Supporting Information

"Modulation of Luminescence Intensity of Lanthanide Complexes by Photoinduced Electron Transfer and its Application to a Long-Lived Protease Probe"

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Synthetic Procedures

General Information. Boc-L-Leu-OH, HBTU, and HOBT·H_2O were obtained from Watanabe Chemical Industries, Ltd. (Japan). All other reagents were purchased from Tokyo Kasei Co., Ltd. (Japan) or Wako Pure Chemical Industries, Ltd. (Japan). They were of the highest grade available, and were used without further purification. ^{1}H-NMR and ^{13}C NMR spectra were recorded on a JEOL JNM-LA300; δ values are given in ppm relative to tetramethylsilane. Mass spectra were recorded on a JEOL JMS-SX102A mass spectrometer (EI^+), a JEOL JMS-700 mass spectrometer (FAB^+), or a JEOL JMS-T100LC (ESI^+, ESI^-). Silica gel column chromatography was performed using silica gel 60N (Kanto Kagaku Co., Ltd.). ODS column chromatography was performed using Chromatorex-ODS (Fuji Silysia Chemical Ltd.). Preparative HPLC purification was performed on a reverse-phase ODS column (GL Sciences, Inertsil Prep-ODS 30 mm x 250 mm) fitted on a JASCO PU-1587 HPLC system.

Complex Formation. Each of the cs124-DTPA derivatives (**1-13**) was reacted with $LnCl_3$ to form the corresponding complex **[Ln-X]** (**X** = **1-13**) in either of the following procedures, both of which gave the same results (65-95% yield). Formation of the complex and its purity was confirmed by mass spectroscopy and reverse-phase HPLC, respectively. The formed complexes were isolated and dissolved in DMSO (10 mM) as stock solutions.

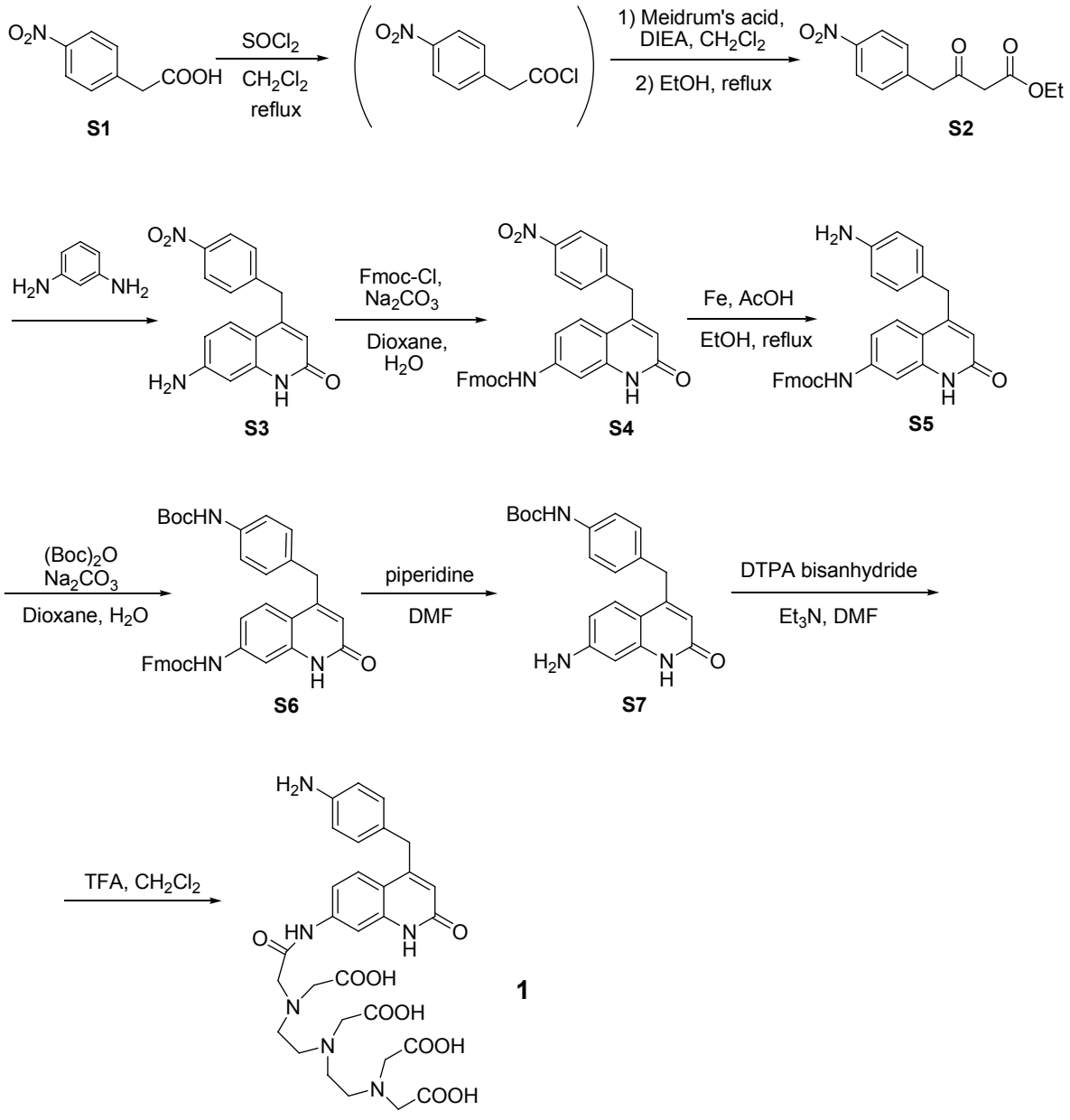
Method 1: To a solution of **X** (about 5 mg) in 200 mM $NaHCO_3$ aq (5 ml) was added an equimolar amount of $LnCl_3·6H_2O$. The solution was stirred at room temperature for 2 h. The formed complex was purified by ODS column chromatography (H_2O/MeOH = 100/0 → 50/50) to give **[Ln-X]** as a colorless solid.

Method 2: To a solution of **X** (about 5 mg) in MeOH was added a slight molar excess of $LnCl_3·6H_2O$. The solution was stirred at room temperature for 1 h, after which the solvent was evaporated. The residue was dissolved in H_2O, and loaded on a C-18 Sep-Pak cartridge (Waters), which was washed thoroughly with H_2O to remove excess metal, and eluted with MeOH. After drying,

[Ln-X] was obtained as a colorless solid.

Synthesis of 1. Compound **1** was prepared by the following procedure, as shown in Scheme S1.

Scheme S1. Synthetic scheme for **1**.



Synthesis of ethyl 4-(4-nitrophenyl)-3-oxobutanoate (S2). $SOCl_2$ (20 ml) was added to a solution of 4-nitrophenylacetic acid (**S1**) (4.40 g, 24.3 mmol) in CH_2Cl_2 (20 ml) and the suspension was refluxed for 2 h. The resulting

solution was evaporated and the residue was dissolved in CH_2Cl_2 (30 ml). This solution was added to a stirred solution of Meldrum's acid (3.50 g, 24.3 mmol) and DIEA (5.84 g, 49.8 mmol) in CH_2Cl_2 (30 ml) under Ar at 0 °C. After the addition was complete (approximately 1 h), the resulting dark orange solution was stirred for 1 h at 0 °C, then at room temperature for 2 h. The solution was diluted with CH_2Cl_2 (40 ml) and the organic phase was washed with 0.1 N HCl and brine, dried over Na_2SO_4 and evaporated to dryness. The resulting orange solid was suspended in EtOH (100 ml) and refluxed for 2 h. The solution was evaporated and the resulting orange oil was left in the freezer overnight to give a yellow solid. The crude solid was recrystallized from EtOH to afford 5.30 g (21.1 mmol) of **S2** as an orange solid, 87% yield. ^{1}H-NMR ($CDCl_3$, 300 MHz) δ 1.29 (t, 3H, J = 7.1 Hz), 3.52 (s, 2H), 4.00 (s, 2H), 4.21 (q, 2H, J = 7.1 Hz), 7.41 (d, 2H, J = 8.8 Hz), 8.21 (d, 2H, J = 8.8 Hz). ^{13}C-NMR ($CDCl_3$, 75 MHz) δ 14.1, 48.9, 49.0, 61.7, 123.8, 130.1, 130.5, 140.6, 166.8, 198.8. MS (EI^+): m/z 251 $[M]^+$.

Synthesis of 7-amino-4-(4-nitrobenzyl)-1*H*-quinolin-2-one (S3). A mixture of m-phenylenediamine (2.29 g, 21.2 mmol) and **S2** (5.30 g, 21.1 mmol) was heated at 140 °C for 16 h under Ar to afford a dark sticky mass, which was solidified by treatment with a small amount of MeOH to give a yellow product. The cooled reaction mixture was suspended in MeOH, and the undissolved solid was collected to give 1.88 g (6.34 mmol) of **S3** as an orange solid, 30% yield. ^{1}H-NMR (DMSO-d_6, 300 MHz) δ 4.15 (s, 2H), 5.75 (br, 2H), 5.91 (s, 1H), 6.35 (d, 1H, J=2.1), 6.39 (dd, 1H, J=8.4, 2.1), 7.34 (d, 1H, J=8.4), 7.55 (d, 2H, J = 8.8 Hz), 8.15 (d, 2H, J = 8.8 Hz), 11.24 (s, 1H). ^{13}C-NMR (DMSO-d_6, 75 MHz) δ 37.0, 96.8, 109.2, 110.6, 115.5, 123.6, 125.7, 129.9, 141.3, 146.1, 147.3, 149.2, 151.1, 162.3. MS (EI^+): m/z 295 $[M]^+$.

Synthesis of 7-(9-fluorenylmethoxycarbonylamino)-4-(4-nitrobenzyl)-1*H*-quinolin-2-one (S4). Compound **S3** (295 mg, 1.00 mmol) and Fmoc-Cl (297 mg, 1.23 mmol) were dissolved in 0.5 N Na_2CO_3 aq-dioxane (1:2, v/v, 60 ml) and the solution was stirred for 12 h at room temperature; a light, white precipitate was formed. The resultant suspension was diluted with H_2O and

the precipitate was collected by filtration and washed with H_2O and MeOH. The crude solid was recrystallized from EtOH to afford 410 mg (0.79 mmol) of **S4** as a colorless solid, 79% yield. ^{1}H-NMR (DMSO-d_6, 300 MHz) δ 4.29-4.33 (m, 3H), 4.48 (d, 2H, J = 6.8 Hz), 6.20 (s, 1H, k), 7.12 (d, 1H, J = 8.7 Hz), 7.31-7.44 (m, 4H), 7.55-7.62 (m, 3H), 7.66 (s, 1H), 7.74 (d, 2H, J = 7.5 Hz), 7.89 (d, 2H, J = 7.5 Hz), 8.17 (d, 2H, J = 8.7 Hz), 10.02 (s, 1H), 11.65 (s, 1H). ^{13}C-NMR (DMSO-d_6,75 MHz) δ 36.8, 46.5, 65.8, 103.6, 113.0, 113.9, 119.7, 120.2, 123.6, 125.1, 125.5, 127.1, 127.7, 130.0, 140.0, 140.8, 141.1, 143.7, 146.2, 146.8, 148.9, 153.2, 161.9. HRMS (ESI$^+$): calcd for $[M+Na]^+$, 540.1535; found, 540.1500.

