Engineering of Switchable Aptamer Micelle Flares for Molecular Imaging in Living Cells

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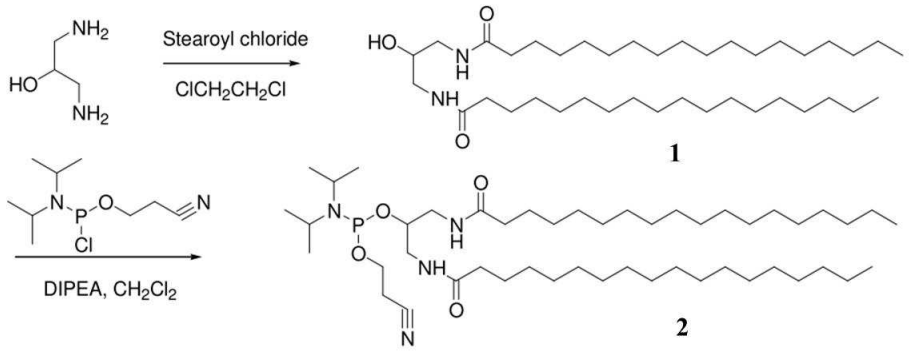
Supporting Information:

Within this supporting information are found:

Experimental Section:

Figure S1: Synthesis of diacyllipid phosphoramidite.



Synthesis of compound 1: A solution of stearoyl chloride (5.00 g) in 50 mL of 1,2-dichloroethane was added dropwise to a solution of 1,3-diamino-2-dydroxypropane (0.73 g) and triethylamine (TEA) (2.57 mL) in 100 mL of 1,2-dichloroethane. The reaction mixture was stirred under a blanket of argon at room temperature for 2h and then at 70 ˚C overnight. The mixture was then cooled to room temperature. The solid product was filtered, washed with CH_2Cl_2, CH_3OH, 5% $NaHCO_3$ and $CH_3CH_2OCH_2CH_3$, in order, and vacuum dried to yield compound 1 as a white solid. The product was identified by ^1H NMR according to data reported in the literature.[1]

Synthesis of compound 2: N,N-diisopropylethylamine (DIPEA) (4.19 mL) was injected into a solution of compound 1 (3.00 g). The solution was cooled on an ice bath under a blanket of argon, and then 2-cyanoethyl-N,N-diisopropylchlorophosphoramidite (2.15 mL) was added dropwise. The reaction mixture was stirred under room temperature for 1h and then at 80˚C for 1.5h. The solution was then cooled to room temperature, washed with 5% $NaHCO_3$ and brine, Na_2SO_4 dried, and vacuum concentrated. The product was precipitated out as a white solid by

adding concentrated solution into CH_3CN. The product was identified by ^1H NMR according to data reported in the literature.[1]

Table S2: Detailed sequence information for all oligonucleotide probes.

Name	Sequences
SAMF	5’ Lipid-$(PEG)_2$-Dabycl-GAC CTG GGG GAG TAT TGC GGA AGG TT-$(PEG)_6$-CCA GGT C-TMR 3’
CSMF	5’ Lipid-$(PEG)_2$-Dabycl-GAC CTG GGG GAG TAA TGC CGA AGG TT-$(PEG)_6$-CCA GGT C-TMR 3’
ASP	5’ $(PEG)_2$-Dabycl-GAC CTG GGG GAG TAT TGC GGA AGG TT-$(PEG)_6$-CCA GGT C-TMR 3’

Red denotes mismatched base.

Figure S3: Agarose gel electrophoresis to demonstrate the formation of SAMF. The nanostructures of SAMF and CSMF result in slower movement in gel electrophoresis and an upper band relative to the band for the aptamer switch probe without diacyllipid conjugation.

