SUPPORTING INFORMATION

Exploring the quinone/inhibitor-binding pocket in mitochondrial respiratory complex I by chemical biology approaches

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General synthetic methods

All moisture- and air-sensitive reactions were performed in oven-dried glassware under nitrogen or argon atmosphere with dry solvents under anhydrous conditions using standard syringe septum techniques. ^{1}H-NMR spectra were recorded at 400 or 500 MHz with Bruker AVANCE III 400 or 500 spectrometers, respectively, using tetramethylsilane (TMS) as the internal standard. ^{13}C-NMR spectra were recorded at 100 or 125 MHz, respectively. Chemical shifts (δ) were given in ppm relative to TMS with coupling constants (J) in Hz. The mass spectra were recorded on a Shimadzu LCMS-8040 with ESI source. Thin-layer chromatography (TLC) was performed on Merk TLC plate Silica-gel 60F254, and the spot was detected by iodine, anis, phosphomolybdic acid, or UV absorbance. Dry solvents were either used as purchased or freshly distilled using common practices where appropriate. HPLC purification was carried out with a Shimadzu LC-10 AS. Elution profiles were monitored at 254 nm with a Shimadzu SPD-10A.

Abbreviations

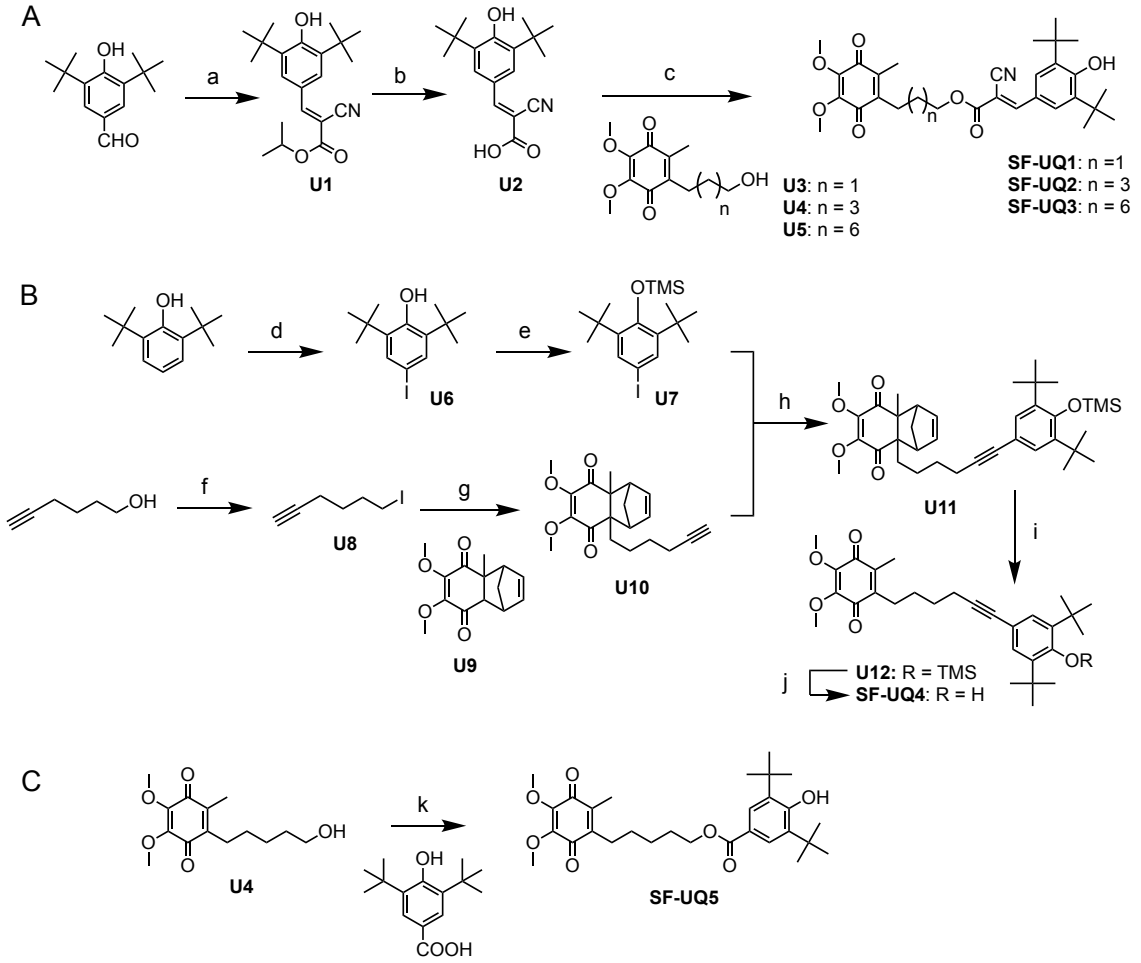
AcOH, acetic acid; Boc, *tert*-butoxycarbonyl; *t*-BuOK, potassium *tert*-butoxide; *t*-BuOH, *tert*-butylalcohol; chloramine T, sodium *p*-toluenesulfonchloramide; DCC, *N,N'*-dicyclohexylcarbodiimide; DDQ, 2,3-dichloro-5,6-dicyano-*p*-benzoquinone; DMAP, 4-dimethylaminopyridine; DMF, *N,N*-dimethylformamide; DMSO, dimethylsulfoxide; EDC, 1-ethyl-3-(3-dimethylaminopropyl)carbodiimide hydrochloride; Et_2O, diethyl ether; NIS, *N*-iodosuccinimide; $MeOP[N(iPr)_2]_2$, methyl *N,N,N',N'*-tetraisopropylphosphorodiamidite; MNBA, 2-methyl-6-nitrobenzoic anhydride; MOM, methoxymethyl; MS, molecular sieves; OMe, methoxy; PMB, *p*-methoxybenzyl; PPh_3, triphenylphosphine; rt, room temperature; $(iPr)_2NH$, diisopropylamine; TBAF, tetrabutylammonium fluoride; TBS, *tert*-butyldimethylsilyl; TEA, triethylamine; TfOH, trifluoromethanesulfonic acid; THF, tetrahydrofuran; TLC, thin-layer chromatography; TMEDA, tetramethylethylenediamine; TMS, trimethylsilyl; TsOH, *p*-toluenesulfonic acid.

*Outline of the syntheses of **SF-UQ1−SF-UQ5***

The synthetic procedures of **SF-UQ1**, **SF-UQ2**, and **SF-UQ3** are outlined in Scheme S1A. Knoevenagel condensation [*1*] of 3,5-di-*tert*-butyl-4-hydroxybenzaldehyde with cyanoacetic acid isopropyl ester afforded **U1**, followed by deprotection. Esterification of **U2** with an appropriate UQ analogue (**U3**-**U5**, *refs2 and 3*) furnished **SF-UQ1**, **SF-UQ2**, and **SF-UQ3**.

SF-UQ4 was synthesized as illustrated in Scheme S1B. Treatment of 2,6-di-*tert*-butylphenol with NIS in the presence of *p*-toluenesulfonic acid gave **U6**, followed by protection of the phenol by TMS to give **U7**. Sonogashira-type cross-coupling [*4*] of alkyne **U10,** which was prepared from alkyl iodide **U8** and Diels-Alder cycloadduct **U9** [*5*], with benzyl iodide **U7** gave **U11.** Cleavage of cyclopentadiene moiety of **U11** and deprotection of TMS group furnished **SF-UQ4**. **SF-UQ5** was synthesized by esterification of **U4** with 3,5-di-*tert*-butyl-4-hydroxybenzoic acid (Scheme S1C).

Scheme S1[a]

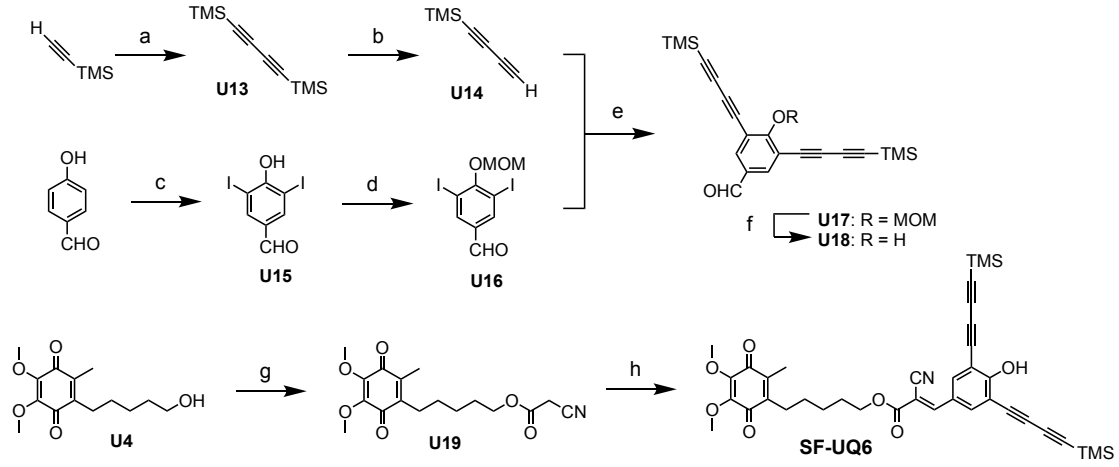


[a]*Reagents and conditions*: (a) piperidine, AcOH, toluene, reflux, 3 h; (b) 10% aq. NaOH, CH_3OH, 50˚C, 18 h, 79% (2 steps); (c) EDC or 2,4,6-trichlorobenzoyl chloride, DMAP, rt, 4−31 h, 21−67%; (d) NIS, TsOH, $CHCl_3$, rt, 17 h, 80%; (e) NaH, TMSCl, DMF, rt, 21 h, 36% ; (f) iodine, PPh_3, imidazole, CH_2Cl_2, rt, 1 h, 63%; (g) **U9**, *t*-BuOK, THF/DMF (1:4), 0˚C, 15 h, 65%; (h) $PdCl_2(PPh_3)_2$, CuI, $(iPr)_2NH$, rt, 2 h, 80%; (i) toluene, reflux, 2 h, quant.; (j) TBAF, AcOH, THF, rt, 1 h, quant.; (k) EDC, DMAP, rt, 31 h, 27%.

*Outline of the synthesis of **SF-UQ6***

The synthetic procedures of **SF-UQ6** are outlined in Scheme S2. Oxidative homo-coupling [*6*] of TMS-acetylenes gave di-TMS-diyne **U13**, followed by the treatment with 1.0~1.3 equivalent of MeLi to give mono-TMS-diyne **U14**. Sonogashira-type cross-coupling of diyne **U14** with diiodide **U16** gave **U17**, whose MOM group was deprotected in the presence of HCl to provide **U18**. Knoevenagel condensation of cyanoacetate **U19** with **U18** provided **SF-UQ6**.

Scheme S2[b]



[b]*Reagents and conditions*: (a) CuCl, TMEDA, acetone, rt, 10 min, 75%; (b) MeLi, Et_2O, rt; (c) $NaIO_4$, NaCl, KI, AcOH/H_2O (9:1), 50°C, 24 h, 65%; (d) NaH, MOMCl, THF, rt, 30 h, 87%; (e) $PdCl_2(PPh_3)_2$, CuI, (iPr)$_2$NH/THF (1:1), rt, 19 h, 46%; (f) THF/6.0 M aq. HCl (2:1), 50°C, 4.5 h, 67%; (g) cyanoacetic acid, EDC, DMAP, CH_2Cl_2, rt, 12 h, 72%; (h) **U18**, piperidine, AcOH, toluene, 70°C, 4 h, 49%.

*Synthesis of **U1***

To a solution of 3,5-di-*tert*-butyl-4-hydroxybenzaldehyde (800 mg, 3.4 mmol) and cyanoacetic acid isopropyl ester (433 mg, 3.4 mmol) in toluene (25 mL), piperidine (30 μL) and AcOH (40 μL) were added at room temperature, and the mixture was heated under reflux for 3 h. The reaction mixture was cooled to room temperature, quenched with water, extracted with toluene and dried over anhydrous $MgSO_4$. The removal of the organic solvent gave **U1**, which was subjected to the next reaction without further purification.

*Synthesis of **U2***

To a solution of crude **U1** in a mixture of methanol (20 mL) and water (10 mL), K_2CO_3 (940 mg, 6.8 mmol) was added in one portion, and the mixture was stirred for 4 h at room temperature. As the reaction did not proceed completely, 10% (w/v) aqueous NaOH (10mL) was added, and the mixture was stirred for further 12 h at room temperature. The reaction was quenched with 1.0 M aqueous HCl, and the MeOH was removed *in vacuo*. The mixture was extracted with diethyl ether and dried over anhydrous Na_2SO_4. The crude product was purified by silica gel column chromatography (Wako gel® C-200, 5% methanol/$CHCl_3$) to give **U2** as a white solid (812 mg, 2.7 mmol, 79%, 2 steps); ^{1}H-NMR (400 MHz, $CDCl_3$): δ 8.24 (s, 1H), 7.96 (s, 2H), 6.01 (s, 1H), 1.49 (s, 18H); ^{13}C-NMR (100 MHz, $CDCl_3$): δ 168.60, 159.91, 157.89, 137.30 (2C), 130.33 (2C), 123.45, 116.24, 97.30, 34.86 (2C), 30.28 (6C); ESI-MS (*m/z*) 300.2 $[M-H]^-$.

*Synthesis of **U3**, **U4**, and **U5***

These compounds were synthesized in 5 steps according to the procedures described in refs. *2* and *3* using commercially available 1,3-propanediol, 1,5-pentanediol, and 1,8-octanediol as a starting material, respectively. **U3**: ^{1}H-NMR (400 MHz, $CDCl_3$): δ 4.00 (s, 3H), 3.99 (s, 3H), 3.61 (t, 2H, J = 6.0 Hz), 2.59 (t, 2H, J = 7.4 Hz), 2.05 (s, 3H), 1.69 (tt, 2H, J = 6.0, 7.4 Hz); ^{13}C-NMR (100 MHz, $CDCl_3$): δ 184.88, 184.62, 144.69, 144.45, 142.45, 139.71, 66.03, 61.83, 61.38, 31.54, 22.56, 12.09. **U4**: ^{1}H-NMR (400 MHz, $CDCl_3$): δ 3.99 (s, 3H), 3.98 (s, 3H), 3.64 (t, 2H, J = 6.5 Hz), 2.47 (br t, 2H, J = 7.0 Hz), 2.01 (s, 3H), 1.60 (m, 2H), 1.46-1.40 (m, 4H); ^{13}C-NMR (100 MHz, $CDCl_3$): δ 184.85, 184.36, 144.52 (2C), 142.95, 139.04, 62.94, 61.35 (2C), 32.61, 28.67, 26.50, 26.15, 12.13; ESI-MS (*m/z*): 269.1 $[M+H]^+$. **U5**: ^{1}H-NMR (400 MHz, $CDCl_3$): δ 3.99 (s, 3H), 3.98 (s, 3H), 3.64 (t, J = 6.6 Hz, 2H), 2.45 (br t, J = 7.3 Hz, 2H), 2.01 (s, 3H), 1.56 (m, 2H), 1.44-1.27 (m, 10H); ^{13}C-NMR (100 MHz, $CDCl_3$): δ 184.91, 184.37, 144.51 (2C), 143.24, 138.90, 63.21, 61.35 (2C), 32.93, 29.92, 29.49, 29.45, 28.88, 26.57, 25.89, 12.11; ESI-MS (*m/z*) 311.2 $[M+H]^+$.

*Synthesis of **SF-UQ1***

To a solution of **U3** (15 mg, 0.063 mmol) and **U2** (21 mg, 0.069 mmol) in anhydrous CH_2Cl_2 (10 mL), 2,4,6-trichlorobenzoyl chloride (17 mg, 0.069 mmol) and DMAP (23 mg, 0.19 mmol) was added at room temperature

under N_2 atmosphere, and the mixture was stirred for 4 h at room temperature. The reaction was quenched with water, extracted with CH_2Cl_2, and dried over anhydrous $MgSO_4$. The crude product was purified by silica gel column chromatography (Wako gel® C-200, 15−20% ethyl acetate/*n*-hexane) to give **SF-UQ1** as an orange oil (7 mg, 0.013 mmol, 21%); ^{1}H-NMR (400 MHz, $CDCl_3$): δ 8.18 (s, 1H), 7.92 (s, 2H), 5.94 (br s, 1H), 4.32 (t, 2H, J = 6.1 Hz), 3.99 (s, 3H), 3.99 (s, 3H), 2.66 (br t, 2H, J = 7.7 Hz), 2.07 (s, 3H), 1.95-1.86 (m, 2H), 1.48 (s, 18H); ^{13}C-NMR (100 MHz, $CDCl_3$): δ 184.66, 184.24, 163.61, 159.36, 156.50, 144.68, 144.63, 141.60, 139.96, 137.15 (2C), 129.89 (2C), 123.61, 116.64, 98.05, 65.72, 61.37 (2C), 34.84 (2C), 30.28 (6C), 27.77, 23.28, 12.21; ESI-MS (*m/z*): 522.30 $[M-H]^-$.

*Synthesis of **SF-UQ2***

SF-UQ2 was prepared from **U4** according to the same procedure described for **SF-UQ1**, with the exception that EDC was used for esterification. The crude product was purified by silica gel column chromatography (Wako gel® C-200, 5% ethyl acetate/*n*-hexane to $CHCl_3$) to give **SF-UQ2** as an orange oil (9 mg, 0.016 mmol, 29%); ^{1}H-NMR (400 MHz, $CDCl_3$): δ 8.16 (s, 1H), 7.91 (s, 2H), 5.92 (br s, 1H), 4.30 (t, 2H, J = 6.6 Hz), 3.99 (s, 3H), 3.99 (s, 3H), 2.49 (br t, 2H, J = 7.1 Hz), 2.03 (s, 3H), 1.79 (m, 2H), 1.50-1.42 (m, 4H), 1.48 (s, 18H); ^{13}C-NMR (100 MHz, $CDCl_3$): δ 184.84, 184.34, 163.73, 159.21, 156.24, 144.56 (2C), 142.78, 139.15, 137.09 (2C), 129.79 (2C), 123.65, 116.69, 98.45, 66.20, 61.36 (2C), 34.83 (2C), 30.29 (6C), 28.54, 28.47, 26.43, 26.21, 12.19; ESI-MS (*m/z*) 550.3 $[M-H]^-$.