Synthesis of 4-(4-aminobenzyl)-7-(9-fluorenylmethoxycarbonylamino)-1*H*-quinolin-2-one (S5). A mixture of **S4** (420 mg, 0.81 mmol), iron powder (273 mg, 4.88 mmol), EtOH (60 ml) and glacial AcOH (20 ml) was refluxed under Ar for 2 h with vigorous stirring. After the mixture had cooled to room temperature, the solvent was evaporated. The residue was purified using silica gel column chromatography (MeOH/AcOEt = 1/20) to give 256 mg (0.53 mmol) of **S5** as a pale yellow solid, 65% yield. ^{1}H NMR (DMSO-d_6, 300 MHz) δ 3.90 (s, 2H), 4.31 (t, 1H, J = 6.6 Hz), 4.48 (d, 2H, J = 6.6 Hz), 4.92 (br, 2H), 6.06 (s, 1H), 6.49 (d, 2H, J = 8.1 Hz), 6.91 (d, 2H, J = 8.1Hz), 7.12 (d, 1H, J = 9.2 Hz), 7.32-7.45 (m, 4H), 7.63-7.66 (m, 2H), 7.75 (d, 2H, J = 7.1 Hz), 7.90 (d, 2H, J = 7.9 Hz), 10.00 (s, 1H), 11.53 (s, 1H). ^{13}C-NMR (DMSO-d_6, 75 MHz) δ 36.6, 46.5, 65.8, 103.5, 112.8, 114.0, 114.3, 118.7, 120.2, 124.8, 125.1, 125.5, 127.1, 127.7, 129.3, 139.9, 140.8, 143.7, 147.1, 151.2, 153.2, 162.1. HRMS (ESI$^+$): calcd for $[M+H]^+$, 488.1974; found, 488.1943.

Synthesis of 7-(9-fluorenylmethoxycarbonylamino)-4-[4-(tert-butoxycarbonylamino)benzyl]-1*H*-quinolin-2-one (S6). A solution of **S5** (200 mg, 0.41 mmol) and $(Boc)_2O$ (300 μl, 1.30 mmol) in 0.5 N Na_2CO_3 aq.-dioxane (1:2 v/v, 30 ml) was stirred for 12 h at room temperature. Then, it was diluted with H_2O, and the resulting aqueous phase was extracted with AcOEt. The combined organic phase was washed with brine, dried over Na_2SO_4 and evaporated to dryness. The solid was purified by silica gel

column chromatography (n-hexane/AcOEt = 3/7) to give 111 mg (0.18 mmol) of **S6** as a colorless solid, 44% yield. ^{1}H-NMR (DMSO-d_6, 300 MHz) δ 1.52 (s, 9H), 4.09 (s, 2H), 4.38 (t, 1H, J = 6.6 Hz), 4.55 (d, 2H, J = 6.6 Hz), 6.15 (s, 1H), 7.17-7.23 (m, 3H), 7.39-7.51 (m, 6H), 7.68-7.71 (m, 2H), 7.82 (d, 2H, J = 7.6 Hz), 7.97 (d, 2H, J = 7.6 Hz), 9.34 (s, 1H), 10.07 (s, 1H), 11.64 (s, 1H). ^{13}C-NMR (DMSO-d_6, 75 MHz) δ 28.1, 36.7, 46.6, 65.8, 78.9, 103.6, 112.9, 114.1, 118.3, 119.0, 120.2, 125.2, 125.5, 127.1, 127.7, 129.1 131.7, 137.9, 140.0, 140.8, 140.9, 143.7, 150.5, 152.8, 153.3, 162.1. MS (FAB$^+$): m/z 588 $[M+H]^+$.

Synthesis of 7-Amino-4-[4-(tert-butoxycarbonylamino)benzyl]-1*H*-quinolin-2-one (S7). Piperidine (1 ml) was added to a solution of **S6** (104 mg, 0.18 mmol) in DMF (10 ml) and the solution was stirred at room temperature for 2 h. After removal of the solvent, the residue was purified by silica gel column chromatography (MeOH/AcOEt = 1/20) to afford 63 mg (0.17 mmol) of **S7** as a light yellow solid, 97% yield. ^{1}H-NMR (DMSO-d_6, 300 MHz) δ 1.39 (s, 9H), 3.87 (s, 2H), 5.66 (s, 2H), 5.74 (s, 1H), 6.28 (d, 1H, J = 2.0 Hz), 6.32 (dd, 1H, J = 9.6, 2.0 Hz), 7.06 (d, 2H, J = 8.4 Hz), 7.28-7.33 (m, 3H), 9.20 (s, 1H), 11.11 (s, 1H). ^{13}C-NMR (DMSO-d_6, 75 MHz) δ 28.1, 36.8, 78.9, 96.8, 109.4, 110.4, 114.9, 118.2, 125.7, 129.0, 132.2, 137.8, 141.1, 150.7, 150.9, 152.8, 162.4. MS (FAB$^+$): m/z 366 $[M+H]^+$.

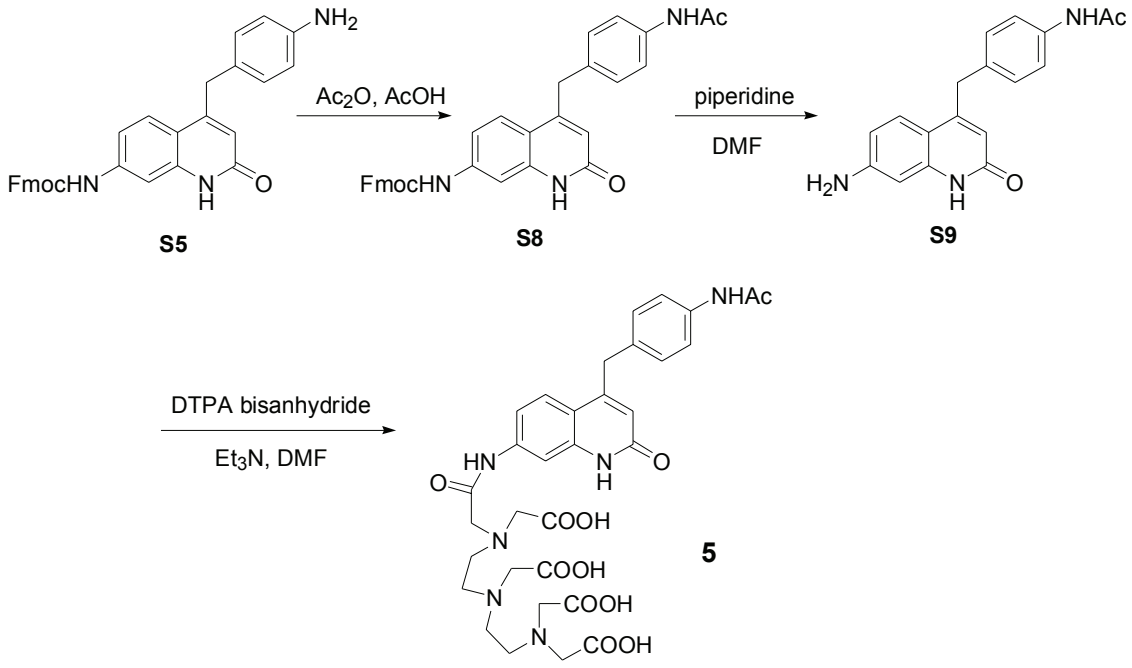
Synthesis of N-{N-[4-(4-aminobenzyl)-1,2-dihydro-2-oxoquinolin-7-yl]carbamoylmethyl}-N,N',N'',N''-diethylenetriaminetetraacetic acid (1). To a solution of DTPA bisanhydride (44 mg, 0.12 mmol) in DMF (10 ml) and triethylamine (400 μl) was added dropwise a solution of **S7** (37 mg, 0.10 mmol) in DMF (10 ml). The solution was stirred at 0 °C for 2 h and the reaction was quenched by adding 5 ml of H_2O. The reaction mixture was evaporated to dryness. The remaining solid was dissolved in CH_2Cl_2-TFA (1:1 v/v, 10 ml) and stirred for 1 h at room temperature. The residue was purified by preparative reverse-phase HPLC; linear gradient from 15% to 30% solvent B (solvent A, 0.1 M triethylammonium acetate (pH 6.5); solvent B, 80% acetonitrile/ 20% 0.1 M triethylammonium acetate (pH 6.5)). Compound **1** (19

mg, 0.03 mmol) was obtained as a colorless solid, 30% yield. ^{1}H-NMR (D_2O/ KD_2PO_4-K_2DPO_4, 300 MHz) δ 2.85-3.10 (m, 8H), 3.17 (s, 2H), 3.25 (m, 4H), 3.39 (s, 2H), 3.44 (s, 2H), 3.89 (s, 2H), 6.24 (s, 1H), 6.59 (d, 2H, J = 8.1 Hz), 6.92 (d, 2H, J = 8.1 Hz), 7.13 (d, 1H, J = 8.3 Hz), 7.54 (m, 2H). ^{13}C-NMR (D_2O/ KD_2PO_4-K_2DPO_4, 75 MHz) δ 38.2, 51.9, 52.6, 52.7, 53.2, 56.9, 59.1, 60.3, 60.7, 108.0, 117.4, 118.0, 119.1, 127.2, 130.1, 131.1, 134.8, 139.5, 140.6, 146.0, 155.0, 165.7, 173.7, 175.1, 176.3, 180.0. HRMS (FAB$^+$): calcd for $[M+H]^+$, 641.2571; found, 641.2587.

Synthesis of [Ln-1]. **[Ln-1]** was synthesized by the method 2 described in Complex Formation. **[Eu-1]**: HRMS (ESI$^+$): calcd for $[M+H]^+$, 791.1549; found, 791.1507. **[Tb-1]**: HRMS (ESI$^+$): calcd for $[M+H]^+$, 797.1548; found, 797.1590.

Synthesis of 5. Compound **5** was prepared according to the following procedure, as shown in Scheme S2.

Scheme S2. Synthetic scheme for **5**.



