INSTRUCTION MANUAL

ISO-NOPF500-Cxx
Microsensor for NO measurement

Serial No._____________________

www.wpiinc.com
ABOUT THIS MANUAL

The following symbols are used in this guide:

⚠️ This symbol indicates a CAUTION. Cautions warn against actions that can cause damage to equipment. Please read these carefully.

⚠️ This symbol indicates a WARNING. Warnings alert you to actions that can cause personal injury or pose a physical threat. Please read these carefully.

NOTES and TIPS contain helpful information.

Fig. 1—The ISO-NOPF500 sensor is a carbon fiber electrode that works like the traditional ISO-NOP sensor.
INTRODUCTION

The ISO-NOPF500 is a nitric oxide sensor designed like the dry, carbon fiber ISO-NOPF sensors, however, it works like a traditional ISO-NOP. The sensor can be ordered in a variety of lengths from 5–10mm. The standard length is 8mm. It incorporates WPI’s proprietary combination electrode technology in which the nitric oxide-sensing element and separate reference electrode are encased within a single shielded sensor design.

The ISO-NOP was the original nitric oxide sensor, ideal for cell cultures, cell suspensions and many other applications. The new ISO-NOPF500 can be used in the same way, but offers several advantages:

- It is much easier to use, because requires no sleeves or filling solutions.
- Like the other ISO-NOPF sensors, it is flexible and durable.
- Its high sensitivity provides a rapid response time. It is ten times more sensitive than the ISO-NOP.
- The ISO-NOPF500 can be used in acidic conditions.
- The sensor tip is longer than the sensing tip of the ISO-NOP.
- It offers a bigger linear range than the ISO-NOP, and the range is based on the length of the sensor tip.
- The sensor can be calibrated with either the SNAP or Nitrite method.

Fig. 2—The ISO-NOPF500-Cxx is used like the other carbon fiber NO microsensors.
Notes and Warnings

The NO carbon fiber microsensors are robust, but not indestructible. Exercise caution when handling the NO sensor to avoid actions that could damage the tip. Do not bring the tip into contact with hard surfaces like stir bars. See “Unpacking” on page 3.

⚠️ CAUTION: DO NOT EXPOSE SENSOR TO ORGANIC SOLVENTS.

⚠️ CAUTION: To avoid the contamination of solutions, thoroughly rinse and dry the sensor before changing calibration methods.

Parts List

After unpacking, verify that there is no visible damage to the sensor. Verify that all items are included:

(2) NO microsensors
(2) Sensor Performance Evaluations (each sensor is tested individually at WPI)
(1) Instruction Manual

Unpacking

Upon receipt of this sensor, make a thorough inspection of the contents and check for possible damage. Missing cartons or obvious damage to cartons should be noted on the delivery receipt before signing. Concealed damage should be reported at once to the carrier and an inspection requested. Please read the section entitled “Claims and Returns” on page 15 of this manual. Please contact WPI Customer Service if any parts are missing at 941-301-1003 or customerservice@wpiinc.com.

Returns: Do not return any goods to WPI without obtaining prior approval (RMA # required) and instructions from WPI’s Returns Department. Goods returned (unauthorized) by collect freight may be refused. If a return shipment is necessary, use the original container, if possible. If the original container is not available, use a suitable substitute that is rigid and of adequate size. Wrap the instrument in paper or plastic surrounded with at least 100mm (four inches) of shock absorbing material. For further details, please read the section entitled “Claims and Returns” on page 15 of this manual.
OPERATING INSTRUCTIONS

Environmental Influences
There are two environmental parameters to which NO sensors are quite sensitive: temperature and electrical interference.

Temperature
The background current (and to a lesser degree) the selectivity of the NO sensor is affected by temperature. This is due to the effects of temperature on the partial pressure of dissolved NO gas in liquid samples, on the permeability of the coatings and on the conductivities of various sensor components. It is recommended that a calibration procedure be performed at the same temperature as the experiment and that temperature be held constant during NO measurement.

