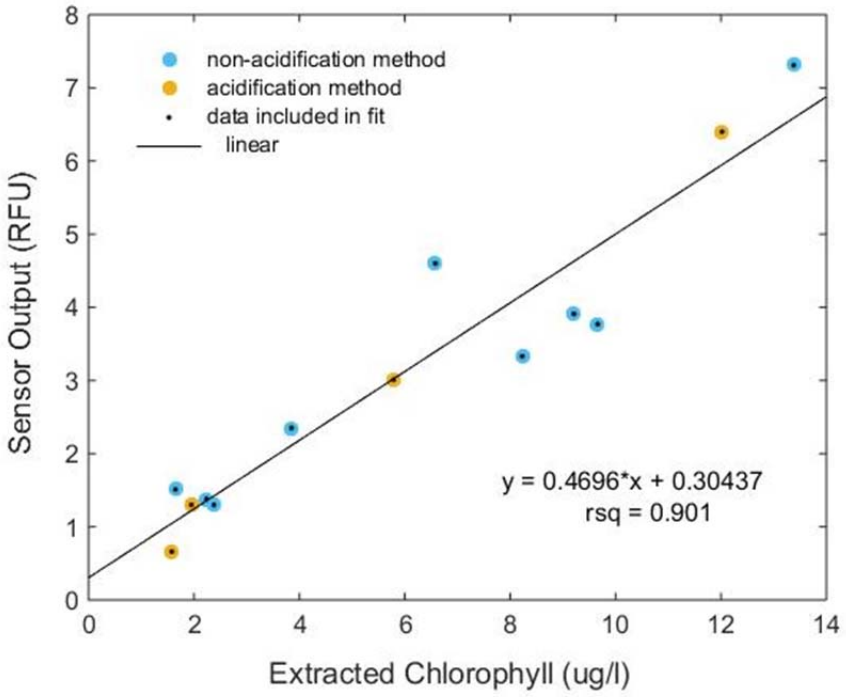
SATURN-04 Chlorophyll Sensor Field Calibration
(March 2010 – December 2012)

Update Notice, August 2016: *The calibration has been updated from the previously released version of data. See the end of this document for a description of changes made and the resulting changes in final data values.*



Calibration Details:

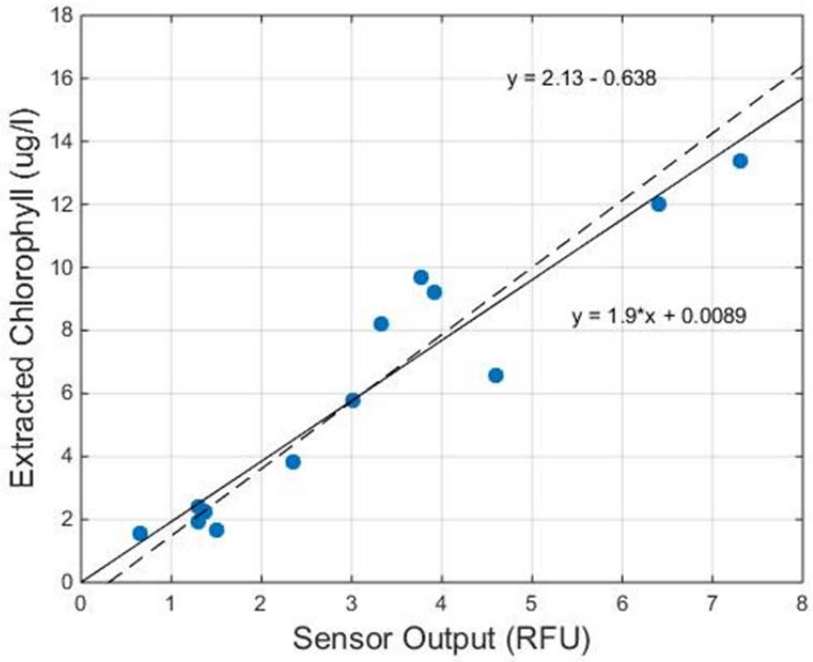
This sensor was calibrated against extracted chlorophyll samples taken from periodic field samples collected between March 2010 and September 2011. Field samples prior to January 2011 were analyzed for extracted chlorophyll using the acidification method for chlorophyll determination, while samples after this date were analyzed using the Welschmeyer non-acidification method of chlorophyll determination. The corresponding sensor reading was determined by interpolating the *in situ* sensor data to the grab sample time point. The resulting calibration equation is: Chlorophyll (µg/L) = 2.130 * rfu - 0.6481. The calibration quality has been flagged as less than good (QL2) because it lacks samples from periods of high biomass (with chlorophyll levels exceeding 14 ug/L) and requires a greater number of samples for a robust calibration (this calibration is based on 13 samples from 10 sampling times). A conservative approach is to apply this calibration only during the time the samples were collected (March 2010 – Sept. 2011), however, this calibration has been extended through the end of 2012 due to a lack of sampling during this time. Subsequent sampling in 2014 and 2015

shows that there was very little shift in the sensor response lending support to the extension of this calibration beyond the sampling period. Shifts in baseline have been addressed and corrected for separately and do not preclude the extension of the calibration through the end of 2012.