Synthesis of 4-(4-acetylaminobenzyl)-7-(9-fluorenylmethoxycarbonylamino) -1*H*-quinolin-2-one (S8). Compound **S5** was synthesized according to the procedure described above. A solution of **S5** (200 mg, 0.41 mmol), Ac_2O (10 ml), AcOH (50 ml) was refluxed for 30 min. Then, the reaction mixture was evaporated to dryness. The resulting solid was purified by silica gel column chromatography (MeOH/AcOEt = 1/20) to give 169 mg (0.32 mmol) of **S8** as a colorless solid, 78% yield. ^{1}H-NMR (DMSO-d_6, 300 MHz) δ 2.00 (s,3H), 3.95 (s, 2H), 5.71 (s, 2H), 5.81 (s, 1H), 6.34 (d, 1H, J = 2.1 Hz), 6.39 (dd, 1H, J = 8.6, 2.1 Hz), 7.16 (d, 2H, J = 8.4 Hz), 7.37 (d, 1H, J = 8.6 Hz), 7.48 (d, 2H, J = 8.4 Hz), 9.86 (s, 1H), 11.16 (s, 1H). ^{13}C-NMR (DMSO-d_6, 75 MHz) δ 24.0, 36.7, 46.6, 65.8, 103.6, 112.9, 120.2, 125.2, 125.5, 127.1, 127.7, 129.1, 132.8, 137.8, 140.0, 140.8, 140.9, 143.7, 150.3, 153.2, 162.1, 168.1, 171.3. MS (FAB^+): m/z 530 $[M+H]^+$.

Synthesis of 4-(4-acetylaminobenzyl)-7-amino-1*H*-quinolin-2-one (S9). The procedure followed was the same as described for **S7**, using piperidine (1 ml) and **S8** (167 mg, 0.32 mmol) in DMF (10 ml), to afford 94 mg (0.31 mmol) of **S9** as a light yellow solid, 97% yield. ^{1}H-NMR (DMSO-d_6, 300 MHz) δ 2.00 (s, 3H), 4.04 (s, 2H), 4.31 (t, 1H, J = 6.9 Hz), 4.47 (d, 2H, J = 6.9 Hz), 6.09 (s, 1H), 7.14 (d, 1H, J = 8.4 Hz), 7.18 (d, 2H, J = 8.7 Hz), 7.31-7.44 (m, 4H), 7.63 (d, 2H, J = 8.7 Hz), 7.61-7.65 (m, 2H), 7.75 (d, 2H, J = 7.6 Hz), 7.90 (d, 2H, J = 7.5 Hz), 9.89 (s, 1H), 10.00 (s, 1H), 11.57 (s, 1H). ^{13}C-NMR (DMSO-d_6, 75 MHz) δ 23.9, 36.8, 96.8, 109.4, 110.4, 114.9, 119.1, 125.7, 129.0, 133.2, 137.6, 141.1, 150.6, 150.9, 162.4, 168.1. MS (FAB^+): m/z 308 $[M+H]^+$.

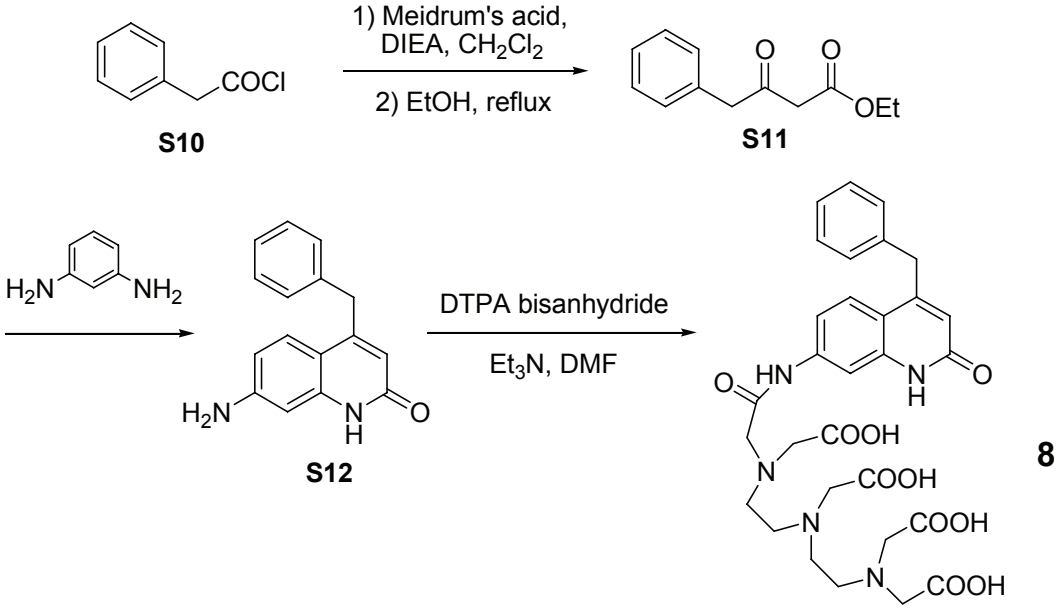
Synthesis of N-{N-[4-(4-acetylaminobenzyl)-1,2-dihydro-2-oxoquinolin-7-yl]carbamoylmethyl}-N,N',N'',N''-diethylenetriaminetetraacetic acid (5). To a solution of DTPA bisanhydride (43 mg, 0.12 mmol) in DMF (10 ml) and trietylamine (400 μl) was added dropwise a solution of **S9** (31 mg, 0.10 mmol) in DMF (10 ml). The solution was stirred at 0 °C for 2 h and the reaction was quenched by adding 5 mL of H_2O. The reaction mixture was evaporated. The residue was purified by preparative reversed-phase HPLC; linear gradient from 15% to 30% solvent B (solvent A, 0.1 M triethylammonium acetate (pH 6.5);

solvent B, 80% acetonitrile/ 20% 0.1 M triethylammonium acetate (pH 6.5)). Compound **5** (30 mg, 0.04 mmol) was obtained as a colorless solid, 40 % yield. ^{1}H-NMR (D_2O/ KD_2PO_4-K_2DPO_4, 300 MHz) δ 1.91 (s, 3H), 2.85-3.10 (m, 8H), 3.17 (s, 2H), 3.27 (m, 4H), 3.38 (s, 2H), 3.43 (s, 2H), 3.72 (s, 2H), 6.14 (s, 1H), 6.94 (d, 2H, J = 7.9 Hz), 7.05 (m, 3H), 7.33 (d, 1H, J = 8.0 Hz), 7.43 (s, 1H). ^{13}C-NMR (D_2O/ KD_2PO_4-K_2DPO_4, 75 MHz) δ 23.8, 37.8, 51.5, 52.0, 52.1, 52.6, 56.5, 58.5, 59.6, 60.0, 106.8, 116.3, 117.0, 118.6, 121.7, 126.3, 130.4, 135.0, 136.5, 138.9, 140.0, 153.3, 164.8, 172.2, 172.9, 174.6, 175.6, 178.8. HRMS (FAB^+): calcd for $[M+H]^+$, 683.2677; found, 683.2673.

Synthesis of [Ln-5]. **[Ln-5]** was prepared by a similar method to that described for **[Ln-1]**. **[Eu-5]**: HRMS (ESI^+): calcd for $[M+H]^+$, 833.1654; found, 833.1645. **[Tb-5]**: HRMS (ESI^+): calcd for $[M+H]^+$, 839.1696; found, 839.1658.

Synthesis of 8. Compound **8** was prepared according to the following procedure, as shown in Scheme S3.

Scheme S3. Synthetic scheme for **8**.



Synthesis of ethyl 4-phenyl-3-oxobutanoate (S11). Phenylacetyl chloride (**S10**) (2.32 g, 15.0 mmol) was added to a stirred solution of Meldrum's acid (2.16 g, 14.9 mmol) and DIEA (3.88 g, 30.0 mmol) in CH_2Cl_2 (30 ml) under Ar at 0 °C. After the addition was complete (approximately 1 h), the resulting dark orange solution was stirred for 1 h at 0 °C, then at room temperature for 2 h. The solution was diluted with CH_2Cl_2 (40 ml) and the organic phase was washed with 0.1 N HCl and brine, dried over Na_2SO_4 and evaporated to dryness. The resulting orange solid was suspended in EtOH (100 ml) and refluxed for 2 h. The solution was evaporated to afford 2.74 g (13.3 mmol) of **S11** as an orange oil, 88% yield. ^{1}H-NMR ($CDCl_3$, 300 MHz) δ 1.16 (t, 3H, J = 6.9 Hz), 3.64 (s, 2H), 3.85 (s, 2H), 4.06 (q, 2H, J = 6.9 Hz), 7.00-7.41 (m, 5H). ^{13}C-NMR ($CDCl_3$, 75 MHz) δ 13.8, 48.0, 49.7, 61.1, 127.0, 128.5, 129.3, 133.0, 166.9, 200.3. MS (FAB^+): m/z 207 $[M+H]^+$.

Synthesis of 7-amino-4-benzyl-1*H*-quinolin-2-one (S12). The procedure was the same as that for **S3**, using m-phenylenediamine (628 mg, 5.82 mmol) and **S11** (1.20 g, 5.82 mmol) to give 555 mg (2.22 mmol) of **S12** as an orange solid, 38% yield. ^{1}H-NMR (DMSO-d_6, 300 MHz) δ 4.01 (s, 2H), 5.73 (br, 2H), 5.83 (s, 1H), 6.34 (d, 1H, J = 2.1 Hz), 6.38 (dd, 1H, J = 8.7, 2.1 Hz), 7.17-7.32 (m, 5H), 7.41 (d, 2H, J = 8.7 Hz), 11.18 (s, 1H). ^{13}C-NMR (DMSO-d_6, 75 MHz) δ 37.3, 96.9, 109.5, 110.4, 115.0, 115.1, 125.7, 126.3, 128.4, 128.8, 138.8, 141.2, 150.5, 151.0, 162.4. MS (FAB^+): m/z 251 $[M+H]^+$.

