# Synthesis and Evaluation of 2-Nonylaminopyridine Derivatives as PPAR Ligands

Shinya Usui,<sup>*a*</sup> Hiroki Fujieda,<sup>*a*</sup> Takayoshi Suzuki,<sup>*a*</sup> Naoaki Yoshida,<sup>*a*</sup> Hidehiko Nakagawa,<sup>*a*</sup> Michitaka Ogura,<sup>*b*</sup> Makoto Makishima,<sup>*b*</sup> and Naoki Miyata<sup>\*,*a*</sup>

<sup>a</sup> Graduate School of Pharmaceutical Sciences, Nagoya City University; 3–1 Tanabe-dori, Mizuho-ku, Nagoya, Aichi 467–8603, Japan: and <sup>b</sup>Nihon University School of Medicine; 30–1 Oyaguchi-kamicho, Itabashi-ku, Tokyo 173–8610, Japan. Received March 26, 2007; accepted May 7, 2007; published online May 9, 2007

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 PPAR $\gamma$  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 PPAR $\gamma$ . Compound 3, with an ethyl group at the C-2 position, was found to be a PPAR $\alpha/\gamma$  dual ligand. Compound 6, the meta isomer of 1, has been shown to be a PPAR $\alpha$  ligand. The introduction of methyl (7) and ethyl (8) groups to the C-2 position of the propanoic acid of 6 further improved PPAR $\alpha$ -binding potency. In cell-based transactivation assay, compounds 3 and 4 showed dual-agonist activity toward PPAR $\alpha$  and PPAR $\gamma$ . Compound 6 was found to be a triple agonist and compound 8 proved to be a selective PPAR $\alpha$  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 (PPAR $\alpha$ , PPAR $\gamma$  and PPAR $\delta$ ) are a set of ligand-activated transcription factors in the nuclear hormone receptor superfamily.<sup>1-4)</sup> 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.<sup>5)</sup> The PPAR $\gamma$  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<sup>6)</sup> and pioglitazone<sup>7)</sup> (Fig. 1) are potent PPAR $\gamma$  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<sup>8</sup>; how-



Fig. 1. Structures of Rosiglitazone, Pioglitazone, 15d-PGJ<sub>2</sub>, Clofibrate, GW7647, GW501516 and Compound 1

ever, the use of TZDs has been limited because of their serious side effects such as hepatic toxicity, weight gain and edema. Meanwhile, PPAR $\alpha$  is highly expressed in metabolically active tissues such as the liver, heart and muscle, and regulates lipid homeostasis. PPAR $\alpha$  agonists such as clofibrate (Fig. 1) have demonstrated the ability to reduce serum triglyceride and increase HDL cholesterol levels,<sup>9)</sup> and are being utilized as hypolipidemic agents. In addition, recent studies revealed that dual agonists of PPAR $\alpha/\gamma$  decrease the free triglyceride plasma concentration and increase plasma HDL concentration in an insulin-resistant animal model.<sup>10,11)</sup> Thus, many groups have ongoing research programs to identify more potent and less toxic PPAR $\alpha$  agonists, PPAR $\gamma$  agonists and PPAR $\alpha/\gamma$  dual agonists.

We previously reported compound **1** (Fig. 1), which was designed based on the structure of rosiglitazone and 15d-PGJ<sub>2</sub>,<sup>12,13</sup> as a potent PPAR $\gamma$  ligand.<sup>14</sup>) To find more potent PPAR $\gamma$  agonists and novel PPAR $\alpha$  agonists, we chose compound **1** as the lead structure, because recent reports indicated that PPAR $\gamma$  affinity can be increased by the introduction of substituents into the C-2 position of propanoic acid,<sup>15—19</sup>) and minor structural modifications can convert PPAR subtype selectivity.<sup>20—22</sup>) 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<sup>23)</sup>: 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<sup>24)</sup> was applied to the conversion of **20a**—**c** into acrylic acid ethyl esters **21a**—

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*-



Fig. 2. Structures of Compounds 1—16

(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**.

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<sub>4</sub> 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**.<sup>25)</sup> Coupling between 2-nonylaminopyridine **18** and propanoic acid methyl esters **26a**—**d** afforded *N*-(2pyridinyl)-*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.



