Bioorganic & Medicinal Chemistry Letters 19 (2009) 3199–3203

Contents lists available at ScienceDirect

Bioorganic & Medicinal Chemistry Letters

journal homepage: www.elsevier.com/locate/bmcl

Tricyclic thienopyridine–pyrimidones/thienopyrimidine–pyrimidones as orally efficacious mGluR1 antagonists for neuropathic pain

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ARTICLE INFO

Article history:
Received 12 March 2009
Revised 21 April 2009
Accepted 23 April 2009
Available online 3 May 2009

Keywords:
Tricyclic thienopyridine–pyrimidone
Thienopyrimidine–pyrimidone
Triazafluorenone
mGluR1 antagonist
Neuropathic pain

ABSTRACT

Introduction of small unsaturated alkylamino groups at the 4-position of the A-ring of the tricyclic framework (triazafluorenone) afforded extremely potent and selective mGluR1 antagonists with desirable properties. Compounds **11q** and **11s** are active in the SNL pain model with ED_{50}s 3.3 and 6.4 mg/kg respectively. Metabolic outcome of propargyl amino moiety was studied.

Glutamate is the major excitatory neurotransmitter in the central nervous system and mediates its actions via activation of both ionotropic and metabotropic receptor families. The metabotropic glutamate receptors (mGluRs) form a family of eight subtypes (mGlu1 to mGlu8) and are assigned to three groups based on their structure, coupling to effector mechanisms and pharmacology. Group I mGluRs (mGluR1 and mGluR5) are post synaptic receptors while Group II (mGluR2 and mGluR3) and Group III (mGluR6, mGluR7 and mGluR8) are located presynaptically. It has been found that the Group I mGluRs play key roles in the central sensitization of pain and other neurologic disorders.[1–6] Over the last decade, there have been tremendous advances in the development of small molecules that selectively activate or inhibit specific mGlu receptor subtypes. Given below are some of the recent examples from literature (Fig. 1).[7–11]

Compound **1** was identified from our high throughput screening as a potent mGluR1 antagonist that was active in an in vivo model for pain (rat spinal nerve ligation, SNL).[12] A recent paper describing the SAR around the tricyclic frame work was published.[13] They have not disclosed any unsaturated alkyl substitutions on the A-ring of the tricyclic structure. Our attempt to replace the dimethyl amino moiety was exceptionally fruitful.[14] We found that small unsaturated alkyls gave extremely potent mGluR1 antagonists. SAR studies involving A-ring and *N*-aryl modifications and some in vivo results are discussed in this Letter (Fig. 2).

Compounds of this type were synthesized according to Scheme 1. Commercially available ethylcyano malonate was condensed with dimethylformamide–dimethylacetal (DMF–DMA) followed by acid catalyzed cyclization gave pyridine derivative **4** in good yields. Upon reaction with $POCl_3$ on **4** gave **5** which was cyclized to a common intermediate **6**. Heating a solution of **6** in DMF with

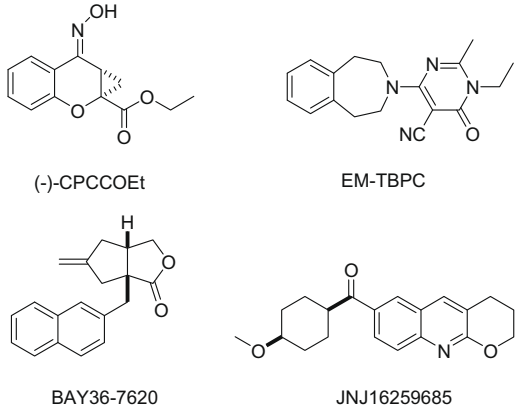

Figure 1. Noncompetitive antagonists of mGluR1.

