Bioorganic & Medicinal Chemistry Letters 23 (2013) 785–790

Contents lists available at SciVerse ScienceDirect

Bioorganic & Medicinal Chemistry Letters

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

Discovery of a new series of [1,2,4]triazolo[4,3-*a*]quinoxalines as dual phosphodiesterase 2/phosphodiesterase 10 (PDE2/PDE10) inhibitors

José-Ignacio Andrés [a,*], Peter Buijnsters [b], Meri De Angelis [a], Xavier Langlois [c], Frederik Rombouts [b], Andrés A. Trabanco [a], Greet Vanhoof [d]

[a] Janssen Research & Development, Medicinal Chemistry, Janssen-Cilag S.A., C/Jarama 75, 45007 Toledo, Spain
[b] Janssen Research & Development, Neuroscience-Medicinal Chemistry, Janssen Pharmaceutica N.V., Turnhoutseweg 30, B-2340 Beerse, Belgium
[c] Janssen Research & Development, Neuroscience-Biology, Janssen Pharmaceutica N.V., Turnhoutseweg 30, B-2340 Beerse, Belgium
[d] Janssen Research & Development, CREATe, Translational Sciences, Janssen Pharmaceutica N.V., Turnhoutseweg 30, B-2340 Beerse, Belgium

ARTICLE INFO

Article history:
Received 1 October 2012
Revised 19 November 2012
Accepted 20 November 2012
Available online 1 December 2012

Keywords:
Neurological disorders
PDE2 inhibitors
PDE10 inhibitors
Triazoloquinoxalines

ABSTRACT

The synthesis, preliminary evaluation and structure–activity relationship (SAR) of a series of 1-aryl-4-methyl[1,2,4]triazolo[4,3-*a*]quinoxalines as dual phosphodiesterase 2/phosphodiesterase 10 (PDE2/PDE10) inhibitors are described. From this investigation compound **31** was identified, showing good combined potency, acceptable brain uptake and high selectivity for both PDE2 and PDE10 enzymes. Compound **31** was subjected to a microdosing experiment in rats, showing preferential distribution in brain areas where both PDE2 and PDE10 are highly expressed. These promising results may drive the further development of highly potent combined PDE2/PDE10 inhibitors, or even of selective inhibitors of PDE2 and/or PDE10.

Phosphodiesterases (PDEs) are a family of enzymes encoded by 21 genes and subdivided into eleven distinct families according to their structural and functional properties. These enzymes metabolically inactivate the ubiquitous secondary messengers, 3′,5′-cyclic adenosine monophosphate (cAMP) and 3′,5′-cyclic guanosine monophosphate (cGMP), hydrolyzing their phosphodiester bonds and converting them into the biologically inactive nucleotide 5′-monophosphates AMP and GMP, respectively.[1] These two messengers regulate a wide variety of biological processes, including pro-inflammatory mediator production and action, ion channel function, muscle contraction, learning, differentiation, apoptosis, lipogenesis, glycogenolysis, and gluconeogenesis. In neurons, they induce the activation of cAMP and cGMP-dependent kinases and subsequent phosphorylation of proteins involved in acute regulation of synaptic transmission as well as in neuronal differentiation and survival.[2] From the 11 identified mammalian PDE families (PDE1–PDE11) three of them selectively hydrolyze cAMP (PDE4, 7, and 8), three are selective for cGMP (PDE5, 6, and 9), and five families hydrolyze both cyclic nucleotides (PDE1, 2, 3, 10, and 11).[3]

Although the PDEs are in general expressed in various tissues and cells throughout the body, the PDE2 and PDE10 enzymes are preferentially expressed in the brain.[4,5] The expression of PDE2 in limbic system and basal ganglia suggests that PDE2 may modulate neuronal signaling involved in emotion, perception, concentration, learning and memory.[6] Additionally, PDE2 is expressed in the nucleus accumbens, the olfactory bulb, the olfactory tubercle and the amygdala, supporting the hypothesis that PDE2 may also be involved in anxiety and depression.[7] PDE10A is the single member identified from the PDE10 family and, in the brain, mRNA and protein are highly expressed in a majority of striatal medium spiny neurons (MSNs).[8] This unique distribution of PDE10A in the brain, together with its increased pharmacological characterization, indicates a potential use of PDE10 inhibitors for treating neurological and psychiatric disorders like schizophrenia.[9] Thus, PDE10 inhibitors may possess a pharmacological profile similar to that of the current antipsychotics which mainly treat positive symptoms of schizophrenia, but also having the potential to improve the negative and cognitive symptoms of the disease while lacking some of the D_2 related side effects, such as the extrapyramidal side effects (EPS) commonly observed with the currently available antipsychotics.[10] Since PDE10 inhibitors can be used to raise levels of cAMP and/or cGMP within cells that express the PDE10 enzyme, for example neurons that comprise the basal ganglia, PDE10 inhibitors may be also useful in treating Parkinson's disease, Huntington's disease, addiction and depression.[11]

