1. Chemistry

1.1. General

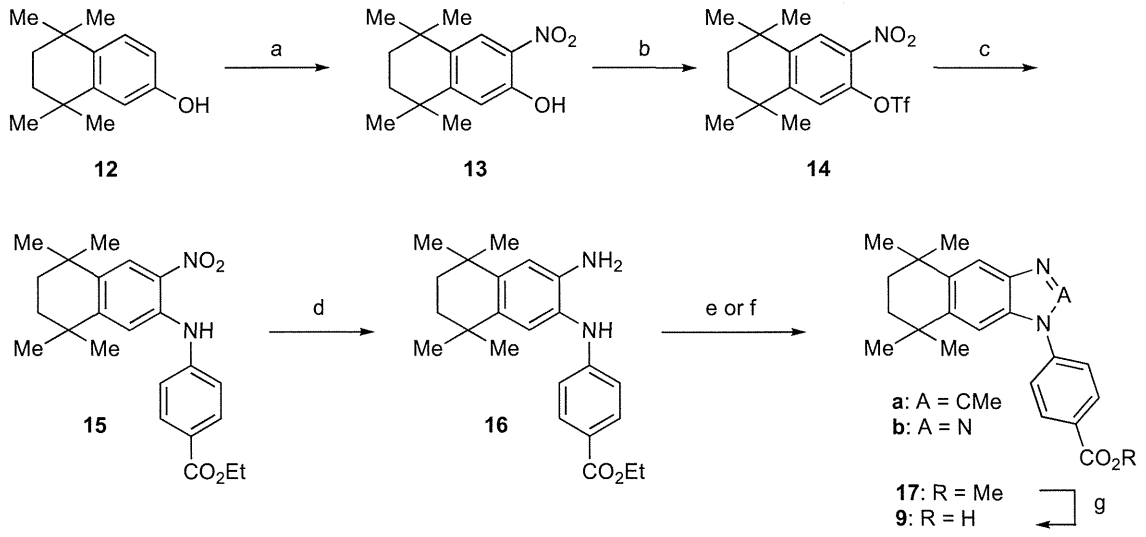
Melting points were determined with a Yanagimoto hot-stage melting point apparatus and are uncorrected. IR spectra were recorded on a JASCO FT/IR350 (KBr). ^{1}H-NMR spectra were recorded on a JEOL JNM-AL300 FT-NMR system (300 MHz) spectrometer, a VarianVXR-300 (300 MHz) or a VarianVXR-500 (500 MHz) spectrometer. Elemental analysis was carried out with a Yanagimoto MT-5 CHN recorder elemental analyzer and results were within ± 0.4% of the theoretical values. FAB-MS was carried out with a VG70-SE.

1.2. Purity determination of compounds by HPLC

Purity of compounds was determined by means of HPLC with a Shimadzu liquid chromatographic system (Kyoto, Japan) consisting of a LC-10AD pump, SPD-10AV UV-Vis spectrophotometric detector, CTO-10AS column oven and C-R5A Chromatopac. The chromatographic analyses were carried out on an Inertsil ODS-3 column (4.6 i.d. x 250 mm, 5 μm, GL Sciences, Tokyo, Japan) with a guard column of Inertsil ODS-3 (4.6 i.d. x 10 mm, 5 μm, GL Sciences) kept at 40°C, using methanol : 25 mM ammonium acetate (adjusted with acetic acid to pH 5.0) (80:20 v/v) as the mobile phase. The flow rate was 0.7 mL/min and the absorbance at 280 nm was monitored.

1.3. Schemes

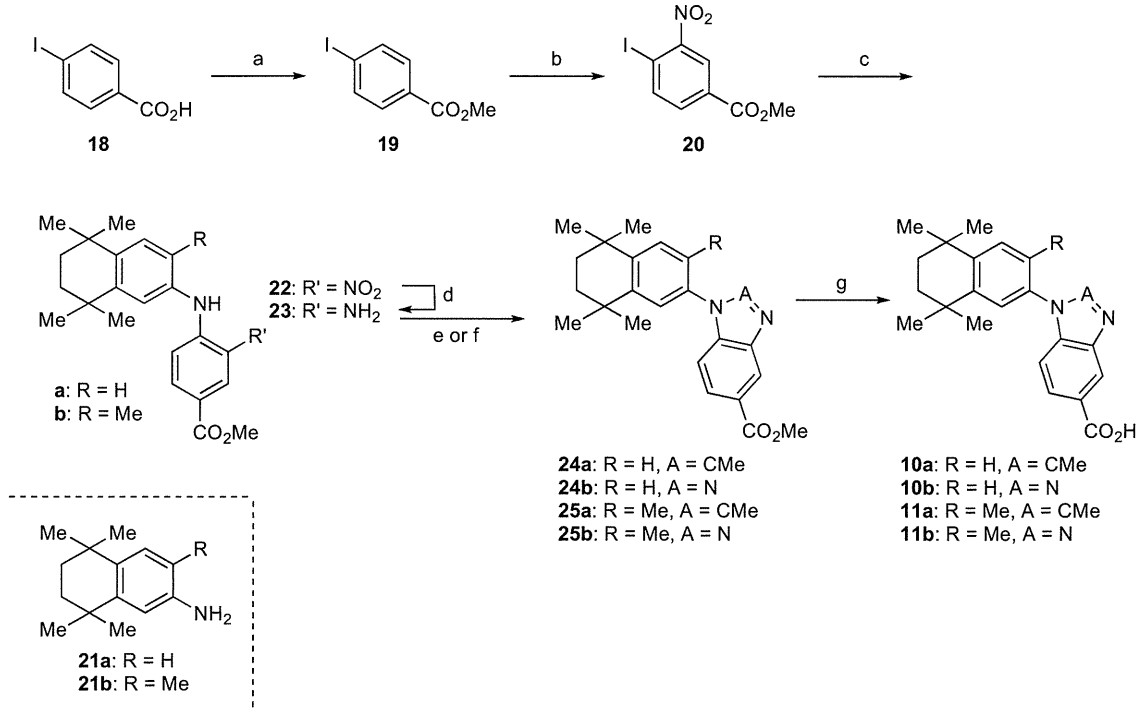
Scheme S1.[a]



[a]**Reagents and conditions:** a) *conc.* HNO_3, CH_2Cl_2, 0°C to rt. b) Tf_2O, pyridine, 0°C to rt. c) Ethyl 4-aminobenzoate, $Pd_2(dba)_3$, (±)-BINAP, Cs_2CO_3, dioxane, 100°C. d) H_2, Pd/C, EtOAc, EtOH, rt. e)

1) Ac_2O, AcOH, rt. 2) *p*-TsOH-H_2O, pyridine, dioxane, 110°C. f) $NaNO_2$, *conc.* H_2SO_4, THF, H_2O, 0°C to rt. g) 1) NaOH, MeOH, 60°C. 2) HCl.

Scheme S2.[a]

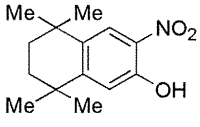


[a]**Reagents and conditions:** a) MeOH, $SOCl_2$, 0-70°C. b) *conc.* HNO_3, *conc.* H_2SO_4, 0°C to rt. c) **21a** or **21b**, $Pd_2(dba)_3$, (±)-BINAP, Cs_2CO_3, toluene, 110°C. d) H_2, Pd/C, EtOAc, rt. e-1) 1)Ac_2O, AcOH, rt. 2) *p*-TsOH-H_2O, pyridine, dioxane, 120°C. e-2) 1)Ac_2O, AcOH, rt. 2) *p*-TsOH-H_2O, dioxane, 120°C. f) $NaNO_2$, H_2SO_4, THF, H_2O, 0°C. g) 1) NaOH, MeOH, 60°C. 2) HCl.

1.4. Compound data

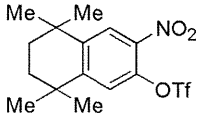
4-[1-(3,5,5,8,8-Pentamethyl-5,6,7,8-tetrahydronaphthalen-2-yl)ethenyl]benzoic acid (**1**)
This compound was prepared according to reference S1.

6-[Ethyl(5,5,8,8-tetramethyl-5,6,7,8-tetrahydronaphthalen-2-yl)amino]nicotinic acid (**5**)
This compound was prepared according to reference S2.



