Pergamon

Bioorganic & Medicinal Chemistry Letters 12 (2002) 3341–3344

BIOORGANIC & MEDICINAL CHEMISTRY LETTERS

SB-656104-A: A Novel 5-HT$_7$ Receptor Antagonist with Improved In Vivo Properties

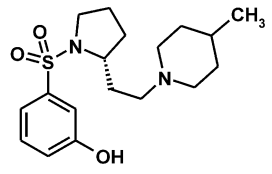
Ian T. Forbes,* Sara Douglas, Andrew D. Gribble, Robert J. Ife, Andrew P. Lightfoot, Ashley E. Garner, Graham J. Riley, Phillip Jeffrey, Alexander J. Stevens, Tania O. Stean and David R. Thomas

GlaxoSmithKline, New Frontiers Science Park, Harlow, Essex CM19 5AW, UK

Received 17 June 2002; accepted 6 August 2002

Abstract—A focused SAR study around the previously reported selective 5-HT$_7$ receptor antagonist, SB-269970-A has resulted in the identification of a structurally related analogue having an improved pharmacokinetic profile. Replacement of the phenolic group in SB-269970-A with an indole moiety, and replacement of the piperidinyl 4-methyl group with a heterocyclic ring system proved to be the key changes leading to the identification of SB-656104-A.

The 5-HT$_7$ receptor is the most recent addition to the serotonin sub-family of G-protein coupled receptors.[1] Receptor localisation studies in various species indicate the presence of 5-HT$_7$ receptors both centrally and peripherally, with the highest receptor densities being located in the thalamus, hypothalamus, limbic and cortical regions of the brain.[2] Although the biological function(s) of this receptor are poorly understood, recent reports suggest that 5-HT$_7$ receptors are involved in the control of circadian rhythms,[3] or in the aetiology of depression.[4] Further clarification of these hypotheses awaits the discovery and evaluation of selective tool compounds. We have previously reported that the selective 5-HT$_7$ receptor antagonist SB-269970-A inhibits rapid eye movement (REM) sleep in rats, suggesting involvement of 5-HT$_7$ receptors in the physiology of sleep.[5]



SB-269970-A

Unfortunately, further studies with SB-269970-A have been hampered by its extremely high in vivo blood clearance in the rat (CL_b 140 mL/min/kg),[5] almost certainly due to the presence of the phenolic hydroxyl group and Phase II metabolism. Thus we embarked upon a chemical programme aimed at replacing the phenolic moiety in SB-269970-A with metabolically more stable bioisosteres.

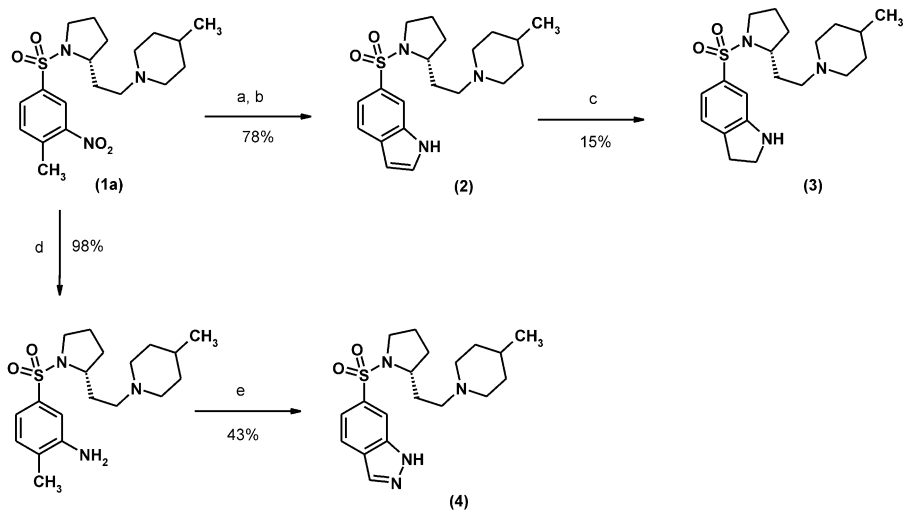
Initially, we investigated the synthesis of heterocyclic analogues of SB-269970-A, since this strategy has proven successful in many related cases.[6] Compounds in Table 1 were synthesised via two main strategies. Route A (Scheme 1) involved the preparation of the key 4-methyl-3-nitrosulfonamide intermediate (**1a**) using the previously reported methodology.[7] Leimgruber–Batcho transformation[8] afforded the indole (**2**), which was reduced with sodium cyanoborohydride to the indoline (**3**). Direct reduction of the nitro group in (**1a**) followed by diazotisation afforded the indazole (**4**).

