Available online at www.sciencedirect.com
SCIENCE DIRECT
Bioorganic & Medicinal Chemistry Letters 16 (2006) 4674–4678
Bioorganic & Medicinal Chemistry Letters

Arylpropionylpiperazines as antagonists of the human melanocortin-4 receptor

Wanlong Jiang,[a] Fabio C. Tucci,[a] Caroline W. Chen,[a] Melissa Arellano,[a] Joe A. Tran,[a] Nicole S. White,[a] Dragan Marinkovic,[a] Joseph Pontillo,[a] Beth A. Fleck,[b] Jenny Wen,[d] John Saunders,[a] Ajay Madan,[d] Alan C. Foster[c] and Chen Chen[a,*]

[a]*Department of Medicinal Chemistry, Neurocrine Biosciences, Inc., 12790 El Camino Real, San Diego, CA 92130, USA*
[b]*Department of Pharmacology, Neurocrine Biosciences, Inc., 12790 El Camino Real, San Diego, CA 92130, USA*
[c]*Department of Neuroscience, Neurocrine Biosciences, Inc., 12790 El Camino Real, San Diego, CA 92130, USA*
[d]*Department of Preclinical Development, Neurocrine Biosciences, Inc., 12790 El Camino Real, San Diego, CA 92130, USA*

Received 3 May 2006; revised 23 May 2006; accepted 30 May 2006
Available online 13 June 2006

Abstract—A series of 3-arylpropionylpiperazines were synthesized as antagonists of the melanocortin-4 receptor. Their potency was found to be increased by replacing the α-methyl substituent of the initial lead **11** with a larger *s*-Bu or *i*-Bu group. Further potency enhancement was observed when a glycine or β-alanine was incorporated onto the benzylamine. Some compounds demonstrated good potency, moderate selectivity, and oral bioavailability.

The melanocortin-4 receptor (MC4R) is a member of the G-protein-coupled receptor (GPCR) superfamily, which plays an important role in the regulation of feeding behavior.[1] MC4R antagonists have been shown to reverse lean body mass loss as well as block the reduction of food intake in animal models, suggesting the possible use for the treatment of cancer cachexia.[2,3] In addition, recent studies have also shown that MC4R is involved in the pathophysiology of anxiety and depression.[4] Therefore, a potent and selective MC4R antagonist has the potential to treat these diseases (Fig. 1).

Several classes of non-peptide MC4R antagonists have been reported. The benzamidine **1** has been shown to be efficacious in a mouse cachexia model.[5] Piperazines such as **2** have demonstrated anxiolytic-like and antidepressant-like activities.[6] Compound **3** potently inhibits NDP-MSH binding to the MC4 receptor.[7] In addition, guanidines such as **4** are potent MC4R antagonists.[8] We have previously reported that a series of β-Ala-D(2,4-Cl)Phe dipeptide derivatives, such as **5**, are functional antagonists and exhibit anti-cachexia activity in a rodent animal model.[9]

By screening a set of compounds designed to target GPCRs, using a radio-labeled binding assay, a lead compound was identified.[10] Thus, **11** possessed a K_i of 490 nM in competition with [^{125}I]NDP-MSH binding to cell membranes stably expressing the human melanocortin-4 receptor. A detailed structure–activity study was then conducted to improve its in vitro potency. Herein, we report on the SAR at both the substituted phenylpropionyl group and the benzylamine of the lead compound **11**.

A general synthetic method was developed to quickly synthesize analogs of **11** (Scheme 1). The piperazine-benzaldehyde **7**, obtained from the corresponding fluorobenzene **6**, was converted to the imines **8** using racemic *tert*-butanesulfinamide. Addition of various alkyllithium reagents to **8** afforded the sulfinamides **9**, which were selectively deprotected at the Boc-group, using trifluoroacetic acid, to give the piperazines **10**. Coupling reactions of **10** with 3-(2,4-dichlorophenyl)propionyl chloride, followed by deprotection with HCl, provided the racemic products **11**–**14** and **18**–**19** in good yields.

