

Seeing the Carbon Cycle

The most important biochemical reactions for life in the ocean and on earth are cellular respiration and photosynthesis. These two reactions play a central role in the carbon cycle, which is projected to impact the lives of today's students, since the ocean-based carbon cycle plays a key role in global warming and hydrocarbon fossil fuels (*Figure 1*). Both have a full subset of additional issues, ranging from disruption of weather patterns, melting of the icecaps, rising sea levels, species extinction, and reusable energy sources. All are related to the carbon cycle. The carbon cycle functions at three temporal rates: 1) Short-term, which consists of respiration, combustion, and photosynthesis; 2) Medium-term, which consists of deforestation, decomposition, and carbon dioxide diffusion in water; and 3) Long-term, which consists of carbonate sedimentation of plankton, sedimentary rock, plate tectonics, and volcanic eruptions. The carbon cycle is highly relevant to today's students.

The carbon cycle is difficult to dissect for the middle school learner. It spans many disciplines and involves chemical reactions that require abstract thinking. However, the significance of the carbon cycle requires that it be explored. The following lessons outline a classroom experiment that was developed to introduce middle school learners to chemistry through the carbon cycle. It allows students to observe the influence of the carbon cycle on algae growth, explore experimental design, collect data, and make a conclusion.

Teaching The Carbon Cycle

These carbon cycle lessons were designed for a seventh grade physical science class based on the Massachusetts Science and Technology Frameworks. Before introducing the carbon cycle, a pre-assessment was given to measure prior knowledge (*Figure 2*). The same form was used as a post-assessment. An introduction included an ocean-focused carbon cycle with deleted terms; the terms were discussed and the students filled in the missing steps of the carbon cycle (*Figure 1*). The students also conducted a carbon cycle scavenger hunt (*Worksheet 1*) and constructed their own carbon cycle from drawings, magazine pictures, and labels.

To make the carbon cycle relevant, we connected it to the contemporary work of researcher Dr. Daniel J Repeta, from the Woods Hole Oceanographic Institute. We discussed Dr. Repeta's research interests of cycling of organic matter and geochemistry of photosynthetic pigments with the students. The students prepared to conduct a carbon cycle experiment in their physical science classes, on the growth of algae using carbon dioxide. At this stage, students reviewed the steps of the scientific method. The objective was to have students write a hypothesis of how the carbon cycle would influence growth of algae. The students wrote their own "If...., then...." hypothesis, such as "If there is more CO₂, then there will be less algae growth" or "If CO₂ increases, then algae growth increases." The students were then introduced to the design of the experiment.

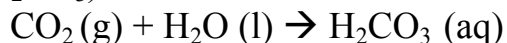
Experimental Design

A single experiment is set up by an adult for multiple classes to observe and analyze; the amount of equipment (*Figure 3*) required is impractical for multiple set-ups. The experiment is simple enough for a science teacher; it is too involved for the middle school learner. It requires multiple and consistent measuring throughout the experiment, and there is a 0.4 molar potassium hydroxide (KOH) solution in the negative control. However, students will make observations, collect data, and perform simulations that will assist them in reaching a final conclusion.

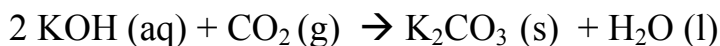
The experiment consists of a set of three half-gallon containers partially filled with solutions that establish different CO₂ levels. Inside each container is a culture of actively growing freshwater *Closterium* algae (*See Figure 4*). The half-gallon container is a closed system that prevents gas exchange with the outside. The beaker of growing algae sits in a liquid reservoir. The reservoir solution can control the amount of CO₂ in the air (*Figure 5*). The *Closterium* algae is grown under a fluorescent light on 12-hour light/dark cycles. Additionally, the system can be modified to trap oxygen gas released by the algae (*Figure 6*).

The experiment consists of three groups: 1) the normal control with a tapwater reservoir; 2) the experimental group with a carbonated water reservoir; and 3) the negative control with a 0.4 M KOH reservoir. The tapwater represents the natural condition of dissolved gases in the environment. The experimental group is a 1:2 mixture of bottled carbonated

water to tapwater, which is enriched with CO₂ gas. CO₂ reacts with water to form carbonic acid (H₂CO₃).



Since 0.4 M KOH reacts with CO₂ to form K₂CO₃ (s), KOH effectively depletes CO₂ from the atmosphere and water of the closed system.



