

Förster-resonance-energy transfer (FRET)

dipole-dipole excitation transfer

Non-radiative excitation transfer between two molecular entities separated by distances considerably exceeding the sum of their van der Waals radii. It describes the transfer in terms of the interaction between the *transition (dipole) moments* of the entities in the very weak dipole-dipole coupling limit. It is a Coulombic interaction frequently called a dipole-dipole coupling. The transfer rate constant from donor to acceptor, k_T , is given by

$$k_T = k_D \left(\frac{R_0}{r} \right)^6 = \frac{1}{\tau_D^0} \left(\frac{R_0}{r} \right)^6$$

where k_D and τ_D^0 are the *emission* rate constant and the *lifetime* of the excited donor in the absence of transfer, respectively, r is the distance between the donor and the acceptor and R_0 is the critical quenching radius or Förster radius, i.e., the distance at which transfer and spontaneous decay of the excited donor are equally probable ($k_T = k_D$) (see Note 3).

R_0 is given by

$$R_0 = \text{Const.} \left(\frac{\kappa^2 \Phi_D^0 J}{n^4} \right)^{1/6}$$

where κ is the *orientation factor*, Φ_D^0 is the fluorescence quantum yield of the donor in the absence of transfer, n is the average refractive index of the medium in the wavelength range where spectral overlap is significant, J is the spectral overlap integral reflecting the degree of overlap of the donor *emission spectrum* with the acceptor *absorption spectrum* and given by

$$J = \int_{\lambda} I_{\lambda}^D(\lambda) \varepsilon_A(\lambda) \lambda^4 d\lambda$$

where $I_{\lambda}^D(\lambda)$ is the normalized *spectral radiant intensity* of the donor so that $\int_{\lambda} I_{\lambda}^D(\lambda) d\lambda = 1$. $\varepsilon_A(\lambda)$ is the *molar decadic absorption coefficient* of the acceptor. See Note 3 for the value of *Const.*

Note 1: The bandpass $\Delta\lambda$ is a constant in spectrophotometers and spectrofluorometers using gratings. Thus, the scale is linear in wavelength and it is convenient to express and calculate the integrals in wavelengths instead of wavenumbers in order to avoid confusion.

Note 2: In practical terms, the integral $\int_{\lambda} I_{\lambda}^D(\lambda) d\lambda$ is the area under the plot of the donor emission intensity versus the emission wavelength.

Note 3: A practical expression for R_0 is:

$$R_0 / \text{nm} = 2.108 \times 10^{-2} \left\{ \kappa^2 \Phi_D^0 n^{-4} \int_{\lambda} I_{\lambda}^D(\lambda) [\epsilon_A(\lambda) / \text{dm}^3 \text{ mol}^{-1} \text{ cm}^{-1}] (\lambda / \text{nm})^4 d\lambda \right\}^{1/6}$$

The *orientation factor* κ is given by

$$\begin{aligned} \kappa &= \cos \theta_{DA} - 3 \cos \theta_D \cos \theta_A \\ &= \sin \theta_D \sin \theta_A \cos \varphi - 2 \cos \theta_D \cos \theta_A \end{aligned}$$

where θ_{DA} is the angle between the donor and acceptor moments, and θ_D and θ_A are the angles between these, respectively, and the separation vector; φ is the angle between the projections of the transition moments on a plane perpendicular to the line through the centres. κ^2 can in principle take values from 0 (perpendicular transition moments) to 4 (collinear transition moments). When the transition moments are parallel and perpendicular to the separation vector, $\kappa^2 = 1$. When they are in line (i.e., their moments are strictly along the separation vector), $\kappa^2 = 4$. For randomly oriented *transition (dipole) moments*, e.g., in fluid solutions, $\kappa^2 = 2/3$.

Note 4: The transfer quantum efficiency is defined as

$$\Phi_T = \frac{k_T}{k_D + k_T}$$

and can be related to the ratio r/R_0 as follows:

$$\Phi_T = \frac{1}{1 + (r/R_0)^6}$$

or written in the following form :

$$\Phi_T = 1 - \frac{\tau_D}{\tau_D^0}$$

where τ_D is the donor excited-state lifetime in the presence of acceptor, and τ_D^0 in the absence of acceptor.

Note 5: FRET is sometimes inappropriately called fluorescence-resonance energy transfer. This is not correct because there is no fluorescence involved in FRET.

Note 6: Foerster is an alternative and acceptable spelling for Förster.

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