Electronic Supplementary Information

An EGFRvIII targeted dual-modal gold nanoprobe for imaging-guided brain tumor surgery

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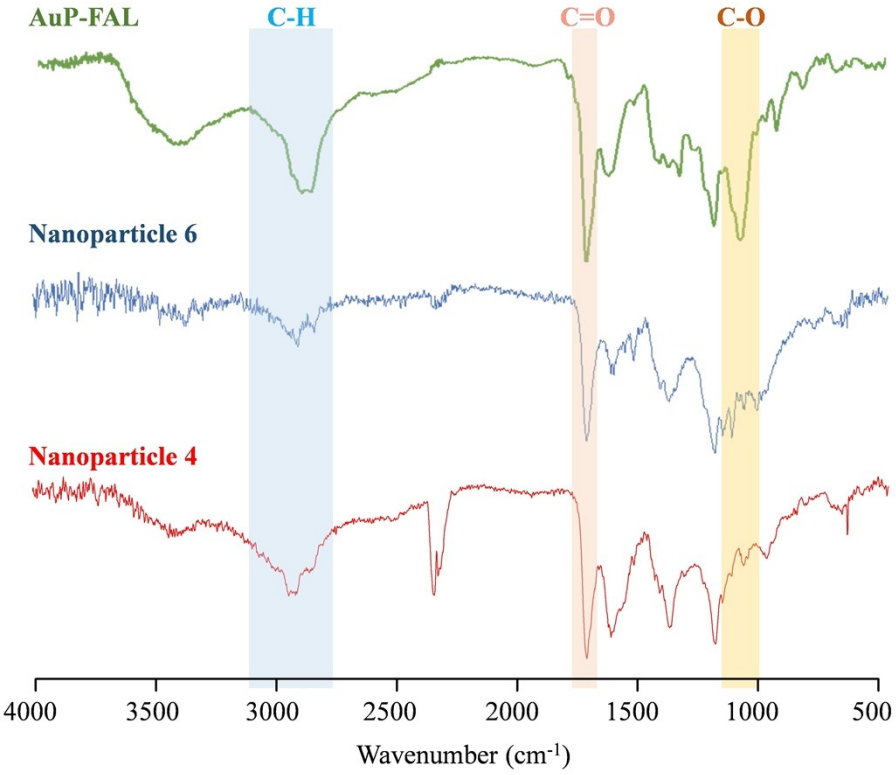

Figure S1. Fourier transform infrared spectroscopy (FTIR) of AuP-FAL and its intermediates. The strong peak at 1720 cm^{-1} was attributed to the C=O stretching in the DTPA chelators, scissor-like peaks at 1044 and 1193 cm^{-1} were assigned to the S=O stretching in the IR783B, a broad peak at 2800–2960 cm^{-1} was attributed to C−H stretching in PEGs, peaks at 1650 and 1550 cm^{-1} were assigned to typical amide bending in FAL peptides.

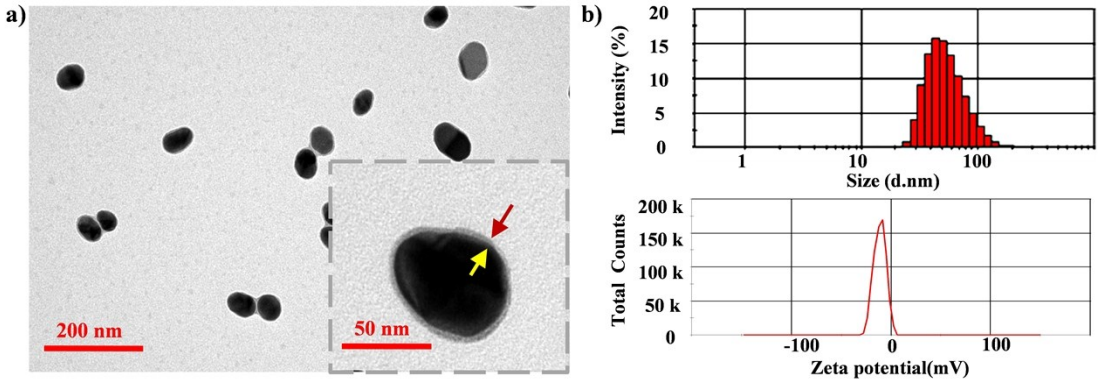

Figure S2. Characterization of **AuP-PEG**. (a) TEM image demonstrated the morphology of AuP-PEG with an average diameter about 60 nm. Insert: amplified image presenting the semi-transparent PEG coating. (b) AuP-PEG showed a hydrodynamic size of 60.7 nm and zeta potential of -21.1 mV.