Abbreviations: CNS, central nervous system; cAMP, 3′,5′-cyclic adenosine monophosphate; cGMP, 3′,5′-cyclic guanosine monophosphate; PDE2, phosphodiesterase 2; PDE10, phosphodiesterase 10; SAR, structure–activity relationship.

* Corresponding author. Tel.: +34 925 245 780; fax: +34 925 245 771.
E-mail address: iandres@its.jnj.com (J.-I. Andrés).

0960-894X/$ - see front matter © 2012 Elsevier Ltd. All rights reserved.
http://dx.doi.org/10.1016/j.bmcl.2012.11.077

The preferential distribution of PDE2 and PDE10 in the brain, together with the preclinical evidence described with the use of selective PDE2 and PDE10 inhibitors for the treatment of multiple CNS disorders, make this combination of potential interest for therapeutic use. It has recently been reported that blockade of PDE2, PDE4 and PDE10 with the atypical anxiolytic drug Tofisopam could improve the negative symptoms of psychosis.[12] Although a series of combined PDE2/PDE10 inhibitors have recently been described, their pharmacological characterization has been limited to in vitro experiments, without any reported pharmacokinetic or pharmacodynamic studies.[13]

We recently started a medicinal chemistry program towards the identification of selective PDE2 inhibitors and/or molecules with combined PDE2/PDE10 activity. A high throughput screen (HTS) of our compound collection identified a series of 1-aryl-4-methyl[1,2,4]triazolo[4,3-*a*]quinoxaline derivatives **A** of which compound **1a** (Fig. 1) showed interesting dual activity, with IC_{50} 30 nM for PDE2 and 325 nM for PDE10. The corresponding regioisomer **1b** presented also dual activity, however the potency was significantly lower. We considered this chemotype and particularly compound **1a** as an interesting starting point for further optimization. This Letter summarizes the preliminary in vitro structure–activity relationships (SAR) and brain penetration results obtained from our exploration focusing on positions C-1 and C-8 of the triazoloquinoxaline scaffold.[14]

The synthesis of the hit **1a** and the key intermediates **7a**–**m** is depicted in Scheme 1. Condensation of commercially available 1,2-diaminophenyl derivatives **2a** and **2b** with pyruvic acid **3a** or ethyl pyruvate **3b** afforded the two possible regioisomers in a 1:1 ratio, which could be separated by column chromatography yielding intermediates **4a**, **b**. In view of the difference in in vitro PDE2 potency between the initial hits **1a** and **1b** only compounds **4a**, **b** were used for the next reaction step. Intermediates **4a**, **b** were halogenated by means of phosphorous oxychloride affording derivatives **5a** and **5b** and cyclized using the appropriate hydrazido compounds **6** yielding **1a** and key intermediates **7a**–**m** in moderate to good yields (60–85%).

The target amides **8**–**26** were prepared by a HATU mediated reaction of the appropriate amines and the acids obtained following hydrolysis of esters **7a**–**l** in good overall yields (Scheme 2).

All the aminomethyl derivatives, **29**–**40**, depicted in Scheme 3 were synthesized starting from the 8-bromo-substituted intermediate **7m**. The key step of the synthesis was the preparation of aldehyde **28** that was synthesized in a two-step reaction procedure. Firstly, palladium-catalyzed Stille cross-coupling reaction of **7m** with tributyl vinyl tin led to the vinyl derivative **27**, which was subsequently oxidized by means of osmium tetroxide and sodium periodate to the target aldehyde intermediate **28**. The reductive amination of primary amines with **28** was performed at room temperature using sodium borohydride as reducing agent, whereas sodium triacetoxyborohydride and microwave irradiation conditions were needed for secondary amines. The target compounds

