Dissolved Oxygen

Primary Importance:
Aquatic organisms require oxygen in the free elemental state as a dissolved gas. The amount of dissolved oxygen in the water is fundamental to the survival of most aquatic plants and animals.

Problem:
Lack of significant levels of dissolved oxygen required by most aquatic organisms for respiration can cause impairment or death.
Some organisms have adapted to low oxygen in water or are able to ingest air directly.

Causes

- Rapid decomposition of organic materials, including dead algae, shoreline vegetation and manure or wastewater sources, decreases oxygen concentrations.
- High ammonia concentrations in the stream use up oxygen in the process of oxidizing NH^{4+} to NO^{3-} (nitrification).
- At higher temperatures, less oxygen can dissolve in water.
- Lack of turbulence or mixing to expose water to atmospheric oxygen results in low dissolved oxygen concentrations in the stream.

Instructions:
These instructions are for use with the HACH Company Dissolved Oxygen test kit, Catalog No. 1469-00, Model OX-2P, for 60 mL sample.

CHECKLIST

- ☐ DO glass collection bottle and glass stopper
- ☐ 23 mL square mixing bottle
- ☐ Plastic measuring tube
- ☐ DO Reagent 1 powder pillows (manganous sulfate)
- ☐ DO Reagent 2 powder pillows (lithium hydroxide)
- ☐ DO Reagent 3 powder pillows (sulfamic acid)
- ☐ Sodium Thiosulfate Solution dropper bottle
- ☐ Waste container
- ☐ Material Safety Data Sheets
- ☐ Testing Instructions
- ☐ Data Sheets

Steps 1-4 of this test MUST be done at the site when the sample is collected. If any oxygen bubbles are seen during these steps, dispose of your sample, rinse and collect a new sample.

1. After rinsing thoroughly with distilled water and sample water, lower the DO bottle (or other clean collection bottle) in an upside-down position to a point 3-4 inches below the water's surface. Turn the bottle upright to an angle tilting upstream to reduce the risk of air bubbles. Allow water to flow into the bottle for approximately 2 minutes until the bottle is full and no air bubbles are present. While the bottle is underwater, place stopper

in the top. Remove the bottle from the stream with the stopper in place. DO NOT pour off the excess water around the rim of the stopper. (Note: If pouring your sample from a collection bottle into the DO bottle, be careful not to agitate or splash the water into the bottle.)

2. Add Dissolved Oxygen 1 Reagent and Dissolved Oxygen 2 Reagent Powder Pillows to the DO bottle (the order does not matter). Stopper the bottle, being very careful not to introduce air bubbles. (Note: Allow the excess water to spill over into a waster container.) If you get an air bubble, start over with step one. With your thumb firmly holding the stopper in place, grip the bottle and shake vigorously until the contents are evenly mixed. A flocculent (floc) precipitate will form. If oxygen is present in the sample, the precipitate will appear brownish-orange in color. A small amount of powdered reagent may remain at the bottom of the bottle. This will not affect the test results.

3. Allow the sample to stand until the floc has settled below the DO bottle's white line. The upper half of the sample will be clear. Shake the bottle again to remix and allow it to resettle in the same manner as above. (Note: the floc will not settle in samples with high concentrations of chloride. Allow a maximum of five minutes for the floc to settle. If no additional progress is made, proceed with the next step.)

4. Add the contents of Dissolved Oxygen 3 Reagent Powder Pillow. Carefully replace the stopper and shake the bottle to mix. The floc will dissolve, creating a yellowish-amber color if DO is present. (Note: Small, rust colored flakes may remain, but will not affect the test results.) At this point, the oxygen is "fixed" and any oxygen bubbles formed after this step will not affect the results of the test.

5. Fill the sturdy, 5 mL measuring tube (1 cm width x 8.5 cm length—if you can stick your little finger inside the tube, it's the wrong one!) to its top with the prepared sample into the square mixing bottle. (Note: Do not discard the rest of the fluid in the DO bottle until you have successfully completed the rest of this test.)