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0960-894X/$ - see front matter © 2009 Published by Elsevier Ltd.
doi:10.1016/j.bmcl.2009.04.104

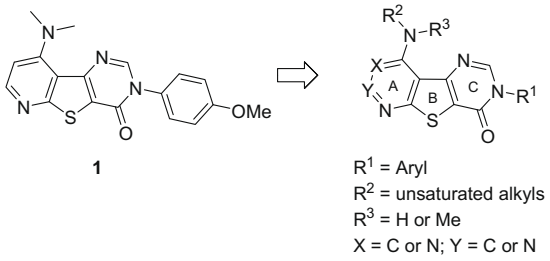


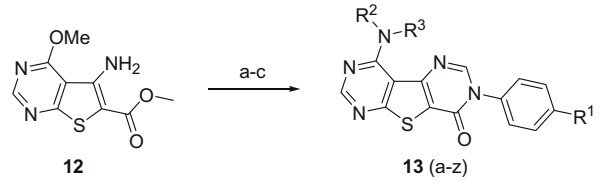
Figure 2. General SAR plans.

DMF–DMA afforded compound **7** which was cyclized to the tricyclic compound **8** in good yields. During this course of our studies, we found that heating a solution of **6** with desired amine in triethylorthoformate in the presence of acetic acid gave the cyclized compound in very high yields.[15] Demethylation of **8** followed by triflation of the phenol afforded **10** in good yields. Intermediate **10** was reacted with unsaturated alkylamines to afford the final compounds in excellent yields. We used allyl, methallyl and propargyl amines in this current SAR study. Similar chemistry was employed in the construction of thienopyrimidine–pyrimidone analogs as shown in Scheme 2. The known amino ester **12**[16] was converted to the final targets in three steps with good overall yields. The final step is the displacement of methoxy group in the pyrimidine nucleus with appropriate amine in polar solvents such as DMSO to get compounds **13a–z**.

The mGluR1 inhibitory potencies of the newly synthesized tricyclic compounds are summarized in Table 1. Initially we turned our attention to the allyl substitution on the A-ring of the pyridine nucleus. Human mGluR1 IC_{50} and rat K_i are shown throughout in this manuscript. Allylamino compounds **11a–i** are generally well tolerated as shown in Table 1. Compound **11a** showed human IC_{50} value of 2 nM with a rat K_i of 1.1 nM. Other substitution on the aromatic ring such as 4-Me, 4-Br, and 4-F are very well tolerated. However the 4-chloro substitution (compound **11c**) gave only moderate activity on the human receptor but showed excellent potency in the rat assay. Similar results were obtained with the 4-Br derivative (**11i**). Disubstitution on the aromatic ring is well tolerated in this scaffold. N-Methylated compounds such as **11g** and **11h** are also tolerated. It has been found that all these tricyclic derivatives are inactive in the mGluR5 assay.

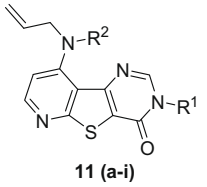
Next, we studied the effect of substitution on the allyl group. Methallyl derivatives generally afforded active analogs with 2–3-fold less mGluR1 affinity as shown in Table 2. Disubstitution on the aromatic ring (compounds **11n**, **11o** and **11p**) generally decreases the human inhibitory potency. Compounds **11k** and **11l** exhibited subnanomolar activity in rat mGluR1 assay. Methallyl substitution was also very effective in the pyrimidine series (Table 3). Simple phenyl substitution (compound **13f**)[17] provided good human affinity but a moderate rat K_i value. It was interesting to note that the 4-pyridyl analog displayed poor activity at both the human and rat receptors. Unlike in the case of pyridine analogs, disubstitution in the pyrimidine series is very well tolerated. For example, compound **13l**, showed human IC_{50} of 8.9 nM and rat K_i of 2.3 nM.