*Synthesis of **SF-UQ3***

SF-UQ3 was prepared from **U5** according to the same procedure described for **SF-UQ1**. The crude product was purified by silica gel column chromatography (Wako gel® C-200, 2% $CH_3OH/CHCl_3$) to give **SF-UQ3** as an orange oil (26 mg, 0.044 mmol, 67%); ^{1}H-NMR (400 MHz, $CDCl_3$): δ 8.16 (s, 1H), 7.91 (s, 2H), 5.92 (brs , 1H), 4.29 (t, 2H, J = 6.7 Hz), 3.99 (s, 3H), 3.99 (s, 3H), 2.45 (br t, 2H, J = 7.2 Hz), 2.01 (s, 3H), 1.80-1.70 (m, 2H), 1.50-1.30 (m, 10H), 1.48 (s, 18H); ^{13}C-NMR (100 MHz, $CDCl_3$): δ 184.90, 184.34, 163.75, 159.16, 156.13, 144.52 (2C), 143.23, 138.89, 137.06 (2C), 129.74 (2C), 123.65, 116.70, 98.56, 66.50, 61.34 (2C), 34.81 (2C), 30.27 (6C), 29.93, 29.41, 29.28, 28.89, 28.73, 26.57, 25.97, 12.11; ESI-MS (*m/z*); 594.4 $[M+H]^+$, 592.3 $[M-H]^-$.

*Synthesis of **U6***

To an ice-cooled solution of 2,6-di-*tert*-butylphenol (1.0 g, 4.9 mmol) in $CHCl_3$ (50 mL), *p*-toluenesulfonic acid (911 mg, 5.3 mmol) and NIS (1.2 g, 5.3 mmol) were added, and the mixture was stirred for 17 h at room temperature. The reaction was quenched with saturated aqueous $NaHCO_3$, extracted with $CHCl_3$, dried over anhydrous $MgSO_4$. The crude product was purified by silica gel column chromatography (Wako gel® C-200, *n*-hexane) to give **U6** as a yellow solid (1.30 g, 3.9 mmol, 80%); ^{1}H-NMR (400 MHz, $CDCl_3$): δ 7.42 (s, 2H),

5.19 (s, 1H), 1.41 (s, 18H); ^{13}C-NMR (100 MHz, $CDCl_3$): δ 154.01, 138.86 (2C), 134.14 (2C), 83.67, 34.57 (2C), 30.29 (6C).

*Synthesis of **U7***

To a suspension of NaH (67 mg, 60% in mineral oil, 1.66 mmol) in anhydrous DMF (12 mL), **U6** (500 mg, 1.50 mmol) was added at 0˚C under N_2 atmosphere. After stirring the mixture at 0˚C for 30 min, TMSCl (180 mg, 1.66 mmol) was added, and the mixture was stirred for 21 h at room temperature. The reaction was quenched with saturated aqueous NH_4Cl, extracted with diethyl ether, and dried over anhydrous $MgSO_4$. The crude product was purified by silica gel column chromatography (Wako gel® C-200, 0-2.5% ethyl acetate/*n*-hexane) to give **U7** as a white solid (243 mg, 0.60 mmol, 36%); ^{1}H-NMR (400 MHz, $CDCl_3$): δ 7.48 (s, 2H), 1.37 (s, 18H), 0.40 (s, 9H); ^{13}C-NMR (100 MHz, $CDCl_3$): δ 153.62, 143.85 (2C), 135.04 (2C), 85.40, 35.35 (2C), 31.24 (6C), 4.09 (3C).

*Synthesis of **U8***

To a suspension of PPh_3 (1.7 g, 6.63 mmol) and imidazole (450 mg, 6.63 mmol) in anhydrous CH_2Cl_2 (15 mL), iodine (1.67 g, 6.63 mmol) was added at room temperature. After the mixture was stirred for 10 min, a solution of 5-hexyne-1-ol (500 mg, 5.09 mmol) in anhydrous CH_2Cl_2 (10 mL) was added dropwise to the mixture, and the mixture was stirred for 1 h at room temperature. The reaction was quenched with saturated aqueous $Na_2S_2O_3$, extracted with hexane, and dried over anhydrous $MgSO_4$. The crude product was purified by silica gel column chromatography (Wako gel® C-200, 0-5% ethyl acetate/*n*-hexane) to give **U8** as a colorless oil (670 mg, 3.22 mmol, 63%); ^{1}H-NMR (400 MHz, $CDCl_3$): δ 3.21 (t, 2H, J = 6.9 Hz), 2.23 (dt, 2H, J = 2.6, 7.0 Hz), 1.96 (t, 1H, J = 2.7 Hz), 1.95 (m, 2H), 1.64 (m, 2H); ^{13}C-NMR (100 MHz, $CDCl_3$): δ 83.81, 69.09, 32.44, 29.29, 17.59, 6.22.

*Synthesis of **U9***

Diels-Alder cycloadduct **U9** was prepared according to the procedure described in ref. *5* using commercially available 2,3-dimethoxy-5-methyl-1,4-benzoquinone and freshly distilled cyclopentadiene; ^{1}H-NMR (400 MHz, $CDCl_3$): δ 6.16 (m, 1H), 6.02 (m, 1H), 3.94 (s, 3H), 3.93 (s, 3H), 3.43 (s, 1H), 3.09 (s, 1H), 2.84 (d, J = 3.1 Hz, 2H), 1.67 (d, J = 7.3 Hz, 1H), 1.55 (d, J = 7.3 Hz, 1H), 1.49 (s, 3H); ^{13}C-NMR (100 MHz, $CDCl_3$): δ 198.64, 195.05, 150.78, 150,72, 138.34, 134.69, 60.83 (2C), 57.26, 53.59, 52.74, 49.02, 46.52, 26.70; ESI-MS (*m/z*) 249.1 $[M+H]^+$.

*Synthesis of **U10***

To a solution of **U9** (200 mg, 0.80 mmol) in a mixture of anhydrous THF (5 mL) and DMF (15 mL), potassium

tert-butoxide (135 mg, 1.2 mmol) was slowly added at -30˚C under N_2 atmosphere. After the mixture was stirred for 10 min at -30˚C, **U8** (200 mg, 0.76 mmol) in THF (3 mL) was added, and the mixture was stirred at 0˚C for 12 h. The reaction was quenched with saturated aqueous NH_4Cl, extracted with diethyl ether, and dried over anhydrous $MgSO_4$. The crude product was purified by silica gel column chromatography (Wako gel® C-200, 5-10% ethyl acetate/*n*-hexane) to give **U10** as a yellow oil (170 mg, 0.52 mmol, 65%); ^{1}H-NMR (400 MHz, $CDCl_3$): δ 6.06 (m, 2H), 3.94 (s, 3H), 3.91 (s, 3H), 3.11 (m, 1H), 3.00 (m, 1H), 2.20 (dt, 2H, J = 2.6, 7.0 Hz), 2.00-1.90 (m, 1H), 1.92 (t, 1H, J = 2.7 Hz), 1.80-1.72 (m, 2H), 1.58-1.38 (m, 5H), 1.49 (s, 3H); ^{13}C-NMR (100 MHz, $CDCl_3$): δ 198.75, 198.19, 150.58, 149.35, 138.18, 137.30, 84.08, 68.80, 60.36, 60.32, 59.48, 56.32, 54.42, 52.78, 43.45, 36.75, 29.08, 25.39, 23.36, 18.24.

*Synthesis of **U11***

To a solution of **U7** (70 mg, 0.17 mmol) and **U10** (52 mg, 0.16 mmol) in anhydrous (iPr)$_2$NH (8 mL), $PdCl_2(PPh_3)_2$ (5.0 mg, 0.008 mmol) and CuI (3.0 mg, 0.016 mmol) were added under Ar atmosphere, and the mixture was stirred for 2 h at room temperature. The reaction was quenched with saturated aqueous NH_4Cl, extracted with diethyl ether, and dried over anhydrous $MgSO_4$. The crude product was purified by silica gel column chromatography (Wako gel® C-200, 15% ethyl acetate/*n*-hexane) to give **U11** as a yellow oil (77 mg, 0.127 mmol, 80%); ^{1}H-NMR (400 MHz, $CDCl_3$): δ 6.06 (m, 2H), 3.89 (s, 3H), 3.86 (s, 3H), 3.12 (br s, 1H), 3.00 (br s, 1H), 2.40 (t, 2H, J = 6.8 Hz), 2.05-1.95 (m, 1H), 1.85-1.75 (m, 2H), 1.53 (s, 3H), 1.65-1.40 (m, 5H), 1.38 (s, 18H), 0.39 (s, 9H); ^{13}C-NMR (100 MHz, $CDCl_3$): δ 198.88, 198.35, 153.42, 150.71, 149.42, 141.19 (2C), 138.32, 137.38, 129.31 (2C), 115.62, 87.50, 81.99, 60.37 (2C), 59.65, 56.43, 54.46, 52.86, 43.53, 36.98, 35.26 (2C), 31.33 (6C), 29.61, 25.71, 23.49, 19.34, 4.01 (3C).

*Synthesis of **U12***

U11 (77 mg, 0.127 mmol) was dissolved in toluene (10 mL), and the solution was heated under reflux for 2 h. Then the solvent was removed under reduced pressure. The crude product was purified by silica gel column chromatography (Wako gel® C-200, 10% ethyl acetate/*n*-hexane) to give **U12** as an orange oil (50 mg, 0.093 mmol, 73%); ^{1}H-NMR (400 MHz, $CDCl_3$): δ 7.28 (s, 2H), 3.988 (s, 3H), 3.986 (s, 3H), 2.52 (brt, 2H, J = 7.6 Hz), 2.44 (t, 2H, J = 6.8 Hz), 2.04 (s, 3H), 1.70-1.65 (m, 4H), 1.38 (s, 18H), 0.39 (s, 9H); ^{13}C-NMR (100 MHz, $CDCl_3$): δ 184.85, 184.30, 153.43, 144.55 (2C), 142.90, 141.18 (2C), 139.16, 129.40 (2C), 115.70, 87.50, 81.92, 61.36 (2C), 35.28 (2C), 31.34 (6C), 29.01, 28.06, 26.04, 19.39, 12.18, 4.04 (3C).

*Synthesis of **SF-UQ4***

To an ice-cooled solution of **U12** (6 mg, 0.011 mmol) in THF (3 mL), AcOH (10 μL) and TBAF (22 μL, 1.0 M solution in THF, 0.022 mmol) were added at 0˚C and the mixture was stirred for 1 h at room temperature.

Then the reaction mixture was quenched with saturated aqueous NH_4Cl at 0˚C, extracted with diethyl ether, and dried over anhydrous $MgSO_4$. The crude product was purified by silica gel column chromatography (Wako gel® C-200, 10% ethyl acetate/*n*-hexane) to give **SF-UQ4** as an orange oil (5.0 mg, 0.011 mmol, quant.); ^{1}H-NMR (400 MHz, $CDCl_3$): δ 7.20 (s, 2H), 5.28 (s, 1H), 3.987 (s, 3H), 3.985 (s, 3H), 2.52 (t, 2H, J = 7.6 Hz), 2.43 (t, 2H, J = 6.8 Hz), 2.04 (s, 3H), 1.70-1.55 (m, 4H), 1.42 (s, 18H); ^{13}C-NMR (100 MHz, $CDCl_3$): δ 184.86, 184.30, 153.99, 144.59, 144.55, 142.91, 139.16, 136.15 (2C), 128.65 (2C), 114.85, 86.97, 82.06, 61.36 (2C), 34.48 (2C), 30.39 (6C), 29.03, 28.05, 26.04, 19.37, 12.18; ESI-MS (*m/z*) 467.3 $[M+H]^+$, 465.3 $[M-H]^-$.

Synthesis of ***SF-UQ5***

To a solution of **U4** (16 mg, 0.060 mmol) and 3,5-di-*tert*-butyl-4-hydroxybenzoic acid (17 mg, 0.066 mmol), EDC (13 mg, 0.066 mmol) and DMAP (8 mg, 0.066 mmol) were added, and the mixture was stirred for 31 h at room temperature. The reaction was quenched with water, extracted with CH_2Cl_2, and dried over anhydrous $MgSO_4$. The crude product was purified by silica gel column chromatography (Wako gel® C-200, 20% ethyl acetate/*n*-hexane) to provide **SF-UQ5** as an orange oil (8 mg, 0.016 mmol, 27%); ^{1}H-NMR (400 MHz, $CDCl_3$): δ 7.89 (s, 2H), 5.66 (s, 1H), 4.28 (t, 2H, J = 6.6 Hz), 3.99 (s, 3H), 3.99 (s, 3H), 2.48 (br t, 2H, J = 7.2 Hz), 2.01 (s, 3H), 1.82-1.75 (m, 2H), 1.50-1.44 (m, 4H), 1.44 (s, 18H); ^{13}C-NMR (100 MHz, $CDCl_3$): δ 184.83, 184.32, 167.36, 158.31, 144.55 (2C), 142.89, 139.06, 135.90 (2C), 127.20 (2C), 121.63, 61.36 (2C), 34.54 (2C), 30.36 (6C), 28.87, 28.61, 26.55, 12.15; ESI-MS (*m/z*) 501.4 $[M+H]^+$, 499.3 $[M-H]^-$.

Synthesis of ***U13***

To a suspension of CuCl (99 mg, 1.0 mmol) in acetone (5 mL), TMEDA (35 mg, 0.30 mmol) was added at room temperature. After the mixture was stirred for 30 min at room temperature, the mixture was poured in a separate funnel. Then, trimethylsilylacetylene (1.0 g, 10 mmol) was added, and the mixture was shaken until the solution became blue. After the solvent was removed under reduced pressure, water was added to the residue, and the mixture was extracted with hexane, and dried over anhydrous $MgSO_4$. The crude product was purified by silica gel column chromatography (Wako gel® C-200, *n*-hexane) to provide **U13** as a white solid (744 mg, 3.83 mmol, 75%); ^{1}H-NMR (400 MHz, $CDCl_3$): δ 0.19 (s, 18H); ^{13}C-NMR (100 MHz, $CDCl_3$): δ 88.20 (2C), 86.19 (2C), -0.28 (6C).

Synthesis of ***U14***

To a solution of **U13** (1.18 g, 6.07 mmol) in anhydrous diethyl ether (25 mL), MeLi (6.1 mL, 1.1 M solution in diethyl ether, 6.70 mmol) was added at -78˚C under N_2 atmosphere. After stirring the mixture for 10 h at room temperature, the reaction was quenched with saturated aqueous NH_4Cl, extracted with diethyl ether, and dried over anhydrous $MgSO_4$. The removal of the organic solvent gave the crude **U14** as a mixture of **U13**,

which was subjected to the next reaction without further purification; ^{1}H-NMR (400 MHz, $CDCl_3$): δ 2.11 (s, 1H), 0.21 (s, 9H). *As the boiling point of **U14** is expected to low (below 120˚C), the solvent should be carefully removed by rotary evaporator (over 70 mmHg, at 30˚C). The ratio of **U13** and **U14** was estimated to be 1: 5 by ^{1}H-NMR.

*Synthesis of **U15***

To a solution of 4-hydorxybenzaldehyde (1.0 g, 8.20 mmol) in a mixture of AcOH (18 mL) and water (2 mL), $NaIO_4$ (1.75 g, 8.20 mmol), NaCl (958 mg, 16.4 mmol), and KI (1.36 g, 8.20 mmol) were added at room temperature, and the mixture was stirred for 1 h at 50 ˚C. After stirring for a further 40 h at room temperature, the reaction was quenched with ice water to form precipitate. The precipitate was filtered off, washed with water and dried in a desiccator to give **U15** as a white solid (2.00 g, 5.35 mmol, 65%); ^{1}H-NMR (400 MHz, $CDCl_3$): δ 9.74 (s, 1H), 8.20 (s, 2H), 6.27 (br s, 1H); ^{13}C-NMR (100 MHz, $CDCl_3$): δ 187.86, 158.55, 141.24 (2C), 133.08, 82.88 (2C); ESI-MS (*m/z*): 372.8 $[M-H]^-$.

*Synthesis of **U16***

To a suspension of NaH (93 mg, 60% in mineral oil, 2.31 mmol) in anhydrous THF (30 mL), **U15** (865 mg, 2.31 mmol) was added at room temperature under N_2 atmosphere. After the mixture was stirred for 30 min at room temperature, chloromethyl methyl ether (188 mg, 2.33 mmol) was added, and the mixture was stirred for 20 h at room temperature. The reaction was quenched with saturated aqueous NH_4Cl, extracted with diethyl ether, and dried over anhydrous $MgSO_4$. The crude product was purified by silica gel column chromatography (Wako gel® C-200, 20% ethyl acetate/*n*-hexane) to provide **U16** as a white solid (784 mg, 1.88 mmol, 81%); ^{1}H-NMR (400 MHz, $CDCl_3$): δ 9.81 (s, 1H), 8.29 (s, 2H), 5.23 (s, 2H), 3.76 (s, 3H); ^{13}C-NMR (100 MHz, $CDCl_3$): δ 188.25, 161.77, 141.60 (2C), 135.57, 100.85, 92.15 (2C), 59.29.

*Synthesis of **U17***

To a solution of **U16** (652 mg, 1.56 mmol) and **U14** (1.01 g, 4.7 mmol) in a mixture of anhydrous THF (10 mL) and $(i\text{Pr})_2\text{NH}$ (15 mL), $PdCl_2(PPh_3)_2$ (56 mg, 0.08 mmol) and CuI (15 mg, 0,08 mmol) were added at room temperature under Ar atmosphere. After stirring for 19 h at room temperature, the reaction was quenched with saturated aqueous NH_4Cl, extracted with diethyl ether, and dried over anhydrous $MgSO_4$. The crude product was purified by silica gel column chromatography (Wako gel® C-200, 2.5% ethyl acetate/*n*-hexane) to provide **U17** as a brown solid (295 mg, 0.73 mmol, 46%); ^{1}H-NMR (400 MHz, $CDCl_3$): δ 9.84 (s, 1H), 7.94 (s, 2H), 5.49 (s, 2H), 3.66 (s, 3H), 0.23 (s, 18H); ^{13}C-NMR (100 MHz, $CDCl_3$): δ 189.11, 165.98, 136.70 (2C), 132.21, 117.43 (2C), 99.73, 93.45 (2C), 87.32 (2C), 80.08 (2C), 71.07 (2C), 58.19, -0.28 (6C).

