

Supporting Information

A reagentless and disposable electronic genosensor: From multiplexed analysis to molecular logic gates

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Experimental Section

Materials: Tris-HCl, 6-mercapto-1-hexanol (MCH) and HAuCl₄ were purchased from Sigma (St. Louis, MO). Disposable screen printed carbon electrode (SPCE), comprised of a carbon working electrode, a carbon counter electrode, and a silver pseudoreference electrode, were obtained from Zensor R&D (Taichung, Taiwan). All solutions were prepared using ultrapure water (specific resistance of 18 MΩ·cm), and the oligonucleotides used in the experiments were ordered from Takara Biotechnology Co., Ltd. (Dalian, China) with the sequences as follows:

stem-loop probe for *gyrB* gene:

5'-SH-(CH₂)₆-GCAGTAACAAGAATAAAACGCCACTGC-(CH₂)₇-NH-MB-3'

stem-loop probe for *K-ras* gene:

5'-SH-(CH₂)₆-CCGTTACGCCACCAGCTCCAAACGG-(CH₂)₇-NH-Fc-3'

gyrB gene target: 5'-GGCGTTTTATTCTTGTT-3'

K-ras gene target: 5'-TTGGAGCTGGTGGCGTA-3'

random sequence: 5'-ACTGGCCGTCGTTTTAC-3'

control sequences: 5'-TCTTTTTTCAGTGGAGAA-3' (*gyrB* gene from *S. Sonnei*); 5'-GTGGAGCTTGTGGCGTC-3' (mismatched sequence).

Multiplexed assay: Gold nanoparticles were electrochemically deposited on the working electrode of the SPCE according to a reported procedure.^{1,2} In brief, the SPCE was preconditioned in 20 mM Tris-HCl buffer (pH 7.4) by cycling the potential between -0.6 and 0.6 V at 0.5 V s⁻¹, followed by controlling the deposition potential at -0.6 V for 60 s in 10 mM HAuCl₄ containing 0.1 M KCl. The SPCE was dried with N₂ and 30 μL of the mixture of stem-loop probes for *gyrB* gene and *K-ras* gene (ratio of 1 to 2) at a final concentration of 2 μM was immediately applied to the working electrode and incubated at room temperature (25 °C) for 2 h.

Next, 30 μL of 2 mM MCH was dropped on the electrode surface for 2 h, followed by incubation in 20 mM Tris-HCl buffer containing 140 mM NaCl, 1 mM MgCl₂, 5 mM KCl and 1 mM CaCl₂ (TB, pH 7.0) for 30 min. The electrodes were then incubated with the appropriate concentration of target DNAs in TB and electrochemical measurements were performed by connecting the electrodes to an EC workstation (CHI 825C). The square wave voltammograms were recorded by scanning the potential from -0.50 V to +0.45 V with a step potential of 4 mV, a frequency of 25 Hz and amplitude of 25 mV. Data processing was made by using the "linear baseline correction" function of the CHI 825C software. Values with error bars represent the standard deviations of three parallel samples at each target concentration.

References:

- 1 J. A. Ho, J. Chiu, J. Hong, C. Lin, K. Hwang and J. R. Hwu, *J. Nanosci. Nanotechnol.*, 2009, **9**, 2324.
- 2 W. Liao and J. A. Ho, *Anal. Chem.*, 2009, **81**, 2470.