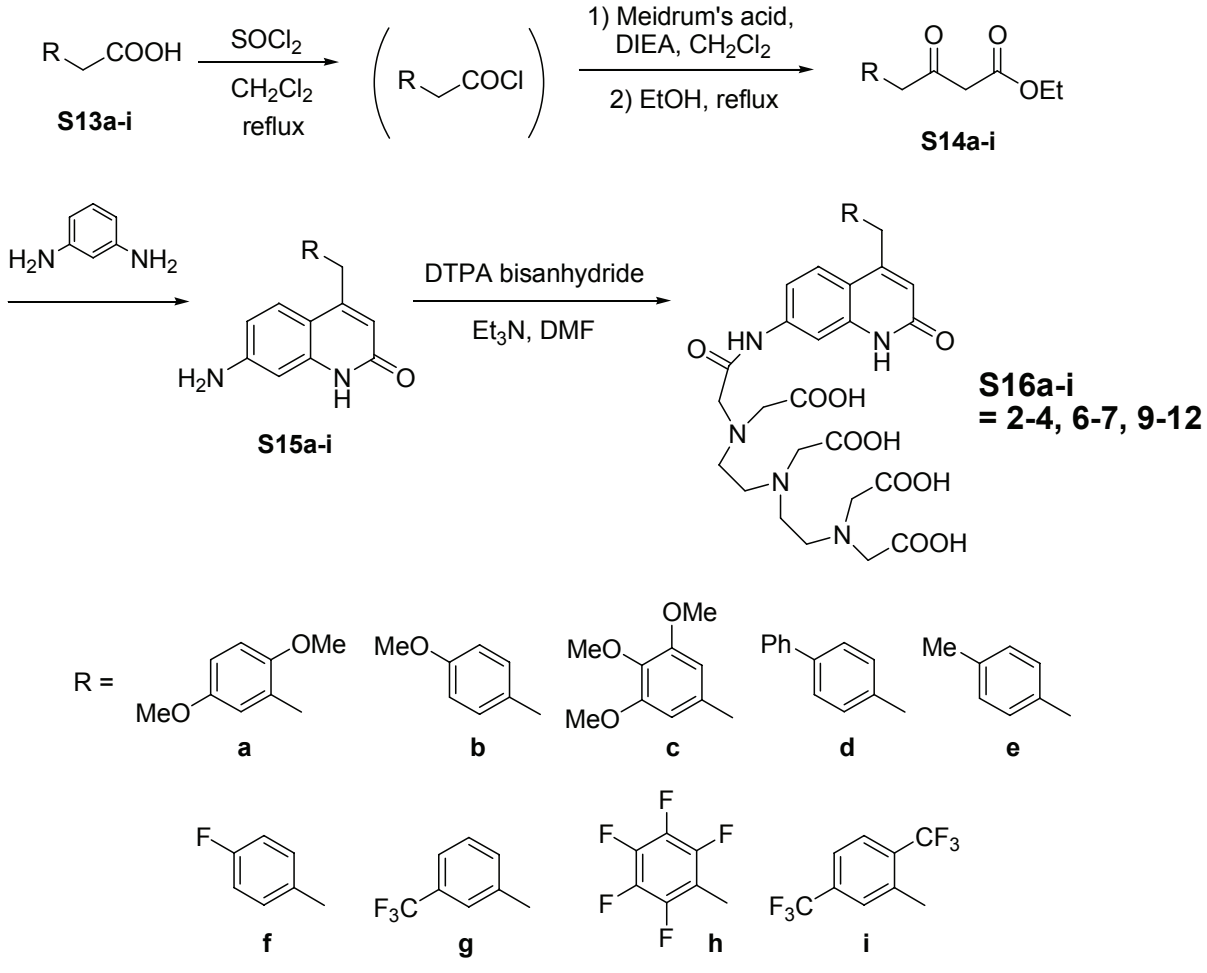
Synthesis of N-{N-[4-benzyl-1,2-dihydro-2-oxoquinolin-7-yl]carbamoylmethyl}-N,N',N'',N''-diethylenetriaminetetraacetic acid (8). The procedure was the same as that for **5**, using DTPA bisanhydride (63 mg, 0.18 mmol) in DMF (10 ml), triethylamine (400 µl) and **S12** (38 mg, 0.15 mmol) in DMF (10 ml). The residue was purified by preparative reverse-phase HPLC under the same conditions as described for **5**. Compound **8** (37 mg, 0.06 mmol) was obtained as a colorless solid, 40% yield. ^{1}H-NMR (D_2O/ KD_2PO_4-K_2DPO_4, 300 MHz) δ 2.84-3.15 (m, 8H), 3.17 (s, 2H), 3.26 (m, 4H), 3.38 (s, 2H), 3.43 (s, 2H), 3.91 (s, 2H), 6.19 (s, 1H), 7.12-7.23 (m, 6H), 7.44 (m, 2H). ^{13}C-NMR (D_2O/ KD_2PO_4-K_2DPO_4, 75 MHz) δ 38.4, 52.3, 52.4, 53.3,

53.4, 58.8, 59.5, 60.1, 60.2, 108.8, 116.7, 117.8, 118.9, 126.5, 127.5, 129.6, 129.7, 139.1, 139.7, 140.9, 153.0, 166.3, 174.2, 179.6, 180.1, 180.2. HRMS (FAB^+): calcd for $[M+H]^+$, 626.2462; found, 626.2458.

Synthesis of [Ln-8]. **[Ln-8]** was prepared by the method 1 described in Complex Formation. **[Eu-8]**: HRMS (ESI^+): calcd for $[M+H]^+$, 776.1440; found, 776.1463. **[Tb-8]**: HRMS (ESI^+): calcd for $[M+H]^+$, 782.1481; found, 782.1443.

Synthesis of 2-4, 6-7, 9-12. Compounds **2-4, 6-7, 9-12** were prepared according to the same procedure as used for **8**. Scheme S4 shows the structures of the compounds.

Scheme S4. Synthetic scheme for **2-4, 6-7, 9-12**.



Synthesis of S14a-i. The following compounds were prepared according to the same procedure as employed for the preparation of **S11**.

S14a: Yield 94%. ^{1}H-NMR (DMSO-d_6, 300 MHz) δ 1.16 (t, 3H, J = 7.0 Hz), 3.53 (s, 2H), 3.67 (s, 3H), 3.69 (s, 3H), 3.71 (s, 2H), 4.06 (q, 2H, J = 7.0 Hz), 6.74 (d, 1H, J = 2.9 Hz), 6.79 (dd, 1H, J = 8.8, 2.9 Hz), 6.89 (d, 1H, J = 8.8 Hz). MS (FAB$^+$): m/z 267 $[M+H]^+$.

S14b: Yield 87%. ^{1}H-NMR (DMSO-d_6, 300 MHz) δ 1.16 (t, 3H, J = 7.1 Hz), 3.60 (s, 2H), 3.72 (s, 3H), 3.77 (s, 2H), 4.05 (q, 2H, J = 7.1 Hz), 6.86 (d, 2H, J = 8.6 Hz), 7.08 (d, 2H, J = 8.6 Hz). MS (FAB $^+$): m/z 237 $[M+H]^+$.

S14c: Yield 97%. ^{1}H-NMR (DMSO-d_6, 300 MHz) δ 1.18 (t, 3H, J = 7.1 Hz), 3.57 (s, 2H), 3.62 (s, 3H), 3.73 (s, 6H), 3.76 (s, 2H), 4.07 (q, 2H, J = 7.1 Hz), 6.56 (s, 2H). MS (FAB $^+$): m/z 297 $[M+H]^+$.

S14d: Yield 97%. ^{1}H-NMR (DMSO-d_6, 300 MHz) δ 1.16 (t, 3H, J = 7.0 Hz), 3.68 (s, 2H), 3.91 (s, 2H), 4.06 (q, 2H, J = 7.0 Hz), 7.32-7.35 (m, 2H), 7.42-7.47 (m, 2H), 7.58-7.65 (m, 5H). MS (FAB $^+$): m/z 283 $[M+H]^+$.

S14e: Yield 92%. ^{1}H-NMR (DMSO-d_6, 300 MHz) δ 1.16 (t, 3H, J = 7.1 Hz), 2.26 (s, 3H), 3.61 (s, 2H), 3.78 (s, 2H), 4.05 (q, 2H, J = 7.1 Hz), 7.04 (d, 2H, J = 8.0 Hz), 7.11 (d, 2H, J = 8.0 Hz). MS (FAB $^+$): m/z 221 $[M+H]^+$.

S14f: Yield 87%. ^{1}H-NMR (DMSO-d_6, 300 MHz) δ 1.16 (t, 3H, J = 7.1 Hz), 3.65 (s, 2H), 3.87 (s, 2H), 4.07 (q, 2H, J = 7.1 Hz), 7.08-7.20 (m, 4H). MS (FAB $^+$): m/z 225 $[M+H]^+$.

S14g: Yield 96%. ^{1}H-NMR (DMSO-d_6, 300 MHz) δ 1.17 (t, 3H, J = 7.1 Hz), 3.70 (s, 2H), 4.02 (s, 2H), 4.07 (q, 2H, J = 7.1 Hz), 7.47 (d, 1H, J = 6.6 Hz), 7.54-7.62 (m, 3H). MS (FAB $^+$): m/z 275 $[M+H]^+$.

S14h: Yield 97%. ^{1}H-NMR (DMSO-d_6, 300 MHz) δ 1.18 (t, 3H, J = 6.9 Hz), 3.82 (s, 2H), 4.09 (q, 2H, J = 6.9 Hz), 4.16 (s, 2H). MS (FAB $^+$): m/z 297 $[M+H]^+$.

S14i: Yield 91%. ^{1}H-NMR (DMSO-d_6, 300 MHz) δ 1.18 (t, 3H, J = 7.1 Hz), 3.74 (s, 2H), 4.09 (q, 2H, J = 7.1 Hz), 4.28 (s, 2H), 7.86 (m, 2H), 7.95 (d, 1H, J = 7.9 Hz). MS (FAB $^+$): m/z 343 $[M+H]^+$.

Synthesis of S15a-i. The following compounds were prepared according to

the same procedure as employed for the preparation of **S12**.

S15a: Yield 45%. ^{1}H-NMR (DMSO-d_6, 300 MHz) δ 3.63 (s, 3H), 3.73 (s, 3H), 3.92 (s, 2H), 5.59 (s, 1H), 5.74 (br, 2H), 6.36 (d, 1H, J = 2.0 Hz), 6.40 (dd, 1H, J = 8.6, 2.0 Hz), 6.65 (d, 1H, J = 3.1 Hz), 6.79 (dd, 1H, J = 8.8, 3.1 Hz), 6.95 (d, 1H, J = 8.8 Hz), 7.38 (d, 1H, J = 8.6 Hz), 11.17 (s, 1H). MS (ESI$^+$): m/z 311 $[M+H]^+$.

S15b: Yield 55%. ^{1}H-NMR (DMSO-d_6, 300 MHz) δ 3.70 (s, 3H), 3.94 (s, 2H), 5.72 (br, 2H), 5.80 (s, 1H), 6.34 (d, 1H, J = 2.0 Hz), 6.38 (dd, 1H, J = 8.8, 2.0 Hz), 6.86 (d, 2H, J = 8.6 Hz), 7.17 (d, 2H, J = 8.6 Hz), 7.40 (d, 1H, J = 8.8 Hz), 11.17 (s, 1H). MS (ESI$^+$): m/z 281 $[M+H]^+$.

S15c: Yield 6%. ^{1}H-NMR (DMSO-d_6, 300 MHz) δ 3.61 (s, 3H), 3.71 (s, 6H), 3.94 (s, 2H), 5.74 (br, 2H), 5.81 (s, 1H), 6.34 (d, 1H, J = 2.0 Hz), 6.41 (dd, 1H, J = 8.8, 2.0 Hz), 6.58 (s, 2H), 7.46 (d, 1H, J = 8.8 Hz), 11.17 (s, 1H). MS (ESI$^+$): m/z 341 $[M+H]^+$.

