Supporting Information

A DNA Octahedron-based Fluorescence Nanoprobe for Dual Tumor-related mRNAs Detection and Imaging

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Table S1. Oligonucleotide sequences.

Name	Sequence (5'-3')
A-1	CACCAGGTTGAATCCTATGCTCGTACATTGTCGCAGTTCAGATACGCTTCATACTGA GAGCGTTCCG
A-Cy5	**Cy5-**CACCAGGTTGAATCCTATGCTCGTACATTGTCGCAGTTCAGATACGCTTCATAC T GAGAGCGTTCCG
A-2	GCGAGTGTCTTTGGCATACTT
A-BHQ3	GCGAGTGTCTTTGGCATACTT-**BHQ3**
B	GGATACATTATACGGTGGTTTGTACGAGCATAGGATTCTTCCTGGTGAAGTATGCCA ATTGGATCCTAAATTCTTGCG
C-1	CCACCGTATAATGTATCCTTCGAGCAGCACGAACTGTCTTCATTCGTCGTCGTCGTA GTTCTGTAGCC
C-Cy3	CCACCGTATAATGTATCCTTCGAGCAGCACGAACTGTCTTCATTCGTCGTCGTCGTA GTTCTGTAGCC-**Cy3**
C-2	TCTTATGCGGATAGTGAAAGC
C- BHQ2	**BHQ2-**TCTTATGCGGATAGTGAAAGC
D	GCGTATCTGAACTGCGACTTCCGCATAAGAGGCTACAGTTGGACCGTAGTTAAATG ACTTCGGAACGCTCTCAGTATGTTTTT**GGTGGTGGTGGTTGTGGTGGTGGTGG**
D-1	GCGTATCTGAACTGCGACTTCCGCATAAGAGGCTACAGTTGGACCGTAGTTAAATG ACTTCGGAACGCTCTCAGTATG
E	CGGTCATCGTCGTCGTCGTTGCACGAATACGAATACTATTCGCAAGAATTTAGGATC CTTGTCATTTAACTACGGTCC
F	CGACGACGACGATGACCGTTCTACGACGACGACGAATGTTGACAGTTCGTGCTGCT CGTTTAGTATTCGTATTCGTGCTTTTT**GGTGGTGGTGGTTGTGGTGGTGGTGG**
F-1	CGACGACGACGATGACCGTTCTACGACGACGACGAATGTTGACAGTTCGTGCTGCT CGTTTAGTATTCGTATTCGTGC
TK1 target	AAGTATGCCAAAGACACTCGC
GalNAc-T target	GCTTTCACTATCCGCATAAGA
c-myc target	CCTCAACGTTAGCTTCACCAA
survivin target	CAAGGAGCTGGAAGGCTGGG
c-raf-1 target	AATGCATGTCACAGGCGGGA

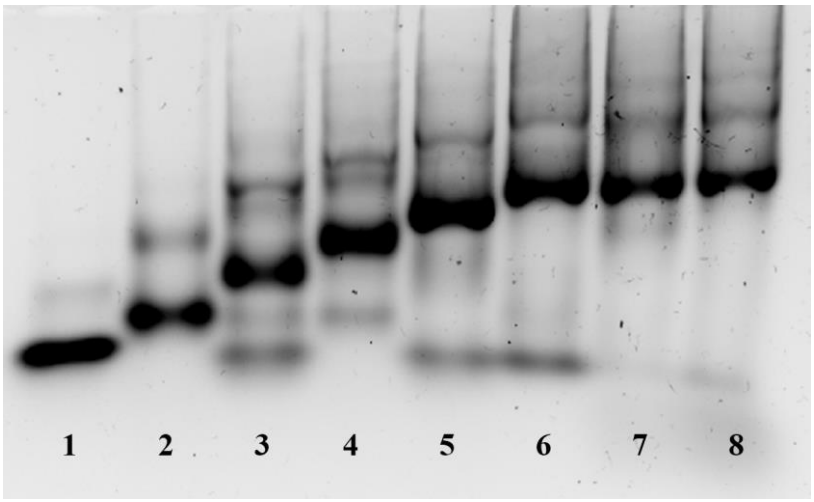

Figure S1. The synthesis of DNA nanoprobe characterized by 1% agarose gel electrophoresis. Lane 1 is strand F; Lane 2 is strand E+strand F; Lane 3 is strand E+strand F+strand C-1; Lane 4 is strand E+strand F+strand C-1+strand B; Lane 5 is strand E+strand F+strand C-1+strand B+strand A-1; Lane 6 is strand E+strand F+strand C-1+strand B+strand A-1+strand D; Lane 7 is strand E+strand F+strand C-1+strand B+strand A-1+strand D+strand A-2; Lane 8 is strand E+strand F+strand C-1+strand B+strand A-1+strand D+strand A-2+strand C-2.

