Supplementary information

Non-invasive detection of gastric cancer relevant D-amino acids with

luminescent DNA/silver nanoclusters

Zhikun Zhang^{#1}, Yang Liu^{#1}, Peifeng Liu², Lu Yang¹, Xingyu Jiang³, Dan Luo⁴ and Dayong Yang^{1*}

¹School of Chemical Engineering and Technology, Key Laboratory of Systems Bioengineering (Ministry of Education), Collaborative Innovation Center of Chemical Science and Engineering (Tianjin), Tianjin University, Tianjin 300350, China ²Renji Hospital, School of Medicine, Shanghai Jiao Tong University, Shanghai 200032, China

 ³National Center for NanoScience and Technology of China, Beijing 100190, China
⁴Department of Biological & Environmental Engineering, Cornell University, Ithaca, New York 14853, United States

[#] These authors contribute equally to this work.

*Corresponding author: Professor Dayong Yang, E-mail: <u>dayong.yang@tju.edu.cn</u>

Effect of DNA sequence on the fluorescence of DNA/Ag NCs.

Ag ions (atoms) have high affinity toward cytosine bases through C-Ag-C. Varying DNA sequence tuned the optical properties of DNA/Ag NCs. The DNA sequences in our study are listed in Table S1.

Table S1 DNA sequences for synthesis of DNA/Ag NCs.

Name	DNA sequence
$(PolyC)_8$	5'-ATCCTCCCACCGGGCCTCCCACCATAAAAACCCCTTAATCCCC-3'
(PolyC) ₇	5'-ATCCTGGGACCGGGCCTCCCACCATAAAAACCCCTTAATCCCC-3'
$(PolyC)_6$	5'-ATCCTGGGACCGGGCCTAAAACCATAAAAACCCTTAATCCCC-3'
(PolyC) ₅	5'-ATCCTGGGACCGGGCCTAAAACCATAAAAGGGTTAATCCCC-3'
(PolyC) ₄	5'-ATCCTGGGACCGGGCCTAAAACCATAAAAGGGTTAATAAAA-3'



Fig. S1 Fluorescence spectra of DNA/Ag NCs with various DNA sequences.

Assay of generation of H₂O₂ from DAA oxidation

The D-Ala solution from 1 μ M to 10 mM, and L-Ala solution at 10 mM were prepared with PBS buffer (pH 6.8, 20 mM), respectively. Then, the D/L-Ala solution were mixed with trinder reagents, incubated at 25 °C for 30 min, and the H₂O₂ was quantified by Visible absorption spectra.



Fig. S2 Assay of generation of H_2O_2 from DAA oxidation. Mechanism (a), spectrum (b) and photographs (c) of solution of colorimetric assay for the generation of H_2O_2 .

Optimization for detection of D-Pro.



Fig. S3 (a) Effect of pH on detection. (b) Effect of temperature on detection. (c) Effect of DAAO concentration on detection. The insets in (a)-(c) are the corresponding fluorescence spectra. Error bars represent standard deviations from three measurements.



Fig. S4 Effect of pH on DAAO activity.



Fig. S5 Effect of temperature on DAAO activity.

Eliminating interference of chloride ion

Chloridion (Cl⁻) are present in gastric juice and saliva, and chloridion decreased fluorescence of DNA/Ag NCs. To eliminate the influence of chloridion, we used AgNO₃ to precipitate Cl⁻.



Fig. S6 Addition of $AgNO_3$ eliminated the influence of chloridion on fluorescence of DNA/Ag NCs.