



Learning Ocean Science through Ocean Exploration

Section 6

Ocean Primary Production

Photosynthesis

Every ecosystem requires an input of energy. The source varies with the system. In the majority of ocean ecosystems the source of energy is sunlight that drives photosynthesis done by micro- (phytoplankton) or macro- (seaweeds) algae, green plants, or photosynthetic blue-green or purple bacteria. These organisms produce ecosystem food that supports the food chain, hence they are referred to as primary producers. The balanced equation for photosynthesis that is correct, but seldom used, is $6\text{CO}_2 + 12\text{H}_2\text{O} = \text{C}_6\text{H}_{12}\text{O}_6 + 6\text{H}_2\text{O} + 6\text{O}_2$. Water appears on both sides of the equation because the water molecule is split, and new water molecules are made in the process. When the correct equation for photosynthesis is used, it is easier to see the similarities with chemosynthesis in which water is also a product.

Systems Lacking Primary Producers

There are some ecosystems that depend on primary production from other ecosystems. Many streams have few primary producers and are dependent on the leaves from surrounding forests as a source of food that supports the stream food chain. Snow fields in the high mountains and sand dunes in the desert depend on food blown in from areas that support primary production. The oceans below the photic zone are a vast space, largely dependent on food from photosynthetic primary producers living in the sunlit waters above. Food sinks to the bottom in the form of dead organisms and bacteria. It is as small as marine snow—tiny clumps of bacteria and decomposing microalgae—and as large as an occasional bonanza—a dead whale.

Chemosynthesis

Ocean exploration has brought to light new ecosystems, dependent on chemosynthetic bacteria which produce food from CO_2 (and sometimes water) using energy from the metabolism of inorganic materials found around them. Chemosynthesis was already well known in terrestrial systems. Nitrifying and anaerobic denitrifying bacteria as well as sulfur-fixing and anaerobic sulfur-reducing bacteria all use energy sources other than the sun. These bacteria had been studied in terrestrial and shallow water systems, but finding entire large ecosystems in the deep sea dependent on chemosynthetic bacteria using sulfur and methane as substrates opened major new areas of research. The discovery of hydrothermal vents and coldwater methane seeps has given us a new vision of primary production in the deep sea.

Hydrothermal Vent Systems

Hydrothermal vents—hot springs associated with spreading ridges on the sea floor—support exotic chemical-based ecosystems. The thriving communities associated with these vents shocked the scientific world when humans first observed a vent on the deep ocean floor in 1977. These vent ecosystems depend on microbes that use chemical energy available in the minerals from the hot spring water. Since they depend upon chemicals for energy instead of the sun, these autotrophic vent-dwelling microbes are called chemoautotrophs. Sulfur in the form of hydrogen sulfide is an energy rich, but toxic molecule. Bacteria that use hydrogen sulfide as an energy source are important to most vent food chains. They exist as both free-living organisms and as mutualistic symbionts within animals. The equation for chemosynthesis based on hydrogen sulfide is $6\text{CO}_2 + 24\text{H}_2\text{S} + 6\text{O}_2 = \text{C}_6\text{H}_{12}\text{O}_6 + 24\text{S} + 18\text{H}_2\text{O}$. The vent food chain supported by chemosynthetic bacteria includes shrimp, tubeworms, clams, fish, crabs, and octopi. These animals are well adapted to the extreme vent environment—complete darkness, water temperatures ranging from the 2°C of the surrounding seawater to 400°C at the vent, hundreds of atmospheres of pressure, and high concentrations of sulfides and other toxic chemicals.

The irony is that once scientists knew what to look for, they went to other well-known ecosystems that were rich in hydrogen sulfides, such as salt marshes, and found the same mutualistic association of chemosynthetic bacteria and animals that had stunned them in the deep vents. No one had ever thought to look for them, but they were there all along.

Both photosynthesis and chemosynthesis require an energy source and use carbon dioxide as a source of carbon to synthesize sugars and new water molecules. Photosynthesis gives off oxygen gas as a by-product, while chemosynthesis produces a wide variety of by-products, depending on what chemical substrate is used. The sugars produced provide both metabolic energy and substrate for synthesis of other biochemical molecules.

Hydrothermal vents have a number of kinds of lithotrophic (chemical-eating) bacteria associated with them in addition to those that use hydrogen sulfide since hydrothermal vents release many inorganic compounds to the surrounding seawater. Not all hydrothermal vents are the same. The very hot vents are called black smokers because some of the minerals released precipitate out as a black cloud in the surrounding colder water. They have high flow rates and very hot water, in addition to the variety of chemicals released. Other vents have cooler temperatures and different chemicals. The Lost City Hydrothermal Field near the Mid-Atlantic Ridge emits fluids at 40 to 75 degrees C and builds carbonate towers up to 60 m high.

Lithotrophic bacteria isolated from deep hydrothermal vents include those that oxidize hydrogen sulfide as well as nitrifying, hydrogen oxidizing, and iron and manganese-oxidizing bacteria. Methylobacterial bacteria that use methane as an energy source also occur there. Those that use hydrogen sulfide are the most studied, in part because they are symbionts in the giant tubeworms.

Cold Seeps

Many of the free-living lithotrophic bacteria form mats around the vent and are grazed upon directly by animals, so they also contribute to primary production around a deep vent.

