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UV-Laser and LED Fluorescence Detection of Trace Organic Compounds in Drinking Water and Distilled Spirits

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1. Introduction

Current methods for the analysis of drinking water and many other liquids often call for the use of reagents and may require extensive sample preparation (American Public Health Association, 1989). For the case of water supplies and water treatment plants, this analysis is usually carried out once every few days or weeks (Killinger & Sivaprakasam, 2006). Most of the analysis is usually conducted using classical analytical chemical techniques, such as mass spectrometry, liquid chromatography, or fluorescence based or tagged reagents (Crompton, 2000). These analytical techniques are sensitive and provide accurate assessment of the chemistry related to the quality of the liquids. However, they often take considerable time and are usually not performed in real-time, especially for the case of a flowing process line. On the other hand, previous fluorescence spectroscopic measurements of ocean water by Coble showed that deep-UV excitation of naturally occurring organic compounds in water can yield significant and unique fluorescence signals in the near UV to visible wavelength range without the need to use additional reagents or sample preparation (Coble, 1996; Coble, 2007). As a result, we have been studying deep-UV laser-induced-fluorescence techniques for the detection of trace species in water and other liquids with the goal of using the natural fluorescence of trace species in the water or liquid samples and being able to provide readings within the time span of a few seconds.

Toward this goal, we have developed a reagentless deep-UV laser and UV-LED induced fluorescence (LIF) system to detect and continuously observe in real time trace levels of colored dissolved organic matter (CDOM) or Dissolved Organic Compounds (DOCs) in water and distilled spirits, such as drinking water, and related water/alcohol based liquids with a sensitivity exceeding that of commercial spectrofluorometers. Our system has been used to detect ppb trace levels of plasicizer Bisphenol-A (BPA) that have leached into drinking water, and has detected and monitored trace levels of DOCs within ocean currents (Killinger & Sivaprakasam, 2006; Sivaprakasam et al. 2003; Sivaprakasam & Killinger, 2003). Recently, our LIF system has been used to measure fluorescence of reverse osmosis processed water and different types of drinking water (Sharikova & Killinger, 2007; Sharikova & Killinger, 2010). These LED/LIF applications have now been extended to additional water related samples, including humic acid samples, tannic acid and chlorinated water samples, juices, coffee, and several wines and distilled spirits; these recent results are presented in this paper.

Our compact LIF system used either frequency tripled or forth harmonic diode pumped Nd:YAG lasers operating at 266 nm and 355 nm, or deep-UV LEDs (265 nm, 300 nm, 335 nm, and 355 nm) as UV excitations sources. The emitted fluorescence was measured over the range of 240–680 nm. Strong emission near 450 nm was observed for the DOCs in water, while emission bands near 340 nm were evident from distilled spirits and wine. It is important to note that one of the main advantages of using a deep-UV excitation wavelength, such as 266 nm, is that the emission fluorescence is separated in wavelength from the Raman emission of water (near 310 nm for 266 nm excitation), and thus yields greater sensitivity and wavelength selectivity than previous systems using lasers operating near 400 to 550 nm. In addition, as a point of reference, our laser based LIF system had a detection sensitivity for the fluorescence standard solution of quinine sulfate on the order of 0.1 ppb. The average laser power was approximately 30 times that of the LED, but differences in the signal intensity due to the difference in the laser and LED excitation intensity were consistent with theory.

Our studies show that deep-UV light emitting diodes (LEDs) are good alternative light sources for our LIF system, which would make the apparatus cheaper and more compact. It should be noted that the research presented in this paper is directed toward the development of new optical spectroscopic measurement techniques which have the potential to offer enhanced capabilities over conventional water monitoring and liquid analysis. However, while its sensitivity has been shown to be in the sub-parts per billion for standard fluorescing compounds used in fluorescence research, such as quinine sulfate, it needs to be further quantified and evaluated against conventional analytical chemistry instruments before it can be used as an on-line analytical instrument for water monitoring. Such comparisons are currently being conducted and will be reported later.

2. Experimental setup

Our Laser and LED induced fluorescence system is similar to that of a conventional spectrofluorometer, but has a sensitivity several orders of magnitude better. Commercial spectrofluorometers often use UV lamps and wavelength selecting spectrometers for the emission source, and single or double monochromators with Photo-multiplier Tubes or CCD detecting arrays for fluorescence detection (Albani, 2007). Often the signal processing is conducted using a chopped CW beam and lock-in amplifier signal detection. Our LIF system uses a high PRF (pulse-repetition-frequency) laser running at about 8,000 pulses/second as the excitation source (or a pulsed LED source running at about 330 pulses/second), and a high-speed boxcar integrator which detects and stores the fluorescence photon signal for each pulse. In addition, our system uses multiple excitation beams and double-pass collection optics to increase the fluorescence signal. Our past work has shown that this combination has enhanced the sensitivity of our laser-induced-fluorescence system by two to three orders of magnitude over conventional spectrofluorometers, depending upon the spectrometer and optical detector configuration used (Sivaprakasam & Killinger, 2003). Details of our current LIF system follow.