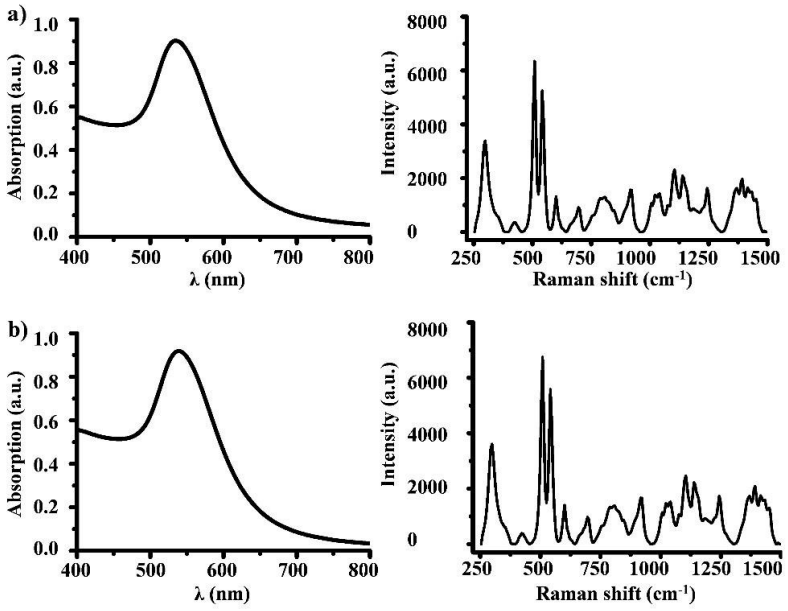

Figure S3. Absorbance and Raman spectra of AuP-FAL (a) and AuP-PEG (b).

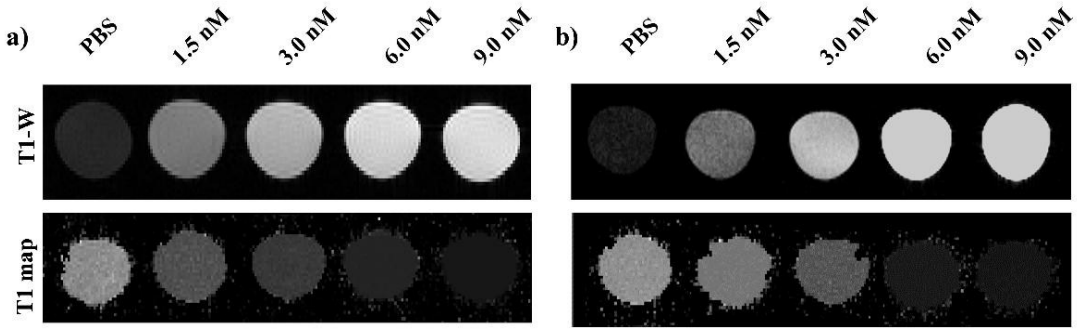

Figure S4. T1-weighted magnetic resonance phantoms (upper panel) and T1 maps (lower panel) of AuP-FAL (a) and AuP-PEG (b) in PBS.