Keywords: Melanocortin; Antagonist; Arylpropinylpiperazine, synthesis.
* Corresponding author. Tel.: +1 858 617 7600; fax: +1 858 617 7967; e-mail: cchen@neurocrine.com

0960-894X/$ - see front matter © 2006 Elsevier Ltd. All rights reserved.
doi:10.1016/j.bmcl.2006.05.088

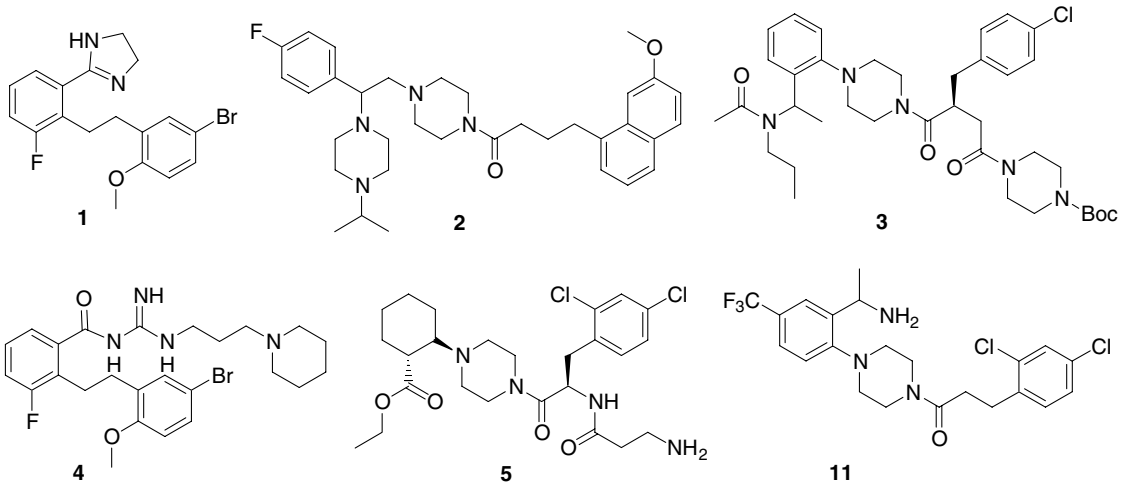
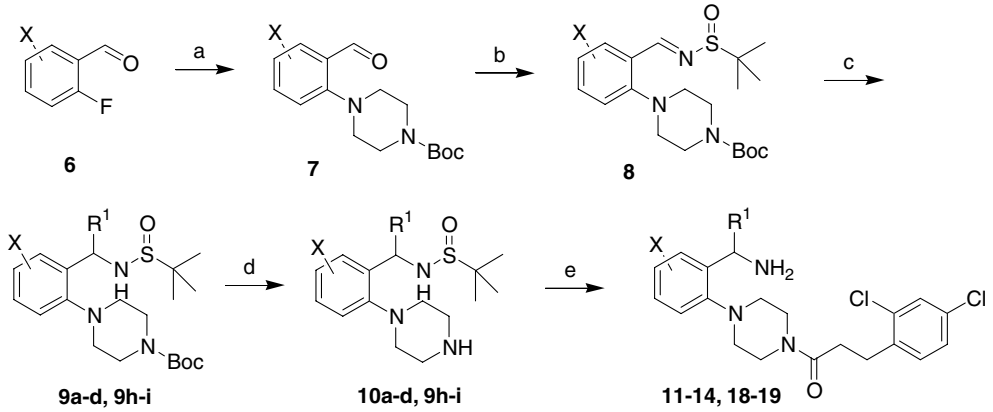


Figure 1. Some small molecule MC4R antagonists.



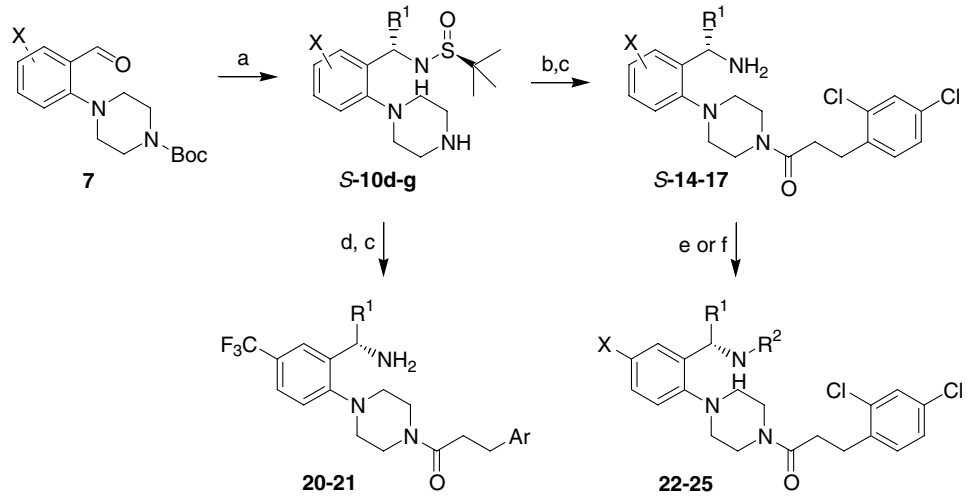
Scheme 1. Reagents and conditions: (a) *N*-Boc-piperazine/DMF/110–140 °C, 32–92%; (b) *t*-BuSONH$_2$/Ti(OEt)$_4$/THF, rt, 90–99%; (c) R^{1}Li/Me$_3$Al/THF, −78 °C, 50–80%; (d) TFA/CH_2Cl_2; (e) 2,4-ClPhCH_2CH_2COOH/EDC/DMF/CH_2Cl_2; then HCl/MeOH, 80–90%.

