Supplementary Material (ESI) for Chemical Communications

Supporting Information

Nanoparticle assisted magnetic resonance imaging of the early-reversible stage of amyloid β self-assembly

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Synthesis of Co@Pt-Au

Co@Pt-Au nanoparticles were synthesized by catalytic hydrogenation of the gold precursor, $AuCl(PPh_3)$, in the presence of Co@Pt nanoparticle seeds. Co@Pt nanoparticles (6 nm, 5.1 mg), $AuCl(PPh_3)$ (19.8 mg, 0.04 mmol), and hexadecylamine (45.6 mg, 0.4 mmol) were dissolved in 1,2-dichlorobenzene (5 mL) and 4 % H_2/Ar mixture gas was continuously bubbled through the solution. The resulting solution was heated to 70 ^oC over 20 min. and kept at this temperature for 15 min. The initially dark brown solution gradually changed to red as the reaction proceeded. The resulting solution was cooled to room temperature and treated with 7 mL of ethanol to precipitate red nanoparticles which were then separated by centrifugation.

Synthesis of LA-PEG_{600}-X (X= COOH, OH)

Lipoic acid-PEG_{600}-X (LA-PEG_{600}-X; X=COOH, OH) was synthesized by using a slight modification of the literature method. LA-PEG_{600}-OH (1.6 g, 2 mmol) and succinic anhydride (220 mg, 2.2 mmol) was mixed in 3 mL pyridine and stirred for 5 h at 50 ^oC. Pyridine was removed under reduced pressure (10^{-2} mm Hg) and carboxylated LA-PEG_{600} was washed several times with deionized (DI) water. The LA-PEG_{600} complexes are analysed by using NMR and MALDI-MS.

LA-PEG_{600}-OH: TLC (chloroform/methanol 95:5) R_f = 0.3. 1H NMR (250 MHz, $CDCl_3$): (ppm) 4.10 (t, 2H), 3.55 (m, 44H), 3.09 (m, 2H), 2.94 (s, 1H), 2.57 (t, 4H), 2.31 (m, 1H), 2.25 (t, 2H), 1.62 (q, 2H), 1.57 (m, 4H), 1.38 (m, 2H). LA-PEG_{600}-COOH: TLC (chloroform/methanol 93:7) R_f = 0.3. 1H NMR (250 MHz, CDCl3): (ppm) 4.15 (t, 2H), 3.55 (m, 44H), 3.09 (m, 2H), 2.94 (s, 1H), 2.31 (m, 1H), 2.25 (t,

2H), 1.62 (q, 2H), 1.57 (m, 4H), 1.38 (m, 2H). MALDI-MS (DHB) calcd for LA-PEG_{600}-O$(M^+Na)^+$: m/z 800.5 and for LA-PEG_{600}-COONa $(M^+Na)^+$: m/z 901.3.