Replacement of the dimethylamino group of our lead compound with propargylamine afforded an extremely potent compound **11q** with human IC_{50} 0f 0.9 nM and a rat K_i of 0.6 nM as



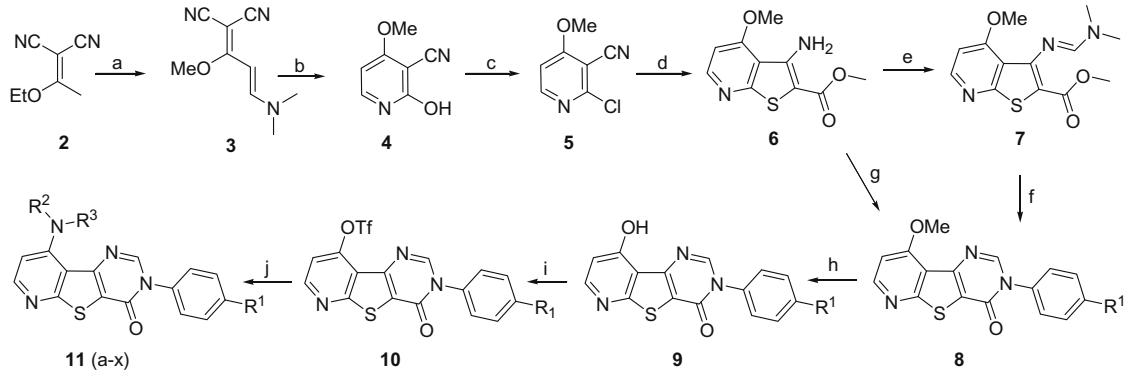
Scheme 2. Reagents and conditions: (a) DMF–DMA, DMF, 100 °C; (b) 4-Cl-aniline (R^1 = Cl), HOAc, toluene, 110 °C; (c) R^2R^3NH, DMSO.

Table 1
mGluR1 receptor binding for compounds **1**, **11a–i**



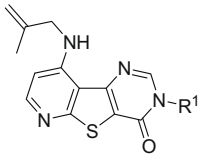
Compd	R^1	R^2	h-mGluR1 IC_{50}[a] (nM)	r-mGluR1 K_i[a] (nM)
1			9.5	7.9
11a	4-MeO-Ph	H	2	1.1
11b	4-Me-Ph	H	11	2.7
11c	4-Cl-Ph	H	60	1.1
11d	4-Br-Ph	H	12.6	0.7
11e	4-F-3-MeO-Ph	H	6.8	14
11f	4-F-Ph	H	4.4	7.7
11g	4-Cl-Ph	Me	1.1	5.4
11h	4-MeO-Ph	Me	6.9	4.1
11i	4-Br-Ph	Me	66	3.2

[a] The IC_{50} and K_i data are an average of at least three measurements, performed on human mGlu1/5 and rat mGlu1 receptors, respectively. The standard error was 10%, and variability was less than twofold from assay to assay. h-mGluR5 IC_{50} > 3 μM.



Scheme 1. Reagents and conditions: (a) DMF–DMA, MeOH, 80 °C; (b) HOAc (80%), 130 °C; (c) $POCl_3$, Et_3N, 100 °C; (d) $HSCH_2CO_2Me$, NaOMe, DMF, 80 °C; (e) DMF–DMA, DMF, 100 °C; (f) 4-Cl-aniline (R^1 = Cl), HOAc, toluene, 110 °C; (g) 4-Cl-aniline (R^1 = Cl), HOAc, $CH(OEt)_3$, 160 °C; (h) BBr_3, CH_2Cl_2; (i) $NPhTf_2$, CH_2Cl_2; (j) R^2R^3NH, DMSO.

Table 2
mGluR1 receptor binding for compounds **11j–p**

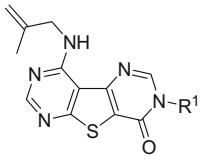


11 j-p

Compd	R^1	h-mGluR1 IC_{50}[a] (nM)	r-mGluR1 K_i[a] (nM)
11j	4-Cl-Ph	10.6	1.2
11k	4-Me-Ph	16.4	0.2
11l	4-MeO-Ph	10.4	0.4
11m	4-Br-Ph	43	1.5
11n	3-F-4-MeO-Ph	103	13
11o	3-(2,3-Dihydrobenzo[*b*] [1,4]Dioxin-6-yl)	196	13
11p	3-(Benzo[*d*][1,3]dioxol-5-yl)	241	14

[a] The IC_{50} and K_i data are an average of at least three measurements, performed on human mGlu1/5 and rat mGlu1 receptors, respectively. The standard error was 10%, and variability was less than twofold from assay to assay. h-mGluR5 IC_{50} > 3 μM.

