

# Multiwavelength ratiometric fluorescence sensing

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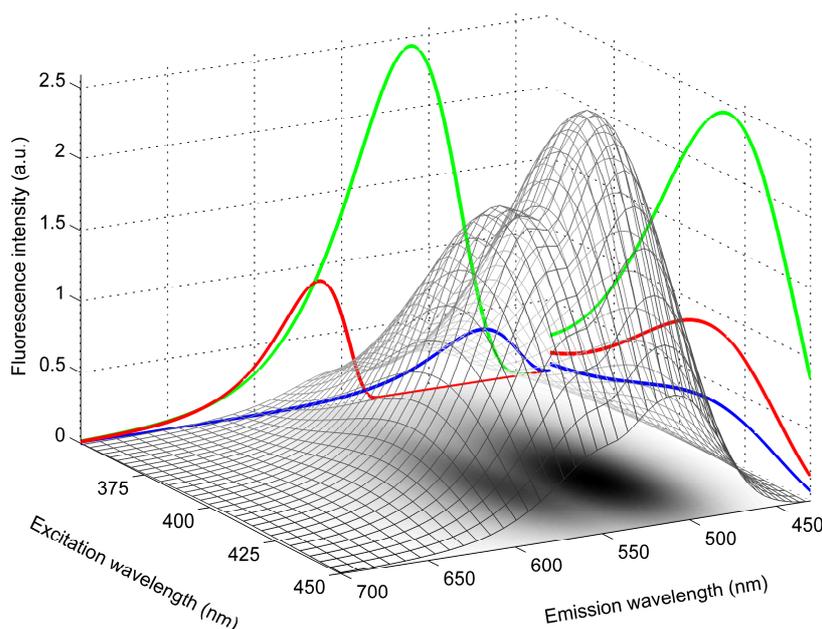
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Intermolecular interactions involve physical forces of different origin. The goal of many sensing and imaging technologies is to characterize these interactions and their dynamics from a single dye. To achieve such a goal we propose the use of dyes exhibiting ground-state and excited-state transformations that switch fluorescence between several emission bands. These transformations can be influenced by properties of the environment of the dye such as polarity, viscosity, electronic polarizability, H-bonding ability, temperature, pressure, pH, analyte concentrations, etc.

We suggest a model-based deconvolution analysis of the excitation-wavelength-dependent fluorescence spectra into basic components<sup>[1]</sup>. The fluorescence response of a 3-hydroxychromone derivative dye (FE<sup>[2]</sup>) that exhibits position-dependent H-bonding equilibrium and excited-state intramolecular proton transfer (ESIPT) was studied in detail. In this case, three fluorescence emission bands can be obtained, the relative fluorescence contributions of which are excitation-wavelength-dependent.

The developed algorithm for spectral deconvolution into individual components is based on their representation as asymmetric Siano-Metzler log-normal functions<sup>[3]</sup>. It is a true 2d approach, modeling both the excitation and emission spectra of the individual bands. The figure to the right shows the fluorescence response of FE in octanol decomposed into components, the projections of which are their respective emission and excitation spectra.



Application of the algorithm to the solvation response of FE allows separating the spectral signatures of polarity and H-bonding of the dye environment. The method has been used for characterizing the binding sites on pathological fibrillar aggregates of  $\alpha$ -synuclein using these dyes as the probes<sup>[4]</sup>.

## References

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