Conformational Change-Induced Fluorescence of Bovine Serum Albumin-Gold Complexes

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Supporting Information

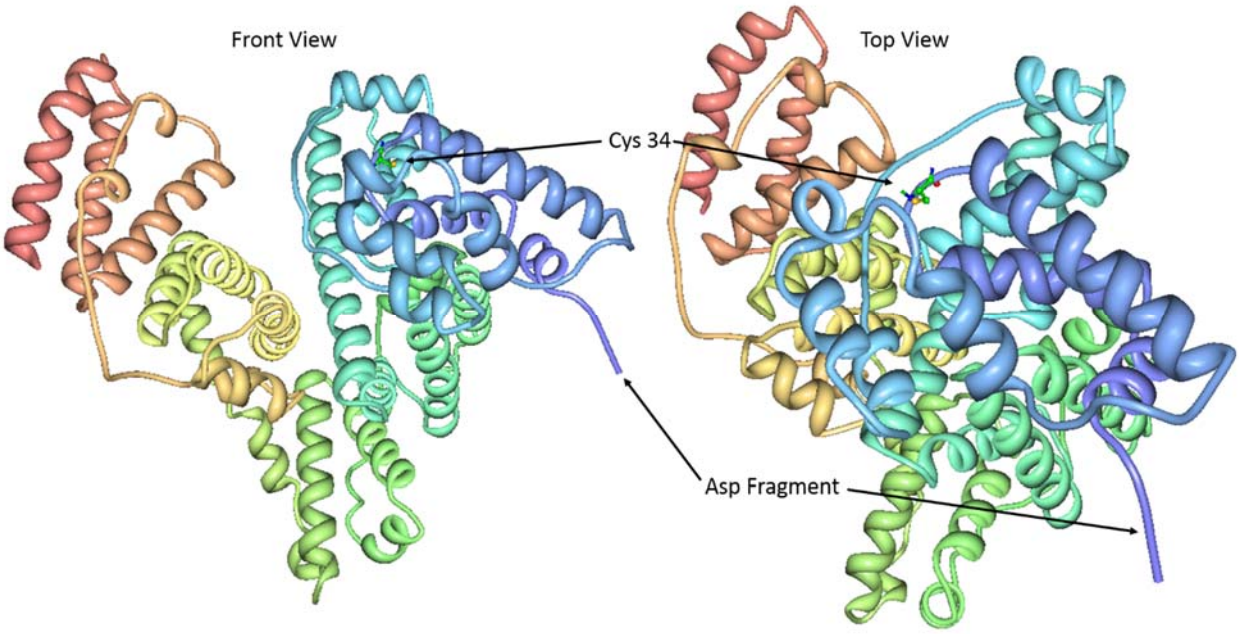

Figure S1. Structure of bovine serum albumin at pH = 7. The "Asp fragment" (Asp-The-His-) is located at the N-terminus. Location of Cys34 is also shown. PDB ID: 3V03.

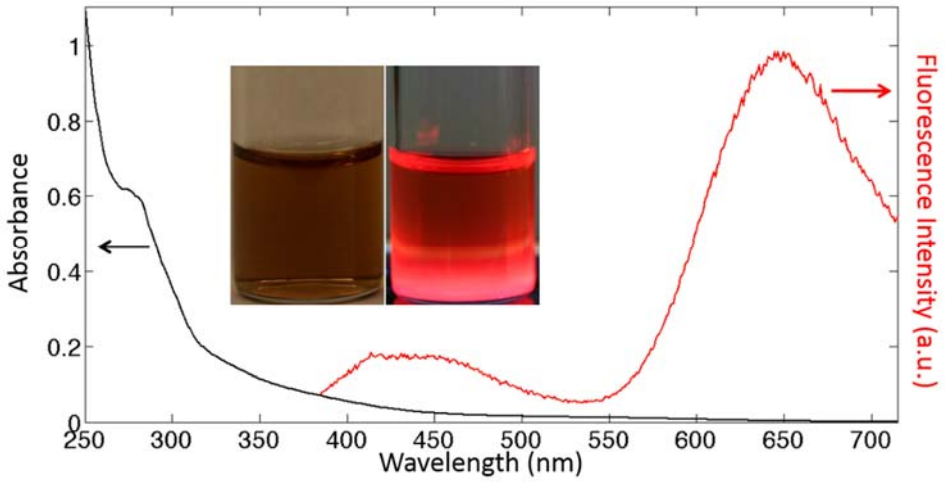

Figure S2. The compound of Xie *et al.* reproduced according to protocol (1).