Cold water seeps form above methane hydrate deposits. Communities develop around methane seeps that depend on the energy from hydrogen sulfide and methane captured by bacteria. Dense huge thickets of organisms surround the seeps. Animals from the nearby ocean may come and feed here as well as those that live only in the seep ecosystem.

This section includes several activities related directly to primary production in the ocean—both by photosynthesis and chemosynthesis. Additional exercises found on the OE web site or OE CD include:

Where to Find More Activities on Ocean Primary Productivity

- *Message in the Bottles* from the 2002 Arctic Ocean Exploration
- *Rock Eaters* of the Gulf of Alaska in Exploring Alaska's Seamounts 2002
- *From the Gulf of Mexico to the Moons of Jupiter* in the 2002 Gulf of Mexico expedition
- *Candy Chemosynthesis* in Submarine Ring of Fire 2002
- *It's a Gas* from Deep East 2001 and Hudson Canyon 2002

Lesson Plan 15

Being Productive in the Arctic Ocean

Focus

Primary productivity and limiting factors

Focus Question

What factors limit primary productivity in the Arctic Ocean?

Learning Objectives

Students will identify the major factors that limit primary productivity in the Arctic Ocean and will describe how these factors exert limiting effects.

Students will infer which factors are limiting in a data set of potentially limiting factors and primary productivity.

Materials

- ☐ Five sets of *Sample Data Cards* – one for each student group
- ☐ One *Data Summary Sheet* for each group

Audio/Visual Equipment

- ☐ None

Teaching Time

One or two 45-minute class periods

Seating Arrangement

Five groups

Key Words

Pelagic
Benthic
Zooplankton
Primary productivity
Phytoplankton

PAR

Chlorophyll *a*

Background Information

This activity focuses on primary productivity in the pelagic realm of the Arctic Ocean. Primary productivity refers to the amount of organic matter, usually expressed as grams of carbon per square meter per day, produced by organisms that make food from simple inorganic substances using energy from sunlight to conduct photosynthesis or the chemical reactions of chemosynthesis. Primary productivity in the Arctic Ocean is largely due to photosynthesis carried out by microscopic drifting algae called phytoplankton. Photosynthesis requires photosynthetic organisms, light, carbon dioxide, water, and mineral nutrients. Lack of any one of these may limit photosynthesis or primary production. Which factors limit primary production in the Arctic Ocean? How much primary production actually occurs? This activity uses data from a scientific paper: Smith, Jr., W. O. 1995. Primary productivity and new production in the Northeast Water (Greenland) Polynya during summer 1992. *Journal of Geophysical Research* 100: 4357-4370. It enables your students to examine the factors that limit primary production using real data.

The Arctic Ocean is the smallest of the world's four ocean basins. It is not easily explored as it is almost entirely covered with ice for eight months of the year, a drifting polar ice pack covers the central and western portions year-round, and sea temperature seldom rises above 0°C. Organisms living in the water column between the ocean surface and the bottom, largely phytoplankton, conduct most

of the primary production in the pelagic water. Melting sea ice allows increased light to enter the sea. Algae grow rapidly since the sun shines for 24 hours a day during the summer. Through photosynthesis, these phytoplankton provide energy for a variety of drifting animals (zooplankton) that include crustaceans and jellyfish. Zooplankton provide food for larger pelagic animals including fishes, squids, seals, and whales. When pelagic organisms of any size die, they settle to the ocean bottom as detritus and become the energy source for benthic organisms, including sponges, bivalves, crustaceans, polychaete worms, sea anemones, bryozoans, tunicates, and ascidians. These animals are food for bottom-feeding fishes, whales, and seals.

LEARNING PROCEDURE

1. Review the background information on the Arctic Ocean with your students. Emphasize that the sea ice, pelagic and benthic communities are connected in the food chain and that photosynthesis by microscopic algae (phytoplankton) provides the energy for all the other organisms in these realms. Use the OE CD or web site for information on the Arctic Ocean. You may mention that a few marine systems, such as those in the vicinity of hydrothermal vents or cold water seeps, are not dependent on photosynthesis for energy, but rely on chemosynthesis instead. Check the OE CD or the web site at <http://oceanexplorer.noaa.gov/explorations/02galapagos/galapagos.html> and <http://oceanexplorer.noaa.gov/explorations/02fire/welcome.html> for information on these systems. If necessary, review the basic concepts of photosynthesis. Be sure students understand that photosynthesis is limited by whichever of the essential components is in limited supply.
2. Students have data on Arctic Ocean primary productivity and measurements of the factors that may limit production. Ten data sets will be examined, representing samples that were taken at 10 different times of the year. As each sample is examined, students are asked to explain the results in terms of what factors seem to be limiting primary productivity.
3. Distribute the five sets of sample data cards to the student groups. One group should receive the Ice Cover cards, a second group should receive the Chlorophyll a cards, a third group should receive the PAR cards, a fourth group should receive the Nitrate cards, and the fifth group should receive the Primary Productivity cards. Each set should contain one card for each of the 10 sampling days.
4. Briefly discuss the meaning of each set of cards:
 - Ice Cover cards show the percent of the sea surface that is covered with ice.
 - PAR cards list the amount of photosynthetically active radiation - the amount of sunlight that is usable for photosynthesis - as a percentage of the maximum radiation that occurs during the year.
 - Chlorophyll a cards show the amount of chlorophyll a - a measure of the photosynthetic-capable algae present in the surface seawater.
 - Nitrate cards list the amount of nitrogen-containing mineral nutrients present such as nitrate or ammonia.
 - Primary Productivity cards show the amount of organic matter produced through photosynthesis at the sea surface.
5. Working with the class as a whole, have each group take turns reading its cards for Sample Day #1. List the readings on the Blank Data Summary Sheet. When each group has read their cards, discuss the results. Repeat this process for two more days. Then have the students fill in the sheet and decide as a group what the limiting factor for each of the days from #4 to #10 is that limits primary production. Then discuss their thoughts as a class.
 - Sample Day #1: This is a fairly high rate of Primary Productivity. Students should note that there is no ice to block sunlight and PAR is fairly high. The significance of Chlorophyll a and Nitrate concentrations will become apparent as other days are examined.