S15d: Yield 40%. ^{1}H-NMR (DMSO-d_6, 300 MHz) δ 4.06 (s, 2H), 5.75 (br, 2H), 5.90 (s, 1H), 6.36 (d, 1H, J = 2.0 Hz), 6.40 (dd, 1H, J = 8.6, 2.0 Hz), 7.32-7.44 (m, 6H), 7.57-7.63 (m, 4H), 11.21 (s, 1H). MS (ESI$^+$): m/z 327 $[M+H]^+$.

S15e: Yield 56%. ^{1}H-NMR (DMSO-d_6, 300 MHz) δ 2.24 (s, 3H), 3.96 (s, 2H), 5.72 (br, 2H), 5.81 (s, 1H), 6.34 (d, 1H, J = 2.0 Hz), 6.37 (dd, 1H, J = 8.6, 2.0 Hz), 7.07-7.15 (m, 4H), 7.38 (d, 1H, J = 8.6 Hz), 11.17 (s, 1H). MS (ESI$^+$): m/z 265 $[M+H]^+$.

S15f: Yield 57%. ^{1}H-NMR (DMSO-d_6, 300 MHz) δ 4.01 (s, 2H), 5.74 (br, 2H), 5.82 (s, 1H), 6.35 (d, 1H, J = 2.0 Hz), 6.38 (dd, 1H, J = 8.6, 2.0 Hz), 7.08-7.13 (m, 2H), 7.27-7.32 (m, 2H), 7.38 (d, 1H, J = 8.6 Hz), 11.20 (s, 1H). MS (ESI$^+$): m/z 269 $[M+H]^+$.

S15g: Yield 50%. ^{1}H-NMR (DMSO-d_6, 300 MHz) δ 4.14 (s, 2H), 5.77 (br, 2H), 5.85 (s, 1H), 6.36 (d, 1H, J = 2.0 Hz), 6.39 (dd, 1H, J = 8.6, 2.0 Hz), 7.41 (d, 1H, J = 8.6 Hz), 7.50-7.64 (m, 4H), 11.24 (s, 1H). MS (ESI$^+$): m/z 319 $[M+H]^+$.

S15h: Yield 42%. ^{1}H-NMR (DMSO-d_6, 300 MHz) δ 4.16 (s, 2H), 5.49 (s, 1H), 5.85 (br, 2H), 6.38 (d, 1H, J = 2.2 Hz), 6.49 (dd, 1H, J = 8.8, 2.2 Hz), 7.53 (d, 1H, J = 8.8 Hz), 11.27 (s, 1H). MS (ESI$^+$): m/z 341 $[M+H]^+$.

S15i: Yield 23%. ^{1}H-NMR (DMSO-d_6, 300 MHz) δ 4.32 (s, 2H), 5.25 (s, 1H),

5.83 (br, 2H), 6.38 (d, 1H, J = 2.0 Hz), 6.44 (dd, 1H, J = 8.9, 2.0 Hz), 7.37 (d, 1H, J = 8.9 Hz), 7.77 (s, 1H), 7.91 (d, 1H, J = 7.9 Hz), 8.05 (d, 1H, J = 7.9 Hz), 11.27 (s, 1H). MS (ESI^+): m/z 387 $[M+H]^+$.

Synthesis of S16a-i (= 2-4, 6-7, 9-12) and their lanthanide complexes. The following compounds were prepared according to the same procedure as employed for the preparation of **8**. They were reacted with $LnCl_3$ to give the complexes **[Ln-X]** (**X** = **2-4**, **6-7**, **9-12**) in the manner described above (method 1 in Complex Formation).

S16a (2): Yield 21%. ^{1}H-NMR (DMSO-d_6, 300 MHz) δ 2.87-3.01 (m, 8H), 3.42 (s, 4H), 3.46 (m, 4H), 3.52 (s, 2H), 3.64 (s, 3H), 3.73 (s, 3H), 4.02 (s, 2H), 5.84 (s, 1H), 6.70 (d, 1H, J = 3.1 Hz), 6.81 (dd, 1H, J = 8.9, 3.1 Hz), 6.96 (d, 1H, J = 8.9 Hz), 7.35 (d, 1H, J = 8.6 Hz), 7.68 (d, 1H, J = 8.6 Hz), 7.92 (s, 1H), 10.32 (s, 1H), 11.57 (s, 1H). HRMS (ESI^+): calcd for $[M+H]^+$, 686.2674; found 686.2706.

S16b (3): Yield 29%. ^{1}H-NMR (DMSO-d_6, 300 MHz) δ 2.87-2.91 (m, 4H), 2.97 (m, 4H), 3.38 (s, 4H), 3.43 (m, 4H), 3.53 (s, 2H), 3.71 (s, 3H), 4.05 (s, 2H), 6.08 (s, 1H), 6.86 (d, 1H, J = 8.5 Hz), 7.20 (d, 1H, J = 8.5 Hz), 7.31 (d, 1H, J = 8.9 Hz), 7.68 (d, 1H, J = 8.9 Hz), 7.91 (s, 1H), 10.57 (s, 1H), 11.57 (s, 1H). HRMS (ESI^+): calcd for $[M+H]^+$, 656.2568; found 656.2606.

S16c (4): Yield 25%. ^{1}H-NMR (DMSO-d_6, 300 MHz) δ 2.88-2.93 (m, 4H), 3.00 (m, 4H), 3.43 (s, 4H), 3.47 (m, 4H), 3.61 (m, 5H), 3.71 (s, 6H), 4.05 (s, 2H), 6.08 (s, 1H), 6.62 (s, 2H), 7.34 (d, 1H, J = 8.6 Hz), 7.75 (d, 1H, J = 8.6 Hz), 7.89 (s, 1H), 10.30 (s, 1H), 11.57 (s, 1H). HRMS (ESI^+): calcd for $[M+H]^+$, 716.2779; found 716.2750.

S16d (6): Yield 26%. ^{1}H-NMR (DMSO-d_6, 300 MHz) δ 2.87-2.91 (m, 4H), 2.97 (m, 4H), 3.42 (s, 4H), 3.45 (m, 4H), 3.53 (s, 2H), 4.18 (s, 2H), 6.19 (s, 1H), 7.34 (d, 2H, J = 7.6 Hz), 7.38 (d, 2H, J = 8.0 Hz), 7.43 (t, 2H, J = 7.6 Hz), 7.61 (m, 4H), 7.92 (s, 1H), 10.31 (s, 1H), 11.61 (s, 1H). HRMS (ESI^+): calcd for $[M+H]^+$, 702.2775; found 702.2820.

S16e (7): Yield 33%. ^{1}H-NMR (DMSO-d_6, 300 MHz) δ 2.20 (s, 3H), 2.82-2.86 (m, 4H), 2.90-2.94 (m, 4H), 3.37 (s, 4H), 3.41 (m, 4H), 3.48 (s, 2H), 4.02 (s, 2H), 6.05 (s, 1H), 7.05 (d, 2H, J = 8.2 Hz), 7.11 (d, 2H, J = 8.2 Hz), 7.26 (d,

1H, J = 8.9 Hz), 7.61 (d, 1H, J = 8.9 Hz), 7.86 (s, 1H), 10.24 (s, 1H), 11.53 (s, 1H). HRMS (ESI^+): calcd for $[M+H]^+$, 640.2619; found 640.2651.

S16f (9): Yield 28%. ^{1}H-NMR (DMSO-d_6, 300 MHz) δ 2.81-2.85 (m, 4H), 2.93 (m, 4H), 3.36 (s, 4H), 3.40 (m, 4H), 3.46 (s, 2H), 4.07 (s, 2H), 6.04 (s, 1H), 7.07 (t, 2H, J = 8.8 Hz), 7.27 (m, 3H), 7.62 (d, 1H, J = 8.8 Hz), 7.86 (s, 1H), 10.25 (s, 1H), 11.55 (s, 1H). HRMS (ESI^+): calcd for $[M+H]^+$, 644.2368; found 644.2364.

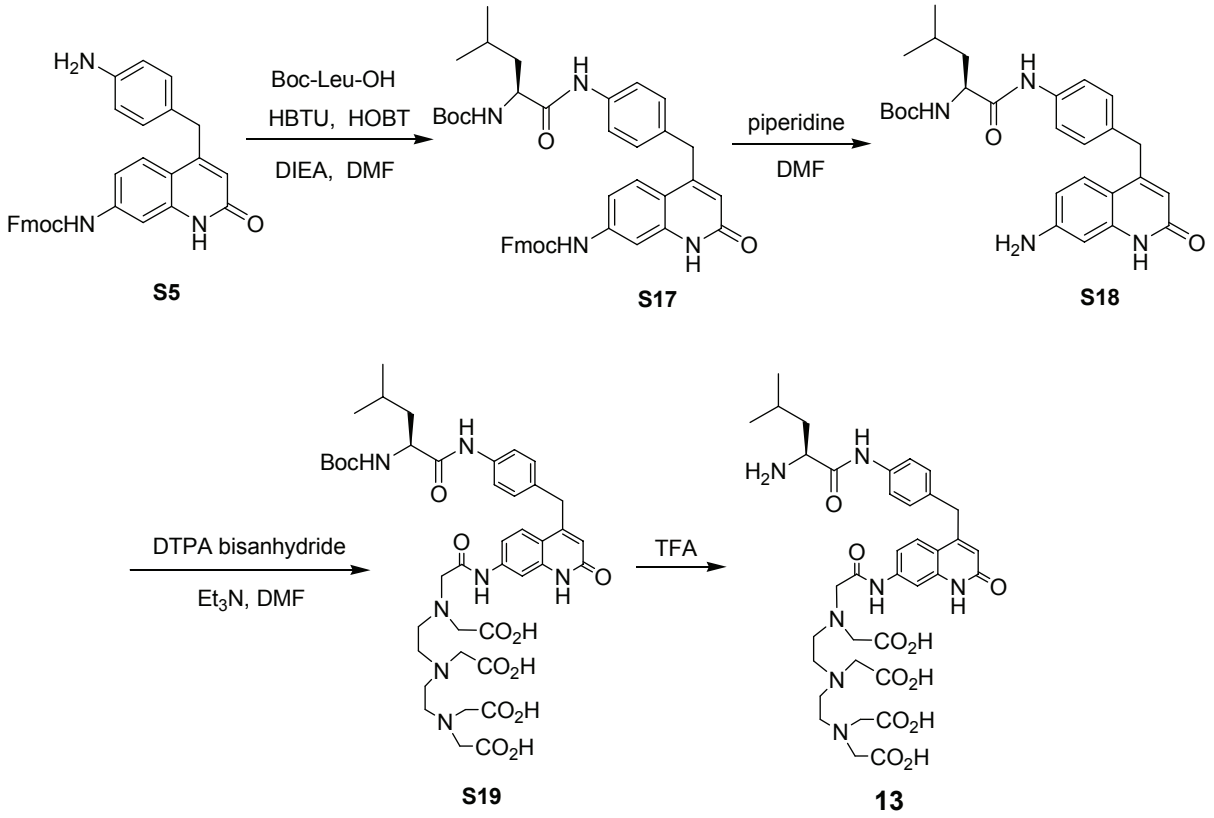
S16g (10): Yield 31%. ^{1}H-NMR (DMSO-d_6, 300 MHz) δ 2.88-2.93 (m, 4H), 3.02 (m, 4H), 3.43 (s, 4H), 3.47 (s, 2H), 3.49 (s, 2H), 3.57 (s, 2H), 4.27 (s, 2H), 6.13 (s, 1H), 7.34 (d, 1H, J = 7.7 Hz), 7.56 (d, 1H, J = 7.7 Hz), 7.61 (m, 2H), 7.71 (m, 2H), 7.92 (s, 1H), 10.31 (s, 1H), 11.64 (s, 1H). HRMS (FAB^+): calcd for $[M+H]^+$, 694.2336; found 694.2334.