*Synthesis of **U18***

To a solution of **U17** (295 mg, 0.73 mmol) in THF (10 mL), 6.0 M HCl (5 mL) was added at room temperature, and the mixture was stirred for 4 h at 50 ˚C. Then, the mixture was cooled to room temperature, extracted with diethyl ether, and dried over anhydrous $MgSO_4$. The crude product was purified by silica gel column chromatography (Wako gel® C-200, 5% ethyl acetate/*n*-hexane) to give **18** as a brown solid (210 mg, 0.58 mmol, 79%); ^{1}H-NMR (400 MHz, $CDCl_3$): δ 9.79 (s, 1H), 7.91 (s, 2H), 0.25 (s, 18H); ^{13}C-NMR (100 MHz, $CDCl_3$): δ 188.99, 164.57, 136.21 (2C), 130.04, 110.32 (2C), 94.27 (2C), 87.00 (2C), 81.57 (2C), 69.21 (2C), -0.30 (6C); ESI-MS (*m/z*): 361.2 $[M-H]^-$.

*Synthesis of **U19***

To a solution of **U4** (30 mg, 0.11 mmol) and cyanoacetic acid (12 mg, 0.14 mmol) in CH_2Cl_2 (8 mL), EDC (32 mg, 0.17 mmol) and DMAP (20 mg, 0.17 mmol) were added at room temperature, and the mixture was stirred for 11 h at room temperature. The reaction was quenched with water, extracted with CH_2Cl_2, and dried over anhydrous $MgSO_4$. The crude product was purified by silica gel column chromatography (Wako gel® C-200, 30% ethyl acetate/*n*-hexane) to give **U19** as an orange oil (27 mg, 0.081 mmol, 72%); ^{1}H-NMR (400 MHz, $CDCl_3$): δ 4.19 (t, 2H, J = 6.6 Hz), 3.98 (s, 3H), 3.97 (s, 3H), 3.46 (s, 2H), 2.45 (br t, 2H, J = 6.8 Hz), 2.00 (s, 3H), 1.69 (m, 2H), 1.43 – 1.45 (m, 4H); ^{13}C-NMR (100 MHz, $CDCl_3$): δ 184.76, 184.31, 163.13, 144.51 (2C), 142.58, 139.16, 113.19, 66.92, 61.35 (2C), 28.30, 28.22, 26.28, 26.04, 24.90, 12.14; ESI-MS (*m/z*); 336.2 $[M+H]^+$.

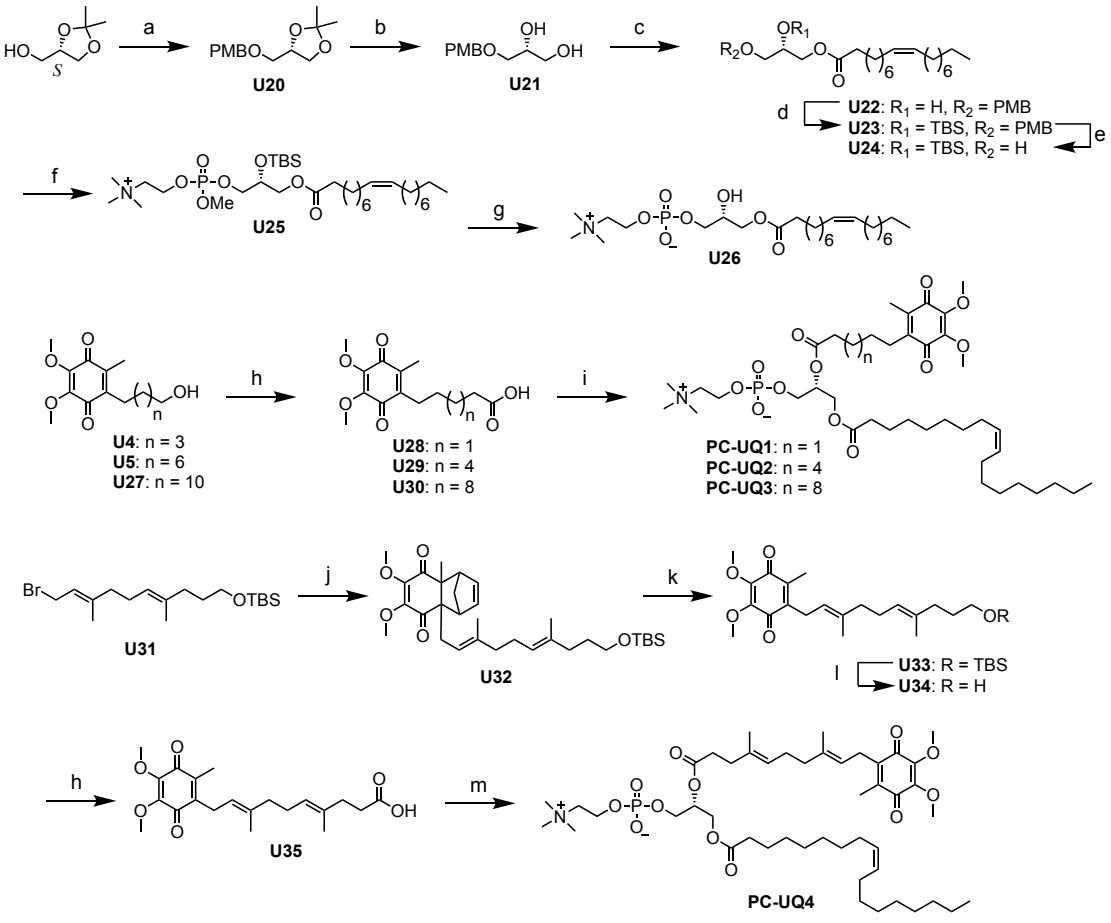
*Synthesis of **SF-UQ6***

To a solution of **U18** (33 mg, 0.090 mmol) and **U19** (27 mg, 0.081 mmol) in toluene (7 mL), piperidine (8 μL) and AcOH (5 μL) were added at room temperature. After stirring for 2 h at 70˚C, piperidine (8 μL) was added again to the mixture, which was stirred for a further 2 h at 70˚C. The mixture was cooled to room temperature, quenched with saturated aqueous NH_4Cl, extracted with ethyl acetate, and dried over anhydrous $MgSO_4$. The crude product was purified by silica gel column chromatography (Wako gel® C-200, 20% ethyl acetate/*n*-hexane) to give **SF-UQ6** as an orange oil (27 mg, 0.040 mmol, 49%); ^{1}H-NMR (400 MHz, $CDCl_3$): δ 8.04 (s, 2H), 8.01 (s, 1H), 4.31 (t, 2H, J = 6.6 Hz), 2.49 (t, 2H, J = 6.6 Hz), 2.03 (s, 3H), 1.79 (m, 2H), 1.50-1.46 (m, 4H), 0.25 (s, 18H); ^{13}C-NMR (100 MHz, $CDCl_3$): δ 184.82, 184.34, 163.52, 162.54, 152.07, 144.56 (2C), 142.70, 139.19, 137.31 (2C), 124.78, 115.28, 110.73 (2C), 102.46, 94.38 (2C), 86.99 (2C), 81.76 (2C), 68.99 (2C), 66.73, 61.36 (2C), 28.42, 28.39, 26.37, 26.13, 12.19, -0.35 (6C); ESI-MS (*m/z*) 678.2 $[M-H]^-$.

*Outline of the syntheses of **PC-UQ1**−**PC-UQ4***

The synthetic procedures of **PC-UQ1**, **PC-UQ2**, **PC-UQ3** and **PC-UQ4** are outlined in Scheme S3. (*S*)-(+)-2,2-dimethyl-1,3-dioxolane-4-methanol was protected with PMB group, followed by the removal of acetal, esterification of *sn*-1 position of **U21** with oleic acid to provide monoacylglycerol **U22**. The free hydroxy group at *sn*-2 position in **U22** was protected with TBS group, then the PMB group was removed in the presence of DDQ to give **U24**. **U25** was prepared by phosphoramidite methodology using choline tosylate, followed by the cleavage of the methyl and TBS groups to provide **U26**. The *lyso*-PC **U26** was treated with appropriate UQ intermediate (**U28**-**U30** and **U35** [*7*]) in the presence of 2-methyl-6-nitrobenzoic anhydride or 2,4,6-trichlorobenzoyl chloride to afford the target compounds.

Scheme S3[c]



[c]*Reagents and conditions*: (a) PMBCl, NaH, DMF, rt, 3 h, 84%; (b) AcOH/water (6:1), 50˚C, 2 h, 85%.; (c) oleic acid, DCC, DMAP, CH_2Cl_2, rt, 2 h, 57%; (d) TBSCl, imidazole, neat, rt, 16 h, quant.; (e) DDQ, CH_2Cl_2/water (9:1), 0 ˚C, 2 h, 77%; (f) i) $MeOP[N(iPr)_2]_2$, *1H*-tetrazole, MS4A, CH_2Cl_2, rt, 2 h; ii) choline tosylate, *1H*-tetrazole, rt, 2 h, iii) Bu_4NIO_4, 0˚C, 30 min, 77% (3 steps); (g) i) TMA, CH_2Cl_2, CH_3CN, *i*PrOH, rt, 12 h; ii) aq. HCl, THF, rt, 12 h, 84% (2 steps); (h) Jone's reagents, acetone, rt, 2 h, 64-92% (i) **U26**, MNBA, DMAP, rt, 3−20 h, 36−68%; (j) **U9**, *t*-BuOK, DMF/THF (1:3), 0˚C, 12 h, 53%; (k) toluene, reflux, 5 h, quant.; (l) THF/1.0 M aq. HCl (4:1), rt, 1 h, 90%; (m) **U26**, 2,4,6-trichlorobenzoyl chloride, DMAP, rt, 16 h, 21%.

*Synthesis of **U20***

To a suspension of NaH (2.64 g, 60% in mineral oil, 66 mmol) in anhydrous DMF (100 mL), (*S*)-(+)-2,2-dimethyl-1,3-dioxolane-4-methanol (8.0 g, 60.5 mmol) was added at 0˚C under N_2 atmosphere. After stirring for 30 min, *p*-methoxybenzyl chloride (10.3 g, 66 mmol) was added to the mixture, followed by the stirring for 3 h at room temperature. The reaction was quenched with water, extracted with 10% ethyl acetate/*n*-hexane, washed with brine and dried over anhydrous $MgSO_4$. The crude product was purified by silica gel column chromatography (Wako gel® C-200, 10−15% ethyl acetate/*n*-hexane) to provide **U20** as a colorless oil (12.8 g, 50.8 mmol, 84%); ^{1}H-NMR (400 MHz, $CDCl_3$): δ 7.26 (d, 2H, J = 8.7 Hz), 6.88 (d, 2H, J = 8.6 Hz), 4.52 (dd, 2H, J = 19.2, 11.7 Hz), 4.28 (tt, 1H, J = 12.0, 6.0 Hz), 4.05 (dd, 1H, J = 8.2, 6.4 Hz), 3.81 (s, 3H), 3.63 (dd, 1H, J = 8.2, 6.4 Hz), 3.53 (dd, 1H, J = 8.8, 5.7 Hz), 3.44 (dd, 1H, J = 9.8, 5.6 Hz), 1.42 (s, 3H), 1.36 (s, 3H); ^{13}C-NMR (100 MHz, $CDCl_3$): δ 159.30, 130.08, 129.38 (2C), 113.82 (2C), 109.38, 74.77, 73.18, 70.81, 66.96, 55.28, 26.79, 25.39.

*Synthesis of **U21***

U20 (12.8 mg, 50.8 mmol) was dissolved in a mixture of AcOH (150 mL) and water (25 mL), then the mixture was heated to 50˚C and stirred for 2 h. The solvent was removed under reduced pressure, and the crude product was purified by silica gel column chromatography (Wako gel® C-200, 40−100% ethyl acetate/*n*-hexane) to give **U21** as a white solid (11.1 g, 41.8 mmol, 82%); ^{1}H-NMR (400 MHz, $CDCl_3$): δ 7.25 (d, 2H, J = 8.8 Hz), 6.89 (d, 2H, J = 8.7 Hz), 4.47 (s, 2H), 3.88 (m, 1H), 3.80 (s, 3H), 3.69 (dd, 1H, J = 11.4, 3.9 Hz), 3.62 (dd, 1H, J = 11.4, 5.5 Hz), 3.55 (dd, 1H, J = 9.6, 4.4 Hz), 3.51 (dd, 1H, J = 9.6, 6.2 Hz), 2.72 (s,1H), 2.29 (s, 1H); ^{13}C-NMR (100 MHz, $CDCl_3$): δ 159.36, 129.78, 129.48 (2C), 113.80 (2C), 73.20, 71.43, 70.72, 64.05, 55.27.

*Synthesis of **U22***

To an ice-cooled solution of **U21** (2.00 g, 9.42 mmol) and oleic acid (1.27 g, 4.49 mmol) in anhydrous CH_2Cl_2 (50 mL), DCC (930 mg, 4.50 mmol) and DMAP (550 mg, 4.50 mmol) were added under N_2 atmosphere. After stirring for 2 h at room temperature, the reaction mixture was filtered, and the filtrate was concentrated under reduced pressure. The crude product was purified by silica gel column chromatography (Wako gel® C-200, 30% ethyl acetate/*n*-hexane) to give **U22** as a colorless oil (1.22 g, 2.56 mmol, 57%); ^{1}H-NMR (400 MHz, $CDCl_3$): δ 7.25 (d, 2H, J = 8.7 Hz), 6.88 (d, 2H, J = 8.7 Hz), 5.39-5.30 (m, 2H), 4.49 (s, 2H), 4.17 (dd, 1H, J = 4.5, 11.5 Hz), 4.12 (dd, 1H, J = 5.9, 11.6 Hz), 4.05-3.97 (m, 1H), 3.81 (s, 3H), 3.52 (dd, 1H, J = 4.3, 8.4 Hz), 3.46 (dd, 1H, J = 6.2, 9.6 Hz), 2.49 (d,1H, J = 4.8 Hz), 2.32 (t, 2H, J = 7.6 Hz), 2.05-1.98 (m, 4H), 1.63-1.59 (m, 2H), 1.38-1.22 (m, 20H), 0.88 (t, 3H, J = 6.9 Hz); ^{13}C-NMR (100 MHz, $CDCl_3$): δ 174.14, 159.62, 130.23, 129.97, 129.95, 129.64 (2C), 113.98 (2C), 73.40, 70.78, 69.16, 65.60, 55.49, 34.36, 32.12, 29.98, 29.91, 29.74, 29.53 (2C), 29.38, 29.32 (2C), 27.44, 27.38, 25.12, 22.90, 14.33.

Synthesis of ***U23***

U22 (190 mg, 0.40 mmol) was stirred with imidazole (52 mg, 0.76 mmol) and TBSCl (226 mg, 0.57 mmol) for 16 h at room temperature under solvent-free (neat) conditions. The mixture was purified by silica gel column chromatography (Wako gel® C-200, 5% ethyl acetate/*n*-hexane) to provide **U23** as a colorless oil (237 mg, 0.40 mmol, quant.); ^{1}H-NMR (400 MHz, $CDCl_3$): δ 7.24 (d, 2H, J = 8.7 Hz), 6.87 (d, 2H, J = 8.7 Hz), 5.34 (m, 2H), 4.46 (s, 2H), 4.18 (m, 2H), 4.01 (m, 2H), 3.80 (s, 3H), 3.42 (d, 2H, J = 5.3 Hz), 2.28 (t, 2H, J = 7.6 Hz), 2.01 (m, 4H), 1.59 (m, 2H), 1.38-1.20 (m, 20H), 0.88 (t, 3H, J = 6.4 Hz), 0.87 (s, 9H), 0.07 (s, 3H), 0.06 (s, 3H); ^{13}C-NMR (100 MHz, $CDCl_3$): δ 173.89, 159.42, 130.45, 130.22, 129.98, 129.45(2C), 113.98(2C), 73.33, 71.76, 69.95, 66.40, 55.49, 34.49, 32.12, 29.99, 29.93, 29.75, 29.54 (2C), 29.41, 29.36, 29.33, 27.44, 27.40, 25.96 (3C), 25.14, 22.90, 18.34, 14.33, -4.48, -4.59; ESI-MS (*m/z*); 591.4 $[M+H]^+$.

Synthesis of ***U24***

To a solution of **U23** (237 mg, 0.40 mmol) in a mixture of CH_2Cl_2 (4.5 mL) and water (0.5 mL), DDQ (182 mg, 0.80 mmol) was added at 0˚C, and the mixture was stirred for 2 h at 0˚C. The reaction was quenched with saturated aqueous $NaHCO_3$, extracted with CH_2Cl_2, washed with brine, and dried over anhydrous $MgSO_4$. The crude product was purified by silica gel column chromatography (Wako gel® C-200, 5−15% ethyl acetate/*n*-hexane) to give **U24** as a colorless oil (182 mg, 0.31 mmol, 77%); ^{1}H-NMR (400 MHz, $CDCl_3$): δ 5.40-5.25 (m, 2H), 4.13-4.04 (m, 2H), 3.96-3.88 (m, 1H), 3.63-3.50 (br m, 2H), 2.31 (t, 2H, J = 7.6 Hz), 2.05-1.93 (m, 4H), 1.65-1.55 (m, 2H), 1.45-1.20 (m, 20H), 0.90 (s, 9H), 0.88 (t, 3H, J = 7.0 Hz), 0.11 (s, 6H); ^{13}C-NMR (100 MHz, $CDCl_3$): δ 173.95, 130.24, 129.96, 70.81, 65.02, 64.13, 34.43, 32.12, 29.99, 29.91, 29.74, 29.54 (2C), 29.38, 29.34, 29.31, 27.44, 27.39, 25.95 (3C), 25.11, 22.90, 18.28, 14.33, -4.43, -4.61; ESI-MS (*m/z*); 471.4 $[M+H]^+$.

