Synthesis and Evaluation of 2-Nonylaminopyridine Derivatives as PPAR Ligands

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To find novel PPAR ligands, we prepared several 3-{3 or 4-[2-(nonylpyridin-2-ylamino)ethoxy]phenyl}-propanoic acid derivatives which were designed based on the structure of our previous PPARγ ligand 1. In PPAR binding affinity assays, compound 4, which had an ethoxy group at the C-2 position of the propanoic acid of 1, showed selective binding affinity for PPARγ. Compound 3, with an ethyl group at the C-2 position, was found to be a PPARα/γ dual ligand. Compound 6, the meta isomer of 1, has been shown to be a PPARα ligand. The introduction of methyl (7) and ethyl (8) groups to the C-2 position of the propanoic acid of 6 further improved PPARα-binding potency. In cell-based transactivation assay, compounds 3 and 4 showed dual-agonist activity toward PPARα and PPARγ. Compound 6 was found to be a triple agonist and compound 8 proved to be a selective PPARα agonist. In the human hypodermic preadipocyte differentiation test, it was demonstrated that the maximal activity of compounds 3 and 4 was higher than that of rosiglitazone.

Key words peroxisome proliferator-activated receptor (PPAR); agonist; selectivity

The peroxisome proliferator-activated receptors (PPARα, PPARγ and PPARδ) are a set of ligand-activated transcription factors in the nuclear hormone receptor superfamily.[1—4] These receptors regulate the expression of a large number of genes involved in lipid metabolism and energy balance by binding to a DNA sequence termed PPAR response elements.[5] The PPARγ is predominantly expressed in adipose tissues, and plays a pivotal role in adipose differentiation, and the regulation of glucose and lipid homeostasis. The clinically useful thiazolidinedione (TZD) class of insulin sensitizers such as rosiglitazone[6] and pioglitazone[7] (Fig. 1) are potent PPARγ agonists used in the treatment of Type 2 diabetes. TZDs are known to improve insulin resistance, which is a key underlying feature of Type 2 diabetes[8]; however, the use of TZDs has been limited because of their serious side effects such as hepatic toxicity, weight gain and edema. Meanwhile, PPARα is highly expressed in metabolically active tissues such as the liver, heart and muscle, and regulates lipid homeostasis. PPARα agonists such as clofibrate (Fig. 1) have demonstrated the ability to reduce serum triglyceride and increase HDL cholesterol levels,[9] and are being utilized as hypolipidemic agents. In addition, recent studies revealed that dual agonists of PPARα/γ decrease the free triglyceride plasma concentration and increase plasma HDL concentration in an insulin-resistant animal model.[10,11] Thus, many groups have ongoing research programs to identify more potent and less toxic PPARα agonists, PPARγ agonists and PPARα/γ dual agonists.

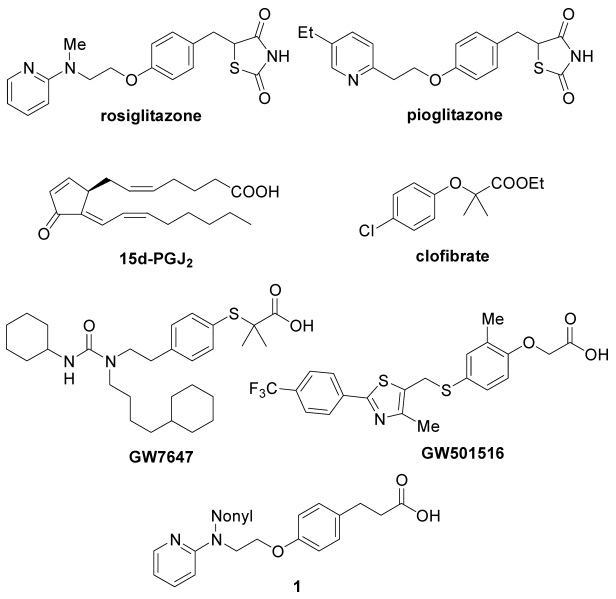

Fig. 1. Structures of Rosiglitazone, Pioglitazone, 15d-PGJ$_2$, Clofibrate, GW7647, GW501516 and Compound **1**

We previously reported compound **1** (Fig. 1), which was designed based on the structure of rosiglitazone and 15d-PGJ$_2$,[12,13] as a potent PPARγ ligand.[14] To find more potent PPARγ agonists and novel PPARα agonists, we chose compound **1** as the lead structure, because recent reports indicated that PPARγ affinity can be increased by the introduction of substituents into the C-2 position of propanoic acid,[15—19] and minor structural modifications can convert PPAR subtype selectivity.[20—22] In this article, we report the synthesis, binding affinity and biological activity of PPAR ligands based on the structure of compound **1**.

Chemistry The compounds prepared for this study are shown in Fig. 2, and the routes used for synthesis are shown in Charts 1—3. Chart 1 shows the preparation of compounds **1**, **3**, **4**, **6**—**10**, **12** and **13**. The 2-nonylaminopyridine **18** was prepared by the method of Buchwald[23]: treatment of *n*-nonylamine **17** with 2-bromopyridine, $Pd_2(DBA)_3$, BINAP, and *t*-BuONa in toluene at 105 °C. *p*-Hydroxybenzaldehyde **19a,** *m*-hydroxybenzaldehyde **19b** and isovaniline **19c** were allowed to react with 1,2-dibromoethane to give ethers **20a**—**c**. The Horner–Wadsworth–Emmons reaction[24] was applied to the conversion of **20a**—**c** into acrylic acid ethyl esters **21a**—

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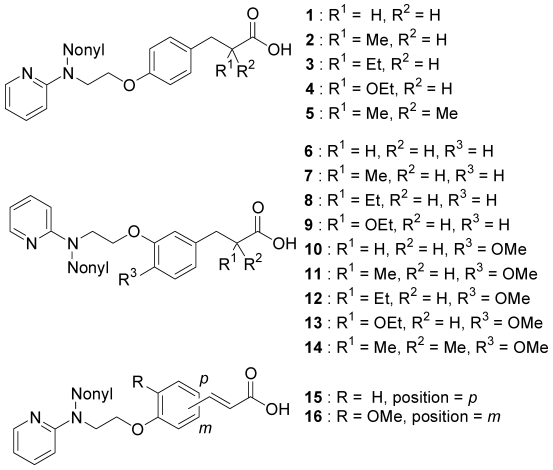
j. The double bonds of **21a**—**j** were hydrogenated to yield compounds **22a**—**j**. Coupling between 2-nonylaminopyridine **18** and propanoic acid ethyl esters **22a**—**j** afforded *N*-(2-pyridinyl)-*N*-nonylpropanoic acid ethyl esters **23a**—**j**. The subsequent hydrolysis of **23a**—**j** gave the desired carboxylic acids **1**, **3**, **4**, **6**—**10**, **12** and **13**.



1 : R^1 = H, R^2 = H
2 : R^1 = Me, R^2 = H
3 : R^1 = Et, R^2 = H
4 : R^1 = OEt, R^2 = H
5 : R^1 = Me, R^2 = Me

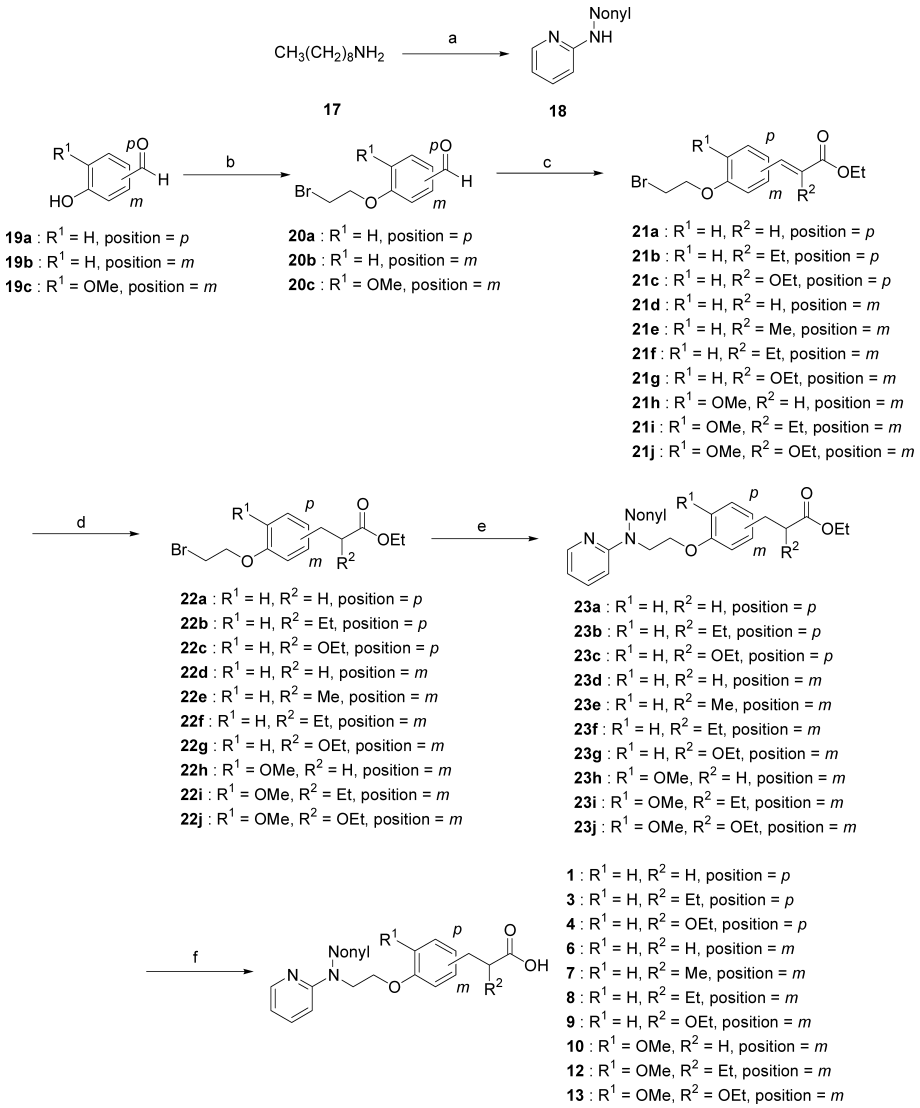
6 : R^1 = H, R^2 = H, R^3 = H
7 : R^1 = Me, R^2 = H, R^3 = H
8 : R^1 = Et, R^2 = H, R^3 = H
9 : R^1 = OEt, R^2 = H, R^3 = H
10 : R^1 = H, R^2 = H, R^3 = OMe
11 : R^1 = Me, R^2 = H, R^3 = OMe
12 : R^1 = Et, R^2 = H, R^3 = OMe
13 : R^1 = OEt, R^2 = H, R^3 = OMe
14 : R^1 = Me, R^2 = Me, R^3 = OMe