(a) 2-bromopyridine, Pd<sub>2</sub>(DBA)<sub>3</sub>, BINAP, *t*-BuOH, toluene, 105 °C, 70%; (b) 1,2-dibromoethane, Cs<sub>2</sub>CO<sub>3</sub>, THF, 65 °C, 33—57%; (c) (EtO)<sub>2</sub>P(O)CH(R)CO<sub>2</sub>Et, NaH, anhydrous THF, 0 °C to rt, 47—95%; (d) H<sub>2</sub>, Pd/C, EtOH, 79—97%; (e) **18**, Et<sub>3</sub>N, KI, 105 °C, 9—17%; (f) 2 N aq. NaOH, EtOH, THF, rt, 82—100%.

13 :  $R^1$  = OMe.  $R^2$  = OEt. position = m



(a) NaBH<sub>4</sub>, EtOH, rt, 81–95%; (b) Ac<sub>2</sub>O, DMAP, rt, 96–97%; (c) 1-methoxy-1-trimethylsilyloxypropene, or dimethylketene methyltrimethylsilyl acetal, Mg(ClO<sub>4</sub>)<sub>2</sub>, rt, 90–94%; (d) **18**, Et<sub>3</sub>N, KI, 105 °C, 5–13%; (e) 2 N aq. NaOH, EtOH, rt, 75–97%.







#### **Results and Discussion**

The binding affinity of the compounds for PPARs was evaluated with the CoA-BAP System (Microsystems).<sup>26)</sup> 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 PPAR $\alpha$ , PPAR $\gamma$  and PPAR $\delta$  were evaluated and the results are shown in Figs. 3, 4





Fig. 3. Binding Affinity for PPAR $\gamma$  of Compounds 1—16 at 0.1, 1.0 and 10  $\mu$ M

Values are the means of at least three experiments.



Fig. 4. Binding Affinity for PPAR $\alpha$  of Compounds 1—16 at 0.1, 1.0 and 10  $\mu$ M

Values are the means of at least three experiments.

and 5, respectively. GW7647<sup>27)</sup> (PPAR $\alpha$ ), rosiglitazone<sup>6)</sup> (PPAR $\gamma$ ) and GW501516<sup>28)</sup> (PPAR $\delta$ ) were used as reference compounds (Fig. 1).

The lead compound 1 showed relatively high affinity for PPAR $\gamma$  and little affinity for PPAR $\alpha$  and PPAR $\delta$  (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 PPAR $\gamma$  and PPAR $\alpha$ .<sup>15–19)</sup> Among compound 1, methyl 2, ethyl 3, ethoxy 4, and dimethyl 5, compound 4 showed the strongest affinity for PPAR $\gamma$ , so compound 4 was founded to be a potent and selective PPAR $\gamma$  ligand (Figs. 3—5). In addition, the PPAR $\gamma$  affinity of compound 3 was comparable to that of compound 1, whereas compound 3 displayed strong affinity for PPAR $\alpha$  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

1056



Fig. 5. Binding Affinity for PPAR $\delta$  of Compounds 1—16 at 0.1, 1.0 and 10  $\mu$ M

Values are the means of at least three experiments.

much higher affinity for PPAR $\alpha$  than 1 (Fig. 4). Furthermore, the affinity for PPAR $\gamma$  of 6 was lower than that of 1 (Fig. 3), and compound 6 exhibited little affinity for PPAR $\delta$  (Fig. 5). To find more potent PPAR $\alpha$  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 PPAR $\alpha$  and compound 8 displayed slightly weak affinity for PPAR $\gamma$  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 PPAR $\alpha$ ,  $\delta$ , and  $\gamma$ . 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 PPAR  $\alpha/\gamma$  dual agonist. Although the binding assay indicated that compound 4 is a selective PPAR $\gamma$  ligand, it showed potent dual-agonist activity toward PPAR $\alpha$  and PPAR $\gamma$ . 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<sub>50</sub> values and selectivity for PPAR $\alpha$  and PPAR $\gamma$ , and the introduction of ethyl group (compound 8) increased the activity and selectivity for PPAR $\alpha$ . Among these compounds, compound 8 showed the highest selectivity for PPAR $\alpha$ .