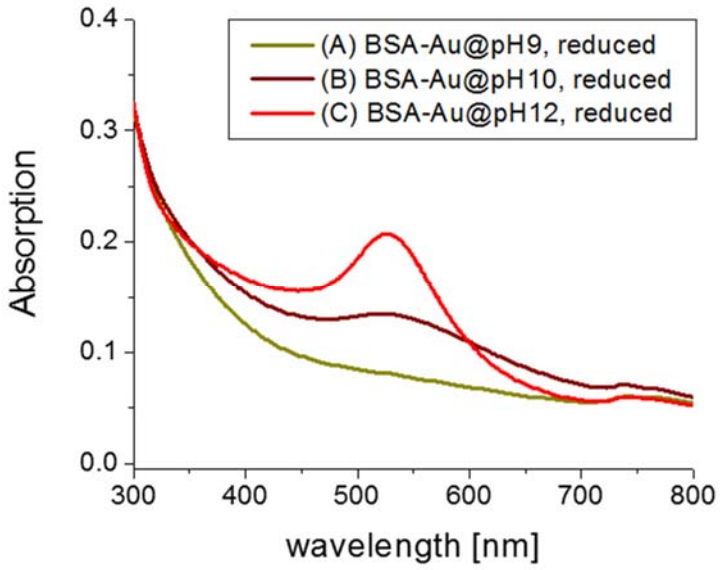

Figure S3. BSA-Au synthesized according to protocol (1) at pH = 9, 10, 12, and then reduced using $NaBH_4$. The absorption spectra show surface plasmon peak at λ = 520 nm for reduced BSA-Au@pH=10,12. This is consistent with the formation of nanoparticles of size ~5 nm observed by transmission electron microscopy. The absorption line-shape is also consistent with small (<2 nm) nanoparticle formation for pH=9 (BSA-Au@pH=9, reduced).