Synthesis of ***U25***

To a solution of **U24** (170 mg, 0.36 mmol) in anhydrous CH_2Cl_2 (7 mL), activated MS4A (500 mg) was added, and the mixture was stirred for 30 min at room temperature under Ar atmosphere. Methyl *N,N,N′,N′*-tetraisopropylphosphordiamidite (132 mg, 0.50 mmol) and *1H*-tetrazole (13 mg, 0.18 mmol) were added to the mixture at room temperature. After stirring for 2 h at room temperature, choline tosylate (303 mg, 1.1 mmol) and *1H*-tetrazole (77 mg, 1.1 mmol) were added to the mixture, followed by the stirring for 2 h at room temperature. The reaction mixture was cooled to 0˚C, then Bu_4NIO_4 (238 mg, 0.55 mmol) was added in one portion, and the mixture was stirred for 30 min at room temperature. The reaction mixture was filtered through Celite, and the filtrate was washed with 10% (w/v) aqueous $Na_2S_2O_3$ and saturated aqueous $NaHCO_3$. The organic layer was dried over anhydrous $MgSO_4$, concentrated under reduced pressure, and the crude product was purified by silica gel column chromatography (Wako gel® C-200, 10% methanol/$CHCl_3$) to provide **U25** as a

colorless oil (220 mg, 0.267 mmol, 74%); ^{1}H-NMR (400 MHz, $CDCl_3$:MeOD=1:1): δ 5.36-5.32 (m, 2H), 4.58-4.50 (m, 2H), 4.24-4.18 (m, 1H), 4.15-4.02 (m, 4H), 3.86 (dd, 3H, J = 0.8, 11.3 Hz), 3.79-3.76 (m, 2H), 3.24 (s, 9H), 2.36 (t, 2H, J = 7.6 Hz), 2.10-1.95 (m, 4H), 1.68-1.58 (m, 2H), 1.40-1.20 (m, 20H), 0.92 (s, 9H), 0.89 (t, 3H, J = 7.0 Hz), 0.14 (s, 6H); ^{13}C-NMR (100 MHz, $CDCl_3$:MeOD=1:1): δ 174.91, 130.77, 130.50, 70.07, 66.49, 65.60, 62.31, 58.21, 55.83, 54.61 (3C), 34.91, 32.77, 30.58, 30.52, 30.34, 30.17, 30.12, 30.01, 29.94, 29.90, 27.97, 27.93, 26.20 (3C), 25.72, 23.49, 18.78, 14.52, -4.41 (2C); ESI-MS (m/z) 651.5 $[M+H]^+$.

*Synthesis of **U26***

To a solution of **U25** (220 mg, 0.267 mmol) in a mixture of CH_2Cl_2 (2 mL), CH_3CN (3 mL), and *i*PrOH (3 mL), trimethylamine (4.5 mL, 45% (w/v) solution in water) was added at room temperature. After stirring for 12 h at room temperature, the solvent was removed under reduced pressure. The crude product was dissolved again in a mixture of THF (6 mL) and water (3 mL), then 1.0 M aqueous HCl (250 µL) was added to the solution, and the stirring was continued for a further 12 h at room temperature. The reaction was quenched with 25% (w/v) aqueous NH_3 at 0˚C, extracted with 10% MeOH/$CHCl_3$ and dried over anhydrous Na_2SO_4. The crude product was purified by silica gel column chromatography (Wako gel® C-200, 2:8:0.2-5:5:0.2 methanol/$CHCl_3$/water) to give **U26** as a colorless oil (118 mg, 0.224 mmol, 84%); ^{1}H-NMR (400 MHz, $CDCl_3$: MeOD=1:1): δ 5.38-5.29 (m, 2H), 4.32-4.26 (m, 2H), 4.18 (dd, 1H, J = 4.7, 11.3 Hz), 4.12 (dd, 1H, J = 6.1, 11.3 Hz), 4.02-3.85 (m, 3H), 3.65-3.62 (m, 2H), 3.23 (s, 9H), 2.36 (t, 2H J = 7.6 Hz), 2.05-1.95 (m, 4H), 1.65-1.59 (m, 2H), 1.38-1.22 (m, 20H), 0.89 (t, 3H, J = 6.9 Hz); ^{13}C-NMR (100 MHz, $CDCl_3$:MeOD=1:1): δ 175.21, 130.72, 130.51, 69.55, 67.65, 67.25, 65.93, 60.02, 54.68 (3C), 34.82, 32.75, 30.56, 30.53, 30.32, 30.14, 30.08, 30.03, 29.95, 29.93, 27.94, 27.93, 25.70, 23.46, 14.51; ESI-MS (*m/z*); 522.3 $[M+H]^+$.

*Synthesis of **U27***

U27 was synthesized in 5 steps according to the procedures described in refs. *1* and *2* using commercially available 1,12-undecanediol as a starting material; ^{1}H-NMR (400 MHz, $CDCl_3$): δ 3.99 (s, 3H), 3.99 (s, 3H), 2.45 (br t, 2H, J = 7.4 Hz), 2.35 (t, 2H, J = 7.5 Hz), 2.01 (s, 3H), 1.67-1.59 (m, 2H), 1.45-1.25 (m, 16H); ^{13}C-NMR (100 MHz, $CDCl_3$): δ 184.91, 184.38, 179.09, 144.51 (2C), 143.32, 138.89, 61.36 (2C), 34.06, 30.23, 29.70, 29.65, 29.58, 29.55, 29.40, 29.24, 28.94, 26.61, 24.89, 12.12.

*Synthesis of **U28***

To an ice-cooled solution of **U4** (30 mg, 0.11 mmol) in acetone (5 mL), Jone's reagent (0.13 mmol) was added, and the mixture was stirred for 2 h at room temperature. The reaction was quenched with water, extracted with diethyl ether, and dried over anhydrous $MgSO_4$. The crude product was purified by silica gel column chromatography (Wako gel® C-200, 60% ethyl acetate/*n*-hexane) to provide **U28** as an orange oil (20 mg, 0.071

mmol, 64%); ^{1}H-NMR (400 MHz, $CDCl_3$): δ 3.99 (s, 3H), 3.99 (s, 3H), 2.49 (br t, 2H, J = 7.8 Hz), 2.39 (t, 2H, J = 7.4 Hz), 2.02 (s, 3H), 1.75-1.65 (m, 2H), 1.52-1.42 (m, 2H); ^{13}C-NMR (100 MHz, $CDCl_3$): δ 184.78, 184.28, 179.17, 144.54 (2C), 142.47, 139.29, 61.37 (2C), 33.78, 28.18, 26.17, 24.85, 12.16.

*Synthesis of **U29***

U29 was prepared from **U5** according to the same procedure described for **U28.** The crude product was purified by silica gel column chromatography (Wako gel® C-200, 50% ethyl acetate/*n*-hexane) to provide **U29** as an orange oil (29 mg, 0.089 mmol, 92%); ^{1}H-NMR (400 MHz, $CDCl_3$): δ 3.99 (s, 3H), 3.99 (s, 3H), 2.45 (br t, 2H, J = 7.2 Hz), 2.35 (t, 2H, J = 7.5 Hz), 2.01 (s, 3H), 1.70-1.60 (m, 2H), 1.45-1.25 (m, 2H); ^{13}C-NMR (100 MHz, $CDCl_3$): δ 184.91, 184.37, 179.22, 144.54 (2C), 143.17, 138.94, 61.36 (2C), 34.02, 29.78, 29.16, 29.09, 28.83, 26.55, 24.82, 12.13.

*Synthesis of **U30***

U30 was prepared from **U27** according to the same procedure described for **U28.** The crude product was purified by silica gel column chromatography (Wako gel® C-200, 30−60% ethyl acetate/*n*-hexane) to provide **U30** as an orange oil (63 mg, 0.17 mmol, 75%); ^{1}H-NMR (400 MHz, $CDCl_3$): δ 3.99 (s, 3H), 3.99 (s, 3H), 2.45 (t, 2H, J = 7.4 Hz), 2.35 (t, 2H, J = 7.5 Hz), 2.01 (s, 3H), 1.67-1.59 (m, 2H), 1.45-1.25 (m, 16H); ^{13}C-NMR (100 MHz, $CDCl_3$): δ 184.91, 184.38, 179.09, 144.51 (2C), 143.32, 138.89, 61.36 (2C), 34.06, 30.23, 29.70, 29.65, 29.58, 29.55, 29.40, 29.24, 28.94, 26.61, 24.89, 12.12.

*Synthesis of **PC-UQ1***

To a solution of **U26** (21 mg, 0.040 mmol) and **U28** (19 mg, 0.067 mmol) in anhydrous $CHCl_3$ (5 mL, containing amylene as stabilizer), MNBA (40 mg, 0.12 mmol) and DMAP (25 mg, 0.20 mmol) were added at room temperature under N_2 atmosphere, and the mixture was stirred for 11 h at room temperature. The reaction was quenched with water, extracted with $CHCl_3$, and dried over Na_2SO_4. The crude product was purified by silica gel column chromatography (Wako gel® C-200, 5% methanol/$CHCl_3$ to 2:8:0.2 methanol/$CHCl_3$/H_2O) to give **PC-UQ1** as a yellow oil (14 mg, 0.018 mmol, 45%); ^{1}H-NMR (400 MHz, $CDCl_3$:MeOD = 1:1): δ 5.38-5.29 (m, 2H), 5.27-5.20 (m, 1H), 4.43 (dd, 1H, J = 3.3, 12.0 Hz), 4.30-4.23 (m, 2H), 4.17 (dd, 1H, J = 6.7, 12.0 Hz), 4.04-3.98 (m, 2H), 4.00 (s, 3H), 3.99 (s, 3H), 3.65-3.62 (m, 2H), 3.24 (s, 9H), 2.50 (br t, 2H, J = 7.8 Hz), 2.40 (t, 2H, J = 7.4 Hz), 2.32 (t, 2H, J = 7.6 Hz), 2.04 (s, 3H), 2.04-1.95 (m, 4H), 1.73-1.64 (m, 2H), 1.64-1.55 (m, 2H), 1.51-1.42 (m, 2H), 1.38-1.20 (m, 20H), 0.89 (t, 3H, J = 6.9 Hz); ^{13}C-NMR (100 MHz, $CDCl_3$:MeOD = 1:1): δ 185.52, 185.04, 174.67, 173.84, 145.17 (2C), 143.17, 140.01, 130.64, 130.37, 71.43, 67.15, 64.38, 63.30, 61.67 (2C), 59.82, 54.67 (3C), 34.69, 34.43, 32.60, 30.43, 30.40, 30.19, 30.00, 29.96, 29.89, 29.80, 29.78, 28.71, 27.84, 27.82, 26.61, 25.55, 25.52, 23.33, 14.47, 12.31; ESI-MS (*m/z*); 786.5 $[M+H]^+$.

*Synthesis of **PC-UQ2***

PC-UQ2 was prepared from **U26** and **U29** according to the same procedure described for **PC-UQ1**. The crude product was purified by silica gel chromatography (Wako gel® C-200, 20% methanol/$CHCl_3$ to 2:8:0.1 methanol/$CHCl_3$/H_2O) to give **PC-UQ2** as a yellow oil (33 mg, 0.040 mmol, 68%); ^{1}H-NMR (400 MHz, $CDCl_3$): δ 5.40-5.30 (m, 2H), 5.28-5.20 (m, 1H), 4.43 (dd, 1H, J = 3.0, 12.0 Hz), 4.32-4.22 (br m, 2H), 4.17 (dd, 1H, J = 6.7, 11.8 Hz), 4.05-3.99 (m, 2H), 4.00 (s, 3H), 3.99 (s, 3H), 3.65-3.60 (m, 2H), 3.24 (s, 9H), 2.47 (br t, 2H, J = 7.1 Hz), 2.38-2.30 (m, 4H), 2.05-1.95 (m, 4H), 2.04 (s, 3H), 1.70-1.55 (m, 4H), 1.45-1.20 (m, 28H), 0.89 (t, 3H, J = 6.7 Hz); ^{13}C-NMR (100 MHz, $CDCl_3$): δ 185.59, 185.05, 174.62, 174.18, 145.06(2C), 143.76, 139.61, 130.61, 130.31, 71.11, 67.13, 64.27, 63.31, 61.64 (2C), 59.73, 54.65 (3C), 34.77, 34.68, 32.54, 30.37 (2C), 30.27, 30.13, 29.94, 29.91, 29.84, 29.75 (2C), 29.66, 29.59, 29.30, 27.77 (2C), 26.94, 25.50, 25.47, 23.28, 14.46, 12.27; ESI-MS (*m/z*): 828.5 $[M+H]^+$.

*Synthesis of **PC-UQ3***

PC-UQ3 was prepared from **U26** and **U30** according to the same procedure described for **PC-UQ1**. The crude product was purified by silica gel column chromatography (Wako gel® C-200, 20% methanol/$CHCl_3$ to 2:8:0.2 methanol/$CHCl_3$/H_2O) to give **PC-UQ3** as a yellow oil (12 mg, 0.014 mmol, 36%); ^{1}H-NMR (400 MHz, $CDCl_3$:MeOD = 1:1): δ 5.38-5.32 (m, 2H), 5.27-5.22 (m, 1H), 4.43 (dd, 1H, J = 3.0, 12.0 Hz), 4.32-4.22 (m, 2H), 4.17 (dd, 1H, J = 6.9, 12.0 Hz), 4.01-3.98 (m, 2H), 3.99 (s, 3H), 3.99 (s, 3H), 3.65-3.58 (m, 2H), 3.23 (s, 9H), 2.47 (br t, 2H, J = 7.4 Hz), 2.34 (t, 2H, J = 7.2 Hz), 2.32 (t, 2H, J = 7.4 Hz), 2.04-1.95 (m, 4H), 2.02 (s, 3H), 1.65-1.55 (m, 4H), 1.45-1.22 (m, 36H), 0.89 (t, 3H, J = 6.8 Hz); ^{13}C-NMR (100 MHz, $CDCl_3$:MeOD = 1:1): δ 185.64, 185.10, 174.63, 174.28, 145.07 (2C), 143.94, 139.57, 130.62, 130.33, 71.15, 67.19, 64.35, 63.35, 61.64 (2C), 59.75, 54.65 (3C), 34.86, 34.70, 32.56, 30.48, 30.39, 30.37, 30.24 (2C), 30.15 (3C), 30.03, 29.96, 29.93, 29.86, 29.76 (3C), 29.38, 27.81, 27.79, 27.00, 25.57, 25.52, 23.29, 14.46, 12.25; ESI-MS (*m/z*) 884.6 $[M+H]^+$.

*Synthesis of **U31***

U31 was synthesized in 7 steps according to the procedures described in ref. *7* using commercially available *trans,trans*-farnesyl acetate as a starting material. ^{1}H-NMR (400 MHz, $CDCl_3$): δ 5.53 (m, 1H), 5.09 (m, 1H), 4.02 (d, 2H, J = 8.4 Hz), 3.58 (t, 2H, J = 6.6 Hz), 2.13-2.04 (m, 4H), 2.00 (br t, 2H, J = 7.7 Hz), 1.73 (s, 3H), 1.64-1.57 (m, 2H) 1.59 (s, 3H), 0.90 (s, 9H), 0.05 (s, 6H); ^{13}C-NMR (100 MHz, $CDCl_3$): δ 143.80, 135.60, 123.63, 120.78, 63.11, 39.73, 35.98, 31.38, 29.87, 26.30, 26.19 (3C), 18.57, 16.25, 16.19, -5.03 (2C).

*Synthesis of **U32***

To a solution of **U31** (390 mg, 1.04 mmol) and **U9** (236 mg, 0.95 mmol) in a mixture of anhydrous THF (3

mL) and DMF (9 mL), potassium *tert*-butoxide (180 mg, 1.60 mmol) was slowly added at -30˚C under N_2 atmosphere. The mixture was allowed to warm up to 0˚C, and stirred for 16 h. The reaction was quenched with saturated aqueous NH_4Cl, extracted with diethyl ether, and dried over anhydrous $MgSO_4$. The crude product was purified by silica gel column chromatography (Wako gel® C-200, 5% ethyl acetate/*n*-hexane) to give **U32** as a slightly yellow oil (340 mg, 0.626 mmol, 66%); ^{1}H-NMR (400 MHz, $CDCl_3$): δ 6.08-6.03 (m, 2H), 5.13-5.05 (m, 2H), 3.91 (s, 3H), 3.88 (s, 3H), 3.57 (t, 2H, J = 6.6 Hz), 3.09 (m, 1H), 3.01 m, 1H), 2.76 (dd, 1H, J = 7.4, 15.1 Hz), 2.43 (dd, 1H, J = 6.4, 15.1 Hz), 2.07-1.93 (m, 6H), 1.79 (br d, 1H, J = 9.4 Hz), 1.63-1.55 (m, 8H), 1.50 (s, 3H), 1.46 (td, 1H, J = 1.5, 9.5 Hz), 0.89 (s, 9H), 0.03 (s, 6H); ^{13}C-NMR (100 MHz, $CDCl_3$): δ 199.09, 198.47, 151.04, 149.37, 138.26, 138.20, 137.41, 135.31, 124.03, 119.86, 63.12, 60.51, 60.27, 59.56, 56.29, 54.68, 53.36, 43.70, 40.17, 36.00, 31.41, 26.74, 26.19 (3C), 23.61, 18.57, 16.59, 16.20, -5.04 (2C).

*Synthesis of **U33***

U33 was prepared from **U32** according to the same procedure described for **U12**. The crude product was purified by silica gel column chromatography (Wako gel® C-200, 5% ethyl acetate/*n*-hexane) to provide **U33** as an orange oil (295 mg, 0.624 mmol, quant.); ^{1}H-NMR (400 MHz, $CDCl_3$): δ 5.07 (br t, 1H, J = 6.4 Hz), 4.94 (br t, 1H, J = 7.0 Hz), 4.00 (s, 3H), 3.98 (s, 3H), 3.57 (t, 2H, J = 6.6 Hz), 3.18 (d, 2H, J = 7.0 H)), 2.08-1.95 (m, 6H), 2.01 (s, 3H), 1.74 (s, 3H), 1.62-1.55 (m, 2H), 1.57 (s, 3H), 0.89 (s, 9H), 0.04 (s, 6H); ^{13}C-NMR (100 MHz, $CDCl_3$): δ 184.99, 184.14, 144.61, 144.47, 141.91, 139.09, 137.84, 135.16, 124.13, 119.08, 63.09, 61.36 (2C), 39.91, 35.99, 31.40, 26.74, 26.19 (3C), 25.52, 18.56, 16.56, 16.20, 12.15, -5.05 (2C).

