

ISE-730 Oxygen Electrode and DO2-100 Current-to-Voltage Adapter

Technical Note



ISE-730 and DO2-100

Overview

In 1954, Dr. Leland Clark invented the first membrane-covered electrode designed to measure the concentration of oxygen in blood, or solution. This electrode was innovative because it was the first electrode to have both the anode and the cathode under the same nonconductive polyethylene membrane. The limited permeability of the membrane reduced the amount of oxygen depleted from the sample which permitted accurate quantitative measurements of the oxygen concentration to be made. Dr. Clark's electrode was the prototype for many of the biosensors used today, including the sensors used in blood gas analyzers and probes, like the iWorx ISE-730, used in teaching and research laboratories.

How They Work

The ISE-730 Oxygen Electrode has a Teflon™ membrane that permits a limited amount of oxygen to diffuse from the solution being measured to the electrolyte solution that covers the platinum cathode and silver anode of the electrode. The ISE-730 is supplied with a polarizing voltage of -0.80V from the DO2-100 Current-to-Voltage adapter. The polarizing voltage creates a flow of electrons, or current, between the platinum and silver elements of the electrode. The amount of current that flows between the anode and the cathode is proportional to the concentration of oxygen in the electrolyte that bathes these two electrodes, which is proportional to the concentration of oxygen in solution.

In addition to providing a polarizing voltage for the oxygen probe, the DO2-100 converts the current output of the oxygen probe to a voltage output that can be recorded. The DO2-100 also amplifies this output voltage to a level that is easily recorded by a data acquisition unit. The output of the DO2-100 is 10 millivolts for every nanoampere of current that is flowing. If a two-point calibration is performed using room temperature deionized water, saturated with oxygen, and then



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deoxygenated water at the same temperature, the voltage output of the DO2-100 adapter can be related to the oxygen concentration in the chamber.

How to Use the ISE-730 Oxygen Electrode

Equipment Setup

- 1) Plug the DIN8 cable into a DIN8 transducer input of an iWorx data acquisition unit or amplifier.
- 2) Attach the cable of the oxygen electrode to the BNC connector on the DO2-100 Current-to-Voltage adapter.
- 3) Place the electrode in the reaction chamber or a beaker of the solution to be measured.

Calibration of the ISE-730 Oxygen Electrode

- 1) Place a magnetic stir bar in a beaker and fill the beaker with fresh deionized water before proceeding with the calibration procedure.
- 2) Place the electrode in the beaker and turn up the speed of the stir bar to the maximum rate that allows the stir bar to rotate evenly. If the solution in the chamber is stirred, changes in oxygen concentration reach the electrode instantaneously. If a stirrer is not used, changes in the rate of oxygen production are limited by the rate of diffusion.
- 3) Begin recording the output of the oxygen electrode. Note on the recording that the measurement is from water saturated with oxygen at room temperature. Note the room temperature.
- 4) When the trace settles down to steady baseline, record for an additional 10 seconds before going to the next step.
- 5) Replace the water in the beaker with a 0% oxygen solution, or club soda from a freshly opened container. Do not stir the liquid. Place the electrode in the liquid and immediately record the oxygen concentration in the chamber until the trace is a flat line at a lower amplitude. Note that this is the recording for a liquid depleted of oxygen at room temperature.
- 6) Stop the recording.
- 7) Determine the temperature ($^{\circ}\text{C}$) and the barometric pressure in the lab. The oxygen concentrations in deionized water, over a short range of temperatures at 760mmHg, have been calculated and listed in Table 1. The absorption coefficients of oxygen and the vapor pressures of water at these temperatures are also listed.



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- 8) The concentration of oxygen dissolved in deionized water, or its solubility (S), can be determined more accurately by using the following equation:

$$S = (a/22.414) ((P-p)/P) (r\%/100)$$

where a is the absorption coefficient of O₂ at temperature, p is the vapor pressure of water at temperature, P is the barometric pressure, and r% is the percent oxygen in the air. At 26°C and 760mmHg, assuming the concentration of oxygen in air is 21%, S = 252µMO₂:

$$(0.02783/22.414\text{L/mole}) ((760-25.09\text{mmHg})/760\text{mmHg}) (21/100) = 252\mu\text{MO}_2$$

