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Refinement and Application of Single Molecule Resonance Energy Transfer

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Single molecule fluorescence measurement of resonance energy transfer (RET) is revealing a new level of detailed information about biomolecular systems that exhibit conformational change. This technique has been particularly successful when applied to processes that involve large, binary intramolecular movements, for example rearrangement of enzymes and the folding of two-state proteins. Despite many advances in the application of single molecule RET, most researchers are still reluctant to convert measured energy transfer efficiencies to distances. A number of experiments inspired by Stryer and Haugland's 1967 "spectroscopic ruler" paper have been carried out at the single molecule level using DNA and polyproline, but the results are not nearly so clear as they were in that original work. I will discuss progress that has been made toward understanding the results of these experiments and the deviations they show from Förster's theory.