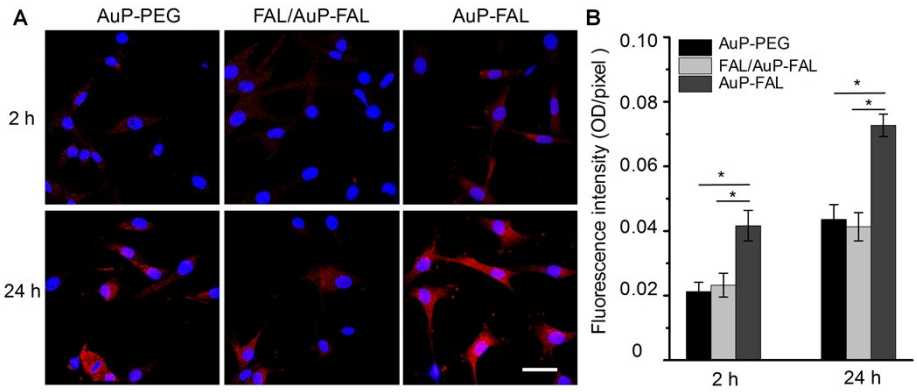

Figure S5. Cellular uptake of nanoprobe in U87-EGFRvIII cell culture. (a) Confocal fluorescence microscopic images of the U87-EGFRvIII cells treated with 0.5 nM AuP-PEG or AuP-FAL for 2 h or 24 h. In the receptor competitive study, U87-EGFRvIII cells were pretreated with 200 nM FAL peptide for 30 min followed by AuP-FAL. Scale bar: 20 μm. (b) Mean intracellular fluorescence intensities (optical density per pixel) after nanoprobe treatment for 2 and 24 h at 37 °C. The values represent mean ± SD (n = 4). * $p < 0.05$ (Mann-Whitney U-test).

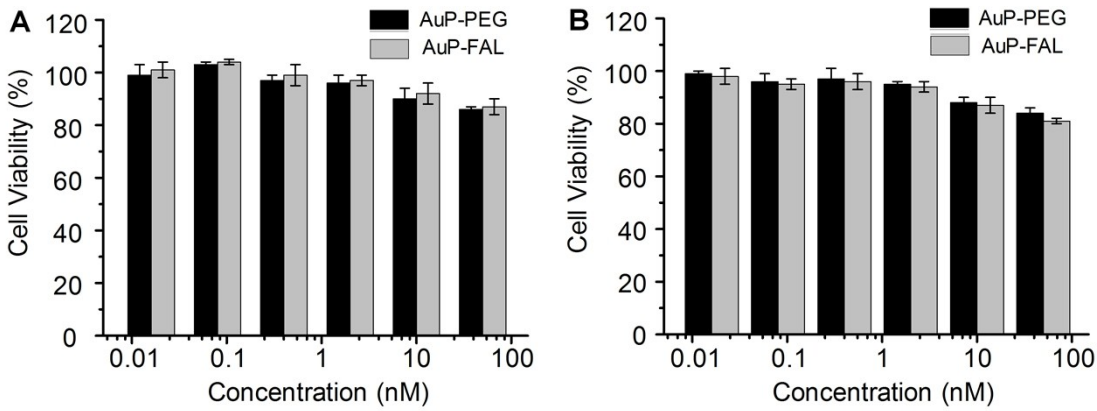

Figure S6. Cytotoxicities of AuP-PEG and AuP-FAL in human glioblastoma U87-EGFRvIII cell line (A) and mouse brain capillary endothelial bEnd.3 cell line (B). Cells were treated with nanoprobe for 24 h with final concentrations in a range of 0.016−50 nM. The CCK8 assay was applied to measure the cytotoxicity.

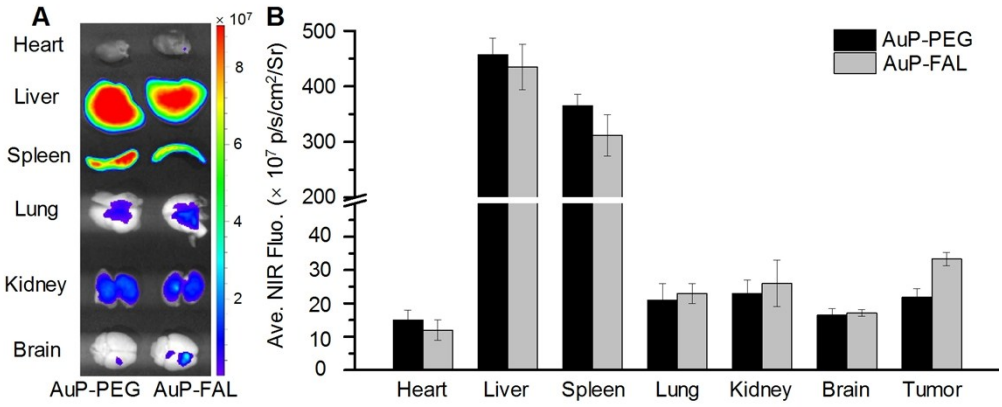

Figure S7. Bio-distribution of AuP-PEG or AuP-FAL in mice bearing 87-EGFRvIII tumor xenograft at 24 h post-injection. (A) *Ex vivo* fluorescence images of the excised mouse organs at 24 h post-injection of the nanoprobe. (B) Bio-distribution of the nanoprobe labeled with near-infrared fluorescent dye IR783B in tumor-bearing mice at 24 h post-injection. The values represent mean ± SD (n = 3).

