

Supporting Information

Dual-Amplified Sensitive DNA Detection Based on Conjugated Polymers and Recyclable Autocatalytic Hybridization of DNA

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1-Mismatch target DNA (**DNA_{1N}**): 5'-GACATGGCCCACTCTTAATCACTAATT-3'

2-Mismatch target DNA (**DNA_{2N}**): 5'-GACATGGCCCACTCTTAATCAGTAATT-3'

Scheme S1. The sequences of DNA_{1N} and DNA_{2N} that are complementary to probe_A with one and two-base mismatches, respectively.

Experimental Section

Materials and Measurements: PFP was synthesized according to the procedure in the literature.¹ The oligonucleotides were purchased from Sangon Biotech (Shanghai) Co., Ltd. The concentrations of oligonucleotides were determined by measuring the absorbance at 260 nm in 160 μ L quartz cuvette. The water was purified using a Millipore filtration system. Fluorescence measurements were carried out in a 3 mL quartz cuvette at room temperature

using a Hitachi F-4500 fluorometer equipped with a xenon lamp excitation source. All of the spectra were measured in HEPES buffer solution (25mM, pH 8.0) at an excitation wavelength of 380 nm. UV-vis absorption spectra were taken on a JASCO V-550 spectrophotometer. A DYY-6C electrophoresis power supply was used in electrophoresis analysis. DNA samples before and after hybridization in the presence and absence of HaeIII endonuclease were loaded onto 2.5% agarose gel with GoldViewTM Nucleic Acid stain (Invitrogen) in a 0.5 X TBE buffer (4.45 mM tris base, 4.45 mM boric acid, 0.1mM EDTA, pH 7.9). The gel was run at 120V for 20 minutes at room temperature and visualized with ChemiDoc XRS system.

DNA Hybridization and Enzyme Digestion: The hairpin DNA A (probe_A) and DNA B labeled with a fluorescein dye at 5'-terminus (probe_B) were prepared in buffer solution (4 mM MgCl₂, 15 mM KCl, and 10 mM Tris-HCl, pH = 8.0) using a snap-cooling procedure: heating at 90°C for 5 minutes and cooling on ice for 1 minute. The hairpins were allowed to equilibrate at room temperature for 30 minutes before use. After adding the target DNA_C ([DNA_C] = 0.25 μM) to the mixture of probe_A and probe_B (1.0 μM each), the solution was incubated at room temperature for 2h, following up by endonuclease HaeIII digestion for 40 min at 37°C.

DNA Detection: To 1.0 mL of HEPES solution (25 mM, pH 8.0) was added all of the digested products and PFP ([PFP] = 1.0×10^{-6} M in repeated units (RUs), [probe_A] = [probe_B] = 1.0×10^{-8} M, [DNA_C] = 2.5×10^{-9} M) at room temperature. Then the fluorescence spectra were measured with an excitation wavelength of 380 nm at room temperature. In the case of

DNA mismatch detection, the assays were performed under the same conditions as above except that target DNAs with one, two, and full base mismatches (named DNA_{1NC}, DNA_{2NC}, and DNA_{NC}, respectively) were used instead of fully matched DNA_C.

References:

- 1 Liu, B.; Wang, S.; Bazan, G. C.; Mikhailovsky, A. *J. Am. Chem. Soc.* **2003**, *125*, 13306-13307.