6. Using the dropper located within the brown bottle marked Sodium Thiosulfate Standard Solution, add this solution drop by drop to the prepared sample in the mixing bottle. Count each drop as it is added and gently swirl to mix the solution until it becomes colorless. Once the prepared sample is clear, add one more drop to ensure a complete color change, if there is no change in color, do not count the last drop. (Note: Hold the dropper vertically above the mixing bottle's mouth when adding drops to ensure the proper volume of titrant. Do not place the dropper inside the mouth of the square bottle as you may contaminate the dropper.) Also, rinse thoroughly any surface, including your hands, that has contacted the above chemical as it may eat holes in your clothing or irritate your skin.)

7. Each drop added to bring about the color change in Step 6 equals the presence of 1.0 mg/L of dissolved oxygen. (Note: If the result of Step 6 is 3 mg/L or less, follow the Low-range instructions provided below.)

8. Use the graph on the next page to calculate percent saturation. By running a straight edge from the appropriate water temperature reading to oxygen mg/L, you will be able to determine percent saturation along the angled scale. Look at the water temperature change directions for the Celsius/Fahrenheit conversion.

9. Record DO to the nearest 1.0 mg/L and record the percent saturation.
   EXAMPLE:
   Water temperature at site=16°C
   Dissolved oxygen=**8 mg/L** = **80% saturation** (look on chart)

10. You may also use the following formula to determine percent saturation:
    DO mg/L (your sample) / DO mg/L needed for your sample to be 100% saturated
    EXAMPLE:
    At your sampling location, you recorded a water temperature of 19°C and a DO of 6 mg/L. Based upon the chart in your book, the DO reading (mg/L) for the water to be 100% saturated at 19°C = 9.3 mg/L. Therefore, 6 / 9.3 * 100 = 64.5%.

Dissolved Oxygen Low-range (0.2-4 mg/L)

1. Use the prepared sample left from Step 5 of the High-range test. Pour off the contents of the DO bottle until the level reaches the 30 mL mark on the bottle.

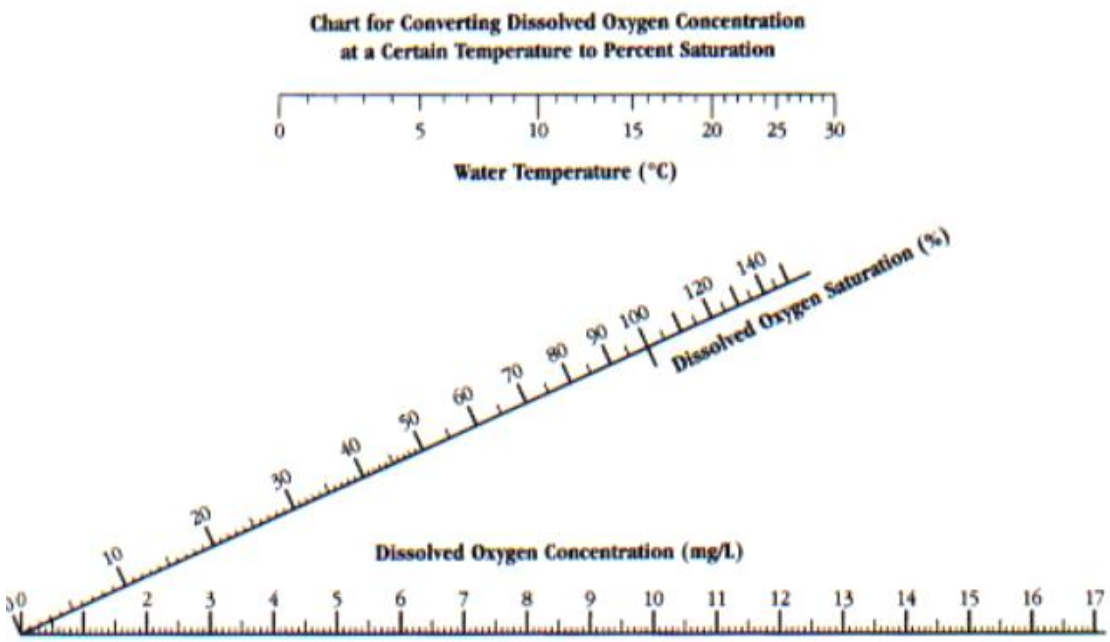
2. Add Sodium Thiosulfate Standard Solution one drop at a time to the DO bottle. Count each drop as it is added and gently swirl to mix the solution until it becomes colorless. Once the prepared sample is clear, add one more drop to ensure a complete color change. If there is no change in color, do not count the drop.