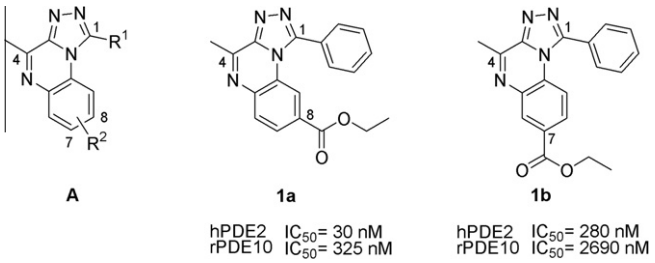


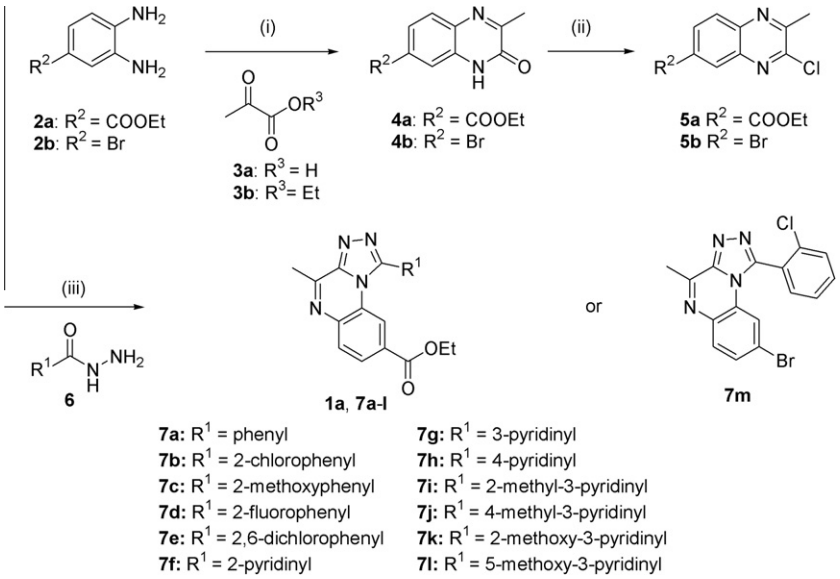
Figure 1. Triazoloquinoxaline hits **1a** and **1b**.

29–**40** were obtained in moderate yields (25–60%). The synthesis of compound **31** starting from the commercially available reagent **2b** is described as an example to illustrate the synthetic methodology.[15]

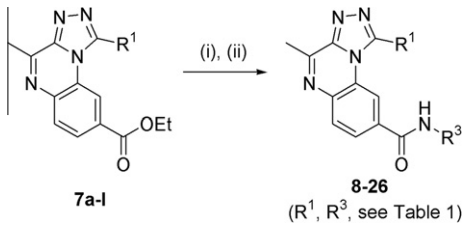
The in vitro inhibitory activity against hPDE2 and rPDE10 enzymes was evaluated for final compounds **1a**, **7b**, **8**–**26** and **29**–**40** (Table 1).[16] Our initial exploration focused on introducing different substituents in the phenyl ring at position C-1 of the triazoloquinoxaline scaffold, while keeping the ethoxycarbonyl moiety fixed at position C-8. From a previous exploration (data not shown) we found that the *o*-chloro-phenyl derivative **7b** seemed to be the most potent PDE2/PDE10 inhibitor, suggesting that distorting coplanarity between the aryl ring and the triazoloquinoxaline core enhanced the inhibitory activity for both enzymes. Replacement of the ethyl ester by the corresponding ethyl-amide analog led to a fivefold increase in potency for PDE2 and to a lesser extent for PDE10 (compare results of **8** with **1a**). Combination of *o*-chlorophenyl at C-1 and ethylamide at C-8 resulted in a 20-fold increase in PDE2 potency and a 12-fold increase in PDE10 potency, increasing its selectivity versus PDE10 to 72-fold (Table 1, compounds **8** and **9**). A small exploration on the C-1 aryl was done while keeping the ethyl-amide fixed at C-8. Replacing the chlorine atom in **9** by other *ortho*-substituents such as methoxy (**10**) or fluorine (**11**) was somewhat detrimental for activity, whereas the introduction of a second chlorine atom at position 6 of the C-1 phenyl ring (**12**) increased the PDE10A inhibition while maintaining the PDE2 activity. Introduction of pyridyl rings at C-1 (**13**–**15**) was found to be detrimental especially for PDE10 activity. However, the 3- and 4-pyridyl derivatives **14** and **15** showed acceptable PDE2 inhibition and a remarkable selectivity versus PDE10 (**14**, 35-fold and **15**, 63-fold selectivity). Introduction of substituents in the 3-pyridyl ring (**16**–**19**) resulted in compounds with comparable selectivity range. From these 3-pyridyl analogs the most interesting one proved to be the 2-methyl-3-pyridyl derivative **16**, with substantially better PDE2 and PDE10 activity when compared to **14**, although it was significantly less potent than the *o*-chlorophenyl compound **9** (20-fold decrease in PDE2 potency and eightfold decrease in PDE10 potency).