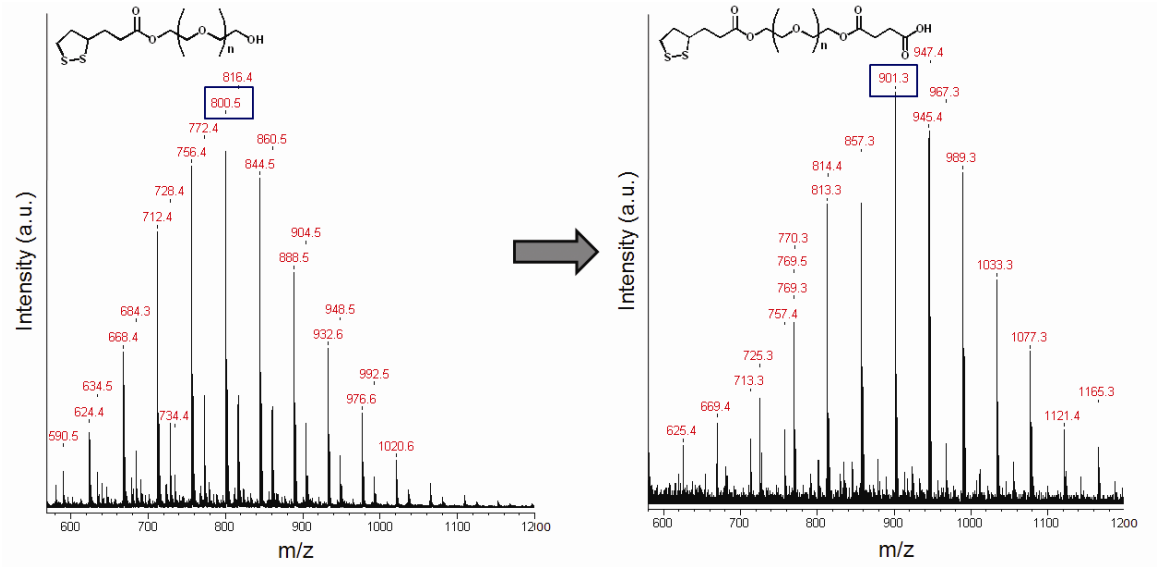

Fig. S1 Matrix-assisted desorption/ionization mass spectroscopy (MALDI-MS) analysis of LA-PEG_{600} – X (X-COOH, OH). The mean m/z of LA-PEG_{600}-OH (a) to LA-PEG_{600}-COOH (b) are measured at 800.5 and 901.3, respectively**.**

Surface modification of Co@Pt-Au

Co@Pt-Au nanoparticles (3 mg) were dispersed in 3 mL of $CHCl_3$ containing LA-PEG_{600}-OH (8 mg, 10 μmol) and LA-PEG_{600}-COOH (2 mg, 2.5 μmol) and the solution was rigorously vortexed for 30 min. Evaporation of $CHCl_3$ by using a gentle Ar strem resulted in a red waxy solid, which was then dissolved in 0.5 mL of 10 mM phosphate buffered solution (PBS, pH 7.2) and titrated with additional NaOH to pH 7. Excess ligands and salts were removed by using DeSalting column (GE healthcare, USA). The nanoparticle solution was concentrated with an appropriate amount of 10 mM PBS to be a 2 mg/ml (Co) concentration using centricon with a cut off of 1,000 kDa.

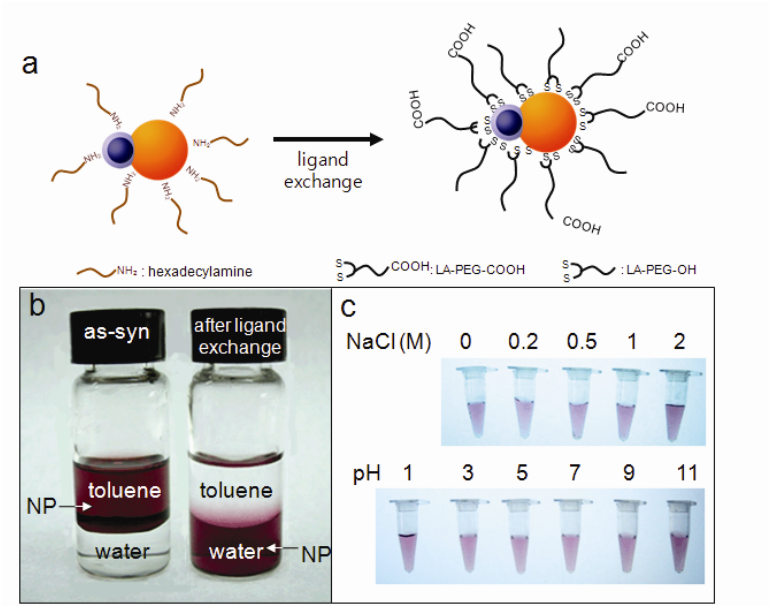

Fig. S2 (a) The schematic of ligand exchange of Co@Pt-Au nanoparticles. (b) Solubility test of synthesized (left) and ligand exchanged (right) Co@Pt-Au nanoparticles. (c) Colloidal stability test of ligand exchanged Co@Pt-Au nanoparticles against salt-concentration (~2M) and pH variation (pH 1~11).