Electrical Interference
External, electrical noise sources (like magnetic stirrers, fluorescent lights, MRI machines, electric motors, computers, pumps and other electrical instruments) may couple into the sensor signal path electromagnetically and impose undesirable signals in the output record. The magnitude of this external noise depends on the environment of the laboratory. If the interference introduced by the electrical signals in the environment is large, identify the noise source and remove it. It is also important to ground and shield the system properly.

TIP: Refer to your free radical analyzer manual for proper grounding and shielding techniques. (In the TBR4100 or Apollo1000 manuals, see “Grounding and Noise Concerns” in the Operating Instructions section.)

Attaching the Sensor to the Microsensor Handle
Once removed from the package, plug the microsensor into a microsensor cable (WPI #91580) connected to the free radical analyzer (Fig. 3). *Be very careful that the sensor tip does not come into contact with anything that could damage it. The sensor should plug in easily. If you encounter resistance, it is probably due to misalignment of the sensor plug with the socket connector inside the microsensor cable. Simply realign the sensor by gently rotating it until it snaps into place.

*NOTE: Current WPI free radical analyzers include the TBR4100, TBR1025 and Apollo1000. Apollo4000 was the original 4-channel WPI free radical analyzer.
Polarizing the Sensor

When a non-polarized microsensor is initially connected to a free radical analyzer, it may display a high (sometimes off-scale) background current. The polarization voltage applied by the instrument causes a reduction of the background current to a stabilized baseline value over time. Set the poise voltage to 865mV. (For the TBR4100/1025, set the Probe Select dial to NO.) The amount of time required to reach a stable baseline current varies for each sensor. New sensors typically take longer, on the order of eight hours.

The Performance Evaluation included with your sensor shows the baseline current and the sensitivity of your sensor when it was quality tested at WPI. (In addition, it shows the polarization time of your sensor in the WPI labs.) The baseline value attainable in your lab may be slightly higher or lower, depending on the temperature* and composition of the test media. For initial performance verification of an ISO-NOPF500 in your lab, WPI recommends using the KI/H$_2$SO$_4$ calibration method described on page 6. Once a stable baseline current is achieved (usually between 3,300-50,000pA), the microsensor is ready for use.

*NOTE: The background current of the sensor will usually increase with increasing temperature of the experiment. Although the sensitivity of the sensor does not change significantly within the range 20-37°C, it is recommended that the calibration procedure be performed at the same temperature as the experiment.

Calibrating the Sensor

This unique sensor can be calibrated in two ways:
- Chemical generation of NO (KI/H$_2$SO$_4$)
- Decomposition of the S-nitrosothiol NO-donor (SNAP)
Calibration by Chemical Generation of NO

**CAUTION:** To avoid the contamination of solutions, thoroughly rinse and dry the sensor before changing calibration methods.

**CAUTION:** This calibration method may only be used with the ISO-NOP and ISO-NOP500 sensors. DO NOT use with other sensors. It will cause permanent damage.

This procedure involves making two solutions (0.1M $\text{H}_2\text{SO}_4 + 0.1$ M KI, and 50µM KNO$_2$ or NaNO$_2$) and then running a calibration procedure. It is based on the following reaction:

$$2\text{KNO}_2 + 2\text{KI} + 2\text{H}_2\text{SO}_4 \rightarrow 2\text{NO} + \text{I}_2 + 2\text{H}_2\text{O} + 2\text{K}_2\text{SO}_4$$

A known amount of KNO$_2$ is added to produce a known amount of NO. The quantity (and so the concentration) of NO generated can be calculated directly from the stoichiometry if the concentrations of the reactants are known. Since KI and $\text{H}_2\text{SO}_4$ are present in great excess, the limiting reagent is KNO$_2$. Experiments have demonstrated that the nitric oxide generated from this reaction will persist long enough to calibrate the NO sensor easily and accurately. Since the reaction goes to completion, the equation above states that the ratio between KNO$_2$ and NO is 1:1. Therefore the amount of NO generated in the solution will be equal to the amount of KNO$_2$ added. The final concentration of NO will be equal to the diluted concentration of KNO$_2$ in the solution.

