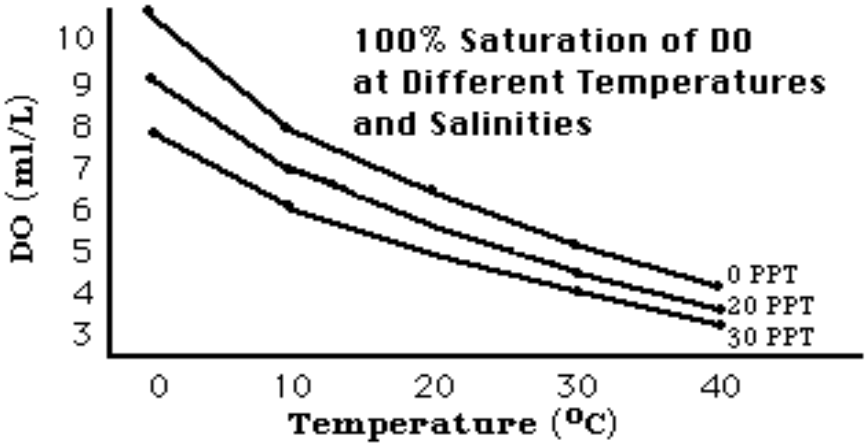
Dissolved Oxygen and Aquatic Primary Productivity[1]

INTRODUCTION[2]

Oxygen is critical to the maintenance of the life processes of nearly all organisms. In the aquatic environment, oxygen must be in solution in a free state (O_2) before it is available for use by organisms. 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 milliliters of oxygen for every liter of air. Conversely, in the aquatic environment there are only about 5 to 10 milliliters of dissolved oxygen (DO) in a liter of water (20 to 40 times less). The measurement of the oxygen in aquatic environments can be a very important indicator of water quality.

Because we live in air, it is hard for us to appreciate the three-dimensional variation in availability of DO that presents itself to aquatic organisms. At 20 ^{o}C, oxygen diffuses 300,000 times faster in air than in 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, tides, or natural flows.[3]

Other factors, both chemical and physical--such as salinity, pH, and especially temperature--can affect the DO concentration and distribution. Salinity, usually expressed in 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. See the figure below.



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 water at higher elevations contains less oxygen. At 4000 meters in elevation (about 13,000 feet), the amount of dissolved oxygen in water is less than two-thirds that at sea level. All these physical factors work together to increase diversity in aquatic habitats with regard to oxygen availability.[2]

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[2]This lab is an abbreviated version of Laboratory 12 of the same name in the *Advanced Placement Biology Laboratory Manual for Students* (Edition C) written and developed by Walt McDonald and William Barstow. The manual is published by The College Board, 1990.

[3]From "Dissolved Oxygen" by Valerie C. Chase in *Carolina Tips*, 5/1/88.

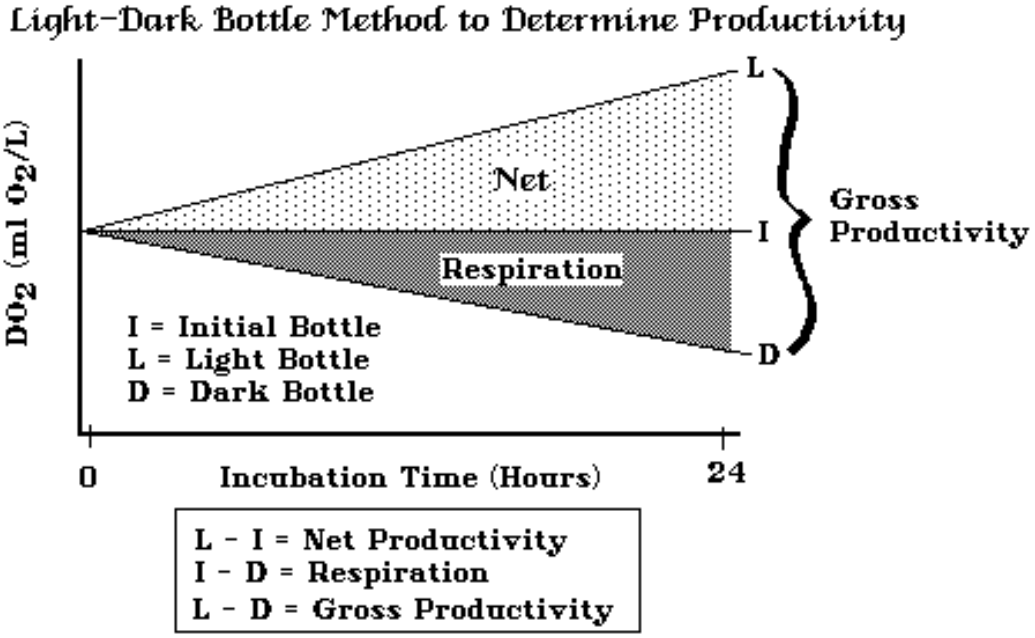
Biological processes, such as photosynthesis and respiration, can also significantly affect DO concentration. Photosynthesis usually increases the DO concentration in water. Respiration usually requires oxygen and will usually decrease DO concentration. The measurement of the DO concentration of a body 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 fertility of any body of natural water depends on the productivity of the green plants within it. The **primary productivity** of an ecosystem is defined as the **rate** at which sunlight is stored by plants in the form of organic materials (carbon-containing compounds). Only those organisms possessing the green pigment chlorophyll can utilize sunlight to create new organic compounds from simple inorganic substances. These green plants use carbon for carbohydrate synthesis from the carbon dioxide dissolved in the water according to the basic equation for photosynthesis:

