Electronic Supplementary Material (ESI) for Chemical Communications

Visible multi-digit DNA keypad lock based on split G-quadruplex DNAzyme and silver microspheres

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Materials

DNA strands were all purchased from Sangon Biotechnology Co., Ltd (Shanghai, China) and their sequences were listed in Table S1. $AgNO_3$, hydroxylamine (50 wt.% solution in water), hemin and TMB were purchased from Sigma-Aldrich (St. Louis, MO, USA). Other chemicals were of reagent grade and were used without further purification. The oligonucleotide was dissolved in water as stock solution and quantified by UV−vis absorption spectroscopy with the following extinction coefficients (ε260 nm, M-1cm-1): A = 15400, G = 11500, C = 7400, T = 8700. UV-vis absorbance measurements were performed on a Cary 50 Scan UV/vis/NIR Spectrophotometer (Varian, USA). The DNA solutions were heated at 88°C for 10 min to dissociate any intermolecular interaction and slowly cooled down to room temperature before use.

Synthesis of SMSs and DNA modification

The SMSs without surfactant protection were synthesized according to our previous reports with some slight modification.[1] 1 mL of 0.495 M $AgNO_3$ was added into 1 mL of a 1.98 M NH_2OH aqueous solution, which lead to a large amount of precipitate in several seconds. After oscillation for 5 min, it was incubated under 4°C for overnight. Then the as-prepared SMSs were washed with distilled water for three times and dispersed in 1.57 mL of water with a final concentration of 34 mg/mL. The large metal microspheres sank to the bottom of the vessel in seconds after oscillation as shown in Figure S4a. The corresponding SEM image was taken by a S4800 scanning electron microscope (Hitachi, Japan) and shown in Figure S4b. For DNA modification, 35 μL of 20 μM DNA strand S solution was added into 20 μL of the as-prepared SMSs. The mixture was oscillated and incubated at room temperature for 2 h. After washed with TAE buffer (40 mM Tris-HAc, 1mM EDTA, 12.5 mM $MgAc_2$, 100 mM NaAc, 20 mM KAc, pH 8.0) for three times, the resulting product was incubated with 30 μL of 1 mM 6-mercapto-1-hexanol (MCH) at room temperature for 30 min to block the surface of the SMSs. Then the DNA modified SMSs were washed with TAE buffer for three times and dispersed in 100 μL TAE buffer at 4°C for further use.

Operation of DNA keypad locks

For each permutation of the DNA keypad locks, 10 μL of the DNA modified SMSs was used. The first 10 μL of 20 μM input DNA was added and incubated for 1h. After washing for three times by TAE buffer, the second input DNA was added in the same amount and incubated for the

same time. This process was repeated until all the inputs had been added. The absorbance value increased with the concentration of input strands (Figure S5). To make the change of color easily recognized by naked eyes, 20 μM was chosen as the input concentration. The precipitate was dispersed in 10 μL TAE buffer and mixed with 10 μL of 2 μM hemin dissolved in TAE buffer for 40 min for each permutation. In the catalytic reaction, the 20 μL final mixture solution was mixed with 480 μL TMB-H_2O_2 substrate solution, which was constituted of 5 μL 0.5% (w/v) TMB, 10 μL 30% (w/v) H_2O_2 and 465 μL substrate buffer (26.6 mM citrate, 51.4 mM disodium hydrogen phosphate, pH 5.0). Ten minutes later, UV-vis spectra were collected and the photo was taken.

PAGE

The 10 μL of 2 μM DNA solution mixed with 2 μL of 6 × loading buffer (25 mM HEPES, pH 8.0, 20 mM KCl, 200 mM NaCl, 50% glycerol, 0.25% bromphenol blue) were analyzed in 15% native polyacrylamide gel. The electrophoresis was conducted in 1 × TBE (pH 8.0) at constant voltage of 110V for 1.5 hour. The gels were scanned by a UV transilluminator after staining with ethidium bromide.

