

## **Fluorometer Chlorophyll a Analysis**

**Manatee County Natural Resources Department; Environmental Protection Division**  
**Standard Operating Procedures; Standardization of Fluorometer for In-Situ Observations**

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## **I. Purpose**

Fluorometers do not measure the analytes they are assessing directly, rather they measure “prescribed situations of fluorescence” and use that data to infer the concentration of the analyte. Because these analytes are not specifically defined enough, calibration can be difficult. As it relates to this project, the level of chlorophyll a in the water gives a value that is representative of the amount of primary production occurring in the water due to algae because it is the pigment used in photosynthesis. Whereas lab analysis dissolves cells away and measures the exact amount of the photosynthetic pigment, these in-situ field measurements are inferring the concentration of chlorophyll a from the irradiant response to UV radiation emitted by the fluorometer. This light response is the result of the saturation of the light harvesting pigment protein complex in the phytoplankton. Because all different types of phytoplankton are present in eutrophication, and each algae have their own specific pigment protein complex, light responses are slightly different. To combat this issue, Eureka recommends a method for standardizing the fluorometer, a process that ensures that the fluorometer always reads the same number in the same solution. This standardization process involves using a commercial standard, in this case a Post-It Note, to find the saturation point and calibrate it to the maximum light response for chlorophyll. In doing this, the fluorometer readings of chlorophyll a concentrations are on a standardized scale of light response, this does not necessarily provide accurate data, but does provide precise or consistent data. This precise data can then be compared against laboratory analysis to find a correction factor that can adjust for the inaccuracy of the readings to convert them into accurate measurements.

## **II. Procedure**

1. Remove storage/calibration cup
2. Clean and dry the fluorometer sensor
3. Point Manta2 multiprobe into the air away from direct light
4. Place a pink Post-It Note within view of the fluorometer
5. Slowly move the Post-It Note closer to the fluorometer until the chlorophyll reading cannot go any higher, this is the saturation point
6. At this distance, press calibrate on the application and calibrate the saturation point to 500  $\mu\text{g/L}$
7. Point the Manta2 multiprobe into the air away from direct light and calibrate to zero