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## **INSTRUCTIONS FOR USE**

### **NEEDLE OXYGEN ELECTRODE**

**PRODUCT #768-20R / #768-21R**

**NOT INTENDED FOR USE ON HUMANS**

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## I. GENERAL INFORMATION

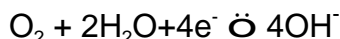
### A. DESCRIPTION

Diamond General's 768-20R / 768-21R needle electrodes are intended for use in biological and physiological or vivo in vitro systems. It is a non-clark style electrode with a built in Ag/AgCl reference electrode.

### B. SYSTEM THEORY

#### 1. THEORY OF POLAROGRAPHIC OXYGEN MEASUREMENTS

When two electrodes are polarized with a potential of slightly less than -1.0 volt in a electrolytic solution containing dissolved oxygen, current will flow as a result of the reduction of oxygen at the cathodic (negatively polarized) surface. This reaction at the cathode is expressed as:

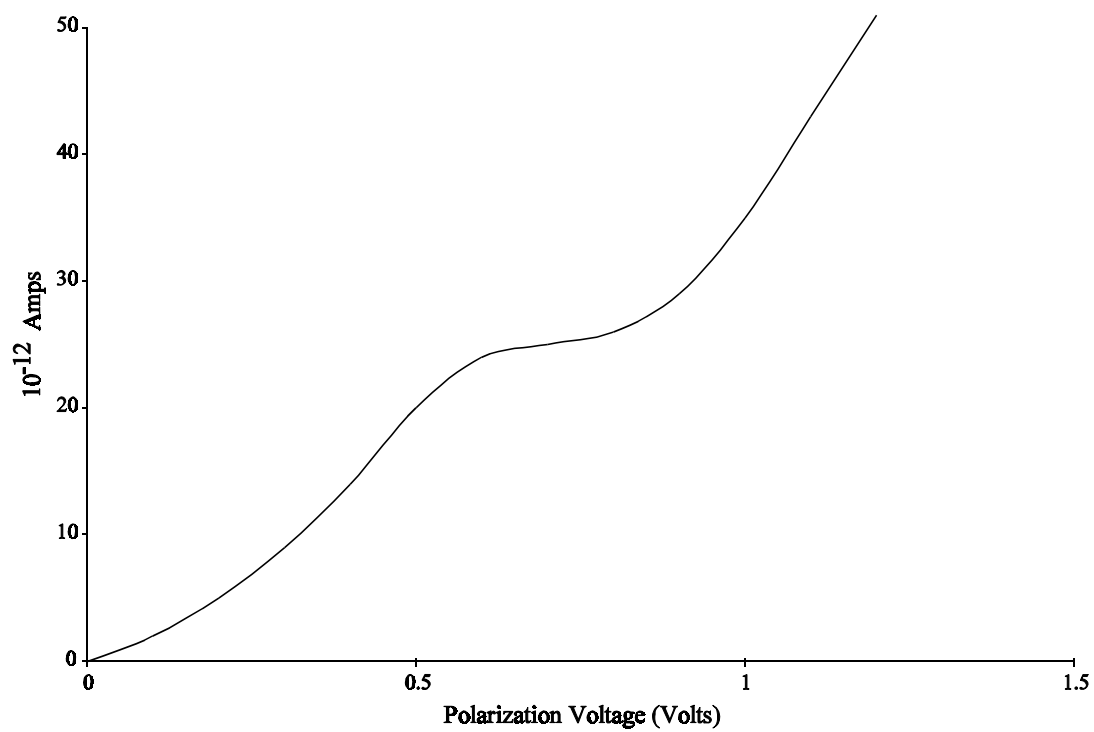


At the other electrode, described as the reference, oxidation takes place. For an Ag/AgCl reference, the reaction is:

