**Supporting Information**

# Investigating the interaction between DNA-templated gold nanoclusters and HSA by spectroscopy

## 1. ESI-MS spectra of DNA.

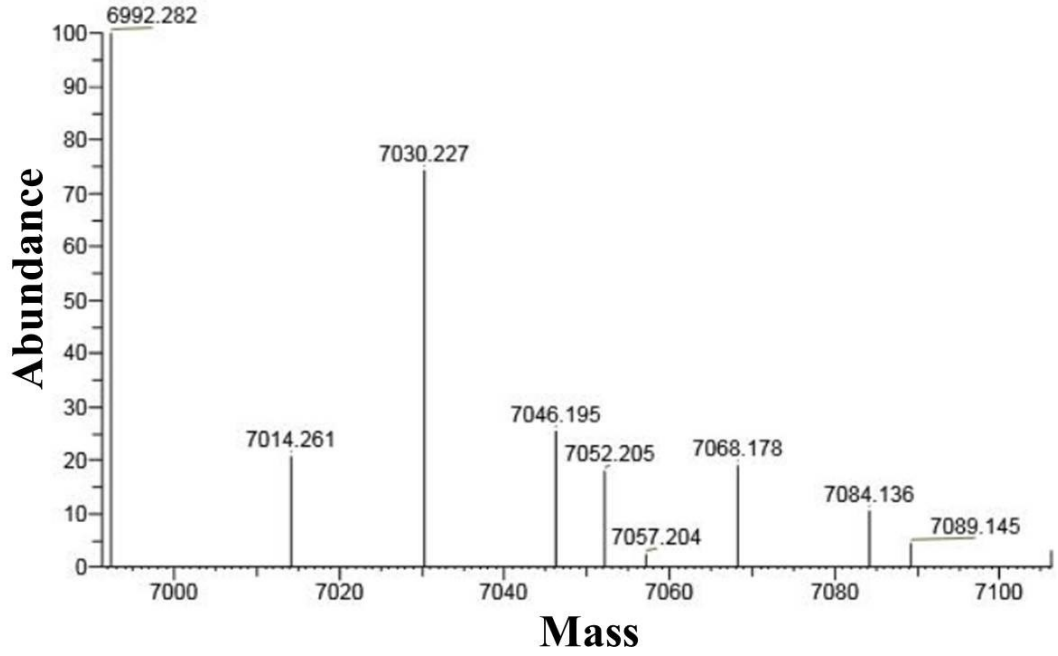


**Figure S1.** Negative-ion ESI-MS of DNA, which shows the molecular weight of DNA is 6992 Da.

## 2. MALDI-TOF-MS of DNA-stabilized DNA-AuNCs

Prior to MALDI-TOF MS measurement, DNA-AuNCs were purified by ultrafiltration using 30 kDa MWCO membranes (Millipore). Next, the AuNCs solution was concentrated 5-fold by ultrafiltration using 3000 Da MWCO membranes (Millipore). Then, a 10 μL aliquot of the AuNCs solution was mixed with an equal volume of methanolic solution of CHCA (0.1 mg/10 μL). The mixture was cast on a stainless steel plate and dried in air for 1 h.