- Sample Day #2: 50% of the sea surface is covered with ice which limits Primary Productivity to less than half the value on Sample Day #1, even though the PAR and Nitrate levels are higher and there are only slightly fewer algae as indicated by Chlorophyll a than on Sample Day #1. Reduction of sunlight by sea ice is a major limiting factor for primary productivity in the Arctic Ocean.
- Sample Day #3: Primary Productivity again is much lower than on Sample Day #1. A combination of ice cover and reduced PAR—perhaps a cloudy day—are probably responsible, since Nitrate and Chlorophyll a levels are similar to previous days.
- Sample Day #4: Primary Productivity is low and the obvious cause is the greatly reduced level of Nitrate.
- Sample Day #5: Everything seems favorable, but Primary Productivity is low. The students may notice that PAR is nearly 100% and may wonder whether there is such a thing as too much light. In the Arctic Ocean, photosynthetic algae can be adapted to rather low light conditions. It is possible for photosynthesis to be inhibited if these algae are exposed to too much light.
- Sample Day #6: Low Primary Productivity again; extensive ice cover is the likely cause.
- Sample Day #7: Time for inferences! When would you expect ice to cover 100% of the sea surface? Winter, of course! So, PAR would be zero because night lasts 24 hours in the polar winter. We would expect Chlorophyll a and Primary Productivity to be pretty close to zero as well.
- Sample Day #8: Reviewing the preceding data sets, students should notice that Nitrate does not appear to limit Primary Productivity except when it is in very limited supply. Since Primary Productivity is relatively high and there is 30% ice cover, students could reasonably infer that Nitrate is not limiting in this case, so it could be any of the previous levels except 0.2.

- Sample Day #9: Low PAR is the key here. It is probably early winter, so students might conclude ice cover is probably fairly high (above 70%) and Primary Productivity is probably quite low.
- Sample Day #10: Since all other factors seem pretty favorable, yet Primary Productivity is low, students should suspect that Nitrate levels are low enough to be limiting.

6. Have students write individual summaries of factors that limit Primary Productivity in the Arctic Ocean.

THE BRIDGE CONNECTION

www.vims.edu/bridge/polar.html

www.vims.edu/bridge/plankton.html

THE “Me” CONNECTION

Have students write a short essay or prepare a brief oral presentation on how knowledge of primary productivity in the Arctic Ocean might benefit them personally, and/or why they think this knowledge is (or is not) important. Ask students to share their thoughts with the class. If they like whales, they have a good connection.

CONNECTION TO OTHER SUBJECTS

English/Language Arts, Mathematics

EVALUATION

Have students write their own interpretations of Sample Days #6 – 10 before these are discussed in class.

EXTENSIONS

Have students use the OE CD or visit <http://oceanexplorer.noaa.gov> to learn about the deep Arctic Ocean and to find out what organisms researchers actually find in the three realms.

Have students research primary productivity in temperate and/or tropical ocean waters and compare these data with primary productivity in the Arctic Ocean.

RESOURCES

<http://oceanexplorer.noaa.gov> – the Arctic Ocean Expedition daily documentaries and discoveries

<http://www.sciencegems.com/earth2.html> – Science education resources

<http://www-sci.lib.uci.edu/HSG/Ref.html> – References on just about everything

<http://photoscience.la.asu.edu/photosyn/education/learn.html>
– Links to many sites and activities about photosynthesis

NATIONAL SCIENCE EDUCATION STANDARDS

Content Standard A: Science As Inquiry

- Abilities necessary to do scientific inquiry
- Understanding about scientific inquiry

Content Standard B: Physical Science

- Chemical reactions

Content Standard C: Life Science

- Interdependence of organisms

Content Standard D: Earth and Space Science

- Energy in the Earth system

Activity developed by Mel Goodwin, PhD, The Harmony Project, Charleston, SC

Student Handout**ICE COVER DATA****Sample Day #1**

Ice Cover = 0%

ICE COVER DATA**Sample Day #2**

Ice Cover = 50%

ICE COVER DATA**Sample Day #3**

Ice Cover = 50%

ICE COVER DATA**Sample Day #4**

Ice Cover = 10%

ICE COVER DATA**Sample Day #5**

Ice Cover = 0%

ICE COVER DATA**Sample Day #6**

Ice Cover = 70%

ICE COVER DATA**Sample Day #7**

Ice Cover = 100%

ICE COVER DATA**Sample Day #8**

Ice Cover = 30%

ICE COVER DATA**Sample Day #9**

Ice Cover = GUESS!