$$6CO_2 + 6H_2O \text{ --------> } C_6H_{12}O_6 + 6O_2$$

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 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 in organic compounds over a period of time. For each milliliter of oxygen produced, approximately 0.536 milligrams 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 after incubation in light and darkness. The difference between the initial and dark bottles is an indication of the amount of oxygen that is being consumed in respiration by the organisms in the bottle. In the bottles exposed to light, the biological processes of photosynthesis and respiration are occurring; therefore, the change in DO concentration from the initial concentration over time is a 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**. See the figure below:



PROCEDURES

You will use the **LaMotte Test Kits** to determine the concentration of DO in the various samples you work with. The process involves adding some chemicals to the water to "fix" the free oxygen (O_2). Once the O_2 is fixed, you will add an acid powder, some starch, and titrate to determine the DO. The endpoint of the titration is fast and dramatic, and easy to see. Follow the instructions inside the lid of the kit, and you will be able to find the DO in PPM (parts per million) or mg O_2 /L. This measurement is read directly from the syringe when you reach your endpoint of the titration. Be sure to wear **safety goggles and gloves** when using the kit.

PART 1: DISSOLVED OXYGEN AND TEMPERATURE

A. Fill three of the water sampling bottles with water of the three different temperatures provided. Determine the DO of each sample. This is a very wet lab so try not to make too big a mess. Record these values in Table 1 below.

B. On the nomograph of oxygen saturation on the next page, use a straightedge or ruler to estimate the percent saturation of DO in your samples and record this value in **Table 1**. Line up the edge of a ruler with the temperature of the water on the top scale and the DO on the bottom scale. Read the percent saturation off of the middle scale.

C. Record your values on the board, and then enter class means in **Table 2**. In your write-up, graph the percent saturation as a function of temperature.

D. Graph % DO saturation as a function of temperature using the class means. Compare this graph to the graph on page 1 of this lab. Discuss the difference when you answer question 1 at the end of the lab.

Table 1
Group Temperature/DO Data:

Temperature	DO (mg/L)	%DO Saturation

Table 2
Class Mean Data:

Temperature	DO (mg/L)	%DO Saturation

PART 2: PRODUCTIVITY AS A FUNCTION OF DEPTH IN A LAKE

Day One:

A. Obtain 7 water sampling bottles (these are also called BOD bottles for "biological oxygen demand"). Fill all the bottles with the water provided. (You may be asked to add a specific weight of aquatic plant leaves into each bottle.) Be careful not to leave an air bubble at the top of the bottle. Using labeling tape, put a small label on the **cap** of each bottle. Mark the labels as follows: I (for "initial"), D (for "dark"), 100%, 65%, 25%, 10%, and 2%.

B. Fix and titrate the "Initial" bottle right away and record this DO value in **Table 3** below and in the data table on the board. Record the class "Initial" bottle mean in **Table 3**. This is the amount of DO that the water has to start with (a baseline). Empty and clean out this bottle.