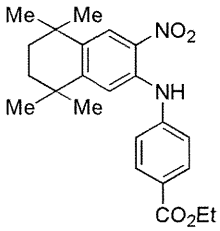
5,5,8,8-Tetramethyl-3-nitro-5,6,7,8-tetrahydronaphthalen-2-ol (**13**).
To an ice-cooled solution of **12** (2.0 g, 9.8 mmol) in CH_2Cl_2 (22 mL) was added *conc.* HNO_3 (0.75 mL) at 0°C. The mixture was stirred at r.t. under an Ar

atmosphere for 30 min, then poured onto ice and extracted with EtOAc (50 mL × 2). The organic layer was washed with H_2O (50 mL × 2) and brine (50 mL), dried over $MgSO_4$, filtered and evaporated under reduced pressure. The residue was recrystallized from MeOH to yield **13** as a pale yellow powder (640 mg, 26%). ^{1}H-NMR (300 MHz, $CDCl_3$) : δ 10.28 (s, 1H), 8.02 (s, 1H), 7.05 (s, 1H), 1.69 (s, 4H), 1.29 (s, 12H).



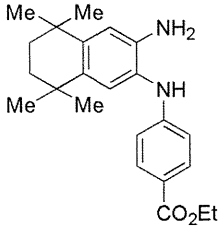
Trifluoromethanesulfonic acid 5,5,8,8-tetramethyl-3-nitro-5,6,7,8-tetrahydronaphthalen-2-yl ester (**14**).

To a solution of **13** (330 mg, 1.3 mmol) in dry pyridine (2.6 mL) was added trifluoromethanesulfonic anhydride (240 mL, 1.5 mmol) at 0°C. The mixture was stirred at r.t. under an Ar atmosphere for 9.0 hr, then poured into 2 mol/L HCl (30 mL) and extracted with EtOAc (50 mL × 3). The organic layer was washed with H_2O (30 mL × 2) and brine (20 mL), dried over $MgSO_4$, filtered and evaporated under reduced pressure. The residue was purified by flash column chromatography (EtOAc : *n*-hexane = 1 : 40) to yield **14** as a yellow powder (470 mg, 94%). ^{1}H-NMR (500 MHz, $CDCl_3$) : δ 8.10 (s, 1H), 7.27 (s, 1H), 1.73 (s, 4H), 1.33 (s, 6H), 1.30 (s, 6H).



4-(5,5,8,8-Tetramethyl-3-nitro-5,6,7,8-tetrahydronaphthalen-2-ylamino)-benzoic acid ethyl ester (**15**).

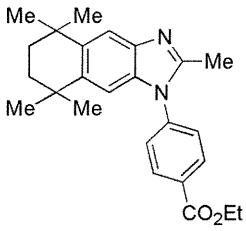
To a solution of **14** (740 mg, 1.9 mmol) and ethyl *p*-aminobenzoate (480 mg, 2.9 mmol) in dry dioxane (20 mL) were added $Pd_2(dba)_3$ (88 mg, 0.097 mmol), (±)-BINAP (90 mg, 0.14 mmol) and Cs_2CO_3 (940 mg, 2.9 mmol). The mixture was refluxed at 100°C for 30 min and filtered through Celite. The filtrate was evaporated under reduced pressure. The residue was purified by flash column chromatography (EtOAc : *n*-hexane = 1 : 60) to yield **15** as a red powder (600 mg, 78%). ^{1}H-NMR (300 MHz, $CDCl_3$) : δ 9.19 (s, 1H), 8.14 (s, 1H), 8.05 (d, J = 9.0 Hz, 2H), 7.43 (s, 1H), 7.25 (d, J = 9.0 Hz, 2H), 4.38 (q, J = 7.0 Hz, 2H), 1.70 (s, 4H), 1.40 (t, J = 7.0 Hz, 3H), 1.31 (s, 6H), 1.23 (s, 6H).



4-(3-Amino-5,5,8,8-tetramethyl-5,6,7,8-tetrahydronaphthalen-2-ylamino)-benzoic acid ethyl ester (**16**).

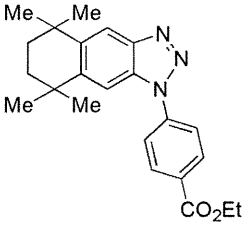
To a solution of **15** (200 mg, 0.50 mmol) in EtOAc (4.0 mL) and EtOH (1 mL) was added Pd/C (catalytic amount). The mixture was stirred at r.t. under an H_2 atmosphere overnight and filtered through Celite. The filtrate was evaporated under reduced pressure to yield **16** as a red oil (180 mg, 97%). ^{1}H-NMR (300 MHz, $CDCl_3$) : δ 7.87

(d, J = 9.0 Hz, 2H), 7.05 (s, 1H), 6.73 (s, 1H), 6.66 (d, J = 9.0 Hz, 2H), 5.68 (s, 1H), 4.31 (q, J = 7.0 Hz, 2H), 3.62 (br s, 2H), 1.65 (s, 4H), 1.35 (t, J = 7.0 Hz, 3H), 1.27 (s, 6H), 1.20 (s, 6H).



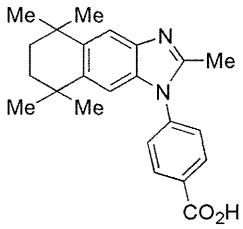
4-(2,5,5,8,8-Pentamethyl-5,6,7,8-tetrahydronaphtho[2,3-*d*]imidazol-1-yl)benzoic acid ethyl ester (**17a**).

To a solution of **16** (90 mg, 0.25 mmol) in AcOH (2.5 mL) was added Ac_2O (230 mL, 2.5 mmol). The mixture was stirred at r.t. for 40 min, poured into H_2O (20 mL) and extracted with EtOAc (40 mL × 2). The organic layer was washed with H_2O (20 mL) and brine (20 mL), dried over $MgSO_4$, filtered and evaporated under reduced pressure. The residue was purified by flash column chromatography (EtOAc : *n*-hexane = 1 : 5) to yield 4-(3-acetylamino-5,5,8,8-tetramethyl-5,6,7,8-tetrahydronaphthalen-2-ylamino)benzoic acid ethyl ester (90 mg). To a solution of this compound (90 mg, 0.22 mmol) in dioxane (2.2 mL) were added *p*-TsOH·H_2O (47 mg, 0.25 mmol) and dry pyridine (20 mL, 0.25 mmol). The mixture was refluxed at 110°C for 2.0 hr, poured into H_2O (30 mL) and extracted with EtOAc (30 mL × 2). The organic layer was washed with H_2O (30 mL) and brine (20 mL), dried over $MgSO_4$, filtered and evaporated under reduced pressure. The residue was purified by flash column chromatography (EtOAc : *n*-hexane = 1 : 10) to yield **17a** as a colorless powder (51 mg, 59%). ^{1}H-NMR (300 MHz, $CDCl_3$) : δ 8.28 (d, J = 8.5 Hz, 2H), 7.71 (s, 1H), 7.46 (d, J = 8.5 Hz, 2H), 7.07 (s, 1H), 4.45 (q, J = 7.0 Hz, 2H), 2.50 (s, 3H), 1.73 (s, 4H), 1.45 (t, J = 7.0 Hz, 3H), 1.37 (s, 6H), 1.26 (s, 6H).



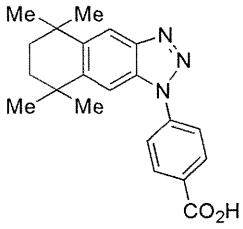
4-(5,5,8,8-Tetramethyl-5,6,7,8-tetrahydronaphtho[2,3-*d*][1,2,3]triazol-1-yl)benzoic acid ethyl ester (**17b**).

To an ice-cooled solution of **16** (86 mg, 0.24 mmol) in THF (6.3 mL) was added *conc*. H_2SO_4 (1.0 mL). Then, a solution of $NaNO_2$ (18 mg, 0.26 mmol) in H_2O (1.0 mL) was added dropwise. The mixture was stirred at r.t. for 1.5 hr, poured into 2 mol/L NaOH (10 mL) and extracted with EtOAc (30 mL × 2). The organic layer was washed with H_2O (40 mL) and brine (30 mL), dried over $MgSO_4$, filtered and evaporated under reduced pressure. The residue was purified by flash column chromatography (EtOAc : *n*-hexane = 1: 20) to yield **17b** as an orange powder (87 mg, 98%). ^{1}H-NMR (300 MHz, $CDCl_3$) : δ 8.31 (d, J = 9.0 Hz, 2H), 8.12 (s, 1H), 7.90 (d, J = 9.0 Hz, 2H), 7.70 (s, 1H), 4.45 (q, J = 7.0 Hz, 2H), 1.79 (s, 4H), 1.45(t, J = 7.0 Hz, 3H), 1.41 (s, 6H), 1.39 (s, 6H).



