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Design and synthesis of oxime ethers of α-acyl-β-phenylpropanoic acids as PPAR dual agonists

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Abstract—Oxime ethers of α-acyl-β-phenylpropanoic acids were prepared to apply as PPARα and γ dual agonists. Among them, compound **11l** proved to exhibit potent in vitro activities with EC_{50} of 19 and 13 nM in PPARα and γ, respectively. It showed better glucose lowering effects than rosiglitazone **1** and ameliorated the lipid profile like plasma triglyceride in *db*/*db* mice model.

Type II diabetes has emerged as one of the biggest problems facing the world today. High glucose level in the blood caused hyperglycemia and insulin resistance, which resulted in the deterioration of the pancreatic β-cell function.[1] Hyperglycemia should be controlled in a careful manner, otherwise higher morbidity and mortality have been found in the related diseases like retinopathy, nephropathy, neuropathy, dyslipidemia, coronary heart disease, hypertension, and obesity.[2] Many targets have been investigated to cure type II diabetes like dipeptidyl peptidase IV,[3] peroxisome proliferator-activated receptors[4] (PPARs), GLP-1 analog,[5] glucokinase,[6] and so on. Among those targets, peroxisome proliferator-activated receptors (PPARs) are known as a good target to lower plasma glucose and to improve metabolic syndrome. PPARs are members of the nuclear receptor superfamily, which consists of three isoforms, PPARα, PPARγ, and PPARδ.[7] Heterodimer of PPARs with retinoid X receptors binds to peroxisome proliferator response element (PPRE) in the promoter region with cofactors, which results in the regulation of many target genes. PPARγ is largely expressed in adipocytes and macrophages.[8] In adipocyte tissue, PPARγ promotes the differentiation and the lipid storage. PPARγ agonists like rosiglitazone **1** and pioglitazone **2** ameliorate insulin sensitivity. In contrast with PPARγ, PPARα is expressed in liver and heart. Activation of PPARα induces fatty acid oxidation in the liver, and reduces serum triglyceride (TG) and increases HDL cholesterol.

PPARγ agonists, rosiglitazone **1** and pioglitazone **2**, with thiazolidinediones (TZDs) have been marketed with a sale of 1.9 billion dollars and 2.0 billion dollars, respectively, in 2004.[9] Recently, 3-year clinical study indicated that pioglitazone reduced the risk of myocardial infarction or stroke significantly.[10] However, long clinical treatment (3–4 months) of TZDs is required to reach their full glucose reduction effect.[11] Although these agents have glucose lowering effect and improve the lipid profile like TG, LDL cholesterols and HDL cholesterol, the crucial side effects[12] like congestive heart failure, edema, fluid retention, and weight gain still remain unsolved. Until now, a large number of PPARα, γ dual agonists have been investigated in many research groups but the successful PPARα, γ dual agonists including tesaglitazar **3** (AstraZeneca) and muraglitazar[13] **4** (BMS-Merck) have not been marketed. Nevertheless, PPARα, γ dual agonist would be well suited for treatment of diabetes and cardiovascular disease as long as it could reduce side effects as mentioned above.

Many PPARα, γ dual agonists contained α-alkoxy acids, α-carbamate acids or thiazolidinediones as shown in tesaglitazar **3**, muraglitazar **4**, rosiglitazone **1**, and pioglitazone **2**. Acid group in PPAR agonists is known

