Name ______________________

Hour _____

AP Biology Lab: Dissolved Oxygen in Aquatic Ecosystem: The Relationship between Temperature, DO, and Respiration Rates

Although water is composed of oxygen and hydrogen atoms, biological life in water depends upon another form of oxygen—molecular oxygen. Oxygen is used by organisms in aerobic respiration, where energy is released by the combustion of sugar in the mitochondria. This form of oxygen can fit into the spaces between water molecules and is available to aquatic organisms.

Fish, invertebrates, and other aquatic animals depend upon the oxygen dissolved in water. Without this oxygen, they would suffocate. Some organisms, such as salmon, mayflies, and trout, require high concentrations of oxygen in their water. Other organisms, such as catfish, midge fly larvae, and carp can survive with much less oxygen. The ecological quality of the water depends largely upon the amount of oxygen the water can hold.

The following table indicates the normal tolerance of selected animals to temperature and oxygen levels. The quality of the water can be assessed with fair accuracy by observing the aquatic animal populations in a stream.

Table 1

Animal	Temperature Range (°C)	Minimum Dissolved Oxygen (mg/L)
Trout	5–20	6.5
Smallmouth bass	5–28	6.5
Caddisfly larvae	10–25	4.0
Mayfly larvae	10–25	4.0
Stonefly larvae	10–25	4.0
Catfish	20–25	2.5
Carp	10–25	2.0
Mosquito	10–25	1.0
Water boatmen	10–25	2.0

A type of relatively unheard of pollution is thermal pollution. One source of thermal pollution comes from nuclear and conventional power plants. Power plants use vast amounts of water that are converted to steam by the thermonuclear and combustion reactions. This steam is used to turn the blades of the turbines which are used to turn the generators producing the electricity. The hot water resulting from condensed steam is partially cooled in specially designed towers before it is released again into the environment by means of a reservoir or stream. The temperature of the returned water is great enough to raise the temperature of the body of water several degrees. Such increases in temperature could greatly affect the organisms living in the water.

In an aquatic environment, oxygen must be in a solution in a free state (O_2) before it is available for use by organisms (bio-available). Its concentration and distribution in the aquatic environment are directly dependent on chemical and physical factors and are greatly affected by biological processes. In the atmosphere, there is an abundance of oxygen, with about 200 mL of oxygen/1 L air. In an aquatic environment, there are about 5-10 mL O_2/1 L water. The concentration of the oxygen in aquatic environments is a very important component of water quality.

At 20°C, oxygen diffuses 300,000 times faster in air than water, making the distribution of oxygen in air relatively uniform. Spatial distribution of oxygen in water, on the other hand, can be highly variable, especially in the absence of mixing by currents, winds, and tides.
Other chemical and physical factors – such as salinity, pH, and especially temperature- can affect the dissolved oxygen (DO) concentration and distribution. Salinity, usually expressed as parts per thousand (ppt), is the content of dissolved salts in water. Generally, as temperature and salinity increase, the solubility of oxygen in water decreases.

The partial pressure of oxygen in the air above the water affects the amount of DO in the water. Less oxygen is present at higher elevations since the air itself is less dense; therefore the water at high elevations contains less oxygen. At 4,000 meters in elevation (13,000 feet), the amount of dissolved oxygen in water is less than two-thirds what it is at sea level. All of these physical factors, along with oxygen concentration, work together to increase diversity in aquatic habitats.

Oxygen from the atmosphere is mixed into the water through diffusion. However, more oxygen is mixed into the water with the help of winds, rain, waves, and currents. The faster the water moves, the more dissolved oxygen the water will contain since it has more contact time with the air. The process of photosynthesis (underwater plants and algae) occurring in the water affects the number and kinds of animals found there. Healthy streams are saturated with oxygen (90 to 110% saturation) during most of the year.

