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Supplementary Information

A water-soluble gadolinium metallofullerenol: facile preparation, magnetic property and magnetic resonance imaging application

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Contents

Experiment Section

1. Materials and Instrumentation

Endohedral metallofullerene Gd@C_{82} was synthesized and purified as described. Other reagents and organic solvents were all commercially available and used as received. Water was purified with Millipore Milli-Q Synthesis purifier (18.0 MΩ cm, Barnstead). X-ray photoelectron spectroscopy (XPS) analysis was performed on ESCALab220i-XL (VG Instruments). Dynamic Light Scattering (DLS) and zeta potential were carried out on a Malvern Instruments Zetasizer Nano ZSI at a 173° scattering angles. Infrared (IR) spectra were recorded in the range from 500 to 4000 cm^{-1} with Nicolet iN10 MX IR microscopy (ThermoFisher Scientific). The UV-Vis-NIR spectrums were obtained by PerkinElmer Lambda 950. SQUID was measured on a Quantum Design MPMS XL-7. Relaxivity and MRI in vitro and vivo were performed on Niumag NMI20-Analyst and Bruker Bio Spec 7.0T/20 cm, respectively.

2. Preparation and Purification of Gd@C_{2v}-C_{82}

Soot containing gadofullerenes and empty fullerenes was prepared by arc discharge method. Graphite rods were doped with Gd/Ni alloy, and then subjected to an arc discharging generator as an anode. The production was performed under a helium atmosphere of 200 Torr and a current of 140 A. The soot obtained was extracted by DMF under reflux in argon atmosphere for 6h, and black extract was obtained after filtration. DMF was totally removed by a rotary evaporator, and the residue was extracted by toluene under ultrasonication. After filtration, a brown toluene extract was obtained for separation by High Performance Liquid Chromatography (HPLC) with Buckyprep-M column (φ 20 ×250 mm; eluent, toluene; flow rate, 12 mL/min). Toluene solution containing Gd@C_{82} of 99% purity was obtained after one-stage HPLC separation.

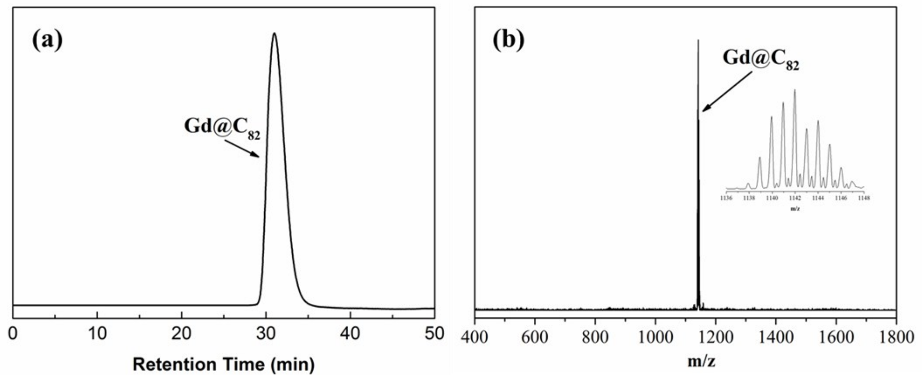

Fig. S1 (a) Chromatogram of the isolated Gd@C_{82} on a Buckyprep-M column (Chromatographic column 20 × 250 mm; flow rate 12 mL/min; toluene as eluent). (b) MALDI-TOF-MS spectrum of the isolated Gd@C_{82}.

3. Synthesis of Gd@$C_{82}(O)_{10}(OH)_{22}$.

Gd@$C_{82}(O)_{10}(OH)_{22}$ was prepared by the tetrabutylammonium hydroxide (40% in water, TBAH) phase-transfer reaction.[1] Toluene solution of purified Gd@C_{82} (~10 mg) was added 3 mL of 50 wt % NaOH solution and 10 μL of TBAH, and the mixtures were stirred at room temperature until the upper layer turned colorless. The toluene was carefully removed from the brown precipitate; with addition of 10 mL water the residue was stirred overnight to give brown aqueous solutions. 50 mL more of alcohol was added to the collected solution to precipitate the product, and red-brown sludge without excessive NaOH and TBAH was obtained by centrifugation. Then the sludge was dissolved in water and placed in a cellulose dialysis bag (MWCO = 3000) for a 7-day dialysis (water changed every 24 h) to remove the residue. Although the reaction aims to introduce hydroxyl groups, some carbonyl groups can be generated on the cage due to pinacol rearrangement.

4. Synthesis of Gd@$C_{82}(O)_{10}(OH)_{16}$ nanoparticle.

Gd@$C_{82}(O)_{10}(OH)_{16}$ nanoparticle was prepared by solid-liquid reaction. Solid Gd@C_{82} (~30 mg) was added 7 mL H_2O_2 (30% in water), 3 mL of 12 wt % NaOH solution, and was stirred at 50 ℃ in the air for 3 h to obtain a red-brown aqueous solution. More than 50 mL of alcohol was added to remove the excessive H_2O_2 and NaOH, and it gave a red-brown sludge after centrifugation. The red-brown aqueous solution of the sludge was purified in a cellulose dialysis bag (MWCO = 3000) for a 7-day dialysis (water changed every 24 h).