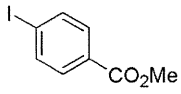
4-(2,5,5,8,8-Pentamethyl-5,6,7,8-tetrahydronaphtho[2,3-*d*]imidazol-1-yl)-benzoic acid (**9a**).

To a solution of **17a** (51 mg, 0.13 mmol) in MeOH (1.3 mL) was added 2 mol/L NaOH (1.0 mL). The mixture was stirred at 60°C for 40 min, poured into 2 mol/L HCl (3 mL) and extracted with EtOAc (30 mL × 3). The organic layer was washed with H_2O (30 mL) and brine (20 mL), dried over $MgSO_4$, filtered and evaporated under reduced pressure to yield **9a** as a colorless solid (41 mg, 88%). The residue was recrystallized from MeOH to yield **9a** as a colorless powder (6.9 mg). Mp: 290.0-293.0°C; HPLC: 13.03 min. 96.6% purity; FAB-MS *m/z*: 363 $[M+H]^+$. ^{1}H-NMR (500 MHz, DMSO-d_6) : δ 8.26 (d, J = 8.5 Hz, 2H), 7.82 (s, 1H), 7.78 (d, J = 8.5 Hz, 2H), 7.26 (s, 1H), 2.51 (s, 3H), 1.70 (s, 4H), 1.36 (s, 6H), 1.23 (s, 6H).



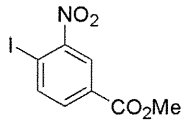
4-(5,5,8,8-Tetramethyl-5,6,7,8-tetrahydronaphtho[2,3-*d*][1,2,3]triazol-1-yl)benzoic acid (**9b**).

To a solution of **17b** (87 mg, 0.23 mmol) in MeOH (2.0 mL) was added 2 mol/L NaOH (1.0 mL). The mixture was stirred at 60°C for 2.5 hr, poured into 2 mol/L HCl (3 mL) and extracted with EtOAc (40 mL × 2). The organic layer was washed with H_2O (30 mL) and brine (20 mL), dried over $MgSO_4$, filtered and evaporated under reduced pressure to yield **9b** as a colorless solid (80 mg, q.y.). The residue was recrystallized from MeOH to yield **9b** as orange needles (33 mg, 41%). Mp: 275.0-279.0°C; HPLC: 13.50 min. 98.4% purity; ; FAB-MS *m/z*: 350 [M+H]+. ^{1}H-NMR (500 MHz, DMSO-d_6) : δ 13.2 (br s, 1H), 8.23 (d, J = 8.5 Hz, 2H), 8.18 (s, 1H), 8.03 (d, J = 8.5 Hz, 2H), 7.88 (s, 1H), 1.74 (s, 4H), 1.38 (s, 12H).



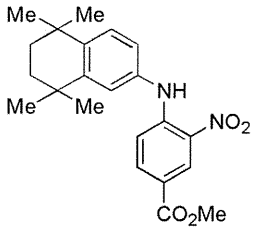
4-Iodobenzoic acid methyl ester (**19**).

To an ice-cooled solution of 4-iodobenzoic acid (5.0 g, 20 mmol) in MeOH (30 mL) was added $SOCl_2$ (2.6 mL, 30 mmol). The mixture was refluxed at 70°C for 1.0 hr, then evaporated with toluene to remove excess $SOCl_2$. The residue was dissolved in EtOAc (150 mL). The solution was washed with *sat.* $NaHCO_3$ (100 mL × 2) and brine (60 mL), dried over $MgSO_4$, filtered and evaporated under reduced pressure. The residue was recrystallized from CH_2Cl_2/*n*-hexane to yield **19** as colorless needles (4.7 g, 91%). ^{1}H-NMR (500 MHz, $CDCl_3$) : δ 7.80 (d, J = 8.0 Hz, 2H), 7.74 (d, J = 8.0 Hz, 2H), 3.91 (s, 3H).



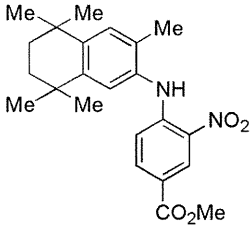
4-Iodo-3-nitrobenzoic acid methyl ester (**20**).

To an ice-cooled solution of **19** (1.3 g, 5.0 mmol) in *conc.* H_2SO_4 (5.0 mL) was added dropwise a solution of *conc.* HNO_3 (6.0 mL) and *conc.* H_2SO_4 (9.0 mL). The mixture was stirred at r.t. for 5.0 hr, then poured onto ice (100 mL), and extracted with EtOAc (60 mL × 2). The organic layer was washed with *sat.* $NaHCO_3$ (50 mL × 2) and brine (50 mL), dried over $MgSO_4$, filtered and evaporated under reduced pressure. The residue was recrystallized from CH_2Cl_2/*n*-hexane to yield **20** as yellow needles (1.2 g, 76%). ^{1}H-NMR (500 MHz, $CDCl_3$) : δ 8.45 (d, J = 2.0 Hz, 1H), 8.15 (d, J = 8.0 Hz, 1H), 7.88 (dd, J = 8.0, 2.0 Hz, 1H), 3.97 (s, 3H).



3-Nitro-4-[*N*-(5,5,8,8-tetramethyl-5,6,7,8-tetrahydro-2-naphthyl)amino]benzoic acid methyl ester (**22a**).

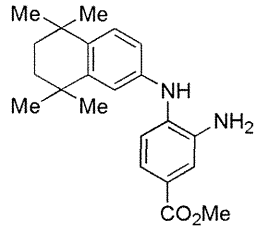
To a solution of **20** (1.1 g, 3.5 mmol) and **21a** (0.70 g, 3.5 mmol) (reference S3) in dry toluene (21 mL) were added $Pd_2(dba)_3$ (160 mg, 0.17 mmol), (±)-BINAP (160 mg, 0.26 mmol) and Cs_2CO_3 (1.6 g, 4.9 mmol). The mixture was refluxed at 110°C under an Ar atmosphere for 5.0 hr, then filtered through Celite. The filtrate was washed with H_2O (40 mL × 2) and brine (30 mL), dried over $MgSO_4$, filtered and evaporated under reduced pressure. The residue was purified by flash column chromatography (EtOAc : *n*-hexane = 1 : 5) and crystallized from *n*-hexane to yield **22a** as red needles (1.2 g, 76%). ^{1}H-NMR (500MHz, $CDCl_3$) : δ 9.77 (br s, 1H), 8.91 (d, J = 2.0 Hz, 1H), 7.94 (dd, J = 9.0, 2.0 Hz, 1H), 7.37 (d, J = 8.0 Hz, 1H), 7.18 (d, J = 2.0 Hz, 1H), 7.16 (d, J = 9.0 Hz, 1H), 7.04 (dd, J = 8.0, 2.0 Hz, 1H), 3.91 (s, 3H), 1.72 (s, 4H), 1.31 (s, 6H), 1.28 (s, 6H).



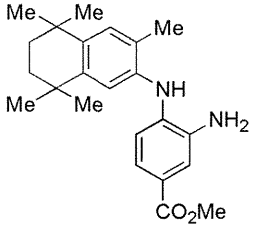
3-Nitro-4-[*N*-(3,5,5,8,8-pentamethyl-5,6,7,8-tetrahydro-2-naphthyl)amino]benzoic acid methyl ester (**22b**).