Having in mind these results, we investigated the effect on PDE2/PDE10 inhibition of different substituents at C-8, while keeping the *o*-chloro-phenyl group at C-1 fixed. Firstly, a diverse set of different amides, exemplified by compounds **20**–**26**, was synthesized. A wide variety of amide moieties was tolerated and proved to be very potent PDE2 inhibitors, with IC_{50} values in the single digit nM range, showing significant combined PDE10A inhibitory activity. Aromatic rings with shorter (**20**, **21**) or longer (**22**) spacers retained good combined activities. Noteworthy is compound **21**, having a tertiary amide at position C-8, which showed a marked decrease in activity for both PDE2 and PDE10A when compared to its direct analogue **20**, indicating that secondary amides are preferred for activity. Interestingly, amides bearing a basic center (**23**–**26**) were also potent inhibitors of both enzymes, with compound **26** showing the most balanced profile among this set in terms of potency (PDE2 IC_{50} 0.42 nM and PDE10 IC_{50} 8.9 nM). The most potent PDE2 inhibitors from this amide series were the ethyl-amides **9** and **12**, whereas the most selective PDE2 inhibitor proved to be the benzyl-amide derivative **20** that showed 32-fold selectivity PDE2 versus PDE10 (IC_{50} 0.38 and 13 nM, respectively).

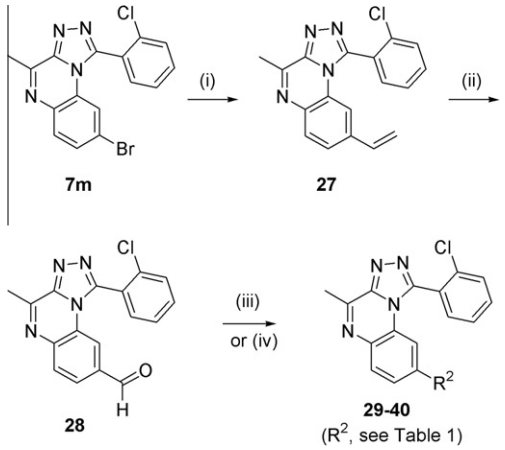
To expand the SAR knowledge, a series of C-8 aminomethyl derivatives containing secondary (**29**, **32**, **34** and **35**) or tertiary amines (**30**, **31**, **33** and **36**–**40**) were prepared. In general, the aminomethyl derivatives were weaker PDE2 inhibitors than the compounds bearing the amide substitution. Thus, compounds **29** and **32** showed 65- and 40-fold less PDE2 inhibition than their corresponding amide pairs **9** and **20**. The PDE10A activity was less



Scheme 1. Reagents and conditions: (i) for **3a** CH_3CO_2H/H_2O, rt, 7 h (42%), for **3b**, toluene, reflux, 3 h (40%); (ii) for **5a** $POCl_3$, reflux, 2 h (65–75%); for **5b**, $POCl_3$, 1,2-dichloroethane, reflux, 4 h (23%); (iii) EtOH, microwaves, 160 °C, 15–25 min (60–85%).



Scheme 2. Reagents and conditions: (i) LiOH, H_2O/THF, rt, 3 h, (quant. yield); (ii) R^3NH_2, Et_3N, HATU, DMF/CH_2Cl_2, rt, 3 h (60–85%).



Scheme 3. Reagents and conditions: (i) LiCl, tributylvinyl tin, $Pd(PPh_3)_4$, toluene, 120 °C, 1 h (93%); (ii) $NaIO_4$, OsO_4, 1,4-dioxane/H_2O, rt, 2 h (75%); (iii) for compounds **29**, **32**, **34** and **35**: primary amine, $NaBH_4$, MeOH, rt (25–45%); (iv) for compounds **30**, **31**, **33**, and **36**–**40**: secondary amine, $NaBH(OAc)_3$, 1,2-dichloroethane, microwaves, 80 °C, 10–20 min (35–60%).

affected between both series, and in this case **29** and **32** were approximately 8- and 4-fold weaker PDE10A inhibitors than **9** and **20**. It is remarkable that compounds showing the most balanced profile regarding dual activity reside within the aminomethyl subseries; the most potent aminomethyl PDE2 inhibitors were **31** and **36**, both compounds showing also good PDE10 inhibition (**31**, IC_{50} 2.8 nM vs 35 nM; **36**, IC_{50} 5.0 nM vs 19 nM). In both the amide and the amine series a similar trend was observed: when the potency for PDE2 increased or decreased, a similar increase or reduction was also observed in PDE10 inhibition.

