N₂O sensor calibration kit

High concentration version

Manual

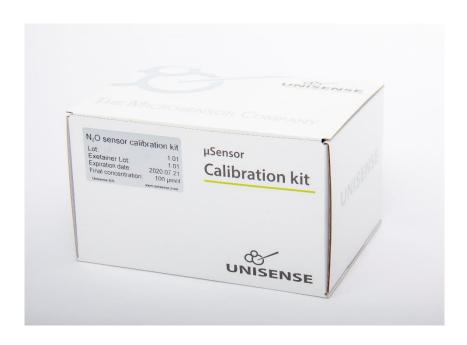






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1 Warranty and liability

1.1 Notice to Purchaser

This product is for research use only. Not for use in human diagnostic or therapeutic procedures.

1.2 Warning

Microsensors have very pointed tips and must be handled with care to avoid personal injury and only by trained personnel. Unisense A/S recommends users to attend instruction courses to ensure proper use of the products.

1.3 Warranty and Liability

The Nitrous Oxide Calibration Kit is guaranteed to give the concentration indicated on the package label until expiry as indicated on the package label. The warranty does not include replacement necessitated by accident, neglect, misuse, unauthorized repair, or modification of the product. In no event will Unisense A/S be liable for any direct, indirect, consequential or incidental damages, including lost profits, or for any claim by any third party, arising out of the use, the results of use, or the inability to use this product.

2 Support, ordering, and contact information

If you wish to order additional products or if you encounter any problems and need scientific or technical assistance, please do not hesitate to contact our sales and support team. We will respond to your inquiry within one working day.

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Further documentation and support are available at our website: www.unisense.com.

3 Content of the calibration kit

Item	Number
Ampoule with water equilibrated with a N₂O gas*	10
1 ml syringe	1
5 ml syringe	1
50 x 1.2 mm needle (red)	2



4 Principle of calibration

Unisense N_2O sensors respond linearly to N_2O concentrations within their linear range (see specifications for your sensor at https://www.unisense.com/N2O). Therefore, a two point calibration is sufficient. One calibration point is the signal for zero N_2O , which is water equilibrated with atmospheric air, and the other calibration point is the signal for one known N_2O concentration.

This calibration kit contains ampoules with water equilibrated with pure N_2O gas. This gives a N_2O concentration of approximately 27 mM. The actual concentration of N_2O in the ampoule is determined for each batch of calibration kits and is shown on the sticker on the calibration kit box.

The content of the ampoule should be diluted into an appropriate amount of water to obtain the desired calibration concentration. This calibration water should be of the same composition of salts, proteins etc. as the water where the measurements will be done. This will ensure that the calibration and measurements are performed in the same environment. This is important because the sensor reacts to the partial pressure of the gas, not the concentration as such, and in the software, this is recalculated into a concentration. The relationship between concentration and partial pressure depends on the salinity and temperature.

WARNING: N_2O sensors must never be exposed to higher concentrations of N_2O than they are designed for. This will result in bubble formation in the tip of the sensor which will then be destroyed.

5 Calibration procedure

For calibration of N₂O sensors a high and a low calibration point are needed.

5.1 Preparation of the calibration solution

The N_2O stock solution in the ampoule must be diluted to the desired concentration in water with the same composition and temperature as where the measurements will be done. Calculation of volume for dilution water and N_2O stock solution is described in 5.1.1.

- 1. Prepare an appropriate volume of water for the dilution, in a flask with a narrow opening (see note A below).
- 2. Make sure this water has the same composition and temperature as where the measurements will be done.
- 3. Open the ampoule with the N_2O containing water by breaking the top off (see note B below). Make sure that you don't shake or mix the content of the ampoule.
- 4. Use a syringe to aspirate the amount from the ampoule needed to create the desired concentration (see note C below).
- 5. Inject the N₂O containing water into the flask prepared in step 1 (see note D below).
- 6. Mix the water in the flask gently to obtain a homogenous distribution of N_2O and avoid mixing air into the water. This will cause N_2O to escape from the water.





Figure 1: Open the ampoule. Leave the tubing on for protection.

5.1.1 How to obtain the desired concentration

The concentration of N_2O in the final calibration solution depends on the volume of N_2O equilibrated water and dilution water used.

