



April 10th, 2021

Dr. Lena von Voithenberg

DKFZ (German Cancer Research Center)

Guest Editor JoVE

Collection of video articles on Förster Resonance Energy Transfer (FRET):
Methods and Applications.

Dear Dr. von Voithenberg

Please enclosed find the manuscript and video entitled “**Chemical modification of the tryptophan residue in an engineered recombinant N-domain from SERCA elucidates FRET between tryptophan and 8-anilidonaphthalene sulfonate**” by José G. Sampedro and Yolanda Cataño. This is a method paper and video submitted as response to your kind invitation for the collection of video articles “Biocatalytic Förster Resonance Energy Transfer (FRET): Methods and Applications” in JoVE. The paper and video are being submitted for review and eventual publication in Journal of Visual Experiments (JoVE).

The manuscript and video describes the NBS mediated chemical modification of Trp residues in the nucleotide binding domain (N-domain) of SERCA as an assay to test the existence of FRET between Trp and ANS. The ANS – N-domain complex shows fluorescence spectra suggesting the existence of FRET, i.e., N-domain fluorescence decreases while ANS fluorescence increases. NBS chemical modification of the N-domain led to quench the intrinsic fluorescence of the protein; hence this allows assaying whether FRET does really exist between these two fluorescent molecules. Results showed that FRET did not take place as ANS fluorescence still is observed when exciting at $\lambda = 295$ nm. Therefore, the assay may be useful to test FRET in other systems whenever this is suspected to occur. The assay is rapid and easy to perform and may also be used with other fluorophores besides ANS.

We hope the paper and video content may fulfill the requirements for



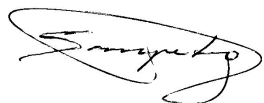
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publication in JoVE.

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Sincerely yours



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