Alternatively, the *S*-configured compounds *S*-**14**–**17** were synthesized using *S*-*tert*-butanesulfinamide as shown in Scheme 2.[11] **20** and **21** were obtained by coupling reactions performed on *S*-**10e** and *S*-**10f**, respectively, with various arylpropionic acids in the presence of EDC. Reductive alkylation of the primary amines *S*-**14**–**15** with various aldehydes, in the presence of a reducing agent, or coupling reactions of *S*-**14**-**15** with *N*-Boc-glycine or *N*-Boc-alanine, gave the products **22**-**25**. In the cases where the N-side chain contained a Boc-protecting group, a TFA treatment was required before final purification through a preparative HPLC system.[12]

All compounds were tested in a ligand binding assay as previously described,[13] and the results are listed in Tables 1–3. The α-*n*-butyl derivative **12**, the isopropyl analog **13**, and the *s*-butyl compound **14** were about 3-fold better than **11** in binding affinity. The *S*-isomer of **14** had a K_i value of 75 nM, which was 2-fold better than the mixture, suggesting that the *S*-enantiomer is the more active species. The *S*-configured isobutyl analog (*S*-**15**, K_i = 74 nM), as well as the 6- and 4-fluoro derivatives *S*-**16** and *S*-**17**, displayed comparable binding affinity. The ethoxymethyl compound **18** was less potent than the *n*-butyl analog **12**, while the benzoxymethyl compound **19** only possessed micromolar binding affinity (Table 1).

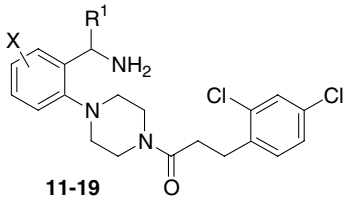
A comprehensive survey of a set of substituted phenylpropionyl groups did not result in compounds with improved binding affinities relative to *S*-**14** or *S*-**15**. Thus, the 4-chlorophenyl analog **20a** and the 2-chloro-4-methoxy compound **20b** displayed comparable potency to *S*-**14**, while the 2-chloro-3,4-dimethoxy compound **20c** (K_i = 2500 nM) had low binding affinity. A more detailed study on analogs of compound *S*-**15** resulted in compounds **21a**–**u**, which displayed low potency (Table 2).

We then introduced a group on the benzylamino nitrogen of *S*-**14** and *S*-**15**. While the *N*-ethyl compound (**22a**, K_i = 92 nM) exhibited similar binding affinity to its parent, other alkylations gave derivatives with lower potency (**22b**–**f**). When an additional amine group was incorporated, the compounds showed improved potency in some cases (**22g**–**k**). For example, the *N*-(aminoethyl) **22g** had a K_i value of 56 nM, and the *N*-(3-aminopropyl) analog (**22k**, K_i = 30 nM) had about 2-fold improvement in binding affinity over its parent (Table 3).



Scheme 2. Reagents and conditions: (a) i–*S*-*t*-BuSONH$_2$/Ti(OEt)$_4$; ii—R^{1}Li/Me$_3$Al/THF, −78 °C, 50–80%; iii—TFA/CH$_2$Cl$_2$; (b) 2,4-ClPhCH$_2$CH$_2$COOH/EDC/DMF/CH$_2$Cl$_2$, 80–90%; (c) HCl/MeOH; (d) ArCH$_2$CH$_2$COOH/EDC/DMF/CH$_2$Cl$_2$, 80–90%; (e) aldehyde/NaB(OAc)$_3$H/CH$_2$Cl$_2$, 80–90%; (f) *N*-Boc-glycine or *N*-Boc-β-alanine/EDC/DMF/CH$_2$Cl$_2$; then TFA/CH$_2$Cl$_2$, 70–80%.