*Synthesis of **U34***

To a solution of **U33** (252 mg, 0.529 mmol) in THF (4 mL), 1.0 M HCl aq. (1 mL) was added at room temperature. After stirring for 1 h at room temperature, the reaction was quenched with saturated aqueous $NaHCO_3$, extracted with ethyl acetate, and dried over anhydrous $MgSO_4$. The crude product was purified by silica gel column chromatography (Wako gel® C-200, 40% ethyl acetate/*n*-hexane) to give **U34** as an orange oil (189 mg, 0.47 mmol, 94%); ^{1}H-NMR (400 MHz, $CDCl_3$): δ 5.08 (br t, 1H, J = 6.8 Hz), 4.92 (br t, 1H, J = 6.9 Hz), 4.00 (s, 3H), 3.98 (s, 3H), 3.60 (t, 2H, J = 6.5 Hz), 3.18 (d, 2H, J = 7.0 Hz), 2.10-1.98 (m, 6H), 2.01 (s, 3H), 1.73 (s, 3H), 1.68-1.59 (m, 2H), 1.59 (s, 3H); ^{13}C-NMR (100 MHz, $CDCl_3$): δ 185.01, 184.16, 144.57, 144.45, 141.88, 139.11, 137.48, 135.09, 124.50, 119.35, 62.91, 61.36 (2C), 39.74, 36.10, 30.90, 26.38, 25.51, 16.42, 16.10, 12.14; ESI-MS (*m/z*): 363.2 $[M+H]^+$.

*Synthesis of **U35***

U35 was prepared from **U34** according to the same procedure described for **U28**. The crude product was purified by silica gel column chromatography (Wako gel® C-200, 30% ethyl acetate/*n*-hexane) to give **U35** as an

orange oil (36 mg, 0.096 mmol, 69%); ^{1}H-NMR (400 MHz, $CDCl_3$): δ 5.09 (br t, 1H, J = 6.9 Hz), 4.90 (br t, 1H, J = 7.0 Hz), 3.99 (s, 3H), 3.98 (s, 3H), 3.18 (d, 2H, J = 6.9 Hz), 2.42 (t, 2H, J = 7.7 Hz), 2.27 (t, 2H, J = 7.7 Hz), 2.10-2.03 (m, 2H), 2.02-1.95 (m, 2H), 2.00 (s, 3H), 1.72 (d, 3H, J = 0.88 Hz), 1.59 (s, 3H); ^{13}C-NMR (100 MHz, $CDCl_3$): δ 184.95, 184.45, 177.72, 144.58 (2C), 141.87, 139.26, 137.31, 133.42, 125.07, 119.46, 61.38 (2C), 39.64, 34.48, 32.82, 26.37, 25.57, 16.40, 16.17, 12.14; ESI-MS (*m/z*); 375.2 $[M-H]^-$, 377.3$[M+H]^+$.

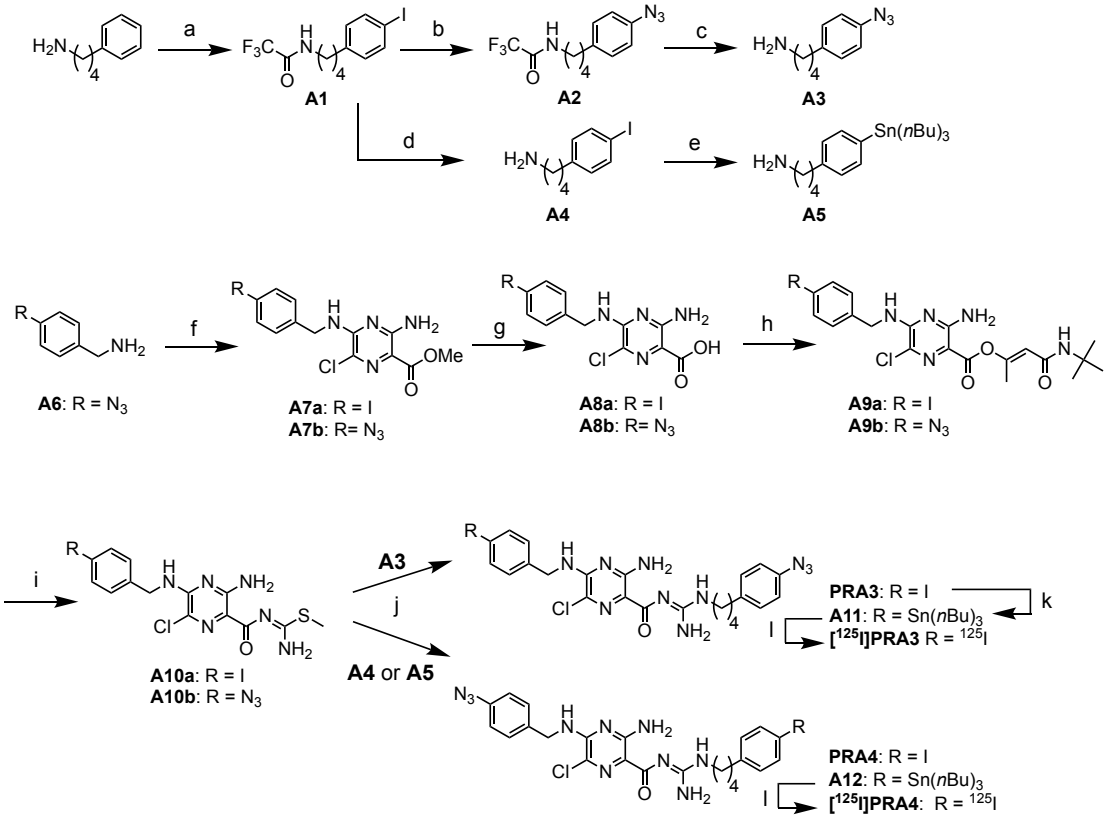
Synthesis of ***PC-UQ4***

PC-UQ4 was prepared from **U26** and **U35** according to the same procedure described for **PC-UQ1**, with the exception that 2,4,6-trichlorobenzoylchloride was used for the esterification. The crude product was purified by silica gel column chromatography (Wako gel® C-200, 20% methanol/$CHCl_3$ to 2:8:0.2 methanol/$CHCl_3$/H_2O) to give **PC-UQ4** as a yellow oil (26 mg, 0.030 mmol, 21%); ^{1}H-NMR (400 MHz, $CDCl_3$:MeOD = 1:1): δ 5.40-5.29 (m, 2H), 5.25-5.19 (m, 1H), 5.11 (br t, 1H, J = 7.0 Hz), 4.95 (br t, 1H, J = 6.9 Hz), 4.40 (dd, 1H, J = 3.3, 12.0 Hz), 4.35-4.25 (m, 2H), 4.18 (dd, 1H, J = 6.6, 12.0 Hz), 4.03 (t, 2H, J = 5.9 Hz), 3.99 (s, 3H), 3.99 (s, 3H), 3.65-3.62 (m, 2H), 3.24 (s, 9H), 3.21 (t, 2H, J = 7.1 Hz), 2.45-2.39 (m, 2H), 2.33 (t, 2H, J = 7.5 Hz), 2.30-2.24 (m, 2H), 2.10-1.95 (m, 8H), 2.03 (s, 3H), 1.75 (s, 3H), 1.65-1.45 (m, 2H), 1.61 (s, 3H), 1.38-1.22 (m, 20H), 0.89 (t, 3H, J = 6.9 Hz); ^{13}C-NMR (100 MHz, $CDCl_3$:MeOD = 1:1): δ 185.75, 184.94, 174.70, 173.87, 145.21, 145.13, 142.57, 139.83, 138.07, 134.07, 130.66, 130.38, 125.69, 119.91, 71.22, 67.13, 64.57, 63.28 (2C), 61.68, 60.04, 54.66 (3C), 40.21, 35.24, 34.71, 33.80, 32.63, 30.45, 30.43, 30.21, 30.03, 29.99, 29.92, 29.82 (2C), 27.86, 27.83, 27.15, 25.95, 25.58, 23.35, 16.70, 16.30, 14.48, 12.32; ESI-MS (*m/z*): 880.7 $[M+H]^+$.

*Outline of the syntheses of **PRA3, PRA4, [^{125}I]PRA3, and [^{125}I]PRA4***

PRA3 and **PRA4** were synthesized according to the previously methods [*8-10*], as summarized in Scheme S4. The amine derivatives **A3**, **A4**, and **A5** were prepared from commercially available 4-phenylbutylamine. They were, in the presence of DIPEA, coupled with methylthiopseudourea **A10a** or **A10b**, which were in parallel synthesized from commercially available methyl 3-amino-5,6-dichloro-2-pyrazinecarboxylate in 4 steps, to provide **PRA3** and **PRA4**, respectively. **[^{125}I]PRA3** and **[^{125}I]PRA4** were prepared by the catalysis of chloramine T [*7*] using tin-precursor **A11** and **A12**, respectively.

Scheme S4[d]



[d]*Reagents and conditions*: (a) i) I_2, $NaIO_3$, conc. H_2SO_4, AcOH, 70˚C, 1.5 h; ii)TFAA, CH_2Cl_2, rt, 2 h, 31% (2 steps); (b) *trans-N,N'*-dimetylcyclohexane-1,2-diamine, CuI, *L*-ascorbic acid Na, EtOH/water, reflux, 1 h, 80%; (c) K_2CO_3, MeOH, reflux, 2 h, 77%; (d) aq. NaOH, MeOH, rt, overnight, 85%; (e) $(Sn(nBu)_3)_2$, $Pd(PPh_3)_4$, dioxane, reflux, overnight, 36%; (f) methyl 3-amino-5,6-dichloro-2-pyrazinecarboxylate, 2-propanol, reflux, 4 h, 98 and 95% (for **A7a** and **A7b**); (g) aq. NaOH, MeOH, reflux, 1 h, 84 and 77% (for **A8a** and **A8b**); (h) 5-methylisoxazole, *t*-BuOH, TfOH, DMF, rt, 1 h, 70 and 62% (for **A9a** and **A9b**); (i) *S*-methylisothiourea sulfate, aq. NaOH, CH_2Cl_2/THF, rt, overnight, 72 and 66% (for **A9a** and **A9b**) ; (j) DIPEA, THF, 65˚C, overnight, 56, 82, and 47% (for **PRA3**, **PRA4**, and **A12**); (k) $(Sn(nBu)_3)_2$, $Pd(PPh_3)_4$, dioxane, 50˚C, 16 h, 9%; (l) [^{125}I]NaI, chloramine T, K_2HPO_4 aq., rt, 10 min, 63 and 39 % (for **[^{125}I]PRA3** and **[^{125}I]PRA4**).

*Synthesis of **A1***

To a solution of 4-phenylbutylamine (8.0 g, 5.4 mmol) in a mixture of acetic acid (150 mL) and sulfuric acid (concentrated, 15 mL), iodine (7.5 g, 59 mmol) and $NaIO_3$ (3.2 g, 16 mmol) were added in portions, and the mixture was stirred for 1.5 h at 70˚C. The mixture was concentrated under reduced pressure, and the residue was resuspended in water, washed with diethyl ether and dichloromethane. The water layer was basified with aqueous NaOH, then extracted with ethyl acetate. The organic layer was dried over anhydrous $MgSO_4$ to give crude 4-(4-iodophenyl)butylamine as a white solid.

To the solution of 4-(4-iodophenyl)butylamine in dichloromethane (150 mL) was added trifluoroacetic anhydride (9.1 g, 43 mmol). The mixture was stirred for 2 h at room temperature, and then it was diluted by dichloromethane and washed with saturated aqueous $NaHCO_3$. The organic layer was dried over $MgSO_4$. The crude product was purified by silica gel column chromatography (Wako gel® C-200, toluene) to give **A1** as a white solid (6.2 g, 17 mmol, 31 %, 2 steps); ^{1}H-NMR (400 MHz/$CDCl_3$): δ 7.60 (dt, J = 8.3, 1.8 Hz, 2H), 6.92 (dt, J = 8.3, 1.8 Hz, 2H), 6.30 (br s, 1H), 3.37 (q, J = 6.5 Hz, 2H), 2.59 (t, J = 7.1 Hz, 2H), 1.68-1.55 (m, 4H); ^{13}C-NMR (100 MHz/$CDCl_3$): δ 157.26, 141.39, 137.68 (2C), 130.68 (2C), 114.60, 91.23, 39.91, 34.95, 28.61, 28.32; ESI-MS (*m/z*): 372.1 $[M + H]^+$.

*Synthesis of **A2***

To a solution of **A1** (700 mg, 1.89 mmol) in EtOH (10 mL) and water (2 mL), *trans-N,N'*-dimethylcyclohecane-1,2-diamine (538 mg, 3.78 mmol), NaN_3 (246 mg, 3.78 mmol), CuI (72 mg, 0.38 mmol), and sodium *L*-ascorbate (75 mg, 0.38 mmol) were added at room temperature. The mixture was heated under reflux for 1 h, then quenched with water and extracted with ethyl acetate. The organic layer was dried over anhydrous $MgSO_4$ and purified by silica gel column chromatography (Wako gel® C-200, 20% ethyl acetate/*n*-hexane) to give **A2** as a yellow solid (431 mg, 1.50 mmol, 80%); ^{1}H-NMR (400 MHz/$CDCl_3$): δ 7.14 (dt, J = 8.5, 2.1 Hz, 2H), 6.95 (dt, J = 8.5, 2.1 Hz, 2H), 6.40 (br s, 1H), 3.37 (q, J = 6.5 Hz, 2H), 2.62 (t, J = 7.1 Hz, 2H), 1.69-1.57 (m, 4H); ^{13}C-NMR (100 MHz/$CDCl_3$): δ 157.27, 138.59, 137.98, 129.89 (2C), 119.24 (2C), 117.47, 114.61, 39.94, 34.81, 28.60, 28.54; ESI-MS (*m/z*): 285.1 $[M+H]^+$.

*Synthesis of **A3***

To a solution of **A2** (73 mg, 0.26 mmol) in MeOH (15 mL), K_2CO_3 (108 mg, 0.781 mmol dissolved in 0.10 mL water) were added, and the mixture was heated under reflux for 2 h. The mixture was concentrated under reduced pressure, and the residue was resuspended in ethyl acetate and extracted with 1.0 M aqueous HCl. The aqueous layer was basified again with 1.0 M aqueous NaOH, and the amine **4** in free form extracted with ethyl acetate. The organic layer was dried over anhydrous $MgSO_4$ and concentrated under reduced pressure to give **A3** as a yellow oil (38 mg, 0.20 mmol, 77%); ^{1}H-NMR (500 MHz/$CDCl_3$): δ 7.16 (d, J = 8.2 Hz, 2H), 6.94 (d, J

= 8.2 Hz, 2H), 2.71 (t, *J* = 6.9 Hz, 2H), 2.60 (t, *J* = 7.6 Hz, 2H), 1.66-1.60 (m, 2H), 1.50-1.44 (m, 2H); ^{13}C-NMR (125 MHz/$CDCl_3$): δ 139.29, 137.46, 129.72 (2C), 118.91 (2C), 41.99, 35.11, 33.19, 28.73; ESI-MS (m/z): 191.1 $[M+H]^+$.

*Synthesis of **A4***

To a stirring solution of **A1** (2.0 g, 5.4 mmol) in MeOH (40 mL), 1.0 M aqueous NaOH (9.6 mL) was added, and the mixture was stirred for 16 h at room temperature. The mixture was concentrated under reduced pressure, and resuspended in ethyl acetate, extracted with 1.0 M aqueous HCl. The aqueous layer was basified with aqueous NaOH, extracted with ethyl acetate. The organic layer was dried over anhydrous $MgSO_4$ to give **A4** as a white solid (1.26 g, 4.58 mmol, 85%); ^{1}H-NMR (400 MHz/MeOD): δ 7.59 (dt, *J* = 8.3, 1.8 Hz, 2H), 6.93 (dt, *J* = 8.3, 1.8 Hz, 2H), 2.93 (br s, 2H), 2.68 (t, *J* = 7.1 Hz, 2H), 2.57 (t, *J* = 7.7 Hz, 2H), 1.65-1.58 (m, 2H), 1.51-1.44 (m, 2H); ^{13}C-NMR (100 MHz/MeOD): δ 141.97, 137.42 (2C), 130.61 (2C), 90.81, 41.52, 35.22, 32.57, 28.53; ESI-MS (*m/z*): 276.0 $[M+H]^+$.

*Synthesis of **A5***

To a solution of **A4** (300 mg, 1.09 mmol) in anhydrous 1,4-dioxane (3 mL), tributylditin (2.2 mL, 4.4 mmol) and $Pd(PPh_3)_4$ (126 mg, 0.110 mmol) were added at room temperature, and the mixture was heated under reflux for 16 h. The mixture was concentrated under reduced pressure, and the residue was purified by silica gel column chromatography (Wako gel® C-200, 2-30% MeOH/chloroform) to afford **A5** as a colorless oil (171 mg, 0.390 mmol, 36%); ^{1}H-NMR (400 MHz/MeOD): δ 7.36 (d, *J* = 7.8 Hz, 2H), 7.18 (d, *J* = 7.8 Hz, 2H), 2.72 (t, *J* = 7.3 Hz, 2H), 2.64 (t, *J* = 7.5 Hz, 2H), 1.72-1.50 (m, 10H), 1.42-1.32 (m, 6H), 1.16-1.00 (m, 6H), 0.92 (t, *J* = 7.3 Hz, 9H); ^{13}C-NMR (100 MHz/MeOD): δ 144.20, 139.83, 138.29 (2C), 130.17 (2C), 42.96, 37.47, 33.33, 31.15 (3C), 30.66, 29.26 (3C), 14.91 (3C), 11.16 (3C); ESI-MS (*m/z*): 440.3 $[M + H]^+$.

*Synthesis of **A6***

To an ice-cooled solution of 4-aminobenzylamine (7.4 g, 60 mmol) in 2.0 M aqueous HCl (50 mL), $NaNO_2$ (4.6 g, 60 mmol) in water (10 mL) was added dropwise. After stirring for 20 min at 0˚C, NaN_3 (4.0 g, 60 mmol) in water (10 mL) was added, and the stirring was continued for 3 h at 0˚C. The reaction was quenched with saturated aqueous $NaHCO_3$, extracted with ethyl acetate, and the organic layer was dried over anhydrous $MgSO_4$. Silica gel column chromatography (Wako gel® C-200, 10% MeOH/chloroform) of the crude product gave **A6** as a brown solid (3.64 g, 24.6 mmol, 38%); ^{1}H-NMR (400 MHz/$CDCl_3$): δ 7.30 (d, *J* = 8.3 Hz, 2H), 6.99 (d, *J* = 8.3 Hz, 2H), 3.84 (s, 2H); ^{13}C-NMR (100 MHz/$CDCl_3$): δ 140.18, 138.65, 128.70 (2C), 119.25 (2C), 46.01.