**Preparing Solution #1: 0.1M $\text{H}_2\text{SO}_4 + 0.1$ M KI**

**NOTE:** Prepare fresh solution daily.

1. Pour 20mL of 0.1M $\text{H}_2\text{SO}_4$ into a 20mL glass vial.
2. Drop a small stirring bar into the solution and position the vial on top of a magnetic stirring plate. Turn on the stirrer so that the bar is stirring at a moderate rate.
3. Add 0.33g KI and mix.

**Preparing Solution #2: 50µM KNO$_2$ (or NaNO$_2$)**

The recommended method for preparing this solution is to purchase an ion chromatography liquid nitrite standard (NaNO$_2$ or KNO$_2$) and dilute it as appropriate. Standard nitrite is available from WPI (#7357).

Alternatively, crystalline reagent KNO$_2$ may be used, however, KNO$_2$ is extremely hygroscopic and degrades once exposed to atmospheric moisture. Therefore, WPI recommends purchasing the reagent packaged under argon if the crystalline reagent is used. (It is available from Eastman Kodak Chem #105 7462.) Store it in a desiccator. While this extends the life of the reagent, it needs to be replaced more frequently than the liquid standard. Store the standard nitrite solution prepared from this compound in a gas-tight bottle in the refrigerator.
Calibration Procedure

Once the sensor is polarized, it can be calibrated. The polarized sensor should already be immersed in Solution #1 and plugged into the free radical analyzer. Set the range to 10nA (or 100nA, as required) and the poise voltage to 865mV. The following example briefly describes the fundamental concepts behind a standard calibration protocol. Known concentrations of NO are generated in Solution #1 by adding a known volume of a the NO standard (Solution #2).

**NOTE:** For additional sensor calibration procedures and calibration theory, refer to the free radical analyzer manual. Most WPI manuals can be downloaded directly from [www.wpiinc.com](http://www.wpiinc.com). This information can also be e-mailed when you contact the WPI Technical Support team at 941-301-1003 or technicalsupport@wpiinc.com.

1. Place the 20mL vial of Solution #1 on the magnetic stirring plate with the stir bar still inside. Turn on the stirrer so that the bar is stirring at a moderate rate.

   **NOTE:** This rate of the stirrer should NOT be modified once it is set.

   **NOTE:** The calibration should be carried out at the same temperature at which the experimental measurements of NO are to be made. This can be accomplished by placing the vial and stand in a water bath at the appropriate temperature and allowing the temperature of the solution in the vial to equilibrate with the water bath.

2. Immerse the tip of the ISO-NOPF500 sensor in the solution and secure it in an electrode holder such as WPI’s Pro-Guide (WPI #47510, 47520, 47530, 47540) or a micromanipulator. The sensor tip should be immersed about 10–15mm into the solution. It should not contact the stir bar, which could damage the fiber.

   **NOTE:** Generally, it is not necessary to pre-purge the calibration solution, since the NO decays slowly in this solution. However, if you feel it is necessary to de-gas Solution #1 prior to calibration, insert a long stainless steel needle through a septum so that the tip is in the solution. Attach the needle through appropriate tubing to a source of pure argon gas (nitrogen may also be used). Insert a short needle through the septum so that the needle tip is clearly exposed inside the vial (not in the solution). The small needle allows gas to escape, thereby avoiding a buildup of pressure. Purge the solution at low pressure (5PSI or less) for 15 minutes. Once purging is complete and the gas source is turned off, remove the purging and pressure relief needles.

3. Wait until the current on the display stabilizes again before continuing and record the value. This may take several minutes if the sensor has undergone a large temperature change. The quiescent baseline current is an indicator of the health of the sensor.