To a solution of **20** (0.92 g, 3.0 mmol) and **21b** (0.65 g, 3.0 mmol) in dry toluene (30 mL) were added $Pd_2(dba)_3$ (140 mg, 0.15 mmol), (±)-BINAP (140 mg, 0.22 mmol) and Cs_2CO_3 (1.4 g, 4.2 mmol). The mixture was refluxed at 110°C under an Ar atmosphere overnight, then filtered through Celite. The filtrate was evaporated under reduced pressure. The residue was purified by flash column chromatography (EtOAc : *n*-hexane = 1 : 10) to yield **22b** as a red foam (1.1 g, 94%). ^{1}H-NMR (300 MHz, $CDCl_3$) : δ 9.63 (br s, 1H), 8.93 (d, J = 2.0 Hz, 1H), 7.93 (dd, J = 9.0, 2.0 Hz, 1H), 7.24 (s, 1H), 7.17 (s, 1H), 6.82 (d, J = 9.0 Hz, 1H), 3.91 (s, 3H), 2.19 (s, 3H) 1.70 (s, 4H), 1.31 (s, 6H), 1.25 (s, 6H).

3-Amino-4-[*N*-(5,5,8,8-tetramethyl-5,6,7,8-tetrahydro-2-naphthylamino)benzoic acid methyl ester (**23a**).

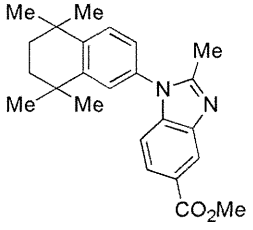


To a solution of **22a** (760 mg, 2.0 mmol) in EtOAc (10 mL) was added Pd/C (catalytic amount). The mixture was stirred at r.t. under an H_2 atmosphere overnight, then filtered through Celite. The filtrate was evaporated under reduced pressure. The residue was recrystallized from CH_2Cl_2/*n*-hexane to yield **23a** as colorless needles (660 mg, 94%). ^{1}H-NM (500 MHz, $CDCl_3$) : δ 7.48 (s, 1H), 7.48 (d, J = 9.0 Hz, 1H), 7.23 (d, J = 8.5 Hz, 1H), 7.14 (d, J = 9.0 Hz, 1H), 6.94 (s, 1H), 6.81 (d, J = 8.5 Hz, 1H), 5.52(s, 1H), 3.87 (s, 3H), 3.57(br s, 2H), 1.69 (s, 4H), 1.28 (s, 6H), 1.26 (s, 6H).



3-Amino-4-[*N*-(3,5,5,8,8-pentamethyl-5,6,7,8-tetrahydro-2-naphthyl)amino]benzoic acid methyl ester (**23b**).

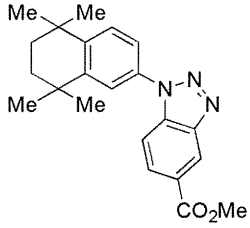
To a solution of **22b** (1.1 g, 2.8 mmol) in EtOAc (20 mL) was added Pd/C (catalytic amount). The mixture was stirred at r.t. under an H_2 atmosphere for 1.0 hr, then filtered through Celite. The filtrate was evaporated under reduced pressure to yield **23b** as a white solid (1.0 g, 97%). ^{1}H-NMR (300 MHz, $CDCl_3$) : δ 7.49 (s, 1H), 7.48 (d, J = 8.0 Hz, 1H), 7.13 (s, 1H), 6.96 (s, 1H), 6.83 (d, J = 8.0 Hz, 1H), 5.39 (br s, 1H), 3.87 (s, 3H), 3.55 (br s, 2H), 2.19 (s, 3H), 1.67 (s, 4H), 1.28 (s, 6H), 1.21 (s, 6H).



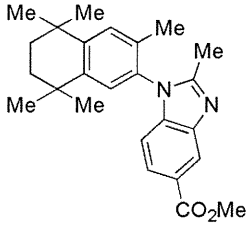
2-Methyl-1-(5,5,8,8-tetramethyl-5,6,7,8-tetrahydronaphthalen-2-yl)-1*H*-benzoimidazole-5-carboxylic acid methyl ester (**24a**).

To a solution of **23a** (180 mg, 0.50 mmol) in AcOH (5.0 mL) was added Ac_2O (0.50 mL). The mixture was stirred at r.t. for 20 min, then poured into ice-cooled 2 mol/L NaOH (50 mL) and extracted with EtOAc (30 mL × 2). The organic layer was washed with H_2O (40 mL × 2) and brine (30 mL), dried over $MgSO_4$, filtered and evaporated under reduced pressure. The residue was recrystallized from CH_2Cl_2/*n*-hexane to yield colorless needles (180 mg). To a solution of the residue (160 mg) in dioxane were added *p*-TsOH·H_2O (76 mg, 0.40 mmol) and pyridine (32 mL, 0.40 mmol). The solution was refluxed at 120°C for 5.0 hr, then poured into H_2O (80 mL) and extracted with EtOAc (30 mL × 2). The organic layer was washed with H_2O (40 mL × 2) and brine (30 mL), dried over $MgSO_4$, filtered and evaporated under reduced pressure. The residue was re-crystallized from CH_2Cl_2/*n*-hexane to yield **24a** as colorless needles (110 mg, 65%). ^{1}H-NMR (500 MHz, $CDCl_3$) : δ 8.44 (s, 1H), 7.93 (d, J = 8.0 Hz, 1H), 7.48 (d, J = 8.0 Hz, 1H), 7.26 (s, 1H), 7.17 (d, J = 8.0 Hz, 1H), 7.10 (d, J = 8.0 Hz, 1H), 3.95 (s, 3H), 2.54 (s, 3H), 1.76 (s, 4H), 1.36 (s, 6H), 1.31 (s, 6H).

1-(5,5,8,8-Tetramethyl-5,6,7,8-tetrahydro-2-naphthyl)-1*H*-benzotriazole-5-carboxylic acid methyl ester (**24b**).

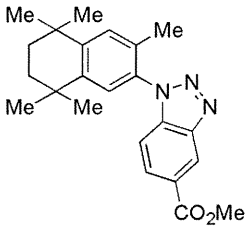


To an ice-cooled solution of **23a** (180 mg, 0.5 mmol) in THF (3.0 mL) was added a mixture of *conc.* H_2SO_4 (1.0 mL) and H_2O (10 mL). Then, a solution of $NaNO_2$ (48 mg, 0.7 mmol) in H_2O (1.0 mL) was added dropwise. The mixture was stirred at 0°C for 20 min, poured into H_2O (30 mL) and extracted with EtOAc (30 mL × 2). The organic layer was washed with H_2O (40 mL) and brine (30 mL), dried over $MgSO_4$, filtered and evaporated under reduced pressure. The residue was purified by flash column chromatography (EtOAc : *n*-hexane = 1: 3) to yield **24b** as an orange solid (180 mg, 99 %). ^{1}H-NMR (500 MHz, $CDCl_3$) : δ 8.87 (s, 1H), 8.23 (d, J = 9.0 Hz, 1H), 7.73 (d, J = 8.0 Hz, 1H), 7.66 (d, J = 2.0 Hz, 1H), 7.55 (d, J = 9.0 Hz, 1H), 7.49 (d, J = 8.0, 2.0 Hz, 1H), 4.00 (s, 3H), 1.77 (s, 4H), 1.36 (s, 12H).



2-Methyl-1-(3,5,5,8,8-pentamethyl-5,6,7,8-tetrahydro-2-naphthyl)-1*H*-benzoimidazole-5-carboxylic acid methyl ester (**25a**).

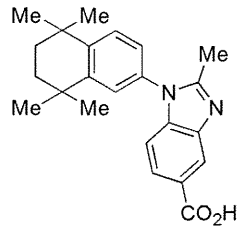
To a solution of **23b** (150 mg, 0.4 mmol) in AcOH (5.0 mL) was added Ac_2O (0.5 mL). The mixture was stirred at r.t. for 15 min, then poured into ice-cooled *sat.* $NaHCO_3$ (70 mL) and extracted with EtOAc (50 mL × 2). The organic layer was washed with *sat.* $NaHCO_3$ (70 mL), dried over $MgSO_4$, filtered and evaporated under reduced pressure. The residue (170 mg) and *p*-TsOH·H_2O (190 mg, 1.0 mmol) were dissolved in dioxane (5.0 mL). The solution was refluxed at 120°C overnight, then poured into H_2O (40 mL) and extracted with EtOAc (30 mL × 2). The organic layer was washed with H_2O (50 mL) and brine (30 mL), dried over $MgSO_4$, filtered and evaporated under reduced pressure to yield **25a** as a brown solid (150 mg, 98%). ^{1}H-NMR (500 MHz, $CDCl_3$) : δ 8.46 (d, J = 1.5 Hz, 1H), 7.92 (dd, J = 8.5, 1.5 Hz, 1H), 7.30 (s, Ar-H, 1H), 7.12 (s, 1H), 6.98 (s, 1H), 3.95 (s, 3H), 2.42 (s, 3H), 1.90 (s, 3H), 1.74 (s, 4H), 1.36 (s, 6H), 1.26 (s, 6H).