*Synthesis of **A7a***

To a solution of methyl 3-amino-5,6-dichloro-2-pyrazinecarboxylate (500 mg, 2.25 mmol) in 2-propanol (10 mL), 4-iodobenzylamine (1.05 g, 4.50 mmol) was added in one portions, and the mixture was heated under reflux for 4 h. After the reaction mixture was concentrated under reduced pressure, the residue was purified by silica gel column chromatography (Wako gel® C-200, 30% ethyl acetate/*n*-hexane) to give **6** as a yellow solid (924 mg, 2.21 mmol, 98%): ^{1}H-NMR (400 MHz/$CDCl_3$): δ 7.68 (d, J = 8.4 Hz, 2H), 7.07 (d, J = 8.4 Hz, 2H), 5.84 (t, J = 5.0 Hz, 1H), 4.60 (d, J = 5.9 Hz, 2H), 3.90 (s, 3H); ^{13}C-NMR (100 MHz/$CDCl_3$): δ 166.68, 155.62, 151.45, 138.10 (2C), 137.50, 129.79 (2C), 121.44, 111.22, 93.36, 52.32, 44.86; ESI-MS (*m/z*): 419.0 $[M+H]^+$.

*Synthesis of **A7b***

A7b was prepared from **A6** according to the procedure described for **A7a**. The crude product was purified by silica gel column chromatography (Wako gel® C-200, 30% ethyl acetate/*n*-hexane) to give **A7b** as a yellow solid (846 mg, 2.53 mmol, 95%); ^{1}H-NMR (400 MHz/$CDCl_3$): δ 7.32 (dt, J = 8.6, 2.3 Hz, 2H), 7.02 (dt, J = 8.6, 2.3 Hz, 2H), 5.82 (br s, 1H), 4.62 (d, J = 5.8 Hz, 2H), 3.90 (s, 3H); ^{13}C-NMR (100 MHz/$CDCl_3$): δ 166.70, 155.66, 151.46, 139.86, 134.48, 129.45 (2C), 121.48, 119.61 (2C), 111.12, 52.32, 44.85; ESI-MS (*m/z*): 334.1 $[M+H]^+$.

*Synthesis of **A8a***

To a solution of **A7a** (374 mg, 0.893 mmol) in MeOH (5 mL), 6.0 M aqueous NaOH (0.45 mL, 2.7 mmol) was added, and the mixture was heated under reflux for 1 h. The reaction mixture was cooled to room temperature and neutralized with 6.0 M aqueous HCl. The white solid, which was formed by addition of water, was collected by filtration to give **A8a** (305 mg, 0.754 mmol, 84%) as a white solid; ^{1}H-NMR (400 MHz/DMSO-d_6): δ 7.68 (d, J = 8.3 Hz, 2H), 7.17 (d, J = 8.3 Hz, 2H), 4.50 (d, J = 6.2 Hz, 2H); ^{13}C-NMR (100 MHz/DMSO-d_6): δ 202.13, 169.76, 155.58, 149.91, 141.00, 137.75 (2C), 130.64 (2C), 117.42, 93.09, 44.02; ESI-MS (*m/z*): 403.0 $[M-H]^+$.

*Synthesis of **A8b***

A8b was prepared from **A7b** according to the procedure described for **A8a**, and the product was obtained as a yellow solid (132 mg, 0.413 mmol, 77%); ^{1}H-NMR (400 MHz/DMSO-d_6): δ 7.50 (br s, 1H), 7.41 (dt, J = 8.6, 2.2 Hz, 2H), 7.09 (dt, J = 8.6, 2.2 Hz, 2H), 4.54 (d, J = 6.2 Hz, 2H); ^{13}C-NMR (125 MHz/DMSO-d_6): δ 202.12, 168.95, 155.85, 150.54, 138.56, 137.80, 130.00 (2C), 119.74 (2C), 118.29, 43.99; ESI-MS (*m/z*): 320.0 $[M+H]^+$.

*Synthesis of **A9a***

To a mixture of 5-methylisoxazole (570 mg, 6.9 mmol) and *t*-BuOH (510 mg, 6.9 mmol), TfOH (1.0 g, 6.9 mmol) was added dropwise at room temperature. The mixture was stirred for 1 h at room temperature, then

A8a (932 mg, 2.30 mmol) in DMF (15 mL) and TEA (700 mg, 6.9 mmol) were added sequentially. After stirring for 16 h at room temperature, the reaction was quenched with iced water to form yellow precipitate. The resulting precipitate was collected by filtration and washed with water to afford **A9a** as a yellow solid (874 mg, 1.61 mmol, 70%); ^{1}H-NMR (400 MHz/$CDCl_3$): δ 7.69 (d, J = 8.4 Hz, 2H), 7.08 (d, J = 8.4 Hz, 2H), 6.62 (br s, 1H), 6.02 (t, J = 5.6 Hz, 1H), 5.42 (d, J = 1.0 Hz, 1H), 4.61 (d, J = 5.8 Hz, 2H), 2.13 (d, J = 0.92, 3H), 1.28 (s, 3H); ^{13}C-NMR (100 MHz/$CDCl_3$): δ 163.73, 162.55, 156.30, 153.20, 151.88, 138.16 (2C), 137.14, 129.85 (2C), 122.05, 114.99, 109.51, 93.51, 51.27, 44.99, 29.05, 20.84; ESI-MS (*m/z*): 544.0 $[M+H]^+$.

*Synthesis of **A9b***

A9b was prepared from **A8b** according to the procedure described for **A9a**, and the product was obtained as a yellow solid (180 mg, 0.392 mmol, 62%); ^{1}H-NMR (400 MHz/$CDCl_3$): δ 7.32 (d, J = 8.5 Hz, 2H),7.01 (dt, J = 8.5, 2.2 Hz, 2H), 6.65 (br s, 1H), 6.07 (t, J = 6.4 Hz, 1H), 5.41 (d, J = 0.83 Hz, 1H), 4.63 (d, J = 5.8 Hz, 2H), 2.12 (d, J = 0.83 Hz, 3H), 1.27 (s, 9H); ^{13}C-NMR (100 MHz/$CDCl_3$): δ163.70, 162.54, 156.35, 153.20, 151.88, 139.92, 134.15, 129.52 (2C), 122.08, 119.60 (2C), 114.99, 109.33, 51.23, 44.92, 29.03, 20.84; ESI-MS (*m/z*): 459.1 $[M+H]^+$.

*Synthesis of **A10a***

To a solution of *S*-methylisothiourea sulfate (1.12 g, 4.02 mmol) in 1.0 M aqueous NaOH (4.8 mL), dichloromethane (4 mL) and THF (4 mL), **A9a** (874 mg, 1.61 mmol) was added in one portion. The mixture was stirred for 16 h at room temperature, then concentrated under reduced pressure. The residue was washed with water, MeOH, and diethyl ether to give **A10a** as a white solid (555 mg, 1.16 mmol, 72%); ^{1}H-NMR (400 MHz/DMSO-d_6): δ 7.92 (t, J = 5.6 Hz, 1H), 7.70 (d, J = 8.2 Hz, 2H), 7.19 (d, J = 8.2 Hz, 2H), 4.54 (d, J = 6.0 Hz, 2H), 2.43 (s, 3H); ESI-MS (*m/z*): 477.0 $[M + H]^+$.

*Synthesis of **A10b***

A10b was prepared from **A9b** according to the procedure described for **A10a**, and the product was obtained as a yellow solid (221 mg, 0.564 mmol, 66%); ^{1}H-NMR (400 MHz/$CDCl_3$): δ 7.31 (dt, J = 8.6, 2.2 Hz, 2H), 7.02 (dt, J = 8.6, 2.2 Hz, 2H), 5.81 (t, J = 5.4 Hz, 1H), 4.62 (d, J = 6.6 Hz, 2H), 2.45 (s, 3H); ^{13}C-NMR (125 MHz/DMSO-d_6): δ 155.74, 155.68, 150.83, 150.77, 138.31, 136.87, 129.71 (2C), 119.39 (2C), 111.31, 107.45, 43.68, 13.75; ESI-MS (*m/z*): 392.0 $[M+H]^+$.

*Synthesis of **PRA3***

To a solution of **A3** (38 mg, 0.20 mmol) in anhydrous THF (2 mL), diisopropylethylamine (74 mg, 0.60 mmol) was added, and the mixture was stirred for 15 min at 65˚C. Then, **A10a** (114 mg, 0.239 mmol) was added to

the mixture, and the reaction mixture was stirred for 16 h at 65˚C. The mixture was concentrated under reduced pressure, and the residue was purified by silica gel column chromatography (Wako gel® C-200, 5% MeOH/chloroform) to afford **PRA3** as a yellow solid (69 mg, 0.11 mmol, 56%); ^{1}H-NMR (400 MHz/MeOD): δ 7.69 (d, J = 8.4 Hz, 2H),7.27 (d, J = 8.4 Hz, 2H), 7.18 (d, J = 8.4 Hz, 2H), 7.01 (d, J = 8.4 Hz, 2H), 4.66 (s, 2H), 3.37-3.36 (m, 2H), 2.71 (t, J = 6.9 Hz, 2H), 1.77-1.73 (m, 4H); ^{13}C-NMR (100 MHz/MeOD): δ 167.33, 157.62, 155.56, 154.27, 140.27, 139.89, 139.34, 138.81 (2C), 131.12 (2C), 130.93 (2C), 122.38, 120.16 (2C), 110.29, 93.13, 45.21, 42.49, 35.73, 29.60, 28.83; ESI-MS (*m/z)*: 619.0 $[M+H]^{+}$.

*Synthesis of **PRA4***

PRA4 was prepared from **A4** and **A10b** according to the procedure described for **PRA3**. The crude product was purified by silica gel column chromatography (Wako gel® C-200, 5% MeOH/chloroform) to give **PRA4** (107 mg, 0.173 mmol, 82%) as a yellow solid; ^{1}H-NMR (400 MHz/MeOD): δ 7.60 (d, J = 8.2 Hz, 2H), 7.43 (d, J = 8.5 Hz, 2H), 7.03-7.00 (m, 4H), 4.65 (s, 2H), 3.38-3.35 (m, 2H), 2.65 (t, J = 7.0 Hz, 2H), 1.79-1.69 (m, 4H); ^{13}C-NMR (100 MHz/MeOD): δ 167.89, 158.12, 156.08, 154.80, 143.75, 141.11, 139.38 (2C), 137.61, 132.55 (2C), 131.37 (2C), 123.09, 120.80 (2C), 110.77, 92.36, 45.81, 43.21, 36.54, 30.11, 29.54; ESI-MS (*m/z*): 619.1 $[M+H]^{+}$.

*Synthesis of **A11***

To a solution of **PRA3** (36 mg, 0.058 mmol) in anhydrous 1,4-dioxane (0.5 mL), tributylditin (0.14 mL, 0.29 mmol) and $Pd(PPh_3)_4$ (7 mg, 0.006 mmol) were added, and the mixture was stirred for 16 h at 50˚C. The mixture was concentrated under reduced pressure, and the residue was purified by silica gel column chromatography (Wako gel® C-200, 2-5% MeOH/chloroform) to afford **A11** as a yellow solid (4.0 mg, 0.0051 mmol, 8.8%); ^{1}H-NMR (400 MHz/MeOD): δ 7.43 (d, J = 8.0 Hz, 2H), 7.34 (d, J = 8.0 Hz, 2H), 7.27 (d, J = 8.4 Hz, 2H), 7.01 (d, J = 8.4 Hz, 2H), 4.62 (s, 2H), 3.37 (t, J = 7.1 Hz, 2H), 2.71 (t, J = 7.0 Hz, 2H), 1.77-1.73 (m, 4H), 1.62-1.54 (m, 6H), 1.39-1.32 (m, 6H), 1.17-1.02 (m, 6H), 0.88 (t, J = 7.3 Hz, 9H); ^{13}C-NMR (100 MHz/$CDCl_3$): δ 166.68, 157.62, 155.56, 154.27, 138.59, 137.98, 137.65, 137.04 (2C), 131.00, 129.78 (2C), 128.84, 127.32 (2C), 118.99 (2C), 110.16, 34.62, 29.07 (3C), 28.97, 28.47, 28.34, 27.37 (3C), 25.69, 13.67 (3C), 9.62 (3C); ESI-MS (*m/z*): 783.2 $[M+H]^{+}$.

*Synthesis of **A12***

A12 was prepared from **A5** and **A10b** according to the procedure described for **PRA3**. The crude product was purified by silica gel column chromatography (Wako gel® C-200, 5% MeOH/chloroform) to give **A12** as a white solid (19 mg, 0.024 mmol, 47%); ^{1}H-NMR (400 MHz/$CDCl_3$): δ 7.36 (d, J = 7.7 Hz, 2H), 7.30 (d, J = 8.4 Hz, 2H), 7.13 (d, J = 7.7 Hz, 2H), 6.99 (d, J = 8.4 Hz, 2H), 4.59 (s, 2H), 3.30 (br s, 2H), 2.61 (br s, 2H), 1.71 (br

s, 4H), 1.71-1.45 (m, 6H), 1.37-1.25 (m, 6H), 1.11-0.94 (m, 6H), 0.88 (t, J = 7.3 Hz, 9H); ^{13}C-NMR (100 MHz/$CDCl_3$): δ 167.89, 155.03, 150.76, 141.45, 139.51, 138.78, 136.54 (2C), 134.57, 131.04, 129.25 (2C), 128.82, 128.13 (2C), 120.38, 119.34, 44.62, 41.67, 35.38, 29.02 (3C), 29.01, 28.52, 27.40 (3C), 13.69 (3C), 9.55 (3C); ESI-MS (*m/z*): 783.3 $[M+H]^+$.

Synthesis of ***[^{125}I]PRA3***

To a solution of **A11** (1.0 mM in EtOH, 20 μL) in a screw-capped 1.5 mL plastic tube, [^{125}I]NaI (Perkin-Elmer, NEZ 033A, 1 mCi, 2,000 Ci/mmol, 10 μL) was added. The radio-iodination was initiated by adding freshly prepared aqueous chloramine T (3.0 mM in 1.0 M KPi buffer (pH 7.4), 10 μL), and the mixture was incubated for 10 min at room temperature. The reaction was quenched with 5% (w/v) aqueous $NaHSO_3$ (50 μL) then the mixture was subjected to HPLC (Shimadzu LC-10AS, Kyoto, Japan) purification using a C18 column (COSMOSIL 5C18-MSII, 4.6 mm x 150 mm, Nacalai Tesque, Kyoto, Japan) at a flow rate of 0.8 mL/min with MeOH/ 0.01% aqueous TFA as an eluent.

The column was eluted with isocratic 10% MeOH in 5 min, then isocratic 80% MeOH in 15 min. The fraction was collected every 30 s (400 μL) and the radioactivity and radiochemical purity were assessed by γ-counting system (COBRA^{TM} II, Packard) and radio-TLC analysis. The radioactive fractions, corresponding to the retention time of cold PRA3 (14.5 min), were combined and the solvent was evaporated by a vacuum-centrifugal evaporator. **[^{125}I]PRA3** was stored as an ethanoic solution (1 mCi/mL) at 4˚C. The radiochemical yield of **[^{125}I]PRA3** from the initial [^{125}I]NaI was 63%. The radiochemical purity and the specific activity were >99% and 2,000 Ci/mmol, respectively (judged from HPLC and radio-TLC).

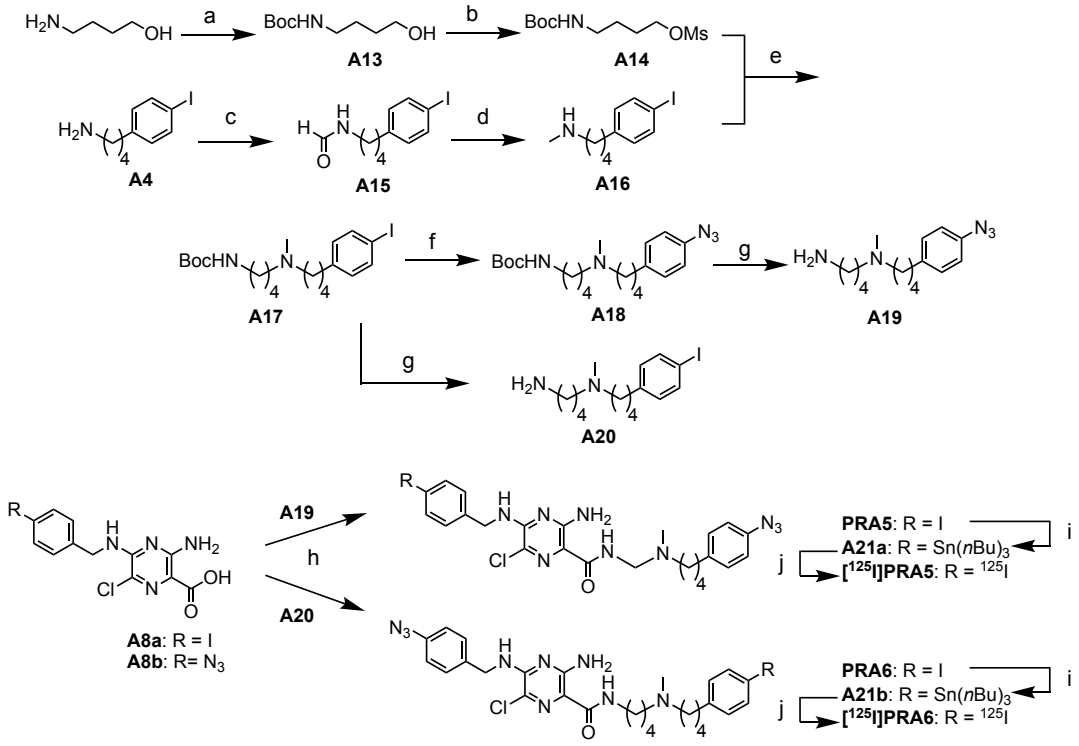
Synthesis of ***[^{125}I]PRA4***

[^{125}I]PRA4 was prepared from **A12** according to the procedure described for **[^{125}I]PRA3**. The radiochemical yield of **[^{125}I]PRA4** from the initial [^{125}I]NaI was 39%. The radiochemical purity and the specific activity were >99% and 2,000 Ci/mmol, respectively (judged from HPLC and radio-TLC). **[^{125}I]PRA4** was stored as an ethanoic solution (1 mCi/mL) at 4˚C.

