Why are we concerned?

- Both aquatic plants and animals depend on dissolved oxygen (D.O.) for survival.

- D.O. concentrations are influenced by many factors including water temperature, the rate of photosynthesis, the degree of light penetration (turbidity and water depth), the degree of water turbulence or wave action, and the amount of oxygen used by respiration and decay of organic matter.

**DEFINITION OF TERMS**

**Cold-blooded:** Animals whose body temperatures match that of their surroundings. Fish, invertebrates, snakes, frogs and toads are cold-blooded.

**Diffusion:** The movement of molecules, for example oxygen molecules, from an area of higher concentration (e.g. the air) to an area of lower concentration (e.g. the water).

**Endpoint:** The completion of a chemical reaction. It is often determined by the change in color of an indicator solution.

**Floc:** Short for flocculent precipitate. These fine, suspended particles look like heavy snow.

**Photosynthesis:** The process in which green plants convert carbon dioxide and water, using the sun’s energy, into simple sugars and oxygen.

**Respiration:** The cellular process in which plants and animals use oxygen and release carbon dioxide. Basically, it is the reverse of photosynthesis because carbon dioxide, water and energy are released in the process.

**Supersaturation:** An indication that more oxygen is dissolved in water than would be in a state of equilibrium. Supersaturation could indicate that some processes are affecting the water’s natural balance found in the state of equilibrium.

**Titrant:** The solution of known strength used for measuring the extent of a chemical reaction, in this case it is sodium thiosulfate.
Oxygen is a clear, colorless, odorless, and tasteless gas that dissolves in water. Small but important amounts of it are dissolved in water. It is supplied by diffusion of atmospheric (air) oxygen into the water and by production of oxygen from photosynthesis by aquatic plants. Wind, waves, and tumbling water in fast-moving streams increase the rate of diffusion.

**Oxygen: Aquatic Life Depends on it**

Both plants and animals depend on dissolved oxygen for survival. Lack of dissolved oxygen can cause aquatic animals (e.g. fish, macroinvertebrates) to quickly leave the area or face death. Under low-oxygen conditions, the aquatic animal community changes quickly. Under extreme conditions, lack of oxygen can kill aquatic plants and animals. Measuring dissolved oxygen is probably the most significant water quality test to determine the suitability of a stream for fish and many other aquatic organisms. However, these measures only provide a snapshot of the oxygen levels at that particular time. Levels can fluctuate widely throughout the day and year. Fish and other organisms have to live and breathe in that water all year long. A short time without oxygen can be fatal.

**Factors Affecting Oxygen Levels**

Oxygen is removed from the water by chemical reactions, the decay process and respiration of living organisms, including fish, bacteria, fungi and protozoans.

Water temperature and atmospheric pressure affect the capacity of water to hold dissolved oxygen. Cold water at high atmospheric pressure holds more dissolved oxygen than warm water at low atmospheric pressure. Oxygen levels also are affected by the degree of light penetration (turbidity, color and water depth) and the degree of water turbulence or wave action. Dissolved oxygen (D.O.) is reported as milligrams of oxygen per liter of water (mg/L) which can be called parts by weight per million (ppm).

Different aquatic organisms have different oxygen needs. Trout and stoneflies, for example, require high dissolved oxygen levels. Trout need water with at least 6 mg/L D.O. Warm water fish like bass and bluegills survive nicely at 5 mg/L D.O. and some organisms like carp and bloodworms can survive on less than 1 mg/L D.O. The oxygen demand of aquatic plants and cold-blooded animals also varies with water temperature. A trout uses five times more oxygen while resting at 80˚ F (26.7˚ C.) than at 40˚ F (4.4˚ C).

Aquatic plants produce oxygen by photosynthesis during daylight hours but they also use oxygen for respiration. During the night or on heavily overcast days, respiration removes oxygen while photosynthesis stops or drastically slows down. Oxygen depletion can occur because of heavy plant growth.

Complete depletion of D.O. can sometimes be detected with your nose. Anaerobic decay results in a rotten egg smell (hydrogen sulfide gas).

**Oxygen in the Balance**

Recording dissolved oxygen differs from other tests in that it requires two distinct calculations. We are interested in both the absolute amount of D.O. and how close the value is to the equilibrium value for that temperature and air pressure, or the percentage of saturation. Values between 90% and 110% of saturation are good. Supersaturated (over 100%) values may sound good but they can also indicate problems, such as excessive plant growth.