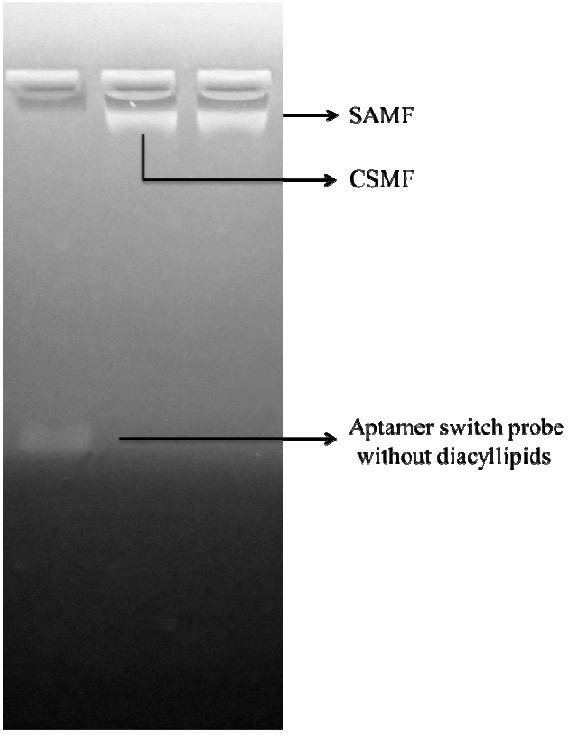


Figure S4: Specificity of SAMF and CSMF to target molecules, λ_{ex}=550 nm, λ_{em}=565-650 nm.

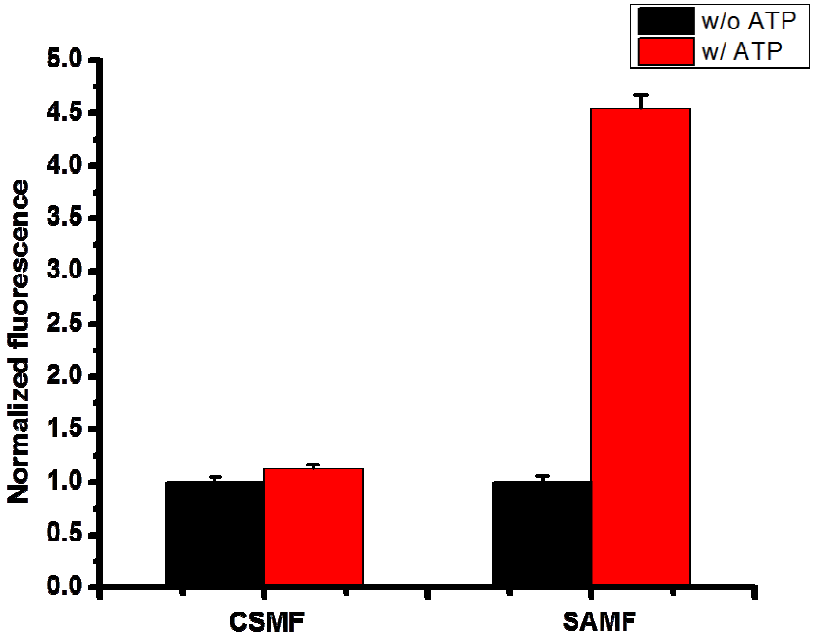


Figure S5: Time lapse fluorescence imaging of SAMF and CSMF in HeLa cells. Confocal laser scanning microscopy images of HeLa cells treated with 1 μM SAMFs or CSMFs for 0.5 h (a, b), 1 h (c, d), 2 h (e, f) and 4 h (g, h). In each image, left panel is TAMRA fluorescence psudo-colored red and right panel is the overlay of TAMRA fluorescence and bright field image. Scale bars: 20 μm.