Several compounds (**1a**, **9**, **17**, **18**, **22**, **31**, **33**–**35** and **37**) were tested for PDE10 inhibition using isolated recombinant expressed human enzyme (hPDE10A) and showed comparable PDE10 inhibition to the one found with the rat PDE10 enzyme (rPDE10A), proving that there are no large differences in inhibition between both species (Table 1).

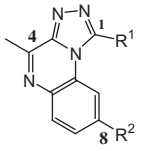
Representative examples from the amide (**9**, **17**, **19**, **24** and **25**) and aminomethyl (**29**, **31** and **40**) series were selected to evaluate their potential to cross the blood–brain barrier (BBB). The studies were conducted in rats to measure brain and plasma concentrations 1 h after subcutaneous (sc) or oral (p.o.) administration of a 10 mg/kg dose. The results obtained are shown in Table 2. Unfortunately, all the amidic derivatives tested had a very low brain uptake, with **9** and **24** being the only compounds that showed some significant brain penetration (brain/plasma (B/P) ratios of 0.11 and 0.14, respectively).

To our delight most compounds from the aminomethyl subseries showed acceptable distribution to the brain. Thus, compounds **29** and **31** showed B/P ratios higher than 0.5 (B/P 0.73 and 0.62, respectively) with acceptable absolute brain levels. On the contrary, compound **40**, having a terminal hydroxyl substituent, showed limited brain uptake (B/P 0.18). From these studies it could be concluded that aminomethyl derivatives, although less potent, had better brain uptake than the more potent amide derivatives.

We decided to further profile and test compound **31** in a microdosing experiment[17] to verify whether the distribution of **31** in the different brain regions would match the PDE2/PDE10 brain expression. The microdosing study was conducted in rats after a 0.03 mg/kg intravenous (iv) dose, in a 2 to 30 min timeframe. As can be observed in Figure 2, compound **31** reached the brain in good levels and showed the highest uptake in the striatum, which is the region where both PDE2 and PDE10 are mainly expressed. The compound also showed higher uptake in cortex and hippocampus (where only PDE2 is highly expressed) compared to cerebellum,[4,5] a region in which expression of both PDE2 and PDE10 is almost negligible.

In addition, compound **31** showed good in vitro selectivity over the other members of the PDE family. A full PDE profile for **31** is shown in Figure 3.

Table 1
In vitro hPDE2 and rPDE10 inhibitory activity of final compounds **1a**, **7b**, **8–26** and **29–40**



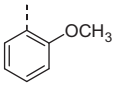
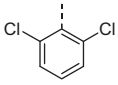
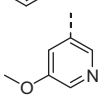
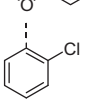
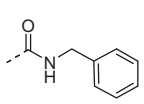
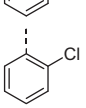
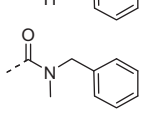
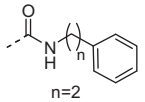
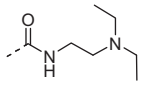
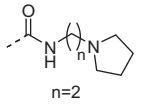
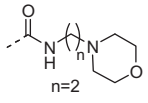
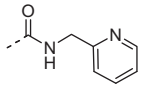
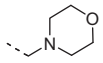
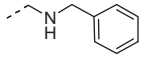
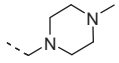
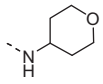
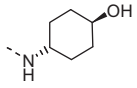
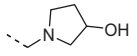
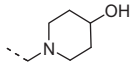
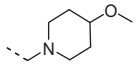
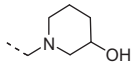
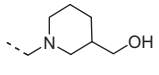
Compd	R^1	R^2	hPDE2 IC_{50}[a] (nM ± SD)	rPDE10A IC_{50}[a,b] (nM ± SD)
1a			30 (n = 1)	325 ± 39
7b			4.4 ± 1.3	35 ± 12(33 ± 6.5)
8			6.2 ± 2.5	50 ± 1.6
9			0.6 ± 0.32	74 ± 36 (29 ± 7.2)
10			5.9 ± 4.2	51 ± 22 (33, n = 1)
11			2.8 ± 1.6	52 ± 17 (41 ± 9)
12			0.28 (n = 1)	19 ± 5.4
13			263 ± 142	39 ± 13 (20, n = 1)
14			39 ± 5.7	39 ± 1.9
15			29 ± 6.6	21 ± 11
16			6.3 ± 1.5	47 ± 18
17			21 ± 2	113 ± 39
18			28 ± 5	35±12(33±6.5)
19			8.9 ± 2.6	50±1.6
20			0.38 ± 0.14	74±36 (29±7.2)
21			19 ± 1.0	51±22 (33, n=1)