*Outline of the syntheses of **PRA5**, **PRA6**, [^{125}I]**PRA5**, and [^{125}I]**PRA6***

PRA5 and **PRA6** were synthesized according to the previously methods [*11*], as illustrated in Scheme S5. The amine derivatives **A19** and **A20** were prepared from 4-amino-1-butanol and 4-phenylbutylamine as starting materials. They were subjected to the conjugation with pyrazine carboxylic acid **A8a** or **A8b** in the presence of *N*-methylmorpholine and HATU to provide **PRA5** and **PRA6**, respectively. **[^{125}I]PRA5** and **[^{125}I]PRA6** were prepared by the catalysis of chloramine T using tin-precursor **A21a** and **A21b**, respectively.

Scheme S5[e]



[e]*Reagents and conditions*: (a) Boc_2O, TEA, CH_2Cl_2, rt, 1 h, quant.; (b) MsCl, TEA, CH_2Cl_2, 0℃, 30 min; (c) ethyl formate, reflux, overnight; (d) $LiAlH_4$, THF, reflux, 3 h; (e) K_2CO_3, MeCN, reflux, overnight, 24% (3 steps from **A4**); (f) *trans-N,N'*-dimetylcyclohexane-1.2-diamine, CuI, *L*-ascorbic acid Na, EtOH/water (4:1), reflux, 4 h; (g) TFA, CH_2Cl_2, rt, 30 min, crude and quant. (for **A19** and **A20**); (h) 4-methylmorpholine, HATU, DMF, rt, 2 h, 23 (2 steps) and 60% (for **PRA5** and **PRA6**); (i) ($Sn(n\text{-}Bu)_3)_2$, $Pd(PPh_3)_4$, dioxane, 50℃, 4 h, 18 and 17% (for **A21a** and **A21b**); (j) [^{125}I]NaI, chloramine T, aq. K_2HPO_4, rt, 10 min, 70 and 61% (for **[^{125}I]PRA5** and **[^{125}I]PRA6**).

*Synthesis of **A13***

To a solution of 4-amino-1-butanol (1.0 g, 11 mmol) in anhydrous dichloromethane (20 mL), Boc_2O (2.9 g, 13 mmol) and trimethylamine (3.3 g, 33 mmol) were added in portions, and the mixture was stirred for 1 h at room temperature. The reaction was quenched with water, extracted with dichloromethane and washed with saturated aqueous NH_4Cl. The organic layer was dried over anhydrous $MgSO_4$. After removal of the solvent, the crude product was purified by silica gel column chromatography (Wako gel® C-200, 5-10% MeOH/chloroform) to give **A13** as colorless oil (2.31 g, quant.); ^{1}H-NMR (400 MHz/$CDCl_3$): δ 4.63 (br s, 1H), 3.66 (t, J = 5.8 Hz, 2H), 3.65 (q, J = 5.7 Hz, 2H), 1.60-1.55 (m, 4H), 1.43 (s, 9H); ^{13}C-NMR (100 MHz/$CDCl_3$): δ 156.34, 79.52, 62.67, 40.51, 29.93, 28.62 (3C), 26.83; ESI-MS (*m/z*): 190.1 $[M + H]^+$.

*Synthesis of **A14***

To a solution of **A13** (1.16 g, 5.5 mmol) in anhydrous dichloromethane (30 mL), triethylamine (650 mg, 6.5 mmol) and MsCl (744 mg, 6.5 mmol) were added carefully, and the mixture was stirred for 30 min at 0˚C. The reaction was quenched with water, extracted with diethyl ether, and the organic layer was dried over anhydrous $MgSO_4$ to give **A14** as a yellow oil (1.55 g, crude); ^{1}H-NMR (400 MHz/$CDCl_3$): δ 4.58 (brs, 1H), 4.24 (t, J = 6.3 Hz, 2H), 3.16 (q, J = 6.5 Hz, 2H), 3.00 (s, 3H), 1.81-1.74 (m, 2H), 1.64-1.56 (m, 2H); ^{13}C-NMR (100 MHz/$CDCl_3$): δ 156.18, 79.52, 69.77, 39.97, 37.59, 28.59, 26.61, 26.49; ESI-MS (*m/z*): 268.1 $[M + H]^+$.

*Synthesis of **A15***

The mixture of **A4** (1.26 g, 4.58 mmol) and ethyl formate (1.0 g, 14 mmol) was heated under reflux for 16 h. The mixture was concentrated under reduced pressure to give **A15** as yellow solid (1.45 g, crude); ^{1}H-NMR (400 MHz/$CDCl_3$): δ 8.15 (s, 1H), 7.59 (dt, J = 8.3, 1.8 Hz, 2H), 6.93 (dt, J = 8.3, 1.8 Hz, 2H), 5.51 (br s, 1H), 3.31 (q, J = 6.6 Hz, 2H), 2.58 (t, J = 7.4 Hz, 2H), 1.67-1.60 (m, 2H), 1.59-1.54 (m, 2H); ^{13}C-NMR (100 MHz/$CDCl_3$): δ 161.35, 141.76, 137.62 (2C), 130.73 (2C), 91.09, 38.10, 35.08, 29.24, 28.56; ESI-MS (*m/z*): 304.0 $[M + H]^+$.

*Synthesis of **A16***

To a suspension of $LiAlH_4$ (261 mg, 6.87 mmol) in anhydrous THF (10 mL), **A15** (1.45 g) in THF (10 mL) was added, and the mixture was heated under reflux for 3 h. After being cooled to 0˚C, the reaction was quenched with 2.0 M aqueous NaOH, then stirred for 30 min at room temperature. The mixture was filtered through a pad of Celite® and the filtrate was concentrated under reduced pressure to give **A16** as a yellow oil (1.06 g, crude); ^{1}H-NMR (400 MHz/$CDCl_3$): δ 7.58 (dt, J = 8.3, 1.8 Hz, 2H), 6.92 (dt, J = 8.3, 1.8 Hz, 2H), 2.66-2.55 (m, 4H), 2.42 (s, 3H), 1.68-1.61 (m, 2H), 1.59-1.52 (m, 2H); ESI-MS (*m/z*): 290.0 $[M + H]^+$.

*Synthesis of **A17***

To a solution **A14** (1.55 g) in acetonitrile (30 mL), **A16** (1.06 g) and K_2CO_3 (1.52 g, 11.0 mmol) was added, and the mixture was heated under reflux for 16 h. The mixture was concentrated under reduced pressure and the residue was resuspended in dichloromethane, washed with water. The organic layer was dried over anhydrous $MgSO_4$, and the crude product was purified by silica gel column chromatography (Wako gel® C-200, 10% MeOH/chloroform) to give **A17**. The crude product was further purified by preparative HPLC (COSMOSIL 5C_{18}-MS-II 20 mm x 250 mm, 80 % MeOH/water) to give pure **A17** as a colorless oil (267 mg, 0.560 mmol, 24%, 3 steps); ^{1}H-NMR (400 MHz/$CDCl_3$): δ 7.58 (dt, J = 8.3, 1.8 Hz, 2H), 6.92 (d, J = 8.3 Hz, 2H), 5.04 (br s, 1H), 3.11 (d, J = 5.4 Hz, 2H), 2.56 (t, J = 7.5 Hz, 2H), 2.33-2.28 (m, 4H), 2.16 (s, 3H), 1.62-1.55 (m, 2H), 1.51-1.46 (m, 6H), 1.43 (s, 9H); ^{13}C-NMR (100 MHz/$CDCl_3$): δ 156.27, 142.36, 137.50 (2C), 130.75 (2C), 90.84, 79.07, 57.82, 57.69, 42.21, 40.74, 35.55, 29.37, 28.67 (3C), 28.22, 27.05, 25.01; ESI-MS (*m/z*): 461.2 $[M + H]^+$.

*Synthesis of **A18***

To a solution of **A17** (136 mg, 0.295 mmol) in a mixture of EtOH (1.2 mL) and water (0.3 mL), *trans-N,N'*-dimethylcyclohecane-1,2-diamine (84 mg, 0.59 mmol), NaN_3 (38 mg, 0.59 mmol), CuI (11 mg, 0.058 mmol), and sodium *L*-ascorbic acid (12 mg, 0.060 mmol) were added. The mixture was heated under reflux for 4 h, then quenched with water and extracted with ethyl acetate. The organic layer was dried over anhydrous $MgSO_4$ and the crude product was purified by silica gel column chromatography (Wako gel® C-200, 10% MeOH/chloroform) to give **A18** as a brown oil (145 mg, crude); ^{1}H-NMR (400 MHz/$CDCl_3$): δ 7.15 (d, J = 8.5 Hz, 2H), 6.94 (dt, J = 8.5, 2.1 Hz, 2H), 5.04 (br s, 1H), 3.11 (d, J = 5.2 Hz, 2H), 2.60 (t, J = 7.5 Hz, 2H), 2.36-2.32 (m, 4H), 2.18 (s, 3H), 1.61-1.56 (m, 2H), 1.51-1.49 (m, 6H), 1.43 (s, 9H); ^{13}C-NMR (100 MHz/$CDCl_3$): δ 156.29, 139.52, 137.64, 129.91 (2C), 119.11 (2C), 57.78, 57.62, 42.14, 40.69, 35.39, 29.55, 28.66 (3C), 28.20, 26.91, 24.84; ESI-MS (*m/z*): 376.3 $[M+H]^+$.

*Synthesis of **A19***

To a solution of crude **A18** (145 mg) in dichloromethane (1.2 mL), TFA (0.8 mL) was added carefully. After stirring for 30 min at room temperature, the mixture was concentrated under reduced pressure. Then, the residue was resuspended in dichloromethane, washed 1.0 M aqueous NaOH, and the organic layer was dried over anhydrous $MgSO_4$. The removal of the solvent gave **A19** as a brown oil (108 mg, crude); ^{1}H-NMR (400 MHz/$CDCl_3$): δ 7.14 (d, J = 8.4 Hz, 2H), 6.92 (d, J = 8.4 Hz, 2H), 2.70 (m, 2H), 2.59 (t, J = 7.5 Hz, 2H), 2.34-2.29 (m, 4H), 2.18 (s, 3H), 1.62-1.55 (m, 2H), 1.49-1.46 (m, 6H); ^{13}C-NMR (100 MHz/$CDCl_3$): δ 139.72, 137.98, 129.91 (2C), 119.08 (2C), 57.90, 57.77, 42.35, 42.25, 35.39, 31.77, 29.23, 27.02, 24.34; ESI-MS (*m/z*): 276.2 $[M+H]^+$.

*Synthesis of **A20***

A20 was prepared from **A17** according to the same procedure described for **A19**, and the product was obtained as a yellow oil (98 mg, 0.27 mmol, 99%); ^{1}H-NMR (400 MHz/$CDCl_3$): δ 7.56 (dt, J = 8.3, 2.0 Hz, 2H), 6.91 (d, J = 8.3 Hz, 2H), 2.68 (t, J = 6.6 Hz, 2H), 2.54 (t, J = 7.5 Hz, 2H), 2.32-2.27 (m, 4H), 2.16 (s, 3H), 1.61-1.52 (m, 2H), 1.49-1.40 (m, 6H); ^{13}C-NMR (100 MHz/$CDCl_3$): δ 142.33, 137.43 (2C), 130.72 (2C), 90.79, 57.91, 57.75, 42.42, 42.35, 35.50, 31.96, 29.33, 27.07, 24.90; ESI-MS (*m/z*): 361.1 $[M+H]^+$.

*Synthesis of **PRA5***

To a solution of **A8a** (119 mg, 0.295 mmol) in anhydrous DMF (2 mL), **A19** (108 mg, crude), 4-methylmorpholine (149 mg, 1.48 mmol), and HATU (168 mg, 0.442 mmol) was added sequentially. The mixture was stirred for 2 h at room temperature, and the reaction was quenched with 2.0 M aqueous NaOH, followed by the extraction with ethyl acetate. The organic layer was dried over anhydrous $MgSO_4$, and the crude product was purified by silica gel column chromatography (Wako gel® C-200, 10% MeOH/chloroform) to afford **PRA5** as a yellow solid (46 mg, 0.069 mmol, 23%, 2 steps); ^{1}H-NMR (400 MHz/$CDCl_3$): δ 7.67 (d, J = 8.3 Hz, 2H), 7.13 (d, J = 8.4 Hz, 2H), 7.07 (d, J = 8.3 Hz, 2H), 6.92 (d, J = 8.4 Hz, 2H), 5.67 (t, J = 5.8 Hz, 1H), 4.57 (d, J = 5.8 Hz, 2H), 3.37 (q, J = 5.8 Hz, 2H), 3.04-2.95 (m, 4H), 2.72 (s, 3H), 2.61 (t, J = 6.9 Hz, 2H), 1.76-1.72 (m, 4H), 1.66-1.63 (m, 4H); ^{13}C-NMR (100 MHz/$CDCl_3$): δ 166.68, 154.03, 151.17, 139.64, 138.27, 137.98 (2C), 137.89, 129.90 (2C), 129.85 (2C), 119.63, 119.21 (2C), 113.16, 93.19, 56.52, 55.99, 44.79, 40.60, 37.64, 34.58, 28.26, 27.28, 23.74, 21.26; ESI-MS (*m/z*): 662.2 $[M+H]^+$.

*Synthesis of **PRA6***

PRA6 was prepared from **A8b** and **A20** according to the procedure described for **PRA5**. The crude product was purified by silica gel column chromatography (Wako gel® C-200, 5−20% MeOH/chloroform) to afford **PRA6** as a yellow solid (107 mg, 0.162 mmol, 60%); ^{1}H-NMR (400 MHz/$CDCl_3$): δ 7.53 (d, J = 8.2 Hz, 2H), 7.30 (d, J = 8.5 Hz, 2H), 6.98 (dt, J = 8.5, 2.2 Hz, 2H), 6.88 (d, J = 8.2 Hz, 2H), 5.69 (t, J = 5.8 Hz, 1H), 4.57 (d, J = 5.8 Hz, 2H), 3.33 (q, J = 6.4 Hz, 2H), 3.05-2,97 (m, 4H), 2.73 (s, 3H), 2.54 (t, J = 7.4 Hz, 2H), 1.70-1.67 (m, 4H), 1.62-1.58 (m, 4H); ^{13}C-NMR (100 MHz/$CDCl_3$): δ 166.82, 154.05, 151.19, 141.28, 139.62, 137.58 (2C), 134.82, 130.73 (2C), 129.48 (2C), 119.72, 119.50 (2C), 112.90, 91.20, 56.76, 56.24, 44.77, 40.93, 37.62, 34.75, 28.09, 27.32, 24.15, 21.56; ESI-MS (*m/z*): 662.2 $[M+H]^+$.

*Synthesis of **A21a***

To a solution of **PRA5** (41 mg, 0.062 mmol) in anhydrous 1,4-dioxane (1.0 mL), tributylditin (0.16 mL, 0.31 mmol), and $Pd(PPh_3)_4$ (7.0 mg, 0.006 mmol) were added. After stirring for 8 h at 50˚C, the mixture was

concentrated under reduced pressure. The residue was purified by silica gel column chromatography (Wako gel® C-200, 25% MeOH/chloroform) to afford crude **A21a** which was further purified by preparative HPLC (COSMOSIL 5C_{18}-MS-II 20 mm x 250 mm, 85% MeOH/ 0.01% aqueous TFA) to give pure **A21a** as a yellow solid (9.0 mg, 0.011 mmol, 18%); ^{1}H-NMR (400 MHz/$CDCl_3$): δ 7.46 (d, J = 7.9 Hz, 2H), 7.28 (d, J = 7.9 Hz, 2H), 7.13 (d, J = 8.4 Hz, 2H), 6.95 (d, J = 8.4 Hz, 2H), 5.64 (t, J = 5.6 Hz, 1H), 4.61 (d, J = 5.6 Hz, 2H), 3.43-3.38 (m, 2H), 3.13-2.92 (m, 4H), 2.74 (s, 3H), 2.62 (t, J = 7.3 Hz, 2H), 1.80-1.75 (m, 4H), 1.65-1.60 (m, 4H), 1.57-1.50 (m, 6H), 1.37-1.28 (m, 6H), 1.34-0.96 (m, 6H), 0.88 (t, J = 7.3 Hz, 9H); ^{13}C-NMR (100 MHz/$CDCl_3$): δ 166.72, 154.15, 151.34, 141.85, 138.20, 138.02, 137.58, 137.14 (2C), 129.87 (2C), 127.51 (2C), 119.72, 119.36 (2C), 113.15, 55.70, 55.36, 45.49, 39.82, 37.76, 34.69, 29.29 (3C), 28.52, 27.58 (3C), 27.46, 23.36, 20.95, 13.88 (3C), 9.81 (3C); ESI-MS (*m/z*): 826.4 $[M+H]^+$.

*Synthesis of **A21b***

A21b was prepared from **PRA6** according to the procedure described for **A21a**. The crude product was purified by silica gel column chromatography (Wako gel® C-200, 2-5% MeOH/chloroform) to give crude **30**, which was further purification by preparative HPLC (COSMOSIL 5C_{18}-MS-II, 20 mm x 250 mm, 85% MeOH/0.01% aqueous TFA) to give pure **A21b** as a yellow solid (11 mg, 0.013 mmol, 17%); ^{1}H-NMR (400 MHz/$CDCl_3$): δ 7.37 (d, J = 7.9 Hz, 2H), 7.31 (d, J = 8.5 Hz, 2H), 7.10 (d, J = 7.9 Hz, 2H), 7.01 (dt, J = 8.5, 2.2 Hz, 2H), 5.64 (t, J = 5.7 Hz, 1H), 4.60 (d, J = 5.8 Hz, 2H), 3.41-3.37 (m, 2H), 3.11-2.92 (m, 4H), 2.73 (s, 3H), 2.61 (t, J = 7.4 Hz, 2H), 1.80-1.76 (m, 4H), 1.67-1.59 (m, 4H), 1.57-1.49 (m, 6H), 1.38-1.28 (m, 6H), 1.14-0.99 (m, 6H), 0.88 (t, J = 7.3 Hz, 9H); ^{13}C-NMR (100 MHz/$CDCl_3$): δ 166.49, 154.06, 151.18, 140.90, 139.76, 139.43, 136.87 (2C), 134.84, 129.43 (2C), 128.74, 128.23 (2C), 119.57 (2C), 113.44, 55.73, 55.37, 44.83, 39.77, 37.81, 35.31, 29.30 (3C), 28.47, 27.58 (3C), 27.44, 23.38, 21.05, 13.88 (3C), 9.76 (3C); ESI-MS (*m/z*): 826.4 $[M+H]^+$.

