"Killer Algae", how do we manage it?

Designed for 10th and 11th Grade

Developer:

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National Science Standards:

Grades 9-12

Content Standard C: The interdependence of organisms

-Organisms both cooperate and compete in ecosystems

-Living organisms have the capacity to produce populations of infinite size

-Human beings live within the world's ecosystems. Human destruction of habitat through pollution is threatening global stability and if not addressed, ecosystems will be irreversibly affected. Invasive species are a type of pollutant.

California Science Standards (many state standards are very similar)

Grades 9-12

Ecology 6, b: Stability in an ecosystem is a balance between competing effects. Students will know how to analyze changes in an ecosystem resulting from changes in human activity and introduction of non-native species.

Investigation and Experimentation 1: Scientific progress is made by asking meaningful questions and conducting careful investigations. Students should develop their own questions and perform investigations. They will:

a. select and use appropriate tools and technology to perform tests, collect data, analyze relationships, and display data.

b. identify and communicate sources of unavoidable experimental error.

c. identify possible reasons for inconsistent results, such as sources of error or uncontrolled conditions.

d. formulate explanations using logic and evidence.

Underlying concepts:

1. Invasive species can negatively impact local ecosystems and economies.
2. Students will understand design and execution of experiments, as well as be able to explain their results.

Overview:

Students will learn content regarding *Caulerpa taxifolia* and other invasive species. The teacher will assign small groups of students sections of background reading that the groups will learn and peer teach to the entire class. Based on their background knowledge, students will identify threats posed by *C. taxifolia* to their local coastal ecosystem. Background reading will center on *C. taxifolia* and its history, biology, geographic distribution, invasive characteristics, human spread, economics, and control/eradication efforts. Students will hypothesize what will happen with different methods of control, and/or different concentrations of control agents. The students will learn to make hypotheses and set up experiments to test those hypotheses. Each group will be assessed on their group participation and clear presentation of their results from the experiments. They should support their results with why they think what happened (including unavoidable experimental error and sources of error when carrying out their experiment) and what would be the best choice for eradication based on their results and the supporting information (see resources section for further information on eradication from researchers in Australia). The teacher should read the content information to be familiar with the material.

Background:
Caulerpa taxifolia, a green marine alga (seaweed), has invaded southern California, southeastern Australia, and the Mediterranean Sea. In 2000, the alga covered over 10,000 hectares in the Mediterranean and has been dubbed "killer algae" because of its major ecosystem and financial impacts in the Mediterranean. The Mediterranean populations are too large to eradicate, while millions of dollars are being spent in the United States (using chlorine and heavy, black tarps) and Australia (using rock salt, reducing salinity, copper sulfate) to eradicate the alga. In the spring of 2005, the eradication of *C. taxifolia* in southern California was declared a success; however the invasion site is still being monitored for new populations. All three invasions are believed to have been introduced by the release of the alga from saltwater aquarium tanks.

As is typical of invasive species, *C. taxifolia* grows quickly, can tolerate a wide range of physical conditions (temperatures, depths, salinities), and has effective methods of dispersal via small fragments, which allow it to displace native flora and decrease species abundance and diversity. In these invaded sites, there is a lack of natural predators and diseases that could curb its spread. It disrupts ecosystem function and is very costly to control or eradicate. There is also an economic loss in areas where the sites have been closed to recreational or commercial boating or tourism declines due to reduced biodiversity.

Setting:
Classroom/laboratory

Objectives:
Students will be able to:

1. Describe how changes from an invasive species introduction, in this case *Caulerpa*, have affected biodiversity, ecosystem function, and economic cost.
2. Construct hypotheses as to which control method or different concentrations of control methods will work best and why. Students can compare different methods of control and/or different concentrations of one control method.
3. Synthesize background information with experimental outcomes. Students will support or refute their hypotheses and explain their results with supporting background information.
4. Communicate their control solution to their peers and substantiate their solution with the supporting background information and material obtained through the resources section.

5. Debate and defend their decision and reasoning.

Prerequisite Skills:
Good reading and comprehension skills, background in biology

Skills Used During Project:
Reading, presentation, experimental design, hypothesis construction, problem solving, synthesis of information and experimental results.

