

Supporting information for:

Water soluble fluorophore-carbazole-Au-DNA nano hybrid: enhanced two-photon absorption for living cell imaging application

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Contents:

1. Synthesis process of compound a-c
2. SEM and TEM images
3. Linear optical properties of L-Au nano hybrid prepared without ct-DNA
4. Schematic molecular arrangement of the L-Au-DNA hybrid
5. Cyclic-voltammetric response of carbazole and H₂AuCl₄
6. FT-IR and Raman scattering spectra of L-Au and L-Au-DNA nano hybrids
7. One-photon and two-photon images of HepG2 cells incubated with **L** after 4 hours of incubation.
8. One-photon and two-photon images of HepG2 cells incubated with L-Au-DNA after 2 hours of incubation.

Synthesis of the compound **a**

9-hexyl-9H-carbazole-3-carbaldehyde (2.80 g, 0.01 mol) and sodium borohydride (0.76 g, 0.02 mol) were dissolved in methanol (20 mL), and the mixture was refluxed for 6 h with stirring. The compound **a** was collected by evaporating the solvent methanol.

Synthesis of the compound **b**

a (2.80 g, 0.01 mol) and triphenylphosphine (3.9 g, 0.015 mol) were dissolved in chloroform (30 mL). Potassium iodide (1.70 g, 0.01 mol) was dissolved in water (1 mL), which was added into the above solution along with glacial acetic acid (5 mL) and 18C6 (10 mg). The mixture was stirred for a week and some precipitate appeared. Then, The precipitate was collected by pouring the mixture into 1,4-dimethylbenzene to get white-yellow powder **b**.

Synthesis of the compound **c**

b (0.65 g, 1.0 mmol) and 1,4-phthalaldehyde (0.13 g, 1.0 mmol) were dissolved in *N,N*-dimethylformamide (5 mL), the mixture was stirred for 12 h. The reaction was monitored by TLC to ensure complete reaction. The mixture was poured into dichloromethane (500 mL) and extracted four times with water (100 mL for each). The organic layer was separated off and dried with anhydrous Na₂SO₄, filtered, the crude product was purified by silica gel column chromatography (petroleum ether: ethyl acetate = 50 : 1) to yield a light yellow powder **c**. ¹H NMR(400 MHz, CDCl₃): δ(ppm) 10.04 (s, 1H); 8.52 (d, 1H); 8.25 (d, 1H, *J* = 7.6 Hz); 7.98 (d, 2H, *J* = 8.4 Hz); 7.90 (d, 2H, *J* = 8.4 Hz); 7.87 (d, 1H, *J* = 8.4 Hz); 7.74~7.01 (d, 1H, *J* = 16.3 Hz), 7.68 (t, 2H, *J* = 8.5 Hz); 7.54 (t, 1H, *J* = 7.2 Hz); 7.47~7.23 (d, 1H, *J* = 16.3 Hz); 7.29 (t, 1H, *J* = 7.2 Hz); 4.46 (t, 2H, *J* = 7.2 Hz); 1.84 (m, 2H); 1.34 (m, 6H); 0.87 (m, 3H, *J* = 6.8 Hz).