4. To the vial containing 20mL of Solution #1, sequentially inject five aliquots of Solution #2 (2µL, 4µL, 8µL, 16µL and 32µL) into the glass vial. Typically, each aliquot is twice the volume of the previous one. The current output jumps rapidly after each addition and then plateaus. As soon as it reaches a plateau, inject the next aliquot.
The reaction produces NO gas. When NO gas passes through the gas permeable coating, it generates an output current that is measurable, and the results can then be graphed using a third party spreadsheet with graphing capability like Microsoft® Excel.

The output from the **TBR4100/1025** looks similar to the example shown in **Fig. 3**. Here five sequential additions of Solution #2 were made to Solution #1.

**Fig. 4**—The Calibration Curve shows five aliquot additions.

5. Calculate the concentration of NO in solution after the each aliquot injection of Solution #2 to 20mL of Solution #1. The concentration of NO produced can be calculated as follows: $M_1 V_1 = M_2 V_2$ where $M$=molarity and $V$=volume

- **1st Aliquot**: $2\mu L + 20mL = 20,002\mu L$
  $50\mu M(2\mu L) = M_2(20,002\mu L)$
  $100\mu M/20,002 = 0.0049\mu M = 5nM$

- **2nd Aliquot**: $4\mu L + 20,002\mu L = 20,006\mu L$
  $50\mu M(4\mu L) = M_2(20,006\mu L)$
  $200\mu M/20,006 = 0.0099\mu M = 10nM$

- **3rd Aliquot**: $8\mu L + 20,006\mu L = 20,014\mu L$
  $50\mu M(8\mu L) = M_2(20,014\mu L)$
  $400\mu M/20,014 = 0.0199\mu M = 20nM$

- **4th Aliquot**: $16\mu L + 20,014\mu L = 20,030\mu L$
  $50\mu M(16\mu L) = M_2(20,030\mu L)$
  $800\mu M/20,030 = 0.0399\mu M = 40nM$

- **5th Aliquot**: $32\mu L + 20,030\mu L = 20,062\mu L$
  $50\mu M(32\mu L) = M_2(20,062\mu L)$
  $1600\mu M/20,062 = 0.0797\mu M = 80nM$
6. Record the calculated values in a table similar to the one below. Record the difference in current output (pA) generated by the addition of the known quantities of KNO₂ in a table similar to the one below.

<table>
<thead>
<tr>
<th>Amount added</th>
<th>Total Volume</th>
<th>[NO] nM</th>
<th>Response (pA)</th>
</tr>
</thead>
<tbody>
<tr>
<td>2µL</td>
<td>20,002µL</td>
<td>4.99nM</td>
<td>162.6pA</td>
</tr>
<tr>
<td>4µL</td>
<td>20,006µL</td>
<td>9.99nM</td>
<td>357.0pA</td>
</tr>
<tr>
<td>8µL</td>
<td>20,014µL</td>
<td>19.9nM</td>
<td>902.8pA</td>
</tr>
<tr>
<td>16µL</td>
<td>20,030µL</td>
<td>39.9nM</td>
<td>1911.1pA</td>
</tr>
<tr>
<td>32µL</td>
<td>20,062µL</td>
<td>79.7nM</td>
<td>3813.7pA</td>
</tr>
</tbody>
</table>

7. Construct a standard calibration curve using the recorded data. Using a third party spreadsheet with graphing capability like Microsoft® Excel, it is possible to generate a linear regression analysis that will display the equation and the R² coefficient. To do this in Excel, enter the data and generate a “scatter plot” graph. Then, select the line and right click. Choose Add Trendline. The Add Trendline dialog box appears. On the Type tab, select Linear, and on the Options tab, select the Display equation on chart and Display R-value on chart.

![Calibration Data](image)

**Fig. 5**—The Calibration Output shows a linear response with an R² factor near 1.

The slope of this curve indicates the sensitivity of the sensor. Once the sensitivity of the sensor is determined, the sensor is ready to use experimentally. (In the above example, the sensitivity is 49.0pA/nM.)

### Calibrating with SNAP

**CAUTION**: To avoid the contamination of solutions, thoroughly rinse and dry the sensor before changing calibration methods.

