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Aggregation-induced emission spectral shift as a measure of local concentration of a pH-activatable rhodamine-based smart probe

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Abstract

Generating activatable probes that report about molecular vicinity through contact-based mechanisms such as aggregation can be very convenient. Specifically, such probes change a particular spectral property only at the intended biologically relevant target. Xanthene derivatives, for example rhodamines, are able to form aggregates. It is typical to examine aggregation by absorption spectroscopy but for microscopy applications utilizing fluorescent probes it is very important to perform characterization by measuring fluorescence spectra. First we show that excitation spectra of aqueous solutions of rhodamine 6G can be very informative about the aggregation features. Next we establish the dependence of the fluorescence emission spectral maximum shift on the dimer concentration. The obtained information helped us confirm the possibility of aggregation of a recently designed and synthesized rhodamine 6G-based pH-activatable fluorescent probe and to study its pH and concentration dependence. The size of the aggregation-induced emission spectral shift at specific position on the sample can be measured by fluorescence microspectroscopy, which at particular pH allows estimation of the local concentration of the observed probe at microscopic level. Therefore, we show that besides aggregation-caused quenching and aggregation-induced emission also aggregation-induced emission spectral shift can be a useful photophysical phenomenon.

Keywords: rhodamine; activatable probes; aggregation; dimerization; aqueous solution; aggregation-induced emission spectral shift