Basic Concepts

Once the experiment is set up, it takes about two weeks to complete. During this time, chemical information is provided on the three main elements that play a part in the carbon cycle: carbon, hydrogen, and oxygen. Students learn about atom structure, electron orbitals, and valence electrons; students determine the electron orbitals and valence electrons for hydrogen through neon (*Worksheets 2 and 3*).

Students are then introduced to covalent bonds (bonds formed when electrons are shared between atoms) and simple molecules (carbon dioxide, hydrogen gas, oxygen gas, methane, ethane, and water.) (*Worksheet 4*). Students demonstrate how to construct the molecules by using marshmallows and toothpicks (*Worksheet 5*).

Students are introduced to the molecules of cellular respiration and photosynthesis: carbon dioxide, glucose, oxygen, and water. The relationship of the products and reactants of the two reactions is discussed; (i.e., that the reactants of photosynthesis are the products of cell respiration and vice versa.) Students are expected to know how to draw the molecules, write out the reactions, and know the relationship between the reactions.



At this stage of the lesson, students should be making connections to organisms and the mode of energy use employed. Students understand that plants use photosynthesis and animals use cellular respiration. Finally, the students acquired knowledge should be connected to the carbon cycle diagram (*Figure 1*).

Skills for Data Collection

Periodically, the students visually examine the algae cultures and record their observations on a data sheet (*See Figure 7*). Students look for visual differences in each container and draw what they observe. Before the end of the experiment, students conduct two simulations: 1) serial dilutions and 2) filtration of algae (*See Figure 8*).

To prepare the students for limiting serial dilutions, students serially dilute out green food color by factors of 10. The students are given four 15 ml test tubes, told to label them 1X, 0.1X, 0.01X, and 0.001X, respectively, and then instructed to transfer 9 ml of water into each test tube with a 10 ml graduated cylinder. The students are then given a test tube with 10 drops of green food color diluted in 10 ml of water. The students are instructed to remove 1 ml from the concentrated food color with a bulb pipette, transfer to the 0.1X test tubes, and mix. The students repeat this process by removing 1 ml from the test tube they just prepared and transferring it to the next test tube. This process is repeated until the food color disappears. The students observe with each dilution, as the color intensity of the dye decreases until it disappears.

Next, the students filter pre-made cultures of algae to see how different amounts of algae produce different color intensities on filter paper. Students label and fold the circular filter paper into a cone, which is placed into a plastic funnel. Groups are given 10 ml each of: undiluted algae, 1:3 diluted algae, 1:10 diluted algae, and 1:100 diluted algae. The students then filter the algae through their filter paper cones. The filtrate is collected into 500 ml plastic soda bottles and the color on the filter paper compared.

Teacher Generated Results

Limiting serial dilutions are done to determine the number of algae growing in each container (*Figure 9*). After the two-week experimentation period, the teacher prepares 12 test tubes for serial dilution: positive control, negative control and another 10 test tubes from 10^{-1} to 10^{-10} . A dozen test tubes are prepared for each container (36 test tubes in all) and placed back under the fluorescent light. Students can observe the test tubes every other day to see if the algae are growing. After 10 days, algae should be in all the test tubes up to the dilution point, where there are no more algae cells.

After the serial dilutions, the teacher filters the remaining algae from each beaker through separate sheets of filter paper. The algae may need to be scraped off the beaker with a spatula. Filtering may take some time, but the filtration setup may run dry briefly without any harm. All of the algae culture should be transferred to the filter paper. Some algae may adhere to the glass beaker, but it should be proportional between the containers. Once the filter paper has dried, the amount of algae from each container can be compared.

The amount of oxygen gas released by each culture is determined by finding the volume between two marks on a test tube. After removing the lid from the plastic container and removing the inverted test tube, a Sharpie marker is used to mark the change in gas volume on the test tube. The test tube is then removed from the inverted funnel, allowing any remaining growth medium to pour out. The gas volume is measured by refilling the test tube with water to the first mark, recording the volume, and then adding more water to the second mark. The difference between the two volumes is the amount of oxygen gas collected.

Results

In our experiment, the students observed that the experimental group with carbonated water grew better than the normal control; the culture was greener and produced more oxygen. The negative control of 0.4 M KOH had little detectable algae growth or oxygen gas production. The serial diluted cultures indicated that the experimental group had 1×10^9 algae per ml, compared to 1×10^7 algae per ml for the normal control and 1×10^5 algae per ml for the negative control. The amount of oxygen gas collected also correlated with algae growth, with 1.6 ml for the experimental group, compared to 0.6 ml for the normal control and no oxygen for the negative control. Each culture was filtered and the results qualitatively supported the above findings (*Figure 10*).