2.1 Description of the apparatus

Our fluorescence measurements were performed using a system shown in Fig. 1. The schematic diagram of the apparatus is shown in Fig. 2. The light source was one of the following: a microchip laser, 266 nm or 355 nm (JDS Uniphase, Models NU-10110-100 and NV-10110), or a LED operating at 265 nm, 300 nm, 335 nm or 355 nm (Sensor Electronic Technology, Inc., UVTOP® series). A silicon APD photodetector (New Focus, Model 1621)

was used to trigger data acquisition with the laser source; the LEDs were software-triggered. The laser beam passed through the optical quartz cell (Spectrocell Inc., Model RF-3010-F) several times, for which plane mirrors on the sides of the cell were used. The fluorescence signal was collected at a right angle to the excitation beam with a concave mirror (Optosigma, Model 035-0130) and a fused silica lens (Optosigma, Model 014-0490). It passed through one of the 19 different narrow (10nm) bandpass optical filters ranging from 265 nm to 685 nm before being focused onto a PMT (Hamamatsu, Model H6780-03). One of the absorption cut-off filters (CVI Laser, Models CG-WG-280-2.00-2 and CG-WG-295-2.00-2) was used to block Rayleigh and Raman scattering. All filters were mounted on a stack of motorized filter wheels (CVI Laser, Models AB-302 and AB-304). The PMT signal was acquired by a gated integrator and boxcar averager unit (Stanford Research System, Model SR-250). Data collection and filter wheel control was handled by LabVIEW software through a computer interface unit (Stanford Research System, Model SR-245) and serial bus.

2.2 Laser and LED characteristics

The Q-switched microchip lasers produced 0.4 ns, 0.3-0.4 μJ pulses at a repetition rate of 8 kHz. The beam size was on the order of 1 mm.

The LEDs were operating in a 10 μ s, 50 mA drive current regime with a 330 Hz repetition rate. The output light pulse energy was approximately 7 nJ (with the exception of 22 nJ for the 355 nm LED). It was noted that the LEDs also had an out-of-band emission in the visible region. As an example, this can be seen in Fig. 3 that shows the measured output power of the 265 nm LED (LED265) as a function of wavelength and of drive current. This is a log plot of the intensity and shows that the out-of-band LED emission had a peak value of about 1% compared to the peak emission at 265 nm.

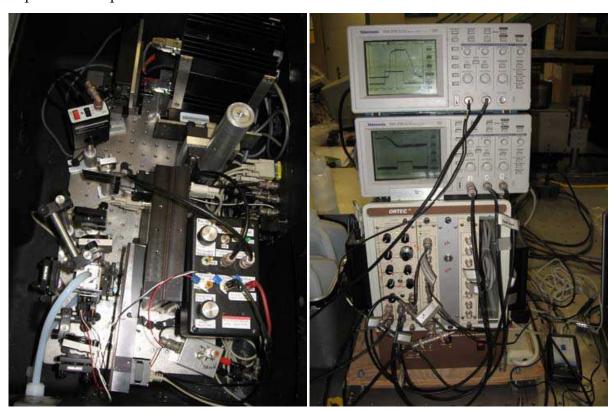


Fig. 1. Experimental system: optics box (left) and electronics box (right)

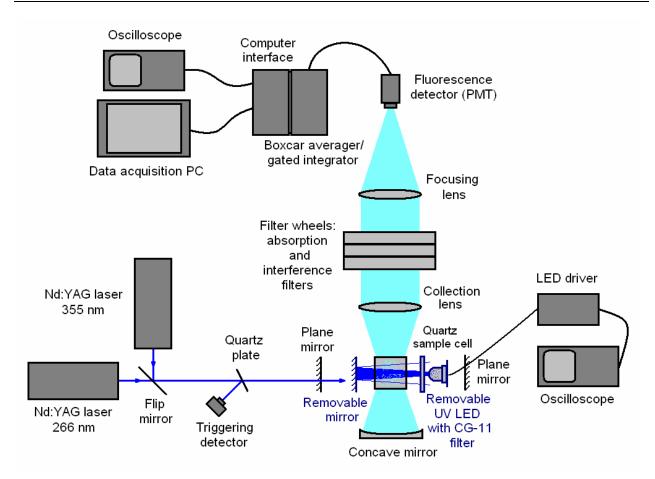


Fig. 2. Schematic diagram of the experimental apparatus

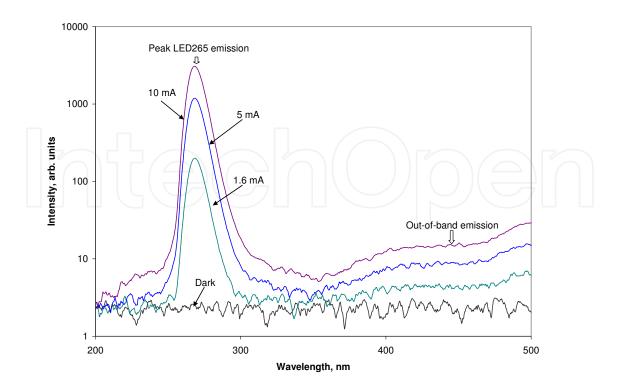


Fig. 3. Emission spectra of LED265 for different CW currents.