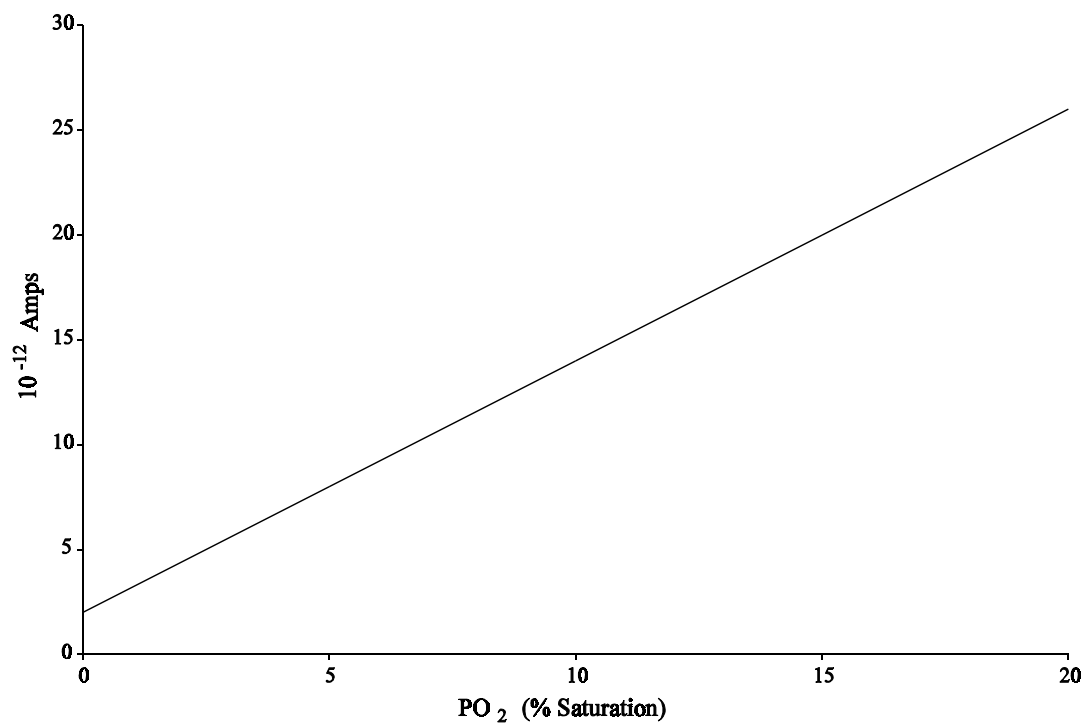


The voltage-current relationship for a polarographic oxygen electrode is represented by the characteristic curve (Figure 1). In the region below approximately -0.5 volt, there is a reasonably linear voltage-current relationship. As the polarization voltage is increased beyond -0.5 volt, the current will tend to reach a plateau in which changes in voltage have little effect on current. In this plateau region the current is limited by the rate at which oxygen can diffuse to the cathode. As the voltage is increased above -1.0 volt, the current will again increase with voltage, due to the reduction of other elements in addition to oxygen.

The electrode is normally operated with the polarization voltage set to the midpoint of the plateau region, in which case the current is diffusion limited. In a diffusion-limited condition, virtually all of the oxygen molecules which reach the cathode are immediately reduced, resulting in a zero oxygen concentration at the cathode surface and a current which is limited by the rate at which oxygen can diffuse to this zero concentration region. The diffusion rate is a function of the oxygen diffusion coefficient of the membrane and media surrounding the cathode and the dissolved oxygen concentration which, in turn, is proportional to the oxygen partial pressure and temperature.



**Figure 1 Characteristic Curve**



**Figure 2 Standard Curve**

The result is that, for a constant temperature, current flow through the electrode will be directly proportional to the partial pressure ( $P_{O_2}$ ) of oxygen.

A plot of the relation between current and  $P_{O_2}$  (at a fixed polarization voltage) is called the standard curve (Figure 2). For most electrodes, the curve is linear. It should be noted that the curve does not intersect the origin, but rather, indicates a small current at zero  $P_{O_2}$ . This current is called the residual, or "dark current," and results from factors such as electrical leakage through insulating materials in the system and reduction of oxygen which was absorbed into the electrode materials.

