Electronic Supplementary Material (ESI) for Dalton Transactions.

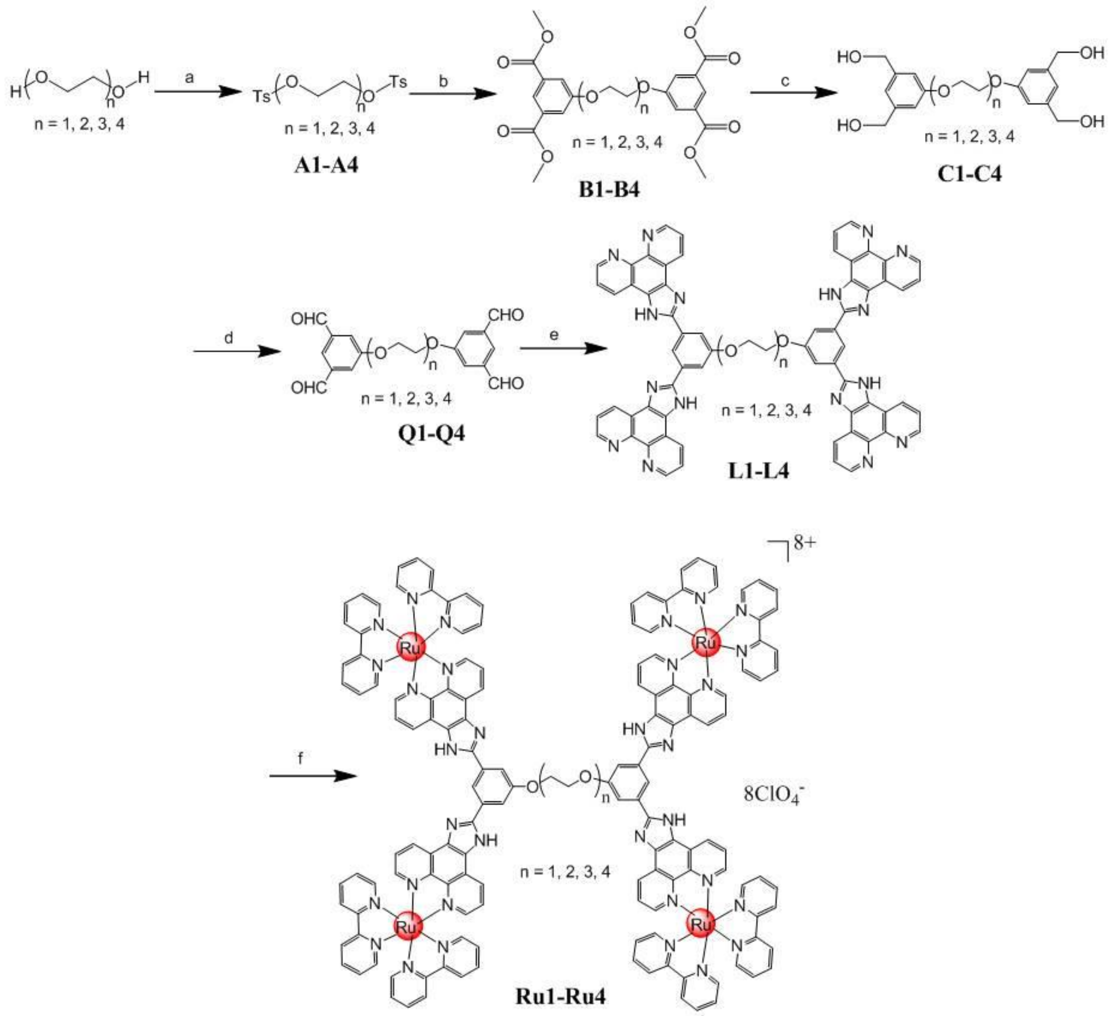
Supporting Information

Tetranuclear ruthenium(II) complexes with oligo-oxyethylene linkers as one- and two-photon luminescent tracking non-viral gene vectors

Kangqiang Qiu, Bole Yu, Huaiyi Huang, Pingyu Zhang,
Liangnian Ji and Hui Chao*

MOE Key Laboratory of Bioinorganic and Synthetic Chemistry, School of Chemistry and
Chemical Engineering, Sun Yat-Sen University, Guangzhou 510275, P. R. China
Tel: +86 20 84110613; Fax: +86 20 84112245; Email: ceschh@mail.sysu.edu.cn

Table of Contents



Scheme. S1. Synthetic routes of **Ru1-Ru4**. (a) *p*-TsCl, yield: 72-73 %; (b) 5-hydroxy-isophthalic acid dimethyl ester, yield: 58-61 %; (c) LAH, yield: 75-76 %; (d) PCC, yield: 49-59 %; (e) 1,10-phenanthroline-5,6-dione, yield: 49-62 %; (f) $[Ru(bpy)_2]Cl_2$, yield: 73-85 %.

Synthesis

Materials

All reagents were purchased from commercial sources and used without further purification unless otherwise specified. All solvents were distilled and/or dried prior their use whenever necessary. The compounds 1,8-Bis-[phenoxy-(3, 5-dicarbaldehyde)]-3,6-dioxo octane (**Q3**), 1,11-Bis-[phenoxy-(3, 5-dicarbaldehyde)]-3,6,9-trioxo undecane (**Q4**),[1] 1,10-phenanthroline-5,6-dione[2] and $[Ru(bpy)_2]Cl_2.2H_2O$[3] were prepared according to literature methods.

Synthesis of dicarbaldehyde compound (Q1-Q2)

Ethylene gloycol ditosylate (A1). Ethylene glycol (0.62 g, 10 mmol) in THF (10 mL) and NaOH (1.0 g, 25 mmol) dissolved in 2.5 mL of water were mixed together under ice-cold conditions. The resulting solution was stirred for 30 min and then to this *p*-tosyl chloride (4.7 g, 25 mmol), dissolved in 5 mL of THF was added drop-wise. The mixture was then allowed to come at room temperature and stirred for further 3 h. Solvent from the reaction mixture was evaporated and water (20 mL) was added. To this ethyl acetate was added and the organic matrrial were extracted in the organic layer which upon rotary evaporation afforded 3.2 g of the crude product. The crude product was then purified using column chromatography (EtOAc/n-hexane) on silica gel (60-100 mesh size) to obtain 2.7 g white solid (yield: 73 %). ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.71 (d, J = 9 Hz, 4H), 7.34 (d, J = 9 Hz, 4H), 4.18 (s, 4H), 2.50 (s, 6H). ESI-MS: m/z = 392.8 $[M+Na]^+$, 424.8 $[M+Na+CH_3OH]^+$.

Diethylene gloycol ditosylate (A2). Diethylene glycol (1.06 g, 10 mmol) in THF (10 mL) and NaOH (1.0 g, 25 mmol) dissolved in 2.5 mL of water were mixed together under ice-cold conditions. The resulting solution was stirred for 30 min and then to this *p*-tosyl chloride (4.7 g, 25 mmol), dissolved in 5 mL of THF was added drop-wise.

The mixture was then allowed to come at room temperature and stirred for further 3 h. Solvent from the reaction mixture was evaporated and water (20 mL) was added. To this ethyl acetate was added and the organic matrrial were extracted in the organic layer which upon rotary evaporation afforded 3.7 g of the crude product. The crude product was then purified using column chromatography (EtOAc/n-hexane) on silica gel (60-100 mesh size) to obtain 3.0 g of white solid (yield: 72 %). ^{1}H NMR (300 MHz, $CDCl_3$) δ 7.75 (d, *J* = 9 Hz, 4H), 7.32 (d, *J* = 9 Hz, 4H), 4.07 (t, *J* = 6 Hz, 4H), 3.59 (t, *J* = 3 Hz, 4H), 2.45 (s, 6H). ESI-MS: m/z = 437.2 $[M+Na]^+$.

1,2-Bis-[phenoxy-(3,5-dicarboxylic acid dimethyl ester)] ethane (B1). A mixture of the **A1** (1.85 g, 5 mmol), 5-hydroxy-isophthalic acid dimethyl ester (2.3 g, 11 mmol) and K_2CO_3 (1.5 g) were taken together in around bottom flask and refluxed in CH_3CN (20 mL) with stirring for 10 h under Ar atmosphere. Reaction mixture was then cooled, filtered and evaporated. To the residue, water (50 mL) was added and extracted with ethyl acetate using a separatory funnel. Upon solvent evaporation from the EtOAc layer, 2.1 g of a crude product was obtained. The crude product was then purified using column chromatography (EtOAc/n-hexane) on silica gel (60-100 mesh size) to obtain 1.3 g of white solid (yield: 58 %). ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.31 (s, 2H), 7.81 (s, 4H), 4.45 (s, 4H), 3.95 (s, 12H). ESI-MS: m/z = 468.9 $[M+Na]^+$.

1,5-Bis-[phenoxy-(3,5-dicarboxylic acid dimethyl ester)]-3-oxo pentane (B2). A mixture of the **A2** (2.07 g, 5 mmol), 5-hydroxy-isophthalic acid dimethyl ester (2.3 g, 11 mmol) and K_2CO_3 (1.5 g) were taken together in around bottom flask and refluxed in CH_3CN (20 mL) with stirring for 10 h under Ar atmosphere. Reaction mixture was then cooled, filtered and evaporated. To the residue, water (50 mL) was added and extracted with ethyl acetate using a separatory funnel. Upon solvent evaporation from the EtOAc layer, 2.3 g of a crude product was obtained. The crude product was then purified using column chromatography (EtOAc/n-hexane) on silica gel (60-100 mesh size) to obtain 1.5 g of white solid (yield: 61 %). ^{1}H NMR (300 MHz, $CDCl_3$) δ 8.26 (s, 2H), 7.76 (s, 4H), 4.24 (t, *J* = 3 Hz, 4H), 3.96 (t, *J* = 3 Hz, 4H), 3.93 (s, 12H). ESI-MS: m/z = 529.2 $[M+K]^+$.

1,2-Bis-[phenoxy-(3,5-dihydroxymethyl)] ethane (C1). B1 (669 mg, 1.5 mmol) was dissolved in anhydrous THF (10 mL) and lithium aluminum hydride (171 mg, 9 mmol) was added to this under ice-cold conditions in Ar atmosphere. Reaction mixture was then allowed to gradually come to room temperature and stirred for 12 h, when TLC indicated complete disappearance of the starting materials. Dilute HCl solution was carefully added to decompose any unreacted excess of lithium aluminum hydride, stirred for half an hour and then the resulting solution was extacted with ethyl acetate to furnish 380 mg of a off-white gum (yield: 76 %). ^{1}H NMR (300 MHz, $CDCl_3$) δ 6.84 (s, 2H), 6.76 (s, 4H), 5.15 (bs, 4H), 4.44 (s, 8H), 4.26 (s, 4H). ESI-MS: m/z=357.2 $[M+Na]^+$.