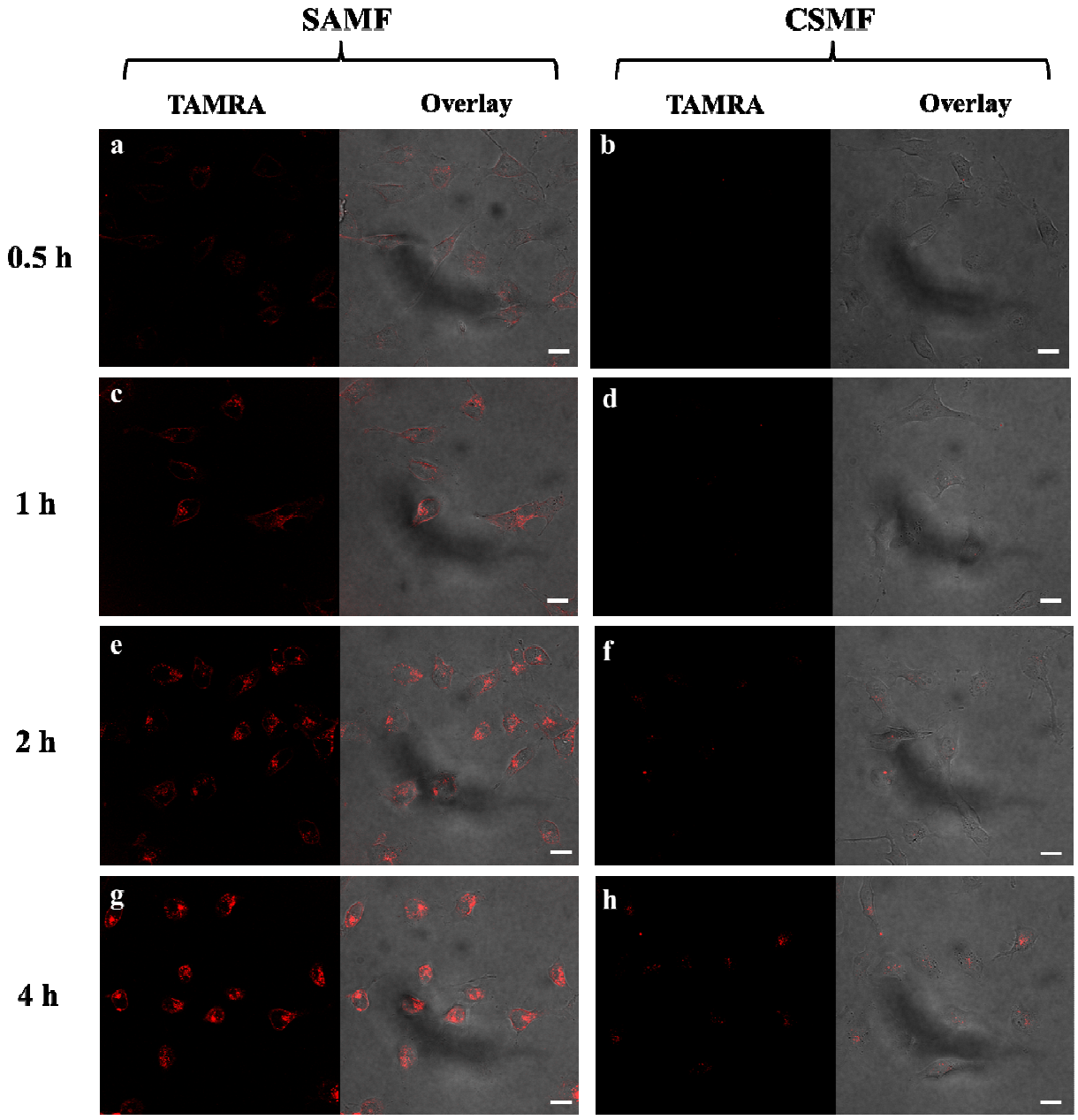


Figure S6: Co-localization assay of (a) SAMFs and (b) CSMFs with LysoSensor. The left panels are LysoSensor fluorescence pseudo-colored green, the second panels are TAMRA fluorescence pseudo-colored red, the third panels are bright field images and right panels are the overlay of LysoSensor fluorescence, TAMRA fluorescence and bright field image. Scale bars: 20 μm.