S16h (11): Yield 40%. ^{1}H-NMR (DMSO-d_6, 300 MHz) δ2.87 (s, 2H), 2.92 (s, 2H), 3.02-3.10 (m, 4H), 3.40 (s, 4H), 3.44 (s, 2H), 3.46 (s, 2H), 3.69 (s, 2H), 4.23 (s, 2H), 5.77 (s, 1H), 7.37 (d, 1H, J = 8.9 Hz), 7.76 (d, 1H, J = 8.9 Hz), 7.87 (s, 1H), 10.29 (s, 1H), 11.62 (s, 1H). HRMS (FAB^+): calcd for $[M+H]^+$, 716.1991; found 716.2007.

S16i (12): Yield 28%. ^{1}H-NMR (DMSO-d_6, 300 MHz) δ 2.90 (s, 2H), 2.96 (s, 2H), 3.04 (m, 4H), 3.45 (s, 4H), 3.48-3.51 (m, 4H), 3.65 (s, 2H), 3.69 (s, 2H), 4.44 (s, 2H), 5.48 (s, 1H), 7.39 (d, 1H, J = 8.8 Hz), 7.70 (d, 1H, J = 8.8 Hz), 7.85 (s, 1H), 7.95 (m, 2H), 8.07 (d, 1H, J = 8.3 Hz), 10.35 (s, 1H), 11.68 (s, 1H). HRMS (FAB^+): calcd for $[M+H]^+$, 762.2210; found. 762.2231.

Synthesis of 13. Compound **13** was prepared according to the following procedure, as shown in Scheme S5.

Scheme 5. Synthetic scheme for **13**.



Synthesis of tert-butoxycarbonyl-L-leucine 4-[7-(9-fluorenylmethoxycarbonylamino)-1*H*-quinolin-2-on-4-yl]methylanilide (S17). Compound **S5** was synthesized according to the procedure described above. To a solution of Boc-L-Leu-OH (50 mg, 0.20 mmol), HBTU (113 mg, 0.30 mmol), HOBT·H_2O (45 mg, 0.30 mmol), and DIEA (58mg, 0.45 mmol) in DMF (2 ml) was added **S5** (73 mg, 0.15 mmol). The solution was stirred at room temperature for 2 h. The solvent was evaporated, and the residue was dissolved in AcOEt (40 ml). This solution was washed successively with water, 0.5 N NaOH, 0.5 N HCl, and brine (30ml each), dried over Na_2SO_4, and evaporated to dryness. The crude material was roughly purified by silica gel column chromatography (MeOH/AcOEt = 1/20) to give 70 mg (0.20 mmol) of **S17** as a pale yellow solid, 67% yield. ^{1}H-NMR ($CDCl_3$, 300 MHz) δ 0.95-1.00 (m, 6H), 1.45 (s, 9H), 1.75 (m, 3H), 4.08 (s, 2H), 4.25-4.29 (m, 2H), 4.60 (d, 2H, J = 6.0 Hz), 6.34 (s, 1H), 6.91 (d, 1H, J = 4.6 Hz), 7.14 (d, 2H, J = 8.4 Hz), 7.30-7.35 (m, 3H), 7.40-7.48 (m, 4H), 7.54 (d, 1H, J = 8.0 Hz), 7.61 (d,

2H, J = 7.4 Hz), 7.79 (d, 2H, J = 7.4Hz), 8.45 (s, 1H). MS (ESI^+): m/z 701 $[M+H]^+$.

Synthesis of tert-butoxycarbonyl-L-leucine 4-(7-amino-1*H*-quinolin-2-on-4-yl)methylanilide (S18). **S17** (40 mg, 0.057 mmol) was dissolved in DMF (2 ml), and piperidine (0.1 ml) was added. The solution was stirred at room temperature for 1 h, and the solvent was evaporated to dryness. The residue was purified by silica gel column chromatography (MeOH/AcOEt = 1/20) to afford 21 mg (0.043 mmol) of **S18** as a white solid, 76% yield. ^{1}H-NMR (CD_3OD, 300 MHz) δ 0.95-0.98 (m, 6H), 1.43 (s, 9H), 1.55-1.57 (m, 3H), 4.12 (s, 2H), 4.19 (m, 1H), 6.04 (s, 1H), 6.50 (d, 1H, J = 2.4 Hz), 6.59 (dd, 1H, J = 8.8, 2.4 Hz), 7.20 (d, 2H, J = 8.7 Hz), 7.49 (d, 2H, J = 8.7 Hz), 7.53 (d, 1H, J = 8.8 Hz). ^{13}C-NMR (CD_3OD, 75 MHz) δ 22.0, 23.5, 28.7, 42.4, 50.5, 55.2, 80.7, 99.1, 105.5, 113.4, 115.4, 121.8, 127.2, 130.4, 135.8, 138.1, 141.9, 152.8, 154.4, 160.7, 165.8, 174.2. HRMS (ESI^+): calcd for $[M+Na]^+$, 501.2478; found, 501.2462.

Synthesis of N-{N-[4-(tert-butoxycarbonyl-L-leucyl)aminobenzyl-1,2-dihydro-2-oxoquinolin-7-yl]carbamoylmethyl}-N,N',N'',N''-diethylenetriaminetetraacetic acid (S19). The procedure was the same as that described for **5**, using DTPA bisanhydride (29 mg, 0.081 mmol) in DMF (2 ml), triethylamine (30 μl) and **S18** (30 mg, 0.063 mmol) in DMF (2 ml). The residue was purified by preparative reverse-phase HPLC; linear gradient from 45% to 70% solvent B (the solvent system was the same as that used for **5**). Compound **S19** (29 mg, 0.034 mmol) was obtained as a colorless solid, 53% yield. ^{1}H-NMR (CD_3OD, 300 MHz) δ 0.87 (m, 6H), 1.34 (s, 9H), 1.48 (m, 2H), 1.63 (m, 1H), 3.14 (s, 4H), 3.43 (s, 4H), 3.54 (s, 6H), 3.60 (s, 2H), 4.09 (m, 3H), 4.31(s, 2H), 6.19 (s, 1H), 7.13 (d, 2H, J = 8.3 Hz), 7.18 (d, 1H, J = 8.5 Hz), 7.42 (d, 2H, J = 8.3 Hz), 7.66 (d, 1H, J = 8.5 Hz), 7.95 (s, 1H). ^{13}C-NMR (CD_3OD, 75 MHz) δ 22.0, 23.5, 28.7, 42.4, 50.7, 51.2, 54.5, 55.0, 55.2, 56.2, 59.0, 80.6, 107.2, 116.4, 117.5, 119.8, 121.9, 126.8, 130.4, 135.2, 138.2, 140.6, 141.7, 153.8, 165.3, 169.5, 172.0, 174.0, 174.6, 174.8. HRMS (ESI^-): calcd for $[M-H]^-$, 852.3780; found, 852.3730.

Synthesis of N-(N-{4-[4-(L-leucylamino)benzyl]-1,2-dihydro-2-oxoquinolin-7-yl}carbamoylmethyl)-N,N',N'',N''-diethylenetriaminetetraacetic acid (13). **S19** (15 mg, 0.018 mmol) was dissolved in TFA (1 ml), and stirred at room temperature for 1 h. The solvent was evaporated, and the residue was dissolved in water and lyophilized to give 12 mg (0.016 mmol) of **13** as a white powder, 84% yield. ^{1}H-NMR (CD_3OD, 300 MHz) δ 0.86 (d, 6H, J = 5.1 Hz), 1.62 (m, 3H), 3.08 (s, 4H), 3.36 (m, 4H), 3.46 (s, 6H), 3.53 (s, 2H), 3.81 (m, 1H), 4.06 (s, 2H), 4.22 (s, 2H), 6.04 (s, 1H), 7.09 (d, 2H, J = 8.3 Hz), 7.16 (d, 1H, J = 8.5 Hz), 7.41 (d, 2H, J = 8.3 Hz), 7.64 (d, 1H, J = 8.5 Hz), 7.88 (s, 1H). ^{13}C-NMR (CD_3OD, 75 MHz) δ 22.0, 23.2, 28.7, 41.8, 50.8, 51.2, 54.6, 54.9, 55.2, 56.0, 56.2, 59.1, 107.3, 116.5, 117.5, 119.7, 121.7, 126.6, 130.7, 135.7, 137.8, 140.6, 141.8, 153.8, 165.3, 169.7, 172.0, 174.6, 174.8. HRMS (ESI^-): calcd for $[M-H]^-$, 752.3255; found, 752.3229.

Synthesis of [Ln-13]. **[Ln-13]** was prepared by a similar method that described for **[Ln-1]**. **[Eu-13]**: HRMS (ESI^-): calcd for $[M-H]^-$, 902.2237; found, 902.2221. **[Tb-13]**: HRMS (ESI^-): calcd for $[M-H]^-$, 908.2274; found, 908.2305.

Table S1. Absorbance, luminescence and phosphorescence properties of Tb^{3+} complexes.