1,5-Bis-[phenoxy-(3,5-dihydroxymethyl)]-3-oxo pentane (C2). B2 (735 mg, 1.5 mmol) was dissolved in anhydrous THF (10 mL) and lithium aluminum hydride (171 mg, 9 mmol) was added to this under ice-cold conditions in Ar atmosphere. Reaction mixture was then allowed to gradually come to room temperature and stirred for 12 h, when TLC indicated complete disappearance of the starting materials. Dilute HCl solution was carefully added to decompose any unreacted excess of lithium aluminum hydride, stirred for half an hour and then the resulting solution was extacted with ethyl acetate to furnish 425 mg of a off-white gum (yield: 75 %). ^{1}H NMR (300 MHz, $CDCl_3$) δ 6.82 (s, 2H), 6.72 (s, 4H), 5.12 (s, 4H), 4.43 (s, 8H), 4.06 (t, J = 3 Hz, 4H), 3.78 (t, J = 3 Hz, 4H). ESI-MS: m/z=401.3 $[M+Na]^+$.

1,2-Bis-[phenoxy-(3,5-dicarbaldehyde)] ethane (Q1). C1 (167 mg, 0.5 mmol) was dissolved in a mixture of dichloromethane and THF (6:4, 20 mL) and 6 equiv. (673.5 mg) of pyridinium chlorochromate was added to this solution. To this solution, 1.5 g of silica was also added. Resulting slurry was stirred at room temperature for 5 h and then directly loaded onto a silica gel column and the required product was eluted with dichloromethane to furnish 80 mg of white solid (yield: 49 %). ^{1}H NMR (300 MHz, $CDCl_3$) δ 10.06 (s, 4H), 8.00 (s, 2H), 7.72 (s,4H), 4.52 (s, 4H). ESI-MS: m/z = 348.9 $[M+Na]^+$, 380.9 $[M+Na+CH_3OH]^+$.

1,5-Bis-[phenoxy-(3,5-dicarbaldehyde)]-3-oxo pentane (Q2). C2 (189 mg, 0.5

mmol) was dissolved in a mixture of dichloromethane and THF (6:4, 20 mL) and 6 equiv. (673.5 mg) of pyridinium chlorochromate was added to this solution. To this solution, 1.5 g of silica was also added. Resulting slurry was stirred at room temperature for 5 h and then directly loaded onto a silica gel column and the required product was eluted with dichloromethane to furnish 109 mg of white solid (yield: 59 %). ^{1}H NMR (300 MHz, $CDCl_3$) δ 10.03 (s, 4H), 7.95 (s, 2H), 7.67 (s,4H), 4.28 (t, *J* = 4.5 Hz, 4H), 3.98 (t, *J* = 4.5 Hz, 4H). ESI-MS: m/z = 392.9 $[M+Na]^+$, 424.9 $[M+Na+CH_3OH]^+$.

Synthesis of ligands (L1-L4)

1,2-bis-(3,5-bis(1H-imidazo[4,5-f][1, 10]phenanthrolin-2-yl)phenoxy) ethane (L1). A mixture of 1,10-phenanthroline-5,6-dione (0.21 g, 1.0 mmol), ammonium acetate (1.55 g, 20 mmol), **Q1** (65.2 mg, 0.20 mmol) and glacial acetic acid (10 mL) was refluxed for about 6 h, giving a suspension. The reaction mixture was filtered hot, and the yellow solid was washed successively with H_2O and Et_2O, then dried in vacuo (115.2 mg, yield: 53 %). The ^{1}H NMR spectrum of **L1** was not obtained due to its poor solubility in common NMR solvents. Anal. Calcd. for $C_{66}H_{38}N_{16}O_2$: C, 72.92; H, 3.52; N, 20.61. Found: C, 72.43; H, 3.57; N, 20.48. FAB-MS: m/z = 1088 [M+1].

1,5-bis-(3,5-bis(1H-imidazo[4,5-f][1,10]phenanthrolin-2-yl)phenoxy)-3-oxo pentane (L2). A mixture of 1,10-phenanthroline-5,6-dione (0.21 g, 1.0 mmol), ammonium acetate (1.55 g, 20 mmol), **Q2** (74.0 mg, 0.20 mmol) and glacial acetic acid (10 mL) was refluxed for about 6 h, giving a suspension. The reaction mixture was filtered hot, and the yellow solid was washed successively with H_2O and Et_2O, then dried in vacuo (140.4 mg, yield: 62 %). The ^{1}H NMR spectrum of **L2** was not obtained due to its poor solubility in common NMR solvents. Anal. Calcd. for $C_{68}H_{42}N_{16}O_3$: C, 72.20; H, 3.74; N, 19.81. Found: C, 71.62; H, 3.91; N, 19.54. FAB-MS: m/z = 1132 [M+1].

1,8-bis-(3,5-bis(1H-imidazo[4,5-f][1,10]phenanthrolin-2-yl)phenoxy)-3,6-dioxo octane (L3). A mixture of 1,10-phenanthroline-5,6-dione (0.21 g, 1.0 mmol), ammonium acetate (1.55 g, 20 mmol), **Q3** (82.8 mg, 0.20 mmol) and glacial acetic acid (10 mL) was refluxed for about 6 h, giving a suspension. The reaction mixture was filtered hot, and the yellow solid was washed successively with H_2O and Et_2O, then dried in vacuo (136.4 mg, yield: 58 %). The ^1H NMR spectrum of **L3** was not obtained due to its poor solubility in common NMR solvents. Anal. Calcd. for $C_{70}H_{46}N_{16}O_4$: C, 71.54; H, 3.95; N, 19.07. Found: C, 70.88; H, 4.01; N, 18.89. FAB-MS: m/z = 1176 [M+1].

1,11-bis-(3,5-bis(1H-imidazo[4,5-f][1,10]phenanthrolin-2-yl)phenoxy)-3,6,9-trioxoundecane (L4). A mixture of 1,10-phenanthroline-5,6-dione (0.21 g, 1.0 mmol), ammonium acetate (1.55 g, 20 mmol), **Q4** (91.6 mg, 0.20 mmol) and glacial acetic acid (10 mL) was refluxed for about 6 h, giving a suspension. The reaction mixture was filtered hot, and the yellow solid was washed successively with H_2O and Et_2O, then dried in vacuo (119.6 mg, yield: 49 %). The ^1H NMR spectrum of **L4** was not obtained due to its poor solubility in common NMR solvents. Anal. Calcd. for $C_{72}H_{50}N_{16}O_5$: C, 70.93; H, 4.13; N, 18.38. Found: C, 70.57; H, 4.24; N, 18.19. FAB-MS: m/z = 1220 [M+1].

Synthesis of complexes (Ru1-Ru4)

$\{[Ru(bpy)_2]_4L1\}(ClO_4)_8$ (Ru1). The yellow solid **L1** (54.3 mg, 0.05 mmol) was mixed with $[Ru(bpy)_2]Cl_2.2H_2O$ (130 mg, 0.25 mmol) in ethylene glycol (10 mL), the mixture was heated to 170 °C for 8 h under Ar. The cooled reaction mixture was diluted with water (10 mL). Saturated aqueous sodium perchlorate solution was added under vigorous stirring, and filtered. The dark red solid was collected and washed with small amounts of H_2O and Et_2O, then dried under vacuum, and purified by column chromatography on alumina using acetonitrile-ethanol as the eluant. The solvent was removed under reduced pressure and red microcrystals were obtained (129 mg, yield = 73%). Anal. Calcd. for $C_{146}H_{102}Cl_8N_{32}O_{34}Ru_4$: C, 49.59; H, 2.91; N, 12.67. Found: C, 49.28; H, 3.01; N, 12.53. ^1H NMR (300 MHz, d_6-DMSO) δ 9.16 (bs, 8H), 8.87 (d, J = 10 Hz, 8H), 8.83 (d, J = 10 Hz, 8H), 8.20 (t, J = 10 Hz, 12H), 8.09 (t, J = 10 Hz, 10H), 7.96 (bs, 8H), 7.87 (d, J = 5 Hz, 16H), 7.58-7.63 (m, 16H), 7.35 (t, J = 10 Hz, 8H), 4.80 (bs, 4H). ESI-MS: m/z = 784.9 $[M-4ClO_4]^{4+}$, 760 $[M-5ClO_4]^{4+}$, 734.6 [M-6ClO_4]$^{4+}$, 709.5 [M-7ClO_4]$^{4+}$, 683.8 [M-8ClO_4]$^{4+}$, 567.7 [M-7ClO_4]$^{5+}$, 547.4 [M-8ClO_4]$^{5+}$, 472.5 [M-7ClO_4]$^{6+}$, 456.1 [M-8ClO_4]$^{6+}$.

{[Ru(bpy)$_2$]$_4$L2}(ClO_4)$_8$ (Ru2). The yellow solid **L2** (56.6 mg, 0.05 mmol) was mixed with [Ru(bpy)$_2$]Cl_2.2H_2O (130 mg, 0.25 mmol) in ethylene glycol (10 mL), the mixture was heated to 170 °C for 8 h under Ar. The cooled reaction mixture was diluted with water (10 mL). Saturated aqueous sodium perchlorate solution was added under vigorous stirring, and filtered. The dark red solid was collected and washed with small amounts of H_2O and Et_2O, then dried under vacuum, and purified by column chromatography on alumina using acetonitrile-ethanol as the eluant. The solvent was removed under reduced pressure and red microcrystals were obtained (152 mg, yield = 85%). Anal. Calcd. for $C_{148}H_{106}Cl_8N_{32}O_{35}Ru_4$: C, 49.65; H, 2.98; N, 12.52. Found: C, 49.40; H, 3.05; N, 12.37. ^{1}H NMR (500 MHz, d$_6$-DMSO) δ 9.40 (bs, 8H), 8.89 (d, *J* = 5 Hz, 8H), 8.85 (d, *J* = 10 Hz, 8H), 8.31 (s, 4H), 8.21 (t, *J* = 7.5 Hz, 8H), 8.10 (t, *J* = 7.5 Hz, 10H), 8.04 (s, 8H), 7.85 (d, *J* = 5 Hz, 16H), 7.65 (s, 8H), 7.58 (t, *J* = 7.5 Hz, 8H), 7.37 (s, 8H), 4.60 (bs, 4H), 4.12 (s, 4H). ESI-MS: m/z = 769.4 [M-5ClO_4]$^{4+}$, 744.6 [M-6ClO_4]$^{4+}$, 719.9 [M-7ClO_4]$^{4+}$, 694.6 [M-8ClO_4]$^{4+}$, 635.0 [M-4ClO_4]$^{5+}$, 596.0 [M-6ClO_4]$^{5+}$, 576.0 [M-7ClO_4]$^{5+}$, 556.1 [M-8ClO_4]$^{5+}$.