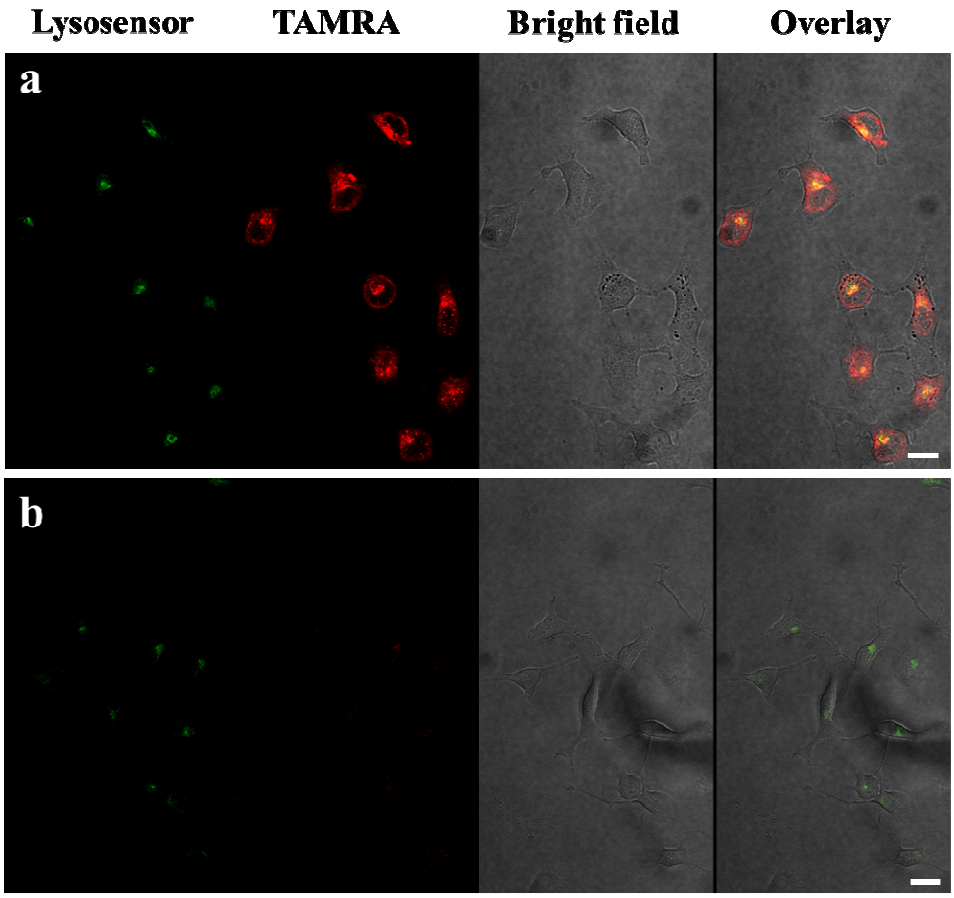


Figure S7: Quantification of intracellular ATP imaging. Live HeLa cells treated with 100 µM etoposide or 3 µg/ml oligomycin compared to cells without treatment and quantified with ImageJ software. The fluorescence intensity of cells without treatment was set to 100.

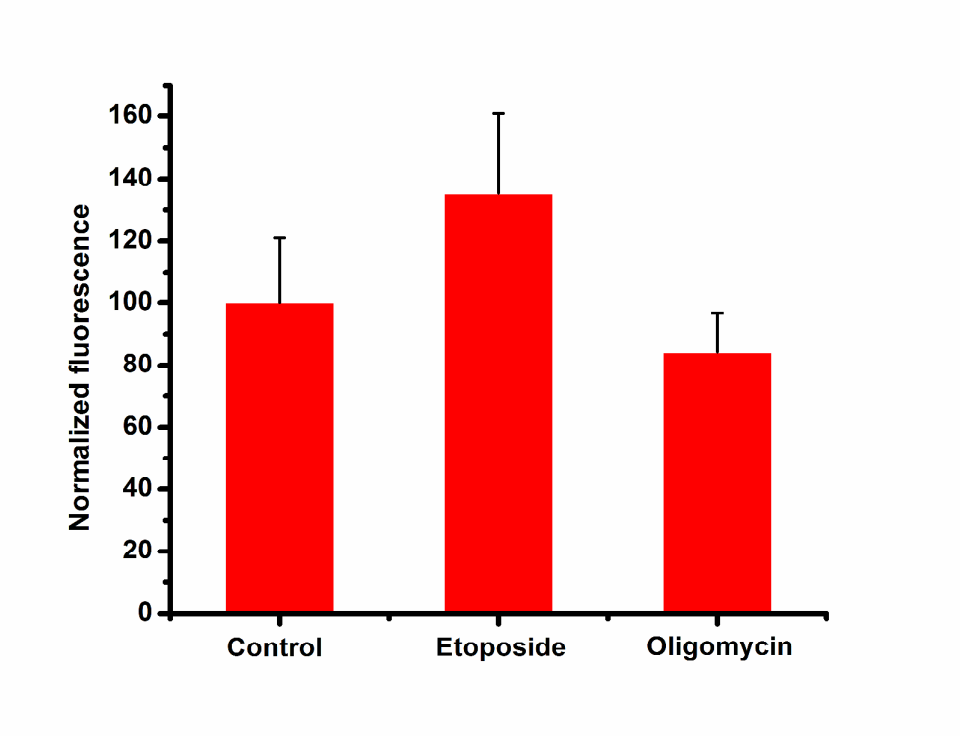


Figure S8: Cellular ATP concentration after treatment with etoposide and oligomycin. Live HeLa cells treated with 100 μM etoposide or 3 μg/ml oligomycin, followed by measurement of cellular ATP concentration with commercial luciferase assay kit.

