Bioorganic & Medicinal Chemistry Letters 21 (2011) 1498–1501

Contents lists available at ScienceDirect

Bioorganic & Medicinal Chemistry Letters

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

Identification of a new series of non-peptidic NK_3 receptor antagonists

Karsten Juhl [a,*], Tore Hansen [a,†], Jan Kehler [a], Nikolay A. Khanzhin [a], Morten B. Nørgaard [a], Thomas Ruhland [a], Dorrit B. Larsen [a], Klaus G. Jensen [a], Björn Steiniger-Brach [b], Søren M. Nielsen [b], Klaus B. Simonsen [a]

[a] Discovery Chemistry and DMPK, H. Lundbeck A/S, Otilliavej 9, DK 2500 Valby, Denmark
[b] Discovery Pharmacology Research, H. Lundbeck, Otilliavej 9, DK 2500 Valby, Denmark

ARTICLE INFO

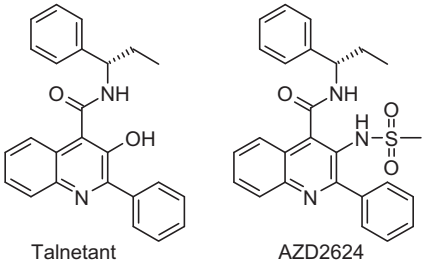
Article history:
Received 2 November 2010
Revised 29 December 2010
Accepted 31 December 2010
Available online 13 January 2011

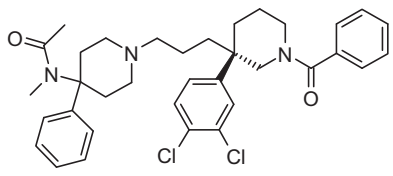
Keywords:
NK_3 antagonist
Schizophrenia
Cyclopropane
Senktide induced hypoactivity

ABSTRACT

The identification and structure–activity relationships of 2-aminomethyl-1-aryl cyclopropane carboxamides as novel NK_3 receptor antagonists are reported. The compound series was optimized to give analogues with low nanomolar binding to the NK_3 receptor and brain exposure, leading to activity in vivo in the senktide-induced hypoactivity model in gerbils.

The NK_3 receptor is one of three known neurokinin receptors. The NK_3 receptor is expressed both centrally and in some peripheral organs.[1–3] The expression in the central nervous system (CNS) and the modulatory effect on various monoaminergic transmitter systems[4] make NK_3 receptor antagonists interesting as potential therapeutic agents for the treatment of CNS diseases such as schizophrenia,[5] Parkinsons disease,[6] panic attacks and major depressive disorder.[7]





Three non-peptide based NK_3 receptor antagonists have entered the clinical trials: Talnetant and AZD2624 from a common 2-aryl-4-carboxamide-quinoline core structure and Osanetant from a piperidine amide series. All three compounds have been withdrawn from further clinical development.

As a result of an NK_3-focused screening campaign including compounds from a previous neurokinin project, we identified a new structural class of non-peptidic NK_3 receptor antagonists. Specifically, we found lead compound **1a** and structurally related analogues that derived from a tri-substituted cyclopropane scaffold. This scaffold is capable of arranging three distinct functionalities (a carboxamide, an aryl and an aminomethyl) in a well-defined orientation. Compound **1a** displays high affinity for the NK_3 receptor (K_i = 5.9 nM) and selectivity towards the NK_1 and NK_2 receptors (K_i = 130 nM and K_i = 88 nM, respectively).[8]

The synthesis of **1a** (and derivatives) commenced from a diazo-transfer from *p*-acetamidobenzenesulfonyl azide (*p*-ABSA) to the phenyl acetic acid allyl ester **2** (see Fig. 1). The resulting diazo compound **3** was converted through intramolecular cyclopropanation to the racemic lactone **4.** The racemate was resolved either through chiral chromatography separation of the two enantiomers (1*S*,5*R*)-**4** and (1*R*,5*S*)-**4** or through a series of chemical modifications involving (a) lewis acid mediated lactone opening with (*R*)-α-methyl benzyl amine, (b) separation of the resulting two diastereomers **5a** and **5b**, (c) acid-mediated conversion of **5a** and **5b** to the enantiomeric pure lactones. Alternatively, analogues of (1*S*,5*R*)-**4** were synthesized from (*R*)-(−)-epichlorohydrin and the appropriate benzyl cyanide.[9] The synthesis continued by opening the lactone ring with various benzyl amines mediated by $AlCl_3$. Conversion of the alcohol **6** to the bromide and subsequent treatment with an amine completed the synthesis of **1a**.