## 2. PRACTICAL CHARACTERISTICS OF OXYGEN ELECTRODES

When using miniature and micro-sized oxygen electrodes, the electrode characteristics are sometimes less than ideal. The smaller the electrode, the more difficult it is to maintain identical characteristics from one electrode to the next. Also, the wide variety of applications in which these electrodes are used makes it difficult to optimize the system for a particular application. However, with a proper understanding of the electrode characteristics, accurate oxygen measurements can be obtained.

The shape of the characteristic curve (Figure 1) can vary considerably between electrodes and also can change with shelf life and use. The plateau is frequently not flat, but might have a small positive or negative slope. Also, it might cover a span of 0.1 to 0.4 volt in width, with a midpoint occurring anywhere between 0.5 and 0.95 volt.

If we define the plateau as the region of the characteristic curve which has the minimum slope and operate the electrode at a voltage occurring near the midpoint of this region, the standard curve will be approximately linear (Figure 2). Occasionally, the characteristic curve might vary considerably from the classic and have a poorly defined plateau, but the linearity of the standard curve is not simultaneously degraded.

## II. ELECTRODE SPECIFICATIONS

Length (needle)	1 1/2 inches
Tip Diameter	768-20R .035 inches
	768-21R .032 inches
Typical Current (Ambient $PO_2$ )	500 x 10 <sup>-12</sup> amps
Time Constant (0-90%)	< 2 seconds
Membrane Thickness	20 microns
Temperature Coefficient	+5% per C
Reference Electrode	Ag/AgCl

### III. PRECAUTIONS

#### A. THIS ELECTRODE IS NOT INTENDED FOR USE ON HUMANS.

#### B. HANDLING

Although the 768-20R electrode is made of a durable construction, care should be taken not to drop the electrode, bend or exert other sharp blows, as the electrode consists of glass tubing within the stainless steel needle.

#### C. Do not immerse the electrode above the hub of the needle.

#### D. To extend the longevity of the electrode, store dry and hydrate only before use.

### IV. PROCEDURES

#### A. OPERATION

**Note:** An internal AgCl reference electrode is used, and must be hydrated before use. Therefore soaking of the electrode in saline is essential. This soaking period is accomplished when the electrode is polarized before use as described in the Stabilization section (C) below.

#### B. THE SELECTION OF POLARIZATION VOLTAGE AND PRODUCTION OF THE CHARACTERISTIC CURVE.

Set up the amplifier following the appropriate instructions. Most oxygen electrodes function well when polarized with a potential of  $-0.70$  volts; therefore, it is not really necessary for the user to produce a characteristic curve. Simply stabilize the electrode at the operating voltage ( $-0.70V$ ) and proceed to Section IV C. It is recommended to use  $-0.70 V$  as the polarizing voltage. If you wish, you may produce your own characteristic curve with following this procedure:

- If the electrode has not yet been in use, you must polarize the electrode in physiological saline (or other calibrating media) for one hour. This polarization period hydrates the reference anode and stabilizes the sensing cathode.

- Set the amplifier to indicate current in  $10^{-11}$  amperes. (If using DG's 1201 Chemical Microsensor, set the **COARSE GAIN** to  $10^{-9}$ , the **COARSE ZERO** to  $10^{-12}$ , both the **FINE GAIN** and **FINE ZERO** full counterclockwise.)

- Equilibrate a beaker of physiological saline (80 ml if using DG's Calibration Cell, Product #1251) with room air (approximately 21% oxygen).

- Set the polarization voltage on the amplifier to an initial value of -1.0 volt.
- Allow several minutes for the reading to stabilize. A strip chart recorder (1.0 volt full scale sensitivity) will aid in observing the stabilization.
- Record the current reading and decrease the polarization voltage to -.95 volts.
- Repeat the above procedure at 0.5 volt intervals allowing the electrode to stabilize at each voltage change.
- Plot the characteristic curve and note the plateau defined as the region of minimum slope. The voltage at the midpoint of the plateau should be chosen as the operating voltage for the electrode.