15 : R = H, position = *p*
16 : R = OMe, position = *m*

Fig. 2. Structures of Compounds **1**—**16**

The preparation of compounds **2**, **5**, **11** and **14**, which have one or two methyl groups at the C-2 position of propanoic acid, is outlined in Chart 2. Aldehydes **20a** and **20c** were reduced by $NaBH_4$ and allowed to react with acetic anhydride to give **25a** and **25b**. Compounds **25a** and **25b** were treated with 1-methoxy-1-trimethylsilyloxypropene or dimethylketene methyltrimethylsilyl acetal in the presence of magnesium perchlorate in anhydrous CH_2Cl_2 to give esters **26a**—**d**.[25] Coupling between 2-nonylaminopyridine **18** and propanoic acid methyl esters **26a**—**d** afforded *N*-(2-pyridinyl)-*N*-nonyl compounds **27a**—**d** and subsequent hydrolysis gave carboxylic acids **2**, **5**, **11** and **14**.

Preparation of the acrylic acid derivatives **15** and **16** is shown in Chart 3. Acrylic acid ethyl esters **21a** and **21h** were allowed to react with 2-nonylaminopyridine **18** to give compounds **28a** and **28b**. Treatment of **28a** and **28b** with aqueous NaOH gave *N*-(2-pyridinyl)-*N*-nonyl acrylic acids **15** and **16**.



19a : R^1 = H, position = *p*
19b : R^1 = H, position = *m*
19c : R^1 = OMe, position = *m*

20a : R^1 = H, position = *p*
20b : R^1 = H, position = *m*
20c : R^1 = OMe, position = *m*

21a : R^1 = H, R^2 = H, position = *p*
21b : R^1 = H, R^2 = Et, position = *p*
21c : R^1 = H, R^2 = OEt, position = *p*
21d : R^1 = H, R^2 = H, position = *m*
21e : R^1 = H, R^2 = Me, position = *m*
21f : R^1 = H, R^2 = Et, position = *m*
21g : R^1 = H, R^2 = OEt, position = *m*
21h : R^1 = OMe, R^2 = H, position = *m*
21i : R^1 = OMe, R^2 = Et, position = *m*
21j : R^1 = OMe, R^2 = OEt, position = *m*

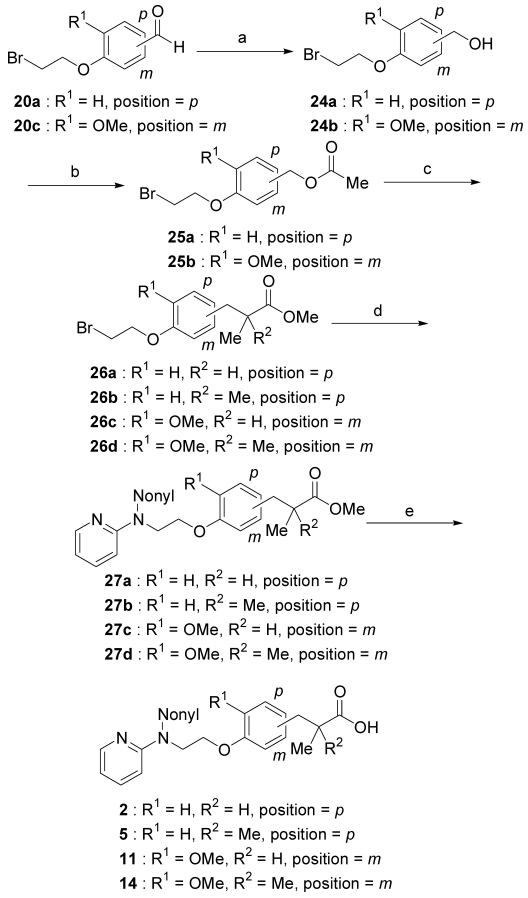
22a : R^1 = H, R^2 = H, position = *p*
22b : R^1 = H, R^2 = Et, position = *p*
22c : R^1 = H, R^2 = OEt, position = *p*
22d : R^1 = H, R^2 = H, position = *m*
22e : R^1 = H, R^2 = Me, position = *m*
22f : R^1 = H, R^2 = Et, position = *m*
22g : R^1 = H, R^2 = OEt, position = *m*
22h : R^1 = OMe, R^2 = H, position = *m*
22i : R^1 = OMe, R^2 = Et, position = *m*
22j : R^1 = OMe, R^2 = OEt, position = *m*

23a : R^1 = H, R^2 = H, position = *p*
23b : R^1 = H, R^2 = Et, position = *p*
23c : R^1 = H, R^2 = OEt, position = *p*
23d : R^1 = H, R^2 = H, position = *m*
23e : R^1 = H, R^2 = Me, position = *m*
23f : R^1 = H, R^2 = Et, position = *m*
23g : R^1 = H, R^2 = OEt, position = *m*
23h : R^1 = OMe, R^2 = H, position = *m*
23i : R^1 = OMe, R^2 = Et, position = *m*
23j : R^1 = OMe, R^2 = OEt, position = *m*

1 : R^1 = H, R^2 = H, position = *p*
3 : R^1 = H, R^2 = Et, position = *p*
4 : R^1 = H, R^2 = OEt, position = *p*
6 : R^1 = H, R^2 = H, position = *m*
7 : R^1 = H, R^2 = Me, position = *m*
8 : R^1 = H, R^2 = Et, position = *m*
9 : R^1 = H, R^2 = OEt, position = *m*
10 : R^1 = OMe, R^2 = H, position = *m*
12 : R^1 = OMe, R^2 = Et, position = *m*
13 : R^1 = OMe, R^2 = OEt, position = *m*

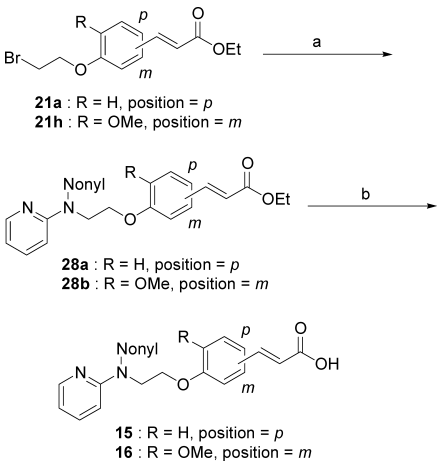
(a) 2-bromopyridine, $Pd_2(DBA)_3$, BINAP, *t*-BuOH, toluene, 105 °C, 70%; (b) 1,2-dibromoethane, Cs_2CO_3, THF, 65 °C, 33—57%; (c) $(EtO)_2P(O)CH(R)CO_2Et$, NaH, anhydrous THF, 0 °C to rt, 47—95%; (d) H_2, Pd/C, EtOH, 79—97%; (e) **18**, Et_3N, KI, 105 °C, 9—17%; (f) 2 N aq. NaOH, EtOH, THF, rt, 82—100%.

Chart 1



(a) $NaBH_4$, EtOH, rt, 81—95%; (b) Ac_2O, DMAP, rt, 96—97%; (c) 1-methoxy-1-trimethylsilyloxypropene, or dimethylketene methyltrimethylsilyl acetal, $Mg(ClO_4)_2$, rt, 90—94%; (d) **18**, Et_3N, KI, 105 °C, 5—13%; (e) 2 N aq. NaOH, EtOH, rt, 75—97%.

Chart 2



(a) **18**, Et_3N, KI, 105 °C, 9—13%; (b) 2 N aq. NaOH, EtOH, THF, rt, 82—99%.