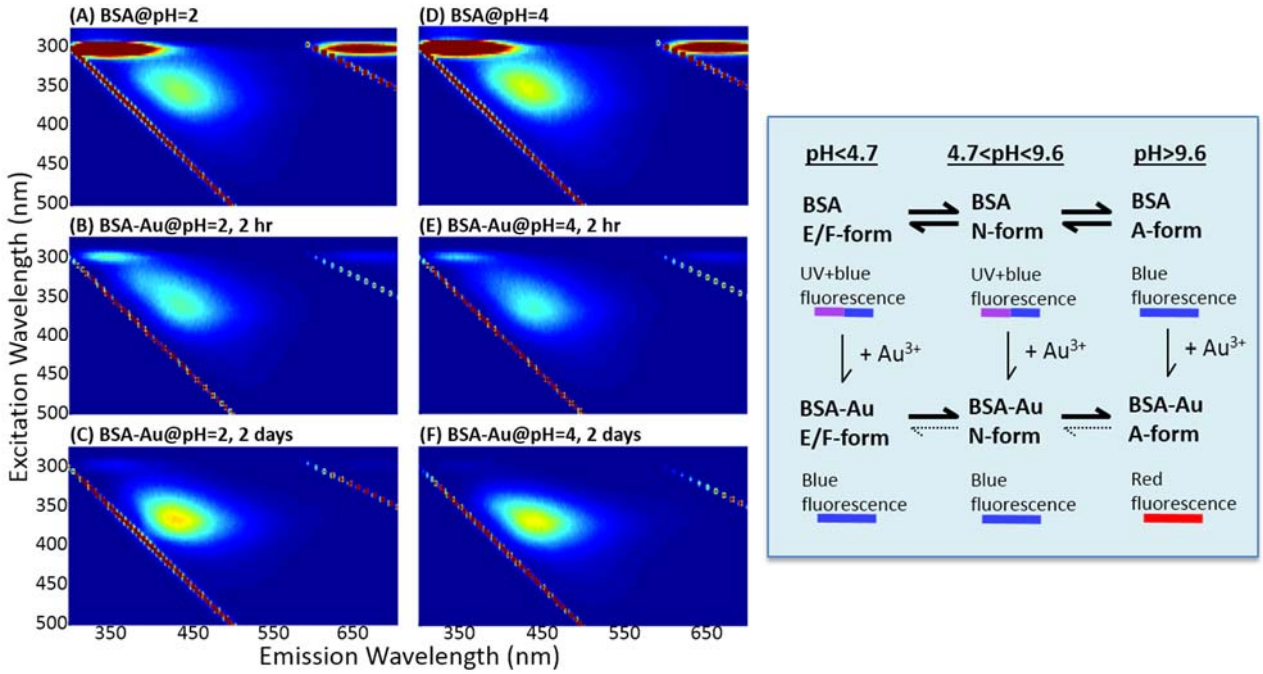

Figure S4. Left panels: Time-course of the excitation-emission map (excitation: 275 – 500 nm; emission: 300 – 700 nm) of BSA and BSA-Au(III) complexes at pH = 2 and 4. (A) BSA@pH=2 (E-form BSA), (B) BSA-Au@pH=2, 2 hours after synthesis, (C) BSA-Au@pH=2, 48 hours after synthesis. (D) BSA@pH=4 (F-form BSA), (E) BSA-Au@pH=4, 2 hours after synthesis, (F) BSA-Au@pH=4, 48 hours after synthesis. The time-course is essentially identical to that of BSA@pH=7 and BSA-Au@pH=7 (Figures 4A-C). BSA-Au@pH=2 and BSA-Au@pH=4 showed no color under ambient light. There was a very small amount of reddish-brown precipitant in BSA-Au@pH=2 after 2 days.

Right panel: A qualitative summary of the relation between pH and the fluorescence of BSA and BSA-Au. The red fluorescence of BSA-Au@pH=12 was found to be stable over weeks in neutral (pH = 7) as well as in acidic (pH = 4) conditions. On the other hand, when BSA-Au@pH=4 and @pH=7 were put into basic (pH = 12), they exhibited red fluorescence within a few hours.

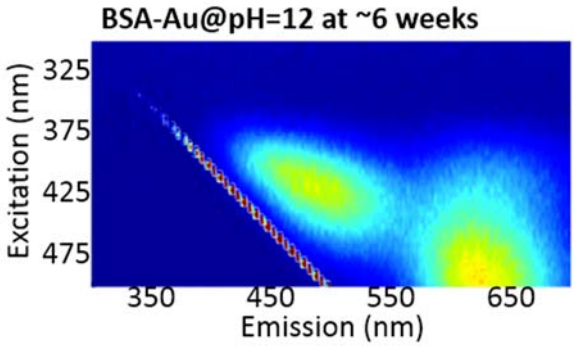

Figure S5. Excitation-emission map of BSA-Au@pH=12 (BSA-to-Au molar ratio 1:13), measured at 6 weeks after synthesis.

