# **Dissolved Oxygen**

#### INTRODUCTION

Oxygen gas dissolved in water is vital to the existence of most aquatic organisms. Oxygen is a key component in cellular respiration for both aquatic and terrestrial life. The concentration of

dissolved oxygen, DO, in an aquatic environment is an important indicator of the environment's water quality.

Some organisms, such as salmon, mayflies, and trout, require high concentrations of dissolved oxygen. Other organisms, such as catfish, mosquito larvae, and carp, can survive in environments with lower concentrations of dissolved oxygen. The diversity of organisms is greatest at higher DO concentrations. Table 1 lists the minimum dissolved oxygen concentrations necessary to sustain selected animals.

Oxygen gas is dissolved in water by a variety of processes—diffusion between the atmosphere and water at its surface, aeration as water flows over rocks and other debris, churning of water by waves and wind, and photosynthesis of aquatic plants. There are many factors that affect the concentration of dissolved oxygen in an aquatic environment. These factors include: temperature, stream flow, air pressure, aquatic plants, decaying organic matter, and human activities.

Table 1: Minimum DO Requirements						
Organism	Minimum dissolved oxygen (mg/L)					
Trout	6.5					
Smallmouth bass	6.5					
Caddisfly larvae	4.0					
Mayfly larvae	4.0					
Catfish	2.5					
Carp	2.0					
Mosquito larvae	1.0					

#### Sources of DO

Diffusion from atmosphere

Aeration as water moves over rocks and debris

Aeration from wind and waves

Photosynthesis of aquatic plants

As a result of plant activity, DO levels may fluctuate during the day, rising throughout the morning and reaching a peak in the afternoon. At night photosynthesis ceases, but plants and animals continue to respire, causing a decrease in DO levels. Because large daily fluctuations are possible, DO tests should be performed at the

#### **Factors that affect DO levels**

**Temperature** 

Aquatic plant populations

Decaying organic material in water

Stream flow

Altitude/atmospheric pressure

Human activities

same time each day. Large fluctuations in dissolved oxygen levels over a short period of time may be the result of an algal bloom. While the algae population is growing at a fast rate, dissolved oxygen levels increase. Soon the algae begin to die and are decomposed by aerobic bacteria, which use up the oxygen. As a greater number of algae die, the oxygen requirement of the aerobic decomposers increases, resulting in a sharp drop in dissolved oxygen levels. Following an algal bloom, oxygen levels can be so low that fish and other aquatic organisms suffocate and die.



Temperature is important to the ability of oxygen to dissolve, because oxygen, like all gases, has different solubilities at different temperatures. Cooler waters have a greater capacity for dissolved oxygen than warmer waters. Human activities, such as the removal of foliage along a stream or the release of warm water used in industrial processes, can cause an increase in water temperature along a given stretch of the stream. This results in a lower dissolved oxygen capacity for the stream.

### **Expected Levels**

The unit mg/L<sup>2</sup> is the quantity of oxygen gas dissolved in one liter of water. When relating DO measurements to minimum levels required by aquatic organisms, mg/L is used. The procedure described in this chapter covers the use of a Dissolved Oxygen Probe to measure the concentration of DO in mg/L. Dissolved oxygen concentrations can range from 0 to 15 mg/L. Cold mountain streams will likely have DO readings from 7 to 15 mg/L, depending on the water temperature and air pressure. In their lower reaches, rivers and streams can have DO readings between 2 and 11 mg/L.

Table 2					
DO Level	Percent Saturation of DO				
Supersaturation <sup>1</sup>	≥ 101%				
Excellent	90 – 100%				
Adequate	80 – 89%				
Acceptable	60 – 79%				
Poor	< 60%				

When discussing water quality of a stream or river, it can be helpful to use a different unit than mg/L. The term percent saturation is often used for water quality comparisons. Percent saturation is the dissolved oxygen reading in mg/L divided by the 100% dissolved oxygen value for water (at the same temperature and air pressure). The manner in which percent saturation relates to water quality is displayed in Table 2. In some cases, water can exceed 100% saturation and become supersaturated for short periods of time.

# **Summary of Methods**

Dissolved oxygen can be measured directly at the site or from water samples transported from the site. Measurements can be made at the site by either placing the Dissolved Oxygen Probe directly into the stream away from the shore or by collecting a water sample with a container or cup and then taking measurements with the Dissolved Oxygen Probe back on the shore. Water samples collected from the site in capped bottles and transported back to the lab must be stored in an ice chest or refrigerator until measurements are to be made. Transporting samples is not recommended, because it reduces the accuracy of test results.