	λ_{Ex} /nm	ε	Φ_{Lum} (%)[a]	τ (H_2O) /ms	τ (D_2O) /ms	q[b]	HOMO level /eV	T_1[c] /cm^{-1}
[Tb-1]	330	13000	0.01	n.d.[d]	n.d.[d]	n.d.[d]	−4.98	22600
[Tb-2]	329	13000	0.38	n.d.[d]	n.d.[d]	n.d.[d]	−5.27	22700
[Tb-3]	330	13000	1.3	1.10	1.72	1.34	−5.72	22600
[Tb-4]	330	13000	3.5	1.18	1.83	1.21	−5.70	22600
[Tb-5]	330	13000	4.8	1.15	1.80	1.27	−5.82	22600
[Tb-6]	330	13000	7.8	1.15	1.83	1.32	−5.90	22600
[Tb-7]	330	13000	9.1	1.22	1.91	1.18	−6.16	22600
[Tb-8]	330	13000	9.6	1.22	1.90	1.17	−6.44	22600
[Tb-9]	330	13000	8.8	1.21	1.95	1.27	−6.54	22600
[Tb-10]	330	13000	9.3	1.15	1.76	1.21	−7.16	22500
[Tb-11]	330	13000	9.3	1.08	1.57	1.14	−7.63	22700
[Tb-12]	330	13000	9.6	1.21	1.83	1.10	−7.77	22700

[a] The quantum yield was determined using quinine sulfate ($\Phi = 0.546$)[S1] as a standard. [b] q values were estimated using the equation $q = 5(\tau_{H_2O}^{-1} - \tau_{D_2O}^{-1} - 0.06)$.[S2] [c] Triplet energy level was evaluated from the wavelength of phosphorescence of Gd^{3+} complex at 77 K measured in EtOH-MeOH glass. [d] Not determined, because the luminescence intensity was too low.

Table S2. Absorbance, luminescence and phosphorescence properties of Eu^{3+} complexes.

	λ_{Ex} /nm	ε	Φ_{Lum} (%)[a]	τ (H_2O) /ms	τ (D_2O) /ms	q[b]	HOMO level /eV	T_1[c] /cm^{-1}
[Eu-1]	330	13000	0.01	n.d.[d]	n.d.[d]	n.d.[d]	−4.98	22600
[Eu-2]	329	13000	1.26	0.58	2.37	1.37	−5.27	22700
[Eu-3]	330	13000	3.29	0.61	2.28	1.26	−5.72	22600
[Eu-4]	330	13000	5.06	0.58	2.31	1.36	−5.70	22600
[Eu-5]	329	13000	5.31	0.59	2.22	1.31	−5.82	22600
[Eu-6]	330	13000	5.56	0.57	2.33	1.39	−5.90	22600
[Eu-7]	330	13000	5.06	0.59	2.37	1.34	−6.16	22600
[Eu-8]	330	13000	4.81	0.59	2.23	1.31	−6.44	22600
[Eu-9]	330	13000	5.56	0.58	2.45	1.38	−6.54	22600
[Eu-10]	330	13000	5.31	0.60	2.37	1.31	−7.16	22500
[Eu-11]	330	13000	5.82	0.62	2.35	1.25	−7.63	22700
[Eu-12]	329	13000	5.31	0.60	2.19	1.27	−7.77	22700

[a] The quantum yield was determined using $[Ru(bipy)_3]Cl_2$ ($\Phi = 0.028$)[S3] as a standard. [b] q values were estimated using the equation $q = 1.2(\tau_{H_2O}^{-1} - \tau_{D_2O}^{-1} - 0.25 - 0.075)$.[S2] [c] Triplet energy level was evaluated from the wavelength of phosphorescence of Gd^{3+} complex at 77 K measured in EtOH-MeOH glass. [d] Not determined, because the luminescence intensity was too low.

Table S3. Comparison of kinetic parameters among probes.

	L-pNA[a]	L-MCA[b]	**[Tb-13]**[c]
K_m (M)	1.13×10^{-4}	8.03×10^{-5}	4.18×10^{-5}
k_{cat} (s^{-1})	81.0	49.2	29.3
k_{cat}/K_m ($M^{-1}s^{-1}$)	7.17×10^5	6.13×10^5	7.01×10^5

Reactions were performed with LAP 0.001 U/ml in 100 mM Tris-HCl buffer at 37 °C. Initial velocity was calculated from the change of absorbance or fluorescence intensity and plotted as a function of the probe concentration. Data were best fitted to a Michaelis-Menten plot. [a] Absorbance at 405 nm was recorded. [b] Emission intensity at 440 nm (ex. 380 nm) was recorded. [c] Emission intensity at 545 nm (ex. 330 nm) was recorded.

Table S4. Inhibition constants of known LAP inhibitors determined with various probes.

	L-pNA	L-MCA	**[Tb-13]**
actinonin	0.11 μM	0.13 μM	0.10 μM
amastatin	0.11 μM	0.12 μM	0.10 μM
bestatin	2.1 μM	1.1 μM	2.9 μM

Assays were performed in 100 mM Tris-HCl buffer (pH 7.4) with LAP (0.01 U) at 37 °C. Substrate concentration was 2 μM (for L-MCA and **[Tb-13]**) or 20 μM (for L-pNA). Initial velocity of the reaction was calculated from the change of fluorescence intensity or absorbance during the first 3 min. Inhibitory activity (%) was determined by comparing the velocity with and without inhibitor. K_i was calculated from the following equation, where v = reaction velocity in the presence of inhibitor, v_0 = reaction velocity in the absence of inhibitor, $[I]$ = inhibitor concentration.

$$\frac{v}{v_0} = \frac{1}{1+[I]/K_i}$$

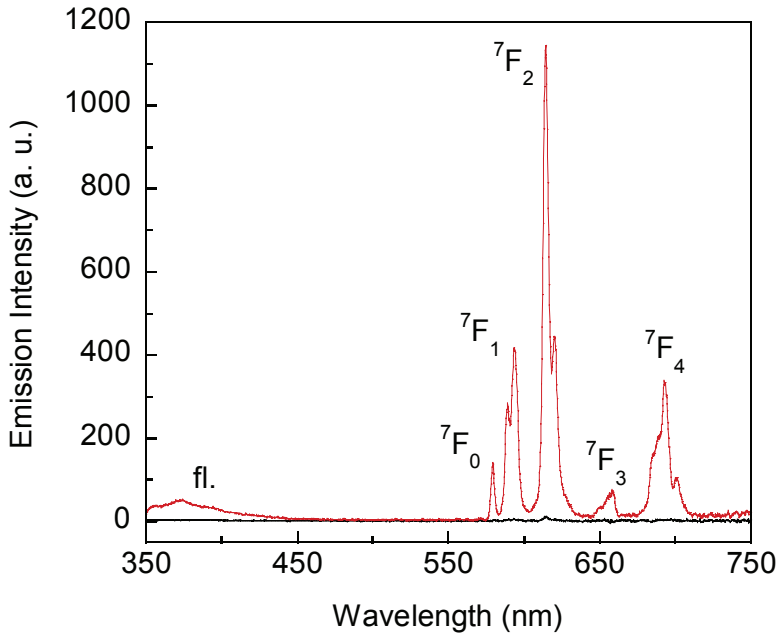


Figure S1. Emission spectra of **[Eu-8]** (red line) and **[Eu-1]** (black line) 5 μM in aqueous solution (100 mM HEPES buffer, pH 7.3, I = 0.1($NaNO_3$)). Excitation wavelength was 330 nm. The attribution of each band is shown in the figure (see main text).

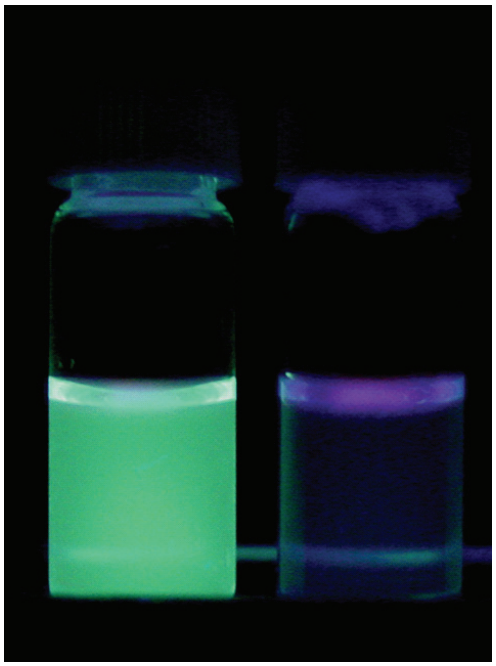


Figure S2. Comparison of luminescence intensity of **[Tb-13]** (5 μM) before (left) and 120 min after (right) the enzymatic reaction. Samples were illuminated with a UV lamp (365 nm).

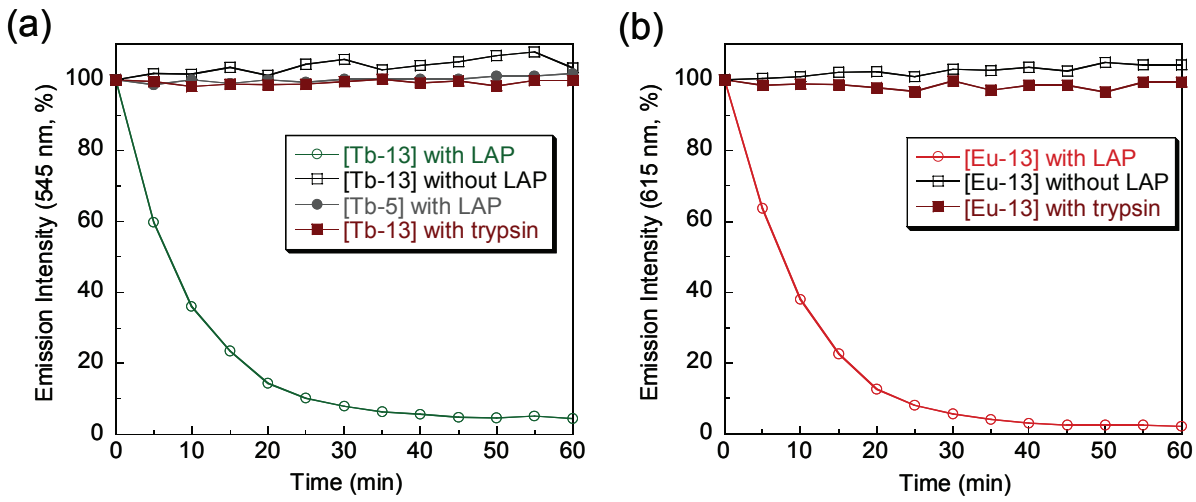


Figure S3. Emission intensity of **[Ln-X]** (2 μM) as a function of time after the addition of LAP (0.01 U) or trypsin (10 BAEE U). (a) Tb^{3+} complexes. (b) Eu^{3+} complexes.