The alternative strategy (Route B) involved the key intermediate (**1b**). Reaction with ammonia or sodium hydroxide afforded the corresponding 4-amino and 4-hydroxy analogues respectively, which were converted using standard methodologies to the benzimidazole (**5**), benzotriazole (**6**), benzimidazolone (**7**) and the benzoxazolidinone (**8**) (Scheme 2).

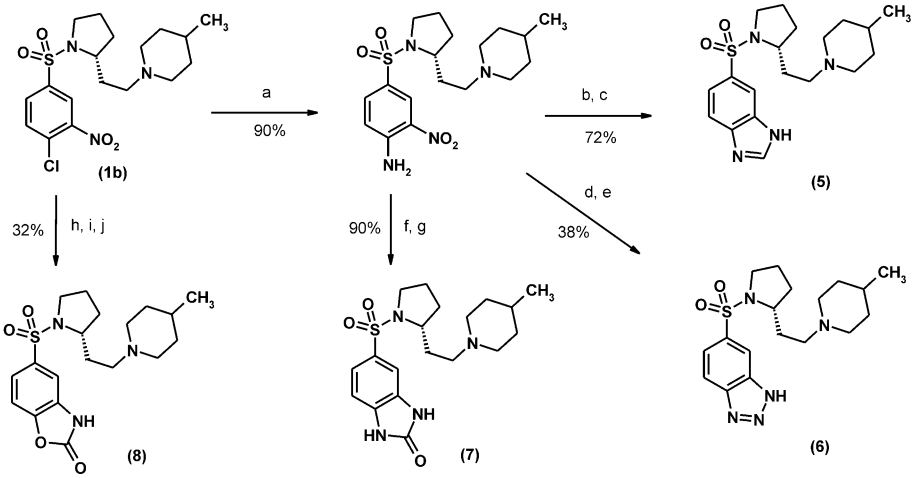
The 5-HT$_7$ binding affinities[9] for compounds (**2**)–(**8**) are shown in Table 1. It can be seen that in all cases, replacement of the phenol with a heterocyclic ring caused a reduction in activity of varying degrees. Thus the indole (**2**), indazole (**4**) and benzimidazole (**5**) retained significant

*Corresponding author. Tel.: +44-1279-622126; fax: +44-1279-622790; e-mail: ian_t_forbes@gsk.com

0960-894X/02/$ - see front matter © 2002 Elsevier Science Ltd. All rights reserved.
PII: S0960-894X(02)00690-X



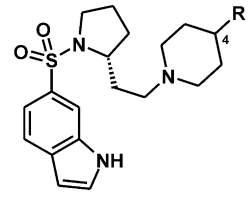
Scheme 1. Reagents and conditions for route A: (a) dimethylformamide dimethylacetal, 100 °C, 6 h; (b) Raney Nickel, hydrazine, MeOH, THF, 45 °C; (c) sodium cyanoborohydride, acetic acid, rt, 4 h; (d) Raney Nickel, hydrazine, MeOH, THF, 45 °C; (e) $NaNO_2$, acetic acid, rt, 24 h.