Conclusion

Students were able to see two parts of the carbon cycle through this classroom experiment: that CO_2 dissolved in water diffuses out into the atmosphere of the closed system, and that it can be used by algae through photosynthesis for growth. The students were able to test their hypothesis of whether atmospheric CO_2 levels influence algae growth: they observed and

concluded that CO₂ has a positive influence on algae growth. The increased amount of CO₂ and increased growth of *closterium* algae is evidenced by Figure 10. It produced 100-times more algae per ml than did the normal control. In addition, the negative control showed the essential requirement of CO₂ for algae growth.

Project History

This experiment grew out of collaborative work in the Ocean Science Education Institute (OSEI), which develops and implements high quality ocean science education for middle school students through projects that connect with existing district curriculums and effective science educational practices. OSEI is a project of the Center for Ocean Science Education Excellence-New England (COSEE-NE), an NSF-funded partnership between the New England Aquarium, the University of Massachusetts/Boston, and the Woods Hole Oceanographic Institution (WHOI). The OSEI format includes a five-day workshop, numerous classroom visits, and two follow-up days. During the 2004-2005 school year, researchers and Massachusetts middle school teachers, district science coordinators, and facilitators teamed up to produce district-wide, inquiry-based science curricula for middle school students based on current ocean science research. To find out more about OSEI and other COSEE-NE programs, please visit our website at www.cosee-ne.net.