Table 1 (*continued*)

Compd	R^1	R^2	hPDE2 IC_{50}[a] (nM ± SD)	rPDE10A IC_{50}[a,b] (nM ± SD)
22		 n=2	1.7 ± 1.09	52 ± 17 (41±9)
23			1.4 ± 0.31	19 ± 5.4
24		 n=2	2.2 ± 0.57	39 ± 13 (20, *n* = 1)
25		 n=2	0.47 ± 0.11	39 ± 1.9
26			0.42 ± 0.05	21 ± 11
29			24 ± 16	47 ± 18
30			20 ± 3.2	113 ± 39
31			2.8 ± 1.0	35 ± 12(33 ± 6.5)
32			15 ± 1.7	50 ± 1.6
33			7.1 ± 2.5	74 ± 36 (29 ± 7.2)
34			7.3 ± 1.7	51 ± 22 (33, *n* = 1)
35			12 ± 3.6	52 ± 17 (41 ± 9)
36			5.0 ± 2.5	19 ± 5.4
37			9.7 ± 2.4	39 ± 13 (20, *n* = 1)
38			15 ± 11	39 ± 1.9
39			6.3 ± 3.9	21 ± 11
40			24 ± 13	47 ± 18

[a] Values are mean ± SD of at least two experiments unless specified.
[b] Values in brackets refer to data generated using isolated recombinant human PDE10A enzyme.

In summary, we have identified and synthesized a series of 1-aryl-4-methyl[1,2,4]triazolo[4,3-*a*]quinoxaline derivatives as novel combined PDE2/PDE10 inhibitors, selective over all other PDEs. From this investigation we identified several highly potent compounds possessing IC_{50} values for PDE2 and PDE10 in the (sub)nanomolar range. Substituted amides were not brain penetrant and therefore we synthesized a series of related aminomethyl derivatives that nicely showed combined PDE2/PDE10 inhibitory activity. Among them, we selected compound **31** in view of its good potency (IC_{50} 2.8 nM for PDE2 and 35 nM for PDE10) for a

Table 2
Brain and plasma levels for dual PDE2/PDE10 inhibitors 1 h after administration[a]

Compd	Route	(*B*)	(*P*)	*B*/*P* ratio
9[b]	p.o.	185 ± 44	1880 ± 805	0.11 ± 0.03
17[c]	sc	98 ± 22	2800 ± 537	0.036 ± 0.01
19[c,d]	p.o.	16 ± 2	467 ± 83	0.034 ± 0.01
24[b,e]	sc	143 ± 36	1073 ± 64	0.14 ± 0.04
25[b]	sc	88 ± 22	2540 ± 295	0.034 ±0.005
29[c]	sc	628 ± 204	845 ± 78	0.73 ± 0.17
31[c,e]	sc	189 ± 78	299 ± 120	0.62 ± 0.03
40[b]	sc	99 ± 8	569 ± 93	0.18 ± 0.02

[a] (*B*): brain levels in ng/g; (*P*): plasma levels in ng/mL. Compounds formulated in 20% HP-β-CD at pH 4. Data are expressed as geometric mean values of at least two runs ± the standard error measurement (SEM).
[b] Study in male Wistar rats dosed at 10 mg/kg.
[c] Study in male Sprague Dawley rats dosed at 10 mg/kg.
[d] Formulated in 0.5% methocel suspension.
[e] Formulated in H_2O + tartaric acid.