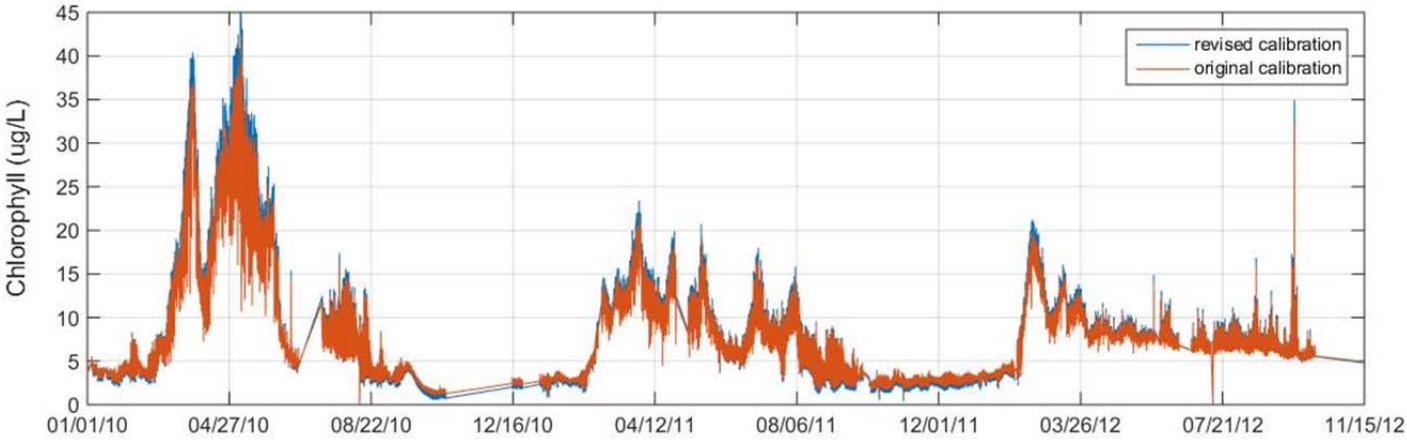
Revised Calibration vs. Previous Calibration:

The originally released calibration has been revised. The regression was corrected so that extracted chlorophyll concentration (µg/L) was defined as the independent variable and sensor output (RFU) as the dependent variable. Revising the regression to the one shown at the top of this document (with sensor output as the dependent variable) changed the resulting equation from: Chl(µg/L) = 1.92 * rfu + 0.009 to Chl(µg/L) = 2.13*rfu - 0.648.

The plot below shows the original calibration and fit as the black solid line. The corrected regression (the one shown at the top of this document, fitting sensor output as the dependent variable) is shown as the dashed line.



After revising the calibration, greater than 99% of the changes in final chlorophyll values are within ± 1 µg/L and the maximum change in values is 3.99 µg/L. The figure below shows the data calibrated with both the previous and revised calibrations:



Field Sample Data included in Calibration:

Method	Sample Date	Depth	Avg Chl	St.dev.	n	Sensor(rfu)
Acid	3/3/10 11:30	0.3	5.79	0.46	2	3.01
Acid	4/7/10 8:20	0.3	12.02	0.56	2	6.40
Acid	10/22/10 11:08	0.3	1.58	0.04	2	0.66
NA	1/10/11 9:47	8.6	2.24	0.21	3	1.38
Acid	1/10/11 9:58	0.3	1.95	0.34	3	1.30
NA	1/10/11 9:58	0.3	2.38	0.32	3	1.30
NA	4/28/11 14:01	8.6	13.38	2.96	3	7.31
NA	5/31/11 15:14	8.6	9.21	0.26	3	3.91
NA	5/31/11 15:12	0.3	9.66	0.59	3	3.77
NA	7/26/11 14:06	8.6	8.23	0.55	3	3.33
NA	7/26/11 14:08	0.3	6.58	0.16	3	4.60
NA	9/7/11 14:52	8.6	1.65	0.00	1	1.51
NA	9/7/11 14:42	0.3	3.85	0.00	1	2.35

Sample Collection: Whole water samples were collected from the pump at the station. The samples were stored in amber HDPE bottles and shipped overnight on blue ice to the Peterson/Needoba lab at OGI in Beaverton. The samples were then stored refrigerated until processing within 24 hours of receipt.

Sample Processing Details:

1. 100mL of sample is filtered onto a Whatman GF/F under low vacuum. The filter is folded, placed into a cryovial and stored at -80°C.
2. When ready to process the samples, the filters are placed in glass test tubes and 5mL of 90% acetone is added (under dimmed lights). Three acetone blank test tub are also prepared.

3. Samples are allowed to extract for 24 hours in the dark at -20°C.
4. After extracting, samples are allowed to warm to room temperature in the dark.
5. Measure the extracted sample on benchtop fluorometer:
    a. Non-Acidification Method (used after January 2011)
        - Vortex test tube.
        - Filter removed with forceps and discarded
        - Wipe test tube with kimwipe.
        - Measure Raw Fluorescence (F0).
        - Measure the 3 acetone blanks
    b. Acidification Method (used prior to January 2011)
        - Vortex test tube.
        - Filter removed with forceps and discarded
        - Wipe test tube with kimwipe.
        - Measure Raw Fluorescence (F0).
        - Add 3 drops 10% HCl.
        - Invert test tube.
        - Measure Raw Fluorescence (Fa = acidified)
        - Measure 3 Blanks without adding acid.