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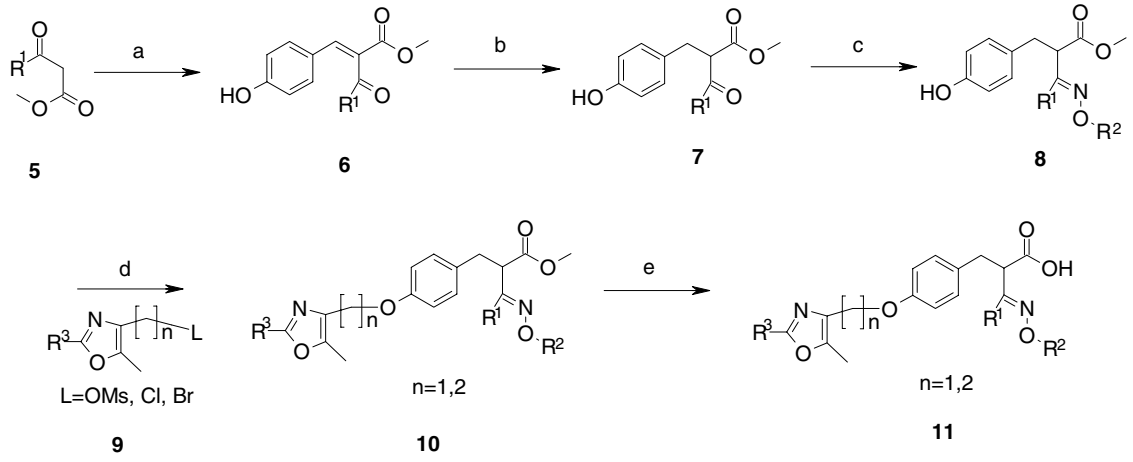
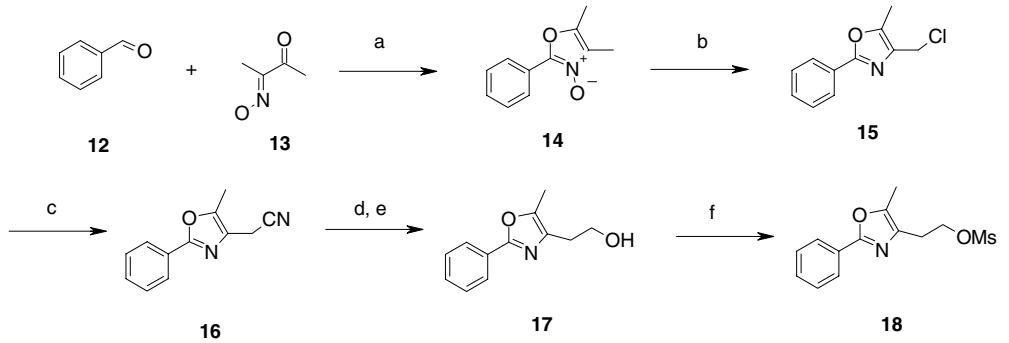
to play a key role in interacting with PPARα and γ.[4b] In an effort to search for the new PPAR agonist, this led us to investigate isostere of α-substituent in acid. At the beginning, introduction of oxime group at α-position in acid was expected to provide a working scaffold for the efficacious PPAR agonist. Therefore, we designed a series of new phenylpropanoic acids containing the oxime functionality as PPARα, γ dual agonists. The synthesis of phenylpropanoic acids having the oxime group is described in Scheme 1. Most of the compounds were synthesized in the recemic forms. Knoevenagel reaction was employed for the synthesis of **6**. Condensation of β-keto ester **5** and 4-hyroxy benzaldehyde was carried out with catalytic amounts of piperidine and acetic acid in benzene using Dean-Stark trap. The yield of condensation was excellent (90–95%) and this type of reaction allowed the modification of R^1 group in **11** by simply changing the commercially available β-keto esters **5**. Hydrogenation of **6** and the following reaction with *O*-alkyl hydroxyl amine afforded phenols **8**. The generated geometric (*E*)- and (*Z*)-isomers were not isolated in this step but separated in the final step using HPLC. Alkylation of phenols **8** with the commonly used oxazoles **9** was carried out by refluxing with Cs_2CO_3 in acetonitrile to furnish esters **10** in good yields. Oxazoles **9** could be prepared by using the published procedures.[14] Scheme 2 shows a simple synthetic route to one of oxazoles **9** with phenyl group as R^3 position. Butane-2,3-dione mono-oxime was reacted with benzaldehyde and hydrochloric acid in dioxane to afford oxazole *N*-oxide **14**. Treatment of *N*-oxide **14** with phosphorus oxychloride in chloroform gave chloride **15**. For the one-carbon elongation, chloride **15** was subjected to sodium cyanide in DMF at 80 °C and then alkaline hydrolysis with KOH in aqueous THF to furnish the corresponding acid. Reduction of the acid with borane dimethylsulfide complex and the following reaction with methanesulfonyl chloride afforded mesylate **18**. Ester **10** was hydrolyzed with sodium hydroxide in aqueous THF and methanol, and then the (*E*)- and (*Z*)-isomers were separated with HPLC to give pure acid **11**.[15] Among them, spectral data for the representative **11l** are given below. Spectra for **11l**: ^{1}H NMR ($CDCl_3$) δ 7.94–7.92 (1H, m), 7.38–7.33 (3H, m), 6.97 (1H, d, J = 8.6), 6.78 (1H, d, J = 8.6), 4.81(2H, s), 3.84–3.78 (1H, m), 3.79–3.73 (1H, m), 3.38 (1H, dd, J = 12, 4.9 Hz), 3.15 (1H, dd, J = 14, 4.9 Hz), 2.84 (1H, dd, J = 14, 12 Hz), 2.29 (3H, s), 1.75 (3H, s), 1.33–1.27 (2H, m), 0.66 (3H, t, J = 7.6 Hz) ^{13}C NMR ($CDCl_3$) δ 178.68, 159.90, 159.35, 156.87, 157.06, 132.43, 132.12, 130.08, 129.97,



