currently being used for biofuel production. Oil extracted from algae can be refined into biofuels.

Potential Lesson Objective - May vary depending on how the lab portion is designed

Students will be able to describe limiting factors for a population of algae.
Students will be able to design an experiment to answer a question.
Students will be able to collect and analyze data to draw a conclusion.
Students will be able to determine how light affects photosynthesis.
Students will develop an awareness of algae as a source for bioenergy.

## Instructional Process

## Introduce Problem

Fossil fuels like coal and oil that are currently used by the United States for energy create harmful emissions that pollute our air and can damage the environment. To mitigate these destructive emissions, Department of Energy scientists are developing new methods of producing energy. One such method is the use of algae to create biofuels.

Algae are protists that produce energy through photosynthesis. That energy is stored in the algae as oils. The oils can be extracted and refined to create biofuel.

This process is not widely used yet and there are still many challenges to overcome before algal biofuel can be commercialized. Crops must be protected from grazers and pests and provided with proper nutrients and water supply. The cost of algal biofuel is high. To make the best use of resources, scientists are working to maximize the amount of algae that can be grown in a pond. Increasing algal biomass increases the amount of oil and thus the amount of biofuel that can be created.

Your task is to grow the most algae with the resources provided so that you would be able to create a maximum amount of biofuel.

## Perform Lab Activity

The basic procedure for growing algae involves choosing a container, media for growth, and algae. Put an equal amount of media in each container. Add an equal amount of algae into each
container and place the algae near a light source. Change only 1 variable for each container of algae. Aerating your algae will help it to grow.

There are a number of procedures for growing algae available online. Here are two options:

Donae College- Resource for Growing Algae to Make Biofuels:
http://doanealgae.wixsite.com/algaegrowingresource/lessonplans
Carolina Biological Supply- Culturing Algae: $\underline{\text { http://www.carolina.com/teacher- }}$
resources/Document/culturing-algae-instructions/tr29112.tr
Once an algae growth method is chosen, students will set up a culture of algae and test a variable to increase the biomass of algae that can be grown in a specific container. Algae can be grown for 1-3 weeks and then measured. There are many ways to vary this lab to make it fit any lab situation or budget.

Algae source: Algae can be harvested from a pond. If the pond has visible green algae floating on the top, simply dip out a sample with a bottle or cup. Shake the stock solution of algae to evenly distribute the cells before pipetting a sample into student growth chambers.

Algae can be purchased from a supply company. Chlorella, Chlamydomonas, and Scenedesmus are recommended species because they are large enough to be easily observed under a microscope.

Container options: Erlenmeyer flasks, plastic water bottles, petri dishes, test tubes, etc... almost anything that can hold water can grow algae. Remove labels to allow light to enter.

## Media options:

Media can be purchased from a supply company.
Algae can be grown in pond water or fish tank water.
Media can be made using Miracle-Gro.
Media can be made using stock chemicals.

## Aeration Methods:

Fish tank aerator
Shake the container
Place the containers on a rocker

## Light Source:

Cool-white fluorescent lights are recommended for growing algae because they don't generate heat. Fluorescent bulbs handing about 18-24" above the algae are ideal. There are a variety of instructions for constructing a light bank available online.

## Variables to test:

Amount of light - students can place algal cultures at several distances from the light source Fertilizer concentration - amount of nitrogen in the media can be altered
Light color - light can be filtered using colored plastic
Monoculture vs polyculture - students can grow more than 1 species of algae in the same culture
Amount of aeration - students can vary length or frequency of aeration

## Measurement Options:

Count cells using a hemocytometer.
Measure cell density by measuring light absorbance using a spectrophotometer or colorimeter.
Harvest algae, place on a coffee filter, and dry on the lab bench or in an oven. Mass dry algae.

## Analyze Data

Data analysis will vary based on the variable students have tested. Students should graph data with the variable tested on the $x$ axis and the algal growth (\# of cells, optical density, or mass) on the $y$ axis.

Sample analysis questions:

1. What can you conclude about algae growth based on your results?
2. What resources did the algae require for growth?
3. What would you try next time to increase the biomass produced?
4. Chlorophyll absorbs red and blue wavelengths of light and the rest of the light is "wasted". Predict what would happen in algae were grown under red light only. Predict what would happen if algae were grown under green light only.
$\qquad$ Date: $\qquad$

## Growing Algae Lab

## Background:

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To measure the algae population, you will use a colorimeter. Light will pass through the sample of algae and strike a photocell. This will measure the absorbance of the sample. Algae cells will block the photons from reaching the photocell so that more light will be absorbed. The absorbance value is proportional to the population of algae in the sample.