Chart 3

Results and Discussion

The binding affinity of the compounds for PPARs was evaluated with the CoA-BAP System (Microsystems).[26] In this system, alkaline phosphatase (AP) activity is directly proportional to the affinity of the ligands for PPARs. The abilities of compounds **1**—**16** to bind PPARα, PPARγ and PPARδ were evaluated and the results are shown in Figs. 3, 4 and 5, respectively. GW7647[27] (PPARα), rosiglitazone[6] (PPARγ) and GW501516[28] (PPARδ) were used as reference compounds (Fig. 1).

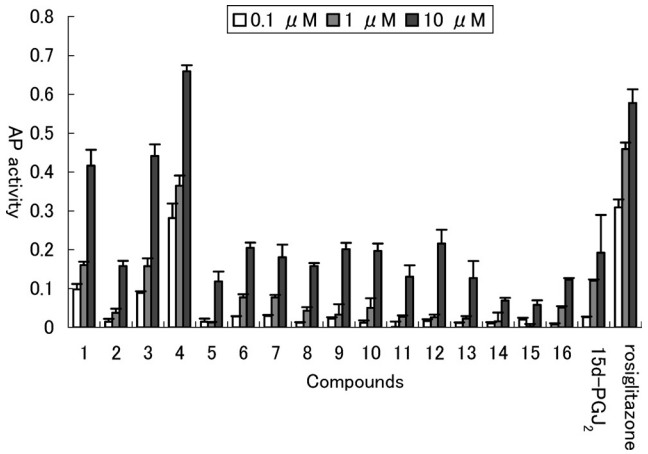


Fig. 3. Binding Affinity for PPARγ of Compounds **1**—**16** at 0.1, 1.0 and 10 μM

Values are the means of at least three experiments.

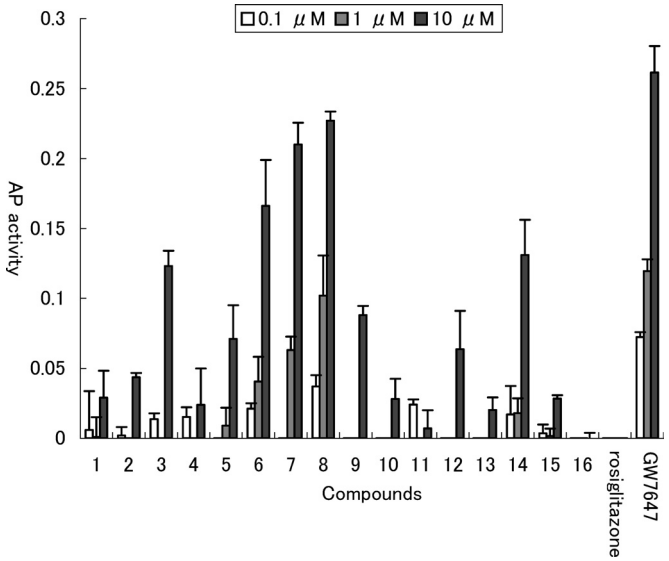


Fig. 4. Binding Affinity for PPARα of Compounds **1**—**16** at 0.1, 1.0 and 10 μM

Values are the means of at least three experiments.

The lead compound **1** showed relatively high affinity for PPARγ and little affinity for PPARα and PPARδ (Fig. 3—5). We initially examined the effect of substituents at the C-2 position of the propanoic acid of **1**, because it has been reported that the introduction of an alkyl or an alkoxy group at this position increases the activity of PPARγ and PPARα.[15—19] Among compound **1**, methyl **2**, ethyl **3**, ethoxy **4**, and dimethyl **5**, compound **4** showed the strongest affinity for PPARγ, so compound **4** was founded to be a potent and selective PPARγ ligand (Figs. 3—5). In addition, the PPARγ affinity of compound **3** was comparable to that of compound **1**, whereas compound **3** displayed strong affinity for PPARα as compared with **1** (Figs. 3, 4).

Next, we investigated the PPARs affinity of compound **6**, the *meta* isomer of compound **1**.[29] Compound **6** showed

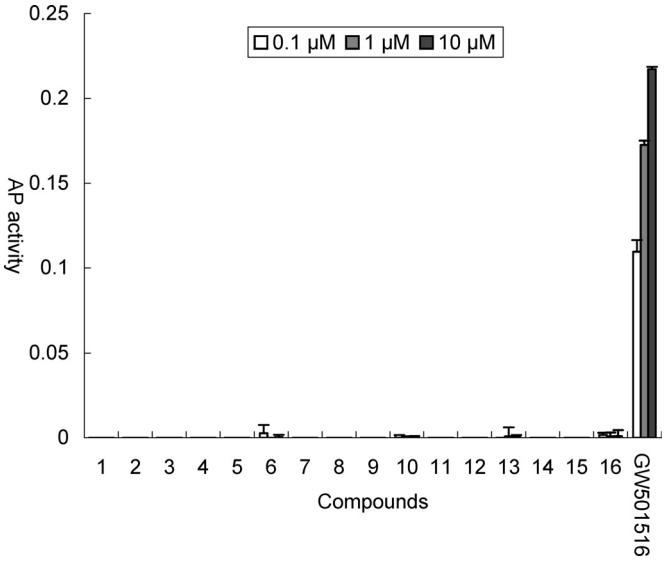


Fig. 5. Binding Affinity for PPARδ of Compounds **1**—**16** at 0.1, 1.0 and 10 μM

Values are the means of at least three experiments.

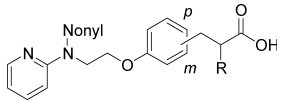
much higher affinity for PPARα than **1** (Fig. 4). Furthermore, the affinity for PPARγ of **6** was lower than that of **1** (Fig. 3), and compound **6** exhibited little affinity for PPARδ (Fig. 5). To find more potent PPARα ligands, we prepared and tested compounds **7**, **8** and **9** in which a methyl, ethyl and ethoxy group was substituted at the C-2 position of the propanoic acid of **6**, respectively. Compounds **7** and **8** showed strong affinity for PPARα and compound **8** displayed slightly weak affinity for PPARγ as compared with the parent compound **6**. The effect of the introduction of a methoxy group at the C-4 position of the benzene ring of **6** was also investigated; however, compounds **10**—**14** showed no pronounced affinity for PPARs as compared to compound **6** (Figs. 3—5).

The acrylic acid derivatives **15** and **16** did not show notable affinity for PPARs (Figs. 3—5).

Based on the findings in the binding assay, we next investigated the PPAR transactivation activity of compounds **1**, **3**, **4**, **6**, **7**, and **8** by reporter gene assay[30] (Table 1). We initially tested the activity of compound **1**, which did not show significant transcriptional activity for PPARα, δ, and γ. We then examined the activity of compounds **3** and **4**, which have an ethyl or ethoxy group at the C-2 position of the propanoic acid of **1**. As expected from the binding assay, compound **3** was a PPARα/γ dual agonist. Although the binding assay indicated that compound **4** is a selective PPARγ ligand, it showed potent dual-agonist activity toward PPARα and PPARγ. Compound **6**, the *meta* isomer of compound **1**, was found to be a triple agonist. Compound **7**, which had an introduced methyl group at the C-2 position of the propanoic acid moiety of **6**, improved the EC_{50} values and selectivity for PPARα and PPARγ, and the introduction of ethyl group (compound **8**) increased the activity and selectivity for PPARα. Among these compounds, compound **8** showed the highest selectivity for PPARα.

As compounds **3** and **4** were found to have relatively high PPARγ agonist activity in our study, we used them for further evaluation. Since it has been reported that the activation

Table 1. *In Vitro* Functional PPAR Transactivation Activity of Compounds



Compound	R	Position	EC_{50}[a] (μM)		
			PPARγ	PPARδ	PPARα
1	H	*p*	>10	>10	>10
3	Et	*p*	2.62	>10	3.68
4	OEt	*p*	0.32	>10	0.74
6	H	*m*	7.76	6.42	3.20
7	Me	*m*	4.72	>10	2.65
8	Et	*m*	>8	>10	1.81

a) Compounds were screened for agonist activity on PPAR-GAL4 chimeric receptors in transiently transfected HEK-293 cells as described. EC_{50} value is the molar concentration of the test compound that affords 50% of maximal reporter activity.[33]

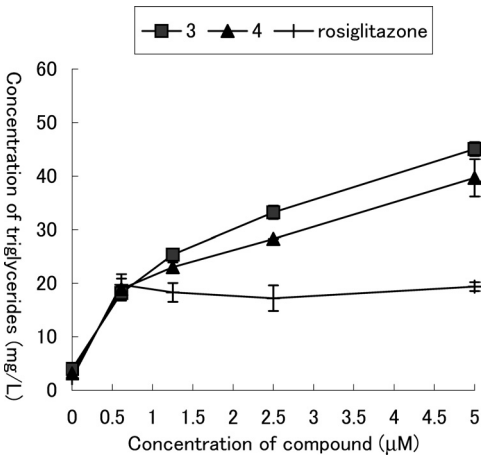


Fig. 6. Accumulation of Fatty Acid in Human Preadipocytes by Compounds **3**, **4** and Rosiglitazone

Values are the means of at least three experiments.

of PPARγ enhances adipocyte differentiation[31] and increases insulin sensitivity, compounds **3** and **4** were subjected to a human hypodermic preadipocyte differentiation test.[32] The accumulation of triglycerides in the cells was observed after the administration of compounds **3** and **4** at concentrations of 0, 0.613, 1.25, 2.5 and 5 μM, and the activity of compounds **3** and **4** was found to be higher than that of rosiglitazone at 1 μM and higher concentrations (Fig. 6).