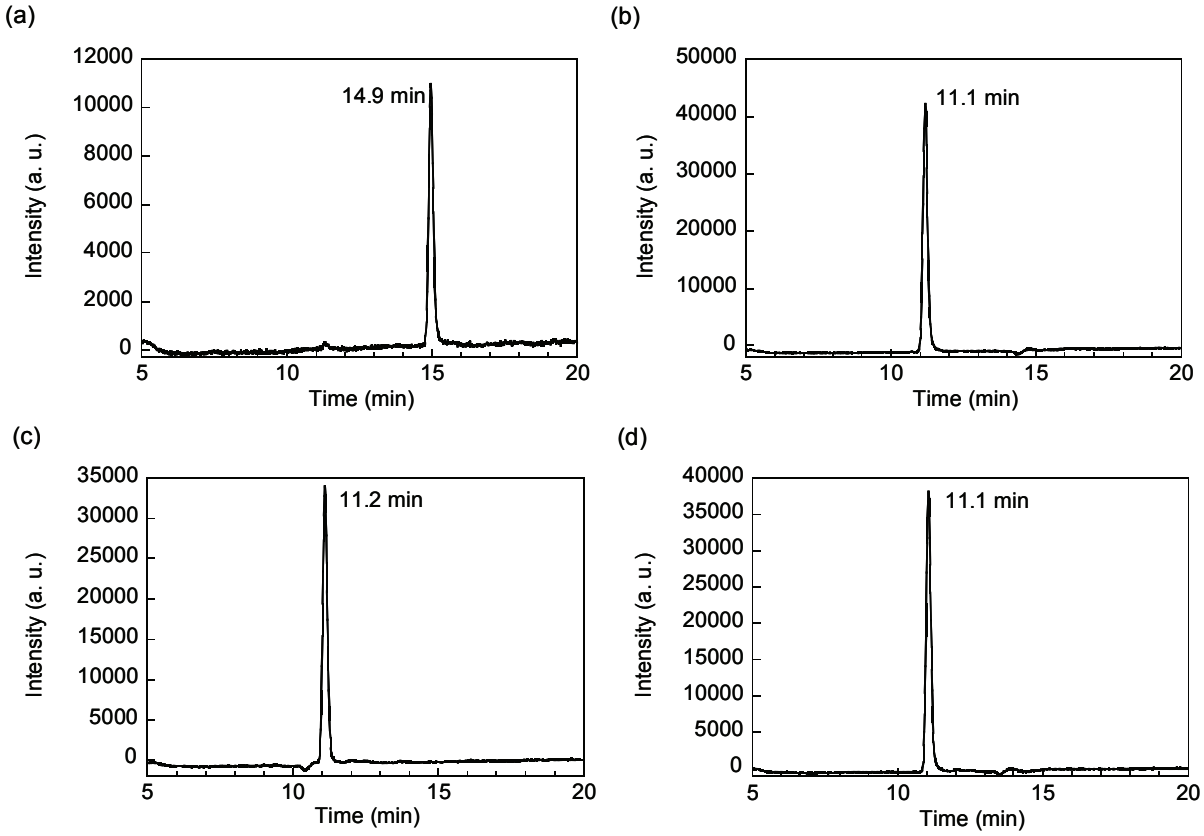


Figure S4. HPLC confirmation of the enzymatic conversion of **[Tb-13]** to **[Tb-1]**: (a) a solution of **[Tb-13]**, (b) a solution of **[Tb-1]**, (c) reaction mixture (90 min after the addition of LAP), (d) mixed solution of **[Tb-1]** and reaction mixture. For all spectra, absorbance at 330 nm was recorded.

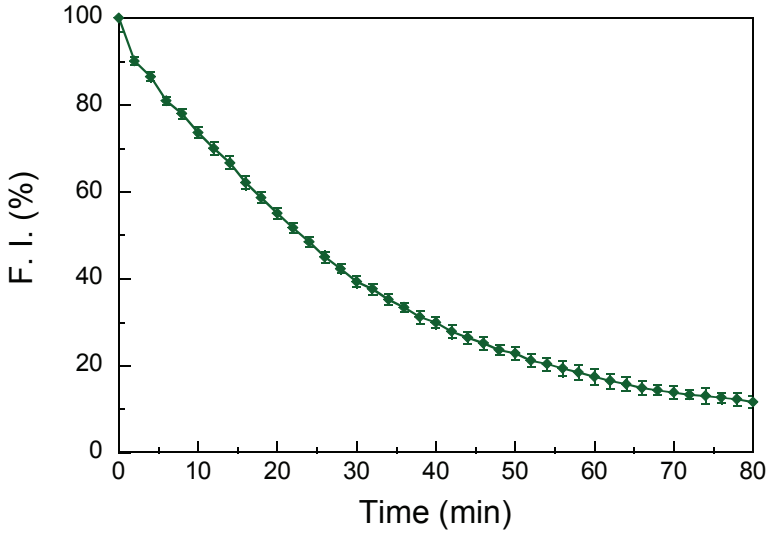


Figure S5. LAP reaction on 96-well microplate. 0.001 U of LAP was added to 2.5 μM **[Tb-13]** in 100 mM Tris-HCl buffer, pH 7.4. Total volume was 200 μl. Luminescence intensity at 545 nm (ex. 330 nm) was recorded every 2 min with a delay time of 50 μs and a gate time of 2.0 ms. Data are shown as mean ± SEM (n = 5).

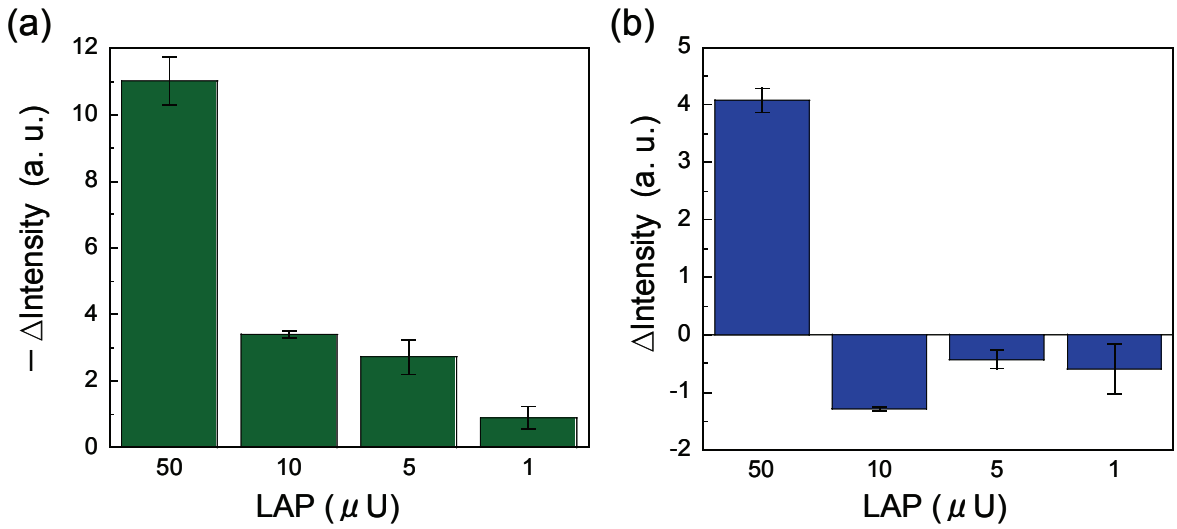


Figure S6. Limit of detection of LAP. Assays were performed on 96-well plates for 60 min with 2.5 μM (a) **[Tb-13]** or (b) L-MCA. TRF measurements were performed for (a). Change of fluorescence intensity (ex./em. = 325/545 nm for (a) and 380/440 nm for (b)) over 90 min was plotted against the amount of added LAP. Data were corrected so that change of intensity without LAP was zero, and are shown as mean ± SEM (n = 3).

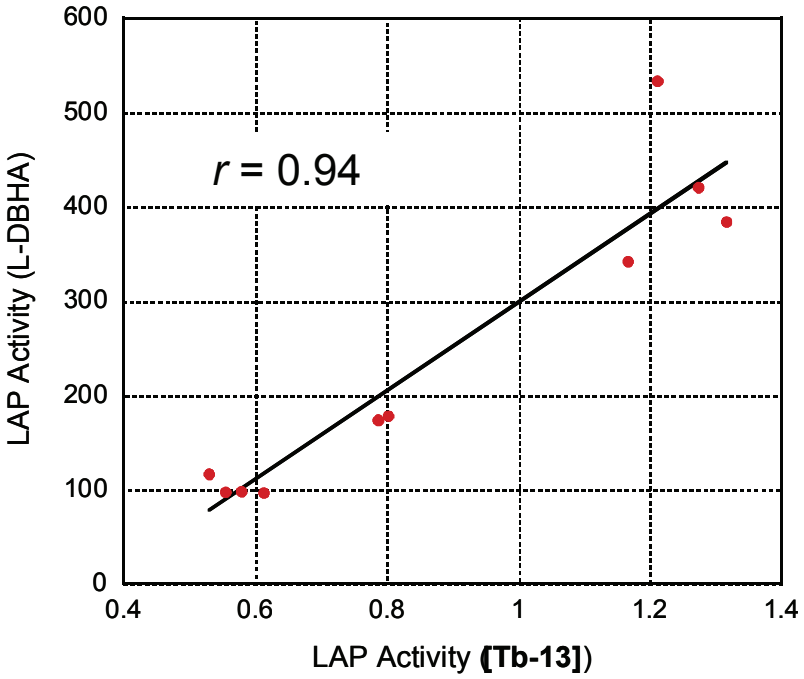


Figure S7. Correlation between serum LAP activities measured by two methods. On the horizontal axis, values determined with **[Tb-13]** (mU/ml serum) are plotted. On the vertical axis, values determined using the commercially performed L-DBHA method (IU/L) are plotted. The correlation coefficient (*r*) is shown in the figure.

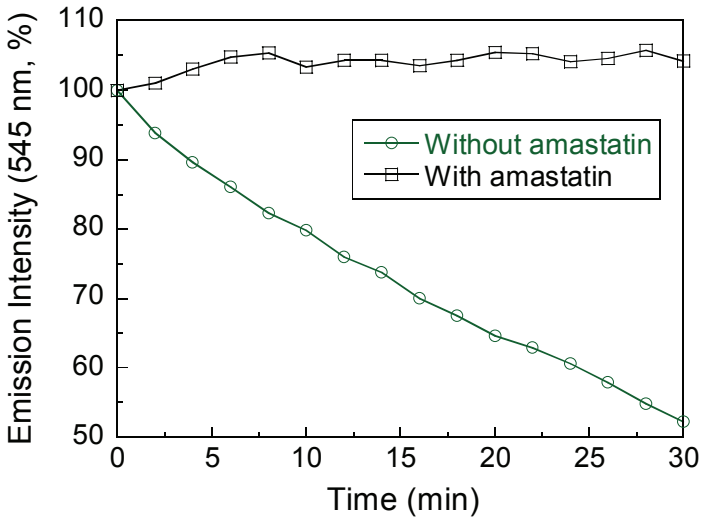


Figure S8. Emission intensity of **[Ln-13]** (1 μM) in human serum as a function of time with (black open square) or without (green open circle) amastatin (10 μM). Experiments were performed in 10% (v/v) human serum/Tris-HCl buffer (0.1 M, pH 7.4) at 37 °C with TRF measurement (delay time = 50 μsec, gate time = 2.0 msec). Excitation/emission wavelengths were 330/545 nm.

List of authors of reference 19(b) in the main text.

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