5. Characterization of the Gd@C_{82} metallofullernols.

X-ray photoelectron spectroscopy (XPS) was used to evaluate the chemical structure of the derivatives according to area integrals for the characteristic bonds. XPS analysis was performed on ESCALab220i-XL (VG Instruments).

Table. S1 Fitting of C1s binding energy

Bond (C1s)	C-C,C=C (284.8 eV)	C-O (286.4 eV)	C=O (288.2 eV)
Gd@$C_{82}(O)_{10}(OH)_{22}$	27266	11888	4392
Gd@$C_{82}(O)_{10}(OH)_{16}$	21588	6517	4000

Considering the relative contents of different chemical states for carbon in Table S1, the average molecular formula could be estimated as Gd@$C_{82}(O)_{10}(OH)_{22}$ and Gd@$C_{82}(O)_{10}(OH)_{16}$, respectively.

Dynamic Light Scattering (DLS) and zeta-potential measurements were performed on a NanoZS ZEN3600, Malvern instrument. Samples were filtered through a membrane with 200 nm pore size before DLS measurement. The zeta potentials were measured three times per sample in Milli-Q water at pH 7.0.

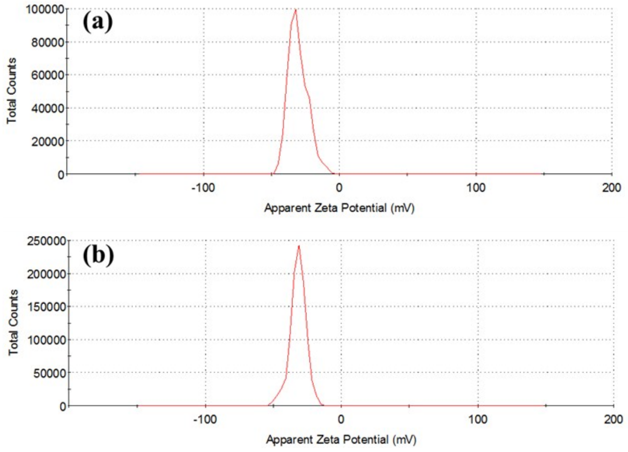

Fig. S2 Zeta potential of (a) Gd@$C_{82}(O)_{10}(OH)_{22}$ and (b) Gd@$C_{82}(O)_{10}(OH)_{16}$ is -30.5 eV and -31.8 eV, respectively.

The much negative zeta potentials suggested these nanoparticles were stable enough in water.

6. SQUID Measurements

Magnetization properties were performed on a Quantum Design MPMS XL-7 system at temperature from 5 K to 300 K at intervals of 5 K in magnetic field of 0.1 Tesla for magnetic susceptibility measurement and in 10 K with magnetic field up to 5 Tesla at intervals of 0.1 T for M-H measurement. All the solid samples were obtained by vacuum drying, and 10 mg of each dry powder was sealed in a capsule with negligible magnetism. As there existed rare residual solvents and crystal water in samples, the accurate mass of samples for SQUID measurement was calibrated by inductively coupled plasma-atomic emission spectrometry (ICP-AES, SHIMADZU ICPE-90000 instrument). 2 mg of each sample was dissolved in 5 mL of HNO_3, sonicated for 60 min and then added 5 mL of H_2SO_4/H_2O_2 (v/v = 4/1) mixture, sonicated for another 60 min. This acid solution was diluted to 100 mL and then detected the Gd^{3+} concentration (c_i, mg/L). The calibrated content of Gd in each sample for SQUID measurement was $0.5*c_i$ (mg), with which the accurate molar magnetic susceptibility (χ) of samples could be obtained. The Curie–Weiss law describes the molar magnetic susceptibility in dependence on temperature above the Curie point:

$$\chi = \frac{C}{T - T_c} + \chi_i$$

(1)

where C is a material-specific Curie constant, T is absolute temperature (measured in kelvins), T_c is the Curie temperature, and χ_i is magnetic susceptibility of the background. The carefully calibrated effective magnetic moment of samples was deduced with $\mu_{eff} = \sqrt{8C}\mu_B$, in which μ_B is Bohr magneton. The Curie–Weiss Law was perfectly fitted for all the samples; and the fitting parameters were available in Table S3.

Table. S2 The parameters of Curie–Weiss fit of molar magnetic susceptibility vs temperature plots for $Gd@C_{82}$, $Gd@C_{82}$-EDA $Gd@C_{82}(O)_{10}(OH)_{22}$ and $Gd@C_{82}(O)_{10}(OH)_{16}$.