Conclusion

To find novel PPAR ligands, we prepared several 2-nonylaminopyridine derivatives which were designed based on the structure of a PPARγ ligand **1**. In PPAR binding affinity assays, compound **4**, which had an ethoxy group at the C-2 position of the propanoic acid of **1**, showed high binding affinity for PPARγ and little affinity for PPARα and PPARδ. Compound **3**, which had an ethyl group at the C-2 position of propanoic acid, was found to be a PPARα/γ dual ligand. Compound **6**, the *meta* isomer of **1**, has been shown to be a PPARα ligand. The introduction of methyl (**7**) and ethyl (**8**) groups at the C-2 position of the propanoic acid of **6** further improved PPARα-binding potency. In cell-based transactivation assay, compounds **3** and **4** showed dual-agonist activity toward PPARα and PPARγ. Compound **6** was found to be a

triple agonist and compound **8** proved to be a selective PPARα agonist. In the human hypodermic preadipocyte differentiation test, it was demonstrated that the activity of compounds **3** and **4** was higher than that of rosiglitazone. The findings of this study will help provide an effective agent for Type 2 diabetes and hyperlipidemia.

Experimental

Melting points were determined using a Yanagimoto micro melting point apparatus or a Büchi 545 melting point apparatus and were left uncorrected. Proton nuclear magnetic resonance spectra (^{1}H-NMR) were recorded on a JEOL JNM-LA500 spectrometer in solvent as indicated. Chemical shifts (δ) were reported in parts per million relative to the internal standard tetramethylsilane. High-resolution mass spectra (HR-MS) were recorded on a JEOL JMS-SX102A mass spectrometer. Reagents and solvents were purchased from Aldrich, Tokyo Kasei Kogyo, Wako Pure Chemical Industries, and Kanto Kagaku, and used without purification. Flash column chromatography was performed using Silica Gel 60 (particle size 0.046—0.063 mm) supplied by Merck.

2-Nonylaminopyridine (18) To a solution of 2-bromopyridine (0.9 ml, 9.20 mmol) in 10 ml of anhydrous toluene were added *n*-nonylamine (**17**, 10.0 ml, 55.2 mmol), $Pd_2(DBA)_3$ (0.169 g, 0.18 mmol), *rac*-BINAP (0.229 g, 0.37 mmol) and sodium *tert*-butoxide (1.24 g, 12.9 mmol). The reaction mixture was stirred at 105 °C for 3 d under Ar, and poured into water. The whole was extracted with AcOEt. The AcOEt layer was separated, washed with water and brine, and dried over Na_2SO_4. The solvent was removed by evaporation *in vacuo*. Purification by silica gel flash chromatography (AcOEt/*n*-hexane=1/5) gave 1.43 g (70%) of **18** as a yellow solid: ^{1}H-NMR ($CDCl_3$, 500 MHz, δ: ppm) 8.07 (1H, dd, *J*=4.9, 1.5 Hz), 7.41 (1H, ddd, *J*=8.5, 6.7, 1.8 Hz), 6.55 (1H, dd, *J*=6.7, 5.5 Hz), 6.37 (1H, d, *J*=8.2 Hz), 4.45 (1H, m), 3.24 (2H, m), 1.65—1.59 (2H, m), 1.41—1.27 (12H, m), 0.88 (3H, t, *J*=6.9 Hz).

4-(2-Bromoethoxy)benzaldehyde (20a) To a solution of *p*-hydroxybenzaldehyde (**19a**, 1.25 g, 10.0 mmol) in 13 ml of THF were added Cs_2CO_3 (4.28 g, 13.0 mmol) and 1,2-dibromoethane (1.76 ml, 20.0 mmol). The mixture was stirred at 65 °C for 14 h. The mixture was poured into 2 N aqueous NaOH. The mixture was extracted with AcOEt. The AcOEt layer was separated, washed with water and brine, and dried over Na_2SO_4. The solvent was removed by evaporation *in vacuo*. Purification by silica gel flash chromatography (AcOEt/*n*-hexane=1/3) gave 0.758 g (33%) of **21a** as a light yellow solid: ^{1}H-NMR ($CDCl_3$, 500 MHz, δ: ppm) 9.90 (1H, s), 7.85 (2H, d, *J*=8.5 Hz), 7.02 (2H, d, *J*=8.8 Hz), 4.38 (2H, t, *J*=6.1 Hz), 3.67 (2H, t, *J*=6.1 Hz).

3-[4-(2-Bromoethoxy)phenyl]acrylic Acid Ethyl Ester (21a) To a suspension of NaH (191 mg, 4.78 mmol) in 4 ml of anhydrous THF was added dropwise a solution of ethyl diethylphosphono acetate (1.07 ml, 5.21 mmol) in 5 ml of anhydrous THF at 0 °C under Ar. The mixture was stirred for 1 h at 0 °C. To the mixture was added dropwise a solution of **20a** (916 mg, 4.00 mmol) in 8 ml of anhydrous THF. The reaction mixture was stirred for 16 h at 0 °C. The reaction mixture was poured into ice-water and extracted with AcOEt. The AcOEt layer was separated, washed with brine, and dried over Na_2SO_4. The solvent was removed by evaporation *in vacuo*. Purification by silica gel flash chromatography (AcOEt/*n*-hexane=1/5) gave 901 mg (76%) of **20a** as a white solid: ^{1}H-NMR ($CDCl_3$, 500 MHz, δ: ppm) 7.64 (1H, d, *J*=15.8 Hz), 7.48 (2H, d, *J*=8.8 Hz), 6.92 (2H, d, *J*=8.5 Hz), 6.32 (1H, d, *J*=15.8 Hz), 4.32 (2H, t, *J*=6.1 Hz), 4.26 (2H, q, *J*=7.3 Hz), 3.65 (2H, t, *J*=6.1 Hz), 1.34 (3H, t, *J*=7.0 Hz).

3-[4-(2-Bromoethoxy)phenyl]propanoic Acid Ethyl Ester (22a) To a solution of **21a** (445 mg, 1.49 mmol) in 5 ml of EtOH was added 64 mg of 7% Pd/C. The reaction mixture was stirred for 1 d under H_2 at room temperature. The catalyst was removed by filtration, and the solvent was removed by evaporation *in vacuo*. 359 mg (80%) of **22a** was obtained as a light yellow oil: ^{1}H-NMR ($CDCl_3$, 500 MHz, δ: ppm) 7.12 (2H, dt, *J*=8.8 , 3.0 Hz), 6.83 (2H, dt, *J*=8.5, 3.0 Hz), 4.26 (2H, t, *J*=6.1 Hz), 4.12 (2H, q, *J*=7.0 Hz), 3.62 (2H, t, *J*=6.1 Hz), 2.89 (2H, t, *J*=7.6 Hz), 2.58 (2H, t, *J*=8.2 Hz), 1.23 (3H, t, *J*=7.0 Hz).

3-{4-[2-(Nonyl-pyridin-2-ylamino)ethoxy]phenyl}propanoic Acid Ethyl Ester (23a) To a solution of **22a** (0.343 g, 1.14 mmol) in 1 ml of THF were added 2-nonylaminopyridine (1.01 g, 4.56 mmol), KI (0.189 g, 1.14 mmol) and Et_3N (0.45 ml, 2.30 mmol). The reaction mixture was stirred for 28 h at 105 °C. The reaction mixture was poured into water and extracted with AcOEt. The AcOEt layer was separated, washed with water and brine, and dried over Na_2SO_4. Purification by silica gel flash chromatography (toluene : $CHCl_3$: AcOEt=45 : 5 : 3) gave 55 mg (11%) of **23a** as a colorless oil: ^{1}H-NMR ($CDCl_3$, 500 MHz, δ: ppm) 8.13 (1H, ddd, *J*=4.8, 1.8, 0.9 Hz), 7.40 (1H, ddd, *J*=8.8, 7.0, 1.8 Hz), 7.09 (2H, d, *J*=8.5 Hz), 6.83 (2H, dt, *J*=8.5, 2.7 Hz), 6.50 (2H, m), 4.14 (2H, t, *J*=6.1 Hz), 4.11 (2H, q, *J*=7.3 Hz), 3.91 (2H, t, *J*=6.1 Hz), 3.47 (2H, t, *J*=7.9 Hz), 2.87 (2H, t, *J*=7.6 Hz), 2.57 (2H, t, *J*=7.9 Hz), 1.66—1.59 (2H, m), 1.36—1.20 (12H, m), 1.23 (3H, t, *J*=7.0 Hz), 0.88 (3H, t, *J*=6.7 Hz); HR-MS Calcd for $C_{27}H_{40}N_2O_3$ 440.3039, Found 440.3036.