To block this out-of-band light, a VIS-blocking, UV-passing filter (CVI Model CG-UG-11) was used with our LED sources. Figure 4 shows the spectral output power of the LEDs with and without the filter using a linear scale for the intensity. The beam size within the sample cell was about 5 mm.

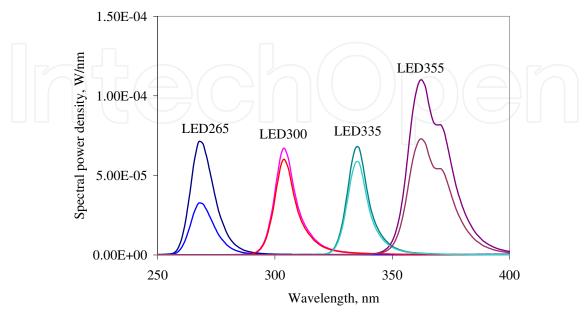


Fig. 4. LED spectral peak power density in 50 mA, 10 µs pulse regime (lower curves with CG-UG-11 filter)

3. Data and results

Our LIF system was used to measure a considerable variety of water related liquids including tap water, Reverse Osmosis (RO) treated ground water, and other water quality related substances. These results are shown in the following.

3.1 Experimental conditions and settings for liquid samples

The liquid samples were stored, when necessary, in the dark and cold. Water samples were not further processed. Wine samples were diluted to 10 mL per 1 L with distilled water. Humic substances (International Humic Substances Society, 1R101N, 1S103H, 1S104H) were prepared as 10 mg per 500 mL of distilled water.

A flow cell with a linear flow rate of 5 cm/s was used to minimize photobleaching. With each bandpass filter, 1000 measurements were taken, which lasted a few seconds per filter setting including filter switching time. Boxcar averaging setting was 300 samples. The sensitivity setting was adjusted for each sample to maximize the signal. The spectra were compensated for filter bandwidth and transmission, PMT quantum efficiency and gating integrator/boxcar averager sensitivity.

3.2 Reverse Osmosis processed ground water and drinking (tap) water

Ground water taken from a shallow well at USF was processed by a Reverse Osmosis unit at Prof. Carnahan's lab (Carnahan, 2006). The fluorescence spectra of ground water before and after RO treatment using 266 nm laser excitation is shown in Fig. 5. The broad peak observed around 470 nm in the untreated water is typical of the organic compounds usually

present in such samples (Coble, 2007). After the RO treatment, the fluorescence signal decreased significantly, especially on the short-wavelength side. The signal from distilled water is shown for comparison.

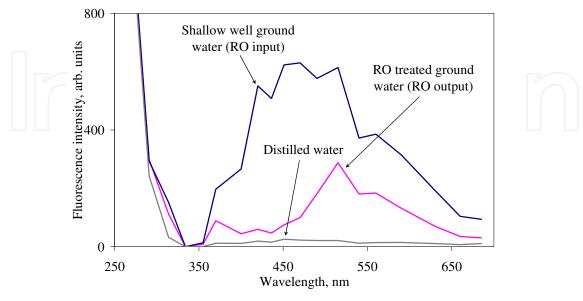


Fig. 5. Fluorescence of ground water before and after reverse osmosis treatment, 266 nm laser excitation

Tap water in our lab was continuously monitored for a period of a week (see Fig. 6). As can be seen, the flowing tap water, Fig. 6(a), had a greater range of variation than a sample recirculated through the system, Fig. 6(b). Certain repetitiveness of the running water signal might be indicative of the water usage patterns at our university. The initial growth in the recirculated signal was due to plastic leaching from the soft tubing used in the pump.

We have also tested tap water collected from different locations in the US (Fig. 7). All samples were taken directly from residential tap water except for the Tampa location, where an on-line water filter in a drinking fountain was present. For all samples, settings were the same during data acquisition. As can be seen, the fluorescence spectra obtained with 266 nm laser excitation, (a), and 265 nm LED, (b), were different only in overall intensity. Comparing the spectra from different locations, one can see that all possessed two large peaks centered around 420 and 460 nm, as well as smaller peaks on the sides. However, both the absolute and relative intensity of the peaks varied with the location, serving as an indication of the difference in both the total concentration and the species of organic compounds present in the sample.

The signal-to-noise ratio (SNR) was calculated for the set of tap water samples as a difference between peak fluorescence of the sample and the distilled water (reference) signals divided by double the standard deviation of 1000 measurements. The results were typically in the 300-900 range for the laser sources, and around 30-190 for the LEDs (Sharikova & Killinger, 2010). For example, at 266 nm, the laser pulse energy was about 100 times greater than the LED pulse energy, but the SNR values differed only by a factor of 10 being about 296 for the laser LIF and about 26 for LED excitation for the Ann Arbor water data. The reason for the stronger than expected signal with the LED excitation may be due to differences in the light excitation and fluorescence overlap volume in the LED configuration or sample photobleaching in the case of the lasers.

