Supporting Information

Highly sensitive and specific detection of tumor cells based on split aptamer-triggered dual hybridization chain reaction

Baoyin Yuan [a,b,c], Linyan Guo [a], Kai Yin [a], Xinyu Wang [a], Qian Liu [a], Meimei He [a], Kangdong Liu [a,b,c,d,e,*] and Jimin Zhao [a,b,c,*]

[a] Department of Pathophysiology, School of Basic Medical Sciences, Zhengzhou University, Zhengzhou, Henan, China

[b] Henan Provincial Cooperative Innovation Center for Cancer Chemoprevention, Zhengzhou, Henan, China

[c] State Key Laboratory of Esophageal Cancer Prevention and Treatment, Zhengzhou University, Zhengzhou, Henan, China

[d] China-US Hormel (Henan) Cancer Institute, Zhengzhou, Henan, China

[e] Cancer Chemoprevention International Collaboration Laboratory, Zhengzhou, Henan, China

* To whom correspondence should be addressed:

E-mail: kdliu@zzu.edu.cn; zhaojimin@zzu.edu.cn

Table S1 Sequences of all DNA used in this work. [a]

Name	Sequence (5'-3')
SA1	ACTGATTTGCTTGAGCTGAAGATCGTACCGTGAACTAGACAGT
SA2	ACTGTCTAGTAGCAAATCAGT
ZY11	TTGACTTGCCACTGACTACCTGGCGCATTGACGTCAGGTTGAGCTGAAG ATCGTACCGTGAAGTCAGTCGGTCGTCATC
T-SA2	GACCCTAAGCATACATCGTCCTTCATTTTTTACTGTCTAGTAGCAAATCAGT
T-SA2-T	GACCCTAAGCATACATCGTCCTTCATTTTTTACTGTCTAGTAGCAAATCAGTT TTTTTACTTCCTGCTACATACGAATCCCAG
H1	ATGAAGGACGATGTATGCTTAGGGTCGACTTCCATAGACCCTAAGCATACAT
H2	GACCCTAAGCATACATCGTCCTTCATATGTATGCTTAGGGTCTATGGAAGTC
H3	TACATACGAATCCCAGATACCTTCAGCTGGGATTCGTATGTAGCAGGAAGTA
H4	CTGAAGGTATCTGGGATTCGTATGTATACTTCCTGCTACATACGAATCCCAG
Random	N (64nt)

[a] The trigger sequences are in blue.

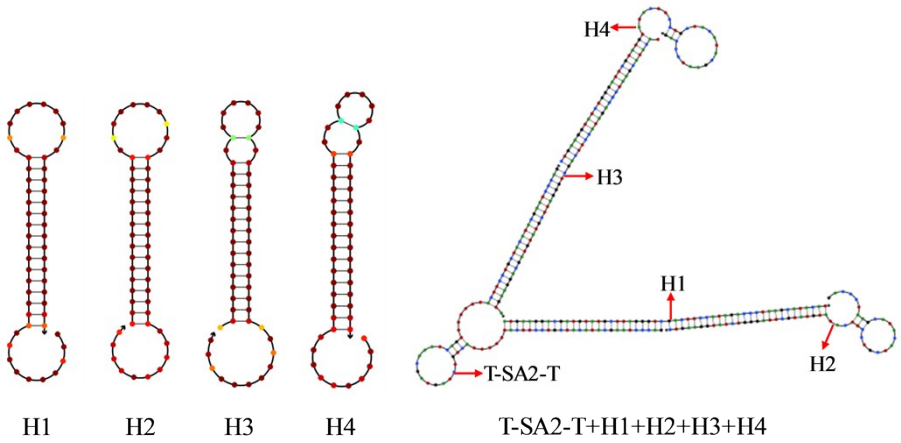

Fig. S1 Structures of H1, H2, H3, H4 and the dual HCR polymerization initiated by T-SA2-T simulated by the NUPACK software under the experimental conditions.

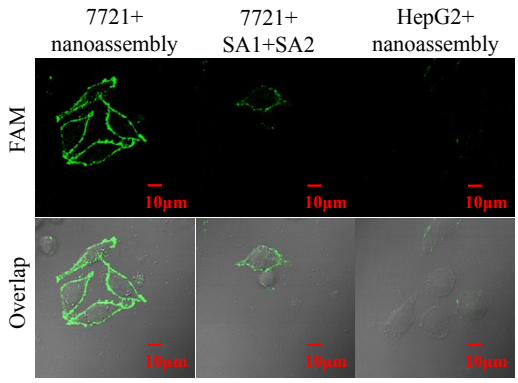

Fig. S2 Confocal imaging of target 7721 cells incubated with the nanoassembly and SA1+SA2, control HepG2 cells incubated with the nanoassembly. The upper are FAM fluorescence images, the lower are the overlays of the fluorescence channel and the bright-field channel. The fluorescence signal was collected by a 100× objective (fluorescence channel: EX 488 nm, EM 505 nm long-pass). The scale bar is 10 μm.