1-(3,5,5,8,8-Pentamethyl-5,6,7,8-tetrahydro-2-naphthyl)-1*H*-benzotriazole-5-carboxylic acid methyl ester (**25b**).

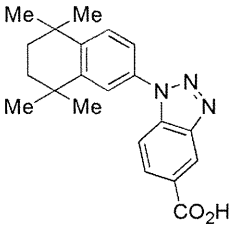
To an ice-cooled solution of **23b** (150 mg, 0.40 mmol) in THF (2.0 mL) was added a mixture of *conc.* H_2SO_4 (1.0 mL) and H_2O (10 mL). Then, a solution of $NaNO_2$ (41 mg, 0.60 mmol) in H_2O (2.0 mL) was added dropwise. The reaction mixture was refluxed at 0°C for 30 min, poured into H_2O (20 mL), and extracted with EtOAc (30 mL × 2). The organic layer was washed with H_2O (40 mL) and brine (30 mL), dried over $MgSO_4$, filtered and evaporated under reduced pressure. The residue was purified by flash column chromatography (EtOAc : *n*-hexane = 1 : 1). The product was again purified by flash column chromatography (EtOAc : *n*-hexane = 1 : 3) to yield **25b** as an orange solid (140 mg, 91 %). ^{1}H-NMR (500 MHz, $CDCl_3$) : δ 8.87 (d, J = 1.5 Hz, 1H), 8.19 (dd, J = 8.5, 1.5 Hz,

1H), 7.40 (d, J = 8.5 Hz, 1H), 7.35 (s, 1H), 7.29 (s, 1H), 4.00 (s, 3H), 2.07 (s, 3H), 1.75 (s, 4H), 1.36 (s, 6H), 1.29 (s, 6H).



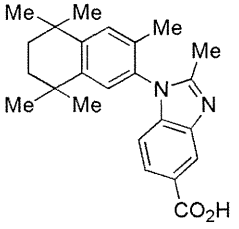
2-Methyl-1-(5,5,8,8-tetramethyl-5,6,7,8-tetrahydro-2-naphthyl)-1*H*-benzoimidazole-5-carboxylic acid (**10a**).

To a solution of **24a** (87 mg, 0.23 mmol) in MeOH (10 mL) was added 2 mol/L NaOH (10 mL). The mixture was stirred at 60°C for 20 min, poured into 1 mol/L HCl (20 mL) and extracted with EtOAc (30 mL × 3). The organic layer was washed with H_2O (60 mL) and brine (40 mL), dried over $MgSO_4$, filtered and evaporated under reduced pressure to yield **10a** as a white solid (65 mg, 78%). Mp > 295°C; ^{1}H-NMR (500 MHz, DMSO-d_6) : δ 12.71 (br s, 1H), 8.19 (s, 1H), 7.82 (d J = 8.5 Hz, 1H), 7.58 (d, J = 8.5 Hz, 1H), 7.51 (s, 1H), 7.29 (d, J = 8.0 Hz, 1H), 7.19 (d, J = 8.0 Hz, 1H), 2.47 (s, 3H), 1.71 (s, 4H), 1.33 (s, 6H), 1.29 (s, 6H); IR (KBr) : ν =2960 (OH), 1698 (CO) cm^{-1}; FAB-MS *m/z*: 363 $[M+H]^+$.



1-(5,5,8,8-Tetramethyl-5,6,7,8-tetrahydro-2-naphthyl)-1*H*-benzotriazole-5-carboxylic acid (**10b**).

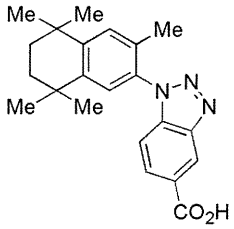
To a solution of **24b** (180 mg, 0.5 mmol) in MeOH (10 mL) was added 2 mol/L NaOH (8.0 mL). The mixture was stirred at 60°C for 15 min, poured into 1 mol/L HCl (20 mL), and extracted with EtOAc (30 mL × 2). The organic layer was washed with H_2O (40 mL × 2) and brine (30 mL), dried over $MgSO_4$, filtered and evaporated under reduced pressure to yield **10b** as an orange solid (170 mg, q.y.). Mp: 260.0-261.0°C; ^{1}H-NMR (300 MHz, DMSO-d_6) : δ 13.39 (br s, 1H), 8.72 (d, J = 1.5 Hz, 1H), 8.19 (d, J = 9.0, 1.5 Hz, 1H), 7.90 (d, J = 9.0 Hz, 1H), 7.77 (d, J = 2.0 Hz, 1H), 7.67 (d, J = 8.5 Hz, 1H), 7.60 (d, J = 8.5, 2.0 Hz, 1H), 1.73 (s, 4H), 1.34 (s, 12H) ; IR (KBr): ν = 2961-2933 (OH), 1713 (CO) cm^{-1}; FAB-MS *m/z*: 350 $[M+H]^+$.



2-Methyl-1-(3,5,5,8,8-pentamethyl-5,6,7,8-tetrahydro-2-naphthyl)-1*H*-benzoimidazole-5-carboxylic acid (**11a** : CBiM-PMN).

To a solution of **25a** (150 mg, 0.4 mmol) in MeOH (10 mL) was added 2 mol/L NaOH (10 mL). The mixture was stirred at 60°C for 20 min, poured into 1 mol/L HCl (20 mL) and extracted with EtOAc (30 mL × 2). The organic layer was washed with H_2O (40 mL) and brine (30 mL), dried over $MgSO_4$, filtered and evaporated under reduced pressure to yield **11a** as a brown solid (140 mg, 93%). The residue was recrystallized from MeOH to yield a colorless powder (84 mg). Mp: 295.0°C; ^{1}H-NMR (300 MHz, DMSO-d_6) : δ 12.71 (br s, 1H), 8.20 (s, 1H), 7.81 (d J = 8.5 Hz, 1H), 7.47 (s, 1H), 7.36

(s, 1H), 6.94 (d, J = 8.5 Hz, 1H), 2.32 (s, 3H), 1.85 (s, 3H), 1.69 (s, 4H), 1.33 (s, 6H), 1.25 (s, 3H), 1.24 (s, 3H); IR (KBr): ν = 2957-2925 (OH), 1701 (CO) cm^{-1}; FAB-MS *m/z*: 377 $[M+H]^+$.



1-(3,5,5,8,8-Pentamethyl-5,6,7,8-tetrahydro-2-naphthyl)-1*H*-benzotriazole-5-carboxylic acid (**11b**: CBt-PMN).

To a solution of **25b** (140 mg, 0.36 mmol) in MeOH (6.0 mL) was added 2 mol/L NaOH (6.0 mL). The mixture was stirred at 60°C for 15 min, poured into 1 mol/L HCl (12 mL), and extracted with EtOAc (20 mL × 2). The organic layer was washed with H_2O (20 mL) and brine (20 mL), dried over $MgSO_4$, filtered and evaporated under reduced pressure to yield **11b** as a pale yellow solid (120 mg, 86%). The residue was recrystallized from EtOAc/*n*-hexane as colorless needles. Mp: 238.0-239.0°C; ^{1}H-NMR (300 MHz, DMSO-d_6) : δ 13.26 (1H, br s), 8.72 (1H, d, J = 1.5 Hz), 8.14 (1H, dd, J = 8.5, 1.5 Hz) , 7.56 (1H, d, J = 8.5 Hz), 7.52 (1H, s), 7.49 (1H, s), 2.00 (3H, s), 1.71 (4H, s), 1.34 (6H, s), 1.27 (6H, s); IR (KBr): ν = 2961 (OH), 1714 (CO) cm^{-1}; FAB-MS *m/z*: 364 $[M+H]^+$.