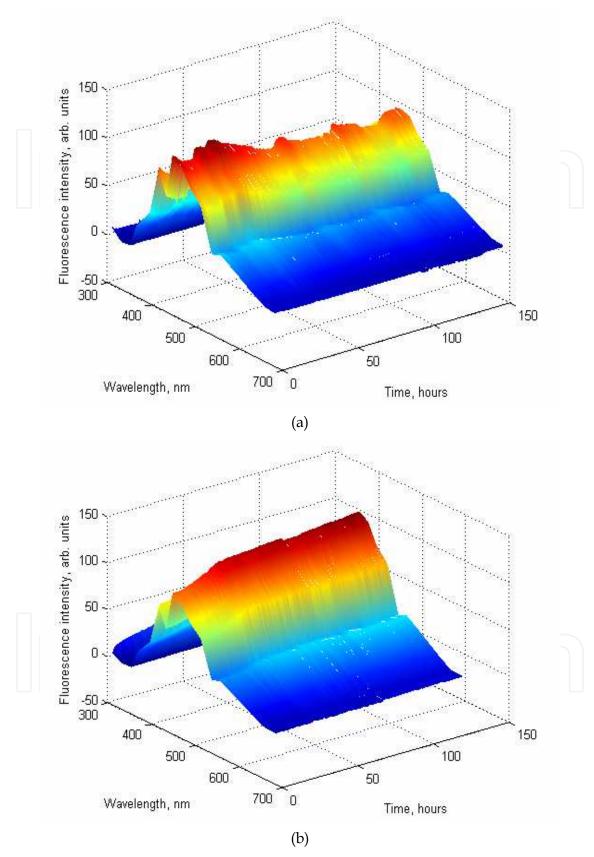
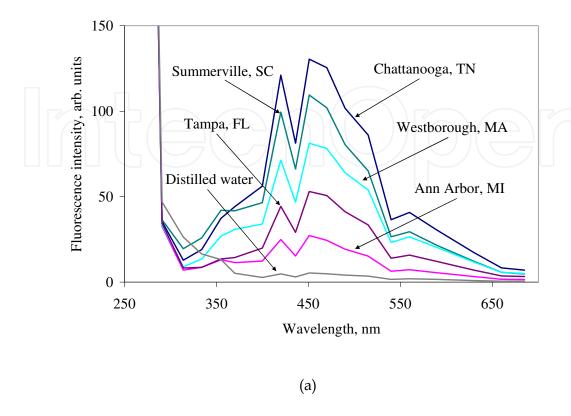


Fig. 6. Week long continuous tap water monitoring with 266 nm laser excitation: (a) running tap water; (b) recirculated tap water



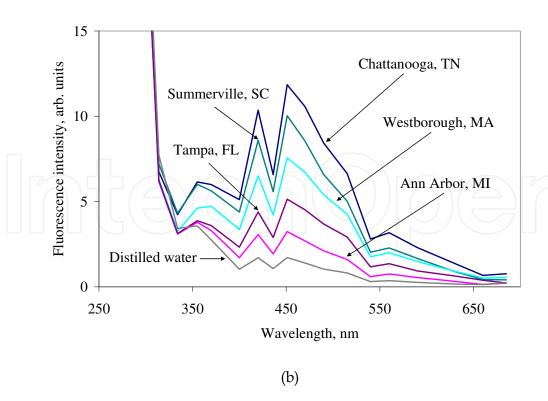


Fig. 7. Fluorescence of tap water with (a) 266 nm laser and (b) 265 nm LED excitation

3.3 Fluorescence spectrum of tannic acid and chlorine added to water sample

The LIF system was used to study long-term changes in the fluorescence of Total Organic Carbon (TOC) and the influence of chlorine on TOC fluorescence. Tannic acid is often used to represent TOC in water analysis measurements and is a specific type of tannin (plant polyphenol) (Hudson et al., 2007). Distilled water and trace solutions of organic compounds were recirculated in the portable LIF system for several hours. Tannic acid representing Total Organic Carbon (3 mg/L in deionized water) and chlorinated tannic acid (3 mg/L TOC and 12 mg/L Cl in deionized water) samples were prepared by Mr. Panagiotis Amitzoglou from the water processing laboratory of Prof. Audrey Levine (Levine, 2006). The solutions were produced by dilution of pure chemicals in nanopure deionized water. Concentrations of TOC and chlorine in the samples shown here were typical of those in drinking water.

The LIF signal for a sample of tannic acid is shown in Fig. 8 and was obtained using the 266 nm laser. As can be seen, the fluorescence in Fig. 8 had the strongest peak at 370 nm, the second-strongest at 420 nm, and a weaker peak at 451 nm. This made it quite distinct from the typical natural fluorescence of water, for example Fig. 7(a), which had a broad peak maximized at 451 nm.

