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APPLICATION NOTE NO. 74

Revised February 2005

Calculating Calibration Coefficients for the Turner Cyclops-7 Fluorometer

The Cyclops-7 provides a 0 to 5 volt output voltage proportional to fluorescence. The Cyclops-7 is factory-configured at Turner to detect one of the following fluorophores:

Fluorophore	Chlorophyll a	Rhodamine WT tracer dye	Phycocyanin	Phycoerythrin
Designation on instrument	С	R	P	Е

The user can customize the 0-5V Cyclops-7 range to correspond to the expected data range by changing the interface cable, thus improving data resolution. The ranges are:

Cable Gain	Chlorophyll a (µg/l)	Rhodamine WT tracer dye (ppb)	Phycocyanin or Phycoerythrin (cells/mL)
X100	0-5	0 - 10	0 - 20,000
X10	0 - 50	0 - 100	0 - 200,000
X1	0 - 500	0 - 1000	0 - 2,000,000

Example: You expect a range of $0-30~\mu g/l$ for chlorophyll *a* fluorescence. If you use an X1 cable, the maximum voltage will be $0.3~V~(=30~\mu g/l~/~5~V]$). This limits the resolution and multiplies the noise level of the instrument. Changing the range to $0-50~\mu g/l$ (using an X10 cable) provides the best results.

When interfacing a Cyclops-7 to a Sea-Bird CTD, Sea-Bird defines the concentration as: concentration = (scale factor * voltage) + offset

The factory range can be used to calculate the factory default scale factor and offset (see page 2 for field calibration): scale factor = Range / 5 V

offset = - scale factor * measured voltage at 0 concentration (i.e., blank voltage)

To determine the blank voltage, place the fluorometer, in its field configuration (with or without a flow-through cap, as applicable) in de-ionized water and take a voltage reading.

Setting Up Configuration (.con) File in SBE Data Processing or SEASAVE

Change the range by changing the interface cable, as needed, before you set up the .con file in Sea-Bird software.

- 1. Use the Configure menu to create / modify the .con file. See the software Help files for details.
- 2. Select the Turner SCUFA **fluorometer** (Note: You must select the SCUFA because the Cyclops is not available as a choice in the current software version. However, the calculation for the SCUFA is of the same form as for the Cyclops, so the software will calculate fluorescence correctly for the Cyclops.).

The software prompts for scale factor, offset, and units, and calculates fluorescence as:

- Chlorophyll *a* Equations shown are for units of μg/l; other units available chlorophyll *a* (μg/l) = (scale factor * voltage) + offset where Scale Factor is in μg/l-volt and Offset is in μg/l
- **Rhodamine** Equations shown are for units of ppb; other units available Rhodamine (ppb) = (scale factor * voltage) + offset *where* Scale Factor is in ppb/volt and Offset is in ppb

Phycocyanin or Phycoerythrin – Equations shown are for units of cells/ml.
 Phycocyanin or Phycoerythrin = (scale factor * voltage) + offset
 where Scale Factor is in cells/mL-volt and Offset is in cells/mL

Note concerning Phycocyanin or Phycoerythrin units: The current software version does not allow the user to select cells/mL as output units for the Turner SCUFA. Calculate and input the scale factor in cells/mL-volt and the offset in cells/mL. Select any units in the software, and be aware when you view and process the data that the units are actually cells/mL.

Note: The software dialog box also shows grayed out fields for MX, MY, and B; these calibration parameters are applicable only when a Turner OBS has been selected for another voltage channel (intended for use with a Turner SCUFA with an optional turbidity channel).

Example of Chlorophyll a Concentration Calculation:

If fluorometer scale factor = $10.5 \mu g/l$ -volts, offset = $-0.03 \mu g/l$, and measured voltage from fluorometer = $3.65 \nu l$ volts, Calculated concentration ($\mu g/l$) = (scale factor * νl voltage) + offset = (10.5 * 3.65) - $0.03 = 38.29 \mu g/l$

Note on Field Calibration

While the nominal scale factor and offset based on factory range and your measured blank voltage can be used to obtain approximate values, **field calibration for chlorophyll** *a*, **Phycocyanin, and Phycoerythrin is highly recommended**. The relationship between fluorescence and these parameters is highly variable, and is not easy to determine in the laboratory. Species distribution, ambient light level, and health of the stock are just some of the factors that affect the relationship.

To accurately measure concentrations, perform calibrations on seawater samples with concentrations of plankton populations that are similar to what is expected in situ. Determine concentrations independently, and use those concentrations, as well as readings from the fluorometer (with or without a flow through cap, as applicable), to determine the correct Scale Factor. It is only through the use of these calibrations that a meaningful and accurate measure of these parameters can be obtained. The scale factor is correct as long as the condition of the plankton population does not change; the condition does change with season and geographic location.

To accurately measure the offset, perform a calibration on a blank (de-ionized water) sample, with the fluorometer in its field configuration (with or without a flow-through cap, as applicable).

See Turner's Cyclops-7 manual for calibration details.