2. Purity

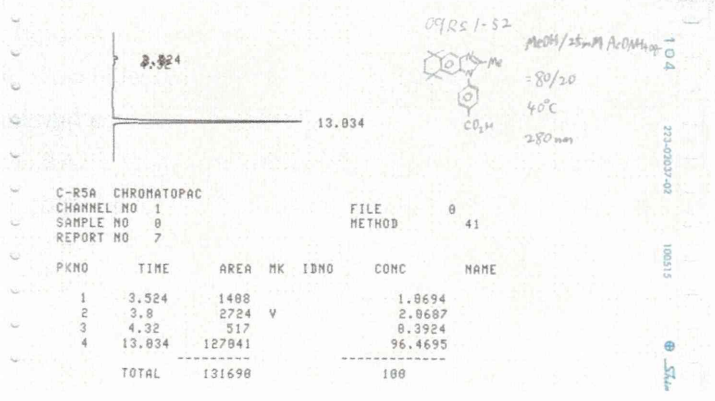
2.1. Combustion analysis data

Table S1. Combustion analysis data for compounds **10a**, **10b**, **11a**, and **11b**

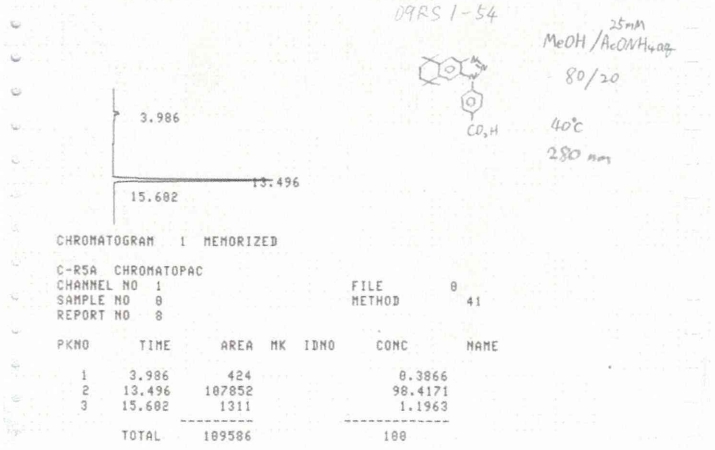
compound	Formula	Calculated			Found		
		C	H	N	C	H	N
10a	$C_{23}H_{26}N_2O_2 \cdot 5/3H_2O$	70.38	7.53	7.14	70.61	7.27	6.97
10b	$C_{21}H_{23}N_3O_2$	72.18	6.63	12.03	72.02	6.79	11.93
11a	$C_{24}H_{28}N_2O_2$	76.56	7.50	7.44	76.31	7.65	7.40
11b	$C_{22}H_{25}N_3O_2$	72.70	72.70	11.56	72.55	6.97	11.54

2.2. HPLC Charts

9a:



9b:



3. Luciferase Reporter Gene Assay

3.1. Culture of COS-1 cells

COS-1 cells were maintained in Dulbecco's modified Eagle's medium supplemented with 10% FBS in a humidified atmosphere of 5% CO_2 in air at 37°C.

3.2. Luciferase reporter gene assay

Luciferase reporter gene assays were performed using COS-1 cells transfected with three kinds of vectors: each receptor subtype, a luciferase reporter gene under the control of the appropriate RXR response element, and secreted alkaline phosphatase (SEAP) gene as a background. CRBPII-tk-Luc, tk-TREPII-Luc, tk-PPREx3-Luc, and tk-rBARx3-Luc reporters were used as RXR, RAR, PPAR, and LXR response elements, respectively. The amounts of each receptor subtype and response element were 1.0 μg and 4.0 μg, respectively. Transfection was performed with QIA Effectene Transfection reagent according to the supplier's protocol. In the case of heterodimer assay, RXRα (0.5 μg), each partner receptor (PPARγ or LXRα, 0.5 μg) and the partner response element (4.0 μg) were transfected into COS-1 cells as described above. Test compound solutions (DMSO concentration below 1%) were added to the suspension of transfected cells, which were seeded at about 2.0×10^4 cells/well in 96-well white plates. After incubation in a humidified atmosphere of 5% CO_2 at 37°C for 18 h, 25 μL of the medium was used for analyzing SEAP activities and the remaining cells were used for luciferase reporter gene assays with a Steady-Glo Luciferase Assay system (Promega) according to the supplier's protocol. The luciferase activities were normalized using SEAP activities. The assays were carried out in triplicate three times.

3.3 Supplementary data 1

Table S2. Transactivation of RXRα by **5**, **9a**, **9b**, **10a**, **10b**, **11a** and **11b** in COS-1 cells[a]

Compound	EC_{50} (nM)[b]	E_{max} (%)	Efficacy at 1 μM (%)	Efficacy at 10 μM (%)
5	3.8 ± 0.2	100 ± 4	96 ± 4	100 ± 4
9a	n.d.[c]	n.d.	1 >[d]	6 ± 2
9b	n.d.	n.d.	1 >	15 ± 2
10a	n.d.	n.d.	23 ± 5	82 ± 1
10b	n.d.	n.d.	5 ± 1	63 ± 7
11a	367 ± 130	94 ± 5	70 ± 4	80 ± 5
11b	143 ± 2	75 ± 3	75 ± 3	68 ± 3

a. COS-1 cells were transfected with three kinds of vectors: RXRα, a luciferase reporter gene under the control of the appropriate RXR response element (CRBPII-tk-Luc), and secreted alkaline phosphatase (SEAP) gene as a background. Data are mean ± s.e.m. Data are the mean values of at

least two separate experiments with triplicate determinations. Luciferase activity of **1** at 1 μM was defined as 100 percent.

b. EC_{50} values were determined from full dose-response curves in COS-1 cells.

c. n.d. means "not determined"

d. 1 > means "less than 1%"

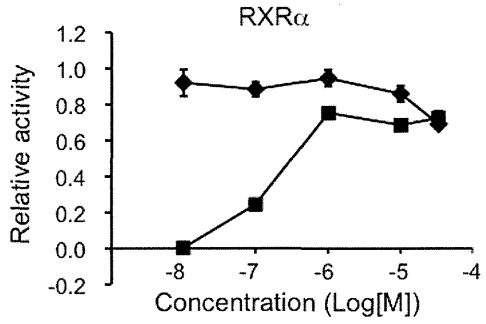


Figure S1. Dose-dependence of RXRα-agonistic activities of CBt-PMN (**11b**) (squares) and RXRα-antagonistic activity of CBt-PMN (**11b**) in the presence of 1 μM LGD1069 (**1**) (diamonds). COS-1 cells were transfected with three kinds of vectors: each RXR receptor subtype, a luciferase reporter gene under the control of the appropriate RXR response element (CRBPII-tk-Luc), and secreted alkaline phosphatase (SEAP) gene as a background. The transactivation activity is shown as relative activity based on the luciferase activity of 1 μM LGD1069 (**1**) taken as 1.0. Error bars are s.e.m.