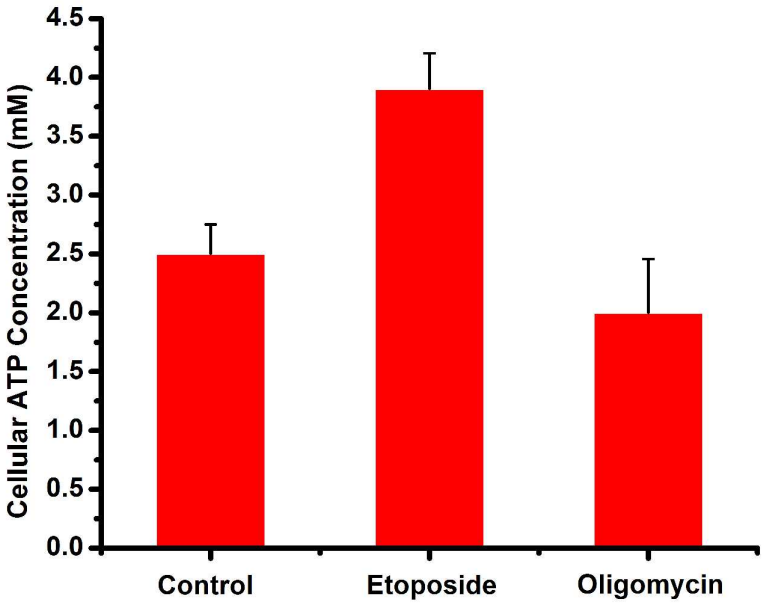
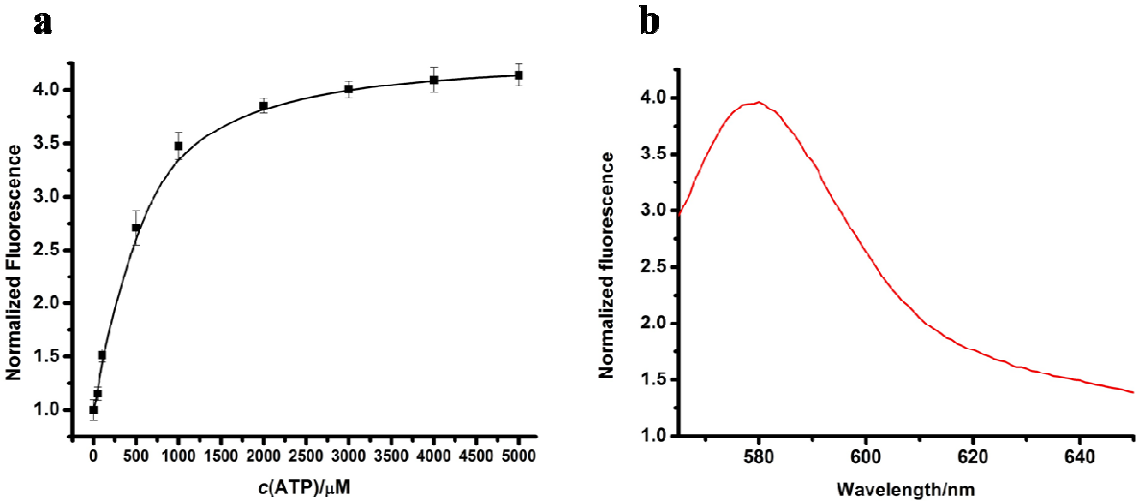


Figure S9: Measurement of cellular ATP concentration with SAMF in HeLa cell lysates. (a) Fluorescence titration of SAMF against different of concentrations of ATP in cytosol buffer. (b) The emission spectrum for TAMRA was normalized in HeLa cell lysates (F_{TMR}=3.95, cellular ATP concentration=2.8 mM).



References:

1. Liu, H.; Zhu, Z.; Kang, H.; Wu, Y.; Sefan, K.; Tan, W. DNA-Based Micelles: Synthesis, Micellar Properties and Size-Dependent Cell Permeability. *Chem. Eur. J.* **2010**, 16, 3791-3797.