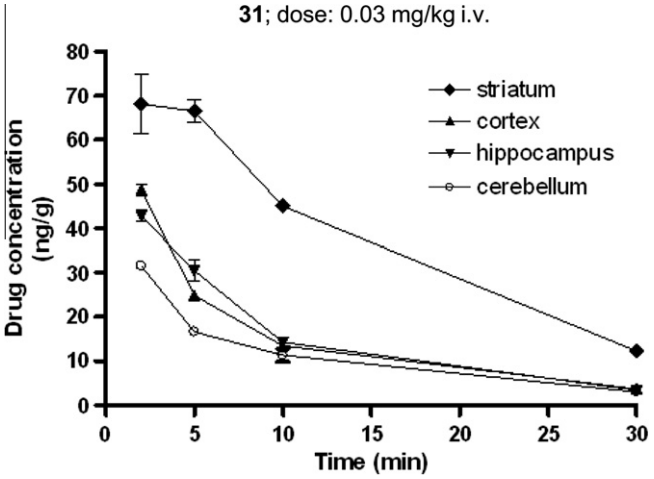


Figure 2. Microdosing experiment for compound **31**.

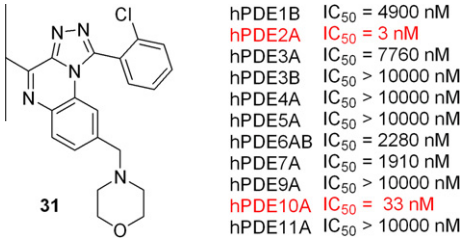


Figure 3. PDE profile of compound **31**.

microdosing study to ascertain its ability to enter the brain and bind specifically to both enzymes. The results obtained with **31** proved that this derivative possessed acceptable brain uptake and was able to bind selectively to PDE2 and PDE10 enzymes. These promising results make **31** a valuable tool compound to further study the implication of dual PDE2/PDE10 inhibitors on several neurological pathologies, opening the way to the synthesis of new more potent and drug-like compounds.

Acknowledgments

The authors thank Ilse Lenaerts for the in vivo experimental work of the microdosing study, and Lieve Dillen for the measurements of compound concentration in the rat brain samples. They also thank members of the analysis and purification group of Janssen R&D, Spain.

Supplementary data

Supplementary data (the experimental details of the synthesis of key intermediates required for the preparation of final compound **31**, and the protocols for in vitro PDE's inhibition are provided) associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.bmcl.2012.11.077.