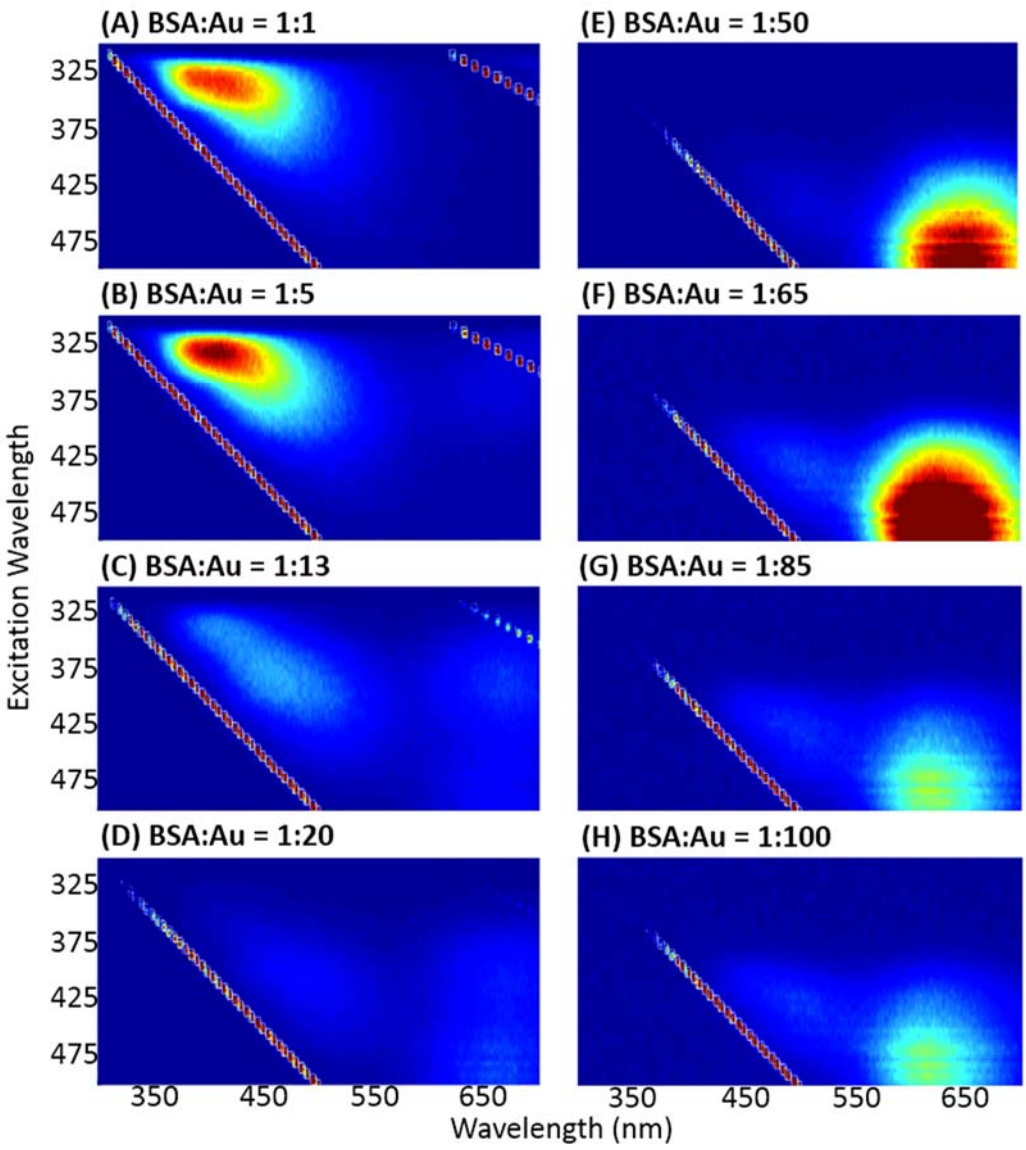

Figure S6. Ratiometric studies of the BSA-Au(III) complexes prepared according to protocol (2), where the pH of BSA solution and $HAuCl_4$ was pre-adjusted to 12, then mixed. The coordination sphere of $AuCl_4^-$ is changed to $Au(OH)_4^-$. Because $(OH)_4^-$ anions are less labile, $Au(OH)_4^-$ stays in solution, thus lesser Au(III) cations are available for reaction. Excitation-emission maps (excitation: 300 – 500 nm; emission: 300 – 700 nm) for the BSA-to-Au molar ratios of (A) 1:1, (B) 1:5, (C) 1:13, (D) 1:20, (E) 1:50, (F) 1:65, (G) 1:85, (H) 1:100.

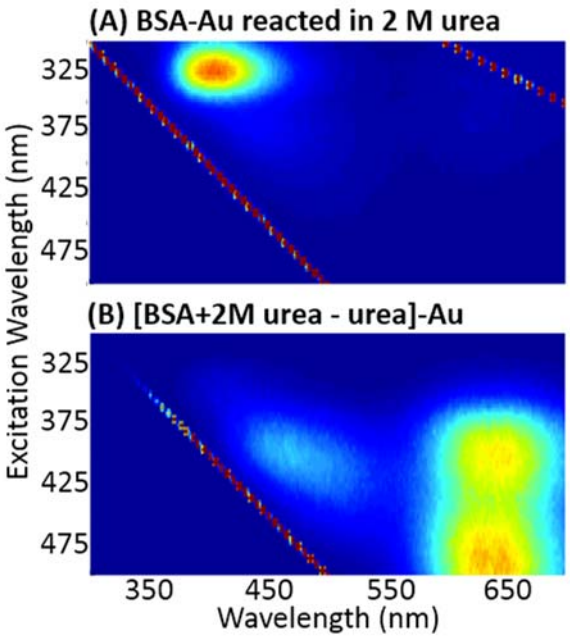

Figure S7. Effect of urea in the reaction of BSA and Au. Urea is used to denature BSA (while Cys-Cys disulfide bonds are preserved), to enable the disulfide bond cleaving by TCEP according to standard procedure. Because urea is known to react with $HAuCl_4$, one needs to confirm the removal of urea from BSA by dialysis before reacting with $HAuCl_4$. (A) BSA reacted with $HAuCl_4$ according to protocol (1) at pH = 12 (BSA-to-Au molar ratio 1 : 13) in the presence of 2 M urea did not yield red-fluorescent compound. (B) BSA was treated with 2 M urea, then dialyzed to remove the urea. The dialyzed BSA was reacted with $HAuCl_4$ according to protocol (1) at pH = 12. This yielded the same red fluorescent BSA-Au compound without the urea processing, resulting in a similar E-E map. We thus confirmed the successful removal of urea by dialysis.