As compounds 3 and 4 were found to have relatively high PPAR $\gamma$  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

Nonyl N N N O R O H					
Compound	R	Position	EC <sub>50</sub> <sup><i>a</i>)</sup> (µм)		
			PPARγ	PPAR $\delta$	PPAR $\alpha$
1	Н	р	>10	>10	>10
3	Et	p	2.62	>10	3.68
4	OEt	p	0.32	>10	0.74
6	Н	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.<sup>33</sup>



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 PPAR $\gamma$  enhances adipocyte differentiation<sup>31)</sup> and increases insulin sensitivity, compounds **3** and **4** were subjected to a human hypodermic preadipocyte differentiation test.<sup>32)</sup> 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  $\mu$ M, and the activity of compounds **3** and **4** was found to be higher than that of rosiglitazone at 1  $\mu$ M 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 PPAR $\gamma$  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 PPAR $\gamma$  and little affinity for PPAR $\alpha$  and PPAR $\delta$ . Compound 3, which had an ethyl group at the C-2 position of propanoic acid, was found to be a PPAR $\alpha/\gamma$  dual ligand. Compound 6, the *meta* isomer of 1, has been shown to be a PPAR $\alpha$  ligand. The introduction of methyl (7) and ethyl (8) groups at the C-2 position of the propanoic acid of 6 further improved PPAR $\alpha$ -binding potency. In cell-based transactivation assay, compounds 3 and 4 showed dual-agonist activity toward PPAR $\alpha$  and PPAR $\gamma$ . Compound 6 was found to be a triple agonist and compound **8** proved to be a selective PPAR $\alpha$  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 (<sup>1</sup>H-NMR) were recorded on a JEOL JNM-LA500 spectrometer in solvent as indicated. Chemical shifts ( $\delta$ ) 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<sub>2</sub>(DBA)<sub>3</sub> (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<sub>2</sub>SO<sub>4</sub>. 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: <sup>1</sup>H-NMR (CDCl<sub>3</sub>, 500 MHz,  $\delta$ : 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<sub>2</sub>SO<sub>4</sub>. 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: <sup>1</sup>H-NMR (CDCl<sub>3</sub>, 500 MHz,  $\delta$ : 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<sub>2</sub>SO<sub>4</sub>. 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: <sup>1</sup>H-NMR (CDCl<sub>3</sub>, 500 MHz,  $\delta$ : 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), 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<sub>2</sub> 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: <sup>1</sup>H-NMR (CDCl<sub>3</sub>, 500 MHz,  $\delta$ : 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 1057

