Electronic Supplementary Material (ESI) for RSC Advances.

Supporting Information

Controllable DNA nanodevices regulated by logic gates for multi-stimulus recognition

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S1: DNA sequences

The length and sequence(strand A,B,M,F,Q) in the sensing module of the nanodevices were adopted from reference [1, 2]. The length and sequence of other strands were designed and analyzed by NUPACK[3] to decrease the unexpected secondary structure and minimize crosstalk between strands.

Table S1. Sequences of the Oligonucleotides (5’ to 3’)

Name	Sequence (5′→3′)	Length (nt)
A	FAM-TCATGTTTGTTTGTTGGCCCCCCTTCTTTCTTA	33
B	ACAAACATGA-BHQ	10
L6	GTGAGATAAGAAAGAAGGGGGGCCAACAA	29
L7	CGCGCAATAAGAAAGAAGGGGGGCCAACAA	30
L8	TAAGTGCATAAGAAAGAAGGGGGGCCAACAA	31
L9	CCAGCACAATAAGAAAGAAGGGGGGCCAACAA	32
K	TTGTTGGCCCCCCTTCTTTCTTATGCACTTA	31
G1	TAAGAAAGAAGGGGGGCCAACAAGTCCGG	29
I1	CCGGACTTGTTGGCCCCCCTTCTTTCTTA	29
G2	CGCCCATAAGTGCATAAGGGGGGCCAACAAGTCCGG	36
I2	TTGTTGGCCCCCCTTATGCACTTATGGGCG	30
I3	CCGGACTTGTTGGCCCCCCTTATGCACTTA	30
G3	TAAGTGCATAAGGGGGGCCAACAAGCGCGTGCCGATATCG	40
T	AGTCAGATTACGATATCGGCACGCGC	26
I4	GCGCGTGCCGATATCGTAATCTGACT	26
I5	ACGCGCTTGTTGGCCCCCCTTATGCACTTA	30
M	CCTGCCACGCTCCGCTCACTGACCTGGGGGAGTATTGCGGAGGAAGGT	48

F	FAM-GCGGAGCGTGGCAGG	15
Q	CCCAGGTCAGTG-BHQ	12
L1	CCATCTACCTTCCTCCGCAATACTCC	26
K1	GGAGTATTGCGGAGGAAGGTAGATGG	26

S2: Optimization for repressing the sensor

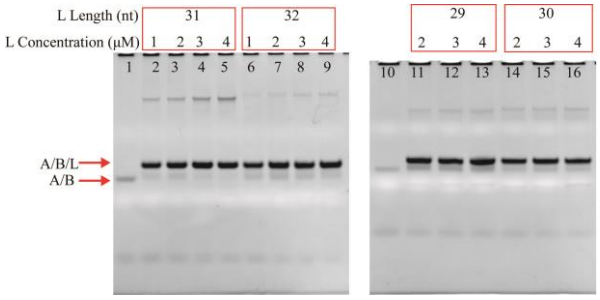

Figure S1. PAGE gel analysis of different lengths and concentrations of strand 'L'. Lane 1 and lane 10: (A/B); Lane 2-5: the sensor (A/B) with addition of different concentrations of 'L8' (31nt) as 1μM, 2μM, 3μM and 4μM; Lane 6-9: the sensor (A/B) with addition of different concentrations of 'L9' (32nt) as 1μM, 2μM, 3μM and 4μM; Lane 11-13: the sensor (A/B) with addition of different concentrations of 'L6' (29nt) as 2μM, 3μM and 4μM; Lane 14-16: the sensor (A/B) with addition of different concentrations of 'L7' (30nt) as 2μM, 3μM and 4μM. [A]=1μM and [B] =4μM.

To obtain better performance, the control experiments of PAGE analysis were carried out to choose the appropriate length and concentration of lock strand 'L' (Figure S1). A series of different lengths of strand 'L' and different concentrations of each length were introduced to suppress the sensor as 29-nt to 32-nt (Lanes 11-13, 14 -16, 2-5, 6-9). It is noted that although some unexpected secondary structure was produced, the remained complex (A/B) was the least when the length of 'L' was 31-nt and the corresponding concentration was 4 times higher than the sensor in lane 5. Therefore 31-nt strand 'L8' was chosen as the lock strand to avoid unexpected background signals.