Table 3
mGluR1 receptor binding for compounds **13a–m**

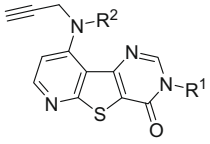


13 (a-m)

Compd	R^1	h-mGluR1 IC_{50}[a] (nM)	r-mGluR1 K_i[a] (nM)
13a	4-Cl-Ph	7.7	2.2
13b	4-Me-Ph	12.4	2.9
13c	4-MeO-Ph	9.6	7.5
13d	4-Br-Ph	14.5	3.8
13e	3-F-4-MeO-Ph	72	68
13f	Ph	4.6	25
13g	4-Py	955	1000
13h	4-F-Ph	10.7	38
13i	3-Cl-Ph	53	62
13j	3-(Benzo[*d*]thiazol-5-yl)	16	43
13k	3-(Benzo[*d*]thiazol-6-yl)	17	49
13l	3-(Benzo[*b*]thiophen-5-yl)	8.9	2.3
13m	3-(2,3-Dihydrobenzo[*b*] [1,4]dioxin-6-yl)	33	64

[a] The IC_{50} and K_i data are an average of at least three measurements, performed on human mGlu1/5 and rat mGlu1 receptors, respectively. The standard error was 10%, and variability was less than twofold from assay to assay. h-mGluR5 IC_{50} > 3 μM.

Table 4
mGluR1 receptor binding for compounds **11q–x**

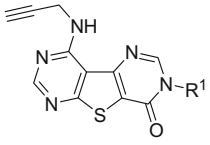


11 (q-x)

Compd	R^1	R^2	h-mGluR1 IC_{50}[a] (nM)	r-mGluR1 K_i[a] (nM)
11q	4-Cl-Ph	H	0.9	0.6
11r	4-Me-Ph	H	1.5	1.6
11s	4-MeO-Ph	H	2.1	3.5
11t	4-Br-Ph	H	1.8	0.7
11u	3-F-4-MeO-Ph	H	12.5	28
11v	4-Cl-Ph	Me	3.8	18
11w	4-MeO-Ph	Me	10.8	27
11x	4-Br-Ph	Me	1.9	8

[a] The IC_{50} and K_i data are an average of at least three measurements, performed on human mGlu1/5 and rat mGlu1 receptors, respectively. The standard error was 10%, and variability was less than twofold from assay to assay. h-mGluR5 IC_{50} > 3 μM.

Table 5
mGluR1 receptor binding for compounds **13n–z**



13 (n-z)

Compd	R^1	h-mGluR1 IC_{50}[a] (nM)	r-mGluR1 K_i[a] (nM)
13n	4-Cl-Ph	3.0	4.4
13o	4-Me-Ph	3.9	8.3
13p	4-MeO-Ph	13	25
13q	4-Br-Ph	6.8	29
13r	3-F-4-MeO-Ph	212	77
13s	4-F-Ph	8.2	39
13t	2-F-4-MeO-Ph	85	32
13u	3-(2,3-Dihydrobenzo[*b*] [1,4]dioxin-6-yl)	106	160
13v	3-(Benzo[d][1,3]dioxol-5-yl)	137	997
13w	3-(Benzo[*d*]thiazol-5-yl)	45	65
13x	3-(Benzo[*d*]thiazol-6-yl)	209	107
13y	3-(Benzofuran-5-yl)	21	63
13z	3-(Benzo[*b*]thiophen-5-yl)	219	69

[a] The IC_{50} and K_i data are an average of at least three measurements, performed on human mGlu1/5 and rat mGlu1 receptors, respectively. The standard error was 10%, and variability was less than 2-fold from assay to assay. h-mGluR5 IC_{50} > 3 μM.

shown in Table 4. 4-Me, 4-MeO, and 4-Br substitution on the *N*-aryl group afforded similarly highly potent compounds. N-Methylation lowered affinity 4–5-fold in all cases except 4-BrPh derivative (**11x**).