Figure 1: Carbon Cycle Diagram

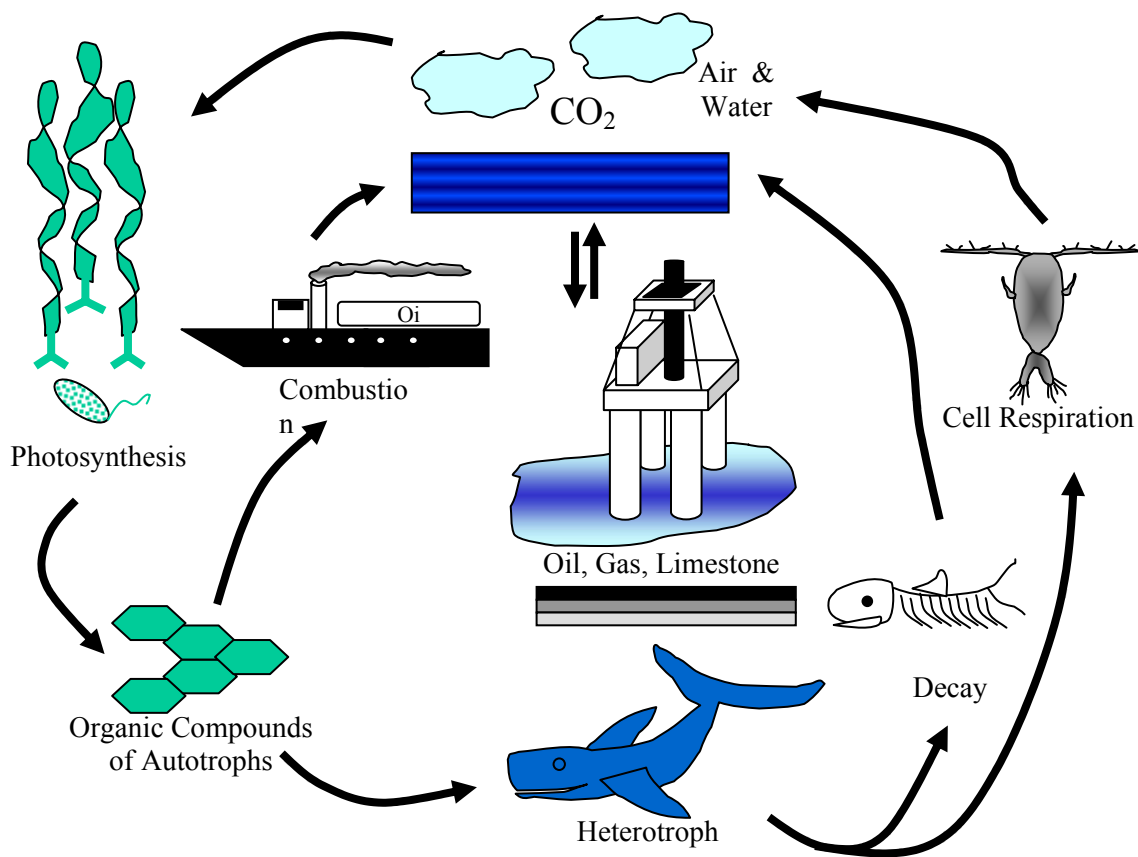


Figure 2. Sample Pre-Assessment Questions

- Carbon dioxide is produced by the human body through _____.
 - excretion
 - respiration
 - photosynthesis
 - sweating
- When humans eat sugar, the sugar is broken down and released as _____.
 - water
 - oxygen
 - carbon dioxide
 - salt
- During the day, plants _____ to make energy.
 - need oxygen
 - need carbon dioxide
 - need carbohydrates
 - produce oxygen
- Carbon dioxide can be found in
 - the oceans
 - forests
 - the atmosphere
 - All of the above

5. Carbon dioxide is found in
 - A. seashells
 - B. limestone
 - C. Crabs shells
 - D. All of the above
6. High amounts of carbon dioxide is thought to be causing
 - A. skin cancer
 - B. hurricanes
 - C. global warming
 - D. All of the above
7. Most of the energy in the USA comes from
 - A. fossil fuels
 - B. solar power
 - C. nuclear power
 - D. wind power
8. High amounts of carbon dioxide in the air comes from
 - A. burning coal
 - B. using gasoline
 - C. burning heating fuel
 - D. All of the above
9. The major store of carbon dioxide is in
 - A. deep ocean
 - B. fossil fuels
 - C. sedimentary rock
 - D. the atmosphere
10. Carbon dioxide is _____ important to me.
 A. very B. somewhat C. a little D. not
11. List all the things that play a role in the carbon cycle.
12. Draw as much of the Carbon Cycle as you can.

Figure 3: Supplies Needed for the Experiment

Numbers in parentheses indicate the number needed.

- 2 liters of Distilled Water (Grocery store)
- Order culture of actively growing *Closterium* algae (Carolina Biological Supply #HT-15-2115, \$ 4.95)
- Tube 50X Alga-Gro® : (Carolina Biological Supply #HT-15-3751, \$27.60)
- Schultz, 10-15-10 Plant Food Plus (Wal-Mart or Kmart, \$2.95)
- (3) 1/2 Gal Rubbermaid ® Screw-cap containers (Wal-Mart or Kmart)
- (4) 600 ml glass Pyrex beakers (Ball Canning jars work as a substitute)
 Must be able to fit inside 1/2 Gal container
 Plastic containers may have impurities that inhibit algae growth
 Beakers are cleaned and rinsed twice with distilled or bottled water

- 0.5 liter of carbonated water (Tonic water)
- 0.6 liter 0.4 Molar potassium hydroxide solution (Chemistry Teacher)
- 500 milliliter (ml) graduated cylinder
- 50 ml graduated cylinder
- Rubberband
- Plastic wrap
- Glass marking pen
- (2) Fluorescent light
- Electric timer

Additional Supplies for Collection of Oxygen Gas

- (3) 1/2 Gal Rubbermaid ® Screw-cap containers
- (3) 600 ml glass beakers
- (3) glass funnels
- (3) 12 milliliter glass test tubes (Need to fit over stem of inverted funnel)
- Parafilm

Figure 4: Growing Closterium Algae

- Add 50X-Alga-Gro® to 1 liter of distilled water to make 1X-Alga-Gro®
- Transfer to the glass beaker 400 ml of 1X-Alga-Gro®
- Add 1 drop of Plant Food
- Transfer half the volume of the *Closterium* algae culture to the beaker
- Cover with plastic wrap and secure with a rubber band
- Punch six holes in the plastic wrap
- Place in a sunny window or under a fluorescent light for about a week
- As the culture grows, the green color will intensify.

