Design and Synthesis of Stimuli Activated Near-Infrared Dyes to Target Cellular Organelle

INDEX NO: 74/19/Chem./26

Abstract: We demonstrated ratiometric acidic pH-activatable visible to NIR switchable fluorescent dye that we synthesised. An organic cell-permeable probe with an acidic pH-activatable oxazolidine moiety and lysosome-targeting morpholine activity makes up the design. Through ring opening of the oxazolidine moiety at acidic pH, the visible closed oxazolidine form (abs418 nm) can be changed to the highly conjugated NIR Cy-7 form (abs780 nm). The pH can be used to influence the ratiometric fluorescent probe's switching, which is very reversible. Spectroscopies for NMR, UV/vis, and fluorescence allowed for the observation of the probe's pH switching behaviour. We have shown how to efficiently synthesise acidic pH-triggered visible to NIR interchangeable ratiometric fluorescent pH sensors using crystal structures. This bioresponsive probe has reversible pH-sensitive absorption/emission characteristics, low cytotoxicity, a huge bathochromic spectral shift at 322 nm with enhanced quantum yield from neutral to acidic pH, high sensitivity and selective targeting of live cell lysosomes with ideal pKa, off-to-on narrow NIR absorption/fluorescence signals with high molar absorption coefficient at acidic lysosomal lumen, and in situ live Using a dual channel confocal laser scanning microscope, selective labelling and ratiometric pH imaging in human cancer live cell lysosomes are observed. The pH-activatable organic fluorescent dye used in this experiment has a morpholine moiety for lysosome targeting and an acidic pH. We have designed and synthesized acidic pH-activatable dual targeting ratiometric fluorescent probe-peptide conjugate using SPPS protocol on Rink amide AM resin for living carcinoma cell specific active targeting successively cell penetration and selective staining of lysosomes alongside real-time monitoring, 3D, and multicolor live-cell imaging. The design consists of a RGDS peptide residue to target cancer cell surface overexpressed receptor

Design and Synthesis of Stimuli Activated Near-Infrared Dyes to Target Cellular Organelle

INDEX NO: 74/19/Chem./26

$\alpha v\beta_3$ integrin, live-cell penetrating organic Changsha chromophore comprising a lysosome targeting morpholine group, and acidic pH openable spiro-lactam ring for visible-to-NIR switchable ratiometric response. The fluorescent probe-peptide conjugate exhibits intramolecular spirolactamization at physiological and basic pH through Arg amide NH of the RGDS peptide residue. The visible spirolactam state (NIR OFF) can be switched to the highly conjugated push-pull NIR open amide state (NIR ON) through spiro-lactam ring opening triggered by acidic pH with huge bathochromic shift ($\Delta\lambda_{abs}$ = 336 nm, $\Delta\lambda_{em}$ = 265 nm) and displays dynamic pH-sensitive ratiometric optical switching. This bioresponsive *in situ* acidic cancer cell lysosome activatable functional fluorophore-peptide conjugate shows enormous bathochromic fluorescence shift of 265 nm from physiological to acidic pH, OFF-to-ON narrow NIR abs/em bands with augmented molar absorptivity, respectable quantum yield, and ultra-brightness at acidic lysosomal pH; negligible cytotoxicity, and dual targeted ratiometric imaging capability of living cancer cell lysosomes with perfect pK_a.

Samit Guha
17th October 2023

Dr. Samit G...
Assistant P...
Departme...
Jadavpur ...
Kolkata-700...

Signature with Seal of the Supervisor

Ayan Mukherjee 17/10/...
Full signature of the ca...