1.Fluorescence anisotropy reloaded… Rafael

Quantitative assessment of excitation energy transfer from 2D polarization portrait.

Difference between excitation and emission polarization are direct evidence of energy transfer. However, even if the modulation depth and phase of excitation and emission are the same, there still could be energy transfer present. Because Mex and Mem are integration parameters of 2D portrait, and **integration can always lead to the loss of information**. For example, there are those situations when there is no clear luminescence shift or no difference in the excitation and emission modulation depth, but the samples have completely different 2D polarization portrait, which implies different EET process (**give a example**). So how can we extract the full information hidden in the 2D polarization portrait to get insight into the EET process within the system.

More widely spread approach is to combine modulation depth analysis and topological model for multichromophoric system. The idea is to fit the experimentally observed M by changing the topological model and/or EET properties. **Average absolute energy transfer rotation angle** is also exploited for systems consisted of know number of chromophores of defined orientations.

The advantage of our model is that it works **without the need for any prior knowledge**, because we made many reasonable approximations.

Energy funneling concept is paramount in natural photosynthesis, artificial light harvesting devices, and become an important topic in single molecule spectroscopy community.

The 2D portrait can be separated into two limiting cases, **A is no EET component and B is EET funneling**.

In A the emissive states are initially photoselected by linearly polarized excitation light. Without EET, the emission polarization of A is dependent on the excitation polarization, on the other word, depends on the orientation of dipoles. However, the emission polarization of B is independent from the excitation polarization, because no matter which chromophore was initially excited, the emission will always occurs from the same pool of emitters after EET process. 𝜀 is called the energy transfer efficiency. It ranges from 0 to 1, for the absence of EET to unity. The higher the EET efficiency, the more excitation information can be lost.

**Motivation:**

The advantage of 2D portrait.

When the fitting mathematically works ideally (residue is very low), does the funnel indeed reflects the energy transfer pathways or is it just a mathematical construction that fits the data?

Furthermore, what is the limitation of the funnel? Whether there is extra information we can obtain even when the fitting is not ideal.

How to improve the usability of the model?

r0 – 𝜀

Fluorescence anisotropy (FA) for characterizing EET only be correctly for isotropically absorbing samples, but 𝜀 is insensitive to sample alignment.

For an isotropically absorbing sample (such as solutoin), FA (r0) and 𝜀 have the relation as shown in the equation: .

However, for the system possessing anisotropic absorption, FA will be dependent on the degree of alignment (which can be characterized by linear dichroism) and the orientation of alignment axis.

FA and 𝜀 can be used for characterizing the **EET process/rotation in solution**, but 𝜀 can also be applied for **oriented bulk samples**, such as stretched polymer films.

2. Quantitative characterization of light harvesting efficiency… Rafael

For a single system consisted of N chromophores, a general description of the fluorescence intensity of a multichromophoric system measured in the 2D polarization setup is:

N is the number of chromophores in the system. If there are EET process between the chromophores then I(𝜑ex, 𝜑em) is expressed as:

The energy transfer matrix Tik decribes the energy redistribution between chromophores. The diagonal element Tik(i=k) belongs to the NoET part, and the other elements contribute to the ET part. The 2D function I(𝜑ex, 𝜑em) can be separated into NoET and ET parts based on that:

Here we define the energy transfer efficiency , after some rearrangement, the above equation can be written as:

The above equation shows why it is difficult to analyze the ET process in as multichromophoric system. A system of N chromophores contains N funnels possessing different modulation depth Mfi, phase 𝜃fi and energy transfer efficiency 𝜀i.

We use **single funnel approximation (SFA)** to simplify the ET part. It is assumed that there is only one ET-emitter and the ET efficiency 𝜀 is the same for all chromophores. In the frame work of SFA, the above equations take following form:

The parameter 𝜀 works as a scaling factor to characterize the light-collecting efficiency over the whole system to an ET-emitter (funnel), and 𝜀 does not depend or affect the chromophore orientation.

The NoET part is simplified by a so called **three-dipole model** possessing very high symmetry. The whole system can be seen as one absorber possessing its own modulation depth Mex and phase 𝜃ex.

3. Light polarization in single molecule spectroscopy… Daniel Thomsson

The advantage of 2D portrait: Excitation and emission properties might be correlated. If the experiment are done independently (linear dichroism and fluorescence anisotropy), the correlation is lost. Two possible mechanisms behind correlated properties are energy transfer and rotation.

**(The definition of funnel.)** A funnel can possess different polarization properties in different situations. In 2000 Barbara et al. were the first to coin the phrase ‘funnel’ in relation to electronic energy transfer. The **funnel** was referred to as a potential energy landscape within a molecule containing a singlet exciton **trap** at the bottom. Funnel can also be an emitting **chromophore**, and the overlapping of funnels lead to a depolarizing emission. The different between a chromophore and a trap is that the trap can only be acceptor, but the chromophore can be both donor and acceptor. The expression chromophore may also be used in the sense of dye molecule or dipole.

The elements 𝜀ik in the energy transfer matrix Tik tell how much energy is transferred from chromophore i to chromophore k. Each column represent one chromophore and the sum of element in column k correspond to the total energy emitted from chromophore k. The emission from each chromophore is all fully polarized but may possess different phase.

The emission follows excitation in the NoET part. If we do not split the 2D portrait into ET and NoET parts, the contribution from the photoselection to the intensity modulation is highly sensitive to the energy transfer efficiency in multichromophoric molecules.

In the following equation, **𝜑ex = 𝜔ext,** where 𝜔ex is the angular speed.

The total available energy for transfer within a multichromophoric system is given by following equation:

Iexc is the excitation density (W cm-2), 𝜎i is the absorption cross section, i is the fluorescence quantum yield in the absence of EET. In most of the simulation, the excitation and emission transition are parallel, and Iexc𝜎i𝜙i = 1.

FFT- real numbers and positive frequencies.

Summary of results:

1. Extension of SFA3 model

In specific conditions, the model can work for multiple systems or one system possessing several emitters.

(write NoET and ET part for these situations.)

2. The symmetric three dipole model cannot fit that the dipoles does not orientate along the main axis of abs. The higher the ε, the lower the residue.

3. Break the symmetry of three dipole model by varying the angles between side dipoles and main dipole. The quality of excitation fitting is improved a lot.

4. Solution.

A. The higher the ε, the lower the anisotropy

B. The equation for computing fundamental anisotropy does not work when the abs is not polarized.

5. For one system, the model does not work well if it cannot be separated as several SFA sub-systems.