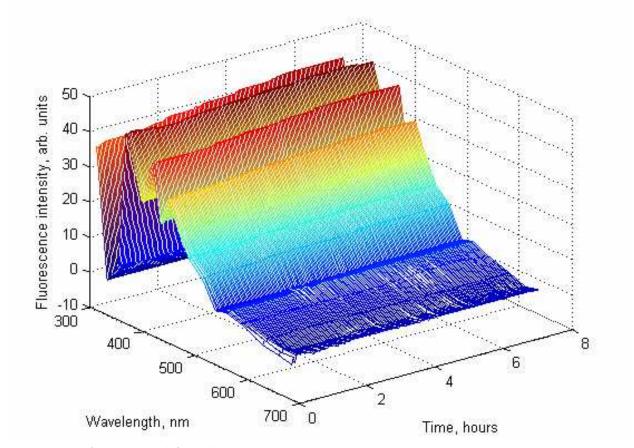


Fig. 8. LIF of tannic acid for 7 hours continuous monitoring; 266 nm excitation.

Fluorescence of a solution containing both tannic acid and chlorine is shown in Fig. 9 for 266 nm excitation; the solution was recirculated for about 5 hours. As can be seen, the spectrum for the 266 nm excitation was significantly different from that of tannic acid alone. The peak at 370 nm was completely suppressed, and the 420 nm peak was reduced drastically, so that the strongest fluorescence was observed at 451 nm.

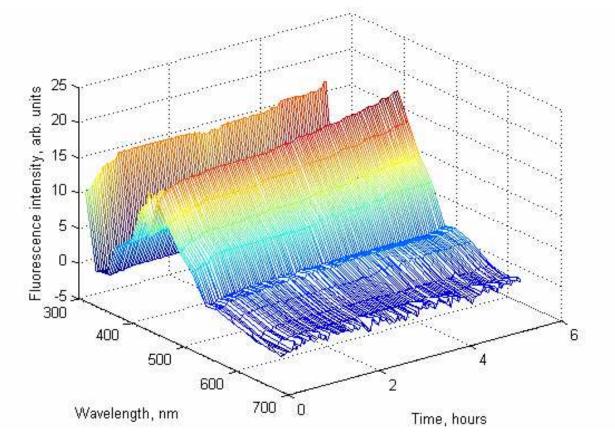


Fig. 9. LIF of tannic acid and chlorine for 5 hours continuous monitoring; 266 nm excitation.

3.3 Wine, juices, and distilled spirits

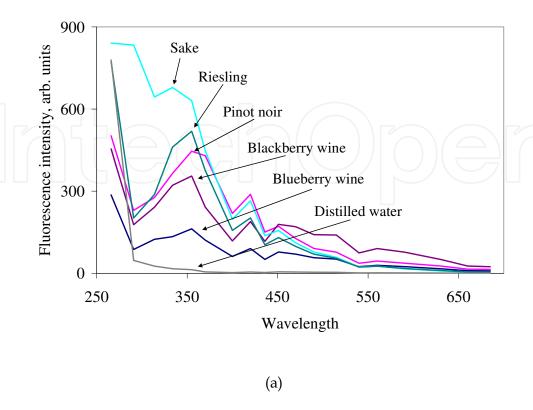
Several types of wine were diluted to ~1% with distilled water to minimize absorption and scattering in the sample cell. All spectra were taken under the same settings and conditions. Fluorescence spectra of these wine samples are shown in Fig. 10 (laser excitation) and Fig. 11 (LED excitation).

Comparing wine spectra (Fig. 10) with those of tap water (Fig. 7) one can see not only the difference in intensity, but also in the shape of the spectrum, particularly at 266 nm excitation. The emission peak at 350 nm dominates in the wine spectra of Fig. 10(a) using 266 nm excitation, but is possibly hidden by the scattering of the 355 nm excitation laser in Fig. 10 (b).

There is also a significant dependence on the excitation wavelength for the wine samples, as can be seen from Figs. 10 and 11. For example, at 266 nm excitation, the signal from sake is strongest, while fruit wines exhibit greater fluorescence at longer excitation wavelengths.

The laser- and LED-excited fluorescence spectra are generally consistent, with the exception of the Riesling wine that showed a much weaker secondary emission peak near 450 nm when the 265 nm LED was used. This might be due to the difference in the overlap between the sample absorption and the excitation line width, or be another indication of strong scattering at this excitation wavelength.

Finally, as part of applying our system toward common drinks, we also investigated the use of the LIF system for other drinkable liquids such as coffee and orange juice. Some of our results are shown in Fig. 12 using diluted samples. The data shows strong fluorescence emission near 350 nm and near 450 to 470 nm.