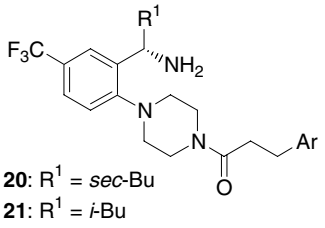
Table 1. SAR of α-alkyl benzylamines at the human MC4R



11-19

Compound	X	R^1	K_i (nM)
11	4-CF$_3$	Me	490
12	4-CF$_3$	*n*-Bu	190
13	4-CF$_3$	*i*-Pr	140
14	4-CF$_3$	*s*-Bu	160
***S*-14**	4-CF$_3$	*s*-Bu	75
***S*-15**	4-CF$_3$	*i*-Bu	74
***S*-16**	6-F	*i*-Bu	110
***S*-17**	4-F	*i*-Bu	94
18	4-F	EtOCH$_2$	690
19	4-F	BnOCH$_2$	1000

Table 2. SAR of aryl propionyl group at the human MC4R



20: R^1 = *sec*-Bu
21: R^1 = *i*-Bu

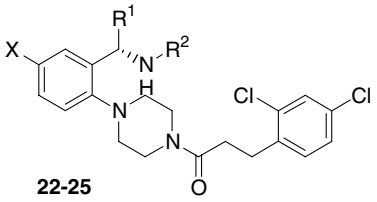
Compound	R^1	Ar	K_i (nM)
20a	*s*-Bu	4-ClPh	86
20b	*s*-Bu	2-Cl,4-MeOPh	57
20c	*s*-Bu	2-Cl,3,4-MeOPh	2500
21a	*i*-Bu	2-FPh	2800
21b	*i*-Bu	2-ClPh	1700
21c	*i*-Bu	2-HOPh	3300
21d	*i*-Bu	2-MeOPh	1400
21e	*i*-Bu	3-MePh	2100
21f	*i*-Bu	3-CF$_3$Ph	2200
21g	*i*-Bu	3-MeOPh	4000
21h	*i*-Bu	4-HOPh	>10,000
21i	*i*-Bu	4-MeOPh	350
21j	*i*-Bu	4-MeSO$_2$Ph	4900
21k	*i*-Bu	2,6-ClPh	2600
21l	*i*-Bu	3,4-OCH$_2$OPh	1500
21m	*i*-Bu	3,4-MeOPh	4300
21n	*i*-Bu	2,5-MeOPh	2500
21o	*i*-Bu	2,4,5-MeOPh	6100
21o	*i*-Bu	2-Thienyl	4600
21q	*i*-Bu	3-Benzothienyl	2300
21r	*i*-Bu	3-Benzoxazolyl	4400
21s	*i*-Bu	3-Indolyl	640
21t	*i*-Bu	1-Me-3-indolyl	3000
21u	*i*-Bu	2-Me-3-indolyl	1600

While the sulfonamide **22l** and carbamate **22m** were only weakly active, the acetyl analog **22n** had a K_i value of 360 nM. By incorporating an amine into **22n**, the resulting compounds (**22o** and **22p**) displayed more than 4-fold increase in potency. These results were confirmed on the isobutyl analogs **23a**–**e**. Thus, the glycine derivative **23d** had a K_i of 19 nM.

Selected compounds were tested for their selectivity over the other melanocortin receptor subtypes. For example, *S*-**16** showed only 10-fold selectivity due to its low affinity at the MC4R. **22p** had good selectivity over the MC1 and MC3 receptors, while it still had good binding affinity at the MC5 receptor (K_i = 170 nM, Table 4). In contrast, **23d** also displayed over 25-fold selectivity at the MC5R. None of the compounds exhibited significant stimulation of cAMP release in cells expressing the MC4 receptor at a 10 μM concentration, demonstrating that they were not functional agonists. Instead, **22p** and **23d** showed dose-dependent inhibition of α-MSH-stimulated cAMP production with IC_{50} values of 1.1 and 1.3 μM, respectively (Table 5).