Scheme 1. Reagents and conditions: (a) 4-hydroxy benzaldehyde, AcOH (cat), piperidine (cat), benzene, Dean-Stark trap, 90–95%; (b) MeOH/ethyl acetate 10:1, H_2, $Pd(OH)_2$, 95%; (c) MeOH, NaOAc, $HCl{\cdot}H_2N{-}OR_2$, 90%; (d) Cs_2CO_3, CH_3CN, reflux, n = 1, 80–90%, n = 2, 50–60%; (e) THF/MeOH/1 N NaOH 1:1:1, 90%.



Scheme 2. Reagents and conditions: (a) 4 M HCl in dioxane, rt, 99%; (b) $CHCl_3$, $POCl_3$, reflux, 99%; (c) NaCN, DMF, 80 °C, 95%; (d) KOH, aq. THF, reflux; (e) $BH_3{\cdot}SMe_2$, THF, rt, 68%; (f) $MeSO_2Cl$, Et_3N, CH_2Cl_2, 0 °C, 99%.

128.72, 127.48, 126.15, 114.56, 74.72, 62.23, 55.82, 34.84, 22.23, 12.68, 10.47, 10.32 mass (EI) *m*/*z* 437 (M^++1) (Fig. 1).

Compound **11** in our hand were screened for PPARα and γ agonist activity on PPAR-GAL4 chimeric receptor in transfected HepG2 cells.[16] Rosiglitazone **1**, tesaglitazar **3**, and GW9578[17] were used as PPARγ, PPARα/γ, and PPARα agonist controls. In Table 1, we summarized functional transactivation data of all the compounds. We initiated to synthesize **11a**, which was found to be a potent agonist in PPARα and γ. It was so encouraging that we decided to scrutinize the influence of R^2 groups. As the size of the substituent increased, PPARα and γ activities appeared to improve. Modification of the chain in R^2 group indicated that the optimal size of the substituent was *n*-butyl group in this study. As shown in Table 1, **11e** had sub-nanomolar activity on PPARγ and 1.3 nM on PPARα. When ethyl group was introduced to R^1 in **11g** and **11h**, they

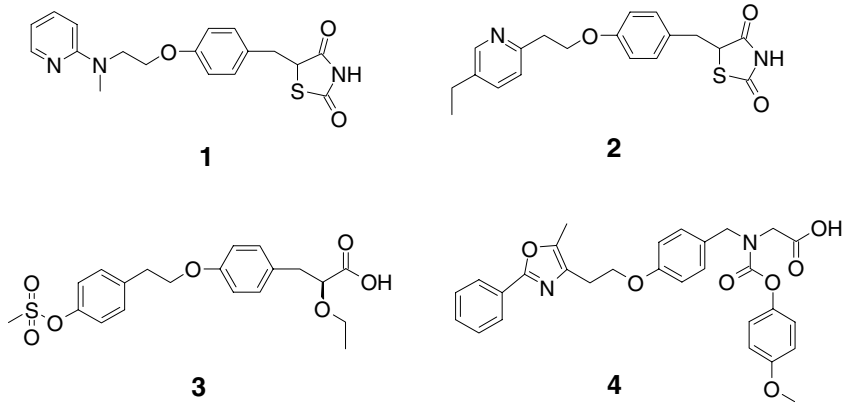
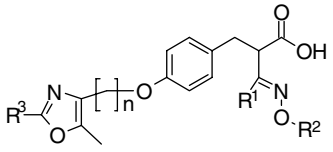


Figure 1. Structures of PPAR agonists.