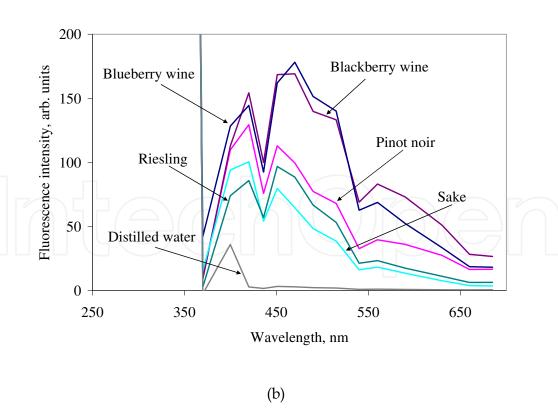


Fig. 10. Fluorescence of wine samples with (a) 266 nm and (b) 355 nm laser excitation

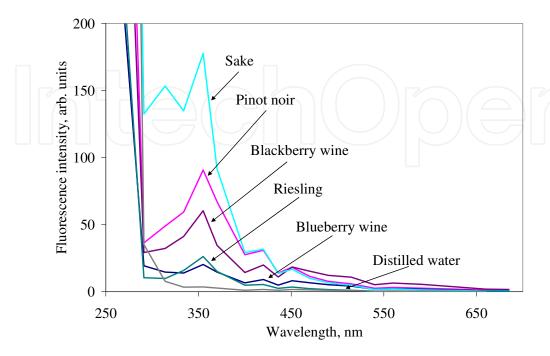


Fig. 11(a)

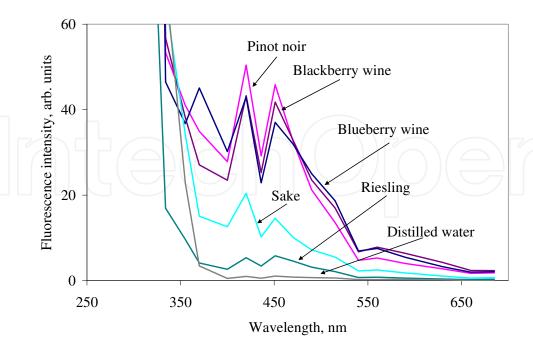


Fig. 11(b)

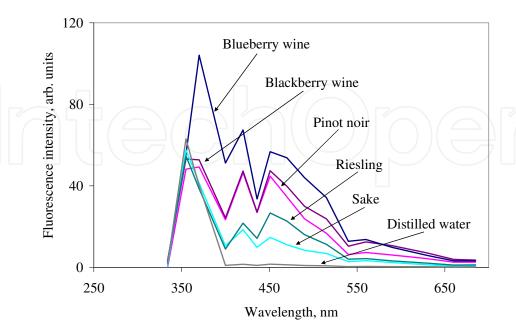


Fig. 11(c)

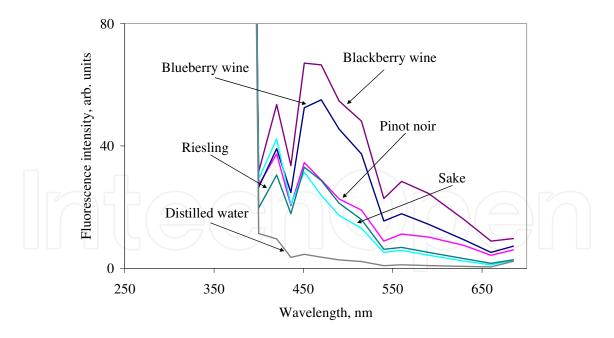


Fig. 11(d)

Fig. 11. Fluorescence of wine samples with (a) 266 nm, (b) 300 nm, (c) 335 nm, and (d) 355 nm LED excitation

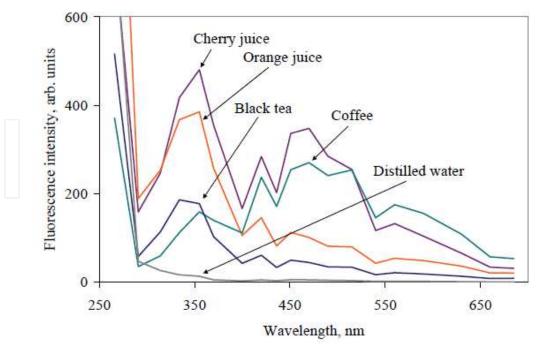


Fig. 12. Fluorescence of various drinks in distilled water; 266 nm laser excitation

3.4 Humic substance standards

Humic acid is a principal component of humic substances which are the major organic constituents of soil, humus, peat, coal, many upland streams, natural lakes, and ocean water. It is usually produced by biodegradation of dead organic matter. Samples of humic acid standards and natural organic matter were obtained from the International Humic Substances Society (www.ihss.gatech.edu). Figures 13 and 14 show fluorescence spectra of these substances with laser and LED excitation, correspondingly. Our data is in general agreement with excitation-emission spectroscopic properties of these materials as reported on the IHSS website (www.ihss.gatech.edu/spectra.html). It is interesting to note that the spectral peaks occur near 450 nm and 500 nm for these humic acid samples. Previous work by Coble has seen shifts in aged humic samples with fluorescence peaks gravitating toward the 500 nm wavelengths (Coble, 2007). Our results here are consistent with these earlier conclusions.

Further work on using LIF as a monitor of these humic substances is being conducted, and is also being explored as a way to monitor other hydrocarbon related substances.

