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APPLICATION NOTE NO. 63 **Revised February 2010**

Calculating Calibration Coefficients for the Turner SCUFA (I, II, or III) Fluorometer/OBS

The SCUFA measures fluorescence and optional turbidity. The fluorescence channel is configured to detect either **chlorophyll *a* (SCUFA II)** or **Rhodamine WT tracer dye (SCUFA III)**.

Notes:
1. SCUFA I measures only chlorophyll a. Discussion of chlorophyll a calibration coefficients for SCUFA II is applicable to users who have a SCUFA I.
2. Turner no longer sells the SCUFA.

As listed in the SCUFA manual, all units are calibrated with primary standards at the Turner factory, with the 0 - 5V range equal to:

- 0 - 80 µg/l of chlorophyll *a* (SCUFA II)
- 0 – 200 ppb of Rhodamine (SCUFA III)
- 0 – 200 NTU (turbidity, SCUFA II or SCUFA III)

These can be used to calculate the factory default scale factor:

scale factor = [value at 5 V - value at 0 V] / 5 V

The user can customize the 0-5V SCUFA range to correspond to the expected data range with Turner's SCUFAsoft software, thus improving data resolution. **Change the range in SCUFAsoft, as needed, before you set up the configuration (.con or .xmlcon) file in the Sea-Bird software**.

Example: You expect a range of 0 – 10 µg/l for chlorophyll *a* fluorescence. If you do not change the default range, the maximum voltage will be 0.62 V (= 10 µg/l / [80 µg/l / 5 V]). This limits the resolution and multiplies the noise level of the instrument. Changing the range using SCUFAsoft to 0 - 10 µg/l provides the best results.

Note on Chlorophyll *a* Calibration

While the nominal Scale Factors based on factory default settings, and 0 Offset, can be used to obtain approximate values, **field calibration for chlorophyll *a* is highly recommended**. 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, 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 Scale Factor. It is only through the use of these calibrations that a meaningful and accurate measure of chlorophyll *a* 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.**

See Turner's SCUFA manual, Sections 2 through 4, for calibration details.

Setting Up Configuration (.con or .xmlcon) File in Seasave V7 or SBE Data Processing

1. In our SEASOFT V2 suite of programs, edit the CTD configuration (.con or .xmlcon) file using the Configure Inputs menu in Seasave V7 (real-time data acquisition software) or the Configure menu in SBE Data Processing (data processing software). See the software Help files for details.

2. (if optional turbidity included in SCUFA) Select the Turner SCUFA **OBS**. The software prompts for Scale Factor and offset and calculates turbidity as:
   turbidity (NTU) = (Scale Factor * Voltage) + Offset
   where
   Scale Factor (NTU/volt) = (turbidity value at 5 volts - turbidity value at 0 volts) / 5 volts
   (Turner calls the Scale Factor the *calibration coefficient*)
   Offset (NTU) = OBS value at 0 volts

3. Select the Turner SCUFA **fluorometer**. The software prompts for Scale Factor, offset, fluorescence units, mx, my, and b (prompts for mx, my, and b only if SCUFA OBS was already entered in configuration file) and calculates fluorescence as:

   Chlorophyll *a* (SCUFA II) – Equations shown are for units of µg/l; other units available
   chlorophyll *a* (µg/l) = (Scale Factor * Voltage) + Offset
   corrected chlorophyll *a* (µg/l) = (mx * chlorophyll *a*) + (my * NTU) + b
   where
   Scale Factor (µg/l-volt) = (chlorophyll *a* value at 5 volts - chlorophyll *a* value at 0 volts) / 5 volts
   (Turner calls the Scale Factor the *calibration coefficient*)
   Offset (µg/l) = chlorophyll *a* value at 0 volts
   mx, my, and b = correction factors for correcting chlorophyll *a* data for turbidity effects

   Rhodamine (SCUFA III) - Equations shown are for units of ppb; other units available
   Rhodamine (ppb) = (Scale Factor * Voltage) + Offset
   corrected Rhodamine (ppb) = (mx * Rhodamine) + (my * NTU) + b
   where
   Scale Factor (ppb / volt) = (Rhodamine value at 5 volts - Rhodamine value at 0 volts) / 5 volts
   (Turner calls the Scale Factor the *calibration coefficient*)
   Offset (ppb) = Rhodamine value at 0 volts
   mx, my, and b = correction factors for correcting Rhodamine data for turbidity effects

   Example of Chlorophyll a Concentration Calculation:
   If fluorometer Scale Factor = 14.5 µg/l-volts and Measured voltage from fluorometer = 4.65 volts,
   Calculated concentration (µg/l) = (Scale Factor * Voltage) + Offset = (14.5 * 4.65) + 0 = 67.4 µg/l