Supplementary Information

Cascade Signal Amplification Strategy for Sensitive and Label-free DNA Detection Based on Exo III-catalyzed Recycling Coupled with Rolling Circle Amplification

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Results and discussion

The time-dependent fluorescence response under different conditions

To demonstrate the feasibility of our cascade amplification strategy, time-dependent fluorescence response under different conditions were carried out. As shown in Fig. S1, in the absence of target DNA, the change of fluorescence intensity was almost negligible in 1 h (cruve b). In contrast, in the present of 5 nM target, the cascade amplification assay could be performed and the fluorescence signal increased significantly (cruve a). Moreover, it could be seen that the RCA process played a key role in the generation of fluorescence signal. Because the intensity of fluorescence signal stayed largely constant without RCA (cruve c).

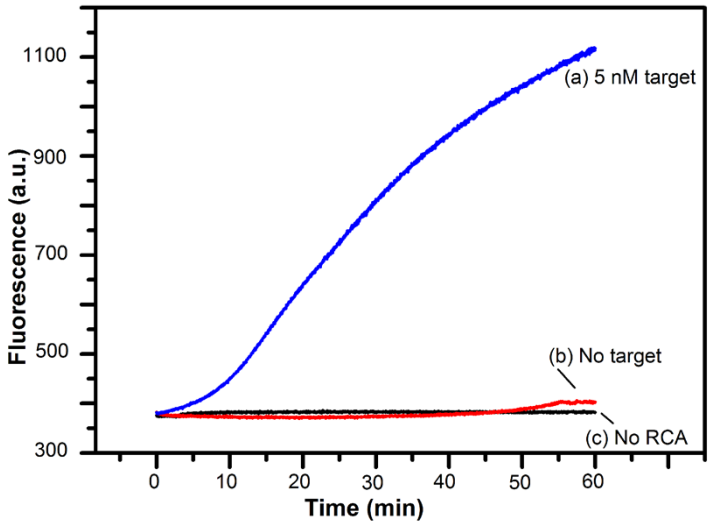

Fig. S1 The time-dependent fluorescence response under different conditions: in the present of 5 nM target DNA (a), in the absence of target DNA (b) and in the absence of RCA process (c). Other expermental conditions: 5.0×10^{-8} M of primer DNA, 2.0×10^{-7} M of circular DNA and 3.0×10^{-6} M of NMM.

The signal amplification capacity of our cascade assay

In order to demonstrate the signal amplification capacity of the cascade amplification strategy, we compared our strategy with the RCA process (Fig. S2). In the cascade amplification experment, three different concentrations of target DNA were chosen (50 pM, 0.5 nM and 5 nM) to initiate the Exo III recycling and followed by RCA. In the RCA experment, prime DNA, with the same concentration as target DNA, was captured directly by the MNB-probe to conduct the RCA process. Under all the three different target and primer DNA concentrations, the cascade amplification strategy produced significantly higher signals than one-step RCA process. These

results implyed that the presented strategy had a good signal amplification capacity for the detection of target DNA.

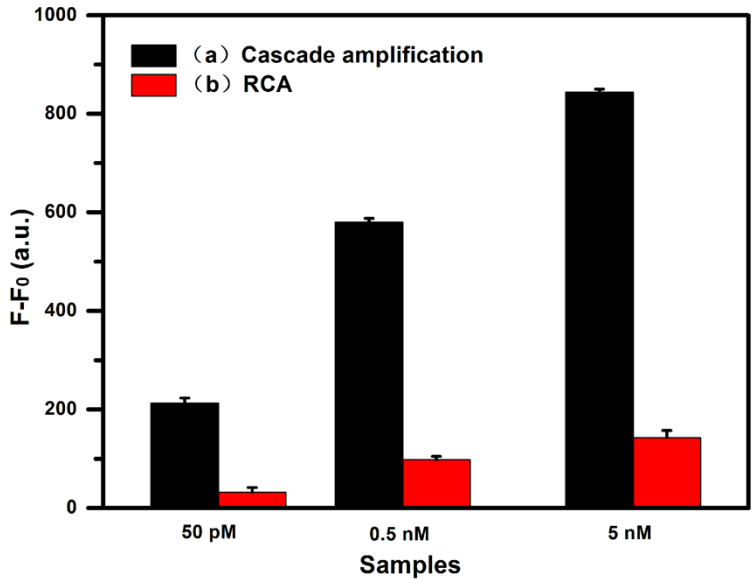

Fig. S2 The fluorescence signal generated by cascade amplification and RCA in three different concentrations of target DNA and primer DNA (50 pM, 0.5 nM, 5 nM). (a) cascade amplification process, template DNA was 5.0×10^{-7} M, probe DNA was 5.0×10^{-7} M, circular DNA was 2.0×10^{-7} M, NMM was 3.0×10^{-6} M; (b) RCA process, probe DNA was 5.0×10^{-7} M, circular DNA was 2.0×10^{-7} M, NMM was 3.0×10^{-6} M