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<sup>&</sup>lt;sup>1</sup> Supersaturation can be harmful to aquatic organisms. It can result in a disease known as Gas Bubble Disease.

<sup>&</sup>lt;sup>2</sup> The unit of mg/L is numerically equal to parts per million, or ppm.



#### **DISSOLVED OXYGEN**

#### **Materials Checklist**

LabPro or CBL 2 interface	100% calibration bottle
TI Graphing Calculator	small plastic or paper cup (optional)
DataMate program	tissues or paper towels
Vernier Dissolved Oxygen Probe	distilled water
250-mL beaker	Sodium Sulfite Calibration Solution
pipet	DO Electrode Filling Solution

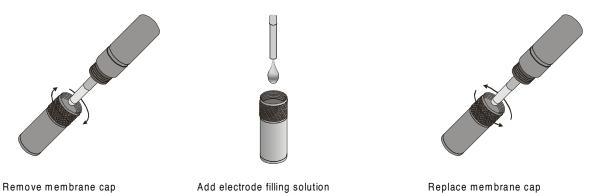
### **Collection and Storage of Samples**

- 1. Before you begin sampling, fill out the site information on the Data & Calculations sheet. Space for observations regarding the site is provided at the bottom of the Data & Calculations sheet. Special things to note about the site are the weather, descriptions of the stream reach (flow, depth, shape), and a description of the riparian zone (density of foliage and width of riparian zone).
- 2. It is important to sample as far away from the shore as is safe and under the surface of the water. Samplers consisting of a rod and container can be constructed for collection of samples from areas of the stream otherwise unreachable. Refer to page Intro-4 of the Introduction of this book for more details. In slow-moving water, it is necessary to take samples below the water's surface at various depths.
- 3. When collecting a sample with a cup or container, prevent mixing of the water sample and air by collecting your sample from below the water surface.
- 4. If you are going to take readings after returning to the laboratory, make sure that there are no air bubbles in the water-sample container and that the container is tightly stoppered. The sample should be stored in an ice chest or refrigerator until measurements are to be made. Storing water samples for later testing decreases sample accuracy and is only recommended in cases where measuring at the site is not possible.
- 5. When taking readings in cold (0–10°C) or warm (25–35°C) water, allow more time for the dissolved oxygen readings to stabilize. Automatic temperature compensation in the Dissolved Oxygen Probe is not instantaneous and readings may take up to 2 minutes to stabilize depending on the temperature.

# **Testing Procedure**

- 1. Prepare the Dissolved Oxygen Probe for use.
  - a. Remove the blue protective cap if it is still on the tip of the probe.
  - b. Unscrew the membrane cap from the tip of the probe.
  - c. Using a pipet, fill the membrane cap with 1 mL of DO Electrode Filling Solution.
  - d. Carefully thread the membrane cap back onto the electrode.
  - e. Place the probe into a container of water.





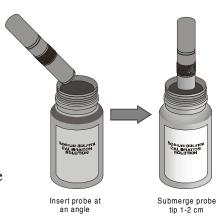
- 2. Plug the Dissolved Oxygen Probe into Channel 1 of the LabPro or CBL 2 interface. Use the link cable to connect the TI Graphing Calculator to the interface. Firmly press in the cable ends.
- 3. Turn on the calculator and start the DATAMATE program. Press CLEAR to reset the program.
- 4. Set up the calculator and interface for the Dissolved Oxygen Probe.
  - a. If CH 1 displays DO (MG/L), proceed to Step 5. If it does not, continue with this step to set up your sensor manually.
  - b. Select SETUP from the main screen.
  - c. Press ENTER to select CH 1.
  - d. Select D. OXYGEN (MG/L) from the SELECT SENSOR menu.
  - e. Select OK to return to the main screen.
- 5. Warm up the Dissolved Oxygen Probe for 10 minutes..
  - a. With the probe still in the water, wait 10 minutes while the probe warms up. The probe must stay connected to the interface at all times to keep it warmed up. If disconnected for a period longer than a few minutes, it will be necessary to warm it up again.
  - b. Select SETUP from the main screen.
- 6. Set up the calibration for the Dissolved Oxygen Probe.

If your instructor directs you to manually enter the calibration values, select CALIBRATE, then MANUAL ENTRY. Enter the slope and intercept values, select OK, then proceed to Step 7.