(toluene : CHCl<sub>3</sub> : AcOEt=45 : 5 : 3) gave 55 mg (11%) of **23a** as a colorless oil: <sup>1</sup>H-NMR (CDCl<sub>3</sub>, 500 MHz,  $\delta$ : 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<sub>27</sub>H<sub>40</sub>N<sub>2</sub>O<sub>3</sub> 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  $2 \times a$  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  $2 \times a$  aqueous HCl. The mixture was subjected to silica gel flash chromatography (CHCl<sub>3</sub>/MeOH=19/1) to give 44 mg (90%) of **1** as a colorless oil: <sup>1</sup>H-NMR (CDCl<sub>3</sub>, 500 MHz,  $\delta$ : 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<sup>+</sup>); HR-MS Calcd for C<sub>25</sub>H<sub>36</sub>N<sub>2</sub>O<sub>3</sub> 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; <sup>1</sup>H-NMR (CDCl<sub>3</sub>, 500 MHz,  $\delta$ : 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<sub>27</sub>H<sub>40</sub>N<sub>2</sub>O<sub>3</sub> 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; <sup>1</sup>H-NMR (CDCl<sub>3</sub>, 500 MHz,  $\delta$ : 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), 3.47 (2H, t, J=7.6 Hz), 3.05 (1H, dd, J=14.0, 4.5 Hz), (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<sub>27</sub>H<sub>40</sub>N<sub>2</sub>O<sub>4</sub> 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; <sup>1</sup>H-NMR (CDCl<sub>3</sub>, 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<sub>25</sub>H<sub>36</sub>N<sub>2</sub>O<sub>3</sub> 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; <sup>1</sup>H-NMR (CDCl<sub>3</sub>, 500 MHz,  $\delta$ : 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<sub>26</sub>H<sub>38</sub>N<sub>2</sub>O<sub>3</sub> 426.2882, Found; M<sup>+</sup> 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; <sup>1</sup>H-NMR (CDCl<sub>3</sub>, 500 MHz,  $\delta$ : 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<sub>27</sub>H<sub>40</sub>N<sub>2</sub>O<sub>3</sub> 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; <sup>1</sup>H-NMR (CDCl<sub>3</sub>, 500 MHz,  $\delta$ : ppm) 8.13 (1H, dd, *J*=4.9, 1.2 Hz), 7.44 (1H, dd, *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), 4.07 (1H, dd, *J*=7.3, 4.9 Hz), 3.90 (2H, m), 3.59 (1H, m), 3.47 (2H, t, *J*=7.0 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<sub>27</sub>H<sub>40</sub>N<sub>2</sub>O<sub>4</sub> 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; <sup>1</sup>H-NMR (CDCl<sub>3</sub>, 500 MHz,  $\delta$ : 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), 2.61 (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<sub>26</sub>H<sub>38</sub>N<sub>2</sub>O<sub>4</sub> 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; <sup>1</sup>H-NMR (CDCl<sub>3</sub>, 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), 3.86 (2H, 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<sub>28</sub>H<sub>42</sub>N<sub>2</sub>O<sub>4</sub> 470.3145, Found 470.3142.

**2-Ethoxy-3-{4-methoxy-3-[2-(nonylpyridin-2-ylamino)ethoxy]phenylpropanoic 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; <sup>1</sup>H-NMR (CDCl<sub>3</sub>, 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<sub>28</sub>H<sub>42</sub>N<sub>2</sub>O<sub>5</sub> 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<sub>4</sub> (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<sub>2</sub>SO<sub>4</sub>. The solvent was removed by evaporation *in vacuo.* 763 mg (81%) of **24a** was obtained as a white solid: <sup>1</sup>H-NMR (CDCl<sub>3</sub>, 500 MHz,  $\delta$ : 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<sub>2</sub>Cl<sub>2</sub> were added Ac<sub>2</sub>O (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<sub>2</sub>SO<sub>4</sub>. Purification by silica gel flash chromatography (AcOEt/*n*-hexane =1/5) gave 2.62 g (96%) of 25a as a light yellow oil: <sup>1</sup>H-NMR (CDCl<sub>3</sub>, 500 MHz,  $\delta$ : 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<sub>2</sub>Cl<sub>2</sub> 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 stirred for 1 d at room temperature. The reaction mixture was diluted with CH<sub>2</sub>Cl<sub>2</sub>. The CH<sub>2</sub>Cl<sub>2</sub> solution was washed with water and brine, and dried over Na<sub>2</sub>SO<sub>4</sub>. 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: <sup>1</sup>H-NMR (CDCl<sub>3</sub>, 500 MHz,  $\delta$ : 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: <sup>1</sup>H-NMR (CDCl<sub>3</sub>, 500 MHz,  $\delta$ : 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<sub>27</sub>H<sub>40</sub>N<sub>2</sub>O<sub>3</sub> 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; <sup>1</sup>H-NMR (CDCl<sub>3</sub>, 500 MHz,  $\delta$ : 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<sub>26</sub>H<sub>38</sub>N<sub>2</sub>O<sub>3</sub> 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-1trimethylsilyloxypropene: light yellow oil; <sup>1</sup>H-NMR (CDCl<sub>3</sub>, 500 MHz,  $\delta$ : 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<sub>27</sub>H<sub>40</sub>N<sub>2</sub>O<sub>3</sub> 440.3039, Found 440.3040.