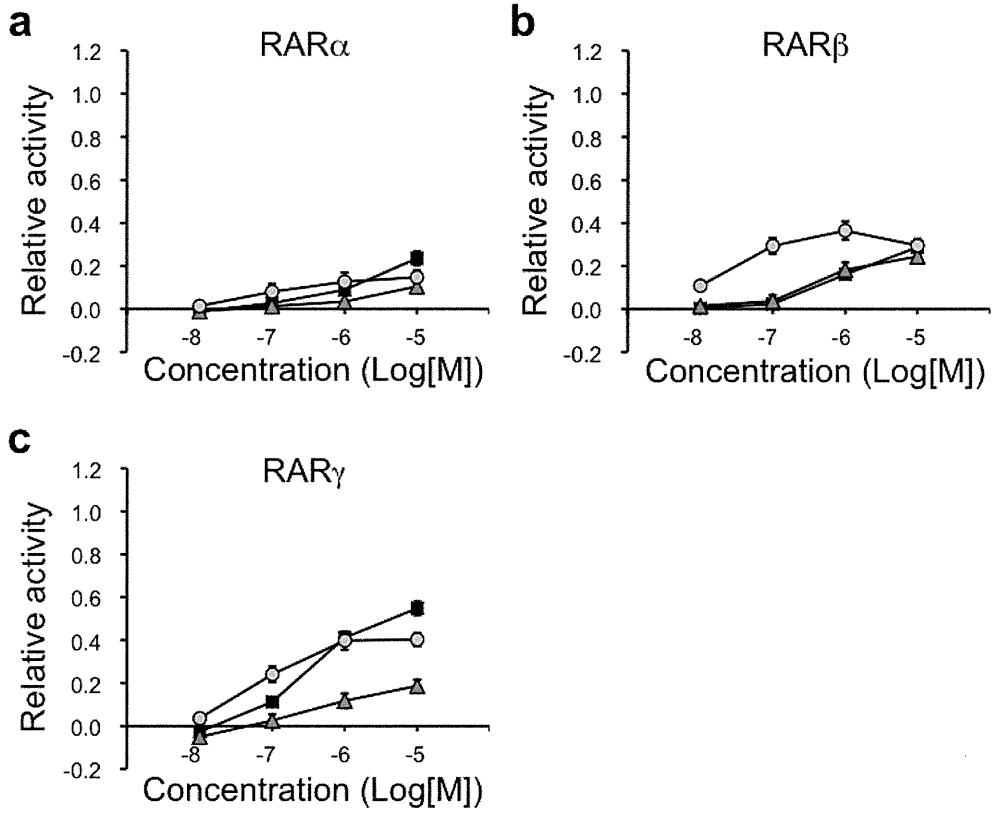


Figure S2. Relative transactivation activities toward RARα, RARβ and RARγ by NEt-TMN (**5**), CBiM-PMN (**11a**), and CBt-PMN (**11b**). COS-1 cells were transfected with three kinds of vectors: each RAR receptor subtype, a luciferase reporter gene under the control of the appropriate RAR response element (tk-TREPII-Luc), and secreted alkaline phosphatase (SEAP) gene as a background. The data are relative activity with respect to the luciferase activity of 1 μM Am80 (RARα/β selective agonist) (reference S3) (toward RARα/β) or ATRA (toward RARγ) taken as 1.0. Circles, triangles, and squares indicate NEt-TMN (**5**), CBiM-PMN (**11a**), and CBt-PMN (**11b**), respectively. The data (n = 3) represent the mean ± s.e.m.

4. In vivo assay

4.1. Measurement of plasma concentration of test compounds after oral administration of 30 mg/kg to mice

Groups of six-week-old ICR male mice (n = 5-9 in each) were treated with solutions of test compound 30 mg/kg (1% ethanol and 0.5% CMC in distilled water) at a volume of 10 mL/kg of body weight by oral administration. At the indicated times, 0.6 mL of blood was taken from the inferior vena cava under diethyl ether anesthesia. Each blood sample was centrifuged at 9,000 rpm for 5 min at r.t. To 100 μL of the resulting plasma were added 100 μL of ice-cold 5 mM ammonium acetate solution (adjusted with acetic acid to pH 5.0) and 1 mL of ice-cold ethyl acetate. The resulting mixture was vortexed for 30 sec, kept at room temperature for 10 min, and centrifuged at 9,000 rpm for 30 sec at room temperature. An 800 μL aliquot of the ethyl acetate phase was removed and concentrated to dryness in a centrifugal evaporator. To the resulting residue was added 100 μL of HPLC-grade methanol. This solution was directly subjected to HPLC analysis, and the concentration of each compound was determined from the peak area of the sample with reference to a calibration plot obtained with the authentic compound.

4.2. HPLC conditions

The HPLC system used in this study was a Shimadzu liquid chromatographic system (Kyoto, Japan) consisting of an SCL-10A system controller, LC-10AD pump, SPD-10AV UV-Vis spectrophotometric detector, SIL-10AD autoinjector, CTO-10A column oven, DGU-14A degasser and C-R7A Chromatopac. The samples (each 20 μL) were injected using a refrigerated autosampler kept at 10°C. The chromatographic analyses were carried out on an Inertsil ODS-3 (4.6 i.d. x 250 mm, 5 μm, GL Sciences, Tokyo, Japan) kept at 40°C, using methanol : 33.3 mM ammonium acetate (adjusted with acetic acid to pH 5.0) (85:15, v/v) as a mobile phase. The flow rate was 0.7 mL/min and the absorbance at 280 nm was monitored.

4.3. Observation of side effects after once-daily oral administration of 30 mg/kg for 7 consecutive days in male ICR mice

Six-week-old male ICR mice were purchased from Charles River Laboratories Japan, Inc.. This experiment was conducted in accordance with the Guidelines for Animal Experiments at Okayama University Advanced Science Research Center, and all procedures were approved by the Animal Research Control Committee of Okayama University. After the arrival of the animals, all were group-housed and acclimated to the colony for 1 day before the experiment. Before the experiment, they were housed with four mice per cage, with free access to water and chow pellets in a light (12 hr on, 8:00 A.M. /12 hr off, 8:00 P.M.), temperature (23 ± 1°C), and relative humidity (50 ± 20%)-controlled environment. At 1 day before experiments, mice were assigned to experimental groups to minimize the variance between groups based on the measured weight (four per cage (17 x 33 x 15

cm)). Their body weights were measured at approximately 10:00 A.M. every day for 7 days before dosing. Mice were administered orally with a solution of compounds at a dose of 30 mg/kg or with the vehicle (1% ethanol and 0.5% carboxymethyl cellulose (CMC) in distilled water) at a volume of 10 mL/kg of animal at approximately 10:00 A.M. every day for 7 days. At the final day of dosing, animals were fasted from 17:00 P.M. and given water *ad libitum*. On the next day at approximately 10:00 A.M., animals were weighed and anesthetized with diethyl ether. Blood and liver were removed immediately, and the liver weight was measured. Approximately 1 mL of blood in an Eppendorf sample tube was centrifuged to afford the serum sample. Each blood sample was centrifuged at 9,000 rpm for 5 min at r.t..

4.4. Observation of side effects after once-daily oral administration of 30 mg/kg for 28 consecutive days in SD rats

Five-week-old male and female SD rats were purchased from Charles River Laboratories Japan, Inc.. This experiment was conducted in accordance with the Guidelines for Animal Experiments at Okayama University Advanced Science Research Center, and all procedures were approved by the Animal Research Control Committee of Okayama University. After the arrival of the rats, all were group-housed and acclimated to the colony for 1 day before the experiment. Before the experiment, they were housed with three rats per cage, with free access to water and chow pellets in a light (12 hr on, 8:00 AM /12 hr off, 8:00 PM), temperature (23 ± 1°C), and relative humidity (50 ± 3%)-controlled environment. At 1 day before experiments, rats were assigned to experimental groups to minimize the variance between groups based on the measured weight (three per cage (34.5 x 40.3 x 17.7 cm)). Their body weights were measured at approximately 10:00 A.M. every day for 28 days before dosing. Rats were administered orally with a solution of compounds at a dose of 30 mg/kg or with the vehicle (1% ethanol and 0.5% carboxymethyl cellulose (CMC) in distilled water) at a volume of 5 mL/kg of animal at approximately 10:00 A.M. every day for 28 days. At the final day of dosing, animals were fasted from 17:00 P.M. and given water *ad libitum*. On the next day at approximately 10:00 A.M., animals were weighed and anesthetized with diethyl ether (maintained with isoflurane). Blood, liver, brain, kidney, spleen, and testis (male only) were removed immediately. The liver, brain, kidney, spleen, and testis were weighed and frozen with liquid nitrogen. Approximately 10 mL of blood treated with heparin in a centrifuge tube was centrifuged at 2,000*g* for 10 min at 4°C to obtain a plasma sample.

4.5. Evaluation of blood glucose-lowering activities in KK-A^y mice

Type 2 diabetic male KK-A^y mice, in which the A^y allele at the agouti locus was transferred to the inbred KK strain by repetitive back-crossing, were used as the congenic strain. The introduction of the A^y allele caused DM and massive hereditary obesity. The KK-A^y mice (CLEA Japan Inc., Tokyo, Japan) were allowed free access to solid food and tap water. This experiment was conducted in

accordance with the Guidelines for Animal Experiments at Kyoto Pharmaceutical University (KPU), and all procedures were approved by the Experimental Animal Research Committee at KPU. The animals were housed in an air-conditioned room at a temperature of 23 ± 1 °C and a humidity of 60 ± 10%, with lights on from 8:00 AM to 20:00 PM. KK-A^y mice were randomly divided into compound-treated and untreated groups. After the measurment of body weight and their blood glucose levels, the mice received daily oral administrations of compounds at 10 mg/kg/day dissolved in polyethylene glycol 400 (PEG) at approximately 10:00 AM for 14 days. Samples for measurements of fed blood glucose level were taken from the tail vein of the mice, and glucose was measured by using the glucose oxidase method (Glucocard, Arkray, Kyoto, Japan).