{[Ru(bpy)$_2$]$_4$L3}(ClO_4)$_8$ (Ru3). The yellow solid **L3** (58.8 mg, 0.05 mmol) was mixed with [Ru(bpy)$_2$]Cl_2.2H_2O (130 mg, 0.25 mmol) in ethylene glycol (10 mL), the mixture was heated to 170 °C for 8 h under Ar. The cooled reaction mixture was diluted with water (10 mL). Saturated aqueous sodium perchlorate solution was added under vigorous stirring, and filtered. The dark red solid was collected and washed with small amounts of H_2O and Et_2O, then dried under vacuum, and purified by column chromatography on alumina using acetonitrile-ethanol as the eluant. The solvent was removed under reduced pressure and red microcrystals were obtained (136 mg, yield = 75%). Anal. Calcd. for $C_{150}H_{110}Cl_8N_{32}O_{36}Ru_4$: C, 49.71; H, 3.06; N, 12.37. Found: C, 49.25; H, 3.10; N, 12.22. ^{1}H NMR (500 MHz, d$_6$-DMSO) δ 9.03 (s, 8H), 8.89 (d, *J* = 5 Hz, 8H), 8.85 (d, *J* = 10 Hz, 8H), 8.21 (s, 12H), 8.11 (s, 10H), 7.87 (bs, 16H), 7.74 (bs, 8H), 7.65 (s, 8H), 7.59 (s, 8H), 7.38 (s, 8H), 4.43 (bs, 4H), 3.96 (s, 4H), 3.80 (s, 4H). ESI-MS: m/z = 781.6 $[M\text{-}5ClO_4]^{4+}$, 756.6 $[M\text{-}6ClO_4]^{4+}$, 731.8 $[M\text{-}7ClO_4]^{4+}$, 707.0 $[M\text{-}8ClO_4]^{4+}$, 625.7 $[M\text{-}5ClO_4]^{5+}$, 605.9 $[M\text{-}6ClO_4]^{5+}$, 585.8 $[M\text{-}7ClO_4]^{5+}$, 564.8 $[M\text{-}8ClO_4]^{5+}$, 504.3 $[M\text{-}6ClO_4]^{6+}$, 488.3 $[M\text{-}7ClO_4]^{6+}$, 470.3 $[M\text{-}8ClO_4]^{6+}$, 418.4 $[M\text{-}7ClO_4]^{7+}$, 403.9 $[M\text{-}8ClO_4]^{7+}$.

$\{[Ru(bpy)_2]_4L4\}(ClO_4)_8$ (Ru4). The yellow solid **L4** (61.0 mg, 0.05 mmol) was mixed with $[Ru(bpy)_2]Cl_2.2H_2O$ (130 mg, 0.25 mmol) in ethylene glycol (10 mL), the mixture was heated to 170 °C for 8 h under Ar. The cooled reaction mixture was diluted with water (10 mL). Saturated aqueous sodium perchlorate solution was added under vigorous stirring, and filtered. The dark red solid was collected and washed with small amounts of H_2O and Et_2O, then dried under vacuum, and purified by column chromatography on alumina using acetonitrile-ethanol as the eluant. The solvent was removed under reduced pressure and red microcrystals were obtained (150 mg, yield = 82%). Anal. Calcd. for $C_{152}H_{114}Cl_8N_{32}O_{37}Ru_4$: C, 49.76; H, 3.13; N, 12.22. Found: C, 49.45; H, 3.19; N, 12.08. ^{1}H NMR (500 MHz, d_6-DMSO) δ 9.05 (bs, 8H), 8.89 (d, *J* = 5 Hz, 8H), 8.86 (d, *J* = 5 Hz, 8H), 8.19-8.24 (q, 12H), 8.10 (t, *J* = 7.5 Hz, 10H), 7.93 (bs, 8H), 7.86 (s, 8H), 7.78 (bs, 8H), 7.57-7.66 (m, 16H), 7.36-7.41 (m, 8H), 4.43 (bs, 4H), 3.90 (s, 4H), 3.72 (s, 4H), 3.67 (s, 4H). ESI-MS: m/z = 791.7 $[M\text{-}5ClO_4]^{4+}$, 767.7 $[M\text{-}6ClO_4]^{4+}$, 741.9 $[M\text{-}7ClO_4]^{4+}$, 613.7 $[M\text{-}6ClO_4]^{5+}$, 593.9 $[M\text{-}7ClO_4]^{5+}$, 574.1 $[M\text{-}8ClO_4]^{5+}$, 494.8 $[M\text{-}7ClO_4]^{6+}$, 478.0 $[M\text{-}8ClO_4]^{6+}$, 424.5 $[M\text{-}7ClO_4]^{7+}$, 410.1 $[M\text{-}8ClO_4]^{7+}$, 359.4 $[M\text{-}8ClO_4]^{8+}$.

References

1 A. Jain, A. Paul, B. Maji, K. Muniyappa, S. Bhattacharya, *J. Med. Chem.*, 2012, **55**, 2981–2993.

2 M. Yamada, Y. Tanaka, Y. Yoshimoto, S. Kuroda, I. Shimao. *Bull. Chem. Soc. Jpn.*, 1992, **65**, 1006–1011.

3 B. Sullivan, D. Salmon, T. Meyer, *Inorg. Chem.*, 1978, **17**, 3334–3341.

Table S1 Photophysical data of Ru1-Ru4 in DMSO/H_2O (v/v=1:99) at 298 K

Complexes	λ_{max}[a]	ε_{max}[b]	λ_{max}[c]	φ[d]	$\tau/\mu S$[e]	δ/GM[f]
Ru1	458	6.15	613	0.070	0.165	60.0
Ru2	460	5.37	613	0.097	0.164	45.4
Ru3	458	6.31	612	0.086	0.185	32.4
Ru4	460	6.46	613	0.093	0.183	26.1

[a] λ_{max} values of the one-photon absorption in nm. [b] Extinction coefficient in 1×10^4 $M^{-1}\cdot cm^{-1}$.
[c] λ_{max} values of the one-photon emission spectra in nm. [d] Liminescence quantum yield. [e] Life time.
[f] Two-photon absorption cross section at 850 nm for **Ru1**, 810 nm for **Ru2**-**4**, measured in methanol.

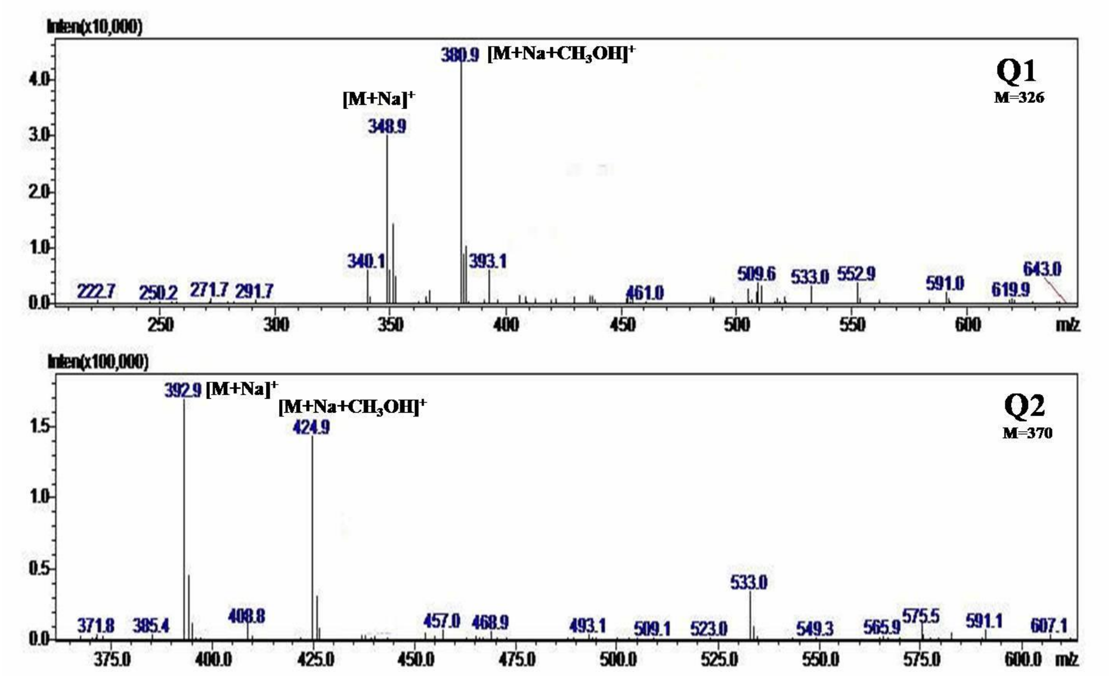


Fig. S1. ES-MS spectrums of **Q1-Q2**.

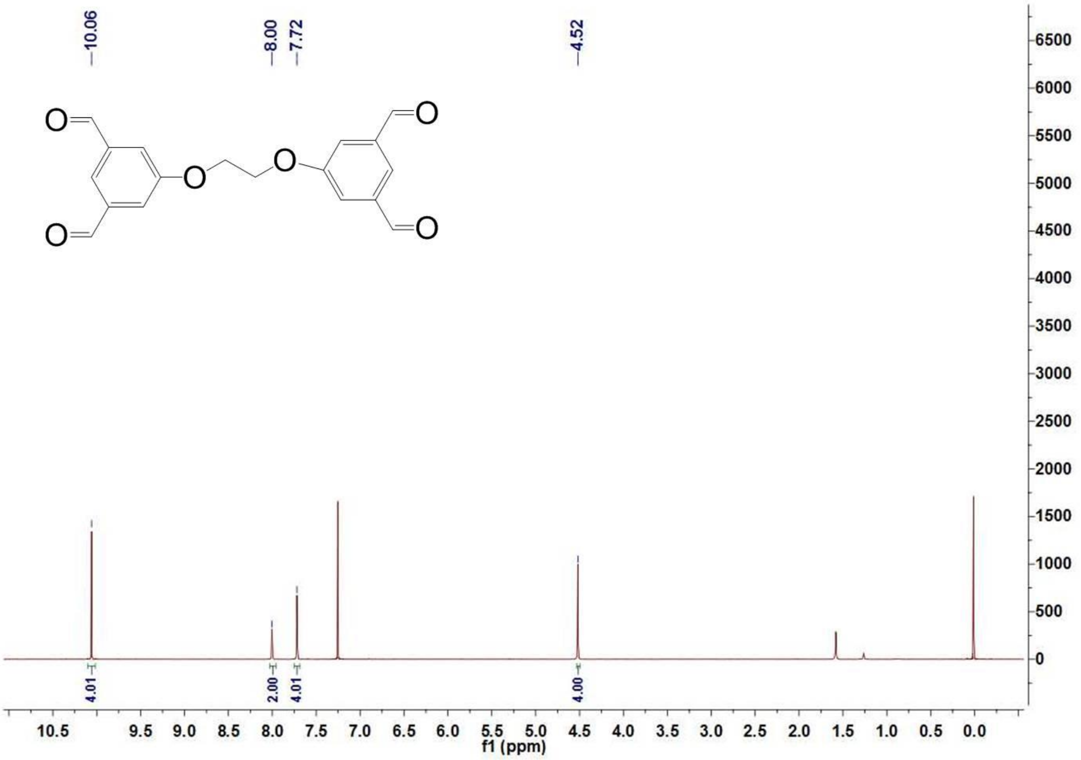


Fig. S2. ^{1}H NMR spectrum of **Q1**.

