Low cost in situ chlorophyll a fluorometer for marine and fresh waters

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Abstract

Microscopic phytoplankton is an essential part of the global oxygen production, climate control as well as being the cornerstone of the ocean food web, and as such is an essential parameter to monitor. Due to its content in chlorophyll a, the reference technique to measure phytoplankton concentration in water compartments is based on chlorophyll a fluorometry. The measurement principle is based on a high energy blue excitation light (between 430 nm to 470 nm typ.), which triggers a low energy red fluorescence signal (between 650nm to 700nm typ.). Most of the in-situ commercial probes are based on this principle, however these systems are considered as too expensive by end-users, with average pricing in the 3 000 range. By combining 3D-printing, low-cost ink based optical filtering, and off-the-shelf electronic components, we evaluate the feasibility of a complete system that can be built for less than 100 [1], including low-power wireless broadcasting capabilities using the LoRa protocol.

Our fluorimeter is based around a common flat face design. Blue light excitation is achieved with three Luxeon REBEL LXML-PR01-500 Royal Blue LEDs. The photodetector is an AMS TSL257, which embeds a silicon photodiode and a high-gain transimpedance amplifier on the same chip, thus keeping noise levels reasonable especially in low-light conditions. An Arduino compatible, Adafruit Feather M0 Lora microcontroller is completed by a micro-SD datalogger and a Real Time Clock shield, and a LED driver has been designed and built atop, the whole system being powered by a 500 mAh Lipo battery. The internal 12-bit Analog to Digital Converter is used to sample the voltage output from the TSL257. A simple point-to-point LoRa connection is used to remotely control as well as retrieving the data from the fluorimeter.

Two different light filters are used in the system: a blue filter which cuts unwanted excitation light above 550 nm, and a red filter which blocks unwanted light below 575 nm. To facilitate the filter integration, we're using ink dyed poly(dimethylsiloxane) (PDMS) filters [2]. These are obtained by mixing the inks (Pelikan red: 14K351221, Pelikan blue: 14K351213) to the PDMS oligomer (Sylgard 184) until a visually homogeneous dispersion is obtained. The curing agent is added to the mix, prior to degassing. The ink dyed PDMS filters are then poured in the 3D printed housing and cured in place at 50°C overnight. The 3D printed housing is made in PRECISA DL260 material with a DWS 029J+ high-resolution SLA (stereolithography) printer, and has been designed to allow precise alignment of the light sources and detectors.

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Chlorophyll a extracts from spinach leaves were characterized by UV-VIS absorption spectroscopy to determine their concentration. For the measurements, the fluorimeter is immersed in an opaque tank containing 400mL of deionized water. Chlorophyll a is then added in the tank by pipetting 500μ L from the stock solution. We investigate the effect of LEDs intensity on the fluorescence signal in order to find the best compromise. While a linear relationship was observed from 5mA to 20mA, above 50mA the fluorescence signal was quickly decreasing, a phenomenon which we attributed to the non-photochemical quenching (NPQ), a well-known protection mechanism in which PSII receptors converts excess energy into heat, to protect the photosynthetic organism [3]. Measurements from 0 to $37\mu g/L$ were performed, showing a good linearity for each excitation current without any sign of saturation. The sensitivity was calculated from the slope of the linear regression fit, and was found to be 1.33, 3.34 and 7.46 counts/(μ g/L) for respectively 5, 10 and 20 mA excitation current. For marine waters, the typical range for chlorophyll a concentration is comprised between 1 μ g/L to 50 μ g/L, so the operational range of our fluorimeter is on par with the requirements. Future works will focus on the integration of an external, 16-bit precision ADC in order to increase the sensitivity, as our objective is to obtain a 0.1 μ g/L precision on the measurement.

Keywords: Marine waters, phytoplankton, 3D printing, LoRa