Figure 5: Experimental Set Up

- Transfer into each half-gallon plastic container 600 ml of reservoir liquid
 - Normal: 600 ml of tap water
 - Experiment: 400 ml of tap water and 200 ml of carbonated water
 - Negative: 600 ml of 0.4 M potassium hydroxide
- Label 600 ml beakers Normal, Experiment, Negative with a Sharpie Pen
- Transfer 350 ml of 1X-Alga-Gro® to each beaker
- Transfer 50 ml of actively growing algae into each beaker
 - Resuspend the algae after each transfer
- Carefully insert beakers into half-gallon container
- Close and wrap with parafilm

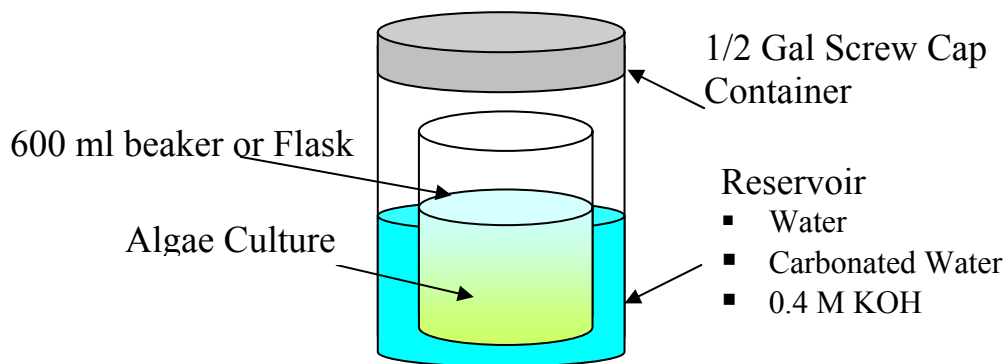


Figure 6: Procedure for Collecting Release of Oxygen Gas.

- Set up the same as in above figure, but do not seal
- Let the algae settle for 10 minutes
- Invert the glass funnel and slowly place on the bottom of flask
- Fill the test tube with 1X-Alga-Gro® to the very top
- Tightly wrap with parafilm (Should be no air)
- Invert the test tube, puncture the parafilm and slide over the funnel stem
- Insure the parafilm is not sealing the funnel stem opening
- If parafilm can not be removed, then repeat last three steps
- With a Sharpie Pen, mark the air-water interface, and seal system

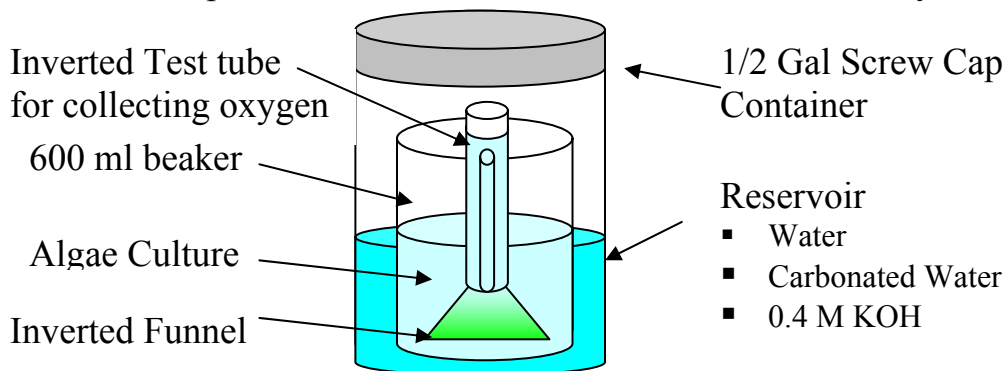


Figure 7: Student Data sheet.

Data sheet for recording observations of carbon dioxide on algae growth. Observe the color of each beakers inside the containers. Record the color of the algae you observe. The intensity of the color matches the intensity of algae growth. Record other data in the rows provided.

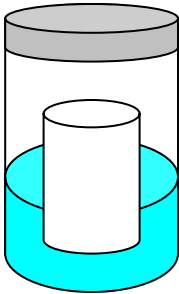
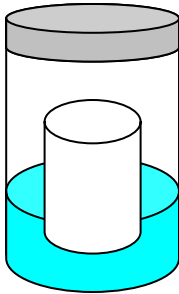
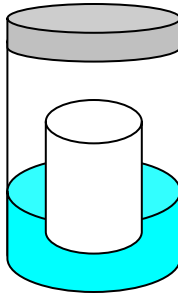
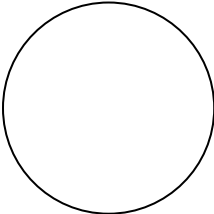
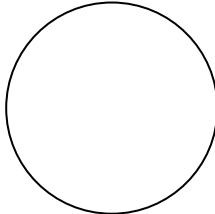
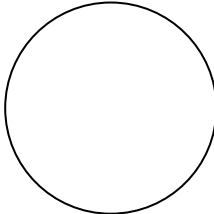
	Normal Control	Experimental Group	Negative Control
Color Intensity			
Volume of oxygen collected	mL	mL	mL
Color intensity of filter paper			
Serial Dilution Limit			