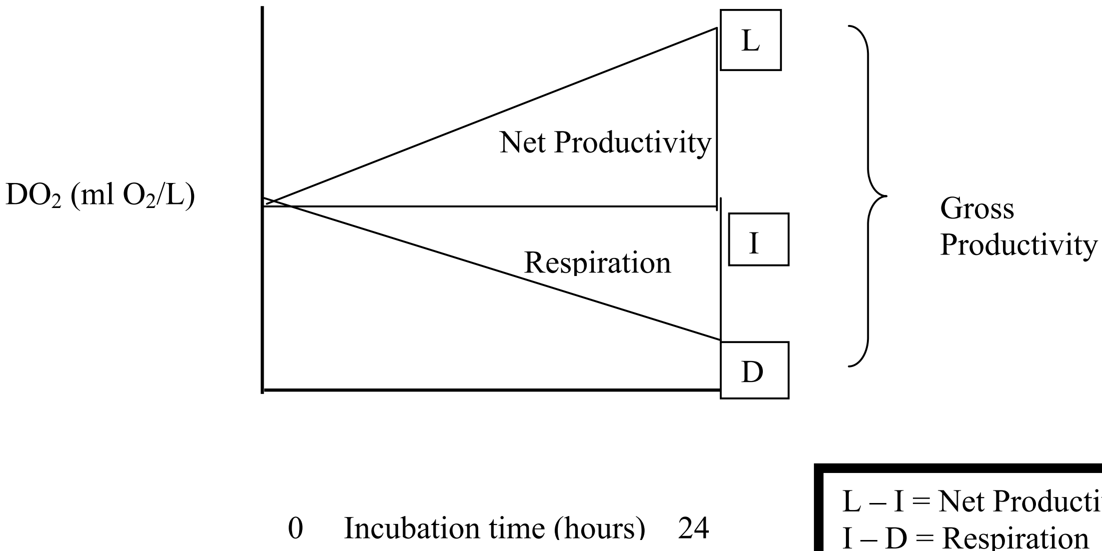
Biological processes, such as photosynthesis and respiration, can also significantly affect DO concentration. Photosynthesis usually increases the DO concentration in water. Aerobic respiration requires oxygen and will usually decrease DO concentration. ***The measurement of the DO concentration of a body of water is often used to determine whether the biological activities requiring oxygen are occurring and consequently, it is an important indicator of pollution.***

The **primary productivity** of an ecosystem is defined as the **rate** at which organic materials (carbon-containing compounds) are stored. Only those organisms possessing photosynthetic pigments can utilize sunlight to create organic compounds from simple inorganic substances. Green plants obtain carbon for carbohydrate synthesis from the carbon dioxide in the water or the air according to the basic equation for photosynthesis:

$$6CO_2 + 6\ H_2O \rightarrow C_6H_{12}O_6 + 6O_2$$

The rate of carbon dioxide utilization, the rate of formation of organic compounds, or the rate of oxygen production can be used as a basis for measuring primary productivity. A measure of oxygen production over time provides a means of calculating the amount of carbon that has been bound (taken up by photosynthesis) over a period of time. For each mL of oxygen produced, approximately 0.536 mg of carbon has been assimilated.

One method of measuring the rate of oxygen production is **the light and dark bottle method**. In this method, the DO concentrations of samples of ocean, lake, or river water or samples of laboratory algal cultures are measured and compared before and after incubation in light and darkness. The difference between the measurements of DO in the initial and dark bottles is an indication of the amount of oxygen that is being consumed in respiration by organisms in the bottle. In the bottles exposed to light, the biological process of photosynthesis and respiration are occurring; therefore the change over time in DO concentration from the initial concentrations is the measure of **net productivity**. The difference over time between the DO concentrations in the light bottle and the dark bottle is the total oxygen production and therefore an estimate of gross productivity.