Scheme 2. Reagents and conditions for route B: (a) aqueous ammonia, dioxan, 90 °C, sealed tube; (b) 50 psi H_2, 10% palladium–charcoal, EtOH; (c) formic acid, $NaHCO_3$, H_2O, 100 °C; (d) 50 psi H_2, 10% palladium–charcoal, EtOH; (e) $NaNO_2$, acetic acid, HCl, rt; (f) 50 psi H_2, 10% palladium–charcoal, EtOH; (g) phosgene, CH_2Cl_2, Hunig's base; (h) NaOH, dioxan, reflux; (i) 50 psi H_2, 10% palladium–charcoal, EtOH; (j) phosgene, CH_2Cl_2, Hunig's base.

levels of 5-HT_7 binding affinity, whereas the benzimidazolidinone (**7**) and the oxazolidinone (**8**) showed the greatest reduction in affinity. Attempts to correlate the pK_a of the heterocyclic NH with the 5-HT_7 receptor affinity failed to reveal any trend. In the case of (**7**) and (**8**) it appears that a carbonyl group at C-2 is not well tolerated, suggesting that polarity and/or steric interactions may be influencing factors. Overall, the most interesting analogue was the indole (**2**), which was one of the few analogues to possess a similar 5-HT_7 receptor affinity to the phenol SB-269970-A. Unfortunately, (**2**) showed high blood clearance and zero bioavailability in the rat.[10] In vitro metabolism studies on closely related structures indicated that hydroxylation may be occurring on the 4-methylpiperidine ring system,[10] so we followed up these leads with modifications to the piperidine ring.

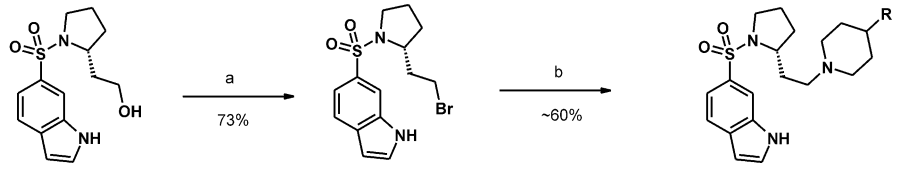
Initially, we investigated the preparation of alternative R groups at the C-4 position in (2). We were particularly interested in aromatic C-4 substituents, since receptor modelling studies indicated the presence of a large lipophilic pocket around this region.



(**2**) R = CH_3

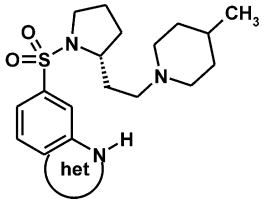
The preparation of analogues (**9**)–(**16**) involved similar methodologies to those for (**2**), except that the indole ring system was introduced prior to introduction of the 4-substituted piperidine moiety, allowing parallel synthesis of target compounds (Scheme 3).

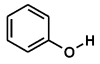
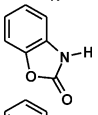
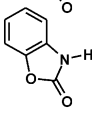
It was most gratifying to see that replacement of the 4-methyl group in (**2**) with a variety of aromatic groups



Scheme 3. Reagents and conditions: (a) carbon tetrabromide, triphenylphosphine, CH_2Cl_2, 0°C, 1 h; (b) 4-substituted piperidine, MeCN, $NaHCO_3$, 80°C, 12 h.

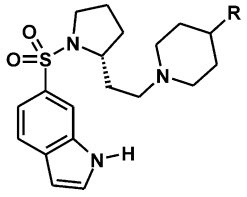
Table 1. Heterocyclic phenol isosteres



No.	het	pK_i[9] 5-HT$_7$	No.	het	pK_i[9] 5-HT$_7$
SB-269970-A		8.90±0.10	**5**		8.19±0.11
2		8.62±0.10	**6**		7.59±0.04
3		7.64±0.10	**7**		7.24±0.04
4		8.19±0.03	**8**		6.78±0.10

significantly enhanced the 5-HT$_7$ receptor affinity (Table 2). The greatest increases were seen with the indole (**9**), the benzimidazolone (**11**) and the 4-fluorophenoxy analogue (**14**). Unfortunately, it was found that these modifications also markedly increased unwanted affinity for the adrenergic α_{1B} receptor, with the greatest potentiation being observed with compounds (**9**) and (**16**). Only compounds (**12**) and (**13**) retained relatively low affinity for this receptor. Overall, the 4-chlorophenoxy derivative (**13**; free base SB-656104; hydrochloride salt SB-656104-A) was selected as the most promising analogue for further studies.