Curriculum Connections:
Science and Reading Comprehension

Time Needed:
2.5 weeks

1. Students work on background information, create hypotheses, and set up experiments (2-3, 55-min class periods).
2. Experiments will take 7 physical days to run. Students will need to make observations and measurements every day or every other day (0.25-0.5 hours per observation). Experiment will run over a weekend and students will not do observations on those two days.
3. Students finish experiments and synthesize information (2, 55-min class periods).
4. Students present to class (2, 55-min class periods).

Teacher Preparation:

1. Read all information.
2. Decide what control agent(s) you will use (see materials). Note: This experimental design can be replicated for various control methods (chlorine bleach, thick plastic or layers of screen for shade, rock salt, copper sulfate). The teacher decides the 3 different concentration/treatments to use (e.g. no shade, a little shade with some screens, or total darkness with thick plastic). Concentrations and instructions are written out in detail for the chlorine bleach treatment.
3. Create a spreadsheet for students to record data (or have the students create a spreadsheet).
4. Collect/purchase all needed materials.
5. Divide class of 30 students into 3 groups of 10. Then split each group of 10 in half. During class you will give each half a separate section of background material to read, comprehend, and then teach to the other half of their group.
6. Split the background material (Attachment A) in half how you see fit (there are seven sections).
7. Create 3 sets of background information for the 3 groups of 10 students.

Vocabulary:
Ecosystem: A community of organisms that function together as a unit with their environment.
Native: An organism that is from the area you are working.

Non-native: An organism that is not from the area you are working.
Invasive: An organism that is not from the area you are working and causes problems in the new ecosystem.
Disperse: The movement of something to a new area (e.g. fragments of seaweed).
Eradicate: Completely remove an organism from an area.
Control: Keep populations of an organism at a constant or lower density than their current levels.
Clone: A genetically identical individual.

<u>Materials</u>:

All experiments use live specimens, so all materials used (e.g. plastic tubs, forceps, etc.) must be free from chemical contamination.

1. Purchase mixed bags (2-3) of live green algae from a biological supply company or saltwater aquarium store. This should be sufficient for a class of 30. (One bag costs approximately $10-$15.)
2. Keep the seaweed in 23 to 25ºC water that is aerated, circulated, or filtered with natural or artificial seawater and a light source. (4 gallons of seawater costs approximately $10.)
3. A minimum of eighteen small buckets/tubs or 50 mL beakers that can be filled with seawater. You will need at least 15, plus 3 buckets for the different concentrations of one control agent. You will need more containers if using more than one control agent.
4. Filtered natural or artificial seawater. You can filter the seawater with 0.45 micron filters within 24 hours of the experimental set up. Filtering may not be necessary with artificial seawater.
5. Air stones to aerate each beaker/tub (at least 15).
6. Paper towels.
7. Rulers (mm).
8. Razor blades (single-edge) or scapels.
9. Forceps.
10. Permanent markers.
11. Control agent(s) (thick plastic or screening for shade, chlorine bleach, rock salt, copper sulfate)

<u>Procedure</u>:

1. Divide class of 30 into 3 groups of 10. Then split each group of 10 in half. Give each half a separate section of background material to read and comprehend (Attachment A). Split the background material in half how you see fit (there are seven sections).
2. Each group of 5 will explain the information they have just read to the other 5 students in the group. This way, all students will learn about all aspects of *C. taxifolia* without having to read everything themselves.
3. Each group of 10 will then come up with questions to ask the other groups. This can be a quiz facilitated by the teacher. The teacher or students can make each question worth a certain amount of points and the students will see who can get the most points.

4. Divide each group of 10 students into small groups (3-4 students).
5. Using the small groups, hand out the experiment they will set up (Attachment B). Several groups or all groups can work on the same experiment, so there will be replicates. Alternatively, each group can test a different control method. The design is flexible.
6. Students will first develop hypotheses, and then set up experiments, and write out initial observations of their seaweed before and right after initial treatment. They will track each fragment throughout the week and into the next if necessary. The teacher or students should create a spreadsheet to keep detailed notes about their experiment. These observations will be the data that they use for their results and may help explain any unexpected results (e.g. unavoidable experimental error).
7. Each group should use background information and resources section to help put their results in a larger context. If they were managing an invasion site, what would they do?
8. Students will document change, if any, in the seaweed on a daily or every other day basis throughout the experiment.
9. Using their new background knowledge and results from their experiments, the students will decide what is the best control method or what is the best concentration to use for a specific control method.
10. Students must then consider and describe what they believe will happen if the alga is allowed to grow and thrive in coastal waters. What are the ecological and economic costs?
11. Discussion Questions:
    1. What are some of the challenges facing the aquarium industry and regulatory agencies?
    2. What organizations can you go to for more information on this issue?

