Supporting Information

Real-Time Monitoring of Enzyme-Free Strand Displacement Cascade by Colorimetric Assays

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Content

1. Sequences

Table S1. The first set of oligonucleotides' sequences:

Name	Sequence (from 5' to 3')
C1	AACCACCAAACTTAT CTCT CCAAACAAAACCTAT
C2	AGAG ATAAGTTTGGTGGTT AGAG
C3	CCTAACACAATCACT CTCT AACCACCAAACTTAT
C4	CCACAAAACAAAACT CTCT AACCACCAAACTTAT
C3-TAMRA	CCTAACACAATCACT CTCT AACCACCAAACTTAT-**(TAMRA)**
C2-DABCL	AGAG A-**(DABCYL)** TAAGTTTGGTGGTT AGAG
blue GNP1	HS-TTTTATAAGTTTGGTGGTTAGAGAGTGATTGTGTTAGG
blue GNP1 poly15	HS-TTTTTTTTTTTTTTTATAAGTTTGGTGGTTAGAGAGTGATTGTGTTAGG
blue GNP1 poly25	HS-TTTTTTTTTTTTTTTTTTTTTTTTTTATAAGTTTGGTGGTTAGAGAGTGATTGTGTTAGG
blue GNP2	HS-TTTTCCTAACACAATCACTCTCTAACCA
blue helper	HS-TTTTA
SNP5	AACCCCCAAACTTAT CTCT CCAAACAAAACCTAT
SNP6	AACCATCAAACTTAT CTCT CCAAACAAAACCTAT
SNP7	AACCACAAAACTTAT CTCT CCAAACAAAACCTAT
SNP8	AACCACCCAACTTAT CTCT CCAAACAAAACCTAT
SNP9	AACCACCACACTTAT CTCT CCAAACAAAACCTAT
SNP10	AACCACCAATCTTAT CTCT CCAAACAAAACCTAT
SNP11	AACCACCAAATTTAT CTCT CCAAACAAAACCTAT
SNP12	AACCACCAAACATAT CTCT CCAAACAAAACCTAT
SNP13	AACCACCAAACTCAT CTCT CCAAACAAAACCTAT
SNP14	AACCACCAAACTTCT CTCT CCAAACAAAACCTAT
SNP15	AACCACCAAACTTAA CTCT CCAAACAAAACCTAT
SNP16	AACCACCAAACTTAT ATCT CCAAACAAAACCTAT
SNP17	AACCACCAAACTTAT CCCT CCAAACAAAACCTAT
SNP18	AACCACCAAACTTAT CTTT CCAAACAAAACCTAT
SNP22	AACCACCAAACTTAT CTCT CCCAACAAAACCTAT
SNP27	AACCACCAAACTTAT CTCT CCAAACATAACCTAT

Table S2. The second set of oligonucleotides' sequences:

Name	Sequence (from 5' to 3')
2-C1	CAACTCTTTAAA CTCA TACTAAACAAAACCC TTAAATT
2-C2	TGAG GGGTTTTGTTTAGTA TGAG
2-C3	TACTAAACAAAACCC CTCA CATCTTCTAACATCA
2-C4	TACTAAACAAAACCC CTCA CACACTATAATTCCA
2-blue GNP1	HS-TTTTTGATGTTAGAAGATGTGAGGGGTTTTGTTTAGTA
2-blue GNP2	HS-TTTTTACTAAACAAAACCCCTCACATCT
blue helper	HS-TTTTA
2-SNP4	CAACTCTTTAAA CTCC TACTAAACAAAACCCTTAAATT
2-SNP5	CAACTCTTTAAA CTCA CACTAAACAAAACCCTTAAATT
2-SNP8	CAACTCTTTAAA CTCA TACCAAACAAAACCCTTAAATT
2-SNP10	CAACTCTTTAAA CTCA TACTACACAAAACCCTTAAATT
2-SNP13	CAACTCTTTAAA CTCA TACTAAACCAAACCCTTAAATT

2. Materials

Ultrapure water with 18.2 MΩ cm (Heal Force) is used in all experiments. All of the chemical reagents are of analytic grade and are used without further purification. The 10 nm gold nanoparticles, Tris(2-carboxyethyl) phosphine hydrochloride (TCEP) are purchased from Sigma. All the DNA oligonucleotides purified via HPLC are synthesized by TaKaRa Bio Inc. (Dalian, China) DNase I is purchased from TaKaRa Bio Inc. (Dalian, China)