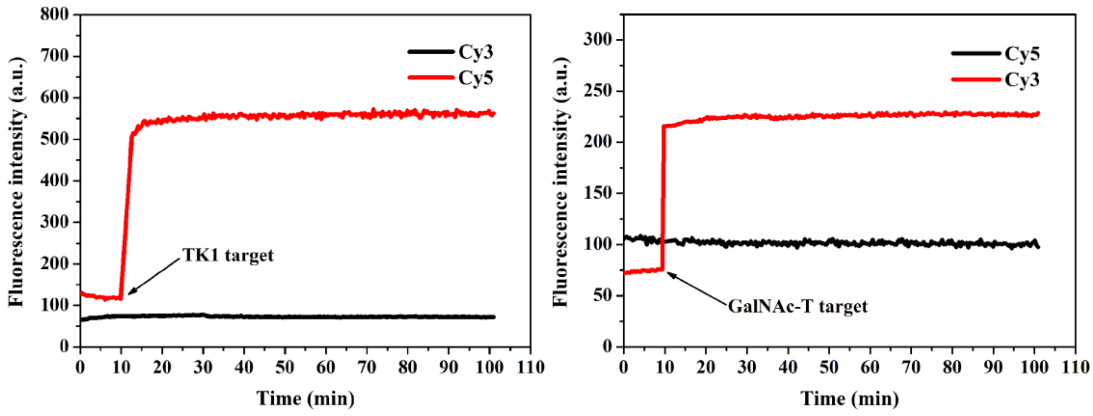

Figure S2. Kinetics Assay. The fluorescence intensities of the nanoprobe were measured at 645 nm and 545 nm excitation wavelength with the emission wavelength of 666 nm and 566 nm, before and after mixing with TK1 target (200 nM, left) or GalNAc-T target (200 nM, right).

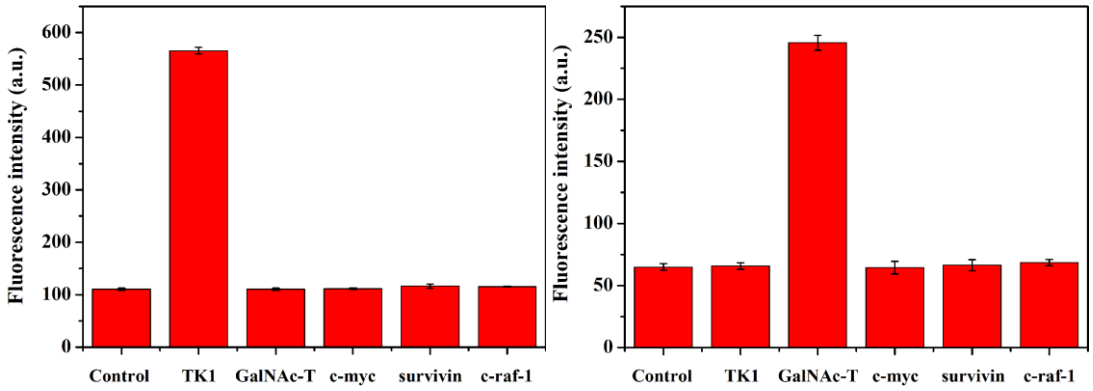

Figure S3. Specificity of the nanoprobe over several DNA targets. The DNA nanoprobe was mixed with different targets (TK1, GalNAc-T, c-myc, survivin and c-raf-1) at concentration of 200 nM, and the fluorescence intensities were measured with 645 nm (left) and 545 nm (right) excitation wavelength, respectively.