### C. STABILIZATION

Allow the electrode to stabilize at the operating voltage (normally -.70V) and temperature at which measurements are going to be made for 1 hour or until stabilization occurs. A strip chart recorder will aid in observing stabilization.

### D. CALIBRATION

Calibration must be carried out at the same temperature at which the measurement site will be. Since oxygen partial pressure and current have a linear relationship, a 2-point calibration will suffice for most applications.

The simplest type of calibration involves using 21% O<sub>2</sub> (ambient room air) and 0% O<sub>2</sub> (100% nitrogen) to produce 2 calibration points. If measurements are limited to a very small range and high accuracy is required, we recommend that an oxygen-nitrogen gas mixture, which has an oxygen concentration similar to that of the substance being tested, be used in place of the air. One calibration should always include the zero point (0% O<sub>2</sub>), and thereby allow compensation for the "dark current".

First, bubble the higher level O<sub>2</sub> gas through the solution containing the electrode. Allow 10 to 15 minutes for the solution to equilibrate with the gas. If using the Chemical Microsensor, dial the **FINE GAIN** until the displayed P<sub>O<sub>2</sub></sub> matches that of the precalibrated gas. (The **COARSE GAIN** should be preset to read 10<sup>-9</sup>. Set the **COARSE ZERO** two orders of magnitude above that of the **COARSE GAIN**. Example: if **COARSE GAIN** is set to 10<sup>-9</sup>, **COARSE ZERO** should be set to 10<sup>-11</sup>).

Next, bubble 100% nitrogen until the solution is saturated. As previously mentioned, there will be a small residual or "dark current" displayed. This small offset (normally less than 10% of the direct current reading at 21% O<sub>2</sub>) must be subtracted electronically at the amplifier. On the Chemical Microsensor, the **FINE ZERO** control is used. Set the **FINE ZERO** until a reading of 00.0 is displayed on the amplifier. Repeat this procedure (with both gases) readjusting the **FINE GAIN** as necessary to complete an accurate calibration. The electrode is now ready to be removed from the calibrating media and inserted into the experimental media (see PRECAUTIONS, III B).

**Note:** If the electrode has been subjected to a temperature change while transferring from the calibrating media to the experimental media, it may take several minutes to restabilize in the experimental media. Also, polarization voltage is temporarily disconnected when electrode is removed from solution. For these reasons, it is important to transfer the electrode as quickly as possible.

#### E. DRIFT

Regardless of the calibration methods chosen, it is important to realize that some electrodes will drift slightly, even after their initial stabilization. For this reason, we recommend that when high accuracy is required, a calibration procedure be utilized both before and after the experiment. If the experiment is over several hours in length and the experiment design will allow for it, additional calibration checks during the experiment would be advisable.

#### F. NOISE

The noise level of the electrode is sufficiently low that it will not interfere with accurate P<sub>O<sub>2</sub></sub> readings. If excessive noise is observed, it is probably due to external sources. The system is susceptible to external interference due to the extremely low currents and high impedances involved. Good grounding and shielding practices should always be observed, and operation in a cage is sometimes advisable. Also, movement of people in the immediate vicinity can cause current fluctuations due to the resulting stray capacitance variations.



## **V. ELECTRODE MAINTENANCE**

### **A. STORAGE**

When not in use, the electrode should be stored dry in a dust free container.

### **B. CLEANING**

After use, submerge the tip of the electrode for 15-30 minutes in deionized H<sub>2</sub>O to remove salts from the membrane.

## **VI. RECOMMENDED ACCESSORIES**

**CALIBRATION CELL (Product No. 1251)**

**CHEMICAL MICROSENSOR (Product No. 1201)**

**CHEMICAL MICROSENSOR II (Product No. 1231)**