3. Reversible assembly (Figure S1)

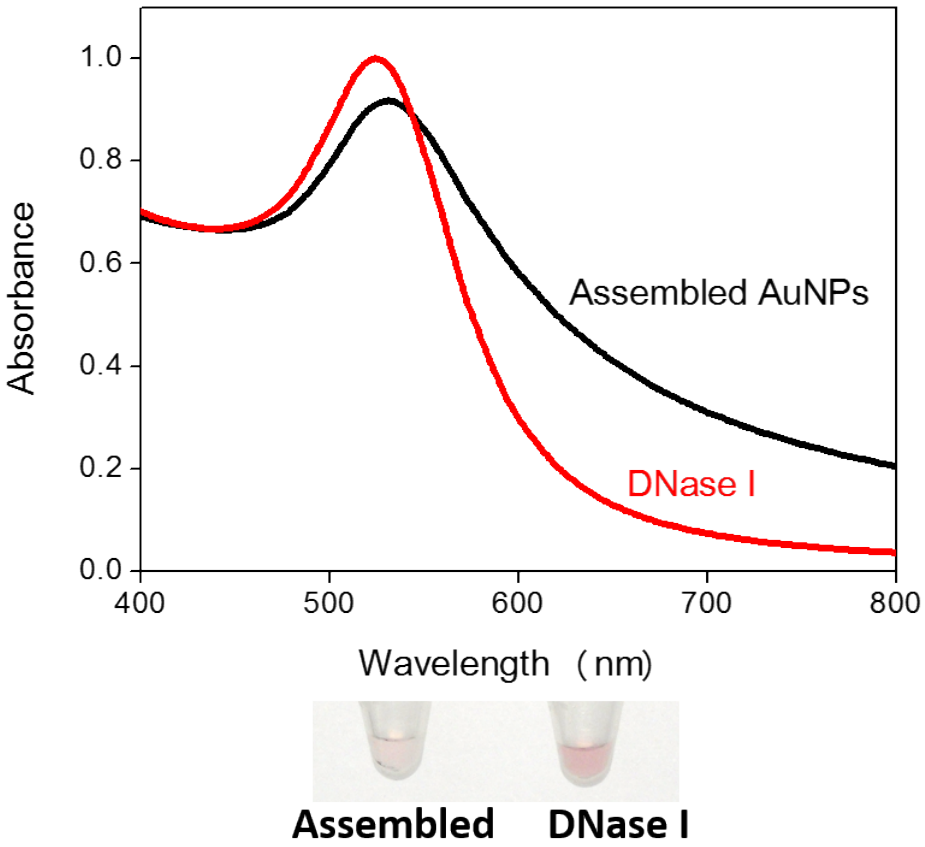

Figure S1. (a) UV-vis spectra of aggregated AuNPs and disassembled AuNPs by DNase I. The reaction time is 1 h at 37°C. After adding DNase I, the wavelength of absorbance peak decreases, proving the disassembly of AuNPs. (b) Corresponding bar graphs and photographs of AuNPs before and after adding DNase I.

4. The effects of the number of polyT (Figure S2)

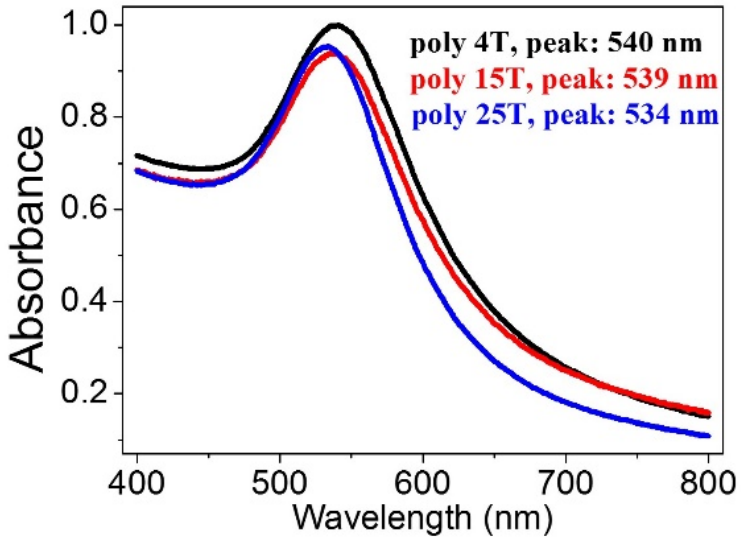

Figure S2. The effects of the number of poly T of the effective oligonucleotides on making AuNPs aggregate. The absorbance peaks of aggregated AuNPs modified with 4-base, 15-base and 25-base poly T are 540 nm, 539 nm and 534 nm respectively.