### Understanding SNAP

SNAP is a stable NO-containing compound that can be used for quantitative generation of NO in solution. SNAP decomposes to NO and a disulfide byproduct when dissolved in water. However, the rate of decomposition is very slow. The kinetics of decomposition for this reagent is a function of several parameters including pH, presence of a catalyst, temperature and light.
In the procedures described here, SNAP is used in combination with a catalyst, cuprous (I) chloride (CuCl) or cupric (II) chloride (CuCl\(_2\)), to generate a known quantity of NO in solution. Note that this protocol does not investigate the effects of all parameters involved in SNAP decomposition, nor does it propose a model by which NO is decomposed. The presented procedures provide an empirical estimation of the amount of generated NO based on the molarity of a standard (stock) solution of SNAP under a controlled set of parameters.

**NOTE:** Remember that most NO sensors are sensitive to temperature changes. It is therefore recommended that the calibration of a NO sensor is performed at the experimental temperature.

**Method 1: SNAP using CuCl**

This method of calibration results in the 100% conversion of SNAP to NO. The amount of NO produced, therefore, is based on the final concentration of SNAP.

**NOTE:** The described calibration procedure requires the use of cuprous chloride, CuCl, where CuCl is the active catalyst for the conversion of SNAP to NO. The calibration curve assumes only the presence of CuCl and hence a 100% conversion efficiency of SNAP to NO. However, in the presence of oxygen CuCl is readily oxidized to CuCl\(_2\). This will happen naturally if the compound is exposed to air and/or there is inadequate storage of CuCl. The oxidation product CuCl\(_2\) is much less efficient at catalyzing the conversion of SNAP to NO, and this would appear during calibration as an apparent low sensitivity of the electrode to NO. Since CuCl is readily oxidized to CuCl\(_2\) special precautions must be taken to keep it in its reduced state prior to any calibration. It is recommended that CuCl be stored under inert conditions, and (if used in solution) the solution must be degassed with inert gas and must be absent of all oxygen.

**NOTE:** If your laboratory is not adequately equipped to satisfy the conditions for storage and use of CuCl, use the CuCl\(_2\) method (Method 2) on page 11.

**Preparing Solutions**

- **Solution #1** (Saturated solution of CuCl): Add 150mg CuCl to 500mL distilled deoxygenated water. The saturated CuCl solution will have a concentration of approximately 2.4mM at room temperature and should be kept in the dark prior to use.
  
  **NOTE:** The distilled water can be deoxygenated by purging with pure nitrogen or argon gas for 15 minutes.

- **Solution #2** (Standard SNAP solution): Add 5.0mg EDTA to 250mL of water and deoxygenate the solution. Then add 5.6mg of SNAP and dissolve it completely.

**Calibration Procedure**

1. Within a nitrogen or argon environment, place 10.0mL of Solution #1 (CuCl) in a 20mL vial.
2. Drop a small stirring bar into the solution, and place the vial on a magnetic stirring plate.
3. Immerse the ISO-NOPF500 sensor into this solution. While stirring allow the sensor to polarize until the background current stabilizes. The sensor should respond and stabilize within 10 seconds and the expected baseline current value should be between 3,000 and 50,000pA, depending on the length of the sensor tip.

4. As soon as the background current stabilizes, begin recording the current output.

5. Sequentially inject five aliquots containing 2µL, 4µL, 8µL, 16µL and 32µL of Solution #2 (SNAP standard solution) into the vial containing Solution #1 (CuCl). Immediately following the first addition of SNAP into Solution #1, the current (pA) output increases rapidly. Within a few seconds the response reaches a plateau. Inject the second aliquot (4µL) as soon as the first signal reaches a plateau. Add the third aliquot (8µL) as the second signal reaches its plateau. If aliquots are not added promptly when reaching the previous plateau, the signal will slowly decline because generated NO is quickly oxidized to nitrite and nitrate, which will not be detected by the sensor. Add the fourth and fifth aliquots in the same manner.