L – I = Net Productivity
I – D = Respiration
L - D = Gross Productivity (or, think of it as L – I + I – D)

Respiration Rate and Temperature

Part A: Respiration Rate Data

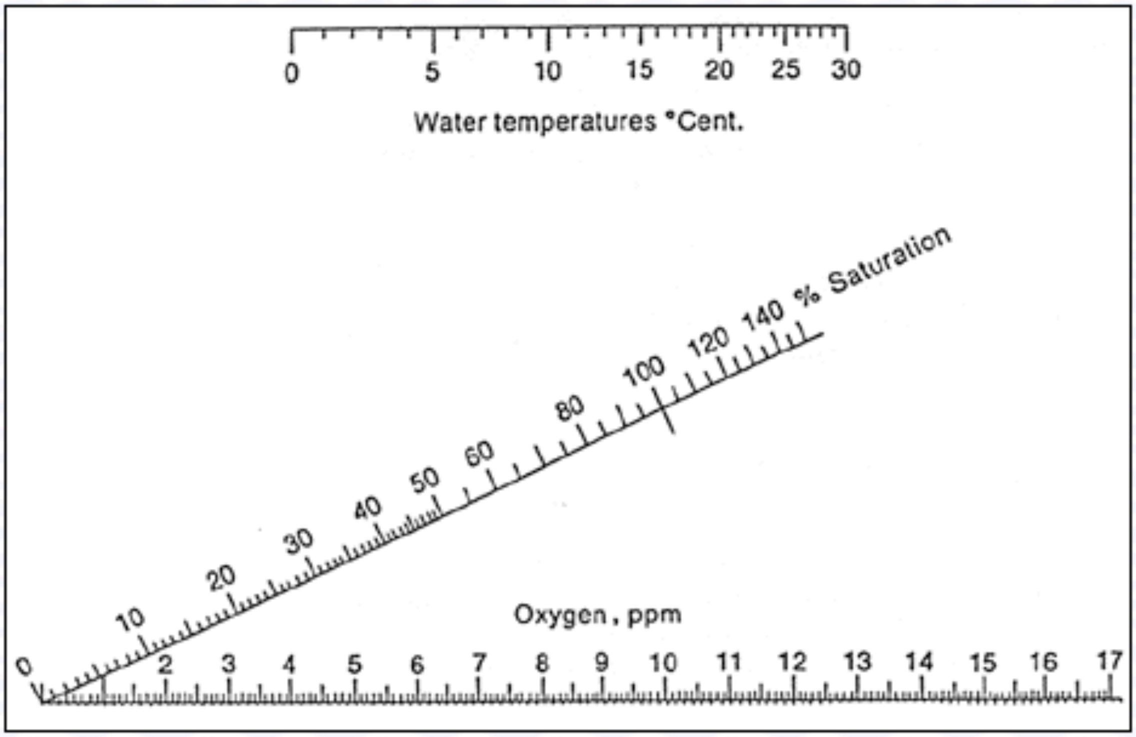
Equipment needed (for each lab group): 1000 ml beaker, 250 mL beaker, , gallon size Ziploc bag, a carp (goldfish), CBL with dissolved oxygen probe, thermometer, hot and cold water baths, stirring rod

1. Place 400 ml. of tap water into a 1000 ml beaker. This will serve as your water bath.
2. Obtain a gallon Ziploc bag. Place a goldfish with 250 mL tank water inside it.
3. Now put the bag with the goldfish into the water bath. Allow the goldfish to sit for at least three minutes before you proceed!
4. Record the temperature of the beaker the goldfish is in. Try not to agitate the goldfish too much!
5. Allow goldfish to quiet down for another minute after taking the temperature.
6. Record the gill beat rate by counting the movement (beat) of the operculum which covers the gills.Take the count for 30 sec. and multiply by two to obtain rate per minute and then record the count in your data table.
7. Remove the bag with the goldfish from the water bath. Change the temperature of your water bath using ice. DO NOT CHANGE THE TEMPERATURE BY MORE THAN 5C
8. Repeat steps 3-7 above two more times.
9. Remove the bag with your goldfish from the water bath. Let the goldfish come back to room temperature by letting it sit for several minutes.
10. Change the temperature of your water bath using warm water. DO NOT CHANGE THE TEMPERATURE BY MORE THAN 5C.
11. Repeat data collection for three trials. Record Results in table.