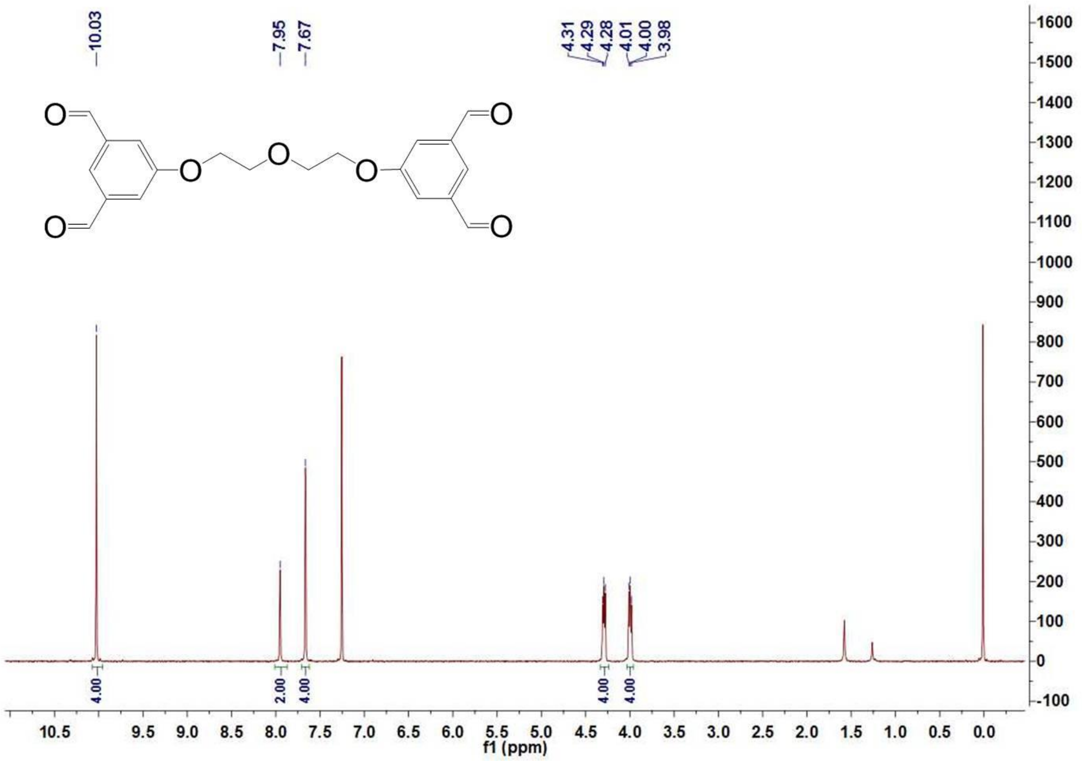


Fig. S3. ^{1}H NMR spectrum of **Q2**.

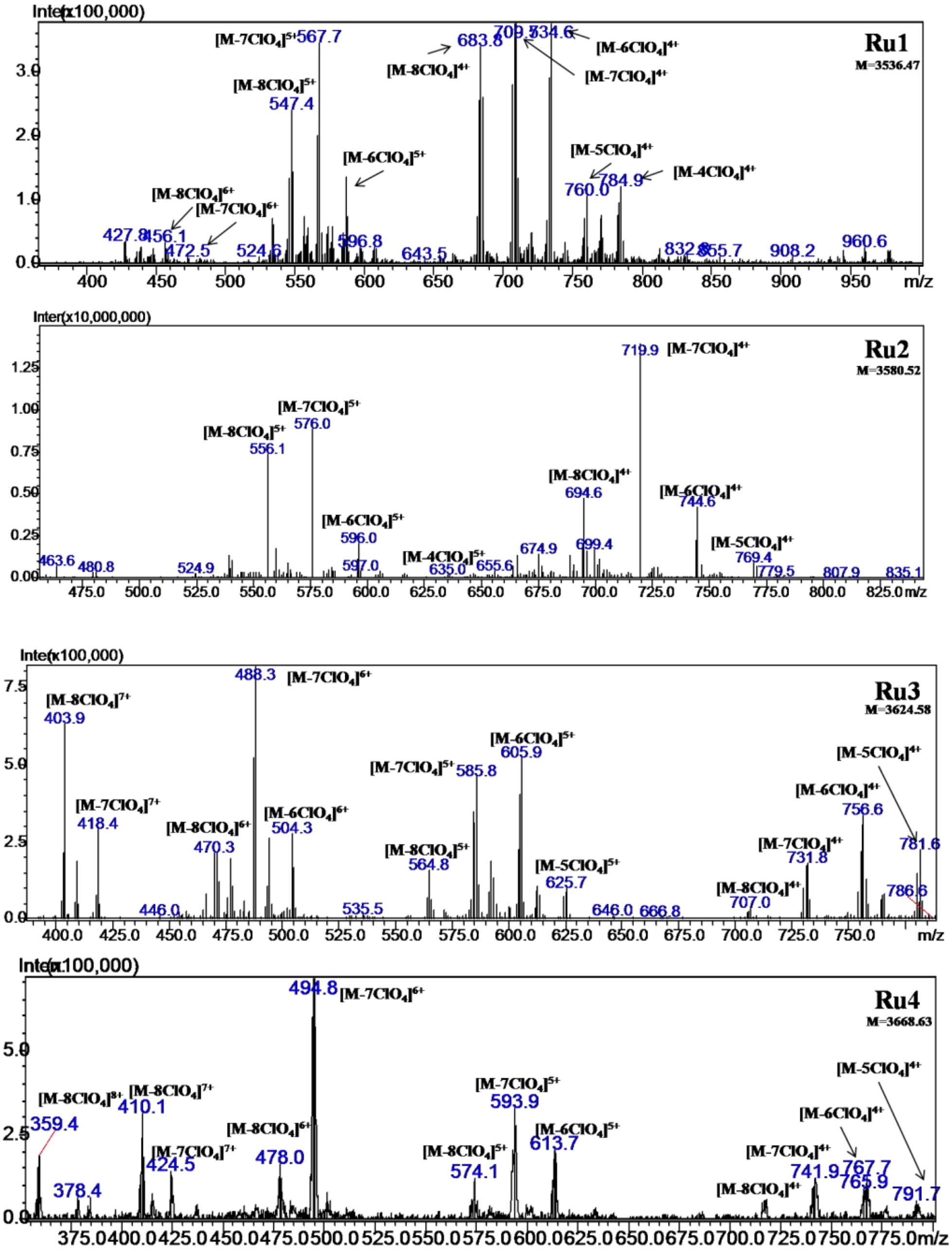


Fig. S4. ES-MS spectrums of **Ru1-Ru4**.

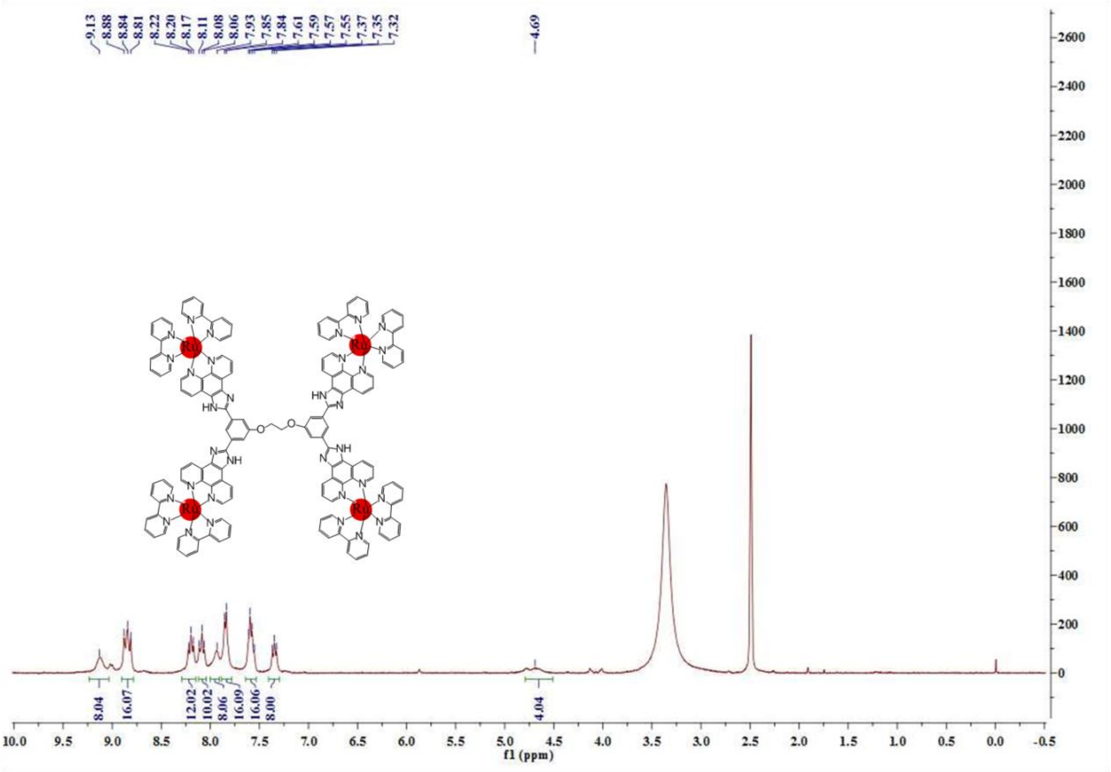


Fig. S5. ^{1}H NMR spectrum of **Ru1**.