6. Construct a calibration curve by plotting the signal output (pA) against the concentration (nM) of SNAP.

Method 2: SNAP using CuCl₂

Method 2 is similar to Method 1 using CuCl₂ as a catalyst. This convenient method is the one used in WPI laboratories. Experimentally it has been shown that CuCl₂ is less efficient as a catalyst in the conversion of SNAP to NO. (The conversion ratio is reduced to approximately 60%) However, it is technically easier to accomplish than Method 1, and it provides reliable data.

Preparing Solutions

- **Solution #1** (CuCl₂ solution): 250mL 0.1M CuCl₂ in distilled water
- **Solution #2** (Standard SNAP solution): Add 5.0mg EDTA to 250mL of water. Then, add 5.6mg of SNAP and dissolve it completely.

Calibration Procedure

1. Place 20.0mL of Solution #1 (CuCl₂) in a 20mL vial.
2. Drop a small stirring bar into the solution, and place the vial on a magnetic stirring plate.
3. Immerse an NO probe into this solution. While stirring allow the background current to stabilize. The sensor should respond and stabilize within 10 seconds and the expected baseline current value should be between 3,000 and 50,000pA, depending on the length of the sensor tip.
4. As soon as the background current stabilizes, begin recording the current output.
5. Sequentially inject five aliquots containing 2µL, 4µL, 8µL, 16µL and 32µL of the SNAP standard solution (Solution #2) into the vial containing Solution #1 (CuCl₂).
Immediately following the first addition of SNAP into Solution#1, the current (pA) output increases rapidly. Within a few seconds the response reaches a plateau. Inject the second aliquot (4µL) as soon as the first signal reaches a plateau. Add the third aliquot (8µL) as the second signal reaches its plateau. If aliquots are not added promptly when reaching the previous plateau, the signal will slowly decline because generated NO is quickly oxidized to nitrite and nitrate, which will not be detected by the probe. Add the fourth and fifth aliquots in the same manner.

**TIP:** You can adjust the volume of injected aliquots according to the concentration of SNAP stock solution. Decrease the volume of aliquot if the electrode is very sensitive or increase the volume of aliquot if the electrode is less sensitive.

6. Construct a calibration curve by plotting the signal output (pA) against the concentration (nM) of SNAP.

**MAINTENANCE**

**Storing the Sensor**

**STANDBY:** When not being used for a short period of time (such as overnight), the microsensor should remain attached to the microsensor cable and kept in solution. Before the next experiment, immerse the sensor in the experimental solution (like, Kreb's Buffer). The background current increases until it reaches a stable value. Do not be alarmed if the background current becomes elevated. This is associated with the hydration of the sensor and will not negatively affect the sensor's performance.

**LONG-TERM:** If the microsensor will not be used for more than three or four days, then it may be stored dry. Remove it from the microsensor cable and clean it. (See below.) Return it to the case in which it was shipped, being very careful to avoid making contact with the sensor tip.

**NOTE:** ALWAYS rinse with distilled water before storing the sensor dry.

The NO microsensor is a maintenance-free consumable sensor. When its performance is no longer satisfactory, remove it from the microsensor cable and dispose of it, replacing it with a new one.

**Cleaning the Sensor**

The sensor should be cleaned after each use by suspending the tip in distilled water for 20–30 minutes to dissolve salts and remove particles which may have accumulated on the fiber. If the sensor was used in a protein-rich solution, the tip should first be soaked in a protease solution for several minutes to remove protein build-up and then rinsed with distilled water. Enzymatic detergent (Enzol, WPI #7363-4) can also be used. The sensor can be sterilized chemically using an appropriate disinfectant (Cidex, WPI #7364). If necessary, gently dab the sensor with a Kimwipes® to remove residue.