Final conc. (mM) = Vol N_2O Ampoule (ml)/Vol cal solution (ml) x Conc Ampoule (mM)

where Final conc. (mM) is the final concentration of N_2O after dilution, $Vol\ N_2O$ Ampoule (ml) is the volume of water transferred from the ampoule to the dilution water, $Vol\ cal\ solution\ (ml)$ is the final volume of the calibration solution, and $Conc\ Ampoule\ (mM)$ is the N_2O concentration in the ampoule as indicated on the calibration box label.

Example:

Vol. N_2O Ampoule = 3.0 ml Vol. cal. solution 300 ml Conc. Ampoule = 27 mM

Final conc. (mM) = $3.0 \text{ ml} / 300 \text{ ml} \times 27 \text{ mM} = 0.27 \text{ mM} = 270 \mu\text{M}$

5.1.2 Conversion of molar concentrations to mg/l

Concentration (mg N_2O/I) = Concentration (μ mol/I) × 44.013 (μ g/ μ mol) × 0.001 (mg/ μ g)

Where Concentration (μ mol/I) is the molar concentration calculated as shown in 5.1.1, 44.013 (μ g/ μ mol) is the molar mass of N₂O and 44.013 (μ g/ μ g) is a conversion factor from μ g to mg.

Example:

Molar concentration = 270 µmol/l

Concentration (mg N_2O/I) = 270 (µmol/I) × 44.013 (µg/µmol) × 0.001 (mg/µg) = 11.88 mg/I



5.2 Calibrating the N₂O sensor

IMPORTANT:

- The pre-activation and polarization of the N_2O sensor must have been completed before doing the calibration. See the N_2O sensor manual for details: https://www.unisense.com/manuals/
- The temperature of the two calibration solutions must be the same.
- Perform the calibration at the same temperature as the measurements if possible. The UniAmp series of amplifiers has a built in temperature compensation within ±3°C of the calibration temperature.
- It is recommended to obtain the low calibration point first to avoid carry over from the N_2O standard.

5.2.1 Obtaining the low calibration point

- 1. Put the sensor, mounted in the protection tube, into a beaker with N_2O free water at the same temperature as used for the high calibration point.
- 2. Allow the sensor to respond and stabilize and record the calibration value in SensorTrace (see the SensorTrace manual for details: https://www.unisense.com/manuals/)

5.2.2 Obtaining the high calibration point

- 1. Put the N_2O sensor, mounted in the protection tube, into the N_2O calibration solution prepared in section 5.1.
- 2. Allow the sensor to respond and stabilize and record the calibration value in SensorTrace (see the SensorTrace manual for details: https://www.unisense.com/manuals/)

NOTES:

- A. It is important to minimize the area of the calibration solution that is exposed to the atmosphere in order to minimize the loss of N_2O to the atmosphere. Therefore, it is recommended to use a narrow mouth bottle, e.g. a conical flask, Erlenmeyer flask.
- B. Opening the ampoule with N_2O containing water: Hold the bottom of the ampoule firmly while grabbing the tubing on the top. Break the top off the ampoule (Figure 1). IMPORTANT: Once an ampoule is opened it must be used immediately.
- C. Pull up N_2O containing water from the ampoule. Point the needle upwards and knock the syringe gently to get all air bubbles to the top. Press the piston to eject these bubbles. Once the syringe is free of bubbles, empty the syringe. Pull up water from the ampoule and adjust the volume to that needed.
 - IMPORTANT: Insert the needle fully in the ampoule and pull up water slowly to avoid bubble formation.
- D. Injection of the N_2O containing water is done with the needle inserted well into the water to avoid splashing and bubble formation. Thereby the injected N_2O containing water will not be lost.



6 Specifications

• Volume of calibration solution in one ampule: 5 ml

 $\begin{array}{ll} \bullet & \text{Content of the ampoule:} & \text{Slightly acidic water with N_2O} \\ \bullet & \text{Lifetime of the calibration kit:} & \text{See label on the calibration kit box} \\ \bullet & N_2O \text{ concentration in the ampoule}^* : & \text{See label on the calibration kit box} \\ \bullet & \text{Certificate of N_2O gas concentration:} & \text{See label on the calibration kit box} \\ \end{array}$

*The ampoule contains water equilibrated with a gas with a certified N_2O content. The concentration is calculated according to Weiss R.F. & Price B.A. 1980. Nitrous oxide solubility in water and seawater. Marine Chemistry 8:347-359. The concentration of N_2O in the ampoule is specified on the label on the calibration kit box. The certificate of the N_2O gas mixture used to produce the ampoule water can be requested from Unisense. See the ID of this certificate on the label on the calibration box.