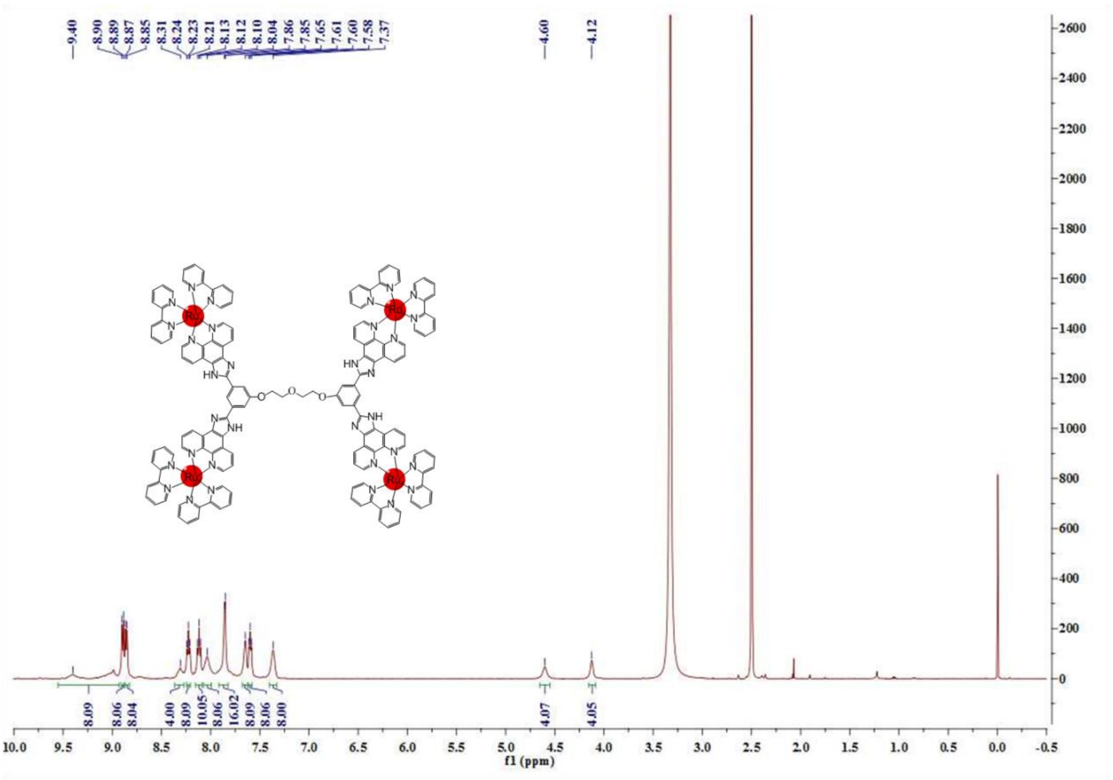


Fig. S6. ^{1}H NMR spectrum of **Ru2**.

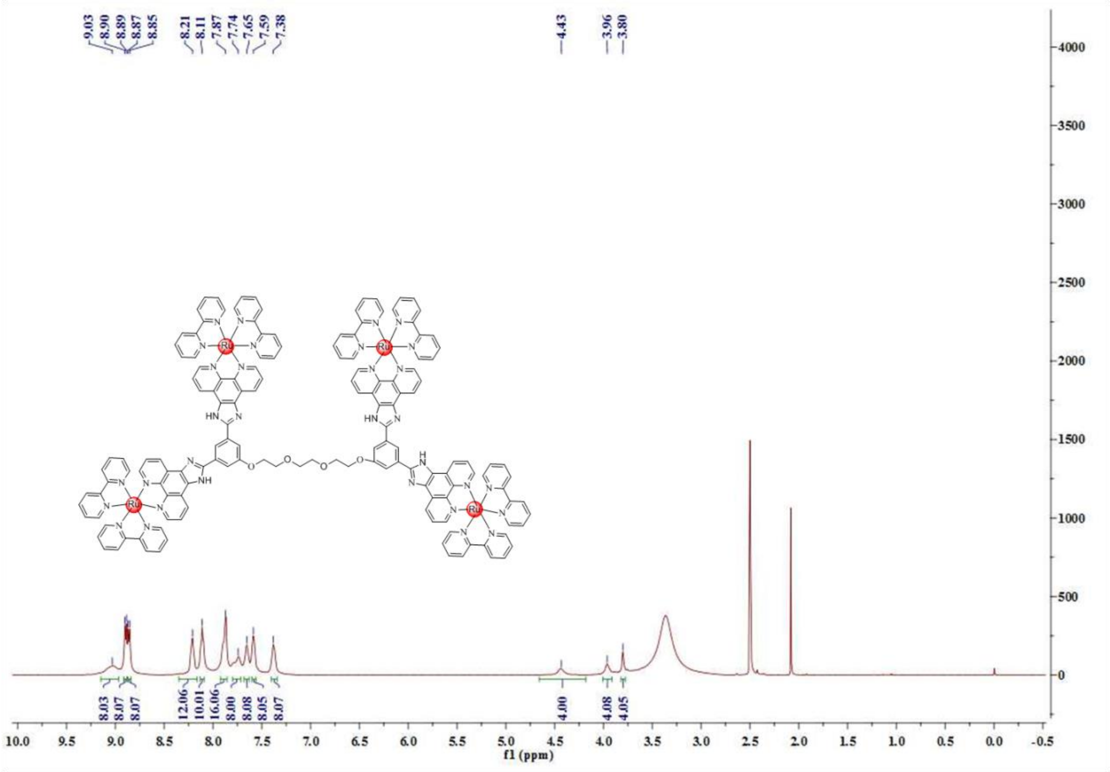


Fig. S7. ^{1}H NMR spectrum of **Ru3**.

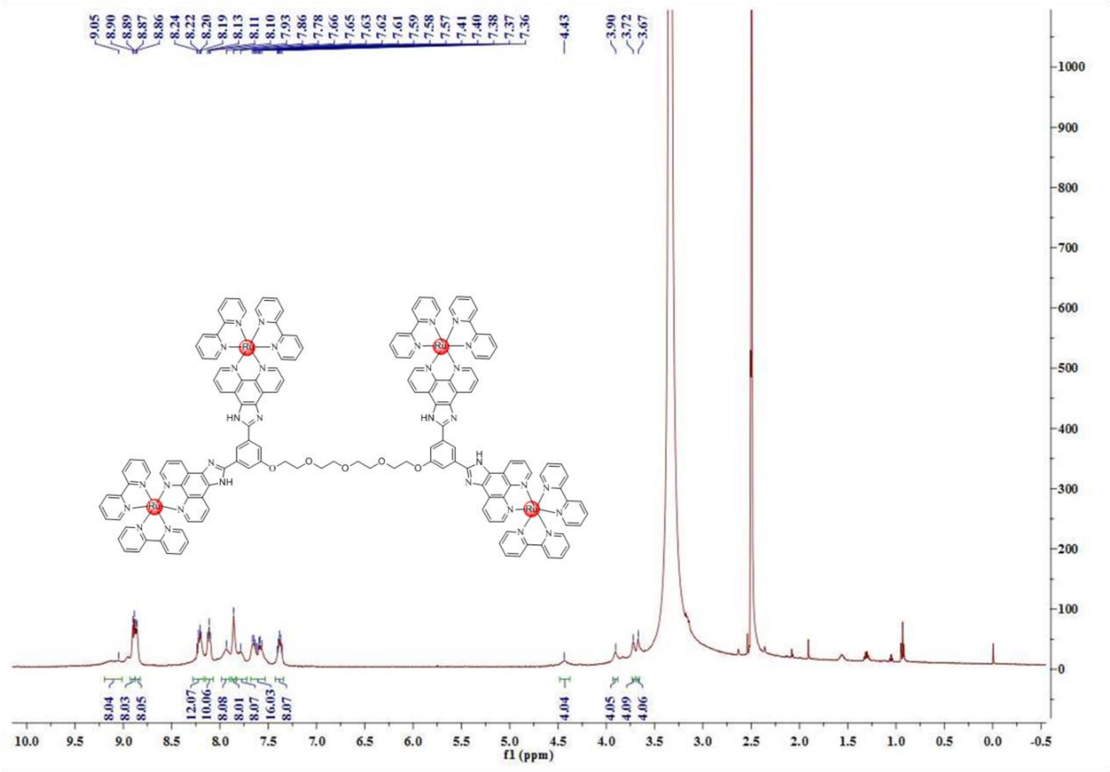


Fig. S8. ^{1}H NMR spectrum of **Ru4**.

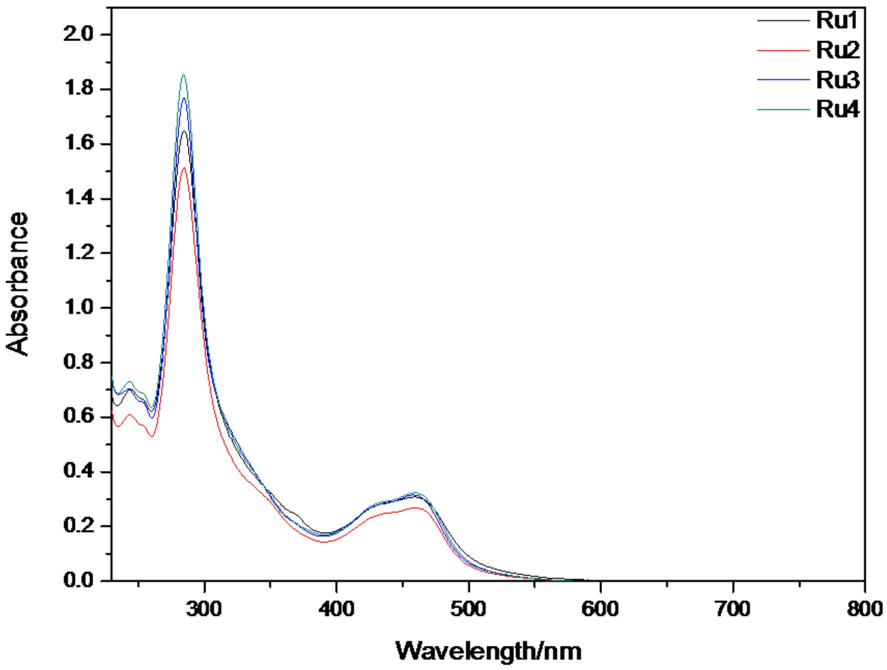


Fig. S9. Absorption spectra of **Ru1-Ru4** (5 µM).