**NOTE:** ALWAYS rinse with distilled water before storing the sensor dry.
## ACCESSORIES

### Table 1: Accessories

<table>
<thead>
<tr>
<th>Part Number</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>7357</td>
<td>Standard Nitrite Solution 1g/L (100mL)</td>
</tr>
<tr>
<td>7363-4</td>
<td>Enzol Enzymatic Detergent, 1 gallon</td>
</tr>
<tr>
<td>7364</td>
<td>Cidex Disinfecting Solution</td>
</tr>
<tr>
<td>91580</td>
<td>Microsensor Cable</td>
</tr>
<tr>
<td>47510</td>
<td>ProGuide Position/Holder with Base</td>
</tr>
<tr>
<td>47520</td>
<td>ProGuide Position/Holder</td>
</tr>
</tbody>
</table>
| 47530       | ProGuide Plus Position/Holder with fine adjust-
|             | ment                                        |
| 47540       | ProGuide Plus with Base                      |
| SNAP25      | SNAP, 25mg vial                              |
| SNAP50      | SNAP, 50mg vial                              |
| SNAP100     | SNAP, 100mg vial                             |

## TROUBLESHOOTING

<table>
<thead>
<tr>
<th>Issue</th>
<th>Possible Cause</th>
<th>Solution</th>
</tr>
</thead>
<tbody>
<tr>
<td>Baseline current is below</td>
<td>The poise voltage (sensor setting) may be incorrectly set.</td>
<td>Set the poise voltage to 865mV. (For the TBR, choosing the NO sensor setting selects 865mV automatically.) Set the range at 10nA or 100nA.</td>
</tr>
<tr>
<td>specified range.</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Unstable baseline</td>
<td>The sensor may be nearing the end of its usable life.</td>
<td>Perform a 5-point calibration set using the standard. If the sensor responds linearly within the desired concentration range, it is still useable. See “Calibrating the Sensor” on page 5.</td>
</tr>
<tr>
<td>If the baseline hasn't</td>
<td>If the baseline hasn't stabilized after 8 hours, the polarizing solution may be contaminated.</td>
<td>Prepare fresh polarizing solution. Use KI/H$_2$SO$_4$ or 0.1M CuCl$_2$.</td>
</tr>
<tr>
<td>stabilized after 8 hours</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Uneven aliquots may have been</td>
<td>External electrical interferences may be the problem.</td>
<td>Identify and isolate electrical interferences.</td>
</tr>
<tr>
<td>used</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Calibration data set is not</td>
<td>Uneven aliquots may have been used.</td>
<td>Check the pipetter calibration.</td>
</tr>
<tr>
<td>linear</td>
<td></td>
<td></td>
</tr>
<tr>
<td>The stock solutions have</td>
<td>The stock solutions have deteriorated.</td>
<td>Prepare fresh standard solution. See “Calibrating the Sensor” on page 5.</td>
</tr>
</tbody>
</table>
### Issue | Possible Cause | Solution
--- | --- | ---
Sensitivity below range specified | Foreign materials have been adsorbed on the sensor’s surface. | If the foreign materials are proteins, use and enzymatic cleanser like Enzol (WPI #7363-4) to remove the contaminant.
| The sensor has reached end of its usable life. | Replace the sensor.

**NOTE:** If you have a problem/issue with the sensor that falls outside the definitions of this troubleshooting section, contact the WPI Technical Support team at 941-301-1003 or technicalsupport@wpiinc.com.

### SPECIFICATIONS

This sensor conforms to the following specifications:

- **Outside Diameter**: \(500\mu\text{m}\)
- **Available Length**: \(5-10\text{mm}\)
  (length varies in 1mm increments—for example, 5mm, 6mm, 7mm. 8mm is standard. Sensor length is proportional to sensitivity.)
- **Response Time**: \(< 10\text{sec.}\)
- **Lowest Detection Limit/Range**: \(0.2\text{nM}\)
- **Nominal Sensitivity-New Sensor**: \(\geq 20 \text{ pA/nM}\)
- **Baseline Drift**: none
- **Poise Voltage**: \(865\text{mV}\)
- **Typical Quiescent Base-line Current, 25ºC**: \(5,000\text{pA}\)
- **Acceptable Baseline Range**: \(3,000-50,000\text{pA}\)
- **Polarization Time**: \(8+\text{hrs}\)
WARRANTY

WPI (World Precision Instruments, Inc.) warrants to the original purchaser that this equipment, including its components and parts, shall be free from defects in material and workmanship for a period of 30 days* from the date of receipt. WPI's obligation under this warranty shall be limited to repair or replacement, at WPI's option, of the equipment or defective components or parts upon receipt thereof f.o.b. WPI, Sarasota, Florida U.S.A. Return of a repaired instrument shall be f.o.b. Sarasota.