ICE COVER DATA**Sample Day #10**

Ice Cover = 15%

Student Handout

PHOTOSYNTHETICALLY ACTIVE RADIATION DATA

Sample Day #1

PAR = 75% of maximum

PHOTOSYNTHETICALLY ACTIVE RADIATION DATA

Sample Day #2

PAR = 78% of maximum

PHOTOSYNTHETICALLY ACTIVE RADIATION DATA

Sample Day #3

PAR = 45% of maximum

PHOTOSYNTHETICALLY ACTIVE RADIATION DATA

Sample Day #4

PAR = 76% of maximum

PHOTOSYNTHETICALLY ACTIVE RADIATION DATA

Sample Day #5

PAR = 98% of maximum

PHOTOSYNTHETICALLY ACTIVE RADIATION DATA

Sample Day #6

PAR = 79% of maximum

PHOTOSYNTHETICALLY ACTIVE RADIATION DATA

Sample Day #7

PAR = GUESS!

PHOTOSYNTHETICALLY ACTIVE RADIATION DATA

Sample Day #8

PAR = 82% of maximum

PHOTOSYNTHETICALLY ACTIVE RADIATION DATA

Sample Day #9

PAR = 10% of maximum

PHOTOSYNTHETICALLY ACTIVE RADIATION DATA

Sample Day #10

PAR = 79% of maximum

Student Handout**CHLOROPHYLL α DATA****Sample Day #1**Chlorophyll α = 87 $\mu\text{g/l}$ **CHLOROPHYLL α DATA****Sample Day #2**Chlorophyll α = 76 $\mu\text{g/l}$ **CHLOROPHYLL α DATA****Sample Day #3**Chlorophyll α = 73 $\mu\text{g/l}$ **CHLOROPHYLL α DATA****Sample Day #4**Chlorophyll α = 82 $\mu\text{g/l}$ **CHLOROPHYLL α DATA****Sample Day #5**Chlorophyll α = 85 $\mu\text{g/l}$ **CHLOROPHYLL α DATA****Sample Day #6**Chlorophyll α = 71 $\mu\text{g/l}$ **CHLOROPHYLL α DATA****Sample Day #7**Chlorophyll α = GUESS!**CHLOROPHYLL α DATA****Sample Day #8**Chlorophyll α = 76 $\mu\text{g/l}$ **CHLOROPHYLL α DATA****Sample Day #9**Chlorophyll α = 5 $\mu\text{g/l}$ **CHLOROPHYLL α DATA****Sample Day #10**Chlorophyll α = 73 $\mu\text{g/l}$

Student Handout

NITRATE DATA

Sample Day #1

Nitrate = 6.2 $\mu\text{mol/l}$

NITRATE DATA

Sample Day #2

Nitrate = 8.4 $\mu\text{mol/l}$

NITRATE DATA

Sample Day #3

Nitrate = 11.3 $\mu\text{mol/l}$

NITRATE DATA

Sample Day #4

Nitrate = 0.2 $\mu\text{mol/l}$

NITRATE DATA

Sample Day #5

Nitrate = 7.1 $\mu\text{mol/l}$

NITRATE DATA

Sample Day #6

Nitrate = 6.7 $\mu\text{mol/l}$

NITRATE DATA

Sample Day #7

Nitrate = 5.2 $\mu\text{mol/l}$

NITRATE DATA

Sample Day #8

Nitrate = GUESS!

NITRATE DATA

Sample Day #9

Nitrate = 7.6 $\mu\text{mol/l}$

NITRATE DATA

Sample Day #10

Nitrate = GUESS!

Student Handout**PRIMARY PRODUCTIVITY DATA****Sample Day #1**

Surface Primary Productivity =
9.3 mg C/m²/day

PRIMARY PRODUCTIVITY DATA**Sample Day #2**

Surface Primary Productivity =
4.3 mg C/m²/day

PRIMARY PRODUCTIVITY DATA**Sample Day #3**

Surface Primary Productivity =
5.1 mg C/m²/day

PRIMARY PRODUCTIVITY DATA**Sample Day #4**

Surface Primary Productivity =
3.4 mg C/m²/day

PRIMARY PRODUCTIVITY DATA**Sample Day #5**

Surface Primary Productivity =
4.3 mg C/m²/day

PRIMARY PRODUCTIVITY DATA**Sample Day #6**

Surface Primary Productivity =
3.4 mg C/m²/day

PRIMARY PRODUCTIVITY DATA**Sample Day #7**

Surface Primary Productivity =
GUESS!

PRIMARY PRODUCTIVITY DATA**Sample Day #8**

Surface Primary Productivity =
6.5 mg C/m²/day

PRIMARY PRODUCTIVITY DATA**Sample Day #9**

Surface Primary Productivity =
GUESS!