	$Gd@C_{82}$	$Gd@C_{82}(O)_{10}(OH)_{22}$	$Gd@C_{82}(O)_{10}(OH)_{16}$
C	5.15	9.58	10.09

Tc	-0.56	-0.17	-0.057
χ_i	0.011	0.0098	0.015
R-Square	0.999	0.999	0.999
μ_{eff} (μ_B)	6.42	8.75	8.98

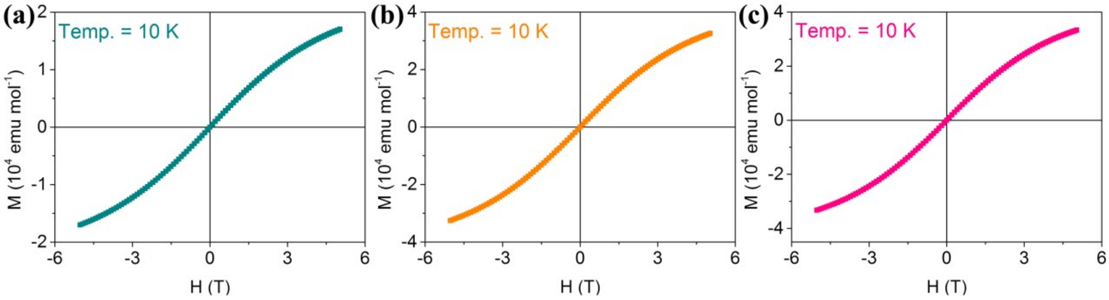

Fig. S3 Magnetization vs. field plots for (a) Gd@C_{82}, (b) Gd@$C_{82}(O)_{10}(OH)_{22}$ and (c) Gd@$C_{82}(O)_{10}(OH)_{16}$.

No hysteresis loop was observed for the magnetization curves for all the samples, indicating that metallofullerenols of Gd@C_{82} were paramagnetic.

7. T_1-Weighted Relaxivity Measurement.

Relaxivity for the metallofullerenols at 0.5 T were measured on a Niumag NMI20-Analyst (22 MHz, 0.5 T, 37 ℃) with parameters as follows: T_R = 1000 ms, T_I = 1 ms, T_I stepping = 200 ms, N_S=4 and N_{IR} = 50. The relaxivity at 7.0 T was obtained on a Bruker Bio Spec 7.0/20 cm at 25 ℃ with T_R = 300 ms, T_E = 6.1 ms, NEX=4 and FA = 90. Solutions of different concentrations were prepared by diluting a stock solution with deionized water, and the concentrations of Gd^{3+} were determined by ICP-AES.

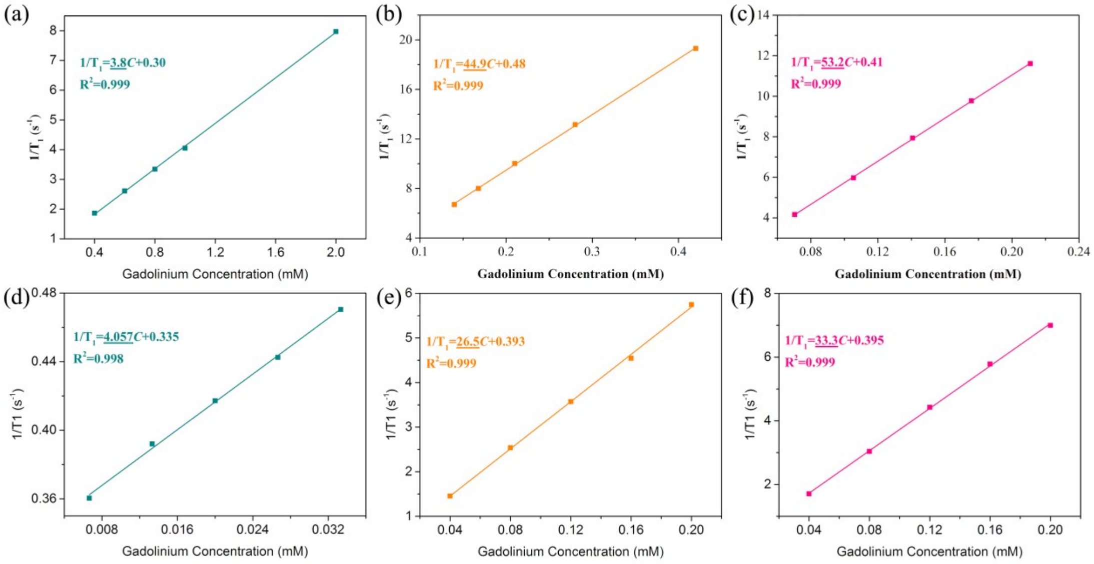

Fig. S4 Measured r_1 relaxivity values for (a) Magnevist, (b) $Gd@C_{82}(O)_{10}(OH)_{22}$, and (c) $Gd@C_{82}(O)_{10}(OH)_{16}$ at a magnetic field of 0.5 T. Measured r_1 relaxivity values for (a) Magnevist, (b) $Gd@C_{82}(O)_{10}(OH)_{22}$, and (c) $Gd@C_{82}(O)_{10}(OH)_{16}$ at a magnetic field of 7.0 T.

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