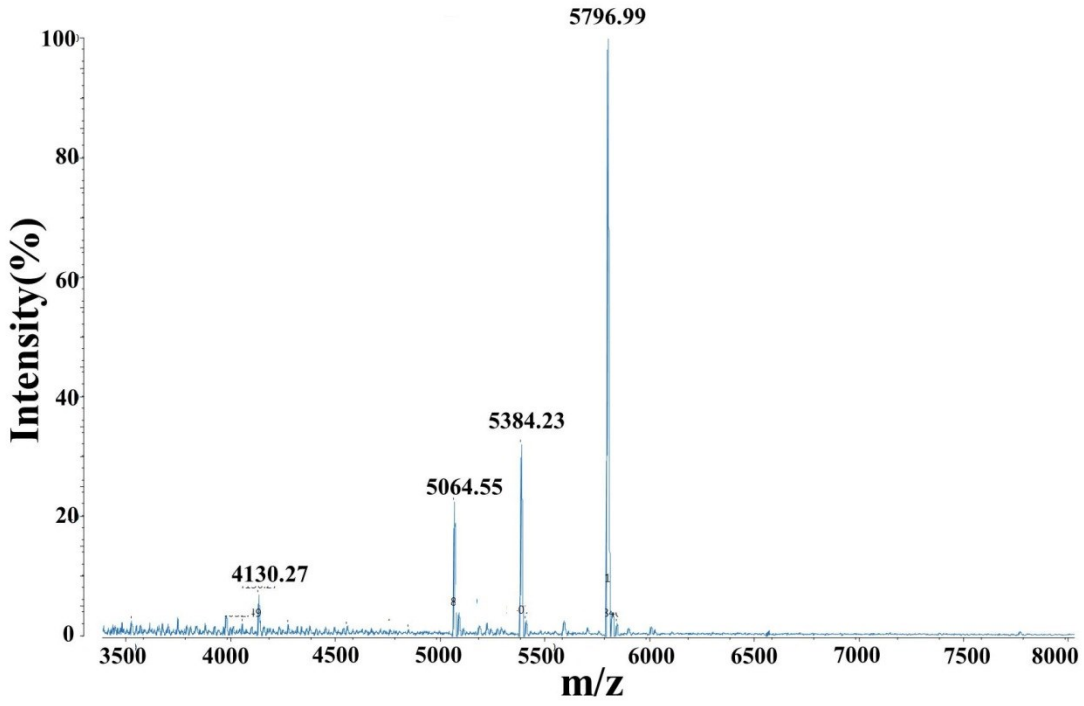


**Figure S2.** Negative-ion MALDI-TOF mass of DNA-AuNCs. The mass spectra of AuNCs consisted of major peaks at *m/z*, 4130.27, 5064.55, 5384.23 and 5796.99, which corresponded to [ 6Au + 3 Na + $H_2O$ + DNA ]$^{2-}$, [16Au + DNA ]$^{2-}$, [ 19Au + Na + H + DNA ]$^{2-}$ and [ 23Au + 3 Na + H + DNA ]$^{2-}$ .

**3. Quenching effect to HSA fluorescence**

In a typical theory, the quenching mechanism is classified into dynamic quenching (collisional encounters) and static quenching (complex formation). They can be distinguished by different dependence on temperature and viscosity, or preferably by fluorescence lifetime measurements.[1] In general, higher temperature results in larger diffusion coefficients and hence increased dynamic quenching; on the contrary, higher temperature usually results in the dissociation of weakly bound complexes, and decreased static quenching. The fluorescence quenching can be described by the well-known Stern-Volmer (SV) [2] equation expressed as:

$$\frac{F0}{F} = 1 + K_{sv}[Q] = 1 + K_q\tau_0[Q] \qquad (S1)$$

where $F_0$ and $F$ are the fluorescence intensity values of the protein HSA in the absence and presence of DNA-AuNCs, respectively. $K_{SV}$ is the Stern-Volmer

quenching constant, and [Q] is the molar concentration of the quencher AuNCs. $K_q$ represents the bimolecular quenching rate, and $\tau_0$ is the average fluorescence lifetime of the HSA in the absence of AuNCs.

For static quenching procedure,[3] the modified Stern-Volmer equation was used for calculating the quenching data.

$$\frac{F0}{F0 - F} = \frac{F0}{\Delta F} = \frac{1}{fa Ka[Q]} + \frac{1}{fa} \qquad (S2)$$

where $f_a$ represents the fraction of accessible fluorescence, and $K_a$ represents the effective quenching constant for the accessible fluorophore.