Conjugation of nanoparticle with NTV

Co@Pt-Au nanoparticles were dissolved in 0.01 mol MES buffer with pH 6.4 and N-(3-dimethylaminopropyl)-N-ethylcarbodiimide hydrochloride (EDC) (9.6 mg, 50 µmol) and N-hydroxysulfosuccinimide (sulfo-NHS) (1.2 mg, 5 µmol) were added to the solution. After standing at room temperature for 10 min, unreacted EDC and sulfo-NHS were removed by using a DeSalting column (GE healthcare, USA) using 0.01 M PBS. Then NTV (1mg) in 50 µl PBS was added to the Co@Pt-Au solution and the reaction was carried out for 2 h at RT. Co@Pt–Au-NTV conjugates were purified by a Sephacryl S-300 column.

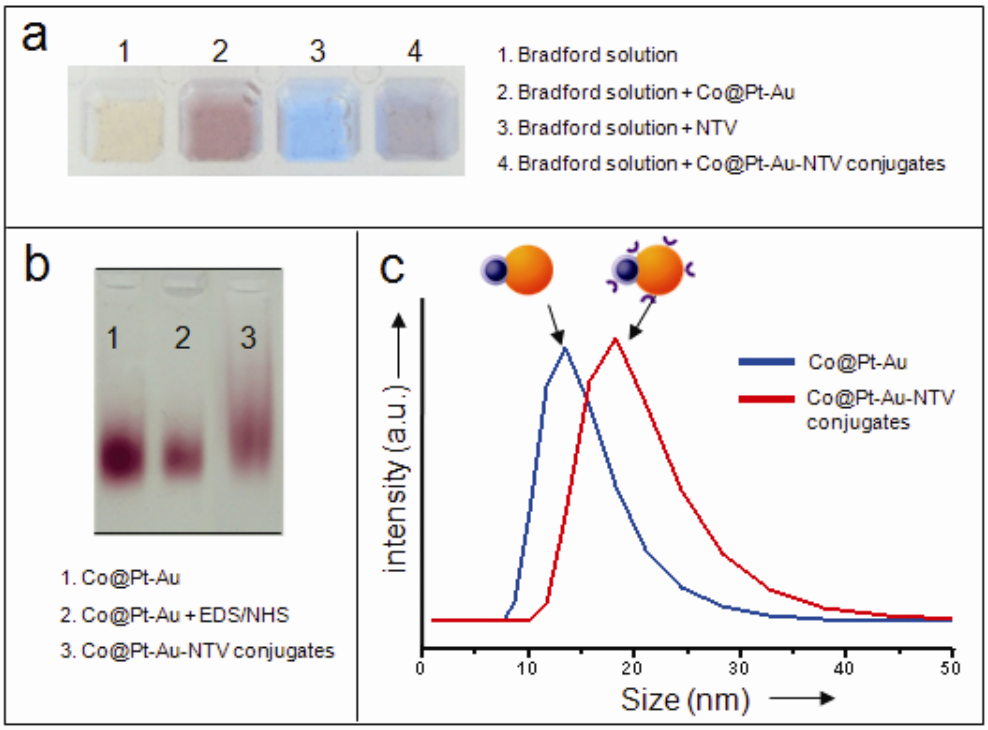

Fig. S3 (a) Bradford solution changed to pale violet color upon mixing with Co@Pt-Au-NTV, while it turned to red color upon mixing with Co@Pt-Au only (b) In gel electrophoresis analysis, nanoparticles conjugated with NTV showed a broadened and retarded band compared to that of free nanoparticles. (c) Similarly, the hydrodynamic size of Co@Pt-Au-NTV increase from 14 nm to 20 nm in DLS measurement compare to free nanoparticle.