Units Conversion

- 1) Scroll to the beginning of the calibration data.
- 2) Use the **Display Time** icons to adjust the **Display Time** of the **Main window** to show the complete calibration data on the same window.
- 3) Click the **2-Cursor** icon on the LabScribe toolbar so that two blue cursors appear on the **Main window**.
- 4) Place **Cursor 1** on the section of data collected when the ISE-730 was in the solution of oxygen-saturated deionized water, and **Cursor 2** on the section of data collected when the electrode was in the oxygen-depleted water.
- 5) Open the channel menu by clicking on the down arrow to the left of the channel's title. Select **Units** from this menu and **Simple** from the submenu to open the **Simple Units Conversion** dialog.
- 6) In the **Simple Units Conversion** dialog window, make sure **2 point cal** is selected in the pull-down menu in the upper left corner of the window. Put check marks in the boxes next to **Apply Units to new data** and **Apply Units to all blocks**.
- 7) Notice that the voltages from the two conditions are automatically entered into the value equations. Enter the values for the two oxygen concentrations in the calibration recording in the corresponding boxes on the right side of the conversion equations. For **Cursor 1**, enter the saturated oxygen concentration taken from Table 1 or derived from the calculation. For **Cursor 2**, enter zero.
- 8) Enter the name of the units, **µM**, in the box below the values.
- 9) Click on the **OK** button to activate the units conversion.
- 10) Select **Save** in the **File** menu and type a name for the file.



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Table 1: Oxygen Concentration [O₂] in Air-Saturated Deionized Water at 760mmHg H₂O Vapor

Temperature (°C)	O ₂ Absorbance Coefficient (a)	Water Vapor Pressure (p) (mmHg)	[O ₂] (μM)
20	0.03102	17.54	284
21	0.03044	18.65	278
22	0.02988	19.83	273
23	0.02934	21.07	267
24	0.02881	22.38	262
25	0.02831	23.76	257
26	0.02783	25.09	252
27	0.02736	26.74	247
28	0.02691	28.35	243
29	0.02649	30.04	238

Experiments

LabScribe experiments using the ISE-730 Oxygen Electrode and the DO2-100 Current-to-Voltage Adapter include:

- **Experiment CM-3: Mitochondrial Respiration** (found in the **Cellular Metabolism** category of the LabScribe **Settings** menu as **Mitorespiration**)
- **Experiment CM-5: Carbon Dioxide Fixation in Intact Cells** (found in the **Cellular Metabolism** category of the LabScribe **Settings** menu as **CO₂Fixation**)
- **Experiment CM-1: Oxygen Consumption and Size** (found in the **Cellular Metabolism** category of the LabScribe **Settings** menu as **O₂Consumption-Size**)
- **Experiment CM-4: Photosynthesis in Isolated Thylakoids** (found in the **Cellular Metabolism** category of the LabScribe **Settings** menu as **Photosynthesis**)
- **Experiment CM-7: Oxygen Consumption and Aerobic Respiration in Goldfish** (found in the **Cellular Metabolism** category of the LabScribe **Settings** menu as **O₂Consumption-AerobicResp**)
- **Experiment GB-3: Water Quality** (found in the **General Biology-Ecology** category of the LabScribe **Settings** menu as **WaterQuality**)

Care and Maintenance of the ISE-730 Micro-electrode

Assembly and Preparation

- 1) Unscrew the acrylic housing with the affixed Teflon™ membrane from the oxygen electrode.
- 2) Check the level of electrolyte solution in this housing. The minimum height of electrolyte in the housing should be 6 mm.



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- 3) If solution needs to be added to the housing, attach a filling tip to the bottle of electrolyte and fill the housing to the minimum height. To fill the housing without adding bubbles, gently place the bubble-free end of the filling tip against the inside of the Teflon™ membrane and add electrolyte until its level reaches the minimum height.
- 4) Insert the electrode tip into the housing. Be careful not to trap any air bubbles near the electrode tip. Screw the housing onto the body of the electrode until it stops.
- 5) Check the tip of the electrode for proper seating within the housing. If the electrode protrudes slightly beyond the end of the housing, it is seated correctly.

Handling

When necessary, the membrane of the electrode can be replaced by following the assembly and preparation procedure above.

Note: When removing and replacing a membrane as well as when calibrating or making measurements, be careful not to apply pressure against the internal electrode. Any excessive pressure against the internal electrode can cause the electrode to crack rendering it useless and unreparable.

Cleaning

When using the electrode in solutions containing protein, the electrode should be soaked in an enzyme cleaning solution after each use for a couple of minutes to remove the protein from the membrane surface. This will prolong the useful life of the membrane.

Storing

Always clean and rinse the electrode before storing. For long-term storage over 1 month:

Remove the membrane housing from the electrode. Rinse the internal electrode with distilled water and pat dry. Place a new, unfilled membrane housing over the internal electrode and attach loosely (do not seat completely). This membrane will serve to keep the dust off of the electrode tip.

For short-term storage, the electrode can be left in room air with the membrane housing still attached.

Electrode Filling Solution (included)

95% Ethylene Glycol

268 mM NaHCO₃

10mM KCl



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