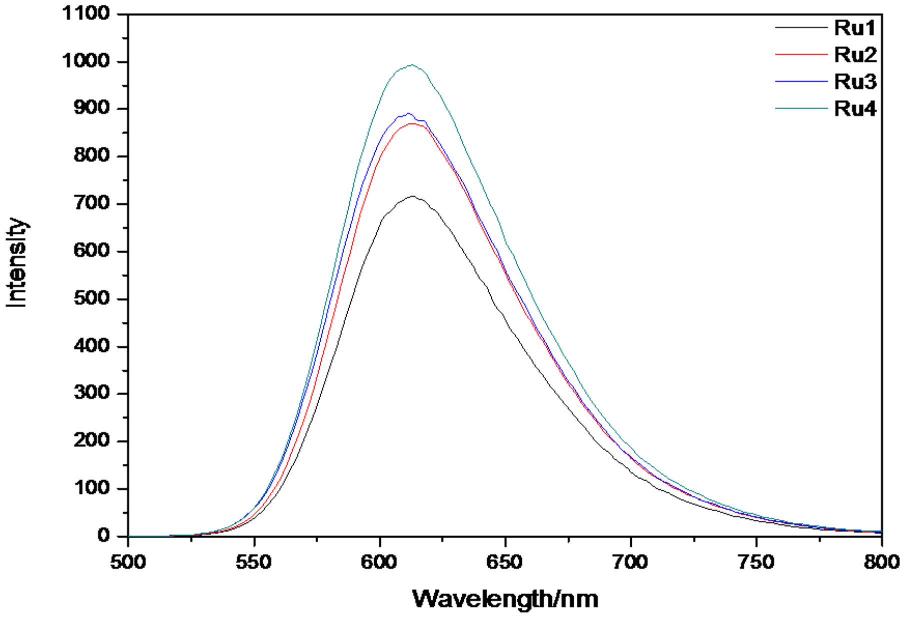


Fig. S10. Emission spectra of **Ru1-Ru4** (5 µM).

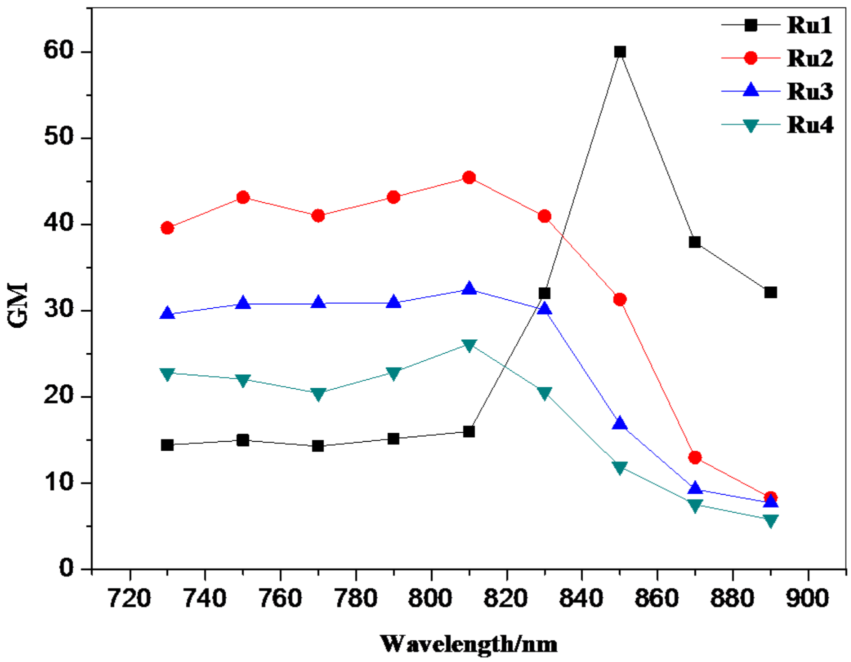


Fig. S11. Two-photon absorption cross-sections of **Ru1-Ru4** at different exitation wavelength from 730 to 890 nm.

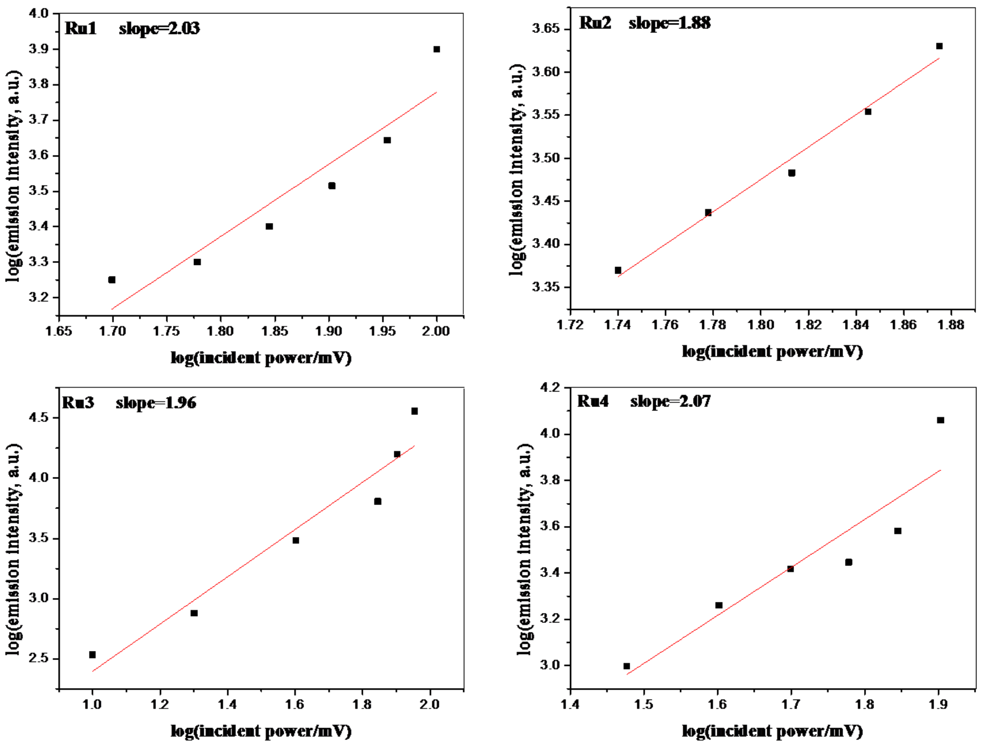


Fig. S12. The logarithmic plots of the power dependence of relative two-photon induced luminescence intensity of **Ru1**, **Ru2-Ru4** as a function of pump power at an excitation wavelength of 850 nm and 810 nm, respectively. The solid lines are the best-fit straight lines with gradient, slope=1.88-2.07, indicating that **Ru1-Ru4** is two-photon excitation active.

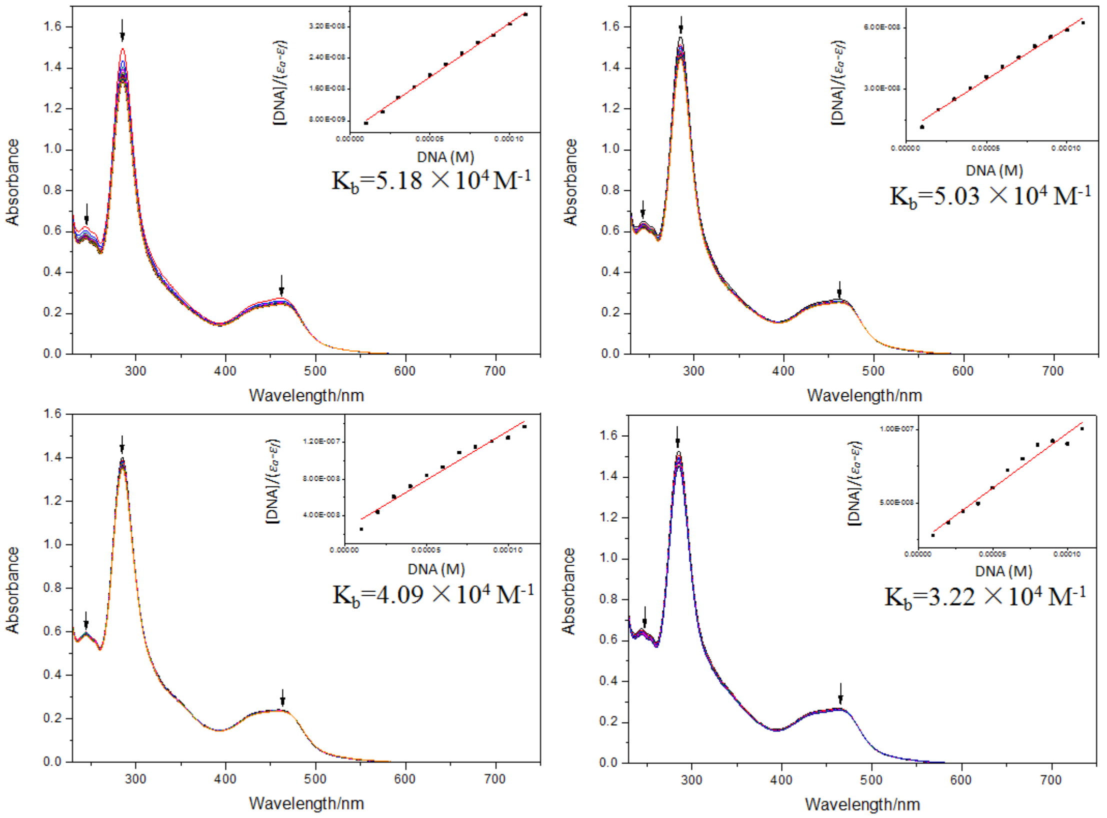


Fig. S13. Changes in absorption spectra of **Ru1-Ru4** (5 μM) upon addition of CT-DNA (0-110 μM). Inset: plot of [DNA]/(ε_a-ε_f) vs [DNA] and the linear fit for the titration of the complex with the DNA.

