

FRET and TR-FRET Peptides

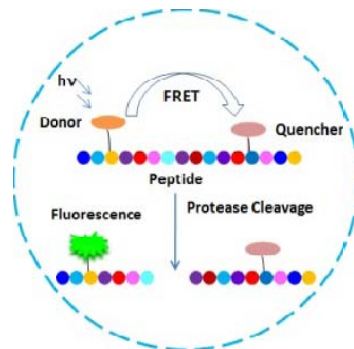


The Peptide Quality Standard

FRET (Fluorescence Resonance Energy Transfer) is a distance dependent dipole-dipole interaction without the emission of a photon resulting in the transfer of energy from an initially excited donor molecule to an acceptor molecule. It allows the detection of molecular interactions in the nanometer range.

FRET peptides are labeled with a donor molecule and an acceptor (quencher) molecule. In most cases the donor and acceptor pairs are two different dyes. The transferred energy from a fluorescent donor is converted into molecular vibrations if the acceptor is a non-fluorescent dye (quencher). When the FRET is stopped (by separating donor and acceptor) an increase of donor fluorescence can be detected. When both the donor and acceptor dyes are fluorescent, the transferred energy is emitted as light of longer wavelength so that the intensity ratio change of donor and acceptor fluorescence can be measured. In order for efficient FRET quenching to take place the fluorophore and quencher molecules must be close to each other ($\sim 10\text{-}100 \text{ \AA}$) and the absorption spectrum of the quencher must overlap with the emission spectrum of the fluorophore. While designing a donor-quencher FRET system, a careful comparison of the donor's fluorescence spectrum with the quencher's absorption spectrum is required.

Peptide is labeled with a fluorescent donor moiety and a non-fluorescent acceptor (quencher) separated by a protease cleavable sequence. After the cleavage the peptide fragment containing donor the dye emits fluorescence due to the loss of FRET



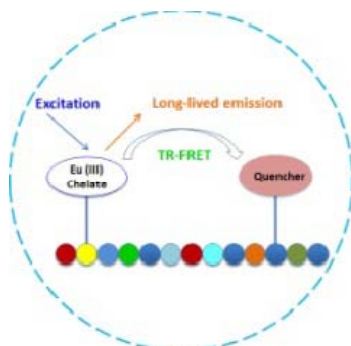
Chinese Peptide Company is the world leader in FRET peptide design and synthesis

- **Sequence modification for FRET design and synthesis**
- **Selection of appropriate donor-acceptor pair**
- **Improvement of FRET substrate solubility and quenching efficiency**
- **Design and synthesis of TR-FRET peptides**

CPC has experience with a wide range of protease peptide substrates including: Aggrecanase, ADAMs, ACE-2, APCE, 2A protease, BACE1, Calpains, Carboxypeptidases, Caspases, Cathepsins, Chymopapain, Complement component C1s, CMV protease, ECE-1, Factor Xa, Furin, GranzymeK, HCV protease, HIV protease, HRV1, Kallikreins, Interferon alpha A, Lethal Factor Protease, Malaria Aspartyl Proteinase, MMPs, Pepsin, Plasmin, Plasmepsin II, Proteinases, Protein Tyrosin Phosphatase, Renin, SARS, TACE, Thrombin, TEV protease, Trypsin and West Nile Virus Protease.

Time-resolved FRET (TR-FRET) peptide

Time-resolved FRET (TR-FRET) has emerged as a method that utilizes long-lived fluorophores to enable the measurements to be delayed by 50–150 microseconds. This time delay allows the signal to be cleared of most nonspecific short-lived emissions.



Eu(III) chelate/ QSY-7
(Ex/Em=340nm/613nm) based TR-FRET
peptide substrates for HTS assay are
available from CPC

CPC offers a variety of Donor-Acceptor pairs for FRET design including long wavelength pairs for better sensitivity

Examples of Donor-Acceptor Pairs for FRET peptide design

Mca/Dnp
Ex/Em=380/460 nm

Edans/Dabcyl
Ex/Em=340/490nm

5-FAM/CPQ 2
Ex/Em=490/520 nm

5-TAMRA/QSY-7
Ex/Em=547/574nm

Cy5/QSY-21
Ex/Em=650/670 nm

An example of Donor-Acceptor pair for TR-FRET HTS assay

Eu(III) Chelate/QSY-7
Ex/Em=340/613nm

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