Application:
Learn effects and impact of *C. taxifolia* and current control methods, as well as a broader understanding of invasive species. In addition, hypothesis generation, experimental design, and the ability of work in groups and present what they've learned will enhance students' knowledge of the scientific process, cooperation and presentation skills.

Evaluation:
Final completed presentation of research findings, as well as participation within their group. The teacher can also score the quality of the quiz questions created and responses by the groups.

Resources for *Caulerpa taxifolia*:
Facts:
http://swr.nmfs.noaa.gov/hcd/caulerpa/factsheet203.htm
http://www.caulerpa.cjb.net/

Literature Review:
http://www.sbg.ac.at/ipk/avstudio/pierofun/ct/caulerpa.htm

Invasive aquatic plants in the United States:
http://aquat1.ifas.ufl.edu/seagrant/cautax2.html

Prevention Program for United States:
http://www.anstaskforce.gov/Caulerpa.htm

Australia: Eradicating and preventing the spread of the invasive alga *Caulerpa taxifolia* in NSW. Gives a long discussion of different control methods.
http://www.deh.gov.au/coasts/imps/caulerpa-taxifolia/index.html

Video on *Caulerpa taxifolia*:
"Deep Sea Invasion" NOVA/PBS, 1 hr, grade level: 4+, ISBN: 1-57807-980-2, available on line from NOVA for $19.95 plus s/h
http://www.pbs.org/wgbh/nova/algae/
http://shop.wgbh.org/webapp/wcs/stores/servlet/ProductDisplay?productId=13334&storeId=11051&catalogId=10051&langId=-1

Southern California *Caulerpa* Action Team
http://www.sccat.net/

Attachments:
Background Information (A) and Experimental Design (B)

Extension:

- Have your students investigate further the response and control/eradication efforts in the United States and Australia. They can learn how the two countries have dealt with the infestations, as well as their obstacles and successes.
- Conduct further research on the aquarium industry in the United States and learn how managers are working on controlling other invasive species that have been released through this trade as well as education programs for the public about these problems (http://www.habitattitude.net/).
- Conduct further trials using the controls that were not tested during the class experiments.

Attachment A: Background Information

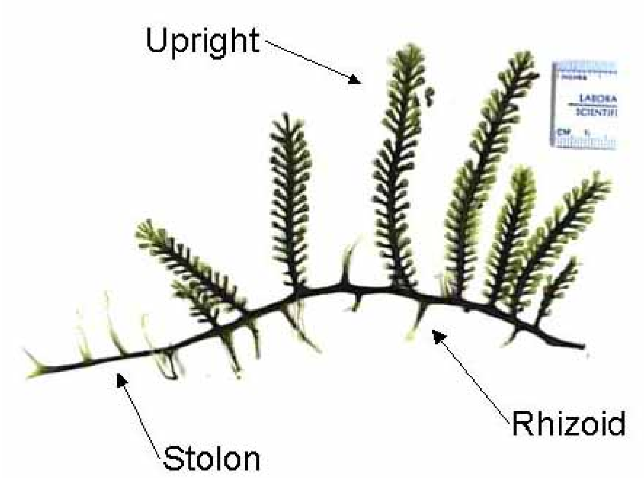
History:

Caulerpa taxifolia was first discovered in the Mediterranean Sea in 1984 as a 1-meter square patch and was believed to have been accidentally introduced from the Monaco Oceanographic Museum. *Caulerpa taxifolia* was noted as a tropical alga, so experts did not think the alga could survive the winter in the Mediterranean Sea. However, they were wrong. By the end of 2000, the alga covered over 10,000 hectares. In 2000, the alga was also discovered in two locations in southern California as well as near Sydney, Australia. The three newer introductions are believed to have occurred from someone dumping their personal aquarium tank into a storm drain or directly into the water bodies.

By the time funding was secured, *Caulerpa taxifolia* covered too much ground in the Mediterranean for authorities to try to eradicate it. Australia came to a similar conclusion when two of their invasion sites had *C. taxifolia* covering 2 and 10 hectares. They began aiming for controlling the population. In the United States, the infestation was much smaller and a rapid response team began working to eradicate the alga. Southern California invasion sites were considered successfully eradicated in the spring of 2005. This will be only the second successful eradication ever of a marine organism.