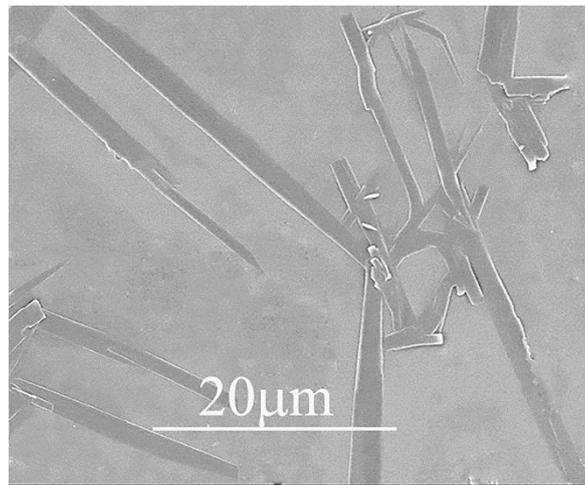


Figure S1 SEM image of L nanostructure

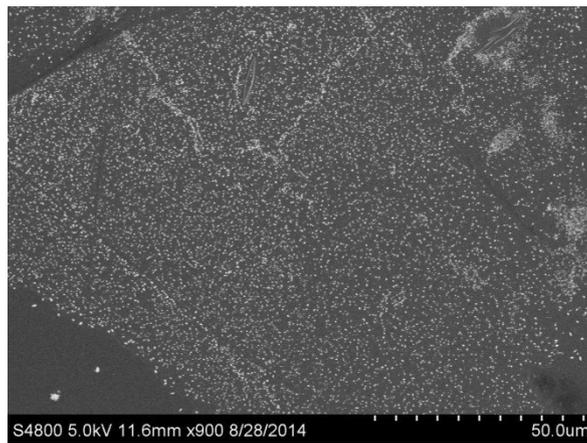


Figure S2 SEM of L-Au-DNA nano hybrid

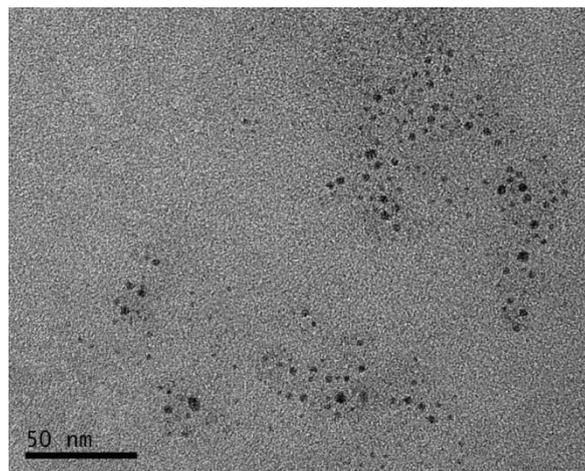


Figure S3 TEM of L-Au-DNA nano hybrid

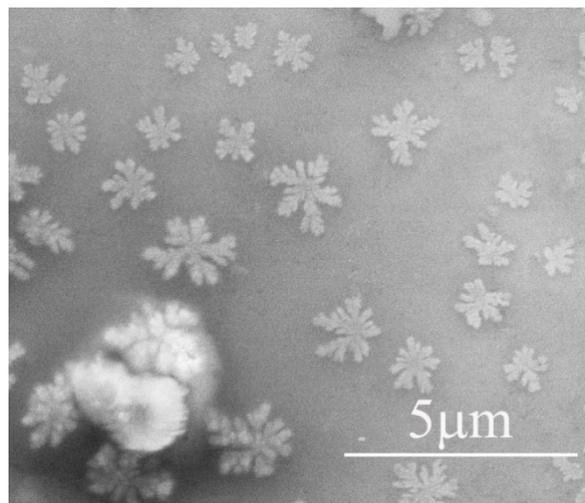


Figure S4 SEM image of L-Au hybrid prepared without ct-DNA

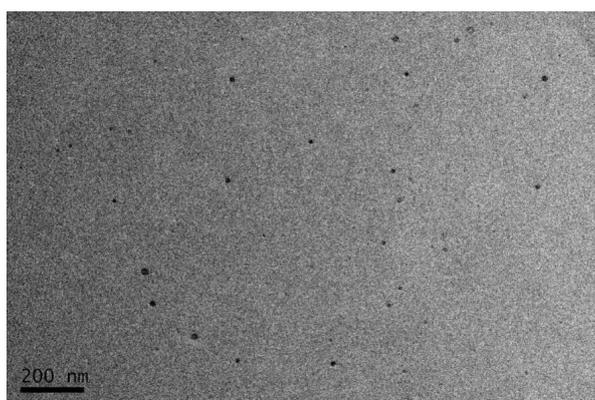


Figure S5 TEM image of pure Au nanoparticles prepared without L and ct-DNA

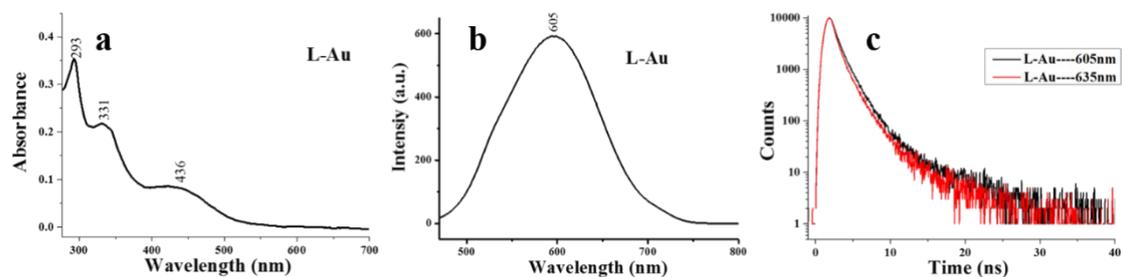


Figure S6 (a) UV-vis absorbance, (b) fluorescence emission and (c) FL lifetime (detected at 605 nm and 635 nm, respectively) spectra of L-Au nanohybrid prepared without ct-DNA

