The Use of Tryptophan-like Fluorescence as an Indicator of Organic Pollution

Fluorescence has long been used to measure a range of water quality parameters, from chlorophyll and algae to hydrocarbons and optical brightening agents. Recent advances in LED technology have led to the development of portable, submersible fluorimeters that can look further than ever before into the ultra-violet region, with some very interesting results...

Background

Fluorimeters have been used for many years in the field of water quality monitoring and are the established and trusted technique for reliably measuring dissolved organic matter (DOM), chlorophyll and algal biomass. Fluorimeters work by emitting light at one wavelength and detecting light emitted by the target molecule at another wavelength. Only certain substances exhibit this property and at very specific pairs of wavelengths – this means that fluorescence can be a very selective and sensitive optical technique.

DOM in freshwaters consists predominately of decaying plant matter (humic or fulvic substances). In many cases plumes of DOM in a water body will also be accompanied by an active microbial community. It is this microbial community that consumes oxygen – leading to high levels of biological oxygen demand (BOD) and the subsequent crashes in oxygen levels that are detrimental to aquatic ecosystems. Proteins found in the cell walls of these micro-organisms have been shown to fluoresce in the same region as the amino-acid, tryptophan. Thus, ‘tryptophan-like’ fluorescence can be used as a measure of the microbial health of a water body and therefore as an indicator of BOD.

Common sources of polluting DOM such as sludge liquor, cattle and pig slurries and human sewerage all fluoresce when excited at the same short ultra violet wavelengths (~280nm). This means that fluorimeters ‘tuned’ to this wavelength could be a uniquely useful tool for a wide range of monitoring applications in both rural and urban catchments.

What’s new?

The original research that identified the different constituents of the fluorescence EEM was carried out in the laboratory using a sophisticated bench-top scanning fluorimeter. Once the specific excitation and emission wavelengths of the tryptophan-like peak (T) were identified then lower cost portable instruments could be developed that focused solely on that excitation/emission pair (i.e. 280nm/340nm). Due to the short wavelengths required to provide the correct excitation, high power xenon flash lamps were used as the source in early portable tryptophan-like fluorimeters. This meant that not only were they relatively large and expensive, but submersion was not possible – instead the sample was introduced via a quartz cuvette. The breakthrough recently occurred when light emitting diodes (LED) were developed that could attain the short wavelengths required. This now means that compact, submersible fluorimeters tuned specifically for tryptophan-like fluorescence can be produced in the same format as established sensors for chlorophyll, algae or optical brightening agents (OBA).

A Practical Solution

For the first time, robust and submersible tryptophan-like fluorimeters can now be deployed as an integral part of the flexible Manta 2 multi-sonde platform from Eureka. This opens up a whole new range of applications for exploiting the power of this novel technique. As the Manta 2 sonde can incorporate up to 12 sensors it will enable...
Potential Applications

The facility to embed multiple water quality sensors – including tryptophan-like fluorimeters – into a single platform opens up a range of new applications as well as offering improved tools for some commonly monitored situations. For example, tryptophan-like fluorescence could be a much more direct measurement of discharging combined sewage overflows (CSO) than using a combination of dissolved oxygen, pH, oxidation reduction potential (ORP), conductivity and depth.

Tryptophan-like fluorescence is also likely to identify inputs of polluting organic matter earlier and more decisively than a suite of sensors designed to measure the effects of DOM. Depending on the type of organic matter, it can sometimes take a while for oxygen levels to drop and for elevated ammonium, nitrate or phosphate levels to develop. This lag-time translates into increased uncertainty over the location and timing of the pollution source. By directly measuring the fluorescent proteins in the cell walls of the microbial population, tryptophan-like fluorescence can also give an indication of the potential severity of subsequent effects of the pollution.

Early trials with the tryptophan sensor show that there is potential for it to be used in more industrial applications. Industries and processes that deal with meat, fish and dairy products often have waste streams that are very high in proteins. This waste is usually discharged into the sewerage system where treatment costs are apportioned according to infrequent sampling and analysis for either BOD or chemical oxygen demand (COD). These tests can be costly in themselves and are unable to provide the high temporal resolution needed to accurately monitor complex processes. The use of tryptophan-like fluorescence sensors for real-time monitoring of waste streams could not only enable better estimation of inputs to treatment plants, but also enable real-time optimisation of active treatment systems, thus reducing costs and penalties associated with under or over-treatment. There is some work to be done in order to optimise anti-fouling for these harsher conditions but fluorimeters are commonly fitted with brushes or wipers for long term deployments in surface water environments.

Perhaps the most exciting aspect of the miniaturisation of tryptophan-like fluorimeters is the ability to combine multiple tryptophan-like fluorimeters in the same platform and gain real-time, continuous data from complimentary sensors. The possibilities are too numerous to mention but already researchers and catchment management professionals are starting to think about how tryptophan-like fluorescence relates to dissolved oxygen levels and whether concurrent chlorophyll data will enable greater understanding of primary production processes. Simultaneous measurement of tryptophan levels and optical brightening agents (OBA) are highly fluorescent and are commonly used in household cleaning products and are considered a ubiquitous tracer of human sewage) should enable the origin of polluting DOM to be established. For example – if a pollution event causes high levels

Read, Print, Share or Comment on this Article at: Envirotech-Online.com/Articles

All of our articles are online! To view and download them, visit: www.envirotech-online.com