4.6. Oral glucose tolerance test (OGTT)

KK-A^y mice treated with each compound for 14 days were fasted for 12 h, and administered orally with a glucose solution (1 M) at a dose of 1 g/kg body weight. At 0, 15, 30, 45, 60, 90 and 120 min after the glucose loading, blood glucose level was measured as described above.

4.7. Measurements of serum or plasma parameters

White blood cells (WBC), red blood cells (RBC), hemoglobin (HGB), hematocrit (HCT), mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC) and platelets (PLT) were measured by using a pocH-100i (Sysmex). Alanine aminotransferase (AST), aspartate aminotransferase (ALT), γ-glutamyl transpeptidase (γ-GTP), alkaline phosphatase (ALP), creatinine (CRE), blood urea nitrogen (BUN), triglyceride (TG), total cholesterol (TCHO), and glucose (GLU) were measured by using a Fuji Dry Chem system (Dry Chem 4000V, Fuji Medical Co., Tokyo, Japan).

4.8. Measurements of metabolic parameters

Blood samples for analysis of glucose level were obtained from the tail vein of the mice and serum glucose levels were measured using Glucocard. Serum concentrations of total choresterol (TCHO) and triglyceride (TG) were determined by a Fuji Dry Chem system (Fuji Medical Co., Tokyo, Japan) as described above. The level of hemoglobin A1C (HbA1C) was measured 24 hr after the final administration of compounds by an immunoassay method (DCA2000). Serum levels of insulin were determined with Glazyme insulin-EIA test (Wako Pure Chemicals Co., Osaka, Japan). Serum levels of adiponectin were determined with Adiponectin immunoassay kit (R&D Systems Inc., Minneapolis, MN, USA).

4.9. RNA Preparation and quantitative real-time RT-PCR

Fifty milligrams of liver tissue from KK-A^y mice described above was resected and mechanically homogenized with a Politron PT 10/35 (Kinematica Inc., Littau-Luzern, Switzerland) in 0.5 mL of Trizol reagent (Invitrogen, Carlsbad, CA). Total RNA was extracted as previously described (reference S4). Quantitative real-time RT-PCR analysis was performed using a LightCycler rapid thermal cycler system (Roche Applied Science, Mannheim, Germany) following the protocol previously reported (reference S5). Two micrograms of total RNA was reverse-transcribed by random hexamer priming using ReverTra Ace (Toyobo, Osaka, Japan). The PCR mixture consisted of 1x SYBR Green PCR Master Mix (Toyobo), which includes DNA polymerase, SYBR Green I Dye, dNTPs, PCR buffer, 10 pmol forward and reverse primers and cDNA of samples in a total volume of 20 μL. The amplification of a housekeeping gene, *Rps18*, was used to normalize the efficiency of cDNA synthesis and the amount of RNA applied. PCR was performed with initial denaturation at 94°C for 30 sec, followed by amplification for 40 cycles, each cycle consisting of denaturation at 95°C for 5 sec, annealing at 60°C for 15 sec, and polymerization at 72°C for 15 sec. Primers used for this study are listed in Table S5.

4.10. Supplementary data 2

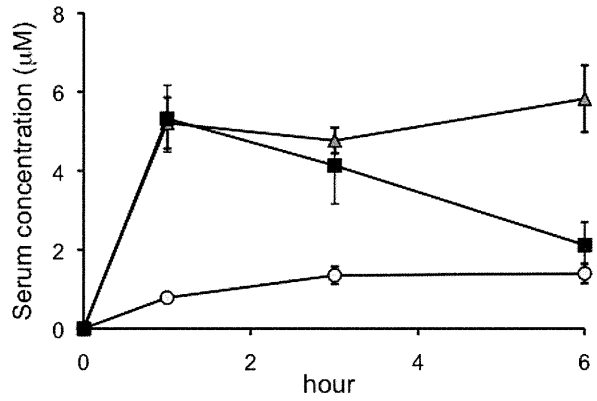


Figure S3. Serum concentrations of NEt-TMN (**5**), CBiM-PMN (**11a**) and CBt-PMN (**11b**) in mice after single oral administration of 30 mg/kg. Circles, triangles, and squares indicate NEt-TMN (**5**), CBiM-PMN (**11a**), and CBt-PMN (**11b**), respectively. The data (n = 5–9) represent the mean ± s.e.m.

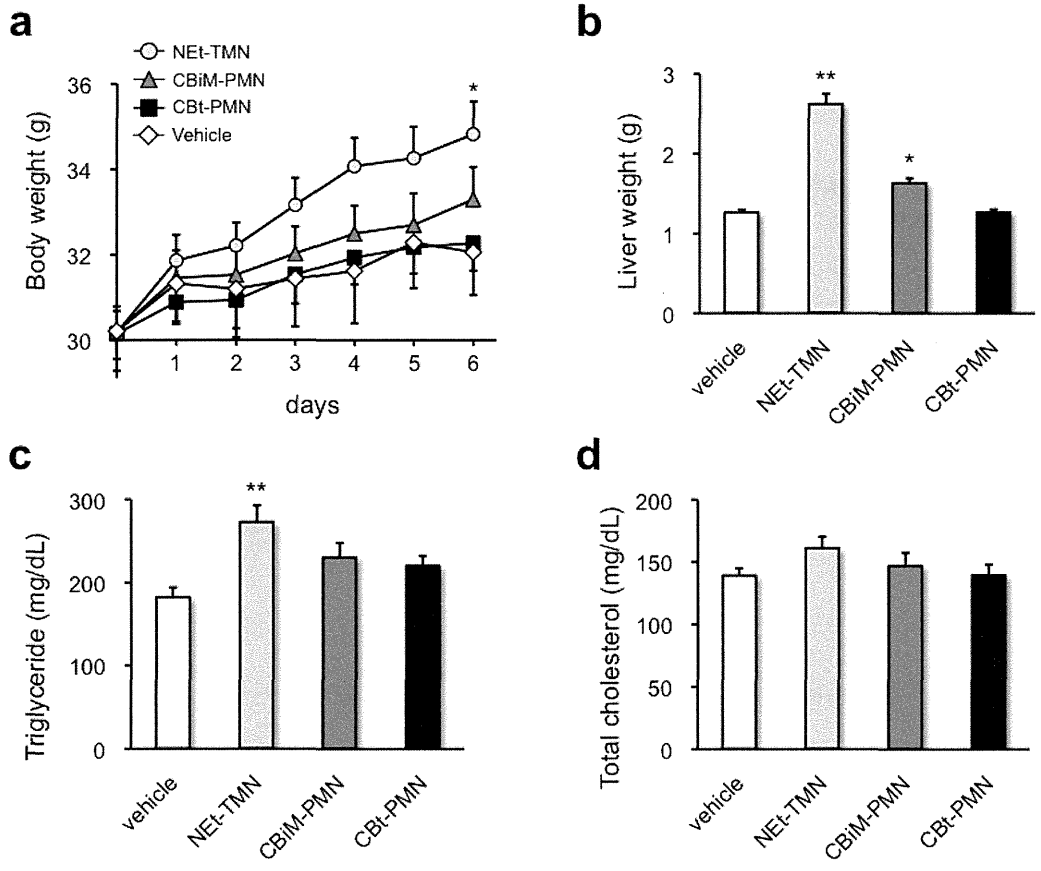


Figure S4. Evaluation of adverse effects of repeated oral administration of compounds at 30 mg/kg/day to male ICR mice for 7 consecutive days. **a**) Time course of body weight. Circles, triangles, and squares indicate NEt-TMN (**5**), CBiM-PMN (**11a**), and CBt-PMN (**11b**), respectively. **b**–**d**) Effects of compounds on liver weight gain, serum triglyceride and total cholesterol, respectively. The data (n = 7–23) represent the mean ± s.e.m. Statistical analysis was performed by analysis of variance (ANOVA). Significant differences: * $p < 0.05$ vs. vehicle. ** $p < 0.01$ vs. vehicle.