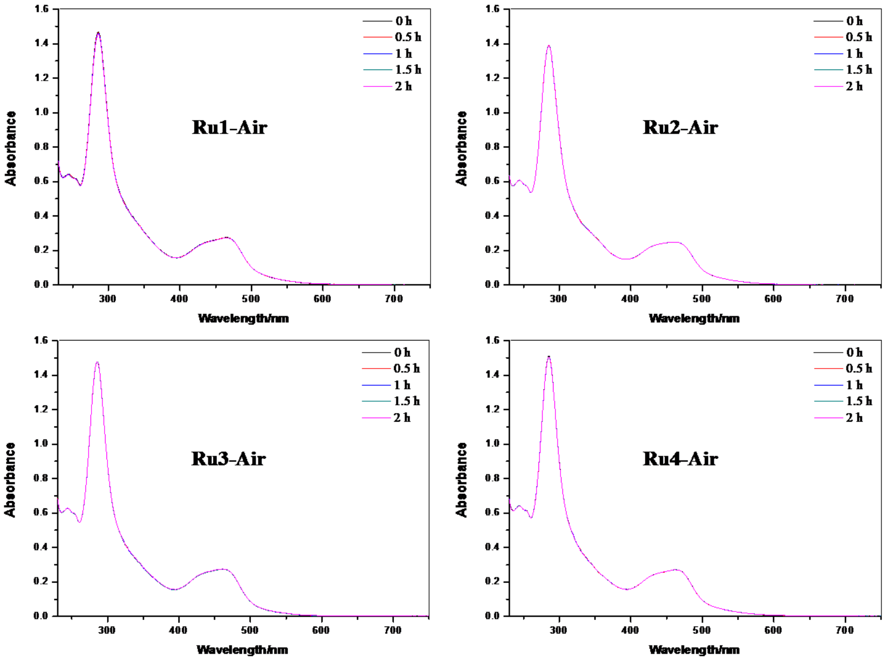


Fig. S14. Changes in absorption spectra of **Ru1-Ru4** (5 μM) under visible irradiation in the presence of CT-DNA (110 μM, base). Air means in Air-saturated solution. Irradiation time = 0, 0.5, 1, 1.5, 2 hours.

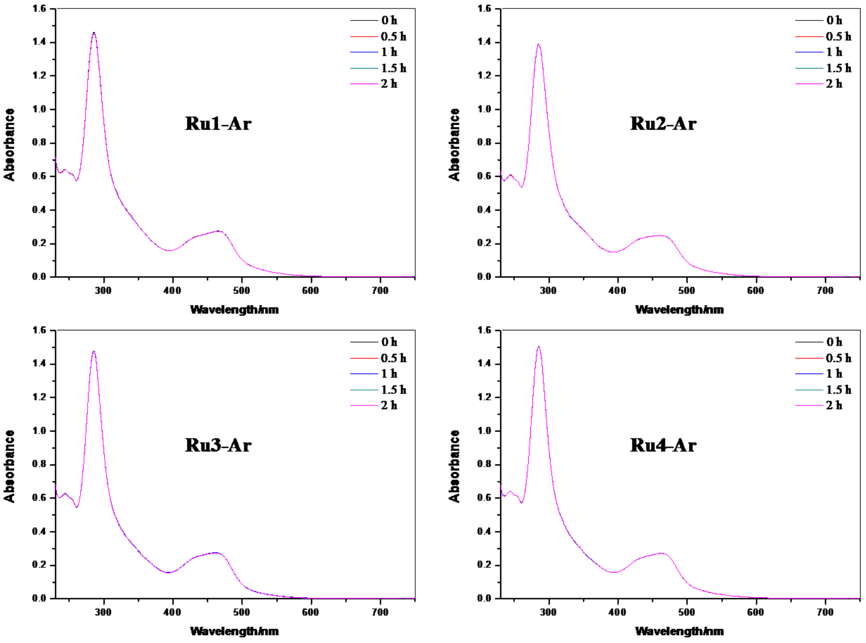


Fig. S15. Changes in absorption spectra of **Ru1-Ru4** (5 μM) under visible irradiation in the presence of CT-DNA (110 μM, base). Ar means in Argon-saturated solution. Irradiation time = 0, 0.5, 1, 1.5, 2 hours.

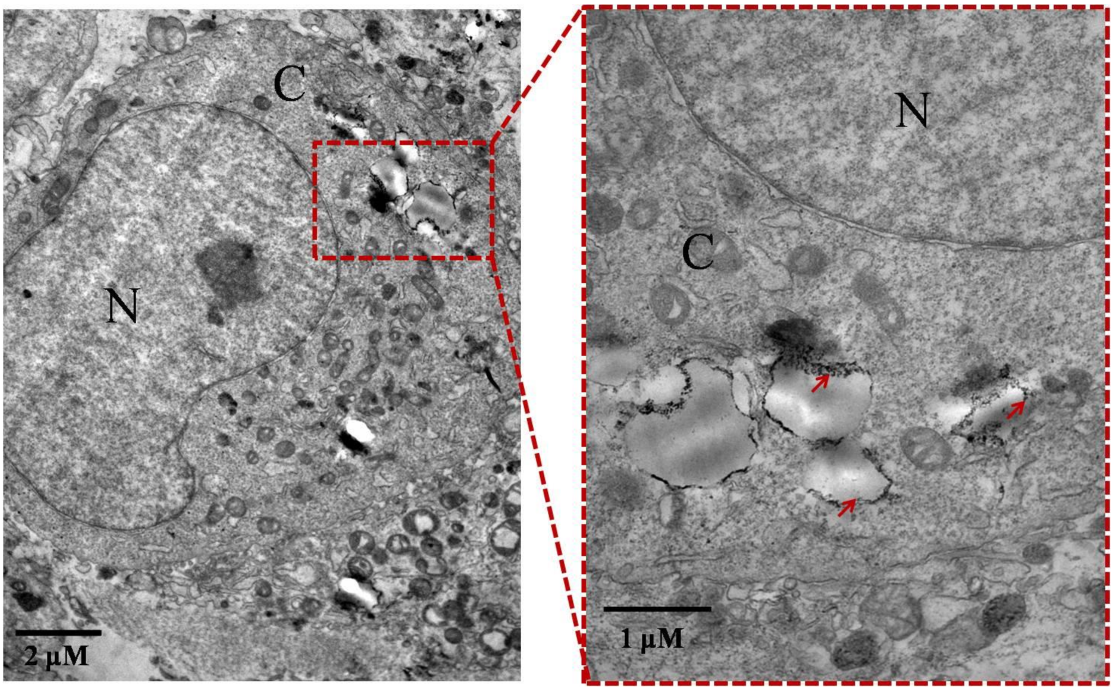


Fig. S16. Cellular uptake and intracellular localization of **Ru2**-pEGFP DNA particles at the +/- ratio of 80.0 monitored by TEM, N: nucleus, C: cytoplasm.

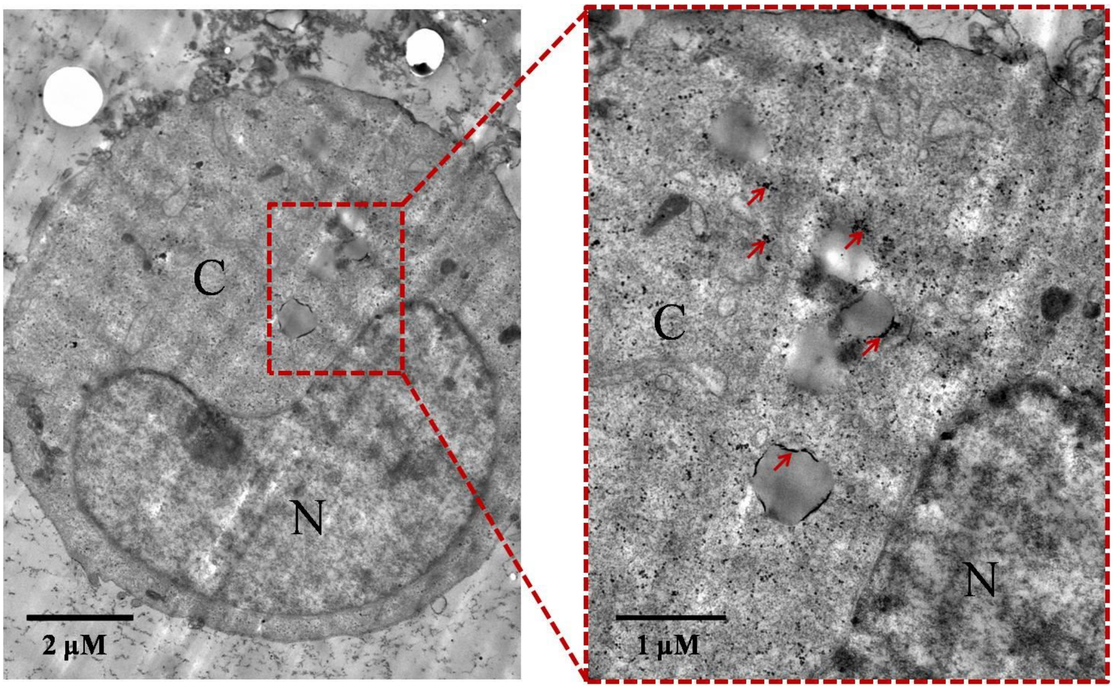


Fig. S17. Cellular uptake and intracellular localization of **Ru3**-pEGFP DNA particles at the +/- ratio of 106.7 monitored by TEM, N: nucleus, C: cytoplasm.

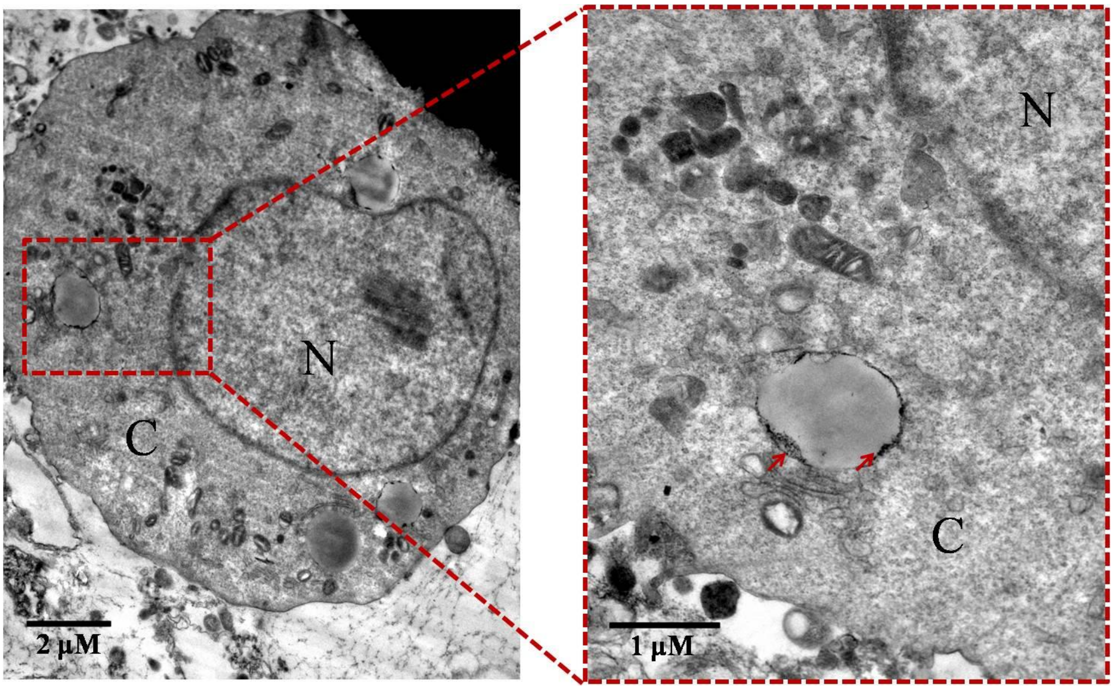


Fig. S18. Cellular uptake and intracellular localization of **Ru4**-pEGFP DNA particles at the +/- ratio of 133.3 monitored by TEM, N: nucleus, C: cytoplasm.