Similar results were obtained in the A-ring pyrimidine series as evidenced from the Table 5. Generally, mono substitution at the *para* substitution of the *N*-aryl ring is generally well tolerated. Compounds **13n**, **13o**, **13q** and **13s** attained single digit nanomolar at the human mGluR1 assay. However disubstituted compounds such as **13x** and **13z** exhibited a large decrease in potency as evidenced from Table 5.

Having achieved excellent potency against mGluR1 receptors, we shifted our attention toward measuring the pharmacokinetics of these types of molecules. Data for representative compounds **11q** and **11s** are shown in Table 6.

Compounds **11q** and **11s** showed moderate rat AUC with high brain to plasma ratio as demonstrated in Table 6. These compounds were inactive in the hERG assay. It has been found that compounds **11q** and **11s** were active in the in vivo pain model (rat spinal nerve ligation, SNL)[12] with ED_{50} values of 3.3 and 6.4 mg/kg, respectively, when dosed orally. The metabolic pathway in rat for high clearance compound **11q** was further investigated using microsome incubation. The metabolic details are shown in Figure 3. The metabolism of acetylenic compounds commonly used in the formulation of pharmaceuticals have been investigated previously using ^{13}C NMR and mass spectrometry.[19,20] Compound **11q**

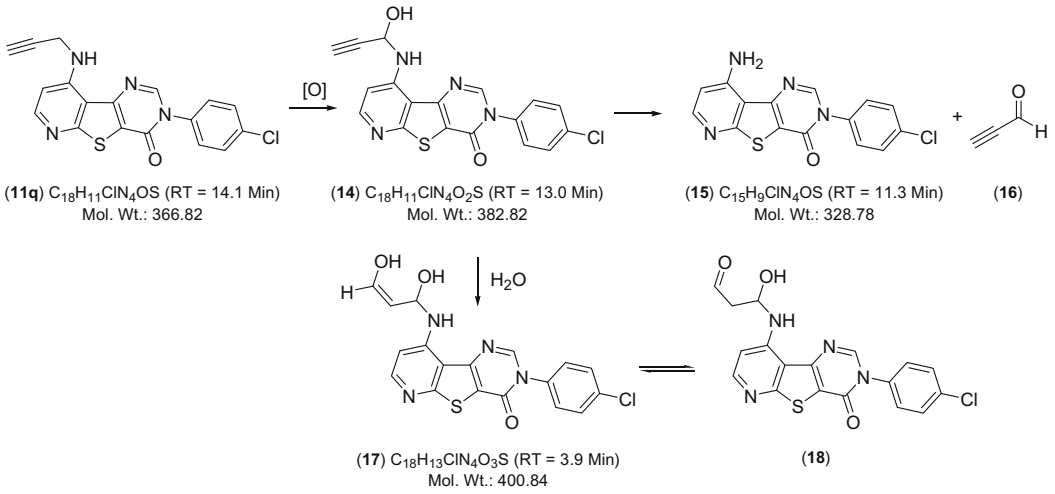
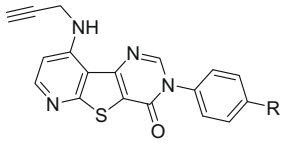


Figure 3. Metabolism of propargyl group (Mass analysis of rat bile samples, 0–24 h).

Table 6
PK Profile and in vivo activity of selected compounds

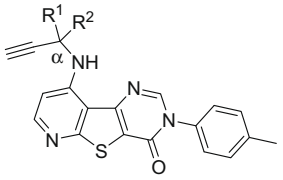


Parameters	R = Cl (**11q**)	R = OMe (**11s**)
Human mGluR1 IC_{50} (nM)	0.9	2.1
Rat mGluR1 K_i (nM)	0.6	3.5
Human mGluR5 IC_{50} (nM)	>3000	>3000
Rat SNL ED_{50}[12]	3.3 mg/kg po	6.4 mg/kg po
Caco-2 permeability	430 nm/s	690 nm/s
Efflux substrate	No	No
Rat PK, (10 mg/kg), AUC (ng.h/mL)[18]	427	682
Brain conc. @ 6 h (ng/g)	67	257
Brain/Plasma	1.8	2.5
Clearance (mL/min/kg)	30	22
Bioavailability (%)	16	34