Table S1 Fluorescence decay lifetime (τ) and amplitude (A) of fluorescence of L-Au hybrid

	λ (nm)	τ 1(ns)	A1(%)	τ 2(ns)	A2(%)	τ 3(ns)	A3(%)	$\bar{\tau}$ (ns)	χ^2	ϕ_{ET} (%)
L-Au	605	0.51	68	1.53	31	5.86	1	0.85	0.97	56.4
	635	0.48	77	1.45	22	4.47	1	0.72	1.08	

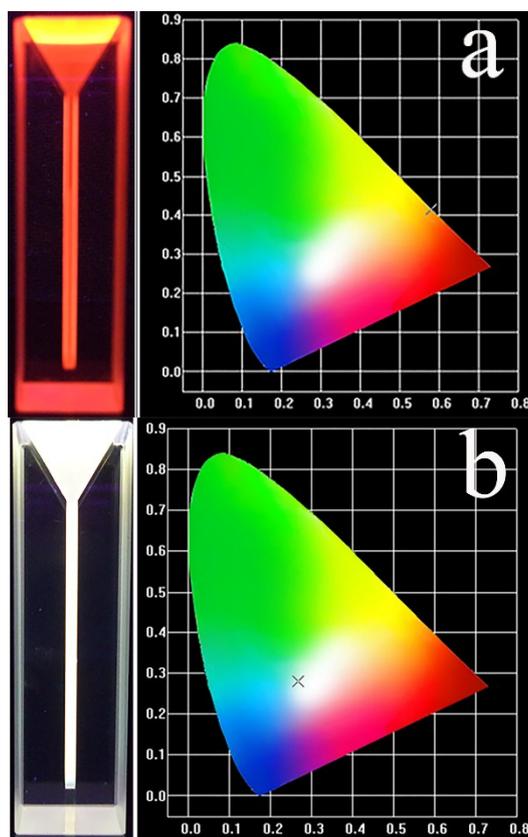


Figure S7 (a) Chromaticity diagram (CIE) of fluorescence emission of **L** and photo of it under 365 nm UV light; (b) Chromaticity diagram (CIE) of fluorescence emission of **L**-Au-DNA nanohybrid and photo of it under 365 nm UV light.