EIS measurement

EIS measurements were conducted on a Zahner Zennium electrochemical workstation (Kronach, Germam). The gold electrode (1.2 mm in diameter), Ag–AgCl electrode (saturated KCl) and Pt coil were used as working electrode, reference electrode and counter electrode, respectively. Before use, the gold electrode was polished with 1.0 μm, 0.3 μm α-Al_2O_3 and then washed ultrasonically in pure water for 3 times successively, followed by electrochemically cleaning in 0.1 M H_2SO_4 until a reproducible cyclic voltammetry was obtained.[2] Each step of modification and hybridization was done by immersing the electrode into the TAE buffer containing 20 μM input DNA for 1h, followed by rinsing with pure water and TAE buffer. EIS measurements were performed under an oscillation potential of 5 mV over the frequency range of 10 KHz–0.1 Hz and in the solution of 5 mM $K_4[Fe(CN)_6]$/$K_3[Fe(CN)_6]$ in 10 mM Tris–HCl buffer, pH 7.4. All the measurements were carried out at room temperature.

References

[1] (a) X. Yang, X. Sun, Z. Lv, W. Guo, Y. Du, E. Wang, *Chem. Commun.*, 2010, **46**, 8818; (b) X. Yang, Y. Du, D. Li, Z. Lv, E. Wang, *Chem. Commun.*, 2011, **47**, 10581.
[2] (a) B. Li, Y. Du, H. Wei, S. Dong, *Chem. Commun.*, 2007, 3780; (b) Y. Du, B. L. Li, F. Wang, S. J. Dong, *Biosens. Bioelectron.*, 2009, **24**, 1979.

Table S1.* Sequences of the oligonucleotides used in this work. The sequences are colored in the same way as in Schemes.

Name	Sequence
S	5'-HS(CH_2)$_6$GCTCCTGGACCC-3'
A	5'-CGTCAGTCACTAGTCT GGGTCCAGGAGC-3'
B	5'-AGACTAGTGACTGACG CATACATCTC TGGG-3'
C	5'-GGGTGGGTGGGT GAGATGTATG-3'
a	5'-TAGTCTGTCAAT GGGTCCAGGAGC-3'
b	5'-ATTGACAGACTA GTGCGTAGATGT-3'
c	5'-CGTCAGTCACTAGTCT ACATCTACGCAC-3'
d	5'-AGACTAGTGACTGACG CATACATCTC TGGG-3'
e	5'-GGGTGGGTGGGT GAGATGTATG-3'
A2M	5'-CGTCAGTCACTAGTCT GGCTCGAGGAGC-3'
A4M	5'-CGTCAGTCAGTACTCT GGCTCGAGGAGC-3'

*To save reagent and simplify design, we have repeatedly used the same DNA strands as different inputs. The mismatched bases are underlined.

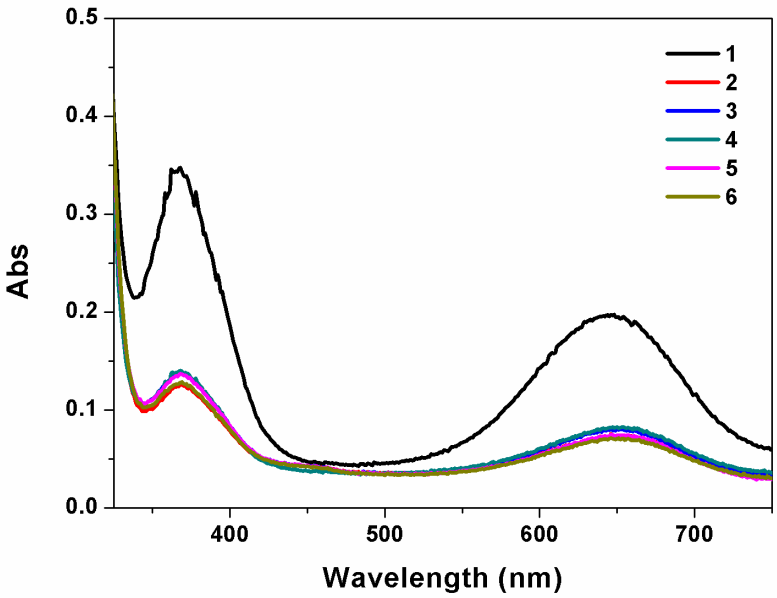

Figure S1. UV-vis absorbance spectra of the six permutations of the three-digit keypad lock. Input sequence of the DNA strands in the different permutations: Permutation 1, ABC; Permutation 2, ACB; Permutation 3, BAC; Permutation 4, BCA; Permutation 5, CAB; Permutation 6, CBA.