5. Average diameter (Figure S3)

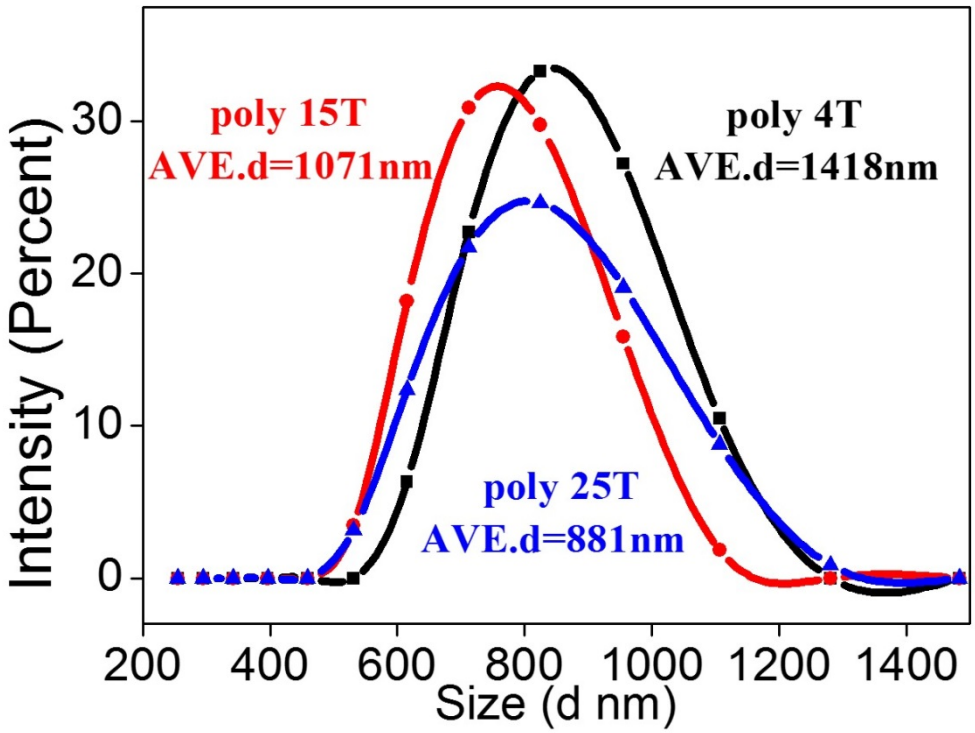

Figure S3. The scattering data shows the average diameter of each kind of AuNPs after annealing. The average diameters of aggregated AuNPs modified with 4-base, 15-base and 25-base poly T are 1418 nm, 1071 nm and 881 nm respectively.