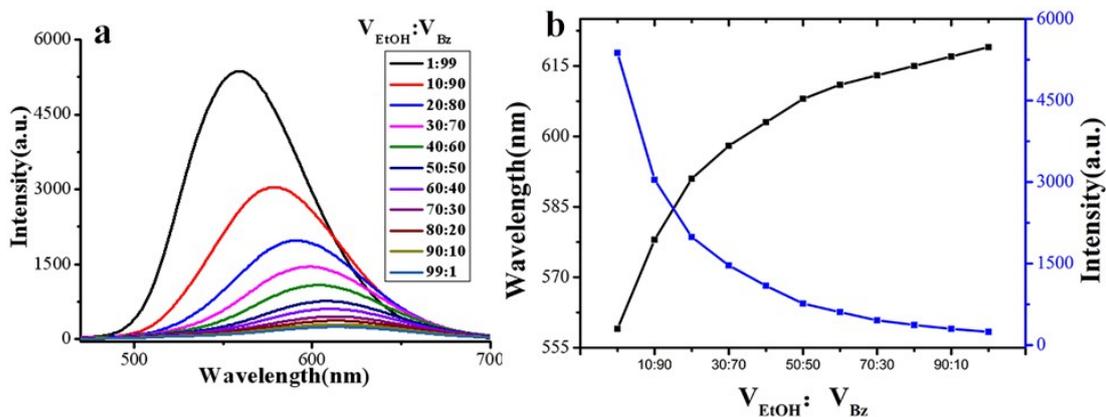


Figure S8 (a) PL emission spectra changes of **L** ($1.0 \times 10^{-4} \text{ mol} \cdot \text{L}^{-1}$) in EtOH-benzene mixtures with different volume ratios; (b) the effect of volume ratio of EtOH to benzene on the maximum emission intensity and wavelength of **L** in EtOH-benzene with the concentration being $1.0 \times 10^{-4} \text{ mol L}^{-1}$.