PRIMARY PRODUCTIVITY DATA**Sample Day #10**

Surface Primary Productivity =
2.6 mg C/m²/day

Teacher's Master Data Summary

Teacher Answer Key

Sample Day	Ice Cover (% of surface)	PAR* (% of maximum)	Chlorophyll <i>a</i> ($\mu\text{g/l}$)	Nitrate ($\mu\text{mol/l}$)	Primary Productivity ($\text{mg C/m}^2/\text{day}$)
1	0	75	87	6.2	9.3
2	50	78	76	8.4	4.3
3	20	45	73	11.3	5.1
4	10	76	82	0.2	3.4
5	0	98	85	7.1	4.3
6	70	79	71	6.7	3.4
7	100	GUESS	GUESS	5.2	GUESS
8	30	82	76	GUESS	6.5
9	GUESS	10	5	7.6	GUESS
10	15	79	73	GUESS	2.6

*photosynthetically active radiation

DATA SUMMARY SHEET**Student Handout**

Sample Day Productivity	Ice Cover (% of surface)	PAR* (% of maximum)	Chlorophyll <i>a</i> ($\mu\text{g/l}$)	Nitrate ($\mu\text{mol/l}$)	Primary ($\text{mg C/m}^2/\text{day}$)
1					
2					
3					
4					
5					
6					
7					
8					
9					
10					

*photosynthetically active radiation

Lesson Plan 16

Chemosynthesis for the Classroom

Focus

Chemosynthetic bacteria

Focus Question

What changes affect succession in the development of chemosynthetic bacterial communities?

Learning Objectives

Students will observe the development of chemosynthetic bacterial communities.

Students will recognize that organisms modify their environment in ways that create opportunities for other organisms to thrive.

Students will be able to explain the process of chemosynthesis.

Students will be able to explain the relevance of chemosynthesis to biological communities in the vicinity of cold seeps.

Materials

- ☐ Directions for setting up Winogradsky columns from <http://topex-www.jpl.nasa.gov/education/activities>
- ☐ Black mud from a local river, lake, or estuary, approximately 500 ml for each student group
- ☐ Water from the same source used to obtain black mud, approximately 3 liters for each student group
- ☐ 500-ml graduated cylinder for each student group
- ☐ Calcium sulfate (plaster of Paris), 80 g for each student group
- ☐ 500-ml jar or beaker
- ☐ Stirring rod
- ☐ Straw (hay) or small pieces of filter paper, 50 g for each student group

- ☐ Sodium bicarbonate (baking soda), 4 g for each student group
- ☐ Crushed multivitamin pill, one for each group
- ☐ Plastic wrap
- ☐ Rubber bands
- ☐ Source of artificial light
- ☐ Tape and markers for labeling graduated cylinders
- ☐ Flashlight with red cellophane over lens
- ☐ Optional: microscopes and materials for making wet mounts

Audio/Visual Equipment

None

Teaching Time

One 45-minute class period to set up columns, approximately 15 minutes at weekly intervals for six weeks to make observations, and one 45-minute class period for presentation and discussion of results

Seating Arrangement

Groups of four students

Key Words

Cold seeps
Methane hydrate ice
Chemosynthesis
Brine pool
Vestimentifera
Trophosome
Succession

Background Information

This activity focuses on chemosynthetic bacteria similar to those that are the base of food webs in

cold seep communities. Black mud from a local water body is incubated in a glass cylinder, called a Winogradsky column, with a source of chemical energy from calcium sulfate and organic material from straw or filter paper. A succession of chemosynthetic bacteria grow over a period of six weeks. This activity was originally developed by the Orange County Marine Institute/San Juan Institute Activity Series and is available on NASA's Jet Propulsion Laboratory Ocean Planet web site at <http://topex-www.jpl.nasa.gov/education/activities>.

Deep-sea chemosynthetic communities occur where hydrocarbon gases, often methane and hydrogen sulfide and/or oil seep out of sediments. These areas, known as cold seeps, are common along continental margins and, like hydrothermal vents, are home to many unique species. Typical features of the seep communities that have been studied so far include mounds of frozen crystals of methane and water called methane hydrate ice, that are home to polychaete worms. Brine pools, containing water four times saltier than normal seawater, also occur. Researchers often find dead fish floating in the brine pool, apparently killed by the high salinity. Where hydrogen sulfide is present, large tubeworms known as vestimentiferans (formerly classified as belonging to the phylum Pogonophora, but recently Pogonophora and Vestimentifera have been included in the phylum Annelida) grow in clusters of millions of individuals. These unusual animals lack a mouth, stomach, or gut. Instead, they have a large organ called a trophosome, that contains chemosynthetic bacteria. Vestimentiferans have tentacles that extend into the water. The tentacles are bright red due to the presence of hemoglobin that absorbs hydrogen sulfide and oxygen. The hemoglobin transports them to bacteria in the trophosome. These bacteria produce organic molecules that provide nutrition to the tubeworm. A similar symbiotic relationship is found in clams and mussels that have chemosynthetic bacteria living in their gills. Bacteria also live independently from other organisms in large bacterial mats. Coldwater seep communities also include snails, eels, sea stars, crabs, lobsters,

isopods, sea cucumbers, and fish which likely use the tubeworms, mussels, and bacterial mats as food.

The Gulf of Mexico has the largest fossil fuel reservoir in the continental U.S. Its geology has been intensively studied for more than 50 years. Yet cold seep communities were only discovered in the Gulf about 20 years ago, and, as of 2001, their biology was examined at only three sites less than 20 km apart.