4. Initial lab bench compact LIF measurement system

A compact version of our LIF system was also studied using a conventional compact spectrometer to replace the set of optical filters for fluorescence wavelength selection. Our earlier work had shown that such a system is several orders of magnitude lower in sensitivity than the system shown in Fig. 1, but the compact design and ease of use has many advantages especially for those cases where the concentration of the DOCs or CDOM is high. Toward this end, we developed a non-optimized compact system.

A schematic diagram of our laboratory bench-top LED-IF/LIF system is shown in Fig. 15. A microchip 266 nm laser or an UV LED equipped with a VIS-blocking CG-UG-11 filter

were used interchangeably to illuminate a sample containing quartz cell. Fluorescence emission passed through a UV-blocking filter to eliminate the second-order peak of the scattered excitation wavelength, and was collected at 90° by an optical fiber connected to the compact spectrometer (Ocean Optics, Inc., USB2000). The spectrometer output was sent to the PC via an USB cable.

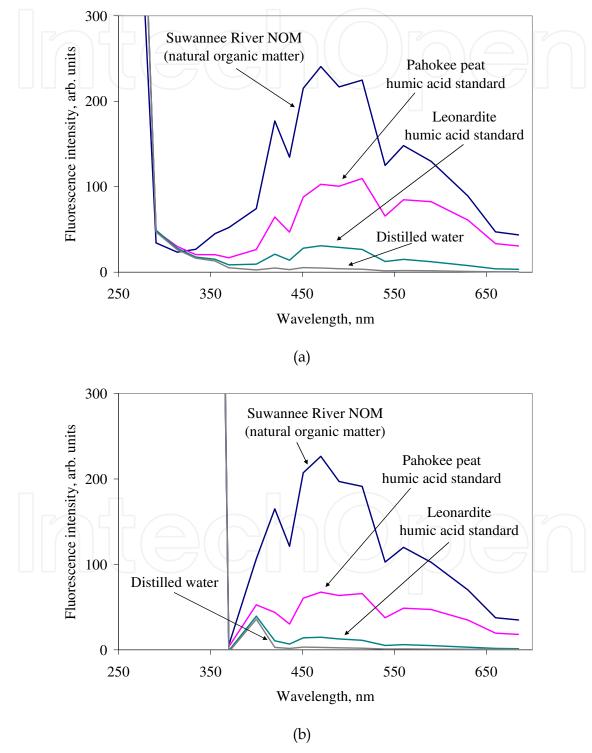


Fig. 13. Fluorescence of humic substances with (a) 266 nm and (b) 355 nm laser excitation

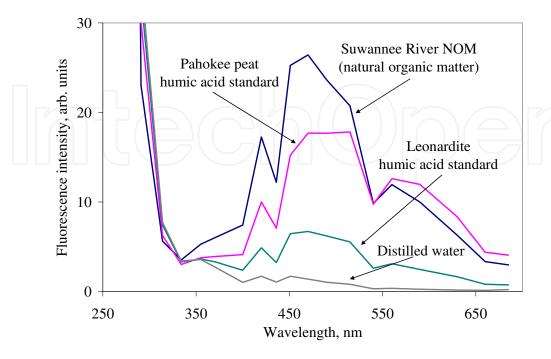


Fig. 14(a)

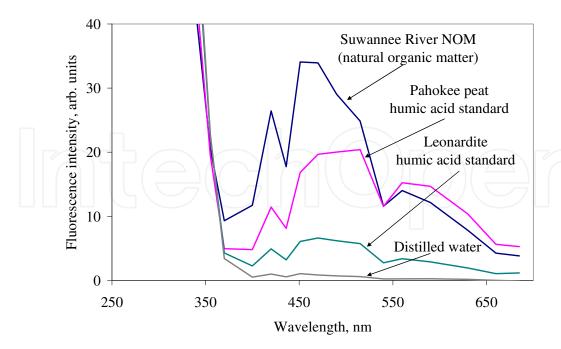


Fig. 14(b)

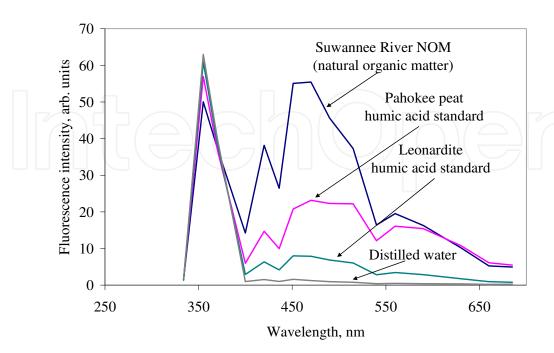


Fig. 14(c)