3-{4-[2-(Nonylpyridin-2-ylamino)ethoxy]phenyl}propanoic Acid (1) (Chart 1) To a solution of **23a** (49 mg, 0.119 mmol) in 1.0 ml of THF and 1.0 ml of EtOH was added a solution of 2 N aqueous NaOH (0.60 ml, 1.19 mmol). The reaction mixture was stirred for 1 d at room temperature, and neutralized to pH 7 with 2 N aqueous HCl. The mixture was subjected to silica gel flash chromatography ($CHCl_3$/MeOH=19/1) to give 44 mg (90%) of **1** as a colorless oil: ^{1}H-NMR ($CDCl_3$, 500 MHz, δ: ppm) 8.14 (1H, dd *J*=4.9, 1.2 Hz), 7.45 (1H, t, *J*=7.1 Hz), 7.10 (2H, d, *J*=8.5 Hz), 6.81 (2H, d, *J*=8.5 Hz), 6.54 (2H, m), 4.12 (2H, t, *J*=5.6 Hz), 3.93 (2H, t, *J*=5.6 Hz), 3.50 (2H, t, *J*=7.9 Hz), 2.88 (2H, t, *J*=7.8 Hz), 2.61 (2H, t, *J*=7.8 Hz), 1.66—1.62 (2H, m), 1.35—1.25 (12H, m), 0.88 (3H, t, *J*=6.8 Hz); MS (EI) *m/z*: 412 (M^+); HR-MS Calcd for $C_{25}H_{36}N_2O_3$ 412.2727, Found 412.2726.

2-{4-[2-(Nonylpyridin-2-ylamino)ethoxy]benzyl}butyric Acid (3) Compound **3** was prepared from **19a** using the procedure described for **1** in 2.5% yield. In the step of the synthesis of **21b**, 2-(diethoxyphosphoryl)butyric acid ethyl ester was used instead of ethyl diethylphosphono acetate: colorless oil; ^{1}H-NMR ($CDCl_3$, 500 MHz, δ: ppm) 8.11 (1H, ddd, *J*=4.9, 1.8, 0.9 Hz), 7.41 (1H, ddd, *J*=8.8, 7.0, 1.8 Hz), 7.08 (2H, d, *J*=8.5 Hz), 6.79 (2H, d, *J*=8.5 Hz), 6.50 (1H, dd, *J*=7.0, 4.8 Hz), 6.48 (1H, d, *J*=7.9 Hz), 4.05 (1H, m), 3.99 (1H, m), 3.85 (2H, m), 3.46 (2H, t, *J*=7.9 Hz), 2.88 (1H, dd, *J*=13.7, 8.5 Hz), 2.70 (1H, dd, *J*=13.7, 6.4 Hz), 2.58 (1H, m), 1.68—1.55 (4H, m), 1.31—1.21 (12H, m), 0.96 (3H, t, *J*=7.6 Hz), 0.88 (3H, t, *J*=6.7 Hz); HR-MS Calcd for $C_{27}H_{40}N_2O_3$ 440.3039, Found 440.3029.

2-Ethoxy-3-{4-[2-(nonylpyridin-2-ylamino)ethoxy]phenyl}propanoic Acid (4) Compound **4** was prepared from **19a** using the procedure described for **1** in 2.1% yield. In the step of the synthesis of **21c**, 2-(diethoxyphosphoryl)ethoxyacetic acid ethyl ether was used instead of ethyl diethylphosphono acetate: colorless oil; ^{1}H-NMR ($CDCl_3$, 500 MHz, δ: ppm) 8.12 (1H, dd, *J*=4.9, 1.2 Hz), 7.43 (1H, ddd, *J*=8.8, 7.0, 1.8 Hz), 7.28 (2H, d, *J*=8.5 Hz), 6.80 (2H, d, *J*=8.5 Hz), 6.51 (2H, m), 4.05 (2H, m), 3.90 (2H, m), 3.60 (1H, m), 3.47 (2H, t, *J*=7.6 Hz), 3.05 (1H, dd, *J*=14.0, 4.5 Hz), 2.94 (1H, dd, 14.3, 7.6 Hz), 1.65—1.59 (2H, m), 1.35—1.20 (12H, m), 1.17 (3H, t, *J*=7.0 Hz), 1.17 (3H, t, *J*=7.0 Hz), 0.88 (3H, t, *J*=7.0 Hz); HR-MS Calcd for $C_{27}H_{40}N_2O_4$ 456.2988, Found 456.2999.

3-{3-[2-(Nonylpyridin-2-ylamino)ethoxy]phenyl}propanoic Acid (6) Compound **6** was prepared from **19b** using the procedure described for **1** in 7.7% yield: colorless oil; ^{1}H-NMR ($CDCl_3$, 500 MHz, δ: ppm) 8.14 (1H, ddd, *J*=4.9, 2.0, 0.8 Hz), 7.42 (1H, ddd, *J*=8.5, 6.7, 2.1 Hz), 7.18 (1H, t, *J*=7.9 Hz), 6.75 (3H, m), 6.5 (2H, m), 4.15 (2H, t, *J*=6.1 Hz), 3.91 (2H, t, *J*=5.8 Hz), 3.47 (2H, t, *J*=7.6 Hz), 2.91 (2H, t, *J*=7.6 Hz), 2.65 (2H, t, *J*=7.9 Hz), 1.65—1.61 (2H, m), 1.35—1.25 (12H, m), 0.88 (3H, t, *J*=6.7 Hz); HR-MS Calcd for $C_{25}H_{36}N_2O_3$ 412.2726, Found 412.2729.

2-Methyl-3-{3-[2-(nonylpyridin-2-ylamino)ethoxy]phenyl}propanoic Acid (7) Compound **7** was prepared from **19b** using the procedure described for **1** in 3.5% yield. In the step of the synthesis of **21e**, 2-(diethoxyphosphoryl)propanoic acid ethyl ester was used instead of ethyl diethylphosphono acetate: light yellow oil; ^{1}H-NMR ($CDCl_3$, 500 MHz, δ: ppm) 8.14 (1H, ddd, *J*=4.8, 1.8, 0.9 Hz), 7.42 (1H, ddd, *J*=8.8, 6.7, 2.1 Hz), 7.17 (1H, t, *J*=7.9 Hz), 6.74—6.78 (3H, m), 6.52 (1H, m), 6.50 (1H, d, *J*=8.8 Hz), 4.14 (2H, t, *J*=6.1 Hz), 3.90 (2H, m), 3.46 (3H, t, *J*=7.6 Hz), 3.02 (1H, dd, *J*=13.4, 6.7 Hz), 2.75 (1H, m), 2.64 (1H, dd, *J*=13.4, 7.6 Hz), 1.66—1.58 (2H, m), 1.35—1.25 (12H, m), 1.17 (3H, d, *J*=7.0 Hz), 0.88 (3H, t, *J*=7.3 Hz); HR-MS Calcd for $C_{26}H_{38}N_2O_3$ 426.2882, Found; M^+ 426.2877.

2-{3-[2-(Nonylpyridin-2-ylamino)ethoxy]benzyl}butyric Acid (8) Compound **8** was prepared from **19b** using the procedure described for **1** in 4.9% yield. In the step of the synthesis of **21f**, 2-(diethoxyphosphoryl)butyric acid ethyl ester was used instead of ethyl diethylphosphono acetate: yellow oil; ^{1}H-NMR ($CDCl_3$, 500 MHz, δ: ppm) 8.13 (1H, ddd, *J* =5.2, 1.2 Hz), 7.42 (1H, ddd, *J*=8.8, 6.7, 2.1 Hz), 7.15 (1H, t, *J*=7.9 Hz), 6.71—6.79 (3H, m), 6.52 (1H, dd, *J*=6.7, 5.2 Hz), 6.50 (1H, d, *J*=8.8 Hz), 4.12 (2H, t, *J*=5.5 Hz), 3.83—3.94 (2H, m), 3.46 (3H, t, *J*=7.6 Hz), 2.93 (1H, dd, *J*=13.7, 8.2 Hz), 2.72 (1H, dd, *J*=13.7, 6.7 Hz), 2.60 (1H, m), 1.69—1.55 (4H, m), 1.35—1.25 (12H, m), 0.96 (3H, t, *J*=7.3 Hz), 0.82 (3H, t, *J*=7.0 Hz); HR-MS Calcd for $C_{27}H_{40}N_2O_3$ 440.3039, Found 440.3043.

2-Ethoxy-3-{3-[2-(nonylpyridin-2-ylamino)ethoxy]phenyl}propanoic Acid (9) Compound **9** was prepared from **19b** using the procedure described for **1** in 2.5% yield. In the step of the synthesis of **21g**, 2-(diethoxyphosphoryl)ethoxyacetic acid ethyl ether was used instead of ethyl diethylphosphono acetate: yellow oil; ^{1}H-NMR ($CDCl_3$, 500 MHz, δ: ppm) 8.13 (1H, dd, J=4.9, 1.2 Hz), 7.44 (1H, ddd, J=8.8, 7.0, 1.8 Hz), 7.17 (1H, t, J=7.6 Hz), 6.83 (2H, m), 6.77 (1H, dd, J=7.9, 1.8 Hz), 6.53 (2H, m), 4.15 (2H, t, J=5.5 Hz), 4.07 (1H, dd, J=7.3, 4.9 Hz), 3.90 (2H, m), 3.59 (1H, m), 3.47 (2H, t, J=7.9 Hz), 3.43 (1H, m), 3.09 (1H, dd, J=13.7, 4.6 Hz), 2.98 (1H, dd, J=13.7, 7.3 Hz), 1.67—1.59 (2H, m), 1.36—1.23 (12H, m), 1.16 (3H, t, J=7.0 Hz), 0.88 (3H, t, J=6.7 Hz); HR-MS Calcd for $C_{27}H_{40}N_2O_4$ 456.2988, Found 456.2985.