LEARNING PROCEDURE

1. Lead a discussion of deep-sea chemosynthetic communities. Contrast chemosynthesis with photosynthesis. In both processes, organisms build sugars from carbon dioxide and a hydrogen source. This process requires energy; photosynthesizers obtain this energy from the sun, while chemosynthesizers obtain energy from chemical reactions. Discuss the variety of chemical reactions that can provide this kind of energy. Contrast hydrothermal vent communities with cold seep communities. Visit http://www.bio.psu.edu/cold_seeps for a virtual tour of a cold seep community.
2. Challenge your students to make their own chemosynthetic communities—closed ecosystems in a bottle. Have them follow the steps given at <http://topex-www.jpl.nasa.gov/education/activities> to set up two kinds of Winogradsky columns, using locally-collected black mud. Divide the groups in half and assign each group to either the light or the dark half. Cover each column tightly with plastic wrap and secure with rubber bands. One half of the columns will be placed in a darkened area and the other in indirect light—not direct sunlight. Students should observe the light and dark columns weekly and record their observations. You may have them make wet mounts for microscopic examination at the end of three and six weeks. Use appropriate safety precautions when making wet mounts, including gloves, antibacterial solution for slide disposal, and hand washing following completion of the activity.

3. Have each group present and discuss its results. Students should have observed a series of changes in the mud's appearance in the columns caused by bacterial species succession—that is, as each kind grows, it changes the environment, making it more favorable for other species. They should infer that changes caused by one species, for example, waste product production, create opportunities for other species. Similarly, changes in the chemical composition of the mud, such as formation of hydrogen sulfide, alters the environment in ways that may favor the growth of other bacterial species. The processes they observed in the Winogradsky columns roughly models the development of deep-sea chemosynthetic communities. Ask the students to speculate about what other organisms might appear in the community if these processes were taking place in the area from which the mud was collected.

THE BRIDGE CONNECTION

www.vims.edu/BRIDGE/vents.html

THE “ME” CONNECTION

Have students write a short essay on why cold seeps might be directly important to their own lives. You may want to offer a hint that perhaps the energy source used by chemosynthetic bacteria could be useful to other species as well. Some estimates suggest that there may be more energy locked up in methane hydrate ices than in all other fossil fuels combined!.

CONNECTION TO OTHER SUBJECTS

English/Language Arts, Biology, Earth Science

EVALUATION

Have students submit records of their observations and their written interpretation of these observations.

EXTENSIONS

Have students investigate more about ancient bacteria and recent findings about physical conditions on some of Jupiter's moons, and report on the implications of chemosynthetic bacteria.

<http://www.bio.psu.edu/People/Faculty/Fisher/fhome.htm> – Web site for the principal investigator on the OE Gulf of Mexico expedition.

<http://www.rps.psu.edu/deep/> – Notes from an expedition exploring deep-sea communities for the origins of life on Earth and extraterrestrial life (<http://www.ocean.udel.edu/deepsea/level-2/chemistry/bacteria.html> and <http://pubs.usgs.gov/publications/text/dynamic.html#anchor19309449> are useful for this).

RESOURCES

<http://oceanexplorer.noaa.gov> –Gulf of Mexico Expedition documentaries and discoveries

<http://ridge.oce.orst.edu/links/edlinks.html> – Links to other deep ocean exploration web sites

<http://www-ocean.tamu.edu/education/oceanworld/resources/>
– Links to other ocean-related web sites

NATIONAL SCIENCE EDUCATION STANDARDS

Content Standard A: Science As Inquiry

- Abilities necessary to do scientific inquiry
- Understanding about scientific inquiry

Content Standard B: Physical Science

- Chemical reactions
- Interactions of energy and matter

Content Standard C: Life Science

- Interdependence of organisms
- Matter, energy, and organization in living systems

Content Standard D: Earth and Space Science

- Energy in the Earth system
- Origin and evolution of the Earth system

Activity developed by Mel Goodwin, PhD, The Harmony Project, Charleston, SC

Lesson Plan 17

Biochemical Detectives

Focus

Biochemical clues to energy-obtaining strategies

Focus Question

How can researchers determine energy and nutritional strategies used by organisms in cold-seep communities?

Learning Objectives

Students will explain the process of chemosynthesis and its relevance to biological communities near cold seeps.

Students will describe three energy-obtaining strategies used by organisms in cold seep communities.

Students will interpret enzyme activity analyses and $\delta^{13}\text{C}$ isotope values and draw inferences from these data about energy-obtaining strategies used by organisms in cold-seep communities.

Materials

- ☐ Flip chart, chalk board, or marker board
- ☐ One copy of *Cold Seep Organism Analysis* results for each student

Audio/Visual Equipment

None

Teaching Time

One or two 45-minute class periods

Seating Arrangement

Groups of four students

Key Words

Cold seeps
Methane hydrate ice
Chemosynthesis
Brine pool
Vestimentifera
Trophosome
Detritus
Isotope analysis
 $\delta^{13}\text{C}$
Enzyme analysis

Background Information

In this activity, students use biochemical analyses of organisms from cold seep communities as a basis for drawing inferences about the energy-obtaining strategies used by these organisms. The teacher introduces coldwater seeps and some of the biochemistry associated with organisms at seeps. Then the students examine real data and draw conclusions from them. This activity assumes the students have a foundation in both chemistry and biology and understand basic physiology. Teachers and students should visit the OE Gulf of Mexico 2002 web site or OE CD and read the *Introduction* plus *Communities* and *Tubeworms* as preparation for this activity.