**3-{4-Methoxy-3-[2-(Nonylpyridin-2-ylamino)ethoxy]phenyl}-2methylpropanoic Acid (11)** Compound **11** was prepared from **20c** using the procedure described for **2** in 3.3% yield: colorless oil; <sup>1</sup>H-NMR (CDCl<sub>3</sub>, 500 MHz,  $\delta$ : 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<sub>27</sub>H<sub>40</sub>N<sub>2</sub>O<sub>4</sub> 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 1methoxy-1-trimethylsilyloxypropene: colorless oil; <sup>1</sup>H-NMR (CDCl<sub>3</sub>, 500 MHz,  $\delta$ : 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<sub>28</sub>H<sub>42</sub>N<sub>2</sub>O<sub>4</sub> 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: <sup>1</sup>H-NMR (CDCl<sub>3</sub>, 500 MHz,  $\delta$ : 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), 4.25 (2H, q, J=7.0 Hz), 4.21 (2H, t, J=5.8 Hz), 3.94 (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<sub>27</sub>H<sub>48</sub>N<sub>2</sub>O<sub>3</sub> 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<sub>3</sub>/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; <sup>1</sup>H-NMR (CDCl<sub>3</sub>, 500 MHz,  $\delta$ : 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*=6.4, 5.5 Hz), 6.50 (1H, d,

 $\begin{array}{l} J{=}8.5\,{\rm Hz}),\ 6.29\ (1{\rm H},\ d,\ J{=}16.1\,{\rm Hz}),\ 4.22\ (2{\rm H},\ t,\ J{=}5.8\,{\rm Hz}),\ 3.95\ (2{\rm H},\ t,\ J{=}5.8\,{\rm Hz}),\ 3.46\ (2{\rm H},\ t,\ J{=}8.2\,{\rm Hz}),\ 1.65{----}1.61\ (2{\rm H},\ m),\ 1.34{----}1.24\ (12{\rm H},\ m),\ 0.88\ (3{\rm H},\ t,\ J{=}7.0\,{\rm Hz});\ Anal.\ Calcd\ for\ C_{25}{\rm H}_{34}{\rm N}_{2}{\rm O}_{3}{\rm :}\ C,\ 73.14{\rm ;}\ H,\ 8.35{\rm ;}\ N,\ 6.82.\ Found:\ C,\ 73.23{\rm ;}\ H,\ 8.46{\rm ;}\ N,\ 6.89. \end{array}$ 

**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; <sup>1</sup>H-NMR (CDCl<sub>3</sub>, 500 MHz,  $\delta$ : 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<sub>26</sub>H<sub>36</sub>N<sub>2</sub>O<sub>4</sub> 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<sup>TM</sup> 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**<sup>30)</sup> 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<sub>2</sub> 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  $\beta$ -galactosidase activities were assayed using a 1420 ARVO<sup>TM</sup> MX multilabel counter (ParkinElmer, Boston, MA, U.S.A.). DNA cotransfection experiments included 58 ng of reporter plasmid, 12 ng of CMX- $\beta$ -galactosidase, and 18 ng of each receptor expression plasmid per well in a 96-well plate. Luciferase data were normalized to an internal  $\beta$ -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<sub>2</sub>. 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<sup>TM</sup> multilabel counter (ParkinElmer, Boston, MA, U.S.A.) after staining with Lipidos Liquid<sup>®</sup> (TOYOBO Co., Ltd, Osaka, Japan).

## **References and Notes**

- 1) Nuclear Receptors Nomenclature Committee. Cell, 97, 161–163 (1999).
- Willson T. M., Brown P. J., Sternbach D. D., Henke B. R., J. Med. Chem., 43, 527—550 (2000).
- Kersten S., Desvergne B., Wahli W., *Nature* (London), 405, 421–424 (2000).
- 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).
- Francis G. A., Annicotte J. S., Auwerx J., Curr. Opin. Parmacol., 3, 186—191 (2003).
- 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).
- 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).
- Cronet P., Petersen J. F., Folmer R., Blomberg N., Sjoblom K., Karlsson U., Lindstedt E. L., Bamberg K., *Structure*, 9, 699–706 (2001).
- Nomura M., Tanase T., Ide T., Tsunoda M., Suzuki M., Uchiki H., Murakami K., Miyachi H., J. Med. Chem., 46, 3581–3599 (2003).
- 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.