6. Not producing ‘false mismatch signal’ (Figure S4)

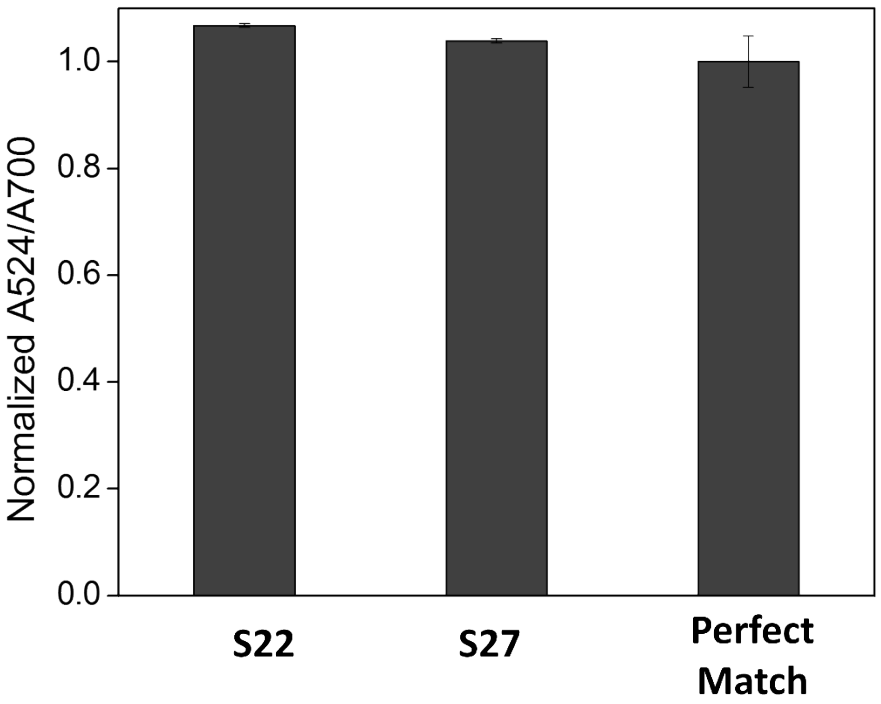

Figure S4. The values of A524/A700 of mismatch positions belonging to the part not participating hybridization are close to that of perfect match sequence, indicating the part not participating hybridization will not produce ‘false mismatch signal’.

7. Another set of mismatch experiment (Figure S5)

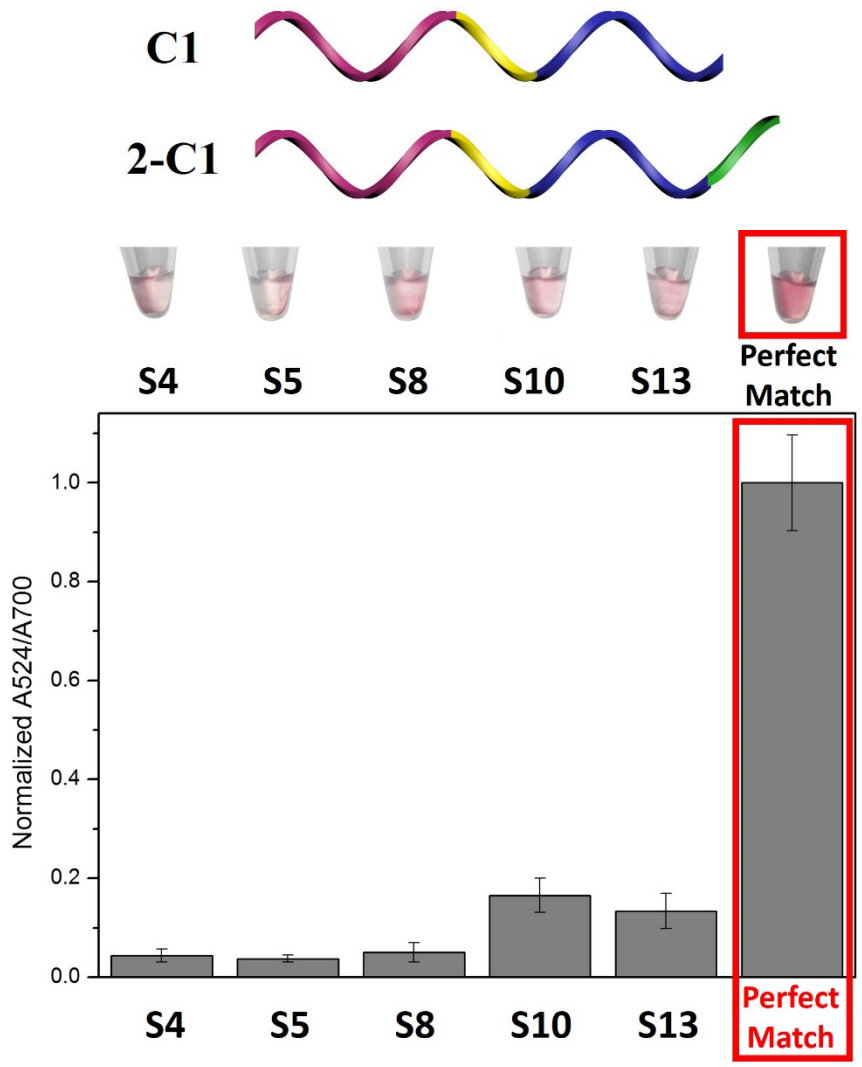

Figure S5. (Upper) Compared with **C1**, **2-C1** has two hang out parts (purple and green) on each side of hybridization part (yellow and blue). (Bottom) Another set of sequences for single-base mismatch detection also work well. The bar graph shows the value of A524/A700 of each sample.

8. References

1. Qian, L. & Winfree, E. Scaling Up Digital Circuit Computation with DNA Strand Displacement Cascades. *Science* **332**, 1196-1201 (2011).

2. Song, S.P. et al. Gold-Nanoparticle-Based Multicolor Nanobeacons for Sequence-Specific DNA Analysis. *Angew. Chem. Int. Ed.* **48**, 8670-8674 (2009).