S3: Upstream 'AND' logic gate for actuation of the Hg^{2+} sensor

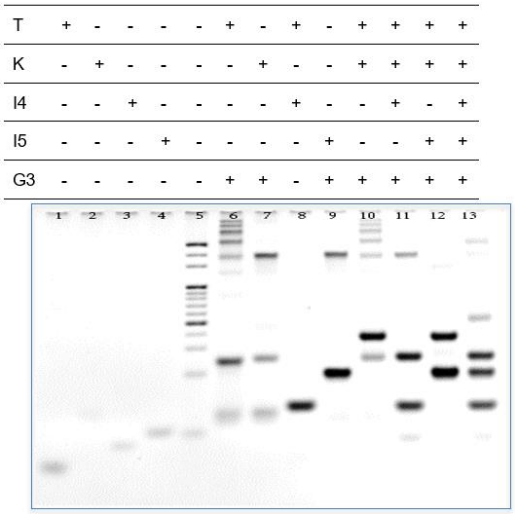

Figure S2. PGAE analysis of the components of 'AND' logic gate and operation upon addition of inputs. Lane 1: T, Lane 2: K, Lane 3: I4, Lane 4: I5, Lane 5: 20-bp DNA ladder, Lane 6: (G3/T), Lane 7: (G3/K), Lane 8: (T/I4), Lane 9: (I5/G3), Lane 10: (T/G3/K), Lane 11: (T/G3/K)+I4, Lane 12: (T/G3/K)+I5, Lane 13: (T/G3/K) +I4+I5.

As shown in Figure S2, gel results confirmed that only both inputs 'I4' and 'I5' were present strand 'K' could be released. Specifically, in lane 11, the band of the complex (T/G3/K) disappeared and yielded the waste (T/I4) and (G3/K) upon addition of 'I4'; in lane 12, the band of the complex (T/G3/K) did not disappear. However, in lane 13, when both 'I4' and 'I5' were added, the band of the complex (T/G3/K) disappeared and yielded the waste (T/I4) and (I5/G3). It is noted that the DNA complex used in this gate had some unexpected secondary structure, but this did not influence the downstream experiment.

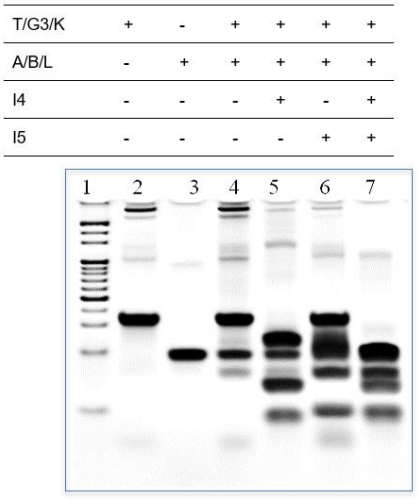

Figure S3. PGAE analysis of the mixture of (T/G3/K) and (A/B/L) upon addition of inputs. Lane 1: 20bp ladder, Lane 2: (T/G3/K), Lane 3: (A/B/L), Lane 4: (T/G3/K)+ (A/B/L), Lane 5: (T/G3/K)+ (A/B/L)+I4, Lane 6: (T/G3/K)+ (A/B/L)+I5, Lane 7: (T/G3/K)+ (A/B/L)+I4+I5.

Further gel analysis shown in Figure S3 confirmed that both inputs ‘I4’ and ‘I5’ could activate the sensor. In lane 6, the band of gate complex (T/G3/K) and the locked sensor (A/B/L) disappeared and yielded the waste upon the addition of both inputs.

S4: NUPACK and Visual DSD simulation results

The state change of the nanodevice in this work utilized the strand displacement reaction between the lock strand and key strand. The sequence of each strand was specially designed according to the hybridization and displacement reaction. Thus, the sequence of a randomly designed oligonucleotide didn’t satisfy the above requirement and couldn’t react with the strands in the nanodevice. Take the reaction between the complex A/B/L and the key strand as an example, a random oligonucleotide 5’-AACTTCCATCCACCACCACCCAACTTAACCA-3’ was designed by NUPACK[3]. From the NUPACK simulation result in Figure S4, when the above randomly designed oligonucleotide mixed with strand A, B, L, K, it hardly affected the reaction product.