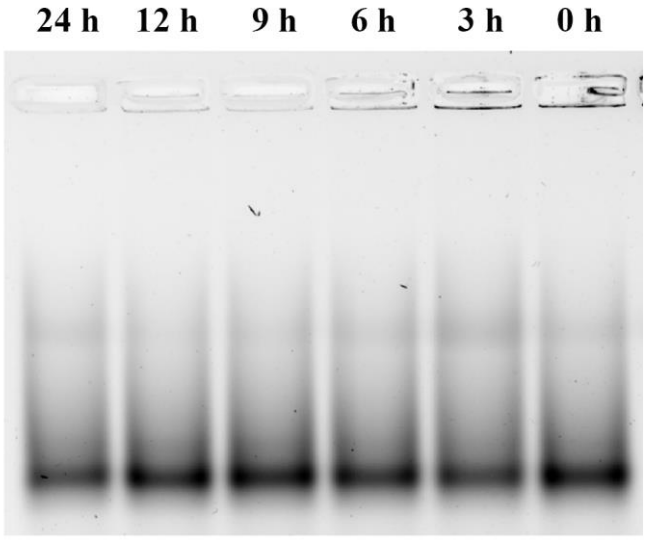

Figure S4. Electrophoretic analysis of the stability of DNA nanoprobe treated with fetal bovine serum. The DNA nanoprobe (250 nM) was incubated with 10% fetal bovine serum (FBS, v:v) at 37 ℃ for 0-24 h and then analyzed with 1% agarose gel electrophoresis.

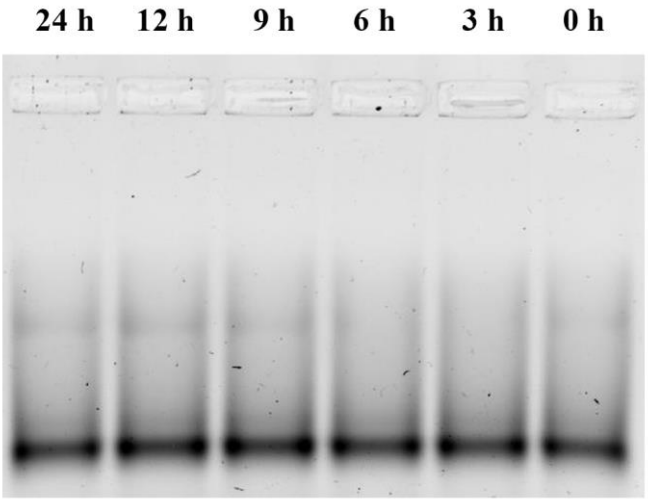

Figure S5. Electrophoretic analysis of the stability of DNA nanoprobe treated with cell lysate. The DNA nanoprobe (250 nM) was incubated with MCF-7 cell lysate at 37 ℃ for 0-24 h and then analyzed with 1% agarose gel electrophoresis.

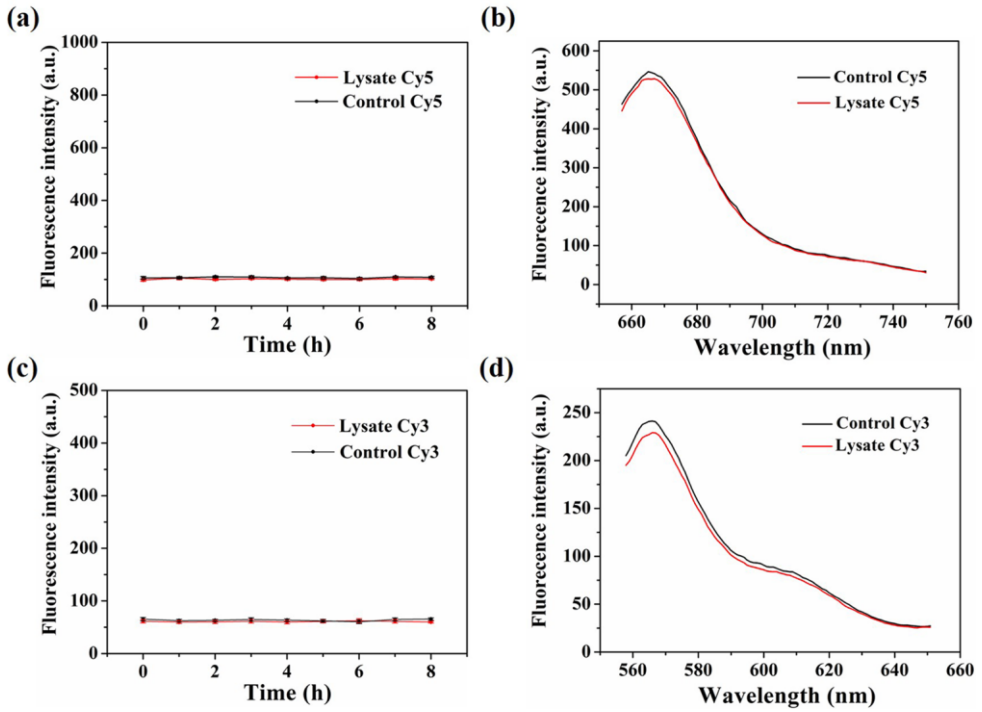

Figure S6. Stability of the DNA fluorescence nanoprobe treated with or without MCF-7 cell lysate. (a) and (c) exhibit the fluorescence changes of the nanoprobe treated with cell lysate (red trace) or without cell lysate (black trace) for 0-8 h. (b) and (d) show the fluorescence spectra of the nanoprobe by hybridizing with TK1 target (200 nM) and GalNAc-T target (200 nM) respectively, after treating with cell lysate for eight hours. TK1 target (Cy5), EX/EM: 645 nm/666 nm; GalNAc-T target (Cy3), EX/EM: 545 nm/566 nm.

