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AQUAREAD
water monitoring instruments

CALIBRATION GUIDE CHLOROPHYLL

OBJECTIVE

Procedure for calibrating the Chlorophyll sensor.

PREPARATORY STEPS

- **Check**

Ensure that the chlorophyll sensor has been correctly assigned to the AUX socket number it's fitted to. Not applicable if using AP/AS-PRO. Ensure that the probes end cap is clean and fitted during calibration and when taking measurements in the field. Failure to use the end cap will lead to incorrect calibration and erroneous readings in the field.

- **Required Items**

- RHOD-CAL calibration stock solution (200g/L Rhodamine WT dye)
- Accurate weighing scales.
- Weighing boat.
- Volumetric flask, 1L.
- Accurate pipette, 0-10ml range.

- **Note on correlation**

There is no direct correlation between Rhodamine concentration and the concentration of chlorophyll. Rhodamine is used as a convenient dye for setting the sensitivity of the sensor. The subsequent display of chlorophyll in terms of mg/L is a generalisation based on research and experience. The only way to obtain a true value in terms of cells/mL is to correlate the values from the Sonde to quantitative data that has been obtained by laboratory analysis of grab samples, then to apply a Grab Sample Factor.

- **Preparation**

The probe, RHOD-CAL and deionised (DI) water should be left on the bench overnight so that their temperatures can equilibrate.

- **Equipment**

- Aquaprobe.
- Calibration instrument; Aquameter / Bluelink / Aquacal PC software
- RHOD-CAL solution.
- Accurate weighing scales
- Weighing boat
- Volumetric flask, 1L

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- Accurate pipette, 0-10ml range.

- **Serial Dilution**

  200g/L stock → 500µg/L is recommended to be done as a two step dilution procedure.

  Step 1: weigh out 0.5g of 200g/L stock solution in a weigh boat and add this to 1L of deionized water in a volumetric flask, use some of the water from the 1L flask to rinse the weigh boat so no stock Rhodamine remains on the boat. Put a lid on the 1L flask and invert 10 times.

  This step results in a 1 in 2000 dilution of the stock, at this point the 1L flask will contain a 100mg/L solution.

  Step 2: Transfer 5ml of the 100mg/L solution to a 1L volumetric flask and top up to 1L with deionized water. Put a lid on the 1L flask and invert 10 times. This step results in a 1 in 200 dilution of the solution from step 1. The concentration of this solution is 500µg/L. This solution can now be used as Point 2 calibration of the CPHYLL sensor.

  The value output by the probe directly after calibration should be approximately 118 µg/L at 20ºC (this value will vary with temperature).

  The dilute solution can be stored in a dark bottle in a refrigerator for up to five days. After that time it must be discarded.

OPERATING MODE

CALIBRATION

Calibration guidance will assume you are using an Aquameter, procedure will be similar when using Bluelink or AquaCal.

Procedure, 0 calibration:

1. Pour the DI water down the side of the calibration vessel so that air bubbles aren't introduced to the liquid.
2. Put the probe into the DI water. If the probe does not have a cleaning arm tap the probe on the bottom multiple times to dislodge bubbles, after you have put it in the liquid and right before starting the calibration. If your probe does have a wiper, run a clean cycle after you put it in the liquid and right before starting the calibration.
3. Leave the probe to sit in the water for two minutes to stabilise.
4. To start the calibration, press the MENU key then select Calibration>Full Cal>AUX ELETRODES>CPHYLL to enter the chlorophyll calibration screen.
5. Move the cursor downwards to point 1, which is the 0 calibration.
6. Press the OK button on the Aquameter to begin the calibration process.
7. A calibration report value will be displayed after the calibration is complete, it is useful to record these values for troubleshooting purposes.

Procedure, 500µg/L calibration:

1. Pour the 500µg/L rhodamine dilution down the side of the calibration vessel so that air bubbles aren't introduced to the liquid.
2. Put the probe into the rhodamine dilution. If the probe does not have a cleaning arm tap the probe on the bottom multiple times to dislodge bubbles, after you have put it in the liquid and right before starting the calibration. If your probe does have a wiper, run a clean cycle after you put it in the liquid and right before starting the calibration.
3. Leave the probe to sit in the rhodamine dilution for two minutes to stabilise.
4. To start the calibration, press the MENU key then select Calibration>Full Cal>AUX ELETRODES>CPHYLL to enter the chlorophyll calibration screen.
5. Move the cursor downwards to point 2, which is the 500µg/L calibration.
6. Press the OK button on the Aquameter to begin the calibration process.
7. A calibration report value will be displayed after the calibration is complete, it is useful to record these values for troubleshooting purposes.

CONTROL

Check the calibration report value is within the acceptable range.
The acceptable range for the 0% calibration is >2000mV
The acceptable range for the 500µg/L calibration is >100mV over 0 point calibration.

Once calibration is complete press the escape button until you can see the live readings. Check that the Chlorophyll readings is displaying a value **close to 118µg/l** at 20 degrees C. This value will vary with temperature.

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Dated	Revision	Document evolutions	Author
02/07/25	0	Creation	CP
	1	Review and fine adjustment	GP

Notes

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