*Synthesis of **[^{125}I]PRA5***

[^{125}I]PRA5 was prepared from **A21a** according to the procedure described for **[^{125}I]PRA3**. The radiochemical yield of **[^{125}I]PRA5** from the initial [^{125}I]NaI was 70%. The radiochemical purity and the specific activity were >99% and 2,000 Ci/mmol, respectively (judged from HPLC and radio-TLC). **[^{125}I]PRA5** was stored as an ethanoic solution (1 mCi/mL) at 4˚C.

*Synthesis of **[^{125}I]PRA6***

[^{125}I]PRA6 was prepared from **A21b** according to the procedure described for **[^{125}I]PRA3**. The radiochemical yield of **[^{125}I]PRA6** from the initial [^{125}I]NaI was 61%. The radiochemical purity and the specific activity were >99% and 2,000 Ci/mmol, respectively (judged from HPLC and radio-TLC). **[^{125}I]PRA6** was stored as an ethanoic solution (1 mCi/mL) at 4˚C.

References (for synthetic procedures)

1. Knoevenagel, E (1898) *Ber. Deutsch. Chem. Ges.* 1898, 31, 2596.
2. Okuda, K., Murai, M., Aburaya, S., Aoki, W., and Miyoshi, H. (2016) *Biochemistry* 55, 470-481.
3. Masuya, T., Okuda, K., Murai, M., and Miyoshi, H. (2016) *Biosci. Biotechnol. Biochem.* 80, 1464-1469.
4. Sonogashira, K. Tohda, Y., and Hagiwara, N. (1975) Tetrahedron Lett. 50, 4467.
5. van der Klei, A., de Jong, R. L. P., Lugtenburg, J., and Tielens, A. G. M. (2002) *Eur. J. Org. Chem.* 2002. 3015-3023.
6. Hay, A. S. (1962) *J. Org. Chem.* 27, 3320-3321.
7. Ito, T., Murai, M., Ninokura, S., Kitazumi, Y., Mezic, K. G., Cress, B. F., Koffas, M. A. G., Morgan, J. E., Barquera, B., and Miyoshi, H. (2017) *J. Biol. Chem.* 292, 7727-7742.
8. Hamprecht, D., Hechel, A., and Kley, J. (2014) WO 2014044849.
9. Hirsh, A. J., Molino, B. F., Zhang, J. Astakhova, N., Geiss, W. B. Sargent,B. J., Swenson, B. D., Usyatinsky, A. Wyle, M. J., Boucher, R. C., Smith, R. T., Zamurs, A., and Johnson, M. R. (2006) *J. Med. Chem.* 49, 4098-4115.
10. Murai, M., Habu, S., Murakami, S., Ito, T., and Miyoshi, H. (2015) *Biosci. Biotechnol. Biochem.* 79, 1061-1066.
11. Ito, T., Murai, M., Morisaka, H., and Miyoshi, H. (2015) *Biochemistry* 54, 3677-3686.

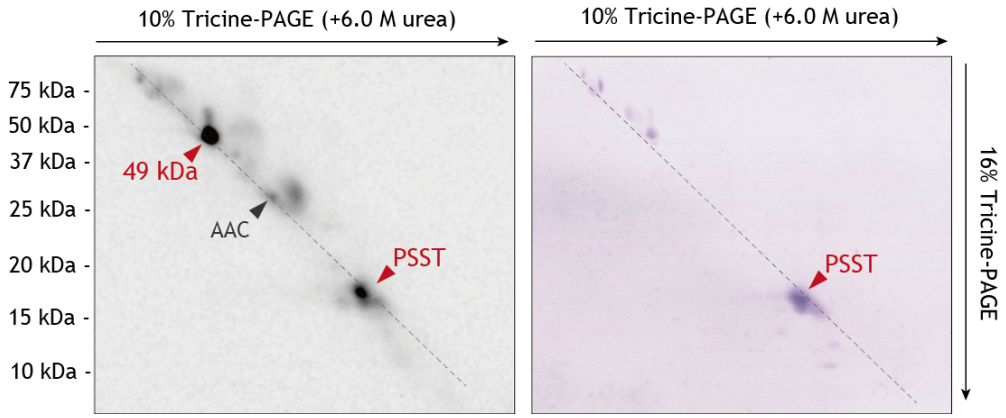


Figure S1

Detection of the PSST subunit on the doubled-SDS gel. (A) SMPs (4.0 mg of proteins/mL) were labeled by [^{125}I]PRA8 (10 nM), and the complex I was isolated by BN-PAGE and electroelution. The complex I labeled by [^{125}I]PRA8 was separated on a first dimensional 10% Schägger-type SDS gel (10% T, 3% C, containing 6.0 M urea) followed by a second dimensional separation on a 16% Schägger-type SDS gel (16% T, 3% C). The SDS gel was subjected to autoradiography. The autoradiogram was identical to that used in Figure 7C. (B) The migration pattern of the PSST subunit on the 2D gel was also profiled by Western blotting using anti-*Paracoccus* Nqo6 antibody (PSST homologue, a kind gift from Dr. Takao Yagi).

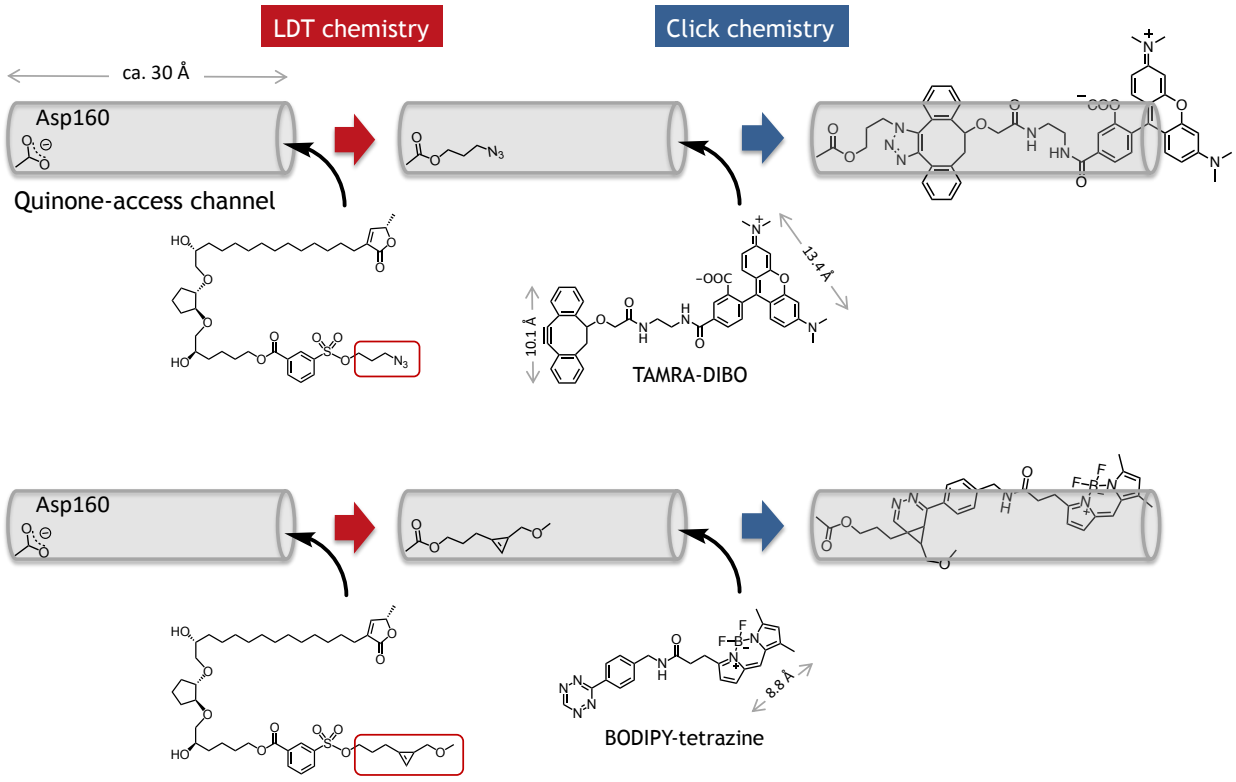


Figure S2

Schematic diagram of the pinpoint chemical modification of the 49 kDa Asp160. The 49 kDa Asp160 can be modified by externally added bulky tags such as TAMRA-DIBO and BODIPY-tetrazine through the two-step conjugation procedure (LDT and click chemistry) (refs. *15* and *18*).

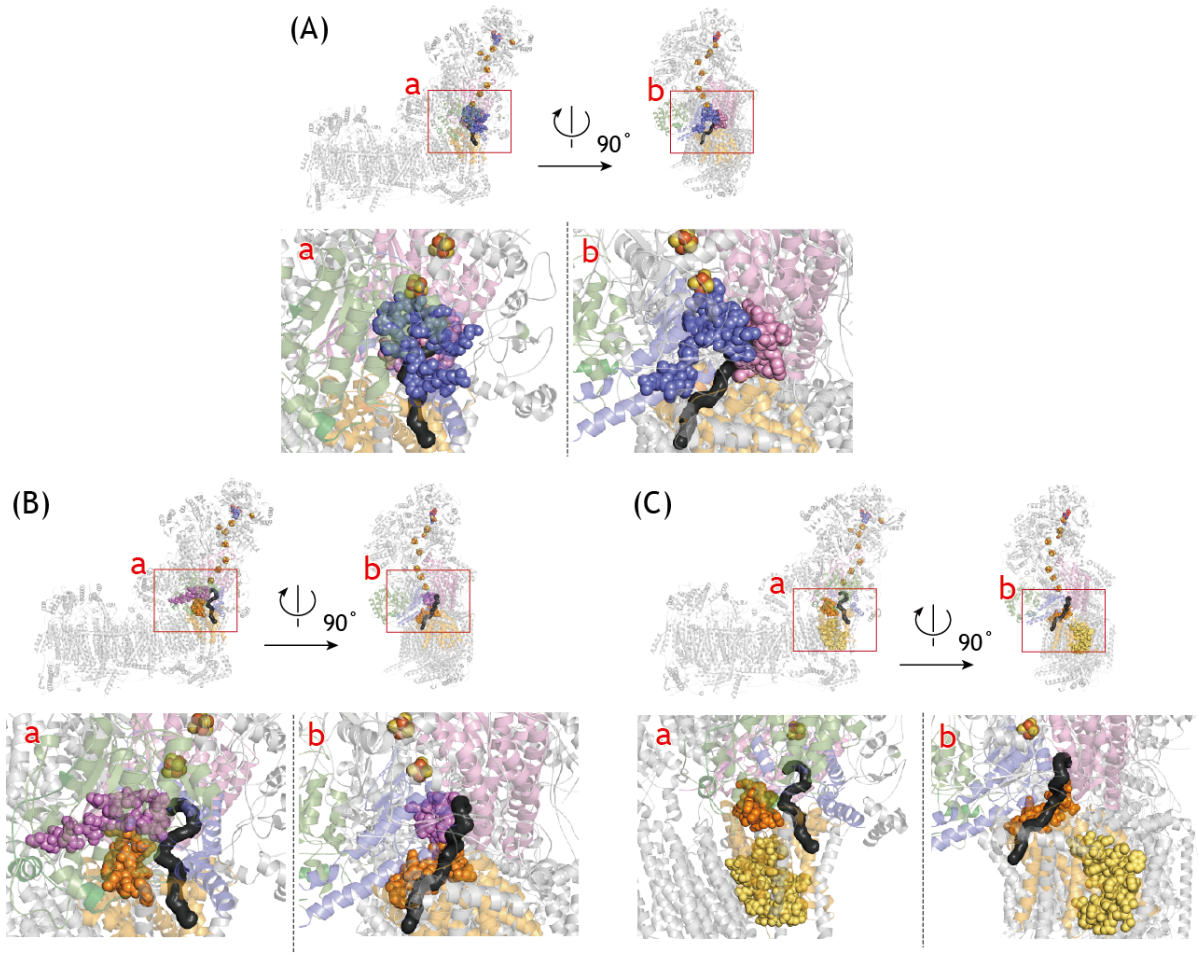


Figure S3

Summary of photoaffinity labeling studies using different quinone-site inhibitors. The regions labeled by photoreactive inhibitors are shown by *spheres* in ovine complex I (PDB entry 5LNK, ref. *9*). The quinone/inhibitor-access channel in the current models is shown in *black*. (A) The region labeled by photoreactive fenpyroximate analogs ([^{125}I]APF and [^{125}I]AIF): the interface between the PSST (Ser43−Arg66) and 49 kDa (Asp160−Arg174) are shown in *blue* and *pink* spheres, respectively. (B) The region labeled by a photoreactive quinazoline ([^{125}I]AzQ): the interface between the *N*-terminal region (Val44−Arg63) in 49 kDa and the matrix side loop connecting the TMH5-6 (Thr201−Ala217) in ND1 are shown in *pink* and *orange* spheres, respectively. (C) The region labeled by photoreactive acetogenin analogs ([^{125}I]TDA and [^{125}I]DANA): the loop connecting the TMH5-6 and the region spanning TMH4-5 (Val144-Glu192) are shown in *orange* and *yellow* spheres, respectively.

Table S1. Proteins identified by MALDI-TOF MS

Protein name (Swiss Prot accession No.)	Matched peptides	MOWSE score [a] (Sequence coverage)	Matched in-gel triptic digests				
			Observed *m/z* (MH^+)	Mr		Peptide sequence	Residues
				Expected	Calculated		
Complex I-49 kDa subunit (P17694)	17	105 (43%)	2014.085	2013.0777	2012.9745	ETAHWKPPPWNDVDPPK	24-40
			2457.451	2456.4437	2456.2924	DTLVSNLTLNFGPQHPAAHGVLR	41-63
			1263.737	1262.7297	1262.6363	LVMELSGEMVR	64-74
			1279.731	1278.7237	1278.6312	LVMELSGEMVR + Oxidation (M)	64-74
			1217.728	1216.7207	1216.6135	CDPHIGLLHR	76-85
			2033.13	2032.1227	2032.0669	LIEYKTYLQALPYFDR	90-105
			1386.803	1385.7957	1385.6979	TYLQALPYFDR	90-105
			934.607	933.5997	933.5284	VLFGEITR	139-146
			1021.517	1020.5097	1020.4375	MFEFYER	177-183
			1037.509	1036.5017	1036.4324	MFEFYER + Oxidation (M)	177-183
			1476.814	1475.8067	1475.6926	IDELEEMLTNNR	222-233
			2441.399	2440.3917	2440.2308	TVDIGIVTAEDALNYGFSGVMLR	239-261
			1031.604	1030.5967	1030.5196	GSGIQWDLR	262-270
			2228.195	2227.1877	2227.0433	TQPYDVYDQVEFDVPIGSR	272-290
			1789.058	1788.0507	1787.8698	AEMKTSMESLIHHFK	336-350
			2269.261	2268.2537	2268.1314	LYTEGYQVPPGATYTAIEAPK	351-371
			1196.723	1195.7157	1195.6349	APGFAHLAGLDK	393-404
Complex I-39 kDa subunit (P34943)	13	103 (45%)	1826.095	1825.0877	1824.9734	SSVSGIVATVFGATGFLGR	14-32
			957.589	956.5817	956.5192	YVVNHLGR	33-40
			1479.922	1478.9147	1478.8205	AVEHSSVVINLVGR	84-97
			1932.987	1931.9797	1931.8578	EWETQNFDFEDVFVK	98-112
			1407.854	1406.8467	1406.767	FIHISHLNADIK	129-140
			1919.148	1918.1407	1918.02	ETFPEATIIKPAEIFGR	158-174
			1157.682	1156.6747	1156.6029	FLNYFANIR	178-186
			1273.797	1272.7897	1272.7231	WFGGVPLISLGK	187-198
			1282.789	1281.7817	1281.7041	GIINAIKDPDAR	213-224
			981.574	980.5667	980.508	TFAFVGPSR	227-235
			1622.923	1621.9157	1621.814	LFEISPFEPWTTR	269-281
			2254.414	2253.4067	2253.262	ILPHLPGLEDLGVEATPLELK	293-313
			1501.849	1500.8417	1500.746	WLSSEIEDVQPAK	326-338

Table S1 (continued)

Protein name (Swiss Prot accession No.)	Matched peptides	MOWSE score[a] (Sequence coverage)	Matched in-gel triptic digests				
			Observed *m/z* (MH^+)	Mr		Peptide sequence	Residues
				Expected	Calculated		
Complex I-PSST subunit (P42026)	14	69 (23%)	1115.603	1114.5957	1114.5771	K.LDDLINWAR	33-41
			715.285	714.2777	714.2643	YDMDR + Oxidation (M)	67-71
			430.264	429.2567	429.2336	ASPR	78-81
			658.376	657.3687	657.3632	MAPALR	96-101
			674.37	673.3627	673.3581	MAPALR + Oxidation (M)	96-101
			1262.641	1261.6337	1261.6125	KVYDQMPEPR	102-111
			1278.636	1277.6287	1277.6074	KVYDQMPEPR + Oxidation (M)	102-111
			1134.547	1133.5397	1133.5175	VYDQMPEPR	103-111
			1150.534	1149.5267	1149.5125	VYDQMPEPR + Oxidation (M)	103-111
			507.231	506.2237	506.1907	GCDR	135-138
			416.351	415.3437	415.2907	IKR	167-169
			432.269	431.2617	431.2492	EKR	170-172
			444.359	443.3517	443.2968	RLR	172-174
			637.362	636.3547	636.3384	IWYR	175-178
Complex I-ND1 subunit (P03887)	7	51 (15%)	979.544	978.5367	978.5321	VLGYMQLR	27-34
			1107.639	1106.6320	1106.6270	VLGYMQLRK	27-35
			2050.167	2049.1601	2049.1622	KGPNVVGPYGLLQPIADAIK	35-54
			1922.077	1921.0701	1921.0673	GPNVVGPYGLLQPIADAIK	36-54
			876.526	875.5187	875.5229	YALIGALR	127-134
			1649.860	1648.8535	1648.8548	FRYDQLMHLLWK	280-291
			1346.698	1345.6908	1345.6853	YDQLMHLLWK	282-291

[a]MOWSE score is -10*logP, where P is the probability that the observed match is a random event. Scores greater than 54 are significant ($p<0.05$).