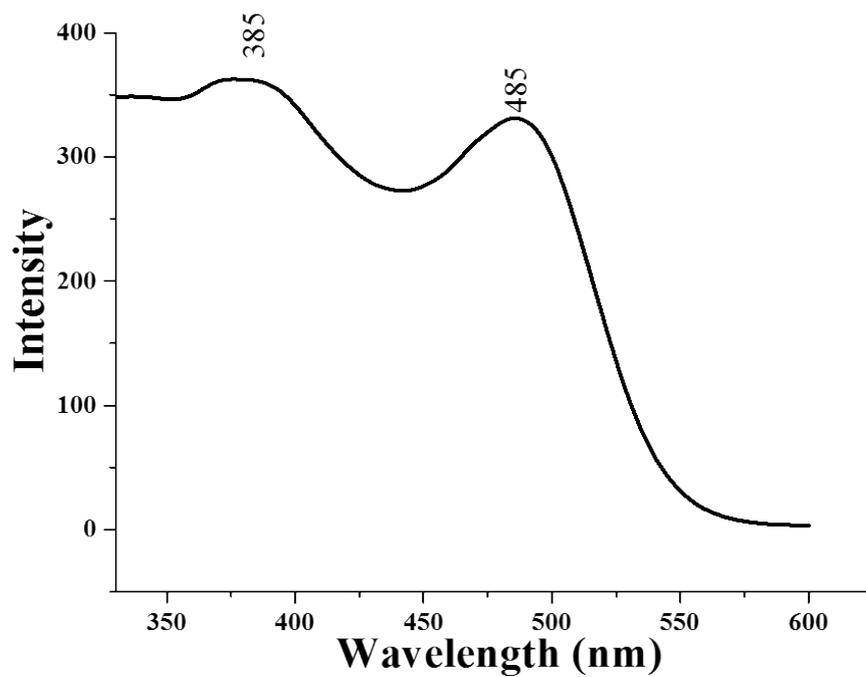


Figure S9 Excitation spectrum of L

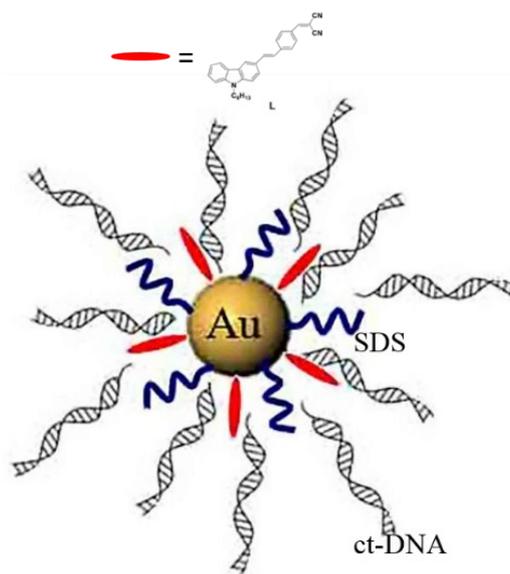


Figure S10 A schematic molecular arrangement of the L-Au-DNA hybrid

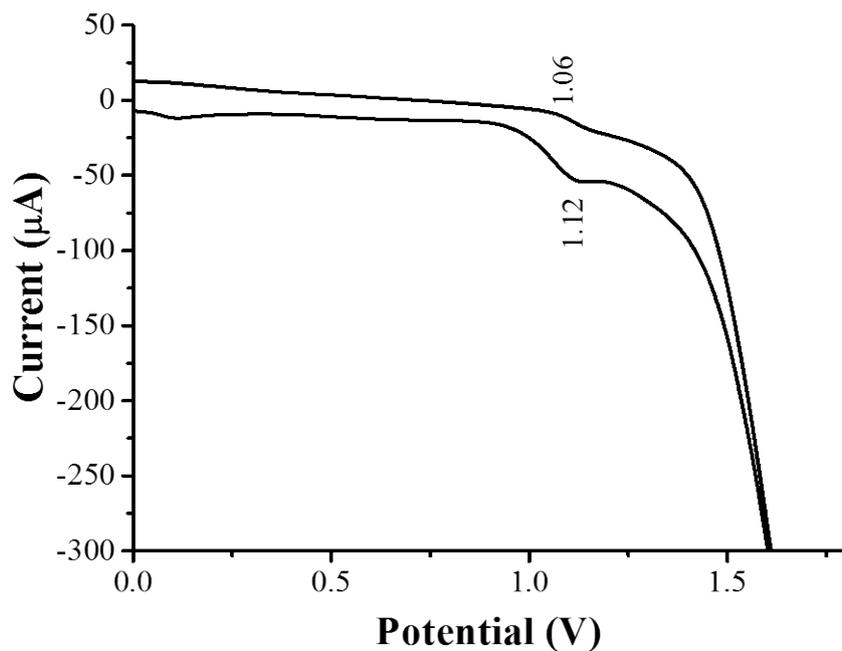


Figure S11 Cyclic-voltammometric response of carbazole in the presence of 0.1 M TBAP *Vs.* Ag/AgCl at room temperature. Scan rate: 100 mV/s

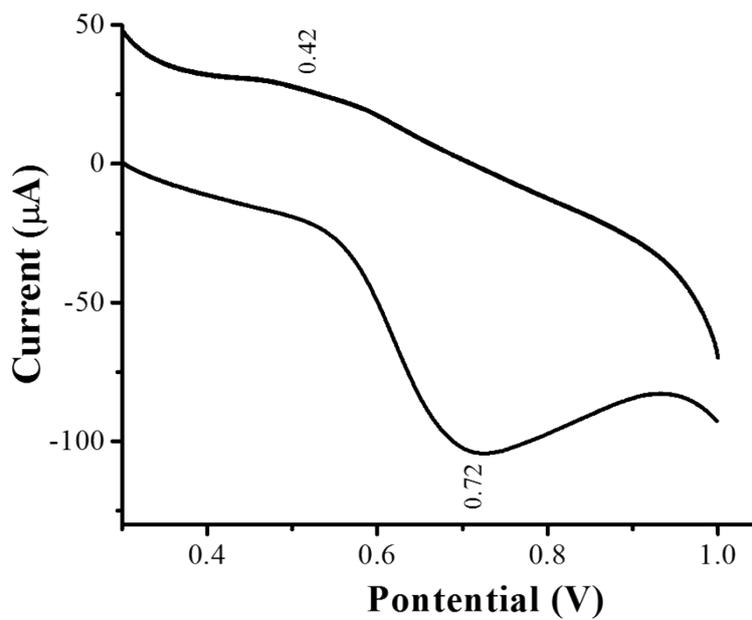


Figure S12 Cyclic-voltammometric response of H[AuCl₄] in the presence of 0.1 M TBAP *Vs.* Ag/AgCl at room temperature. Scan rate: 100 mV/s