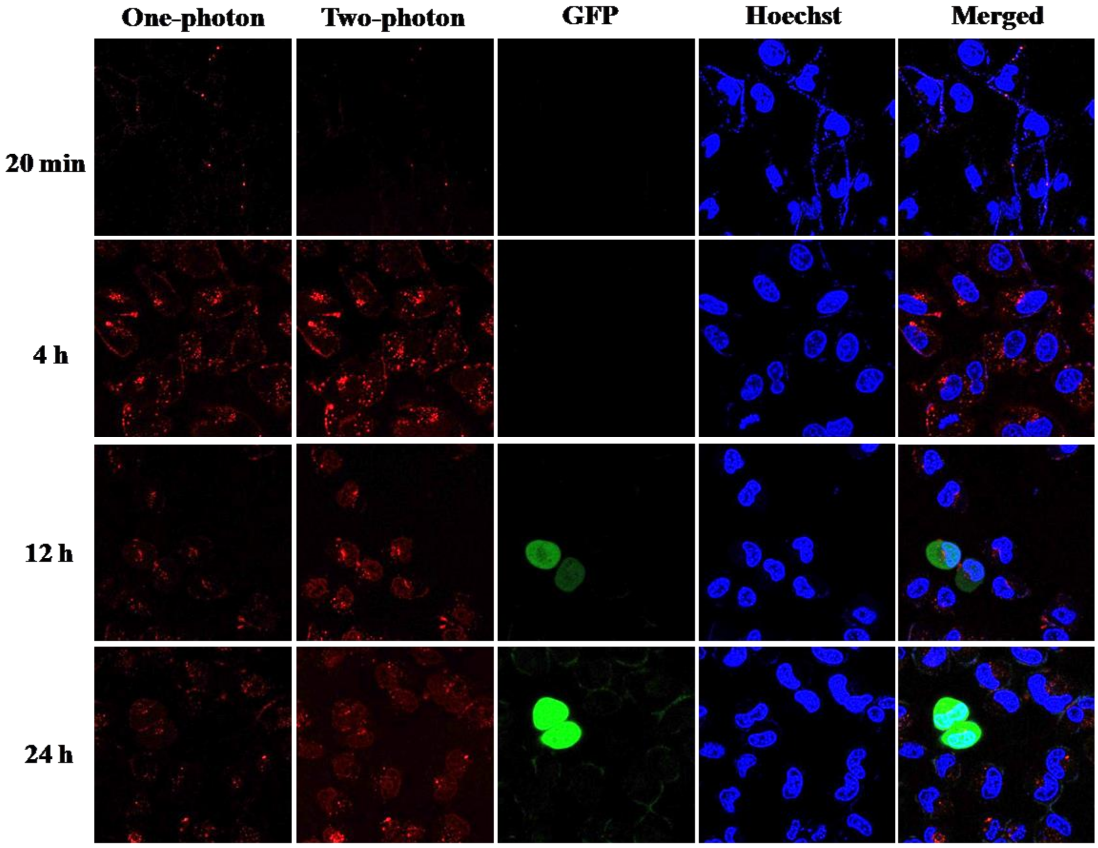


Fig. S19. Time-dependent confocal microscopy images of entry and transportation of **Ru2**-pEGFP DNA particles at the +/- ratio of 80.0 in Hela cells. The DNA concentration is 0.75 μM. The red luminescence was **Ru2**, the blue florescence was Hoechst 33258 and the green florescence was GFP.

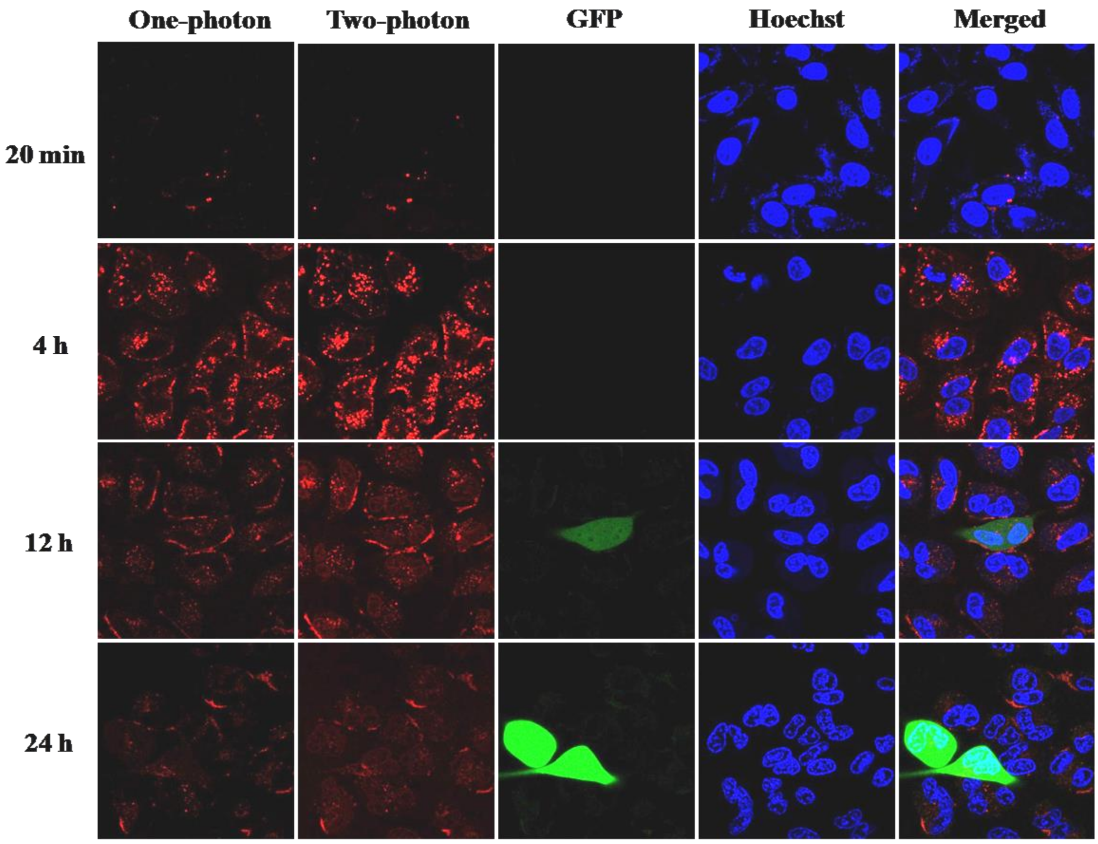


Fig. S20. Time-dependent confocal microscopy images of entry and transportation of **Ru3**-pEGFP DNA particles at the +/- ratio of 106.7 in Hela cells. The DNA concentration is 0.75 μM. The red luminescence was **Ru3**, the blue florescence was Hoechst 33258 and the green florescence was GFP.

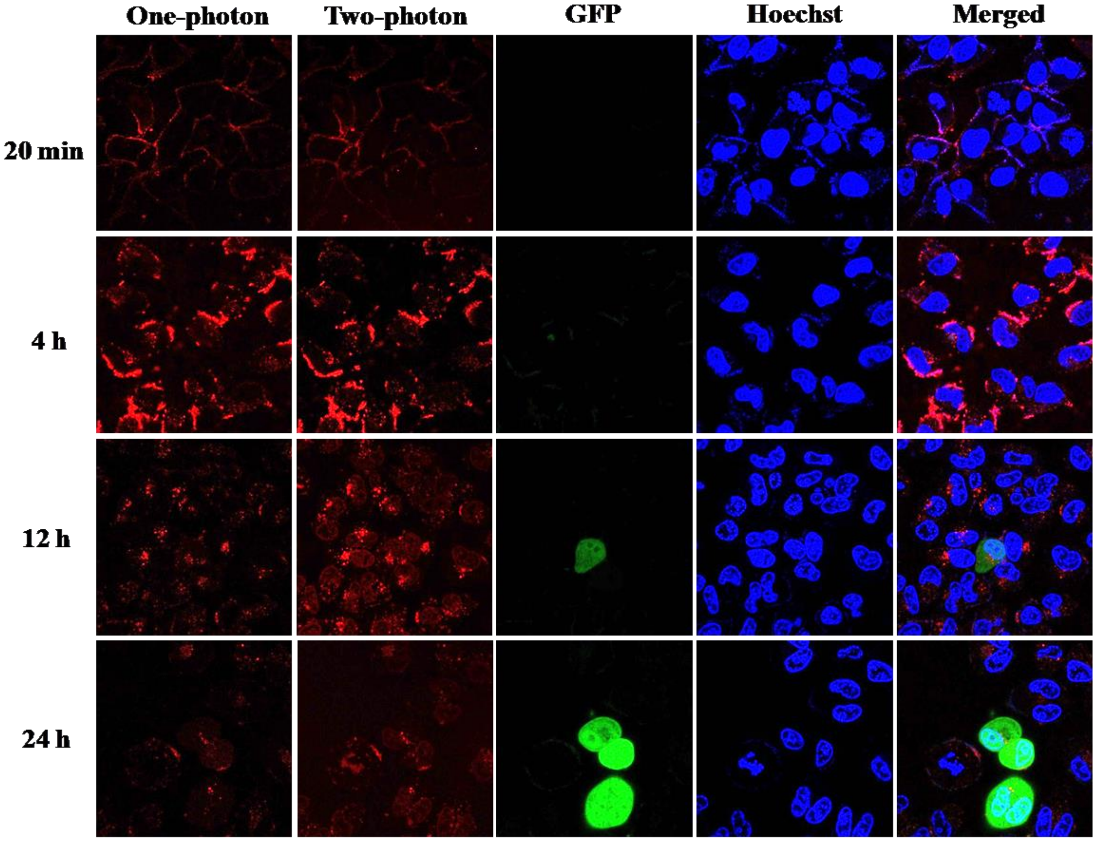


Fig. S21. Time-dependent confocal microscopy images of entry and transportation of **Ru4**-pEGFP DNA particles at the +/- ratio of 133.3 in Hela cells. The DNA concentration is 0.75 μM. The red luminescence was **Ru4**, the blue florescence was Hoechst 33258 and the green florescence was GFP.