Table 2. 4-Substituted piperidines



No.	R	pK_i[9] 5-HT$_7$	pK_i[9] α_{1B}
2	Me	8.62±0.10	<5
9	Indol-3-yl	9.17±0.03	8.89±0.12
10	Indol-2-yl	8.20±0.04	7.05±0.05
11	Benzimidazol-2-on-3-yl	9.15±0.08	7.36±0.03
12	Benzoxazol-2-on-3-yl	8.74±0.02	6.51±0.11
13	4-Chlorophenoxy	8.70±0.10	6.66±0.01
14	4-Fluorophenoxy	9.30±0.06	7.52±0.01
15	4-Chlorobenzoyl	9.10±0.12	7.03±0.07
16	4-Fluorobenzoyl	9.08±0.02	7.72±0.06

SB-656104 was evaluated in a previously described functional model of 5-HT$_7$ receptor activation,[5] blocking 5-carboxamidotryptamine (5-CT) stimulated adeny-

Table 3. Receptor binding selectivity profile of SB-656104

Receptor	Affinity (pK_i)
5-HT$_{1A}$	6.25±0.06
5-HT$_{1B}$	6.20±0.05
5-HT$_{1D}$	7.60±0.02
5-HT$_{1E}$	<5.3
5-HT$_{1F}$	<5.7
5-HT$_{2A}$	7.20±0.02
5-HT$_{2B}$	7.04±0.08
5-HT$_{2C}$	6.57±0.08
5-HT$_4$	5.72±0.10
5-HT$_{5A}$	6.74±0.01
5-HT$_6$	6.07±0.06
5-HT$_7$	8.70±0.10

Table 4. In vivo comparison of SB-269970-A and SB-656104[11]

In vivo data	SB-269970-A	SB-656104[a]
CL_b (rat)	140 mL/min/kg	57±4 mL/min/kg
$t_{1/2}$ (rat)	<0.5 h	2.0±0.2 h
Steady state brain:blood ratio	0.8	0.9
F_{po} (rat)	Not measured	16±8%
ED_{50} to antagonise 5-CT induced hypothermia in guinea pig[5]	3.0±0.2 mg/kg ip (Maximal inhibition at 1 h)	2.0±0.01 mg/kg po (Maximal inhibition at 2 h)

[a]Free base used for DMPK studies; hydrochloride salt used for guinea pig hypothermia model.

lyl cyclase activity with a calculated pA_2 of 8.1±0.1, confirming its antagonist profile. The selectivity profile for SB-656104 across the range of serotonin receptor sub-types is shown in Table 3. Encouragingly, SB-656104 shows ≥100-fold binding selectivity over most other 5-HT receptor subtypes, with the exception of 5-HT_{2A} (30-fold), 5-HT_{2B} (50-fold) and 5-HT_{1D} (12-fold).

Of crucial importance was the in vivo profile of SB-656104, which is summarised in Table 4, in comparison with the former tool compound SB-269970-A. Importantly, SB-656104 shows a greatly improved pharmacokinetic profile in the rat compared to SB-269970-A, possessing a lower blood clearance and a significantly improved half life. Brain penetration with SB-656104 is similar to that measured for SB-269970-A (Table 4). Significantly, the presence of the 4-chlorophenoxy piperidine ring in SB-656104 confers oral bioavailability (16%) which was notably lacking in the earlier lead (**2**). Evaluation of SB-656104-A (hydrochloride salt) in an in vivo functional assay (5-CT induced hypothermia in guinea pigs)[5] resulted in a similar ED_{50} profile to SB-269970-A, *but with administration being* via *the oral route*.