Table 1. In vitro activity of oxime ethers of α-acyl-β-phenylpropanoic acids **11**[a]



Compound	Geometric isomer	*N*	R^1	R^2	R^3	EC_{50}(nM)[b] of hPPARγ	EC_{50}(nM)[b] of hPPARα
11a	*E*	2	Me	Me	Ph	2.1	19
11b	*E*	2	Me	Et	Ph	1.0	12
11c	*E*	2	Me	*n*-Pr	Ph	0.55	4.9
11d	*E*	2	Me	CH_2CHMe_2	Ph	0.3	2.2
11e	*E*	2	Me	*n*-Bu	Ph	0.19	1.3
11f	*E*	2	Me	CH_2-cyclopropyl	Ph	0.36	3.1
11g	*E*/*Z* = 3/1	2	Et	Me	Ph	0.31	32
11h	*E*/*Z* = 4/1	2	Et	Et	Ph	0.1	26
11i	*E*	2	Me	4-*F*-PhenylCH_2	Ph	0.54	10.6
11j	*Z*	2	Me	4-*F*-PhenylCH_2	Ph	5	596
11k	*E*/*Z* = 5/1	1	Me	Et	Ph	4	91
11l	*E*	1	Me	Pr	Ph	13	19
11m	*E*	1	Me	Et	4-*F*-Phenyl	6.8	12
11n	*E*	1	Me	*n*-Pr	4-*F*-Phenyl	28	11.7
11o	*E*	1	Me	2-*F*-Ethyl	4-*F*-Phenyl	7.7	23
11p	*E*	2	Me	Et	4-*F*-Phenyl	3.0	11.5
11q	*E*	2	Me	Pr	4-*F*-Phenyl	2.1	4
11r	*E*	2	Me	2-*F*-Ethyl	4-*F*-Phenyl	2.9	3.5
11s	*E*/*Z* = 5/1	1	Me	Et	*i*-Propyl	48	1237
11t	*E*/*Z* = 5/1	1	Me	Pr	*i*-Propyl	5	105
11u	*E*	2	Me	Et	*i*-Propyl	20	139
11v	*E*	2	Me	Pr	*i*-Propyl	5.2	10
Rosiglitazone **1**						82	
Tesaglitazar **3**						3528	9798
GW9578							78

[a] Agonist activities were measured in human PPAR-GAL4 chimeric HepG2 cells using the published procedure.[16]
[b] Concentration of the test compounds which produced 50% of the maximal reporter activity.

retained good PPARα and γ activities. Comparison of the activities of the geometric isomers **11i** and **11j** demonstrated that (*E*)-isomer had more potent activities in PPARα and γ than (*Z*)-isomer. **11k** and **11l,** having shorter linkers**,** exhibited well-balanced activities as well. 4-Fluorophenyl methyl group was installed in **11m**–**11r** because phenyl group is sometimes metabolically vulnerable. They were prepared for the pharmacokinetic studies and in vivo experiment. We also attached isopropyl group as R^3. Compounds **11s** and **11t** with a short linker turned out to decrease PPARα and γ activities as compared with compounds **11u** and **11v**. In general, a series of oxime ethers of α-acyl-β-phenylpropanoic acids **11** exhibited excellent PPARα and γ activities, which provide a useful scaffold for PPARα and γ dual agonist. Finally, X-ray crystallographic data of **11l** were obtained to verify which enantiomer was potent one in PPARα and γ, unequivocally. (R)-**11l** was found to be a potent enantiomer as shown in Figure 2. Therefore, we isolated a pure enantiomer (R)-**11l** by utilizing chiral HPLC with a Chiracel® OD-RH column, which revealed that it had EC_{50} of 11 and 5 nM in PPARα and γ.

To investigate the interactions of these compounds with the protein, the compound **11l** was chosen to co-crystallize with PPARα LBD (ligand binding domain) and the co-activator peptide fragment (SRC-1).

The U-shaped compound **11l** binds to the PPARα LBD using four hydrogen bonds and many van der Waals contacts. The carboxylate group of the compound **11l** forms hydrogen bonds with Ser280 on helix 3, Tyr314 on helix 5, and Tyr464 on helix 12 as in the structure of tesaglitazar **3**.[18] Its oxime group is located in the so-called 'benzophenone' pocket and makes contacts with Ile272, Phe273, Cys276, and Ile354. The oxime-binding pocket can accommodate various sizes of functional groups, which correlated well with SAR shown in Table 1. The phenyl oxazole group fits into a hydrophobic pocket formed by helix 2′; helix 3 and the β sheet,