was incubated with rat liver microsome for 0–24 h and the metabolites were identified by LC–MS methodology. The oxidation of the carbon adjacent to the amine lead to the intermediate **14** followed by degradation to the final molecules **15** and **16**. LC–MS peak at 13 min with an observed mass of 383 is attributed to the intermediate **14**. LC–MS analysis showed a peak at 3.9 min. with a mass of 401 could be water addition product (**17**) to intermediate **14** as shown in Figure 3. The parent amino compound **15** displayed in LC–MS at 11.3 min. This retention time of the amine **15** was independently confirmed via analysis of pure synthetic sample. Compound **15** was found to be the major metabolite in the rat bile. The side product of this whole sequence, compound **16**, is a known glutathione scavenger described in the literature.[19]

In order to avoid the metabolic issues, we introduced methyl groups adjacent to the amino group. Unfortunately monomethylation and dimethylation of our lead compounds afforded 200–300-fold less active compounds (**19**–**21**) as shown in Figure 4. Identical results were obtained for compounds with different substitution on the right hand side aromatic ring.

In summary, we have achieved a large number of single digit nanomolar mGluR1 antagonists in the tricyclic series. This excellent potency sometimes extrapolates to good in vivo efficacy as seen in compounds **11q** and **11s**. These types of compounds show moderate PK and high brain plasma ratio. It has also been found that these compounds are inactive in the hERG assay. Further data will be published elsewhere.



19; R^1 = Me, R^2 = H; h-IC_{50} = 314 nM
20; R^1 = Me, R^2 = Me; h-IC_{50} = 171 nM
21; R^1 = Et, R^2 = Et; h-IC_{50} = 358 nM

Figure 4. Effect of α-alkylation on human mGluR1 potency.

Acknowledgments

We thank Drs. Deen Tulshian, Julius Matasi and Peter Korakas for helpful discussions. We also thank Lisa Broske's group for animal dosing and Sam Wainhaus' group for PK sample analysis.

References and notes

1. Higgins, G. A.; Miczek, K. A. *Psychopharmacology* **2005**, *179*, 1.
2. Kew, J. N. C.; Kemp, J. A. *Psychopharmacology* **2005**, *179*, 4.
3. Steckler, T.; Lavreysen, H.; Oliveira, A. M.; Aerts, N.; Van Craenendonck, H.; Prickaerts, J.; Megens, A.; Lesage, A. S. J. *Psychopharmacology* **2005**, *179*, 198.
4. Niswender, C. M.; Jones, C. K.; Conn, P. J. *Curr. Top. Med. Chem.* **2005**, *5*, 847.
5. Kunishima, N.; Shimada, Y.; Tsuji, Y.; Sato, T.; Yamamoto, M.; Kumasaka, T.; Nakanishi, S.; Jingami, H.; Morikawa, K. *Nature* **2000**, *407*, 971.
6. Constantino, G.; Macchiarulo, A.; Pellicciari, R. *Bioorg. Med. Chem* **2001**, *9*, 847.
7. Layton, M. E. *Curr. Top. Med. Chem* **2005**, *5*, 859.
8. Ott, D.; Floersheim, P.; Inderbitzin, W.; Stoehr, N.; Francotte, E.; Lecis, G.; Richert, P.; Rihs, G.; Flor, P. J.; Kuhn, R.; Gasparini, F. *J. Med. Chem.* **2000**, *43*, 4428.
9. Malherbe, P.; Kratochwil, N.; Knoflach, F.; Zenner, M.-T.; Kew, J. N. C.; Kratzeisen, C.; Maerki, H. P.; Adam, G.; Mutel, V. *J. Biol. Chem.* **2003**, *278*, 8340.
10. Carroll, F. Y.; Stolle, A.; Beart, P. M.; Voerste, A.; Brabet, I.; Mauler, F.; Joly, C.; Antonicek, H.; Bockaert, J.; Müller, T.; Pin, J. P.; Prézeau, L. *Mol. Pharmacol.* **2001**, *59*, 965.
11. Lavreysen, H.; Wouters, R.; Bischoff, F.; Pereira, S. N.; Langlois, X.; Blokland, S.; Somers, M.; Dillen, L.; Lesage, A. S. J. *Neuropharmacology* **2004**, *47*, 961.
12. Varty, G. B.; Grilli, M.; Forlani, A.; Fredduzzi, S.; Grzelak, M. E.; Guthrie, D. H.; Hodgson, R. A.; Sherry, X. L.; Nicolussi, E.; Pond, A. J.; Parker, E. M.; Hunter, J. C.; Higgins, G. A.; Reggiani, A.; Bertorelli, R. *Psychopharmacology* **2005**, *179*, 207.