Figure 8: Supplies for Simulations per group

Serial Dilutions	
▪	(4) 15 ml test tubes
▪	Test tube rack
▪	10 ml graduated cylinder or 10 ml seriological pipet
▪	A 1 ml bulb pipet or 1 ml seriological pipet with bulb
▪	Glass marking pen
▪	1 test tube of green food coloring for dilution (10 drops into 10 ml of water)
Algae Filtration	
▪	(4) 100 mm pieces of circular filtration paper
▪	Plastic funnel (size ???)
▪	500 ml plastic bottle (Soda pop bottle)

Figure 9: Set up of Serial Dilutions

Label	Dilution	1X-Alga-Gro	Addition of Algae
P	1/2	5 ml	5 ml of cultured Algae
1	10^{-1}	9 ml	1 ml of cultured Algae
2	10^{-2}	9 ml	1 ml of Algae diluted to 10^{-1}
3	10^{-3}	9 ml	1 ml of Algae diluted to 10^{-2}
4	10^{-4}	9 ml	1 ml of Algae diluted to 10^{-3}
5	10^{-5}	9 ml	1 ml of Algae diluted to 10^{-4}
6	10^{-6}	9 ml	1 ml of Algae diluted to 10^{-5}
7	10^{-7}	9 ml	1 ml of Algae diluted to 10^{-6}
8	10^{-8}	9 ml	1 ml of Algae diluted to 10^{-7}
9	10^{-9}	9 ml	1 ml of Algae diluted to 10^{-8}
10	10^{-10}	9 ml	1 ml of Algae diluted to 10^{-9}
N	None	10 ml	None

Figure 10: Summary of Experimental Results

	Normal	Experimental	Negative
Growing Algae Color Intensity	Green	Dark Green	Light Green
Algae Concentration (Cells/ml)	1×10^7	1×10^9	1×10^5
Oxygen Gas (ml)	0.6	1.6	0.1
Filter Algae Color Intensity	+++	+++++	+

Worksheet 1: Scavenger Hunt

Name : _____ **Date:** _____ **Group :** _____ **Form: Answer Sheet**

Your teacher created 8 different worksheets, each worksheet has only one part of the carbon cycle filled in. Students will work individually to go around the room and find other students that have the parts of the carbon cycle that they were missing. Students fill in their sheets until they had all parts of the carbon cycle. Students must orally share answers, no copying. Students could accomplish the same results using their notebooks and texts.

1. Photosynthesis What is it? Examples of this:	2. Organic compounds of Autotrophs What is it? Examples of this:	3. Heterotrophs What is it? Examples of this:
4. Cell Respiration What is it? Examples of this:	5. CO₂ What is it? Examples of this: ...in the air ...in the ocean	6. Combustion What is it? Examples of this:
7. Oil and Gas How is it formed? Examples of this:	8. Limestone How is it formed? Examples of this:	9. Decay What is it? Examples of this:

Worksheet 2: Electron Placement

Name : _____ Date: _____ Group: _____ Form: *Answer Sheet*

Use a Periodic Table of Elements to answer the following questions then follow the directions for electron placement. You can diagram elements 1-18 using these rules.

For each element fill in the following information on the diagram.

1. What is the name of the element?
2. What is the chemical symbol?
3. What is the atomic number for this element?
4. How many electrons are in the atom?
5. How many shells in your diagram have electrons placed on them?
6. How many electrons did you draw in your last (outer) shell?
7. How many valance electrons are available for bonding?

Rules for electron placement:

1. An element's atomic number is equal to the number of its protons.
2. Atoms have the same number of protons and electrons.
3. An atom's protons and neutrons are found in the nucleus. The electrons are found in the shells.
4. Each shell can hold only a certain number of electrons.

Working from the inside to the outside,

The 1st shell holds 2 electrons.

The 2nd shell holds 8 electrons.