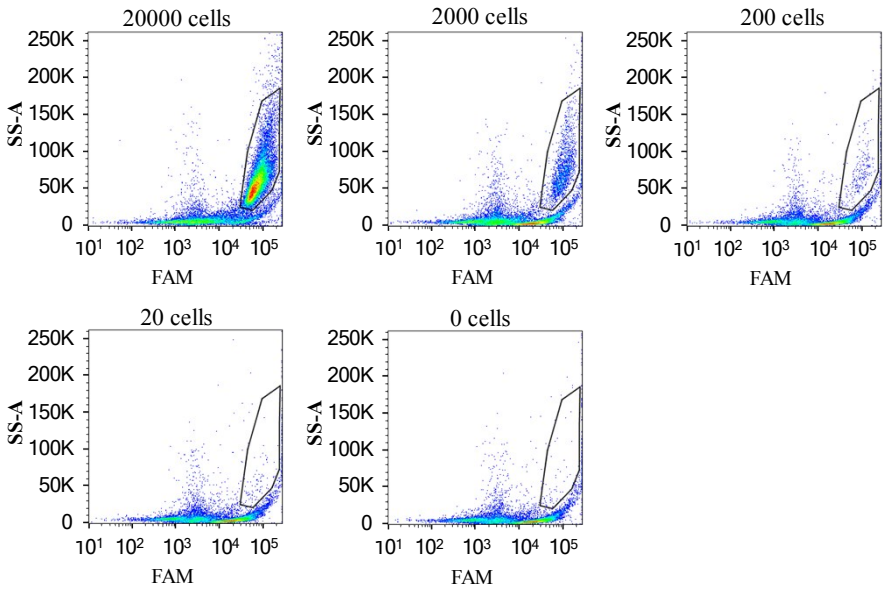

Fig. S3 Flow cytometry assays of 7721 cells with cell number ranging from 0 to 20,000 in 200 μL binding buffer containing 50% human serum by using the dual-HCR strategy. 7721 cells appeared in black frame, and serum fragments located outside black frame.

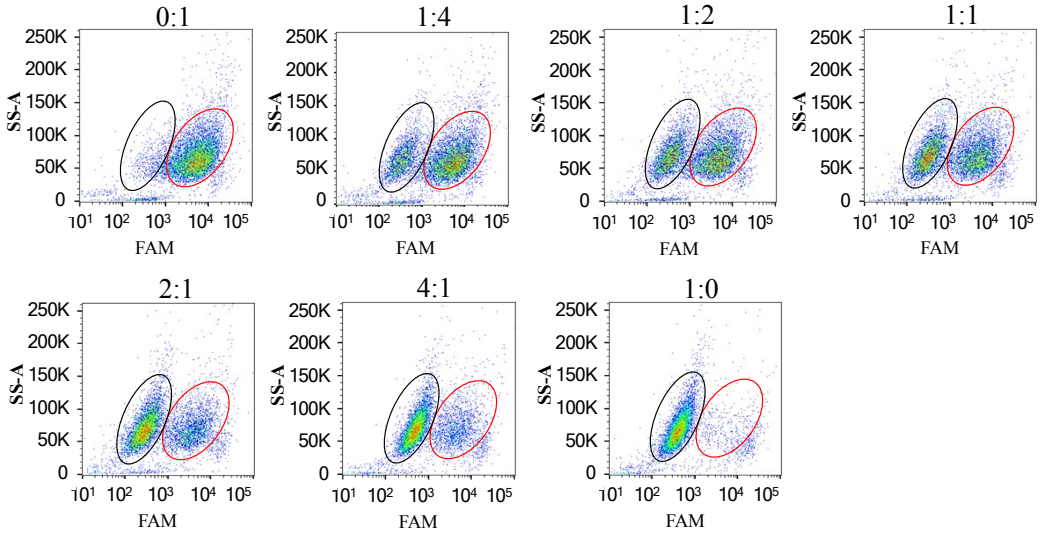

Fig. S4 Flow cytometry scatter plots of cell mixtures incubated with the nanoassembly. The ratios of control 7404 cells to target 7721 cells were 0:1, 1:4, 1:2, 1:1, 2:1, 4:1 and 1:0. The total number of cells was 100,000 and the final volume was 200 μL in each sample.