C. Cover the "Dark" bottle with aluminum foil so that no light can enter. In this bottle no photosynthesis can occur, so the only thing that will change the DO will be the process of respiration by <u>all</u> of the organisms present.

D. The attenuation (decrease) of natural light that occurs due to depth in a body of water will be simulated by using neutral density plastic window screens. Wrap screen layers around the bottles in the following pattern: 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 lie on their sides under the lights so remember to cover the bottom of the bottles to prevent light from entering. Use rubber bands or clothespins to keep the screens in place.

E. Place the bottles on their sides under the bank of lights in the classroom. Be sure to turn the bottles so that their labels are down and not blocking the light from getting to the contents.

Day Two:

A. Using the **LaMotte kits**, determine the DO in all the bottles that have been under the lights. Record the "Dark" bottle DO in **Table 4**. Calculate the respiration rate using the formula in the table. Record the values for the other bottles in **Table 5**. Complete the calculations in Table 5 to determine the Gross and Net Productivity in each bottle. The calculations will be based on a time period of one day. Enter your respiration rate, gross, and net productivity in the data table on the board to determine class means. Enter these means in **Tables 6 and 7**.

B. **<u>Graph both net and gross productivity as a function of light intensity (use both group data and class means)</u>**. The two kinds of productivity may be plotted on the same graph. Make sure you include a key (4 colors or 4 symbols). The y-axis will need to show negative values.

Table 3:

Initial DO in PPM (Group Data)	**Initial DO** (Class mean)

Table 4:

DO (PPM) Dark Bottle (Group Data)	**Respiration Rate** [Initial Bottle (PPM)-Dark Bottle (PPM) /day]

Table 5:

% Light	**DO in PPM** (Group Data)	**Gross Productivity** [Light Bottle (PPM) - Dark Bottle (PPM)/day]	**Net Productivity** [Light Bottle (PPM) - Initial Bottle (PPM)/day)]
100%			
65%			
25%			
10%			
2%			

CLASS MEANS

Table 6
Class Productivity Means:

% Light	**Gross Productivity** (PPM/Day)	**Net Productivity** (PPM/Day)
100%		
65%		
25%		
10%		
2%		

Table 7
Class Respiration Mean:

Respiration Rate (PPM/Day)	

QUESTIONS **Use complete sentences on a separate sheet of paper**.

1. Using your graph of the temperature data, what is the effect of temperature on the amount of oxygen water at different temperatures can hold? Why does the graph of % saturation as a function of temperature go up while the one on the first page of this lab goes down?

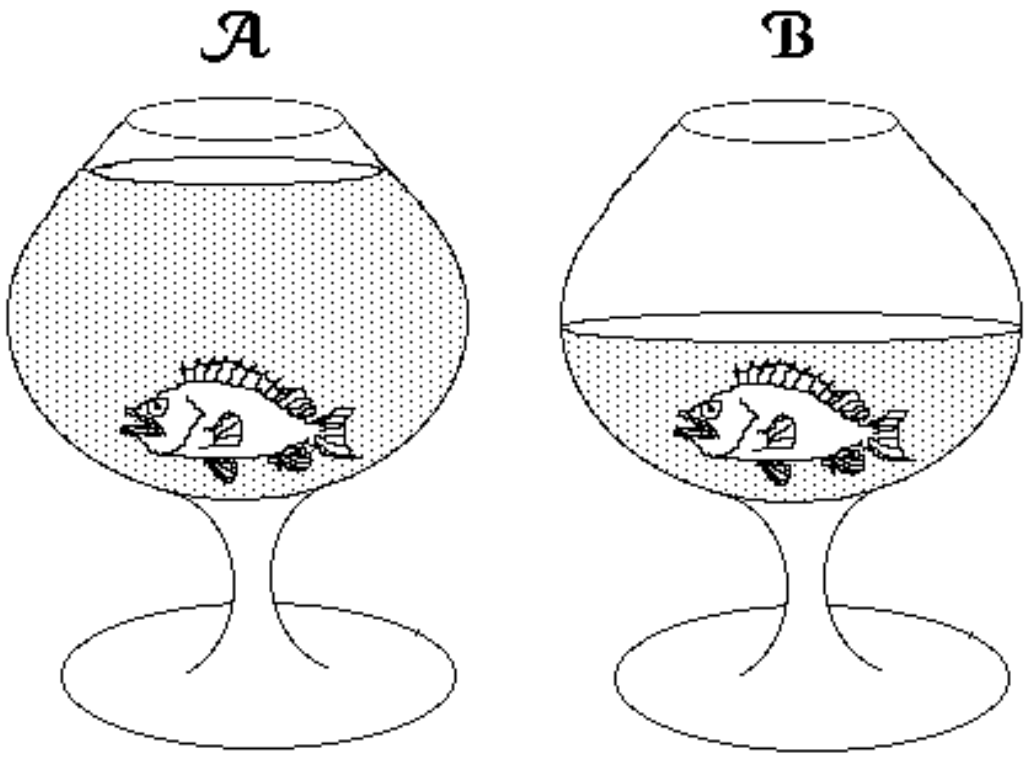
2. At what light intensity do you expect there to be no gross productivity? Net productivity? [Use your graph – go with the class data]