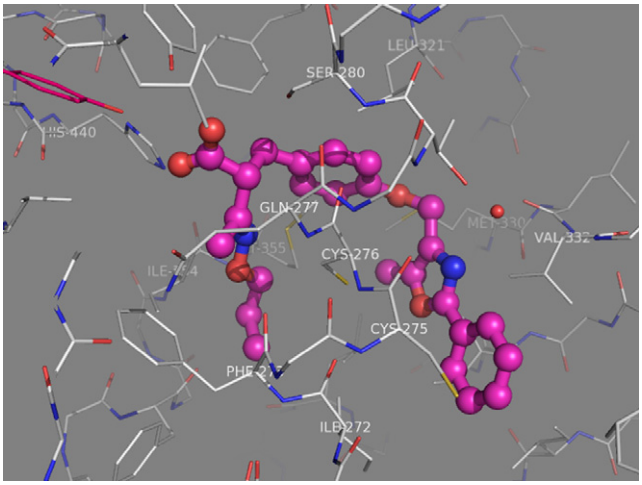


Figure 2. Crystal structure of the PPARα LBD in complex with compound **11l**. Compound **11l** is shown in purple and Tyr464 from helix 12 is highlighted in red. All of the residues within 4 Å from the compound are labeled.

Table 2. Pharmacokinetic data of **11l** in SD rats[a]

Pharmacokinetic parameters	po	iv
Dose (mg/kg)	10	5
AUC_{0-24h}(μg min/ml)	9708	6773
Cmax (μg/ml)	22.6	
Tmax (min)	30	
T1/2 (min)	483	355.9
Clearance (ml/min/kg)		0.8
Vd (ml/kg)		329
Bioavailability (%)	75.5	

[a] $n = 2$.

Table 3. In vivo data of compound **11l** in *db*/*db* mice[20]

Compound	Dose (mg/kg)	Plasma glucose[a] (%)	Plasma TG[a] (%)
Rosiglitazone **1**[b]	10	65	65
Tesaglitazar **3**[c]	1	68	38
11l[c]	0.1	8	43
	1	56	41
	10	73	58

[a] Reduction of plasma glucose and plasma TG were calculated as the percent of reduction and increase with respect to control value.
[b] Blood samples were collected after 12 days.
[c] Blood samples were collected for glucose after 11days and for TG after 14 days.

and the nitrogen atom of the oxazole ring makes a water-mediated hydrogen bond with Thr279.[19] The complex structure of the compound **11l** with PPARγ LBD revealed similar binding interactions.

Based on the cell-based activities and the pharmacokinetic studies in rats, **11l** was selected to adjust the in vivo experiment in male *db*/*db* mice for further evaluation. Its pharmocokinetic behavior in Sprague–Dawley (SD) rats is described in Table 2. When dosed orally at 0.1, 1, 10 mg/kg in *db*/*db* mice[20] for 14 days, compound **11l** exhibited a does-dependent decrease in plasma glucose and plasma triglyceride (TG) as depicted in Table 3. Treatment of 10 mg/kg of **11l** showed a glucose reduction of 73% as compared with that of 65% and 68% at 10 mg/kg of rosiglitazone **1** and 1 mg/kg of tesaglitazar **3**, respectively. At the same time, **11l** reduced plasma TG by 58% at a dose of 10 mg/kg. Glucose lowering effect was also found at a dose of 1 mg/kg of **11l.** Consequently, compound **11l** displayed better glucose lowering effect than rosiglitazone **1**, whereas it showed diminished efficacy as compared with tesaglitazar **3**. Compound **11l** turned out to have glucose lowering effect and ameliorate lipid profile like TG in *db*/*db* mice model. In conclusion, we found that a series of oxime ethers of α-acyl-β-phenylpropanoic acids were potent agonists of PPARα and γ, and exhibited good in vivo efficacy, which will be further developed.