The above warranty is contingent upon normal usage and does not cover products which have been modified without WPI's approval or which have been subjected to unusual physical or electrical stress or on which the original identification marks have been removed or altered. The above warranty will not apply if adjustment, repair or parts replacement is required because of accident, neglect, misuse, failure of electric power, air conditioning, humidity control, or causes other than normal and ordinary usage.

To the extent that any of its equipment is furnished by a manufacturer other than WPI, the foregoing warranty shall be applicable only to the extent of the warranty furnished by such other manufacturer. This warranty will not apply to appearance terms, such as knobs, handles, dials or the like.

WPI makes no warranty of any kind, express or implied or statutory, including without limitation any warranties of merchantability and/or fitness for a particular purpose. WPI shall not be liable for any damages, whether direct, indirect, special or consequential arising from a failure of this product to operate in the manner desired by the user. WPI shall not be liable for any damage to data or property that may be caused directly or indirectly by use of this product.

Claims and Returns

Inspect all shipments upon receipt. Missing cartons or obvious damage to cartons should be noted on the delivery receipt before signing. Concealed loss or damage should be reported at once to the carrier and an inspection requested. All claims for shortage or damage must be made within ten (10) days after receipt of shipment. Claims for lost shipments must be made within thirty (30) days of receipt of invoice or other notification of shipment. Please save damaged or pilfered cartons until claim is settled. In some instances, photographic documentation may be required. Some items are time-sensitive; WPI assumes no extended warranty or any liability for use beyond the date specified on the container.

Do not return any goods to us without obtaining prior approval and instructions from our Returns Department. Goods returned (unauthorized) by collect freight may be refused. Goods accepted for restocking will be exchanged or credited to your WPI account. Goods returned which were ordered by customers in error are subject to a 25% restocking charge. Equipment which was built as a special order cannot be returned.

Repairs

Contact our Customer Service Department for assistance in the repair of apparatus. Do not return goods until instructions have been received. Returned items must be securely packed to prevent further damage in transit. The Customer is responsible for paying shipping expenses, including adequate insurance on all items returned for repairs. Identification of the item(s) by model number, name, as well as complete description of the difficulties experienced should be written on the repair purchase order and on a tag attached to the item.

* Electrodes, batteries and other consumable parts are warranted for 30 days only from the date on which the customer receives these items.
USA
International Trade Center, 175 Sarasota Center Blvd., Sarasota FL 34240-9258
Tel: 941-371-1003 • Fax: 941-377-5428 • E-mail: sales@wpiinc.com

UK
1 Hunting Gate, Hitchin, Hertfordshire SG4 0TJ
Tel: 44 (0)1462 424700 • Fax: 44 (0)1462 424701 • E-mail: wpiuk@wpi-europe.com

Germany
Zossener Str. 55, 10961 Berlin
Tel: 030-6188845 • Fax: 030-6188670 • E-mail: wpide@wpi-europe.com

China & Hong Kong
WPI Shanghai Trading Co., Ltd.
Rm 29a, No8 Dongfang Rd., Pudong District, Shanghai, 200120 PR China
Tel: +86 21 6888 5517 • E-mail: chinasales@china.wpiinc.com

Brazil
Av. Conselheiro Nébias, 756 sala 2611, Santos-CEP: 11045-002, São Paulo Brazil • Tel: (013) 406-29703 • E-mail: info@brazil.wpiinc.com

Internet
www.wpiinc.com • www.wpi-europe.com • www.wpiinc.cn