Due to their desirable in vitro properties, **22p** and **23d** were profiled for their pharmacokinetic properties in

Table 3. SAR of the *N*-alkyl or acyl group at the human MC4R



22-25

Compound	X	R^1	R^2	K_i (nM)
22a	CF_3	*s*-Bu	Et	92
22b	CF_3	*s*-Bu	$CH_2CH(Me)CH_2CH_3$	2800
22c	CF_3	*s*-Bu	CH_2-2-tetrafuranyl	780
22d	CF_3	*s*-Bu	$CH_2C_6H_5$	5900
22e	CF_3	*s*-Bu	$CH_2C_6H_4F$-2	>10,000
22f	CF_3	*s*-Bu	CH_2CH_2Ph	2600
22g	CF_3	*s*-Bu	$CH_2CH_2NH_2$	56
22h	CF_3	*s*-Bu	*S*-$CH_2CH(NH_2)CH_3$	89
22i	CF_3	*s*-Bu	*R*-CH_2-2-pyrrolidyl	110
22j	CF_3	*s*-Bu	*S*-CH_2-2-pyrrolidyl	64
22k	CF_3	*s*-Bu	$CH_2CH_2CH_2NH_2$	30
22l	CF_3	*s*-Bu	SO_2Me	1800
22m	CF_3	*s*-Bu	COOMe	2400
22n	CF_3	*s*-Bu	COMe	360
22o	CF_3	*s*-Bu	$COCH_2NH_2$	18
22p	CF_3	*s*-Bu	$COCH_2CH_2NH_2$	13
23a	CF_3	*i*-Bu	Me	1900
23b	CF_3	*i*-Bu	Et	170
23c	CF_3	*i*-Bu	CH_2CH_2Ome	620
23d	CF_3	*i*-Bu	$COCH_2NH_2$	19
23e	CF_3	*i*-Bu	$COCH_2CH_2NH_2$	25
24a	F	$EtOCH_2$[a]	$COCH_2CH_2NH_2$	300
25a	F	$BnOCH_2$[a]	$CH_2CH_2NH_2$	400
25b	F	$BnOCH_2$[a]	$COCH_2CH_2NH_2$	210

[a] Racemic mixture.

Table 4. Selectivity profiles of *S*-**16**, **22p**, and **23d**[a]

Compound	K_i (nM)			
	MC1R	MC3R	MC4R	MC5R
S-**16**	(21%)	1400	110	1200
22p	(17%)	800	13	170
23d	(18%)	1200	19	500

[a] Binding affinity at the human melanocortin receptors expressed in HEK293 cells with [^{125}I]NDP-MSH as radio-labeled ligand.

Table 5. Pharmacokinetic parameters of compounds **22p** and **23d** in mice[a]

Compound	**22p**	**23d**
iv dose (mg/kg)	5	5
CL (mL/min kg)	3.5	26.9
V_d (L/kg)	1.6	8.8
$t_{1/2}$ (h)	5.2	3.8
AUC (ng/mL h)	23,132	3,071
C_{brain} (ng/g) at 1, 4 h	62, 70	43, 33
C_{brain}/C_{plasma}	0.02, 0.03	0.08, 0.17
po dose (mg/kg)	10	10
C_{max} (ng/mL)	1,166	115
T_{max} (h)	6	2
AUC (ng/mL h)	12,067	687
F (%)	26.1	11.2

[a] Average of three animals.

mice. After an intravenous injection at 5 mg/kg, **22p** exhibited a very low clearance (CL = 3.5 mL/min kg) and low volume of distribution (V_d = 1.6 L/kg), resulting in a long half-life ($t_{1/2}$ = 5.2 h) in this species. At 1- and 4-h postdosing, the whole brain concentrations were 62 and 70 ng/g, which exhibited low brain/plasma ratio of 0.02 and 0.03, respectively. After an oral dose of 10 mg/kg, **22p** reached a maximal concentration of 1166 ng/mL at 6 h, and an area under the curve (AUC) of 12,067 ng/mL h, which gave an absolute bioavailability of 26.1%. The high plasma exposure might reflect high plasma protein binding of this compound, indicated by its low V_d value for a highly lipophilic molecule (measured log*D* was 4.0).[14] In comparison, the more lipophilic **23d** (measured log*D* of 4.5) had a moderate CL of 26.9 mL/min kg, a high V_d of 8.8 L/kg, and a moderate $t_{1/2}$ of 3.8 h. However, despite its high volume of distribution, the brain penetration of **23d** was still low, presumably caused by efflux mechanism at the blood–brain barrier.[15] **23d** had a moderate oral bioavailability of 11.2%.

In conclusion, a series of 3-arylpropionylpiperazines were synthesized and studied as antagonists of the melanocortin-4 receptor. The potency was increased when the α-methyl of **11** was replaced by a larger *s*-butyl or *iso*-butyl group. Further enhancements were observed when either a glycine or β-alanine was incorporated onto the benzylamine. Some compounds demonstrated good potency, moderate selectivity, and reasonable oral bioavailability.