Coldwater seeps are areas where hydrocarbon gases, generally methane, as well as hydrogen sulfide and/or oil seep out of sediments. They commonly occur along continental margins and, like hydrothermal vents, are home to many species not found anywhere else. Typical features of communities currently known include mounds of frozen crystals of methane and water called methane hydrate ice, occupied by ice worms. Brine pools of

water four times saltier than normal seawater have also been found. Researchers often find dead fish floating in the brine pool, apparently killed by high salinity. As with hydrothermal vents, chemosynthetic bacteria form the base of the food web in cold seep communities. These bacteria exist in thick bacterial mats or may live symbiotically in mutually beneficial association with other organisms.

One conspicuous association is that of chemosynthetic bacteria and large vestimentiferan tubeworms formerly classified within the Phylum Pogonophora. (Recently Pogonophora and Vestimentifera have been placed in the Phylum Annelida). Pogonophora means “beard bearing” and refers to the one or more tentacles at the anterior end. Vestimentiferan tentacles are bright red because they contain hemoglobin, the same molecule that makes our blood cells red. Vestimentiferans can grow more than 10 feet long, occur in huge clusters and are estimated to live to over 200 years. They lack a mouth, stomach, and gut. Instead, they have a large organ called a trophosome that holds chemosynthetic bacteria. Hemoglobin in the tubeworms’ blood transports hydrogen sulfide and oxygen to bacteria living in the trophosome. The bacteria produce organic molecules that provide nutrition to the tubeworm. Similar relationships are found in clams and mussels that have chemosynthetic bacteria living in their gills. Other seep organisms use tubeworms, mussels, and bacterial mats as food. These include snails, eels, sea stars, crabs, isopods, sea cucumbers, and fish. As the seep community ages, species composition changes. The area becomes less suitable for the unique species and other organisms are able to forage there.

This activity focuses on different energy-obtaining strategies studied in cold seep organisms. There are two methods of obtaining energy (food) in the deep sea. One is to feed on organic material that derives from photosynthetic organisms living in the upper water column. In bottom dwelling communities, this organic material is present primarily as detritus—bits of dead algal and animal material along with

decomposing bacteria settling to the bottom. Some organic material originates from pelagic organisms that move vertically through the water column.

The second source of food and energy is chemosynthesis. The chemosynthetic energy source is a variety of chemicals. In cold seep communities methane and hydrogen sulfide appear to be the primary sources. Chemosynthesis also requires a source of dissolved carbon—CO₂ in seawater or from hydrocarbons from the seep.

The source of carbon and energy used by species gives important clues about food webs and community structure. Researchers use measurements of carbon isotope ratios to determine carbon sources. The amount of the stable carbon isotope ¹³C varies, depending upon the source of the energy and carbon used in fixing the carbon in organic molecules. Isotope content is compared with a standard. The results are expressed as delta values, abbreviated d(x) in parts-per-thousand (‰; also called parts per mille). Scientists have found that δ¹³C of carbon in photosynthetically-derived detritus is -18 to -20‰; δ¹³C in carbon derived from seawater is -0‰; δ¹³C in carbon from organisms that feed on methane is -40‰ or lower. δ¹³C in carbon from organisms that depend upon sulfur as an energy source is between -30 and -40‰.

Scientists also study energy-obtaining strategies using biochemical studies of cold-seep community organisms. The enzymes adenosine triphosphate sulfurylase (ATPS), adenosine-5 phosphosulfate reductase (APR), and sulfide oxidase (SuO) are common in organisms that use sulfur, while ribulose-bisphosphate carboxylase (RuBP) is more abundant in autotrophic organisms, and methanol dehydrogenase (MeD) is found in organisms that use methanol as an energy source.

LEARNING PROCEDURE

1. Lead a discussion of deep-sea chemosynthetic communities with an emphasis on cold seeps.

Contrast chemosynthesis with photosynthesis. Discuss the variety of chemical reactions that can provide energy for chemosynthesis. Visit http://www.bio.psu.edu/cold_seeps for a virtual tour of a cold seep community. Have the students access the Gulf of Mexico 2002 OE web site or CD.

2. Review the various energy-obtaining options available to cold-seep organisms. Briefly discuss the use of $\delta^{13}\text{C}$ isotope analysis and enzyme analysis for obtaining clues about specific energy-obtaining strategies. Have the students work with you to construct a chart showing what the presence of certain enzymes or $\delta^{13}\text{C}$ values tells researchers about the probable source of energy and carbon.
3. Distribute *Cold-Seep Organism Analysis Results* sheets. These are results of actual biochemical studies on gill tissues of bivalves and trophosome tissues of vestimentiferans. These tissues are used because they are the sites of energy-producing activity within the organisms. Students should plot the Bivalve $\delta^{13}\text{C}$ Analysis data as histograms and draw inferences about the energy-obtaining strategy used by clams versus mussels at cold seeps. The Enzyme and $\delta^{13}\text{C}$ Analysis data can be averaged and compared among organisms and inferences made about the energy-obtaining strategy used by each organism.
4. Have groups present their results and summarize these on a flip chart, chalkboard, or marker board. Lead a discussion of these results. Students who examined $\delta^{13}\text{C}$ isotope data for bivalves should recognize three distinct groups: mussels with $\delta^{13}\text{C}$ values of -40 or more, suggesting a methane-based strategy; clams with $\delta^{13}\text{C}$ values of -20 to -40, suggesting a sulfur-based strategy; and clams with $\delta^{13}\text{C}$ values of -14 to -20, suggesting a heterotrophic strategy—filter-feeding algae or algal detritus. Students who examined data from the vestimentiferan *Lamellibrachia* sp. and the clam *Pseudomilthia* sp. should recognize that the enzyme activity and $\delta^{13}\text{C}$ values suggest a

sulfur-based strategy, probably involving bacterial symbionts. Similarly, results for the unidentified mussel suggest a methane-based strategy.

