Efficient enzyme-powered micromotor device fabricated by a cyclic alternate hybridization assembly for DNA detection

Shizhe Fu, Xueqing Zhang, Yuzhe Xie, Jie Wu,* and Huangxian Ju

State Key Laboratory of Analytical Chemistry for Life Science, School of Chemistry and Chemical Engineering, Nanjing University, Nanjing 210023, P.R. China.

* E-mail: wujie@nju.edu.cn

Experimental

DNA sequences

All oligonucleotides used in this work were synthesized and purified by Sangon Biological Engineering Technology & Co. Ltd. (Shanghai, China) and their corresponding sequences were shown as following (from 5' to 3'):

DNA1: SH-(CH2)6-AAAAAAACCATGTTTACC

DNA2: TAACACGGTCTAAGGTGACGGTAAACATGGACGTCG

DNA3: TAGACCGTGTTAGCGGCGATCTGGGGGAGTAC

S1: COOH-GCCGTGCCAGCACGTCTACCTA GTACTCCCCCAGATCGCCGCT

S2: COOH-AGGTAGACGTGCTGGCACGGCT GGTAGACGTGAGCAATCAGAGCA

S2': COOH-AGGTAGACGTGCTGGCACGGCT

Target DNA: CGACGTCCATGTTTACCGTCACCTTAGACCGTGTTA

Single-base mismatch DNA:

CGACGTCCATGTTTAC G GTCACCTTAGACC GTGTTA

Three-base mismatch DNA:

CGACGTC G ATGTTC G GTGCCTTAGACCG C TGTTA

Non-complementary DNA:
The complementary bases of DNA2 with DNA1 and DNA3, and DNA3 with S1 and S2 (S2') were shown in bold and bold italic, respectively. The mismatch bases were shown in bold with underline.

**Apparatus**

Template-assisted electrochemical growth of outer polymer layer was carried out with a CHI660B electrochemical workstation (CH Instruments Inc., USA). The morphology of the microtube was examined with scanning electron microscope (SEM) (Hitachi S-4800, Japan). Zeta potential analysis was performed on a Zetasizer (Nano-Z, Malvern, UK), which can detect the Zeta potentials of materials with a size range of 3.8 nm – 100 μm, and is suitable for characterizing the PEDOT-PSS microtubes with the length of ~13.5 μm. Polyacrylamide gel electrophoresis (PAGE) analysis was performed on an electrophoresis analyser (Bio-Rad, USA) and imaged on Bio-rad ChemDoc XRS (Bio-Rad, USA). The images and videos were captured by Leica DMI 3000B inverted microscope equipped with a Photometrics Evolve 512/SC camera (Roper Scientific, Duluth, GA), and acquired at a frame rate of 10 frames/s using the Leica MM AF 1.5 software.
Supporting video captions

**Video S1.** Autonomous motion of micromotor device fabricated with different DNA assembly layers in the absence and presence of target DNA.

**Video S2.** Autonomous motion of 4 micromotor devices fabricated with uncontrollable DNA assembly.

**Video S3.** Autonomous motion of micromotor device in H$_2$O$_2$ fuel solutions at different concentrations.

**Video S4.** Autonomous motion of micromotor device in response to target DNA with different concentrations.
**Supporting figures**

**Figure S1.** Schematic representation of target DNA hybridization with DNA2 and the release of DNA multi-layer architecture.

**Figure S2.** Gel electrophoresis image of S1, S2 and the mixture of S1 and S2.
**Figure S3.** (A) Conjugation of catalase on COOH-DNA via EDC and NHS linker. (B) Gel electrophoresis image and (C) protein-staining image treated with brilliant blue R for S1 (lane 1), catalase (lane 2), mixture of catalase and S1 (lane 3), and catalase-S1 conjugate (lane 4).

**Figure S4.** Durability of motion speed of micromotor device in 2% H$_2$O$_2$ solution containing 0.8% (w/v) NaCh.
Figure S5. Decomposition percentage of 2% H$_2$O$_2$ as a function of reaction time in the presence of micromotor device or native catalase.

Figure S6. Decomposition percentage of 2% H$_2$O$_2$ by micromotor device or native catalase for 3 periods of 10 min.

Figure S7. Decomposition percentage of 11 mM H$_2$O$_2$ for 10 min in the presence of (A) 5.5×10$^{-5}$ mg mL$^{-1}$ catalase, (B) 5.5×10$^{-5}$ mg mL$^{-1}$ catalase after exposed to 2% H$_2$O$_2$ for 10 min, and (C) micromotor device after exposed to 2% H$_2$O$_2$ for 10 min.