Biology:

Caulerpa species are green unicellular (one giant cell), multinucleate, macroalgae (seaweed, not microscopic) that have multiple uprights (fronds) from a long stolon. Rhizoids (root-like structures) grow from the stolon (runner) and anchor the alga on rocks, sand, or mud. *Caulerpa* species colonize new areas by extending their stolons to new substrates and producing new rhizoids and uprights. They also spread by vegetative fragmentation. Vegetative fragments are small pieces of the original alga that are somehow broken off the alga, can plug their wounds, and form clonal individuals in new locations. Fragments are frequently created by water motion, herbivores or humans (see below). Pieces of *Caulerpa* as small as 10 mm can break off, travel in the water currents, settle in a new area and rapidly produce new rhizoids and stolons, making invasive growths of this genus especially difficult to contain. Asexual reproduction via vegetative fragmentation is the primary way that *C. taxifolia* spreads once it has been introduced. *Caulerpa* species do have sexual reproduction. However, evidence suggests that all invasive populations, thus far, are only reproducing by fragmentation and are considered one large clone.



Caulerpa racemosa showing uprights (fronds), stolon, and rhizoids.

Geographic Distribution:
Caulerpa species are found throughout tropical waters worldwide and some species also occur in temperate (colder) waters. *Caulerpa taxifolia* was initially believed to be exclusively a tropical species, but it is found in temperate waters as well. The invasive strain of *C. taxifolia* has been linked to a population from eastern Australia that experiences cold winter temperatures. Growing in both warm and cold temperatures, this alga is able to successfully invade a wide range of areas.

Invasive Characteristics:
Invasive strains of *Caulerpa taxifolia* are different from non-invasive strains because they are larger, grow in higher densities, grow deeper in the water column, and form thick mats or carpets along the sea floor. This form of growth outcompetes native plant and algal species, which decreases biodiversity and alters ecosystem function. By out-competing native plants, especially seagrasses, *C. taxifolia* removes food sources for native animals as well as removing their hiding places in the native flora.

Caulerpa taxifolia is similar to other invasive species because it can tolerate a large range of salinity, temperature, and sunlight. For example, it can survive for 10 days out of water on a boat anchor.

Caulerpa taxifolia from an aquarium store (scale bar = 2cm).

Human spread:
Species of *Caulerpa* are widely available in saltwater aquarium stores and through internet outlets (retailers and auctions). They are used in aquaria for their beauty as well as to clean the water of the aquarium tanks (they take up nitrogen excreted by fishes). As more seaweed is sold, there is a higher likelihood of more introductions by people who want to discard excess growth from their tanks or "set their pets free"* directly in the ocean or the bay that flows into the ocean. Their tanks may contain species of *Caulerpa*.

If a boater anchors in a *Caulerpa* meadow and then moves to a new location, fragments of *Caulerpa* may be created and then transported by the anchor. Surprisingly, fragments of *Caulerpa* can easily survive for many days out of water. Scuba divers, swimmers, and anything else that may come into contact with *Caulerpa* can also generate fragments and increase its dispersal (e.g. fishes swimming or rooting through a *Caulerpa* bed).

Economics:
Since *Caulerpa* spreads by vegetative fragments, entire areas have been shut down to all boat activity (recreation and commercial), fishing, and swimming by people and their pets. This has multiple negative economic impacts on communities with infestations. One could examine the fishing and tourism industries to get a better idea of how large the economic impact would be if they were shut down in certain areas.

Caulerpa is also very expensive to eradicate. The United States spent over $4.1 million in two years on eradication (2000-2002). More resources were spent after 2002 on eradication and currently funds are being spent to survey and look for more invasion sites.

Control/Eradication Efforts:
In the Mediterranean, resource managers and scientists knew that the population was too large to eradicate, so they have tried control methods. Both hand-removal and vacuuming the seaweed into boats with disposal on land have been attempted. Both were good at removing large biomasses of *Caulerpa*, but it also fragmented the seaweed promoting dispersal. The Mediterranean infestation was too large for chemical treatment, so in most

places the invasive seaweed has gone untreated. A few *Caulerpa*-free sanctuaries are presently being maintained in support of the dive industry.

Australians have tried killing *Caulerpa* by changing the salinity – they have dumped large amounts of rock salt in some locations and have replaced saltwater with freshwater in some channels. Salt was deemed somewhat successful on a scale up to a few hectares, but expensive. Replacing saltwater with freshwater is also very expensive, but has been effective over large areas. It was also considered less damaging to organisms downstream than copper sulfate.