If your instructor directs you to perform a new calibration, follow this procedure.

#### Zero-Oxygen Calibration Point

- a. Select CALIBRATE, then CALIBRATE NOW.
- b. Remove the probe from the water and place the tip of the probe into the Sodium Sulfite Calibration Solution. **Important:** No air bubbles can be trapped below the tip of the probe or the probe will sense an inaccurate dissolved oxygen level. If the voltage does not rapidly decrease, tap the side of the bottle with the probe to dislodge the bubble. The readings should be in the 0.2- to 0.5-V range.

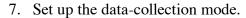




- c. When the voltage stabilizes (~1 minute), press [ENTER].
- d. Enter "0" as the known value in mg/L.

#### Saturated DO Calibration Point

- e. Rinse the probe with distilled water and gently blot dry.
- f. Unscrew the lid of the calibration bottle provided with the probe. Slide the lid and the grommet about 1/2 inch onto the probe body.
- g. Add water to the bottle to a depth of about 1/4 inch and screw the bottle into the cap, as shown. **Important:** Do not touch the membrane or get it wet during this step.
- h. Keep the probe in this position for about a minute. The readings should be above 2.0 V. When the voltage stabilizes, press ENTER.
- i. Enter the correct saturated dissolved-oxygen value (in mg/L) from Table 3 (for example, "8.66") using the current barometric pressure and air temperature values. If you do not have the current air pressure, use Table 4 to estimate the air pressure at your altitude.
- j. Select OK to return to the setup screen.



- a. To select MODE, press once and press ENTER).
- b. Select SINGLE POINT from the SELECT MODE menu.
- c. Select OK to return to the main screen.
- 8. Collect dissolved oxygen concentration data in SINGLE POINT mode.
  - a. Rinse the tip of the probe with a sample of water.
  - b. Place the tip of the probe into the stream at Site 1, or into a cup with sample water from the stream. Submerge the probe tip to a depth of 4-6 cm. Gently stir the probe in the water sample. **Note:** It is important to keep stirring until you have collected your DO value.
  - c. When the readings stabilize (stable to the nearest 0.1 mg/L), select START to begin sampling. Continue stirring. After 10 seconds, the dissolved oxygen concentration will appear on the calculator screen.
  - d. Record this value and the site number on the Data & Calculations sheet (round to the nearest 0.1 mg/L).
  - e. Press ENTER to return to the main screen.
  - f. Repeat Steps 8 a-e to test a second sample or if collecting data for a second site.