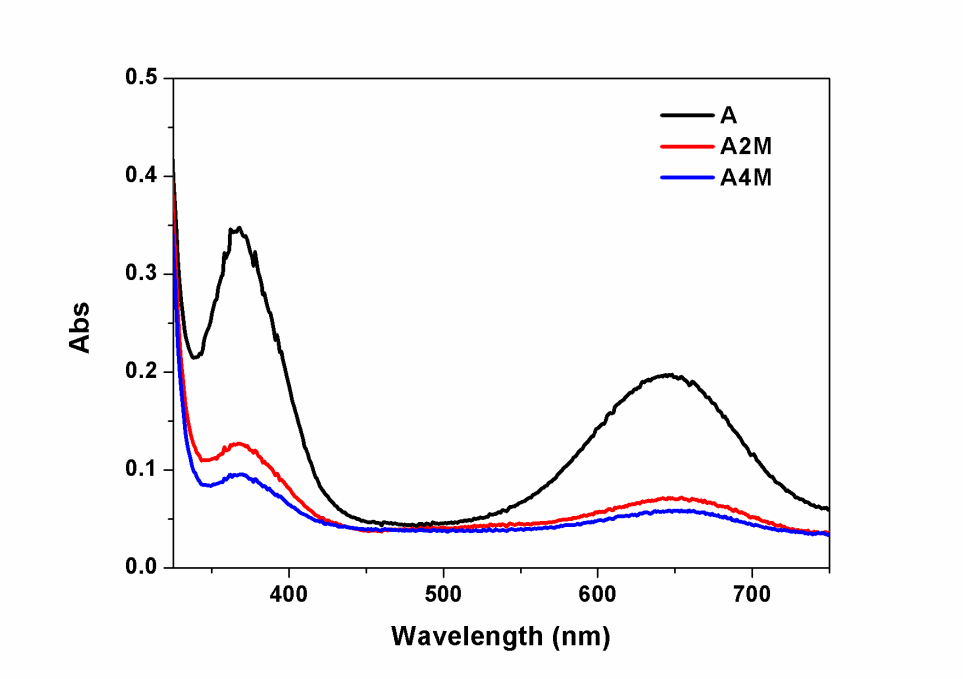

Figure S2. UV-vis absorbance spectra of Permutation 1 of the three-digit keypad lock. Curve A is the spectra of the Permutation 1 (ABC). Strand A was replaced with strand A2M and A4M in curve A2M and A4M, respectively.