3. An animal 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 effort to collect oxygen. [Read lab introduction and think carefully]

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

5. Would you expect the DO concentration of water samples taken from a school's lake at 7:00 AM to be higher or lower than samples taken at 5:00 PM? Explain.

6. In the following drawings of identical containers with identical fish, but with different volumes of water, which one, A or B, would have more oxygen available to the fish? Explain. [Be careful, this requires a fair degree of logical explanation]



7. What is eutrophication? Explain why it can be very hazardous to put nitrogen or phosphorous fertilizers in a body of water?

8. Describe the relationship between dissolved oxygen and the processes of photosynthesis and respiration.

Preparator's Guide

Time Requirements: Two or three 45-minute laboratory periods. Procedures Part 1 and Day One of Part 2 can be done during the first period, and Day Two of Part 2 can be done during the second period.

Student Materials and Equipment:

Item	Quantity / group
LaMotte Dissolved Oxygen Kits	1 (LaMotte #7414/EDO)
BOD Bottles	7 (LaMotte #0688DO)
Aluminum foil	(enough to wrap one BOD bottle)
Plastic window screen	17 (9" x 12" or smaller to cover BOD bottles)
safety goggles	1 for each student in group
plastic gloves	1 pair for each student in group
tap water	500 mL of each temperature
lake water	1 liter
straight edge	1
bright light source for 24 hours	1 per class
masking tape for labeling	1 roll shared among several groups
Chlorella or *Elodea* for lake water spiking	

Preparation Suggestions:

1. I have had better luck with real lake water than with algal cultures, but this was in the summer time. If you spike your culture with *Chlorella* or some other purchased algae, don't put in too much because this makes it difficult for the LaMotte kits to function within their normal limits. Sprigs or leaves of *Elodea* may make a good substitute for algae.

2. It is a good idea to use an aquarium air stone to bubble the culture overnight before the lab.

3. The three temperature baths should be 0^{o}C, 20^{o}C, and 30^{o}C. The nomograph provided does not go over 30^{o}C. These water samples should be allowed to equilibrate for some time before the lab.

4. The plastic window screen can be purchased from the hardware store where I have found it to be abundant and cheap.

5. In advance, the teacher should design class data tables to be put on the board or on an overhead transparency so that class means can be calculated in front of the class.

6. Putting the bottles under cool fluorescent tubes is preferable to incandescent spot lights due to the heat variable the spot lights add to the results.

7. It is a good idea to "walk" the students through their first use of the LaMotte test kit. It is not difficult, but it will save many false starts by those who do not read the directions carefully.

Suggested Answers to Selected Questions:

3. Assuming 100% oxygen removal, a land animal needing 1 liter of oxygen must process 4.8 liters of air at 0^{o}C. That same liter of oxygen is spread through 98 liters of freshwater and 125 liters of seawater. Since water is much more viscous than air (about 100 times more at 0^{o}C), an aquatic

animal not only has to process a vastly greater volume of water than air to obtain oxygen, but also has to work very hard just to move it.[4] Gills, with their countercurrent exchange mechanisms are actually more efficient than lungs, but the aquatic animals must work hard for their oxygen.

6. Although glass A has a greater volume of water and can potentially hold more DO at any particular temperature, the greater concern here is the fact that the fish is going to use the oxygen up. Glass B has a much larger surface area for the diffusion of oxygen into the water from the air.

[4]From "Dissolved Oxygen" by Valerie C. Chase in Carolina Tips, 5/1/88.