Table 3: 100% Dissolved Oxygen Capacity (mg/L)												
	770 mm	760 mm	750 mm	740 mm	730 mm	720 mm	710 mm	700 mm	690 mm	680 mm	670 mm	660 mm
0°C	14.76	14.57	14.38	14.19	13.99	13.80	13.61	13.42	13.23	13.04	12.84	12.65
1°C	14.38	14.19	14.00	13.82	13.63	13.44	13.26	13.07	12.88	12.70	12.51	12.32
2°C	14.01	13.82	13.64	13.46	13.28	13.10	12.92	12.73	12.55	12.37	12.19	12.01
3°C	13.65	13.47	13.29	13.12	12.94	12.76	12.59	12.41	12.23	12.05	11.88	11.70
4°C	13.31	13.13	12.96	12.79	12.61	12.44	12.27	12.10	11.92	11.75	11.58	11.40
5°C	12.97	12.81	12.64	12.47	12.30	12.13	11.96	11.80	11.63	11.46	11.29	11.12
6°C	12.66	12.49	12.33	12.16	12.00	11.83	11.67	11.51	11.34	11.18	11.01	10.85
7°C	12.35	12.19	12.03	11.87	11.71	11.55	11.39	11.23	11.07	10.91	10.75	10.59
8°C	12.05	11.90	11.74	11.58	11.43	11.27	11.11	10.96	10.80	10.65	10.49	10.33
9°C	11.77	11.62	11.46	11.31	11.16	11.01	10.85	10.70	10.55	10.39	10.24	10.09
10°C	11.50	11.35	11.20	11.05	10.90	10.75	10.60	10.45	10.30	10.15	10.00	9.86
11°C	11.24	11.09	10.94	10.80	10.65	10.51	10.36	10.21	10.07	9.92	9.78	9.63
12°C	10.98	10.84	10.70	10.56	10.41	10.27	10.13	9.99	9.84	9.70	9.56	9.41
13°C	10.74	10.60	10.46	10.32	10.18	10.04	9.90	9.77	9.63	9.49	9.35	9.21
14°C	10.51	10.37	10.24	10.10	9.96	9.83	9.69	9.55	9.42	9.28	9.14	9.01
15°C	10.29	10.15	10.02	9.88	9.75	9.62	9.48	9.35	9.22	9.08	8.95	8.82
16°C	10.07	9.94	9.81	9.68	9.55	9.42	9.29	9.15	9.02	8.89	8.76	8.63
17°C	9.86	9.74	9.61	9.48	9.35	9.22	9.10	8.97	8.84	8.71	8.58	8.45
18°C	9.67	9.54	9.41	9.29	9.16	9.04	8.91	8.79	8.66	8.54	8.41	8.28
19°C	9.47	9.35	9.23	9.11	8.98	8.86	8.74	8.61	8.49	8.37	8.24	8.12
20°C	9.29	9.17	9.05	8.93	8.81	8.69	8.57	8.45	8.33	8.20	8.08	7.96
21°C	9.11	9.00	8.88	8.76	8.64	8.52	8.40	8.28	8.17	8.05	7.93	7.81
22°C	8.94	8.83	8.71	8.59	8.48	8.36	8.25	8.13	8.01	7.90	7.78	7.67
23°C	8.78	8.66	8.55	8.44	8.32	8.21	8.09	7.98	7.87	7.75	7.64	7.52
24°C	8.62	8.51	8.40	8.28	8.17	8.06	7.95	7.84	7.72	7.61	7.50	7.39
25°C	8.47	8.36	8.25	8.14	8.03	7.92	7.81	7.70	7.59	7.48	7.37	7.26
26°C	8.32	8.21	8.10	7.99	7.89	7.78	7.67	7.56	7.45	7.35	7.24	7.13
27°C	8.17	8.07	7.96	7.86	7.75	7.64	7.54	7.43	7.33	7.22	7.11	7.01
28°C	8.04	7.93	7.83	7.72	7.62	7.51	7.41	7.30	7.20	7.10	6.99	6.89
29°C	7.90	7.80	7.69	7.59	7.49	7.39	7.28	7.18	7.08	6.98	6.87	6.77
30°C	7.77	7.67	7.57	7.47	7.36	7.26	7.16	7.06	6.96	6.86	6.76	6.66
31°C	7.64	7.54	7.44	7.34	7.24	7.14	7.04	6.94	6.85	6.75	6.65	6.55

Table 4: Approximate Barometric Pressure at Different Elevations								
Elevation (feet)	Pressure (mm Hg)	Elevation (feet)	Pressure (mm Hg)	Elevation (feet)	Pressure (mm Hg)			
0	760	2000	708	4000	659			
250	753	2250	702	4250	653			
500	746	2500	695	4500	647			
750	739	2750	689	4750	641			
1000	733	3000	683	5000	635			
1250	727	3250	677	5250	629			
1500	720	3500	671	5500	624			
1750	714	3750	665	5750	618			



# **DATA & CALCULATIONS**

Disso	lved	Oxygen

t name:  t name:  t name:  t name:  100% dissolved oxygen (mg/L)  lg 9.5 mg/L	E
t name:  t name:  t name:  100% dissolved oxygen (mg/L)	E  d Percent saturation (%)
t name:  D  ric 100% dissolved oxygen (mg/L)	E  d Percent saturation (%)
ric 100% dissolved oxygen (mg/L)	Dercent saturation (%)
ric 100% dissolved oxygen (mg/L)	d Percent saturation (%)
oxygen (mg/L)	saturation (%)
lg 9.5 mg/L	86 %
Average %	
Probe or thermomet or by using known a alue using measured	altitude (see Table
	or by using known

Test Completed: \_\_\_\_\_ Date: \_\_\_\_



#### ADDITIONAL INFORMATION

### **Tips for Instructors**

- 1. Before calibrating or taking measurements with the Dissolved Oxygen Probe, it is necessary to warm up, or polarize, the probe for 10 minutes. Think of it like a clothes iron that has to warm up before you can use it to iron your clothing. You must also keep the probe plugged in until you are finished taking all of your measurements. When the probe is unplugged from an active interface, it begins to cool down just like the clothes iron. All channels of LabPro and CBL 2 will provide constant power to the probe as long as DataMate knows that there is a Dissolved Oxygen Probe attached and you are at the main screen of the DataMate program. Whenever possible, use the AC Adapter so that the interface batteries do not run down during the warm up
- 2. The probe tip should be in water during the warm-up period. You could place the probe into a cup or beaker with water or you could use the DO calibration bottle. Simply fill the DO calibration bottle with water, fit the probe down into the lid, and tighten the lid onto the bottle. The probe tip should be submerged in the water until you calibrate or take samples.

period. If this is not possible, use a fresh set of batteries.