Preparation of $A\beta_{40}$ solution

Biotinylated $A\beta_{40}$ peptides were purchased from Biosource (CA, USA). $A\beta_{40}$ solutions were prepared by following the literature method. $A\beta_{40}$ was suspended in 100 % 1,1,1,3,3,3 hexafluoro-2-propanol (HFIP) at 6 mg/mL and incubated for 30 min. at RT. HFIT was removed by using a gentle Ar stream. Then, $A\beta_{40}$ was solved in dimethyl sulfoxide (DMSO) at 5 mM concentration as a stock solution. The stock solution was diluted to 0, 0.5, 1, 5, 10, 32, 80, and 250 μM in 10 mM phosphate containing 137 mM NaCl and 27 mM KCl and kept for 2 days without shaking.

MR measurement

For the MRI detection, the Co@Pt-Au-NTV and $A\beta_{40}$ mixture was fixed by adding agarose gel with the final concentration of 0.2 %. MR imaging experiments were performed with a 3 T MRI (Achieva 3.0 T, Philips, USA) using Seuse-Flex-M coil. PCR tubes containing nanoparticle treated $A\beta_{40}$ were placed on the instrument and a T2-weighted MRI acquisition was performed by using the following parameters: field of view = 10 × 10 cm^2, point resolution of 256 × 256 μm^2, section thickness of 1 mm, TE = 100 ms, TR = 4000 ms, number of acquisitions = 2.

Disassembly experiments of Co@Pt-Au conjugated $A\beta_{40}$ assemblies

Solutions of the Co@Pt-Au conjugated $A\beta_{40}$ assemblies were diluted in PBS to 0.1 µM and stored without shaking. After 4 days, the solutions were analyzed by DLS and TEM. At 5 µM, only free nanoparticle was detected in DLS data and nanoparticles dispersed without any aggregation were observed in TEM image.

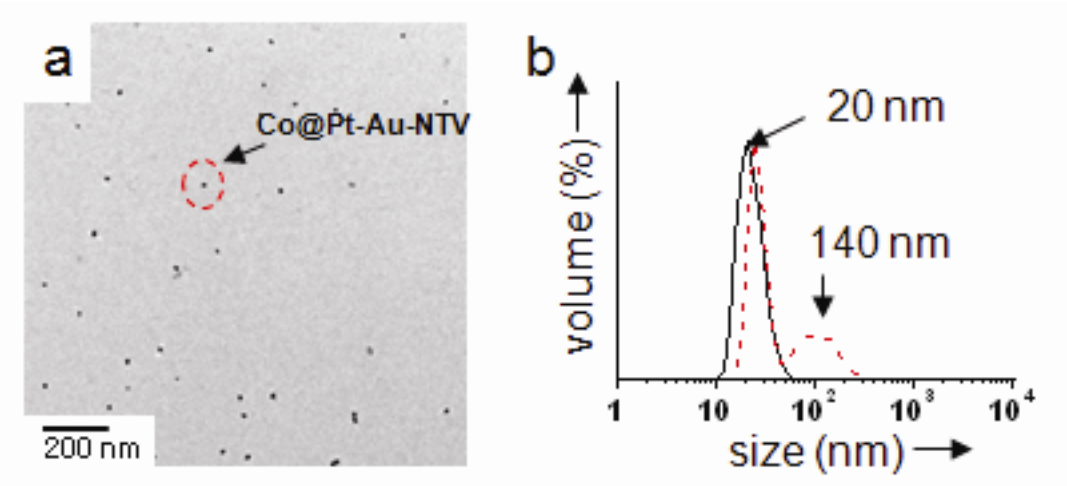

Fig. S4 Dilution on $A\beta_{40}$ assemblies. Assembled $A\beta_{40}$ is diluted to 0.1 µM for 4 days and analyzed using TEM and DLS. (a) TEM images show morphological changes of Co@Pt-Au-NTV-$A\beta_{40}$ assemblies of 5 µM which are completely dissociated after dilution. (b) Size change of Co@Pt-Au-NTV-$A\beta_{40}$ assemblies, measured with DLS also matches with the trends seen in the TEM data. (t = 0: red dotted line and t = 4 days: black solid line)