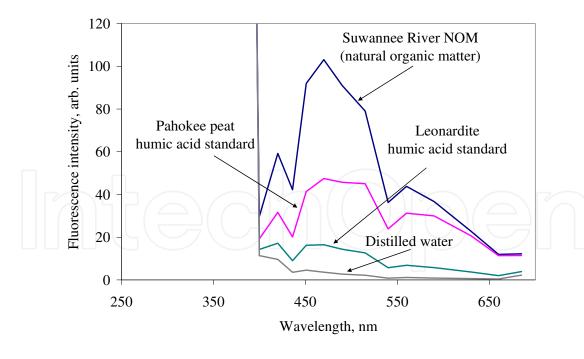


Fig. 14(d)

Fig. 14. Fluorescence of humic substances with (a) 266 nm, (b) 300 nm, (c) 335 nm, and (d) 355 nm LED excitation

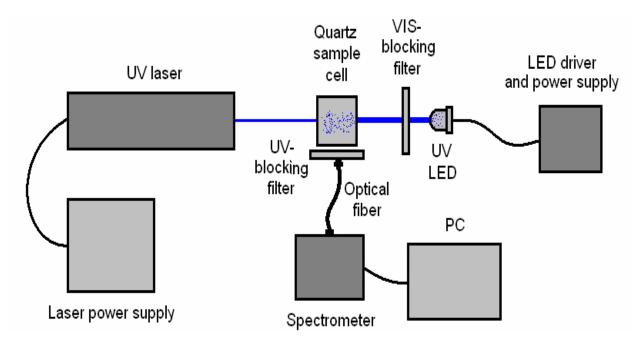


Fig. 15. Schematic diagram of the CW laboratory bench-top LED-IF / LIF system

Fluorescence of natural and drinking water samples were recorded using the system shown in Fig. 15 to compare the signal-to-noise ratio with the laser and LED excitation, and to determine the feasibility of CW LED sources in this compact setup. Lake water with its strong CDOM fluorescence was tested first, and then examples of drinking water were studied as well. As an example, Fig. 16 shows the fluorescence emission from lake water. As can be seen, the emission peak is near 500 nm, and is shifted somewhat from that observed for the tap water samples given earlier in Fig. 7. However, while the fluorescence signal is weak, the results are encouraging enough that more work is being conducted to better optimize the system and optical collection efficiency.

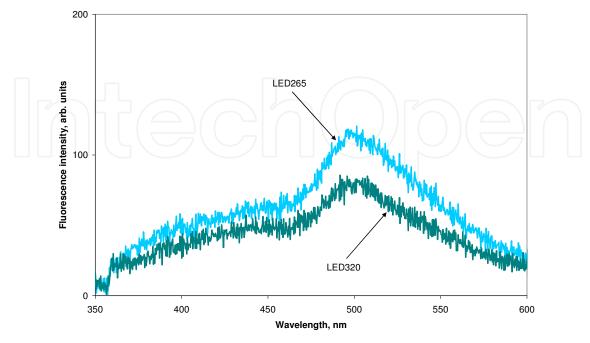


Fig. 16. Lake water fluorescence; 265 nm and 320 nm LED excitation; UV-blocking filter.

For comparison, a similar LIF spectrum for lake water, but using the 266 nm laser source, is shown in Fig. 17. As can be seen, the spectral features are similar, but the SNR is higher.

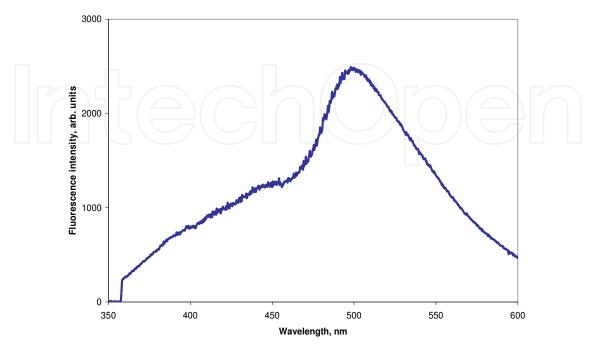


Fig. 17. Lake water fluorescence; 266 nm laser excitation; UV-blocking filter

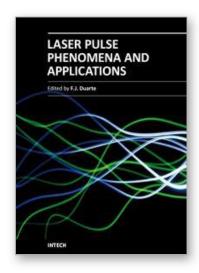
5. Conclusion

The laser- and LED-induced fluorescence system developed in our lab has been used to study the spectra of organic contaminants in drinking water and other liquids. Spectra obtained using LED and laser excitation at the same wavelength exhibited great similarity, while differing in overall intensity. Greater than expected SNR observed with LED excitation is a good indication that a compact, less expensive LED based system might be used for detection and/or monitoring of trace organics and appropriate species in these liquids. Overall, our results are important in that they show that deep-UV LIF spectroscopy may offer some advantages for measurements of trace species in water in real time. In some cases the fluorescence spectrum is unique, but in many cases similar spectral peaks are observed. In the latter case, our LIF system may not be suitable for selective discrimination and identification of different trace species in water, but can still be used for real time monitoring of a known substance if correctly calibrated. It is anticipated that further work will be required to better quantify these techniques, and further cross-comparisons will have to be made using convention analytical water quality instrumentation. However, the deep UV laser and LED LIF spectroscopic techniques described in this paper offer potentially new instruments for such measurements.

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