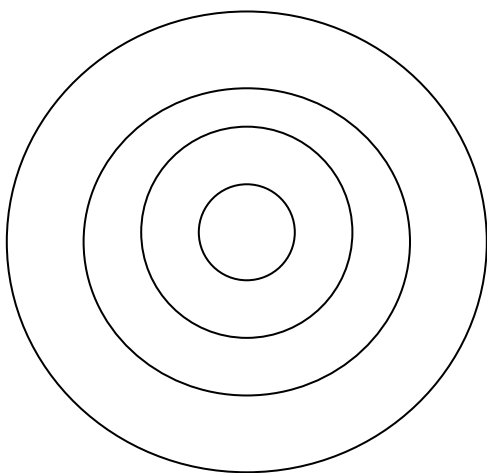
The 3rd shell holds 8 electrons.

The 4th shell holds 18 electrons.

The 5th shell holds 18 electrons.

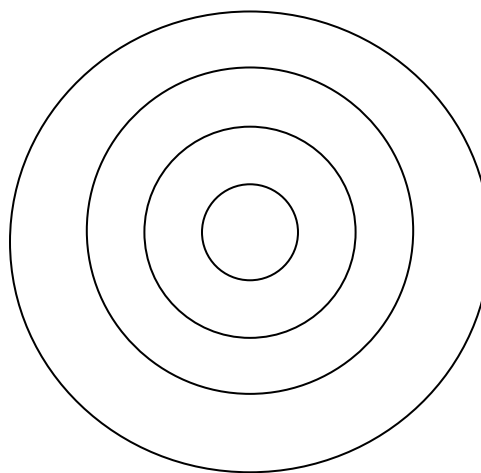
5. Working from the inside out, you must fill one shell completely before adding shells.

Element Name _____
Chemical Symbol _____
Atomic Number _____
Number of Electrons _____



Number of Shells _____
Number of electrons on last shell _____
Valance Electrons _____

Element Name _____
Chemical Symbol _____
Atomic Number _____
Number of Electrons _____



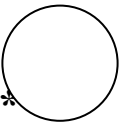
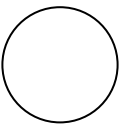
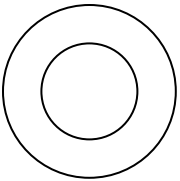
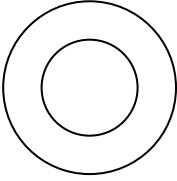
Number of Shells _____
Number of electrons on last shell _____
Valance Electrons _____

This was adapted from the Massachusetts DOE, Adult Basic Education "Science Box" curriculum.

Worksheet 3: Electron Placement / Configuration Activity

Name _____ Date _____ Group _____ Form: *Answer Sheet*

Draw the placement of the electrons on each of the diagrams below for each element. Record the number of shells it has and the number of valance electrons. In the last column draw the Lewis Dot structure for each element.

Atomic # symbol	Element name	Diagram	Number of shells	Number of valance electrons	Lewis Dot
1. H	Hydrogen		1	1	H ·
2. He	Helium				
3. C	Carbon				
4. O	Oxygen				

This activity was adapted from DOE Chemistry Institute, Mark Greenman, 2004.

Worksheet 4: Simple Molecules

Name _____ Date _____ Group _____ Form: *Answer Sheet*

Using covalent bonds and the number of valence electrons for the number of atoms in the molecule sketch the indicated molecule.

Molecule	Chemical Formula	Draw Structural Formula
Hydrogen Gas	H ₂	
Oxygen Gas	O ₂	
Water	H ₂ O	
Carbon Dioxide	CO ₂	
Methane	CH ₄	
Ethane	C ₂ H ₈	
Glucose	C ₆ H ₁₂ O ₂	

Worksheet 5: How Many Molecules?

Name _____ Date _____ Group _____ Form: *Answer Sheet*

Each group will need a plastic bag filled with 45 white mini-marshmallows, 15 of another color and 50 toothpicks. Go to page 116 in Chemical Building Blocks and follow the directions for building hydrocarbons. After building each molecule, draw its structural formula. Answer questions 1-5 on the back of this paper.

Hydrocarbon	Chemical Formula	Draw Structural Formula
Methane	CH ₄	
Ethane	C ₂ H ₆	
Propane	C ₃ H ₈	
Butane	C ₄ H ₁₀	
Isobutane	C ₄ H ₁₀	
Pentane	C ₅ H ₁₂	
Isopentane	C ₅ H ₁₂	

This lab was adapted from Prentice Hall's Science Explorer Chemical Building Blocks page 116.