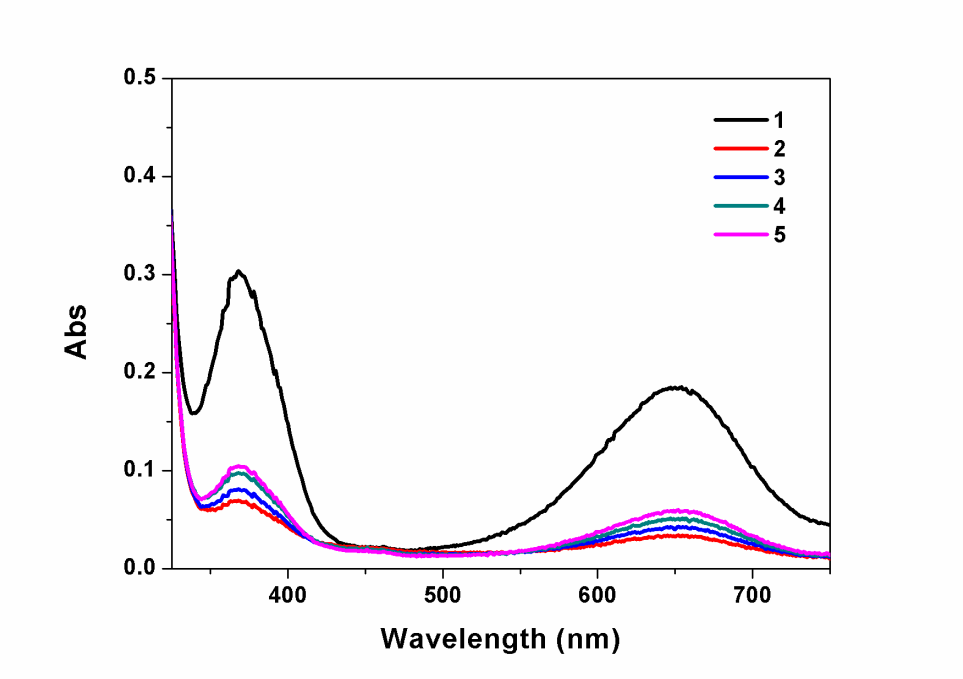

Figure S3. UV-vis absorbance spectra of the five permutations of the five-digit keypad lock. Input sequence of the DNA strands in the different permutations: Permutation 1, abcde; Permutation 2, abced; Permutation 3, abdce; Permutation 4, acbde; Permutation 5, bacde.

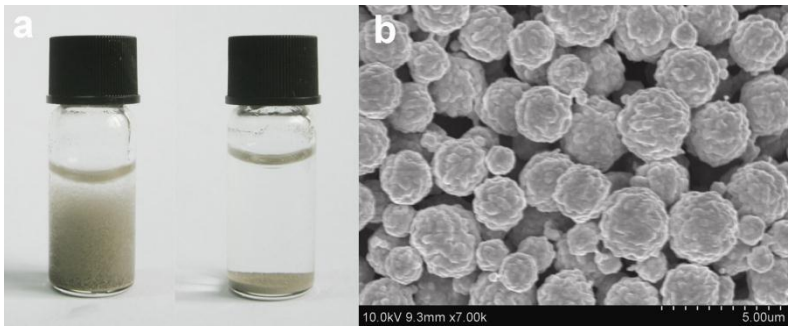

Figure S4. (a) Photos of SMSs after being oscillated (left) and standing for 20 s later (right). (b) SEM image of SMSs.

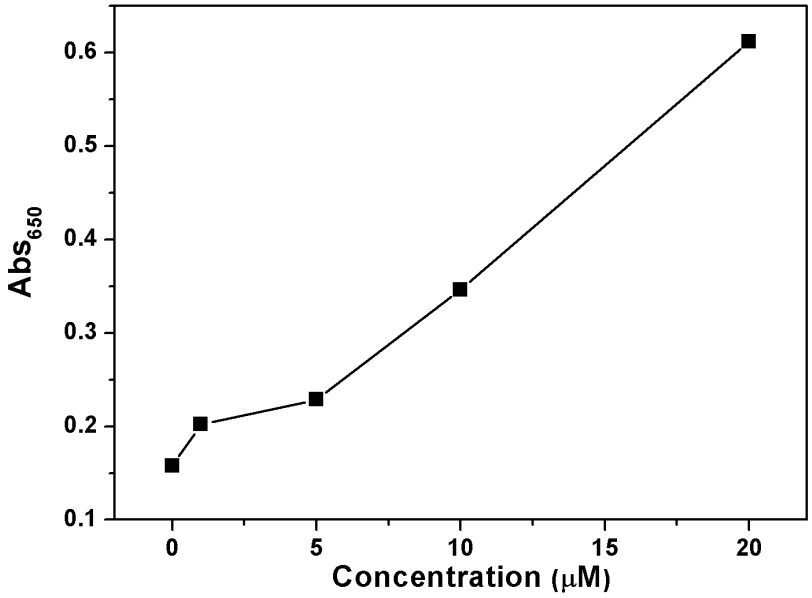

Figure S5. UV-vis absorbance at 650 nm of the Permutation 1 (ABC) of the three-digit keypad lock using the input DNA strands in different concentrations.