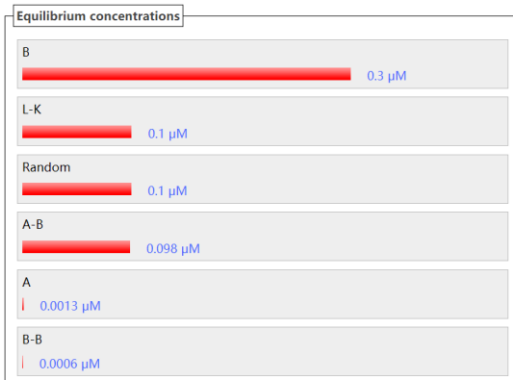

Figure S4. (a) NUPACK simulation results of the reaction of strand A, B, L, and K. [A] =0.1μM, [B] =0.4μM, [L] = 0.1μM , [K] = 0.1μM.

The results could also be confirm by Visual DSD[4]. As can be seen in Figure S5, when the complex ABL(A/B/L) was mixed with the key strand K, they reacted and the complex ABL (red curve) was exhausted quickly. Howerver, in Figure S6, when the complex ABL(A/B/L) was mixed with the random strand, they didn’t react and the complex ABL (red curve) was remained unchanged.

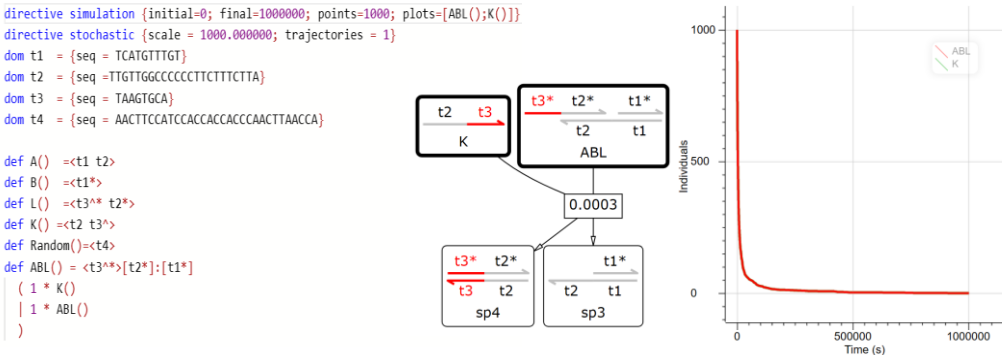

Figure S5. Visual DSD simulation results of the reaction between the complex ABL and the key strand K.

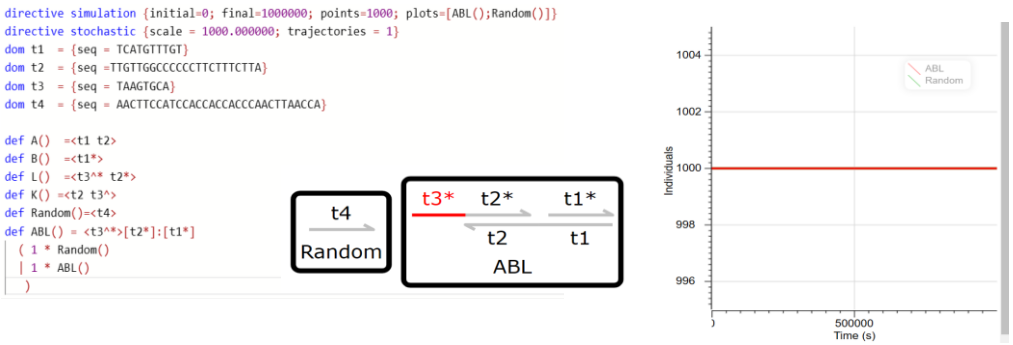

Figure S6. Visual DSD simulation results of the reaction between the complex ABL and the random strand.

References

1. Z. Wang, J. H. Lee and Y. Lu, *Chem. Commun.*, 2008, 6005-6007.
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3. J. N. Zadeh, C. D. Steenberg, J. S. Bois, B. R. Wolfe, M. B. Pierce, A. R. Khan, R. M. Dirks and N. A. Pierce, *J. Comput. Chem.*, 2011, **32**, 170-173.
4. M. R. Lakin, S. Youssef, F. Polo, S. Emmott and A. Phillips, *Bioinformatics*, 2011, **27**, 3211-3213.