THE BRIDGE CONNECTION

www.vims.edu/bridge/vents.html

THE “Me” CONNECTION

Have students write a short essay on their personal strategy for obtaining energy, and how their strategy might involve some form of chemosynthesis. What chemical energy source(s) would their chemosynthetic strategy utilize?

CONNECTION TO OTHER SUBJECTS

English/Language Arts, Biology, Earth Science

EVALUATION

Have students prepare individual written statements of their conclusions prior to oral presentations. Create a grading rubric that includes the group (oral) and individual (written) components.

EXTENSIONS

Have students find biological information and pictures of the organisms they investigated in this exercise using web sources.

RESOURCES

<http://oceanexplorer.noaa.gov> – Gulf of Mexico Expedition 2002 documentaries and discoveries

<http://www.bio.psu.edu/People/Faculty?Fisher/thome.htm> – Web site for the principal investigator on the Gulf of Mexico 2002 Expedition

<http://www.rps.psu.edu/deep/> – Notes from another expedition exploring deep-sea communities

<http://www.ridge.oce.orst.edu/links/edlinks.html> – Links to other deep ocean exploration web sites

<http://www-ocean.tamu.edu/education/oceanworld/resources/> – Links to other ocean-related web sites

NATIONAL SCIENCE EDUCATION STANDARDS

Content Standard A: Science As Inquiry

- Abilities necessary to do scientific inquiry
- Understanding about scientific inquiry

Content Standard B: Physical Science

- Chemical reactions
- Interactions of energy and matter

Content Standard C: Life Science

- Interdependence of organisms
- Matter, energy, and organization in living systems

Content Standard D: Earth and Space Science

- Energy in the Earth system

*Activity developed by Mel Goodwin, PhD,
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Student Handout**Cold Seep Organism Analysis Results
Bivalve $\delta^{13}\text{C}$ Analysis**

Animal	$\delta^{13}\text{C}$ (‰)	Animal	$\delta^{13}\text{C}$ (‰)	Animal	$\delta^{13}\text{C}$ (‰)
mussel	-56	clam	-18	mussel	-46
clam	-32	clam	-34	clam	-18
mussel	-56	clam	-32	mussel	-46
mussel	-56	clam	-16	mussel	-46
clam	-36	mussel	-50	clam	-16
clam	-32	mussel	-50	mussel	-44
mussel	-52	clam	-30	mussel	-44
mussel	-52	mussel	-50	clam	-16
clam	-32	clam	-34	mussel	-44
clam	-36	clam	-18	mussel	-44
mussel	-52	clam	-36	clam	-38
clam	-34	clam	-32	mussel	-44
clam	-32	mussel	-50	mussel	-44
clam	-18	clam	-36	clam	-16
mussel	-52	mussel	-48	mussel	-44
clam	-36	clam	-30	mussel	-42
mussel	-52	mussel	-48	mussel	-40
clam	-36	clam	-32	clam	-38
clam	-32	clam	-34		
clam	-30	clam	-18		
mussel	-50	mussel	-46		
mussel	-50	clam	-30		
clam	-14	mussel	-46		
clam	-34	clam	-38		
clam	-32	mussel	-46		
mussel	-50	clam	-16		
clam	-18	mussel	-46		
mussel	-50	mussel	-46		
clam	-36	clam	-36		
clam	-16	mussel	-46		
clam	-30	mussel	-46		
clam	-32	clam	-36		
mussel	-50	mussel	-46		
clam	-36	mussel	-46		

Student Handout

Cold Seep Organism Analysis Results Enzyme and $\delta^{13}\text{C}$ Analysis

Enzyme Activity						$\delta^{13}\text{C}$ (‰)
Animal	RuBP	ATPS	APR	MeD	SuO	
<i>Pseudomilthia</i> sp. (clam)						
sample #1	0.43	12.86	0.83	nd	2.1	-33.5
sample #2	0.41	2.47	0.66	nd	1.94	-33.6
sample #3	0.44	15.43	1.36	nd	2.04	-32.5
Unidentified Mussel						
sample #1	0.011	nd	nd	0.66	0.7	-51.8
sample #2	0.017	nd	nd	0.53	0.75	-52.0
sample #3	0.021	nd	nd	0.4	1.09	-52.6
<i>Lamellibrachia</i> sp. (vestimentiferan)						
sample #1	0.24	4.24	0.70	nd	1.77	-36.6
sample #2	4.03	1.03	nt	nd	3.15	-36.8
sample #3	4.97	0.51	0.78	nd	5.47	-37.4

nd = not detected

nt = not tested