	Room Temp	5 C colder	5 C warmer
Gill count			
Average			
DO			
% O2 Sat			

Part B: **Dissolved Oxygen and Temperature**

1. Fill three of the water sampling bottles with water of the three different temperatures provided. Use the water baths for this portion of the lab.
2. Using the dissolved oxygen test kits provided, determine the amount of DO per sample.
3. You will be provided with a nomograph for oxygen saturation. Use a straight edge or ruler to determine the percent DO in each of your temperature samples. (To use a nomograph, you line up the edge of a ruler with the temperature of the water on the top scale and the DO on the bottom scale, and read the percent saturation on the middle scale.)



Part C: A Model of Productivity as a Function of Depth in a Lake (Here is the procedure, you have been given the DO to complete the lab.)

1. Obtain seven water-sampling bottles. Fill all of the bottles COMPLETELY with the lake water provided.
2. Using tape, mark the cap of each bottle. Mark as follows: I (for initial), D (for dark), 100%, 65%, 25%, 10%, 2%.
3. Determine the DO for the "Initial" bottle now and record its value. We will also calculate a class mean. This is the amount of DO that the water has to start with (a baseline).
4. Cover the "Dark" bottle with aluminum foil so that no light can enter. In this bottle no photosynthesis can occur, so the only things that will change DO will be the process of respiration by all of the organisms present.
5. The attenuation of natural light that occurs due to depth in a body of water will be simulated by using window screen. Wrap screen layers around the bottles in the following way: 100% light – no screens; 65% light – 1 screen layer; 25% light –3 screen layers; 10% light –5 screen layers; and 2% light- 8 screen layers. The bottles will be on their sides under the lights, so remember to cover the bottom of the bottles to prevent light from entering there. Use rubber bands or clothes pins to keep the screens in place.
6. Place the bottles on their sides under the bank of lights in the classroom. Tomorrow we will continue…

Day Two

1. To complete this experiment, determine the DO in all of the bottles that have been under the lights as well as the dark bottle's DO. Use the "dark" DO in the table below to calculate gross productivity. Calculate the respiration rate using the formula in the table. Record values for other bottles. Complete the calculations to determine the Gross and Net Productivity for each bottle. The calculations will be based on a time period of one day.

Data: Productivity of Screen-Wrapped Sample

# of screens	% light	DO mL O2/ L	Gross Productivity (light bottle – dark bottle)	Net Productivity (light bottle – initial bottle)	Gross Productivity (mgC/ m^3)
0	100%	6.4			
1	65%	5.8			
3	25%	4.5			
5	10%	3.7			
8	2%	4.0			
Foil		4.6			
Initial		9.2			

Graph both net and gross productivities as a function of light intensity (class means).

Questions:

1.What is thermal pollution? How does it affect dissolved oxygen levels. How does this impact the rate of respiration in fish?

2. What happens to fish respiration as dissolved oxygen levels drop? Why? What is the relationship between oxygen production and assimilation of carbon?

3. Refer to your graph of productivity and light intensity…At what light intensity do you expect there to be:

   No gross productivity?___________ No net productivity?_________

4. A mammal uses only 1 to 2 percent of its energy in ventilation (breathing air in and out) while a fish must spend about 15 percent of its energy to move water over its gills. Explain this huge difference in their efforts to collect oxygen, i.e., why is it necessary for the fish to invest so much more energy in the "oxygen acquisition" process?

5. Would you expect the DO in water taken from a stream entering a lake to be higher or lower than the DO taken from the lake itself? Explain.

6. Would you expect the DO concentrations of water samples taken from a lake at 7:00 a.m. to be higher or lower than samples taken at 5:00 p.m.? Explain.

7. In general, how would you expect turbidity to affect the productivity of a lake? Why?