Eradication in the United States was successful by a combination of the use of thick black tarps to block out the sunlight and putting chlorine under the tarps to more directly kill the *C. taxifolia*. Initially, liquid chlorine was used. It was later replaced with chlorine tablets. The eradication process went from 2000-2005.

Experts believe that it may take multiple approaches to control and possibly eradicate *Caulerpa*. Right now experiments are underway around the globe to determine what is most effective.

*Note: “setting pets free” can also lead to invasive fish problems.

Attachment B:
Experimental Design: Green algae control/eradication methods

Materials:
All experiments use live specimens, so all materials used (e.g. plastic tubs, forceps, etc.) must be free from chemical contamination.

12. Purchase mixed bags (2-3) of live green algae from a biological supply company or salt water aquarium store. This should be sufficient for a class of 30. (One bag costs approximately $10-$15.)
13. Keep the seaweed in 23 to 25ºC water that is aerated, circulated, or filtered with natural or artificial seawater and a light source. (4 gallons of seawater costs approximately $10.)
14. You will need a minimum of 18 small buckets/tubs or 50ml beakers that can be filled with seawater. This includes at least 15, plus 3 buckets for the different concentrations of one control agent. You will need more containers if using more than one control agent.
15. Filtered natural or artificial seawater. You can filter the seawater with 0.45 micron filters within 24 hours of the experimental set up. This may not be necessary with artificial seawater.
16. Air stones to aerate each beaker/tub (at least 15).
17. Paper towels.
18. Rulers (mm).
19. Razor blades (single-edge) or scalpels.
20. Forceps (tweezers).
21. Indelible markers.
22. Control agent (thick plastic or screening for shade, chlorine bleach, rock salt, copper sulfate)

Chlorine Bleach Procedure:
Overview: You will have 3 treatments and 5 replicates per treatment. So, you will need a total of 15 fragments and each fragment will be placed in its own beaker or small tub for 7 days (15 containers).

1. Using razor blades or scalpels create 15 seaweed fragments that are each 4 cm long. Use forceps to transfer fragments to and from seawater. Note: you may want to make more than 15 fragments in case some don't do well overnight. Make cuts on a hard surface that is covered with paper toweling that has been soaked in saltwater.
2. For the first 24 hours, each group should keep their fragments in seawater that is not circulating. This will allow for the wounds of the seaweed to heal. Use only healthy looking fragments in this experiment.
3. The 3 chlorine concentrations to test are 0, 50, and 125 ppm bleach in seawater or 0.00, 0.10, and 0.25% respectively. The 0 concentration = the control treatment.
4. Each group of 5 fragments will be dunked in the chosen concentration of chlorine seawater bath for 30 minutes. For example, 1 group of 5 fragments in 0 ppm, 1 group of 5 in 50 ppm, and 1 group of 5 in 125 ppm. Use forceps to transfer fragments to and from seawater and bleach seawater baths. The control, 0 ppm,

must be dunked in a separate seawater bath with no chlorine to account for handling of the fragments; all procedures must be the same for all treatments.

5. After exposure, rinse all fragments with filtered seawater.
6. Place the 15 rinsed fragments in separate 50 ml glass beakers or tubs that contain filtered seawater, is aerated and placed next to a window or under an artificial light. Remember, you need 15 beakers, one for each fragment, to have independent replicates.
7. Make observations for 7 days; record as dead (no green color, disintegrating) or alive (no loss of color or partial loss of green color, along with location where color remains on each fragment). You can use drawings of fragments to illustrate results. Maintain data in a spreadsheet.
8. Graph your results with a simple bar graph.

Note: This experimental design can be replicated for other control methods (thick plastic or layers of screen for shade, rock salt, copper sulfate). The teacher would decide the 3 different concentration/treatments to use (e.g. no shade, partial shade with some screens, or total darkness with thick plastic).

Note: **You may have invasive seaweed in the mixed bags** that you buy from the biological suppliers or saltwater aquarium stores. **Please follow these instructions to prevent any accidental introductions.** Filter seawater after you are done with the experiment and put filters along with all fragments in the freezer overnight. If there is left over seaweed from the experiment, that must be frozen as well. After being in the freezer overnight, all filters and seaweed can be thrown in the trash. **DO NOT DISPOSE IN ANY OTHER MANNER.**