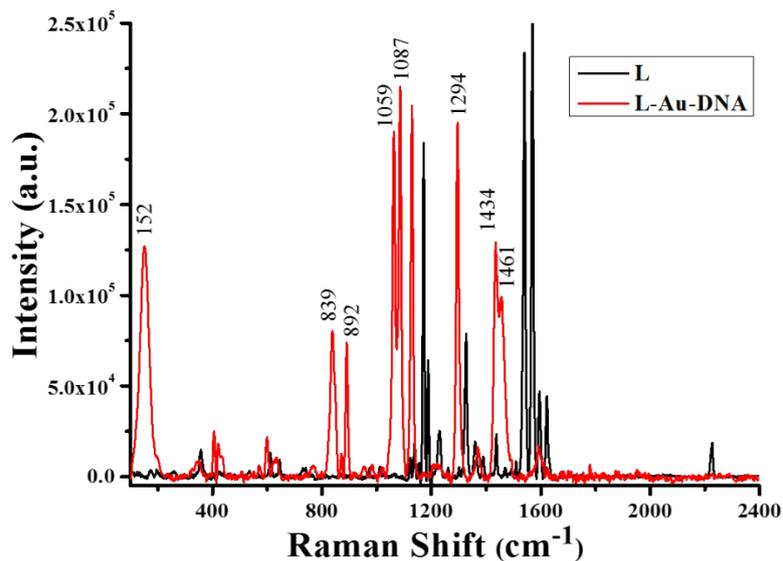


Figure S13 Raman spectra of L and L-Au-DNA nanohybrid

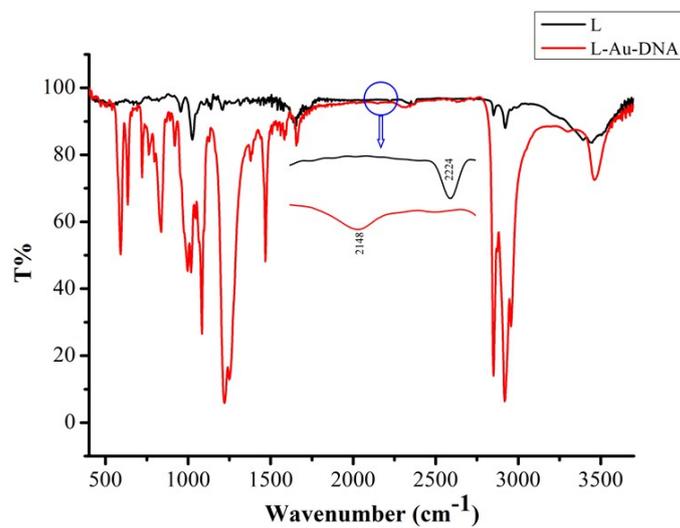


Figure S14 FT-IR spectra of L and L-Au-DNA hybrid

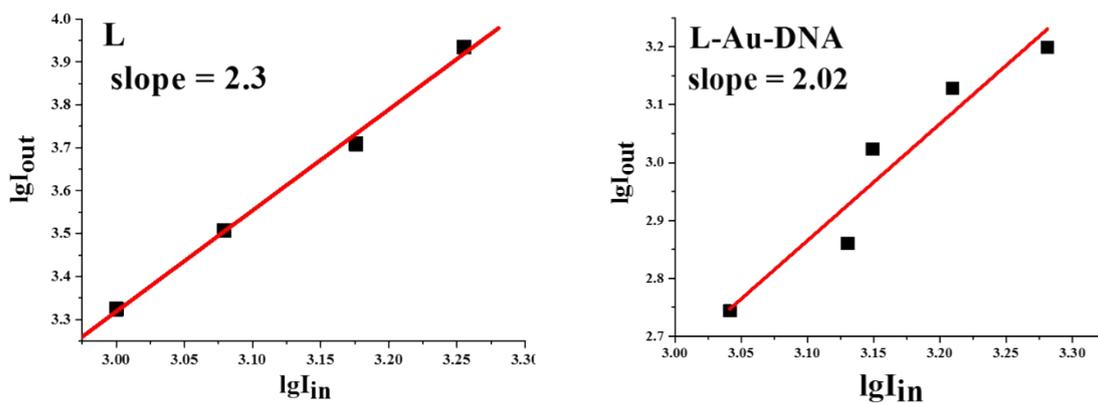


Figure S15 The logarithmic plots of the output two-photon excited fluorescence (I_{out}) vs the square of input laser (I_{in}) of L and L-Au-DNA nanohybrid, respectively.



Figure S16 The photo of L-Au-DNA nano hybrid in water solution

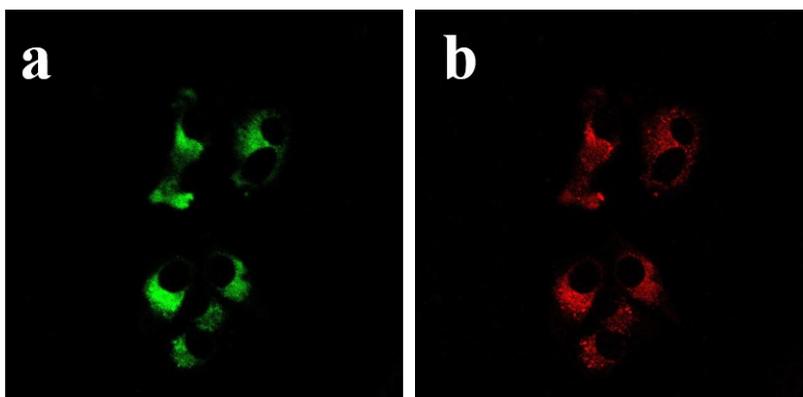