3. When calibrating the Dissolved Oxygen Probe, it is important to be patient and permit the readings to stabilize.

At the zero oxygen point, the voltage should be somewhere between 0.2 V and 0.5 V. If it is not, make sure there is not an air bubble at the tip of your electrode. If you suspect your sodium sulfite solution may have gone bad, mix up some fresh or obtain a new bottle from Flinn Scientific (order code SO426).

At the saturated oxygen point, the voltage should be above 2.0 V. If it is not, make sure the electrode is not actually touching the water in the bottle. Thoroughly rinse the electrode with distilled water again and gently blot it dry with a paper towel being careful not to touch the membrane with your finger.

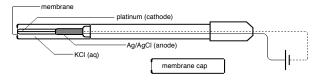
- 4. As the Dissolved Oxygen Probe measures dissolved oxygen, it removes O<sub>2</sub> from the water sample at the junction of the probe membrane. If you leave the probe in one spot in the water sample, you will see your dissolved oxygen readings drop. To prevent this, it is important that students stir the probe gently and slowly through the sample as they take readings.
- 5. The gas-permeable plastic membrane on the Dissolved Oxygen Probe can become clogged by dirt and oil over time. Advise students to avoid touching the membrane at any time. If the water being sampled is murky or dirty, rinse the probe tip with distilled water after each use.
- 6. The electrode of the Dissolved Oxygen Probe is water tight and will not be damaged by water. The junction at the top of the electrode where the cable enters is not water tight and should not be submerged in water for any period of time. To take dissolved oxygen readings at various depths, use a Water Depth Sampler (order code WDS, \$57). This device can be lowered to any desired depth and triggered to collect a representative water sample.
- 7. The SINGLE POINT data-collection mode was designed to make measurements easier and more accurate. When SINGLE POINT mode is used, the interface takes readings for 10 seconds. These readings are averaged and this average value is displayed on the calculator. This method has several advantages over other data-collection modes: (1) It eliminates the need for students to choose one value over another if that value is fluctuating; (2) If the



readings are fluctuating a little, an average of the values is desirable; (3) It requires the students to hold the sensor in the water longer that they might tend to otherwise.

## **How the Dissolved Oxygen Probe Works**

The Vernier Dissolved Oxygen Probe is a Clark-type polarographic electrode that senses the oxygen concentration in water and aqueous solutions. A platinum cathode and a silver/silver chloride reference anode in KCl electrolyte are separated from the sample by a gas-permeable plastic membrane.



A fixed voltage is applied to the platinum electrode. As oxygen diffuses through the membrane to the cathode, it is reduced:

$$\frac{1}{2}$$
 O<sub>2</sub> + H<sub>2</sub>O + 2e<sup>-</sup>  $\longrightarrow$  2 OH<sup>-</sup>

The oxidation taking place at the reference electrode (anode) is:

$$Ag + Cl^- \longrightarrow AgCl + e^-$$

Accordingly, a current will flow that is proportional to the rate of diffusion of oxygen, and in turn to the concentration of dissolved oxygen in the sample. This current is converted to a proportional voltage, which is amplified and read by any of the Vernier lab interfaces.

## Storage of the Dissolved Oxygen Probe

Follow these steps when storing the electrode:

Long-term storage (more than 24 hours): Remove the membrane cap and rinse the inside and outside of the cap with distilled water. Shake the membrane cap dry. Also rinse and dry the exposed anode and cathode inner elements (blot dry with a lab wipe). Reinstall the membrane cap loosely onto the electrode body for storage. Do not screw it on tightly.

**Short-term storage (less than 24 hours):** Store the Dissolved Oxygen Probe with the membrane end submerged in about 1 inch of distilled water.