3-{4-Methoxy-3-[2-(nonylpyridin-2-ylamino)ethoxy]phenyl}propanoic Acid (10) Compound **10** was prepared from **19c** using the procedure described for **1** in 2.7% yield: yellow oil; ^{1}H-NMR ($CDCl_3$, 500 MHz, δ: ppm) 8.16 (1H, ddd, J=4.9, 1.8, 0.9 Hz), 7.43 (1H, ddd, J=8.8, 7.0, 1.8 Hz), 6.93 (1H, d, J=1.5 Hz), 6.79 (1H, d, J=8.2 Hz), 6.74 (1H, dd, J=8.2, 1.8 Hz), 6.53 (1H, dd, J=7.0, 5.2 Hz), 6.51 (1H, d, J=8.8 Hz), 4.24 (2H, t, J=6.7 Hz), 3.94 (2H, t, J=6.4 Hz), 3.83 (3H, s), 3.45 (2H, t, J=7.6 Hz), 2.88 (2H, t, J=7.6 Hz), 2.61 (2H, t, J=7.6 Hz), 1.64—1.58 (2H, m), 1.31—1.25 (12H, m), 0.88 (3H, t, J=7.0 Hz); HR-MS Calcd for $C_{26}H_{38}N_2O_4$ 442.2832, Found 442.2846.

2-{4-Methoxy-3-[2-(nonylpyridin-2-ylamino)ethoxy]benzyl}butyric Acid (12) Compound **12** was prepared from **19c** using the procedure described for **1** in 3.0% yield. In the step of the synthesis of **21i**, 2-(diethoxyphosphoryl)butyric acid ethyl ester was used instead of ethyl diethylphosphono acetate: yellow oil; ^{1}H-NMR ($CDCl_3$, 500 MHz, δ: ppm) 8.16 (1H, ddd, J=4.8, 1.8, 0.6 Hz), 7.43 (1H, ddd, J=9.1, 7.3, 2.1 Hz), 6.91 (1H, d, J=1.8 Hz), 6.76 (1H, d, J=8.2 Hz), 6.71 (1H, dd, J=8.2, 2.1 Hz), 6.53 (1H, dd, J=6.4, 5.1 Hz), 6.50 (1H, d, J=8.8 Hz), 4.23 (2H, m), 3.98 (2H, m), 3.86 (1H, m), 3.83 (3H, s), 3.43 (2H, m), 2.87 (1H, dd, J=13.7, 8.5 Hz), 2.71 (1H, dd, J=13.6, 6.1 Hz), 2.56—2.50 (1H, m), 1.70—1.51 (4H, m), 1.34—1.23 (12H, m), 0.95 (3H, t, J=7.0 Hz), 0.88 (3H, t, J=7.0 Hz); HR-MS Calcd for $C_{28}H_{42}N_2O_4$ 470.3145, Found 470.3142.

2-Ethoxy-3-{4-methoxy-3-[2-(nonylpyridin-2-ylamino)ethoxy]phenyl-propanoic Acid (13) Compound **13** was prepared from **19c** using the procedure described for **1** in 3.8% yield. In the step of the synthesis of **21j**, 2-(diethoxyphosphoryl)ethoxyacetic acid ethyl ether was used instead of ethyl diethylphosphono acetate: yellow oil; ^{1}H-NMR ($CDCl_3$, 500 MHz, δ: ppm) 8.13 (1H, dd, J=4.9, 1.8 Hz), 7.45 (1H, ddd, J=8.8, 7.0, 1.8 Hz), 6.96 (1H, s), 6.78 (2H, m), 6.54 (1H, dd, J=6.7, 5.5 Hz), 6.52 (1H, d, J=8.5 Hz), 4.25 (2H, m), 4.04 (1H, m), 3.93 (2H, m), 3.83 (3H, s), 3.59 (1H, m), 3.44 (2H, t, J=7.6 Hz), 3.05 (1H, dd, J=13.7, 4.9 Hz), 2.97 (1H, dd, J=13.7, 6.4 Hz), 1.65—1.58 (2H, m), 1.35—1.21 (12H, br), 1.17 (3H, t, J=7.0 Hz), 0.88 (3H, t, J=6.7 Hz); HR-MS Calcd for $C_{28}H_{42}N_2O_5$ 486.3094, Found 486.3111.

{4-(2-Bromoethoxy)phenyl}methanol (24a) To a solution of **20a** (890 mg, 3.89 mmol) in 10 ml of EtOH was added $NaBH_4$ (163 mg, 3.89 mmol). The reaction mixture was stirred for 6 h at room temperature. The reaction mixture was poured into water, and extracted with AcOEt. The AcOEt layer was separated, washed with water and brine, and dried over Na_2SO_4. The solvent was removed by evaporation *in vacuo*. 763 mg (81%) of **24a** was obtained as a white solid: ^{1}H-NMR ($CDCl_3$, 500 MHz, δ: ppm) 7.30 (2H, d, J=8.5 Hz), 6.91 (2H, d, J=8.8 Hz), 4.62 (2H, d, J=5.1 Hz), 4.29 (2H, t, J=6.1 Hz), 3.64 (2H, t, J=6.4 Hz), 1.58 (1H, t, J=5.5 Hz).

Acetic Acid 4-(2-Bromoethoxy)benzyl Ester (25a) To a solution of **24a** (2.31 g, 10.0 mmol) in 20 ml of CH_2Cl_2 were added Ac_2O (3.10 ml, 30.0 mmol) and a catalytic amount of DMAP. The reaction mixture was stirred for 1 d at room temperature and diluted with AcOEt. The AcOEt solution was washed with water and brine, and dried over Na_2SO_4. Purification by silica gel flash chromatography (AcOEt/*n*-hexane =1/5) gave 2.62 g (96%) of **25a** as a light yellow oil: ^{1}H-NMR ($CDCl_3$, 500 MHz, δ: ppm) 7.29 (2H, d, J=8.8 Hz), 6.90 (2H, d, J=8.8 Hz), 5.04 (2H, s), 4.28 (2H, t, J=6.1 Hz), 3.63 (2H, t, J=6.1 Hz), 2.07 (3H, s).

3-{4-(2-Bromoethoxy)phenyl}-2-methylpropionic Acid Methyl Ester (26a) To a solution of **25a** (0.948 g, 3.47 mmol) and 1-methoxy-1-trimethylsilyloxypropene (1.00 g, 6.24 mmol) in 35 ml of anhydrous CH_2Cl_2 was added magnesium perchlorate (0.129 g, 0.694 mmol) under Ar. The reaction mixture was stirred for 1 d at room temperature. The reaction mixture was diluted with CH_2Cl_2. The CH_2Cl_2 solution was washed with water and brine, and dried over Na_2SO_4. The solvent was removed by evaporation *in vacuo*. Purification by silica gel flash chromatography (AcOEt/*n*-hexane=1/10) gave 0.935 g (90%) of **26a** as a colorless oil: ^{1}H-NMR ($CDCl_3$, 500 MHz, δ: ppm) 7.07 (2H, dt, J=8.5, 3.0 Hz), 6.82 (2H, dt, J=8.5, 3.0 Hz), 4.26 (2H, t, J=6.4 Hz), 3.63 (3H, s), 3.62 (2H, t, J=6.1 Hz), 2.95 (1H, dd, J=13.4, 7.1 Hz), 2.69 (1H, m), 2.61 (1H, dd, J=13.4, 7.6 Hz), 1.14 (3H, d, J=7.0 Hz).

3-{4-[2-(Nonylpyridin-2-ylamino)ethoxy]phenyl}-2,2-dimethylpropanoic Acid Methyl Ester (27a) Compound **27a** was prepared from **26a** using the procedure described for **23a** in 11% yield: ^{1}H-NMR ($CDCl_3$, 500 MHz, δ: ppm) 8.13 (1H, ddd, J=4.9, 1.8, 0.9 Hz), 7.41 (1H, ddd, J=8.8, 7.0, 2.1 Hz), 7.04 (2H, d, J=8.5 Hz), 6.82 (2H, dt, J=8.5, 1.8 Hz), 6.5 (2H, m), 4.14 (2H, t, J=6.1 Hz), 3.91 (2H, t, J=5.8 Hz), 3.63 (3H, s), 3.47 (2H, t, J=7.9 Hz), 2.94 (1H, dd, J=13.4, 6.7 Hz), 2.68 (1H, m), 2.59 (1H, dd, J=13.4, 7.6 Hz), 1.64—1.57 (2H, m), 1.35—1.25 (12H, m), 1.13 (3H, d, J=7.0 Hz), 0.88 (3H, t, J=7.2 Hz); HR-MS Calcd for $C_{27}H_{40}N_2O_3$ 440.3039, Found 440.3046.