References and notes

1. Conti, M.; Beavo, J. *Annu. Rev. Biochem.* **2007**, *76*, 481.
2. Mennitti, F. S.; Faraci, W. S.; Schmidt, C. J. *Nat. Rev. Drug Disc.* **2006**, *5*, 660.
3. (a) Conti, M. *Mol. Endocrinol.* **2000**, *14*, 1317; (b) Soderling, S. H.; Beavo, J. A. *Curr. Opin. Cell Biol.* **2000**, *12*, 174; (c) Mehats, C.; Andersen, C. B.; Filopanti, M.; Jin, S. L.; Conti, M. *Trends Endocrinol. Metab.* **2002**, *13*, 29.
4. (a) Van Staveren, W. C.; Steinbusch, H. W.; Markerink-Van Ittersum, M.; Repaske, D. R.; Goy, M. F.; Kotera, J.; Omori, K.; Beavo, J. A.; De Vente, J. *J. Comp. Neurol.* **2003**, *467*, 566; (b) Yanaka, N.; Kurosawa, Y.; Minami, K.; Kawai, E.; Omori, K. *Biosci. Biochem.* **2003**, *67*, 973.
5. (a) Soderling, S. H.; Bayuga, S. J.; Beavo, J. A. *Proc. Natl. Acad. Sci. U.S.A.* **1999**, *96*, 7071; (b) Fujishige, K.; Kotera, J.; Michibata, H.; Yuasa, K.; Takebayashi, S.; Okumura, K.; Omori, K. *J. Biol. Chem.* **1999**, *274*, 18438; (c) Loughney, K.; Snyder, P. B.; Uher, L.; Rosman, G. J.; Ferguson, K.; Florio, V. A. *Gene* **1999**, *234*, 109.
6. (a) Blokland, A.; Schreiber, R.; Prickaerts, J. *Curr. Pharm. Des.* **2006**, *12*, 2511; (b) Prickaerts, J.; de Vente, J.; Honig, W.; Steinbusch, H. W.; Blokland, A. *Eur. J. Pharmacol.* **2002**, *436*, 83.
7. Xu, Y.; Zhang, H.-T.; O'Donnell, J. M. *Neurobiol. Depress.* **2012**, 419.
8. Lakics, V.; Karran, E. H.; Boess, F. G. *Neuropharmacology* **2010**, *59*, 367.
9. Siuciak, J. A.; Chapin, D. S.; Harms, J. F.; Lebel, L. A.; McCarthy, S. A.; Chambers, L.; Shrikhande, A.; Wong, S.; Menniti, F. S.; Schmidt, C. J. *Neuropharmacology* **2006**, *51*, 386.
10. Grauer, S. M.; Pulito, V. L.; Navarra, R. L.; Rachel, L.; Kelly, M. P.; Kelley, C.; Graf, R.; Langen, B.; Logue, S.; Brennan, J.; Jiang, L.; Charych, E.; Egerland, U.; Liu, F.; Marquis, K. L.; Malamas, M.; Hage, T.; Comery, T. A.; Brandon, N. J. *J. Pharmacol. Exp. Ther.* **2009**, *331*, 574.
11. (a) Giampà, C.; Laurenti, D.; Anzilotti, S.; Bernardi, G.; Menniti, F. S.; Fusco, F. R. *PLoS ONE* **2010**, *5*, e13417; (b) Giorgi, M.; Melchiorri, G.; Nuccetelli, V.; D'Angelo, V.; Martorana, A.; Sorge, R.; Castelli, V.; Bernardi, G.; Sancesario, G. *Neurobiol. Dis.* **2011**, *43*, 293.
12. Rundfeldt, C.; Socala, K.; Wlaz, P. *J. Neural Transm.* **2010**, *117*, 1319.
13. (a) Stange, H.; Langen, B.; Egerland, U.; Hoefgen, N.; Priebs, M.; Malamas, M. S.; Erdei, J.; Ni, Y. PCT Int. Appl. WO2010054253, May 14, **2010**.; (b) Stange, H.; Langen, B.; Egerland, U.; Hoefgen, N.; Priebs, M.; Malamas, M. S.; Erdei, J. J.; Ni, Y. PCT Int. Appl. WO2010054260, May 14, **2010**.
14. While we were elaborating this manuscript a patent application describing closely related derivatives as PDE2 and/or PDE10 inhibitors has been published. Lankau, H.-J.; Langen, B.; Grunwald, C.; Höfgen, N.; Stange, H.; Dost, R.; Egerland, U. PCT Int. Appl. WO2012104293, August 9, **2012**.
15. Synthesis of 1-(2-chlorophenyl)-4-methyl-8-(morpholin-4-ylmethyl)[1,2,4]triazolo[4,3-a]-quinoxaline (**31**). The synthesis of intermediates **4b**, **5b**, **7k**, **27**, and **28** are provided as Supporting Information. Morpholine (1.37 mL, 15.67 mmol) was added to a stirred solution of intermediate **28** (2.3 g, 7.12 mmol) dissolved in 1,2-dichloroethane (50 mL) and the mixture was heated at 80 °C for 15 min under microwave irradiation (the reaction was divided in three batches). Then sodium triacetoxyborohydride (1.81 g, 8.55 mmol) was added portionwise and the mixture was heated again at the same conditions as before for 20 min. The mixture was then quenched with H_2O and extracted with CH_2Cl_2. The organic layer was separated, dried (Na_2SO_4), filtered and the solvent evaporated in vacuo. The crude compound was purified by chromatography (silica, MeOH in EtOAC 2/98 to 10/90) the desired fractions were collected and the solvent evaporated to yield final compound **31** as pale yellow solid that was further triturated with diethyl ether/DIPE and dried in vacuo (1.6 g, 57%). ^{1}H NMR (400 MHz, $CDCl_3$) δ ppm 2.24–2.41 (m, 4 H), 3.08 (s, 3 H), 3.42 (s, 2 H), 3.53–3.69 (m, 4 H), 7.37 (d, *J* = 1.2 Hz, 1 H), 7.49 (dd, *J* = 8.3, 1.6 Hz, 1 H), 7.54–7.62 (m, 1 H), 7.64–7.75 (m, 3 H), 7.99 (d, *J* = 8.3 Hz, 1 H). Mp 160.4 °C.
16. Protocols for in vitro PDE's inhibition are provided as Supplementary data.
17. Male Wistar rats (180–200 g) were injected intravenously with 0.03 mg/kg of compound **31** and sacrificed at 2, 5, 10 and 30 min after injection (3 rats per time point). Brains were rapidly removed and dissected. Striatum, hippocampus, whole cortex and cerebellum were homogenized using an ultrasonic dismembrator probe (Branson) in 4 volumes of 75% acetonitrile. After centrifugation, drug concentration was measured in each tissue using liquid chromatography coupled to mass spectroscopy.