## Materials:

Alga-Gro Freshwater Medium from Carolina Biological
Screw top culture tubes
Test tube rack
Green algae mixture
Plastic micropipettes
3 Light banks with fluorescent tube lights
Tape measure
Vernier LabQuest and Colorimeter
Cuvettes

## Procedure:

1. Measure 8 mL of medium into each of 4 culture tubes.
2. Label each culture tube with a different light distance: 6 inches, 18 inches, 30 inches, dark
3. Gently swirl the green algae mixture to evenly distribute cells. Collect .25 mL of the mixture and add to each of the culture tubes. Place the cap on loosely to allow for air flow in the tube.
4. Measure the initial population of algae by using the colorimeter to measure absorbance of your solution. See procedure below.
5. Place 1 test tube rack 6 inches from the light source. Place the second test tube rack 18 inches from the light source, and the $3^{\text {rd }}$ test tube rack 30 inches from the light source. Place the final test tube rack inside of the lab bench, in the dark.
6. Place the properly labeled culture tube in the appropriate test tube rack.
7. Observe growth in the tubes for 7 days.
8. On the $7^{\text {th }}$ day, measure the final population of algae using the colorimeter and the procedure below.

## Measuring Absorbance Using the Colorimeter:

1. Prepare a blank by filling a cuvette with 4 mL distilled water. To correctly use a cuvette, remember:
a. Wipe the outside of each cuvette with a lint-free tissue.
b. Handle cuvettes only by the top edge of the ribbed sides.
c. Dislodge any bubbles by gently tapping the cuvette on a hard surface.
d. Always position the cuvette so the light passes through the clear sides.
2. Connect the Colorimeter to the LabQuest.
3. Calibrate the Colorimeter.
a. Open the Colorimeter lid.
b. Holding the blank cuvette by the upper edges, place it in the cuvette slot of the Colorimeter. Close the lid.
c. Press the < or > button on the Colorimeter to select a wavelength of 635 nm (Red) for this experiment.
d. Press the CAL button until the red LED begins to flash, then release. When the LED stops flashing, the calibration is complete.
4. Put 4 ML of distilled water into a second cuvette. Add 3 drops of algae solution from the culture tube.
5. Invert the cuvette to evenly distribute algae cells. Place the cuvette in the Colorimeter and close the lid. Allow 10 seconds for the readings displayed in the meter to stabilize. Record the absorbance value.
6. Repeat steps $3-5$ for all algae samples. Be sure to use the blank between each sample to calibrate the colorimeter.

## Extension:

Now that you know how to grow algae and measure the algal population, your challenge is to grow the largest amount of algae possible in 1 week. You may use any available materials in the lab. Each group will begin with 0.25 mL of the algae solution and you may adjust any other variable. Do some research to see what variables you may want to change. Good luck!

## Using Miracle Gro to make Algae Media

There are several methods for using Miracle Gro as media for your algae. The method you will use depends on why you are growing algae. If algae is being grown to use as stock, to feed fish or to observe, option 1 is fine. However, if you need to be more precise in order to keep the nutrient concentration equal between different samples, option 2 or 3 should be used.

## Option 1:

Fill each bottle/flask about $3 / 4$ full with distilled water. Add a pinch of Miracle Gro. Stir to mix. Add another pinch of Miracle Gro about every week.

## Option 2:

Measure 350 mL of water. Add $1 / 16$ tsp of Miracle Gro. Stir to mix. Pour into bottle or flask that will be used to grow algae.

## Option 3:

From http://doanealgae.wixsite.com/algaegrowingresource/lessonplans

1. Weigh out 9.0 grams of Miracle-Gro All Purpose Plant Food.
2. Add the Miracle-Gro to 1 gallon of water in a sealable, shakable container. Note: make sure the container is clean to prevent microbial growth.
3. Shake the gallon container vigorously until no solid particles remain.
4. Using a 1 M strong base solution and pH strips or a pH probe, bring the pH of the medium to neutral ( pH 7 ). (We use about 8 mL of 1 M KOH )