3-{4-[2-(Nonylpyridin-2-ylamino)ethoxy]phenyl}-2-methylpropanoic Acid (2) Compound **2** was prepared from **27a** using the procedure described for **1** in 97% yield: colorless oil; ^{1}H-NMR ($CDCl_3$, 500 MHz, δ: ppm) 8.13 (2H, d, J=4.8 Hz), 7.41 (1H, ddd, J=8.8, 6.7, 1.8 Hz), 7.08 (2H, d, J=8.5 Hz), 6.82 (2H, d, J=8.5 Hz), 6.5 (2H, m), 4.12 (2H, m), 3.90 (2H, t, J=5.8 Hz), 3.47 (2H, t, J=7.9 Hz), 2.98 (1H, dd, J=13.4, 7.0 Hz), 2.73 (1H, m), 2.63 (1H, dd, J=13.4, 7.6 Hz), 1.65—1.58 (2H, br), 1.35—1.25 (12H, m), 1.17 (3H, d, J=6.7 Hz), 0.88 (3H, t, J=7.0 Hz); HR-MS Calcd for $C_{26}H_{38}N_2O_3$ 426.2882, Found 426.2889.

3-{4-[2-(Nonylpyridin-2-ylamino)ethoxy]phenyl}-2,2-dimethylpropanoic Acid (5) Compound **5** was prepared from **20a** using the procedure described for **2** in 4.2% yield. In the step of the synthesis of **26b**, dimethylketene methyltrimethylsilyl acetal was used instead of 1-methoxy-1-trimethylsilyloxypropene: light yellow oil; ^{1}H-NMR ($CDCl_3$, 500 MHz, δ: ppm) 8.14 (2H, dd, J=4.9, 1.5 Hz), 7.41 (1H, ddd, J=8.8, 6.7, 1.8 Hz), 7.05 (2H, d, J=8.5 Hz), 6.80 (2H, d, J=8.5 Hz), 6.50 (2H, m), 4.12 (2H, t, J=5.8 Hz), 3.90 (2H, t, J=5.8 Hz), 3.47 (2H, t, J=7.6 Hz), 2.81 (2H, s), 1.65—1.59 (2H, m), 1.35—1.25 (12H, m), 1.17 (6H, s), 0.88 (3H, t, J=7.0 Hz); HR-MS Calcd for $C_{27}H_{40}N_2O_3$ 440.3039, Found 440.3040.

3-{4-Methoxy-3-[2-(Nonylpyridin-2-ylamino)ethoxy]phenyl}-2-methylpropanoic Acid (11) Compound **11** was prepared from **20c** using the procedure described for **2** in 3.3% yield: colorless oil; ^{1}H-NMR ($CDCl_3$, 500 MHz, δ: ppm) 8.16 (1H, ddd, J=4.9, 1.8, 0.9 Hz), 7.44 (1H, ddd, J=8.8, 6.7, 1.8 Hz), 6.95 (1H, d, J=1.8 Hz), 6.78 (1H, d, J=7.9 Hz), 6.71 (1H, dd, J=8.2, 1.8 Hz), 6.54 (1H, dd, J=7.0, 5.1 Hz), 6.50 (1H, d, J=8.8 Hz), 4.25 (2H, m), 3.92 (2H, m), 3.83 (3H, s), 3.43 (2H, t, J=7.3 Hz), 2.92 (1H, m), 2.70 (2H, m), 1.64—1.54 (2H, m), 1.31—1.20 (12H, m), 1.16 (3H, d, J=6.7 Hz), 0.88 (3H, t, J=6.7 Hz); HR-MS Calcd for $C_{27}H_{40}N_2O_4$ 456.2988, Found 456.3007.

3-{4-Methoxy-3-[2-(Nonylpyridin-2-ylamino)ethoxy]phenyl}-2,2-dimethylpropanoic Acid (14) Compound **14** was prepared from **20c** using the procedure described for **2** in 8.4% yield. In the step of the synthesis of **26d**, dimethylketene methyltrimethylsilyl acetal was used instead of 1-methoxy-1-trimethylsilyloxypropene: colorless oil; ^{1}H-NMR ($CDCl_3$, 500 MHz, δ: ppm) 8.16 (1H, ddd, J=5.1, 1.8, 0.6 Hz), 7.45 (1H, ddd, J=8.8, 7.0, 1.8 Hz), 7.00 (1H, d, J=2.1 Hz), 6.76 (1H, d, J=8.2 Hz), 6.68 (1H, dd, J=8.2, 1.8 Hz), 6.55 (1H, dd, J=7.0, 5.2 Hz), 6.50 (1H, d, J=8.8 Hz), 4.27 (2H, t, J=7.0 Hz), 3.90 (2H, t, J=7.3 Hz), 3.84 (3H, s), 3.40 (2H, t, J=7.6 Hz), 2.79 (2H, s), 1.63—1.57 (2H, m), 1.35—1.25 (12H, m), 1.20 (6H, s), 0.88 (3H, t, J=7.0 Hz); HR-MS Calcd for $C_{28}H_{42}N_2O_4$ 470.3145, Found 470.3144.

3-{4-[3-(Nonylpyridin-2-ylamino)propyl]phenyl}acrylic Acid Ethyl Ester (28a) (Chart 3) Compound **28a** was prepared from **21a** using the procedure described for **23a** in 13% yield: ^{1}H-NMR ($CDCl_3$, 500 MHz, δ: ppm) 8.14 (1H, ddd, J=4.8, 2.1, 0.9 Hz), 7.63 (1H, d, J=16.2 Hz), 7.45 (2H, d, J=8.8 Hz), 7.42 (1H, ddd, J=8.8, 7.0, 1.8 Hz), 6.91 (2H, d, J=8.5 Hz), 6.52 (1H, dd, J=7.0, 5.5 Hz), 6.50 (1H, d, J=8.8 Hz), 6.30 (1H, d, J=15.8 Hz), 4.25 (2H, q, J=7.0 Hz), 4.21 (2H, t, J=5.8 Hz), 3.94 (2H, t, J=5.8 Hz), 3.46 (2H, t, J=7.9 Hz), 1.64—1.60 (2H, m), 1.36—1.30 (3H, t, J=7.0 Hz), 1.33—1.23 (12H, br), 0.88 (3H, t, J=7.0 Hz); HR-MS Calcd for $C_{27}H_{38}N_2O_3$ 438.2882, Found 438.2855.

3-{4-[3-(Nonylpyridin-2-ylamino)propyl]phenyl}acrylic Acid (15) To a solution of **28a** (84 mg, 0.192 mmol) in 2 ml of THF and 2 ml of EtOH was added a solution of 2 N aqueous NaOH (0.29 ml, 0.580 mmol). The reaction mixture was stirred for 18 h at room temperature, and neutralized to pH 7 with aqueous 2 N HCl. Purification by silica gel flash chromatography ($CHCl_3$/MeOH=19/1) gave 78 mg (99%) of white solid. The 41 mg of the white solid was recrystallized from AcOEt–*n*-hexane and collected by filtration to give 22 mg (52%) of **15** as a white solid: mp 142—143 °C; ^{1}H-NMR ($CDCl_3$, 500 MHz, δ: ppm) 8.15 (1H, dd, J=4.9, 1.8 Hz), 7.70 (1H, d, J=15.9 Hz), 7.47 (2H, d, J=8.8 Hz), 7.43 (1H, ddd, J=8.8, 7.0, 1.8 Hz), 6.93 (1H, d, J=8.8 Hz), 6.53 (1H, dd, J=6.4, 5.5 Hz), 6.50 (1H, d,

J=8.5 Hz), 6.29 (1H, d, J=16.1 Hz), 4.22 (2H, t, J=5.8 Hz), 3.95 (2H, t, J=5.8 Hz), 3.46 (2H, t, J=8.2 Hz), 1.65—1.61 (2H, m), 1.34—1.24 (12H, m), 0.88 (3H, t, J=7.0 Hz); *Anal.* Calcd for $C_{25}H_{34}N_2O_3$: C, 73.14; H, 8.35; N, 6.82. Found: C, 73.23; H, 8.46; N, 6.89.

3-{4-Methoxy-3-[2-(nonylpyridin-2-ylamino)ethoxy]phenyl}acrylic Acid (16) Compound **16** was prepared from **21h** using the procedure described for **15** in 7.4% yield: colorless oil; ^{1}H-NMR ($CDCl_3$, 500 MHz, δ: ppm) 8.22 (1H, dd, J=4.9, 1.8 Hz), 7.69 (1H, d, J=15.8 Hz), 7.44 (1H, ddd, J=8.8, 7.0, 1.8 Hz), 7.31 (1H, d, J=1.5 Hz), 7.09 (1H, dd, J=8.2, 1.8 Hz), 6.86 (1H, d, J=8.2 Hz), 6.56 (1H, dd, J=7.0, 5.2 Hz), 6.51 (1H, d, J=8.5 Hz), 6.31 (1H, d, J=15.8 Hz), 4.31 (2H, t, J=6.4 Hz), 3.99 (2H, t, J=6.1 Hz), 3.90 (3H, s), 3.45 (2H, t, J=7.6 Hz), 1.65—1.61 (2H, m), 1.32—1.22 (12H, m), 0.88 (3H, t, J=7.0 Hz); HR-MS Calcd for $C_{26}H_{36}N_2O_4$ 440.2675, Found 440.2716.

