Absolute fluorescence quantum yield spectra of light scattering samples determined using thermal lens spectroscopy aided by optical absorbance and fluorescence measurements

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It has been demonstrated that thermal lens spectroscopy can be a useful tool to characterize the quantum yields of scattering [1] and fluorescence [2] in samples that experience separately these phenomena in their interaction with light. However, to the authors knowledge there are not reports about the measurement of this parameter when both phenomena are present simultaneously, as can occur often, for example in biological media. In this work, we implemented a pump-probe thermal lens spectrometer that uses a broadband high pressure Xe-arc lamp and a monochromator to generate the pump light source of variable wavelength between 350 and 700nm. The light is focused onto the sample to generate the thermal lens effect that is sensed by a collimated probe light beam provided by a low-power HeNe laser. The intensity of the pump beam is periodically modulated at a low frequency and the probe beam intensity is measured at the far field using a photodiode whose signal is fed to a Lock-in amplifier synchronized at the light modulation frequency. This home-made system is fully automatized to record a photothermal spectrum of a sample in a few minutes. A commercial UV- Vis Spectrophotometer is coupled to the experimental system to also record the sample´s absorbance and fluorescence excitation spectra. Combining these measurements, the quantum yield efficiency spectrum of fluorophores in a scattering solution can be determined and compared with that obtained without scattering. The test-samples studied were ethanol solutions of the Rhodamine 6G fluorophore, and scattering was produced by colloidally suspended polystyrene microbeads. The effects of luminescence quenching by a KI salt were also studied. We discuss an application of the technique for the characterization whole blood.

References

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