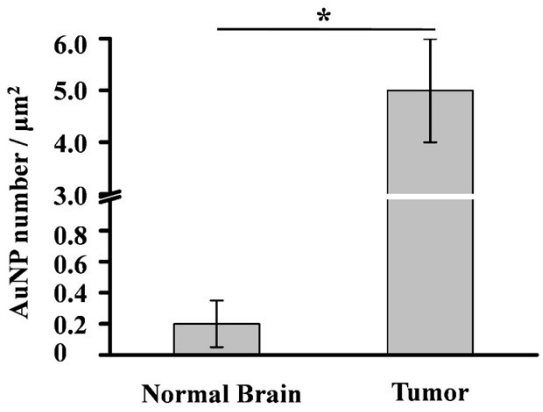

Figure S8. Average number of nanoprobe distributed in a selected area of tumor periphery and normal brain tissue. Above data were calculated from TEM images. All data are presented as mean ± SD. *, $P < 0.05$.

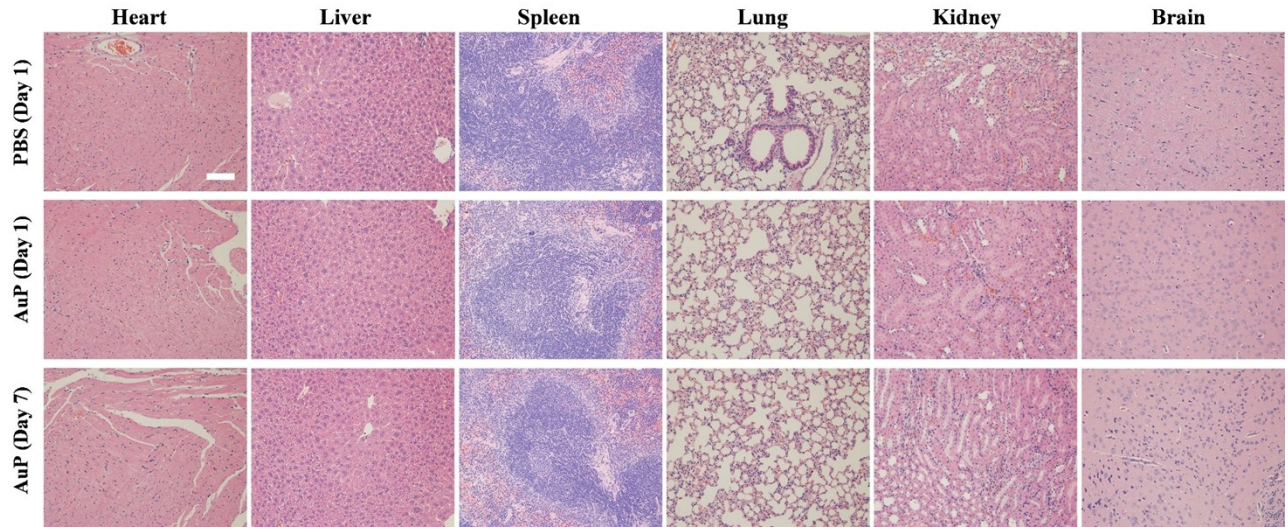

Figure S9. H&E staining of major organs (heart, liver, spleen, lung, kidney, brain) from healthy mice treated with PBS or AuP-FAL. The organs were harvested at 1 day or 7 days after intravenous injection, and then sectioned for histological staining. No obvious lesions were observed. Scale bar = 50 μm.

Table S1. Physical parameters of the gold nanoprobes.

Nanoprobe	d (nm)[a]	PDI [a]	ζ[a]	λ_{abs}[b] (nm)	Molar ratios [c]
AuP-FAL	61.2	0.178	-23.3	535	1/10780/1257/36,152/32
AuP-PEG	60.7	0.195	-21.1	535	1/10147/1308/37,916/N.A.

[a]Diameters (d), polydispersity index (PDI) and zeta potentials (ζ) were measured by dynamic light scattering (DLS). [b]Maximal absorption wavelength. [c]The molar ratios of gold nanoparticle/Gd^{3+}/IR783B/PEG/FAL peptide in the nanoprobe. Number of Gd^{3+} ions were determined by inductively coupled plasma atomic emission spectroscopy (ICP-AES). The average number of IR783B, PEG or FAL peptide labeled on the nanoprobe was quantified by gravimetric analysis. N.A. means Not Applicable.