Figure S17 (a) One-photon image ($\lambda_{ex} = 450$ nm, emission wavelength from 600 to 650 nm), (b) two-photon image of HepG2 cells incubated with 10 μ M of L ($\lambda_{ex} = 880$ nm, emission wavelength from 610 to 660 nm), after 4 hours of incubation, washed by PBS buffer. The scale bar was 20 μ m.

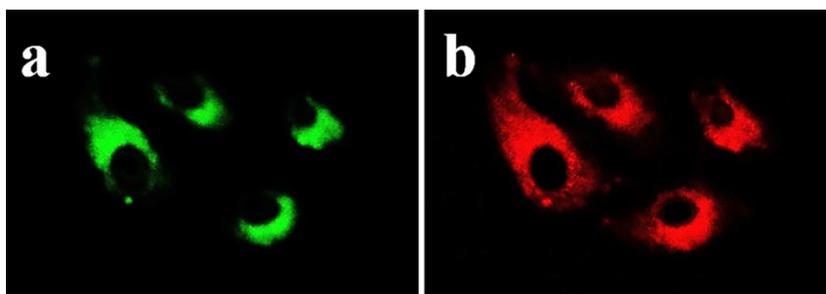


Figure S18 (a) One-photon image ($\lambda_{ex} = 420$ nm, emission wavelength from 530 to 580 nm), (b) two-photon image of HepG2 cells incubated with 10 μ M of the L-Au-DNA nano hybrid ($\lambda_{ex} = 800$ nm, emission wavelength from 550 to 600 nm), after 2 hours of incubation, washed by PBS buffer. The scale bar was 20 μ m.