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References and notes

1. (a) Rhodes, C. J. *Science* **2005**, *307*, 380; (b) Berger, J.; Moller, D. E. *Ann. Rev. Med.* **2002**, *53*, 409; (c) Bell, G. I. *Nature* **2001**, *414*, 788.
2. Zimmet, P.; Alberti, K. G. M. M.; Shaw, J. *Nature* **2001**, *414*, 782.
3. (a) Kim, D.; Wang, L.; Beconi, M.; Eiermann, G. J.; Fisher, M. H.; He, H.; Hickey, G. J.; Kowalchick, J. E.; Leiting, B.; Lyons, K.; Marsilio, F.; McCann, M. E.; Patel, R. A.; Petrov, A.; Scapin, G.; Patel, S. B.; Roy, R. S.; Wu, J. K.; Wyvratt, M. J.; Zhang, B. B.; Zhu, L.; Thornberry, N. A.; Weber, A. E. *J. Med. Chem.* **2005**, *48*, 141; (b) Villhauer, E. B.; Brinkman, J. A.; Naderi, G. B.; Burkey, B. F.; Dunning, B. E.; Prasad, K.; Mangold, B. L.; Russell, M. E.; Hughes, T. E. *J. Med. Chem.* **2003**, *46*, 2774; (c) Gill, I.; Patel, R. *Bioorg. Med. Chem. Lett.* **2006**, *16*, 705.
4. (a) Savkur, R. S.; Miller, A. R. *Expert Opin. Invest. Drugs* **2006**, *15*, 763; (b) Henke, B. R. . *J. Med. Chem.* **2004**, *47*, 4118; (c) Staels, B.; Fruchart, J. *Diabetes* **2005**, *54*, 2460.
5. Joy, S. V.; Rodgers, P. T.; Scates, A. C. *Ann. Pharmacother.* **2005**, *39*, 110.
6. Matschinsky, F. M. *Diabetes* **2002**, *51*, S394.
7. Willson, T. M.; Brown, P. J.; Sternbach, D. D.; Henke, B. R. *J. Med. Chem.* **2000**, *43*, 527.
8. Semple, R. K.; Chatterjee, V. K. K.; O'Rahily, S. *J. Clin. Invest.* **2006**, *116*, 581.
9. Commercial insight: Antidiabetics; Dataminitor, June 2005.
10. Dormandy, J. A.; Charbonnel, B.; Eckland, D. J. A.; Erdmann, E.; Massi-Benedetti, M.; Moules, I. K.; Skene, A. M.; Tan, M. H.; Lefèbvre, P. J.; Murray, G. D.; Standl, E.; Wilcox, R. G.; Wilhelmsen, L.; Betteridge, J.; Birkeland, K.; Golay, A.; Heine, R. J.; Korányi, L.; Laakso, M.; Mokáň, M.; Norkus, A.; Pirags, V.; Podar, T.; Scheen, A.; Scherbaum, W.; Schernthaner, G.; Schmitz, O.; Škrha, J.; Smith, U.; Tatoň, J. *Lancet* **2005**, *366*, 1279.
11. Boden, G.; Zhang, M. *Expert Opin. Invest. Drugs* **2006**, *15*, 243.
12. Bloomgarden, Z. T. *Diabetes Care* **2005**, *28*, 2.
13. Nissen, S. E.; Wolski, K.; Topol, E. *JAMA* **2005**, *294*, 2581.
14. Goto, Y.; Yamazaki, M.; Hamana, M. *Chem. Pharm. Bull.* **1971**, *19*, 2050.
15. All compounds described in this paper gave consistent ^{1}H NMR and LC/MS data. Synthetic experimental procedures and spectral data can be found in the patent: Kim, G. T.; Koh, J. S.; Han, H. O.; Kim, S. H.; Kim, K; Chung H.; Kim, Y. C.; Kim, M.; Koo, K. D.; Yim, H. J.; Hur, G.; Lee, S. H.; Lee, C.; Woo, S. H. WO 2005040127.
16. Bility, M.; Thompson, J.; McKee, R. H.; David, R. M.; Butala, J. H.; Heuvel, J. P. V.; Peters, J. M. *Toxicol. Sci.* **2004**, *82*, 170.
17. Brown, P. J.; Winegar, D. A.; Plunket, K. D.; Moore, L. B.; Lewis, M. C.; Wilson, J. G.; Sundseth, S. S.; Kolbe, C. S.; Wu, Z.; Chapman, J. M.; Lehmann, J. M.; Kliewer, S. A.; Wilson, T. M. *J. Med. Chem.* **1999**, *42*, 3785.
18. Cronet, P.; Petersen, J. F. W.; Folmer, R.; Blomberg, N.; Sjöblom, K.; Larlsson, U.; Lindstedt, E.-L.; Bamberg, K. *Structure* **2001**, *9*, 699.
19. The coordinates have been deposited in the Protein Data Bank: PDB ID 2NPA.
20. Berger, J.; Leibowitz, M. D.; Doebber, T. W.; Elbrecht, A.; Zhang, B.; Zhou, G.; Biswas, C.; Cullinan, C. A.; Hayes, N. S.; Li, Y.; Tanen, M.; Ventre, J.; Wu, M. S.; Berger, G. D.; Mosley, R.; Marquis, R.; Santini, C.; Sahoo, S. P.; Tolman, R. L.; Smith, R. G.; Moller, D. E. *J. Biol. Chem.* **1999**, *274*, 6718.