References and notes

1. Cone, R. D. *Nat. Neurosci.* **2005**, *8*, 571.
2. Scarlett, J. M.; Marks, D. L. *Expert Opin. Invest. Drugs* **2005**, *14*, 1233.
3. Foster, A. C.; Chen, C.; Markison, S.; Marks, D. L. *IDrugs* **2005**, *8*, 314.
4. Chaki, S.; Okuyama, S. *Peptides* **2005**, *26*, 1952.
5. (a) Vos, T. J.; Caracoti, A.; Che, J. L.; Dai, M.; Farrer, C. A.; Forsyth, N. E.; Drabic, S. V.; Horlick, R. A.; Lamppu, D.; Yowe, D. L.; Balani, S.; Li, P.; Zeng, H.; Joseph, I. B.; Rodriguez, L. E.; Maguire, M. P.; Patane, M. A.; Claiborne, C. F. *J. Med. Chem.* **2004**, *47*, 1602; (b) Nicholson, J. R.; Kohler, G.; Schaerer, F.; Senn, C.; Weyermann, P.; Hofbauer, K. G. *J. Pharmacol. Exp. Ther.* **2006**, online ahead of print.
6. (a) Chaki, S.; Hirota, S.; Funakoshi, T.; Suzuki, Y.; Suetake, S.; Okubo, T.; Ishii, T.; Nakazato, A.; Okuyama, S. *J. Pharmacol. Exp. Ther.* **2003**, *304*, 818; (b) Chaki, S.; Oshida, Y.; Ogawa, S. I.; Funakoshi, T.; Shimazaki, T.; Okubo, T.; Nakazato, A.; Okuyama, S. *Pharmacol. Biochem. Behav.* **2005**, *82*, 621.
7. Xi, N.; Hale, C.; Kelly, M. G.; Norman, M. H.; Stec, M.; Xu, S.; Baumgartner, J. W.; Fotsch, C. *Bioorg. Med. Chem. Lett.* **2004**, *14*, 377.
8. Vos, T. J.; Balani, S.; Blackburn, C.; Chau, R. W.; Danca, M. D.; Drabic, S. V.; Farrer, C. A.; Patane, M. A.; Stroud, S. G.; Yowe, D. L.; Claiborne, C. F. *Bioorg. Med. Chem. Lett.* **2006**, *16*, 2302.
9. (a) Tucci, F. C.; White, N. S.; Markison, S.; Joppa, M.; Tran, J. A.; Fleck, B. A.; Madan, A.; Dyck, B. P.; Parker, J.; Pontillo, J.; Arellano, L. M.; Marinkovic, D.; Jiang, W.; Chen, C. W.; Gogas, K. R.; Goodfellow, V. S.; Saunders, J.;

Foster, A. C.; Chen, C. *Bioorg. Med. Chem. Lett.* **2005**, *15*, 4389; (b) Markison, S.; Foster, A. C.; Chen, C.; Brookhart, G. B.; Hesse, A.; Hoare, S. R.; Fleck, B. A.; Brown, B. T.; Marks, D. L. *Endocrinology* **2005**, *46*, 2766.
10. For a reference, please see: Lavrador, K.; Murphy, B.; Saunders, J.; Struthers, S.; Wang, X.; Williams, J. *J. Med. Chem.* **2004**, *47*, 6864.
11. Jiang, W.; Chen, C.; Marinkovic, D.; Tran, J. A.; Chen, C. W.; Arellano, L. M.; White, N. S.; Tucci, F. C. *J. Org. Chem.* **2005**, *70*, 8924.
12. Chen, C.; Pontillo, J.; Fleck, B. A.; Gao, Y.; Wen, J.; Tran, J. A.; Tucci, F. C.; Marinkovic, D.; Foster, A. C.; Saunders, J. *J. Med. Chem.* **2004**, *47*, 6821.
13. Nickolls, S. A.; Cismowski, M. I.; Wang, X.; Wolff, M.; Conlon, P. J.; Maki, R. A. *J. Pharmacol. Exp. Ther.* **2003**, *304*, 1217.
14. Pajouhesh, H.; Lenz, G. R. *NeuroRx* **2005**, *2*, 541.
15. Lin, J. H.; Yamazaki, M. *Drug Metab. Rev.* **2003**, *35*, 417.