13. Recently, scientists from Abbott Laboratories disclosed similar mGluR1 antagonists based on the tricyclic triazafluorenone core structure. See: Zheng, G. Z.; Bhatia, P.; Daanen, J.; Kolasa, T.; Patel, M.; Latshaw, S.; Kouhen, O. F. E.; Chang, R.; Uchic, M. E.; Miller, L.; Nakane, M.; Lehto, S. G.; Honore, M. P.; Moreland, R. B.; Brioni, J. D.; Stewart, A. O. *J. Med. Chem.* **2005**, *48*, 7374.
14. Our efforts in the modification of tricyclic structure is being published in the following patents, WO **2006**/002051 and US **2006**/167029. Also see: Wu, W-L.; Burnett, D. A.; Domalski, D.; Greenlee, W. J.; Li, C.; Bertorelli, R.; Fredduzzi, S.; Lozza, G.; Veltri, A.; Reggiani, A. *J. Med. Chem* **2007**, *50*, 5550. Biology materials and methods were described in this Letter.
15. *Typical one step synthesis of tricyclic compound is described as follows*: Compound **6** (2 g, 0.0084 mol) was suspended in 15 mL triethyl orthoformate and treated with acetic acid (2 mL) and 4-chloroaniline (2 g, 0.0156 mol). The contents were heated in a sealed tube at 160 °C for 16 h. The reaction mixture was cooled to room temperature and the precipitated product was washed several times with ether. The product was dried in vacuo to get 2.5 g of compound **8** (R^1 = Cl) as white solid. ^{1}H NMR ($CDCl_3$): δ 8.65 (d, 1H), 8.29 (s, 1H), 7.56 (d, 2H), 7.40 (d, 2H), 6.93 (d, 1H), 4.17 (s, 3H). Mass Spectrum (M^{+1}): *m/z* calcd for $C_{16}H_{11}ClN_3O_2S^+$ = 344.03, found *m/z* = 344.2.
16. Clark, J.; Shahhet, M. S. *J. Heterocycl. Chem.* **1993**, *30*, 1065.
17. *Spectral data for compound* **13f**: ^{1}H NMR ($CDCl_3$): δ 8.62 (s, 1H), 8.27 (s, 1H), 7.91 (s, 1H), 7.58 (m, 3H), 7.45 (m, 2H), 4.99 (s, 1H), 4.93 (s, 1H), 4.32 (d, 2H), 1.86 (s, 3H). Mass Spectrum (M^{+1}): *m/z* calcd for $C_{18}H_{16}N_5OS^+$ = 350.11, found *m/z* = 350.2.
18. Korfmacher, W. A.; Cox, K. A.; Ng, K. J.; Veals, J.; Hsien, Y.; Wainhaus, S.; Broske, L.; Prelusky, D.; Nomeir, A.; White, R. E. *Rapid Commun. Mass. Spectrom.* **2001**, *15*, 335.
19. Banijamali, A. R.; Xu, Y.; DeMatteo, V.; Strunk, R. J.; Sumner, S. J. *J. Agric. Food Chem.* **2000**, *48*, 4693.
20. DeMaster, E. G.; Sumner, H. W.; Kaplan, E.; Shirota, F. N.; Nagasawa, H. T. *Toxicol. Pharmacol.* **1982**, *65*, 390.