High day-time levels of D.O. are often countered with low night-time levels due to respiration and the cessation of photosynthesis. Wide daily fluctuations of D.O. stress fish and other aquatic animals.

Dissolved oxygen levels are reduced by excessive amounts of organic matter such as sewage, manure, or leaves that wash into streams. Warm water released from industrial outlets, flowages, or storm sewers can also reduce dissolved oxygen levels. Erosion from any number of sources is another factor that lowers dissolved oxygen levels. However, good management practices such as planting or maintaining vegetation that filters rainwater runoff and shades the water, cooler water temperatures and protecting the stream channel in other ways to maintain or increase turbulence all promote good dissolved oxygen levels.
Collecting the Sample

These directions, with some minor modifications, are written for the Hach water testing kit for dissolved oxygen.

Remember that photosynthesis and respiration will continue after a sample is collected, so water can gain or lose oxygen while sitting in the sample bottle. Therefore, you should begin D.O. testing immediately upon reaching the shore after you have collected the sample.

1. Use bottle with the stopper included in the Hach kit.
2. Immerse thermometer in water for about two minutes, record temperature.
3. Collect your sample in roughly one-foot deep, normally moving water.
4. Facing upstream, slowly lower the bottle so opening of the bottle faces away from you and water current is entering bottle.
5. Allow the bottle to fill with water gradually, turning it to allow air bubbles to float out.
6. Cap bottle while still submerged and leave extra water in the neck of the bottle.
7. When lifting out of water, look for bubbles. If you see any, take another sample using the same procedure.

Testing for Dissolved Oxygen

Note: If you see any air bubbles trapped in the sample bottle during steps 2-4, discard the sample and start over.

1. Put on protective gloves and safety goggles. If your skin comes in contact with any powder or titrant, rinse the area liberally with water.
2. Remove the stopper and add the contents of D.O. powder pillow #1 (manganous sulfate powder) and D.O. powder pillow #2 (alkaline iodide azide powder) to the sample.
3. Insert the stopper, being careful not to trap an air bubble and shake vigorously, holding on to the top. If oxygen is present, a brownish-orange floc will form.
4. Allow the sample to stand until the floc settles halfway. Shake the bottle a second time and allow the floc to settle halfway again.
5. Remove the stopper and slowly add the contents of D.O. powder pillow #3 (sulfamic acid), taking care not to displace any floc.
6. Stopper and shake vigorously to dissolve the floc. Shake and wait until all the floc is dissolved. The yellow color is from iodine. This is called the prepared sample. Prepared samples can be stored in the dark for a short time if it is more convenient or comfortable to return to your home/school to complete the analysis.
7. Transfer two plastic measuring tubes full of prepared sample to the square glass mixing bottle (your Hach kit instructions probably say one measuring tube full). Two measuring tubes allows you to determine D.O. to the nearest 0.5 mg/L instead of 1 mg/L.
8. a.) Holding the dropper vertically, add one drop at a time of sodium thiosulfate standard solution titrant (some kits use PAO titrant instead of sodium thiosulfate) to the square mixing bottle, and count each drop. b.) Swirl the solution after each drop. c.) Continue adding sodium thiosulfate drops until the —continued on next page
sample is a very light yellow. d.) Add 3 to 4 drops of starch solution. The prepared sample will turn blue from the added starch solution. If you do not have starch solution, proceed with the next step but be aware that your sample will turn from yellow to colorless instead of blue to colorless. e.) Continue adding drops, mixing and counting until the prepared sample turns from blue (or yellow) to colorless (the end point). Often this is just one or two more drops, so be careful.

9. The dissolved oxygen content of the water in mg/L is the total number of drops of titrant used to get to the endpoint divided by two if two measuring tubes of prepared sample were used. If only one measuring tube of prepared sample was used, the dissolved oxygen content is equal to the number of drops of titrant used. Example: If you used two tubes of sample, you need to divide by two (13 drops divided by two tubes = 6.5 mg/L). If you only used one tube of sample, it’s the actual number of drops of titrant used (6 drops with one tube = 6 mg/L).

10. Report dissolved oxygen (mg/L) and temperature on the record form.

11. Use instructions and chart below to convert D.O. to % saturation. Report % saturation on the record form.

### Temperature Conversion Chart

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**How to Find Percentage of Saturation:** Using a straight edge, find your water temperature (convert from Fahrenheit if necessary using above chart). Align with the Oxygen mg/Liter scale. The percentage of saturation is on the same line. For example, 5°C with 10 mg/L of oxygen aligns with 75% saturation, which is your answer.

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