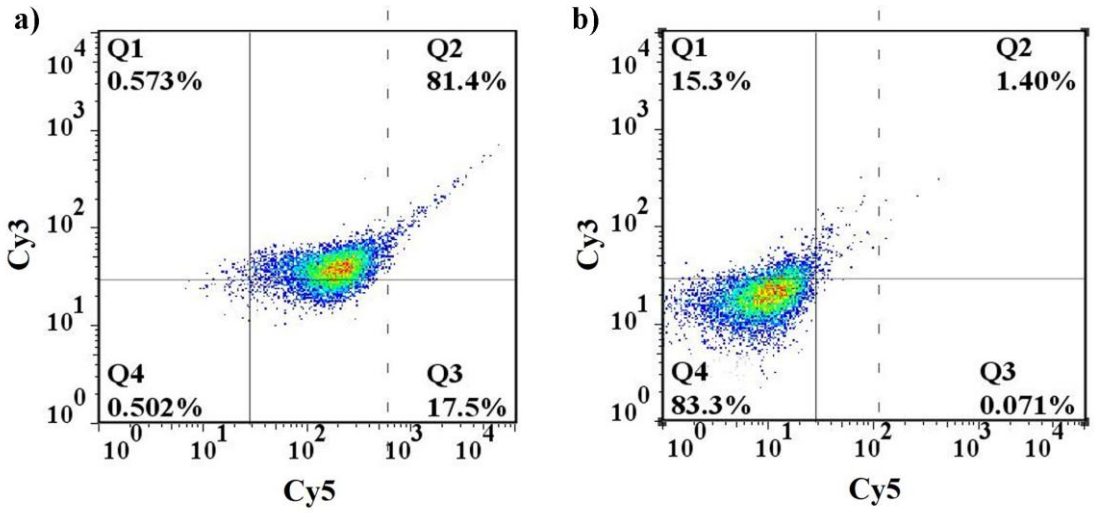

Figure S7. Simultaneous detection of TK1 and GalNAc-T mRNA by flow cytometry. (a) MCF-7 and (b) MCF-10A cells were incubated with the nanoprobe (100 nM) for 4 h at 37 ℃. Then they were analysed via flow cytometry after thoroughly washing with PBS.