3. Multiply the number of drops used by 0.2 to obtain the mg/L Dissolved Oxygen.
   EXAMPLE: 15 drops x 0.2=3 mg/L DO

4. Record DO in mg/L and percent saturation.

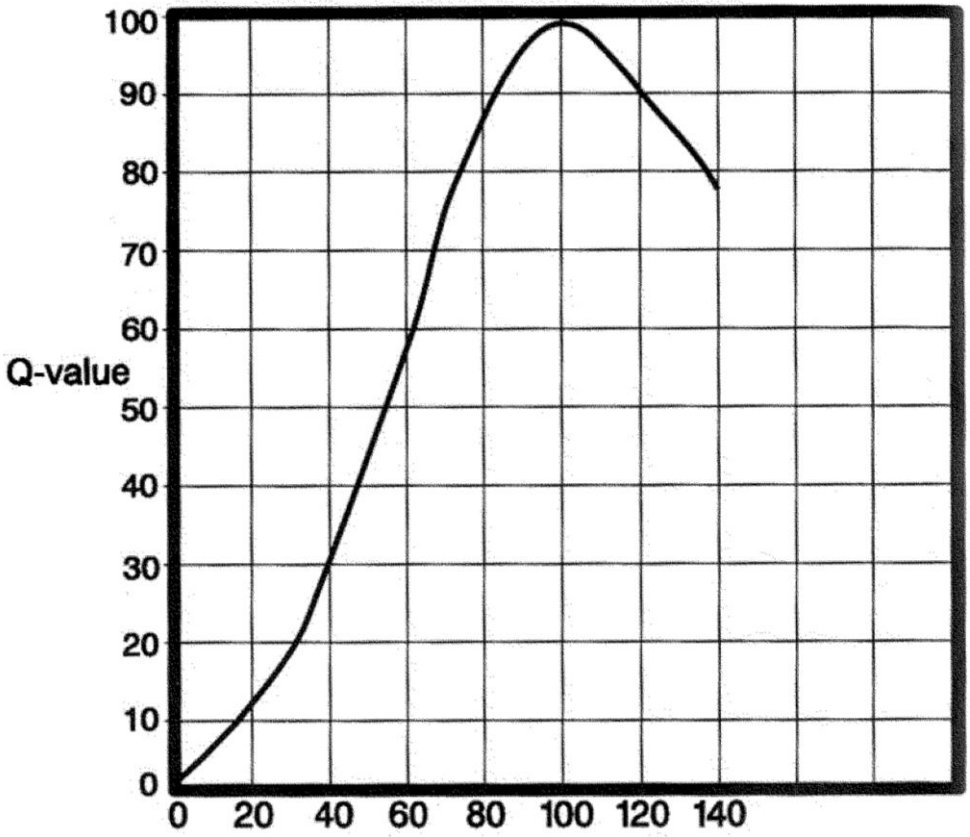
TYPICAL RANGE FOR DO = 5.4 to 14.2mg/L

First find percent saturation…



Then find the Q-value…

Dissolved Oxygen Q-values



DO % Saturation)	Q-Value
0	0
10	8
20	13
30	20
40	30
50	43
60	56
70	77
80	88
85	92
90	95
95	97.5
100	99
105	98
110	95
120	90
130	85
140	78
>140	50

Note: if DO % saturation > 140.0, Q = 50.0