



Application Note

Cyanobacteria Quantification Using Handheld Fluorometer

Background:

Cyanobacteria, also known as **blue-green algae**, is a phylum of bacteria that obtain their energy through photosynthesis. Cyanobacteria can be found in almost every terrestrial and aquatic habitat—oceans, fresh water, damp soil, temporarily moistened rocks in deserts, bare rock and soil, and even Antarctic rocks. They are found in almost every endolithic ecosystem. Aquatic cyanobacteria are known for their extensive and highly visible blooms that can form in both freshwater and marine environments. The blooms can have the appearance of blue-green paint or scum. These blooms can be toxic, and frequently lead to the closure of recreational waters when spotted. For these reasons alone it should be clear that there is an interest in measuring concentrations of Cyanobacteria.

Cyanobacteria contain accessory pigments from the phycobiliprotein family. The primary phycobilin pigments are **phycocyanin (PC)** and **phycoerythrin (PE)** that happen to have strong fluorescent signatures that do not interfere with the fluorescence of the chlorophylls. This allows for the *in vivo* detection of cyanobacteria without interference from other groups of algae. PC is the predominant phycobilin in freshwater environments, while PE is the predominant pigment in marine environments. From this perspective, fluorometric technique is the most versatile, sensitive, and easy way to measure the concentrations of Cyanobacteria in water. Application examples include, but are not limited to, the monitoring of Cyanobacteria in natural marine and freshwater environments, reservoirs, water and sewage treatment plants, and aquacultural systems.

Fluorescence Detection:

Phycoerythrin: In marine species such as *Synechococcus*, phycoerythrin is the dominant accessory pigment. Narrow band interference filters are used for excitation and emission wavelengths of 530 nm and 590 nm to minimize background interferences from the matrix and other pigments.

Phycocyanin: In contrast, fresh water species such as *Anabaena*, *Microcystis*, and *Spirulina*, are rich in phycocyanin. For detection of this pigment, we use narrow band interference filters with excitation and emission wavelengths of 600 nm and 650 nm, respectively. Generally, the fluorescence signal is directly proportional to the concentration of the material in question.

In vivo analysis is the fluorescent detection of phycobilin in living algal and Cyanobacterial cells in water. In this technique, the excitation light from the fluorometer passes through the untreated sample water and excites the phycobilin within the living cells of the algae present. *In vivo* fluorescence data supplies information on the relative distribution of phycobilin concentrations and usually correlate well with extracted phycobilin samples, as well as the Cyanobacteria concentration. *In vivo* detection has several very useful applications. An example is the monitoring of general trends in Cyanobacteria concentrations in real time. It is very easy to obtain large amounts of data using *in vivo* instrumentation and is an excellent means of following trends and estimating Cyanobacteria concentration.

In vitro analysis (extracted analysis), on the other hand, entails the concentration of phycobilin containing cells onto a filter followed by the extraction of the phycobilin from the cells. This method provides the best quantitation accuracy of phycobilin concentration, but requires laboratory setup to conduct the analytical procedures.

hVI handheld fluorometers have been proven to detect low level of cyanobacteria in water and in extracted samples. Due to its high portability and low cost, it can be used anywhere in the field to conduct environmental study of natural water resources.