## **Automatic Temperature Compensation**

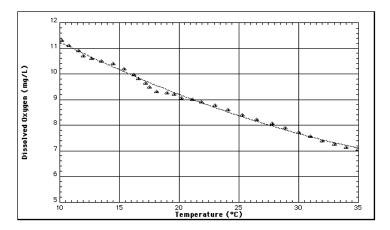
Your Vernier Dissolved Oxygen Probe is automatically temperature compensated, using a thermistor built into the probe. The temperature output of this probe is used to automatically compensate for changes in permeability of the membrane with changing temperature. If the probe was not temperature compensated, you would notice a change in the dissolved oxygen reading as temperature changed, even if the actual concentration of dissolved oxygen in the solution did not change. Here are two examples of how automatic temperature compensation works:

If you calibrate the Dissolved Oxygen Probe in the lab at 25°C and 760 mm Hg barometric pressure (assume salinity is negligible), the value you entered for the saturated oxygen calibration point would be 8.36 mg/L (see Table 3). If you were to take a reading in distilled water that is saturated with oxygen by rapid stirring, you would get a reading of 8.36 mg/L. *If* the water sample is then cooled to 10°C with no additional stirring, the water would no longer



be saturated (cold water can hold more dissolved oxygen than warm water). Therefore, the reading of the temperature-compensated Dissolved Oxygen Probe would still be 8.36 mg/L.

If, however, the solution was cooled to 10°C *and* continually stirred so it remained saturated by dissolving additional oxygen, the temperature-compensated probe would give a reading of 11.35 mg/L—the value shown in Table 3. **Note:** Temperature compensation *does not mean* that the reading for a saturated solution will be the same at two different temperatures—the two solutions have different concentrations of dissolved oxygen, and the probe reading should reflect this difference.



Saturated Dissolved Oxygen vs. Temperature Data

### Sampling in Ocean Salt Water or Tidal Estuaries

(at salinity levels greater than 1000 mg/L)

Dissolved Oxygen concentration for air saturated water at various salinity values,  $DO_{(salt)}$ , can be calculated using the formula:

$$DO_{(salt)} = DO - (k \cdot S)$$

- $-DO_{(salt)}$  is the concentration of dissolved oxygen (in mg/L) in salt-water solutions.
- − DO is the dissolved oxygen concentration for air-saturated distilled water as determined from Table 3.
- − S is the salinity value (in ppt). Salinity values can be determined using the Vernier Chloride Ion-Selective Electrode or Conductivity Probe as described in Test 15.
- -k is a constant. The value of k varies according to the sample temperature, and can be determined from Table 5.



Table 5: Salinity Correction Constant Values								
Temp. (°C)	Constant, k	Temp. (°C)	Constant, k	Temp. (°C)	Constant, k	Temp. (°C)	Constant, k	
1	0.08796	8	0.06916	15	0.05602	22	0.04754	
2	0.08485	9	0.06697	16	0.05456	23	0.04662	
3	0.08184	10	0.06478	17	0.05328	24	0.04580	
4	0.07911	11	0.06286	18	0.05201	25	0.04498	
5	0.07646	12	0.06104	19	0.05073	26	0.04425	
6	0.07391	13	0.05931	20	0.04964	27	0.04361	
7	0.07135	14	0.05757	21	0.04854	28	0.04296	

**Example:** Determine the saturated DO calibration value at a temperature of 23°C and a pressure of 750 mm Hg, when the Dissolved Oxygen Probe is used in seawater with a salinity value of 35.0 ppt.

First, find the dissolved oxygen value in Table 3 (DO = 8.55 mg/L). Then find k in Table 5 at  $23^{\circ}\text{C}$  (k = 0.04662). Substitute these values, as well as the salinity value, into the previous equation:

$$DO_{(salt)} = DO - (k \cdot S) = 8.55 - (0.04662 \times 35.0) = 8.55 - 1.63 = 6.92 \text{ mg/L}$$

Use the value 8.46 mg/L when performing the saturated DO calibration point (water-saturated air), as described in Step 6. The Dissolved Oxygen Probe will now be calibrated to give correct DO readings in salt-water samples with a salinity of 2.0 ppt.

**Important:** For most dissolved oxygen testing, it is *not* necessary to compensate for salinity; for example, if the salinity value is 0.5 ppt, using 25°C and 760 mm Hg, the calculation for DO(s) would be:

$$DO_{(salt)} = DO - (k \cdot S) = 8.36 - (0.04498 \times 0.5) = 8.36 - 0.023 = 8.34 \text{ mg/L}$$

At salinity levels less than 1.0 ppt, neglecting this correction results in an error of less than 0.2%.