Binding Assay The assay for PPAR binding activity was performed using a CoA-BAP System kit (NuLigand series, Microsystems). Briefly, glutathione-*S*-transferase-human nuclear receptor ligand binding domain (GST-hNR LBD) fusion proteins were incubated in a glutathione-fixed micro-well plate at 4 °C overnight. After excessive proteins were removed, human transcriptional intermediary factor 2 (hTIF2)-bacterial alkaline phosphatase (BAP) fusion proteins were added to the well with test chemicals. After 1 h incubation at 4 °C, excessive proteins were removed carefully. The enzyme reaction was allowed to start with the addition of *p*-nitrophenylphosphoric acid (NPP) as a substrate, and incubated at 30 °C. After 3 h, the reaction was stopped by the addition of 0.5 N aqueous NaOH. The product was measured by reading absorbance at 405 nm with a 1420 ARVO™ multilabel counter (ParkinElmer, Boston, MA, U.S.A.). AP activity was determined by subtracting background absorption from the reading at 405 nm.

Cell-Based Transactivation Assay[30] Human embryonic kidney (HEK) 293 cells were cultured in DMEM containing 5% fetal bovine serum at 37 °C in a humidified atmosphere of 5% CO_2 in air. Transfections of PPAR and reporter gene constructs were performed by calcium phosphate coprecipitation. Eight hours after transfection, ligands were added. Cells were harvested 12—16 h after treatment, and luciferase and β-galactosidase activities were assayed using a 1420 ARVO™ MX multilabel counter (ParkinElmer, Boston, MA, U.S.A.). DNA cotransfection experiments included 58 ng of reporter plasmid, 12 ng of CMX-β-galactosidase, and 18 ng of each receptor expression plasmid per well in a 96-well plate. Luciferase data were normalized to an internal β-galactosidase control and reported values are the means of triplicate assays.

Preadipocyte Differentiation Test Human preadipocytes from hypodermic tissues, a preadipocyte growth medium and a preadipocyte differentiation medium were purchased from TOYOBO. Co., Ltd (Osaka, Japan). Human preadipocytes were cultured for 8 d in preadipocyte growth medium in a humidified incubator at 37 °C and 5% CO_2. The medium was renewed every other day. When the preadipocytes reached confluence, the cells were treated with preadipocyte differentiation medium containing compounds **3**, **4**, or rosiglitazone. The cells were cultured for a further 7 d with the differentiation medium renewed every 3 d. The accumulation of triglycerides was evaluated by measuring the absorbance at 570 nm with a 1420 ARVO™ multilabel counter (ParkinElmer, Boston, MA, U.S.A.) after staining with Lipidos Liquid® (TOYOBO Co., Ltd, Osaka, Japan).

References and Notes

1) Nuclear Receptors Nomenclature Committee. *Cell*, **97**, 161—163 (1999).
2) Willson T. M., Brown P. J., Sternbach D. D., Henke B. R., *J. Med. Chem.*, **43**, 527—550 (2000).
4) Kersten S., Desvergne B., Wahli W., *Nature* (London), **405**, 421—424 (2000).
6) Cantello B. C. C., Cawthorone M. A., Cottam G. P., Duff P. T., Haigh D., Hindley R. M., Lister C. A., Smith S. A., Thurlby P. L., *J. Med. Chem.*, **37**, 3977—3985 (1994).
7) Momose Y., Meguro K., Ikeda H., Hatanaka C., Oi S., Sohda T., *Chem. Pharm. Bull.*, **39**, 1440—1445 (1991).
8) Hauner H., *Diabetes Metab. Res. Rev.*, **18**, S10—S15 (2002).
9) Francis G. A., Annicotte J. S., Auwerx J., *Curr. Opin. Parmacol.*, **3**, 186—191 (2003).
10) Lohray B. B., Lohray V. B., Bajji A. C., Kalchar S., Poondra R. R., Padakanti S., Chakrabarti R., Vikramadithyan R. K., Misra P., Juluri S., Mamidi N. V., Rajagopalan R., *J. Med. Chem.*, **44**, 2675—2678 (2001).
11) Duran-Sandoval D., Thomas A. C., Bailleul B., Fruchart J. C., Staels B., *Med. Sci.*, **19**, 819—825 (2003).
12) Forman B. M., Tontonoz P., Chen J., Brun R. P., Spiegelman B. M., Evans R. M., *Cell*, **83**, 803—812 (1995).
13) Kliewer S. A., Lenhard J. M., Willson T. M., Patel I., Morris D. C., Lehmann J. M., *Cell*, **83**, 813—819 (1995).
14) Usui S., Suzuki T., Hattori Y., Etoh K., Fujieda H., Nishizuka M., Imagawa M., Nakagawa H., Kohda K., Miyata N., *Bioorg. Med. Chem. Lett.*, **15**, 1547—1551 (2005).
15) Buckle D. R., Cantello B. C. C., Cawthorne M. A., Coyle P. J., Dean D. K., Faller A., Haigh D., Hindley R. M., Jefcott L. J., Lister C. A., Pinto I. L., Rami H. K., Smith D. G., Smith S. A., *Bioorg. Med. Chem. Lett.*, **6**, 2121—2126 (1996).
16) Buckle D. R., Cantello B. C. C., Cawthorne M. A., Coyle P. J., Dean D. K., Faller A., Haigh D., Hindley R. M., Jefcott L. J., Lister C. A., Pinto I. L., Rami H. K., Smith D. G., Smith S. A., *Bioorg. Med. Chem. Lett.*, **6**, 2127—2130 (1996).
17) Hulin B., Newton L. S., Lewis D. M., Genereux P. E., Gibbs E. M., Clark D. A., *J. Med. Chem.*, **39**, 3897—3907 (1996).
18) Sauerberg P., Pettersson I., Jeppesen L., Bury P. S., Mogensen J. P., Wassermann K., Brand C. L., Sturis J., Woldike H. F., Fleckner J., Andersen A. S., Mortensen S. B., Svensson L. A., Rasmussen H. B., Lehmann S. V., Polivka Z., Sindelar K., Panajotova V., Ynddal L., Wulff E. M., *J. Med. Chem.*, **45**, 789—804 (2002).
19) Cronet P., Petersen J. F., Folmer R., Blomberg N., Sjoblom K., Karlsson U., Lindstedt E. L., Bamberg K., *Structure*, **9**, 699—706 (2001).
20) Nomura M., Tanase T., Ide T., Tsunoda M., Suzuki M., Uchiki H., Murakami K., Miyachi H., *J. Med. Chem.*, **46**, 3581—3599 (2003).
21) Weigand S., Bischoff H., Dittrich-Wengenroth E., Heckroth H., Lang D., Vaupel A., Woltering M., *Bioorg. Med. Chem. Lett.*, **15**, 4619—4623 (2005).
22) Kasuga J., Makishima M., Hashimoto Y., Miyachi H., *Bioorg. Med. Chem. Lett.*, **16**, 554—558 (2006).
23) Wagaw S., Buchwald S. L., *J. Org. Chem.*, **61**, 7240—7241 (1996).
24) Maryanoff B. E., Reitz A. B., *Chem. Rev.*, **89**, 863—927 (1989).
25) Grieco P. A., Handy S. T., *Tetrahedron Lett.*, **38**, 2645—2648 (1997).
26) Kanayama T., Mamiya S., Nishihara T., Nishikawa J., *J. Biochem.* (Tokyo), **133**, 791—797 (2003).
27) Brown P. J., Stuart L. W., Hurley K. P., Lewis M. C., Winegar D. A., Wilson J. G., Wilkison W. O., Ittoop O. R., Willson T. M., *Bioorg. Med. Chem. Lett.*, **11**, 1225—1227 (2001).
28) Sznaidman M. L., Haffner C. D., Maloney P. R., Fivush A., Chao E., Goreham D., Sierra M. L., LeGrumelec C., Xu H. E., Montana V. G., Lambert M. H., Willson T. M., Oliver W. R., Jr., Sternbach D. D., *Bioorg. Med. Chem. Lett.*, **13**, 1517—1521 (2003).
29) Usui S., Fujieda H., Suzuki T., Yoshida N., Nakagawa H., Miyata N., *Bioorg. Med. Chem. Lett.*, **16**, 3249—3254 (2006).
30) Fukuen S., Iwaki M., Yasui A., Makishima M., Matsuda M., Shimomura I., *J. Biol. Chem*, **280**, 23653—23659 (2005).
31) Henke B. R., *J. Med. Chem.*, **47**, 4118—4127 (2004).
32) Lagace D. C., Nachtigal M. W., *J. Biol. Chem.*, **279**, 18851—18860 (2004).
33) Relative transactivation activity at 10 μM: compound **4**, 163% of WY14643 for α-activity, and 88% of rosiglitazone for γ-activity, compound **8**, 275% of WY14643 for α-activity.