Supporting Information

Development of a near infrared Au-Ag bimetallic nanocluster for ultrasensitive detection of toxic Pb$^{2+}$ ions in vitro and inside cells

Achinta Sannigrahi$^{1,*}$, Sourav Chowdhury$^{1,2,*}$, Indrani Nandi$^{1,3}$, Dwipanjan Sanyal$^{1}$, Sayantani Chall$^{1,*}$ and Krishnananda Chattopadhyay$^{1,3,*}$

$^{1}$Structural Biology & Bio-Informatics Division, CSIR-Indian Institute of Chemical Biology, 4, Raja S. C. Mallick Road, Kolkata 700032, India.

$^{2}$Department of Chemistry and Chemical Biology, Harvard University, 12, Oxford Street, Cambridge Massachusetts, USA.

$^{3}$Academy of Scientific and Innovative Research (AcSIR), Ghaziabad 201002, India

*Corresponding Author Email: krish@iicb.res.in, sayantani.chall@yahoo.com

# contributed equally.
Table S1. Volume and molar ratio of Au\(^{3+}\) and Ag\(^+\) used for the synthesis of Au-Ag@BSA NCs.

| Volume ratio (ml) | Molar ratio (M) |
|-------------------|-----------------|
| 2.0:0.5           | 3.6:0.8         |
| 1.5:1.0           | 2.7:1.6         |
| **Au:Ag**         |                 |
| 1.25:1.25         | 2.3:2.0         |
| 1.0:1.5           | 1.8:2.4         |
| 0.5:2.0           | 0.9:3.1         |
Table S2. Time resolved study of Au-Ag@BSA NCs with simultaneous variation of Au and Ag molar ratios.

| [AuCl₄]⁻ (mM) | [Ag⁺](mM) | λ_{ems} | τ₁ (ns) | τ₂ (ns) | τ_{av} (ns) | χ²  |
|---------------|-----------|---------|---------|---------|-------------|-----|
| 4.5           | 0         | 680     | 1.74    | 260     | 211         | 1.08|
| 3.6           | 0.8       | 665     | 1.48    | 157     | 133         | 1.13|
| 2.7           | 1.6       | 703     | 5.64    | 112     | 100         | 1.16|
| 2.3           | 2.0       | 718     | 1.04    | 98.6    | 14.2        | 1.25|
| 1.8           | 2.3       | 766     | 1.37    | 116     | 11          | 1.16|
| 0.9           | 3.1       | 815     | 0.9     | 64      | 2.5         | 1.04|
Table S3. Different sensors for the detection of contaminants (toxic ions).

| Sensing Probes                  | Contaminant | LOD       | Ref. |
|---------------------------------|-------------|-----------|------|
| Au NP                           | Pb\(^{2+}\) | 100 nM    | 1    |
| Au NP                           | Pb\(^{2+}\) | 3 nM      | 2    |
| Au NC                           | Hg\(^{2+}\) | 0.1 nM    | 3    |
| Au NC                           | Fe\(^{3+}\) | 5.4 μM    | 4    |
| Au NC-QD conjugate              | Hg\(^{2+}\) | 9 nM      | 5    |
| Au NP-Graphene Oxide conjugate  | Pb\(^{2+}\) | 0.1 nM    | 6    |
| Au NC                           | CN\(^{-}\)  | 2 nM      | 7    |
| Au nanofilm                    | Pb\(^{2+}\) | 0.1 μmolL\(^{-1}\) | 8 |
| DNAzyme-Au NP hybrid           | Pb\(^{2+}\) | 0.028 nM  | 9    |
| Au NC                           | Pb\(^{2+}\) | 10 nM     | 10   |
| Au-Ag NC                        | Pb\(^{2+}\) | 96.02 nM  | Present work |
Figure S1. (a) Absorption spectra of Au-Ag@BSA NCs where Au-Ag molar ratio was 2.3:2, inset shows broad shoulder due to the incorporation of Ag (zoomed in from Figure S1a); (b) Zeta potential of Au-Ag@BSA NCs.
Figure S2. Decrease of fluorescence intensity of Au-Ag@BSA NCs with increasing concentration of Ag, inset: excitation spectrum of Au-Ag@BSA NCs.
Figure S3. (a) TEM image of as prepared Au-Ag@BSA NCs, Scale bar was 10 nm; (b) DLS size distribution of Au-Ag@BSA NCs showed the existence of higher population of particles ~2 nm in diameter; (c) EDAX spectrum showed the composition and apparent atomic ratio of Au and Ag in Au-Ag@BSA NCs. Inset table showed the elemental composition (Au/Ag) of Au-Ag@BSA NCs.
Figure S4. MALDI mass spectrum of as-prepared Au-Ag@BSA NC.
Figure S5. X-ray photoelectron spectra (XPS) of (a) Au 4f and (b) Ag 3d of Au-Ag@BSA NCs.
Figure S6. X-ray photoelectron spectra of S 2\textit{p} states.
Figure S7. Fluorescence Stability of the as prepared Au-Ag@BSA nanocluster in presence of (a) EDTA and NaCl; (b) at different pH; (c) Au-Ag@BSA NCs fluorescence stability with time.
Figure S8. Fluorescence intensity of Au-Ag @BSA NCs containing three different molar ratios of Au/Ag (a) 4.5:0; (b) 2.3:2.0 and (c) 0.9:3.1 in presence(red) and absence of Pb^{2+}(black).(d) plot of fluorescence intensity against three ratio of Au/Ag suggesting the significant enhancement of intensity only for the ratio 2.3:2.0.
Figure S9. Fluorescence intensity increment of Au-Ag @BSA NCs (Au/Ag = 2.3:2.0) with gradual addition of Pb\(^{2+}\) in (a) tap water, (b) pond water and (c) drinking water (obtained by reverse osmosis of the tap water). Concentration dependence plot for the determination of recovery of Au-Ag@BSA NCs in the presence of (d) tap water, (e) pond water and (f) drinking water (obtained by reverse osmosis of the tap water).

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