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APPLICATION NOTE NO. 61

Calculating Calibration Coefficients for the Chelsea Minitracka Fluorometer

The Chelsea Minitracka fluorometer supplied with your Sea-Bird CTD measures Chlorophyll-a concentrations in the range of 0.03 to 100 μ g/l with a linear output of 0 to +4 volts. If the Chelsea Minitracka is selected when setting up the configuration (.con) file, Sea-Bird data processing software (DOS-SEASOFT or SBE Data Processing for Windows) prompts for $V_{acetone-100}$, and Offset and calculates Chlorophyll-a concentration as:

concentration (
$$\mu g/I$$
) = (100 * [V - V_{acetone}] / [V_{acetone-100} - V_{acetone}]) + Offset

where:

V (volts) = in situ output of the Chelsea Minitracka

 $V_{acetone}$ (volts) = measured output for pure acetone with 0 μ g/l chlorophyll-a; also called $V_{acetone-blank}$

 $V_{\text{acetone-100}}$ (volts) = measured output with 100 μ g/l chlorophyll-a dissolved in 1 liter of acetone

Offset $(\mu g/I)$ = term used to adjust to fit field data; nominally 0.0

The fluorometer comes with a calibration sheet that lists values for $V_{acetone}$ and $V_{acetone-100}$. (see Note at bottom of page)

Example of Concentration Calculation:

 $V_{acetone-100} = 3.102$ volts and $V_{acetone} = 0.076$ volts (from calibration sheet)

Offset = 0

Measured voltage from fluorometer = V = 2.53 volts

Calculated concentration ($\mu g/I$) = (100 * [V - V_{acetone}] / [V_{acetone-100} - V_{acetone}]) + Offset = (100 * [2.53 - 0.076]) / [3.102 - 0.076]) + 0 = 81.10 $\mu g/I$

The relationship between fluorescence and chlorophyll-*a* 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 chlorophyll-*a* concentration with a fluorometer, perform calibrations on seawater samples with concentrations of plankton populations that are similar to what is expected in situ. Determine chlorophyll-*a* concentrations independently, and use those concentrations, as well as readings from the fluorometer, to determine the correct calibration coefficients. It is only through the use of these calibrations that a meaningful and accurate measure of chlorophyll-*a* can be obtained. The calibration coefficients are only correct as long as the condition of the plankton population does not change; the condition does change with season and geographic location.

Shown below is an example of how to adjust $V_{acetone-100}$ in Sea-Bird's data processing software to reflect calibration data.

Example of Calculation of $V_{acetone-100}$ from field calibration for Sea-Bird's data processing software: Seawater sample analysis indicates chlorophyll-a concentration is 90 µg/l when fluorometer reads 3.2 volts. Calculated concentration (µg/l) = $(100 * [V - V_{acetone}] / [V_{acetone-100} - V_{acetone}] + Offset$ $90 = (100 * [3.2 - 0.076] / [V_{acetone-100} - 0.076]) + 0$

Solving for V_{acetone-100}:

 $V_{\text{acetone-100}} = (100 / 90) (3.2 - 0.076) + 0.076 = 3.547 \text{ volts}$ (input in the data processing software)

Factors such as fouling, scratches, or lamp degradation reduce the sensitivity of the sensor. Enter an *Offset* to adjust the calibration to fit the field data.

NOTE: For chlorophyll in acetone calibrations, always use the calibration cell, **as direct contact with acetone will permanently damage the fluorometer**.