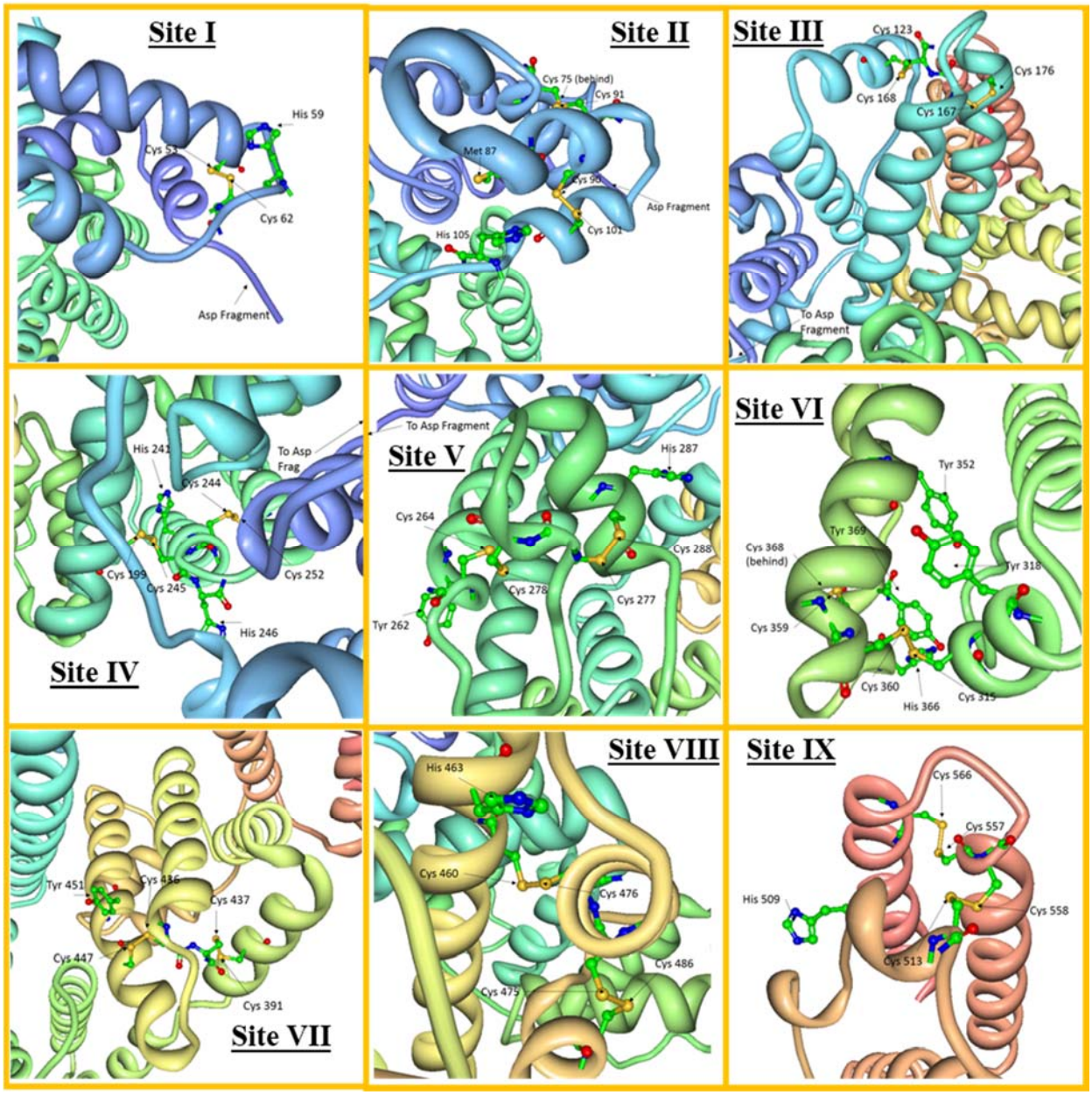

Figure S8. Cys-Cys disulfide bond sites (Site I – IX) in BSA.