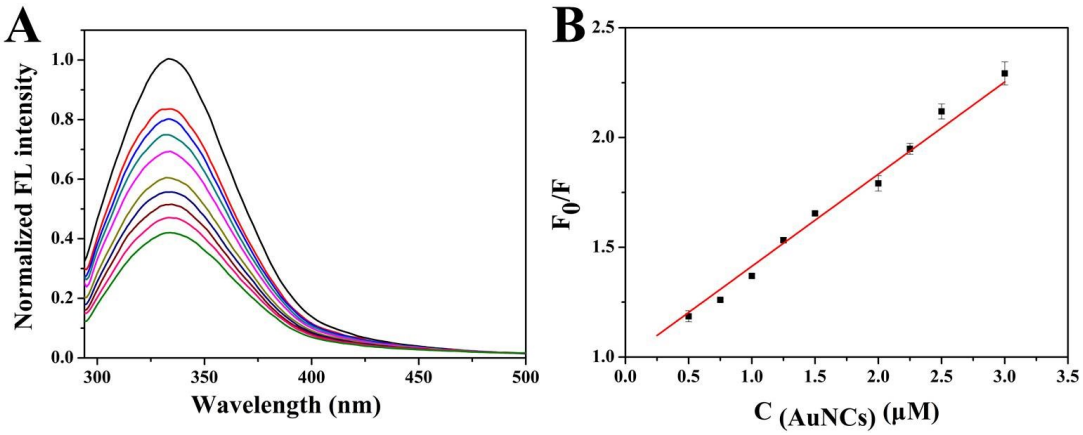


**Figure S3.** (A) Fluorescence emission spectra of the HSA solution upon the addition of various concentrations of DNA-AuNCs (from top to bottom, 0, 0.5, 0.75, 1.0, 1.25, 1.5, 2.0, 2.25, 2.5 and 3.0 μM ) . (B) Relative fluorescence intensity ($F_0/F$) of HSA in the presence DNA-AuNCs (0.5-3.0 μM). The concentration of HSA is 2.0 μM. $\lambda_{ex}$= 280 nm.

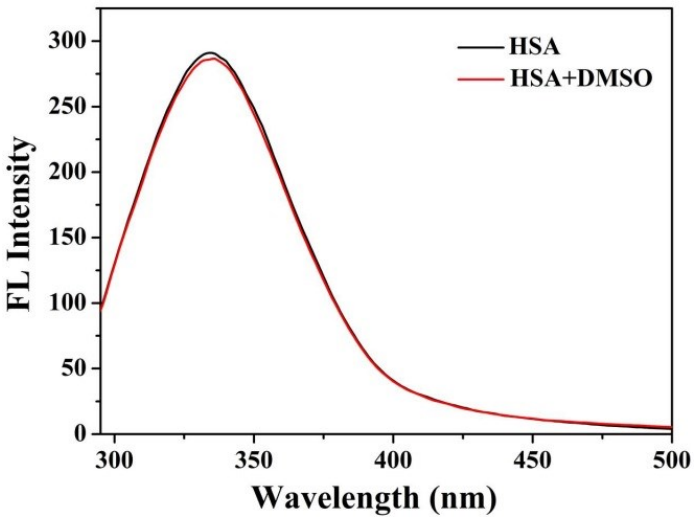

**Figure S4.** Fluorescence spectra of HSA (black line) in the presence of 10 μL DMSO (red line).

**Table S1**. Stern–Volmer quenching constants $K_{SV}$, associative binding constants $K_a$ and relative thermodynamic parameters.

| pH | T (K) | $K_{SV}$ ($10^5$L mol$^{-1}$) | $K$a ($10^5$L mol$^{-1}$) | n | $\Delta G$ (kJ mol$^{-1}$) | $\Delta H$ (kJ mol$^{-1}$) | $\Delta S$ (J mol$^{-1)}$ |
|---|---|---|---|---|---|---|---|
| 7.40 | 298 | 4.23 | 2.31 | 1.20±0.01 | -30.62 | -88.47 | -194.13 |
| | 303 | 3.10 | 1.38 | 1.27±0.05 | -29.65 | | |
| | 308 | 2.35 | 0.72 | 1.32±0.06 | -28.68 | | |

## Reference


1. D. Gao, Y. Tian, S. Bi, Y. Chen, A. Yu and H. Zhang, *Spectrochim. Acta. A Mol. Biomol. Spectrosc.*, 2005, **62**, 1203-1208.
2. B. Tu, Y. Wang, R. Mi, Y. Ouyang and Y.-J. Hu, *Spectrochim. Acta. A Mol. Biomol. Spectrosc.*, 2015, **149**, 536-543.
3. L. A. Sklar, B. S. Hudson and R. D. Simoni, *Biochem.*, 1977, **16**, 5100-5108.