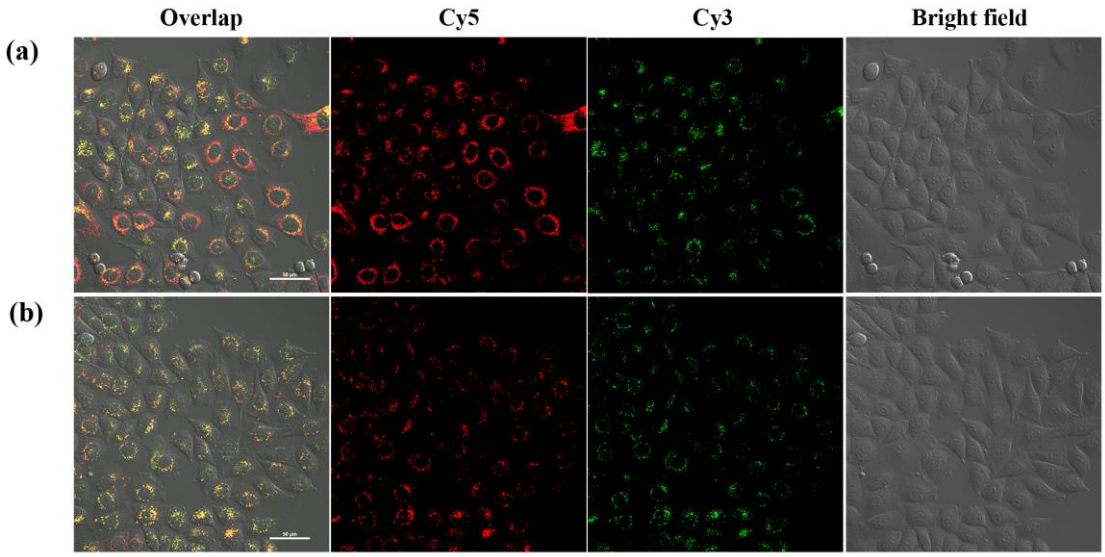

Figure S8. Targeting ability of DNA fluorescence nanoprobe. The MCF-7 cells were incubated with the nanoprobe (100 nM) modified with (a) or without (b) AS1411 aptamer for 4 h at 37 ℃. The two mRNAs were recorded by Cy5 with 640 nm excitation, and Cy3 with 561 nm excitation, respectively. Scale bar: 50 μm.

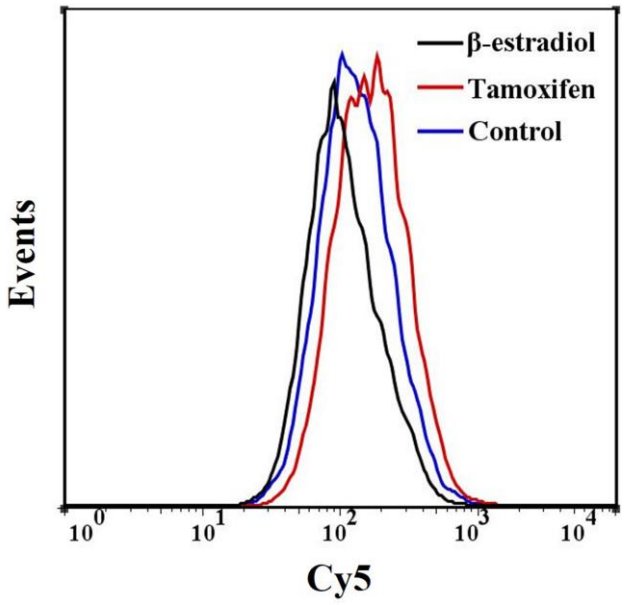

Figure S9. Detection of TK1 mRNA level in MCF-7 cell against varied drug stimuli via flow cytometry. The left group was treated with tamoxifen (10^{-6} M) for 48 h. The middle group was the control group, and the right group was treated with β-Estradiol (10^{-8} M) for 48 h. Then these groups were incubated with the nanoprobe (100 nM) for 4 h at 37 ℃ and analysed via flow cytometry after thoroughly washing with PBS.

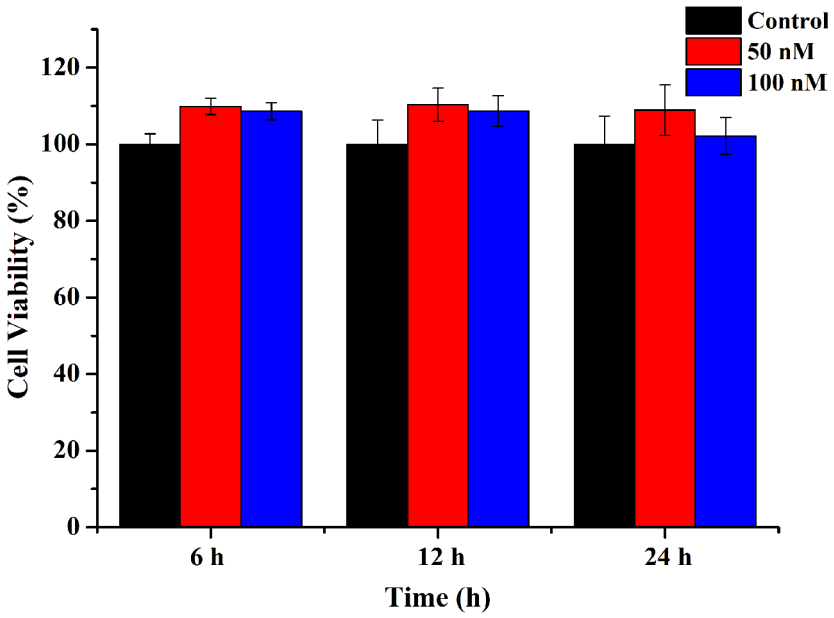

Figure S10. CCK-8 Assay. MCF-7 cells were incubated with nanoprobe (50 nM and 100 nM) for 6 h, 12 h and 24 h. Black bar stands for the control group (0 nM); Red bar stands for the nanoprobe (50 nM); Blue bar stands for the nanoprobe (100 nM).