In summary, through a rational SAR study based around SB-269970-A we have identifed SB-656104-A as a novel, potent and selective 5-HT_7 receptor antagonist with a greatly improved pharmackinetic profile, which represents an improved tool compound for characterisation of 5-HT_7 receptors in the CNS. Further studies with SB-656104-A will be the subject of a future publication.[12]

References and Notes

1. For a recent review of serotonin receptors see: Hoyer, D.; Hannon, J. P.; Martin, G. R. *Pharmacol. Biochem. Behav.* **2002**, *71*, 533.
2. (a) Bard, J. A.; Zgombick, J.; Adham, N.; Vaysee, P.; Branchek, T. A.; Weinshank, R. L. *J. Biol. Chem.* **1993**, *268*, 23422. (b) Ruat, M.; Traiffort, E.; Leurs, R.; Tardivel-Lacombe, J.; Diaz, J.; Arrang, J.-M.; Schwarz, J. C. *Proc. Natl. Acad. Sci.* **1993**, *90*, 8547. (c) To, Z. P.; Bonhaus, D. W.; Eglen, R. M.; Jakeman, L. B. *Br. J. Pharm.* **1995**, *115*, 107. (d) Shen, Y.; Monsma, F. J.; Metcalf, M. A.; Jose, P. A.; Hamblin, M. W.; Sibley, D. R. *J. Biol. Chem.* **1993**, *268*, 18200.
3. Lovenberg, T. W.; Baron, B. M.; Lecea, L. de.; Miller, J. O.; Prosser, R. A.; Rea, M. A.; Foye, P. E.; Rucke, M.; Slone, A. L.; Siegel, B. W.; Danielson, P. E.; Sutcliffe, J. G.; Erlander, M. G. *Neuron* **1993**, *11*, 449.
4. Guscott, M. Poster Presentation, Society for Neurosciences Satellite Meeting on Serotonin, New Orleans, November, 2000.
5. Hagan, J. J.; Price, G. W.; Jeffrey, P.; Deeks, N. J.; Stean, T.; Piper, D.; Smith, M. I.; Upton, N.; Medhurst, A. D.; Middlemiss, D. N.; Riley, G. J.; Lovell, P. J.; Bromidge, S. M.; Thomas, D. R. *Br. J. Pharm.* **2000**, *130*, 539.
6. For a recent example see: Wright, J. L.; Gregory, T. F.; Kesten, S. R.; Boxer, P. A.; Serpa, K. A.; Meltzer, L. T.; Wise, L. D.; Espitia, S. A.; Konkoy, C. S.; Whittemore, E. R.; Woodward, R. M. *J. Med. Chem.* **2000**, *43*, 3408.
7. Lovell, P. J.; Bromidge, S. M.; Dabbs, S.; Duckworth, D. M.; Forbes, I. T.; Jennings, A. J.; King, F. D.; Middlemiss, D. N.; Rahman, S. K.; Saunders, D. V.; Collin, L. L.; Hagan, J. J.; Riley, G. J.; Thomas, D. R. *J. Med. Chem.* **2000**, *43*, 342.
8. Batcho, A. D.; Leimgruber, W. *Org. Synth.* **1985**, *63*, 214.
9. pK_i values represent the mean of at least three determinations using cell lines stably expressing the cloned human receptor. For details of the radioligand binding assays, see ref 7.
10. Unpublished DMPK data from GlaxoSmithKline
11. DMPK protocol for SB-269970-A as previously published.[5] For SB-656104 (free base), iv dosing in male Sprague–Dawley rats ($n=3$): constant infusion over 1 h to achieve target dose of 1 mg free base/kg; oral dosing: an oral suspension of SB-656104 was administered to male Sprague–Dawley rats ($n=3$) at a target dose of 3 mg free base/kg. Blood and brain homogenate samples were analysed by HPLC/MS/MS.
12. Forbes, I. I.; Douglas, S.; Gribble, A. D.; Ife, R. J.; Lightfoot, A. P.; Garner, A. E.; Riley, G. J.; Jeffrey, P.; Massagrande, M.; Melotto, S.; Stevens, A. J.; Stean, T. O.; Thomas, D. R. manuscript in preparation.