* Corresponding author. Tel.: +45 36434041; fax: +45 36438237.
E-mail address: kaju@lundbeck.com (K. Juhl).
† Present address: University of Oslo, Department of Chemistry, Sem Sælands vei 26, 0315 Oslo, Norway.

0960-894X/$ - see front matter © 2011 Elsevier Ltd. All rights reserved.
doi:10.1016/j.bmcl.2010.12.135

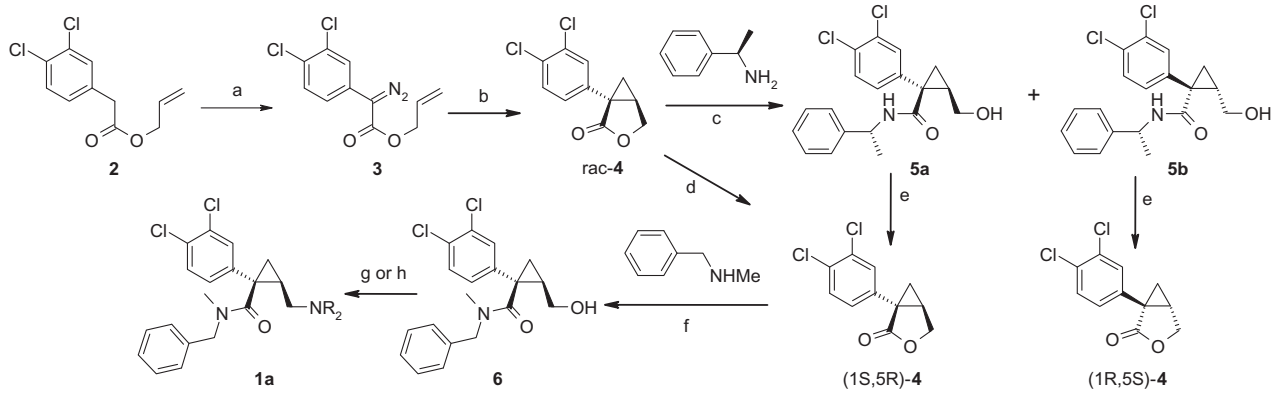
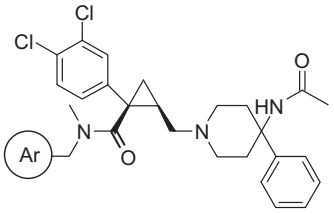


Figure 1. Reagents and conditions: (a) *p*-ABSA (1.2 equiv), DBU (1.3 equiv) MeCN, 0 °C to rt, 20 h; (b) $Rh_2(Oct)_4$ (0.005 equiv), DCM, rt, 4 h; (c) $AlCl_3$ (2 equiv), DCE, column chromatography; (d) chiral HPLC separation; (e) H_2SO_4; (f) benzyl amine, $AlCl_3$ (2 equiv), DCE, 30 min; **4**, 60 min; (g) (i) PBr_3 (3 equiv), DCM, 3 h (ii) amine, Et(*i*Pr)$_3$N, MeCN, 85 °C, 3 h or (i) IBX, 1,2-dichloroethane, (ii) amine, $Na(CN)BH_3$.

The variation of the carboxamide group was the most synthetically feasible approach to systematically analyze the structure–activity relationships (SAR) of this part of the scaffold. In parallel manner, we varied the *N*-methyl-*N*-benzyl amide moiety while keeping the remainder of the molecule unchanged as shown in Table 1. The results indicate that the NK_3 receptor affinities are not dramatically influenced by substituents on the *N*-benzyl moiety. A screen of other amides with substituents different from *N*-methyl and *N*-benzyl was also performed. However, this led to compounds with lower affinity for the NK_3 receptor (data not shown). On the basis of these SAR, the *N*-benzyl-*N*-methyl and *N*-4-fluorobenzyl-*N*-methyl amide moieties were selected to be the optimal substitution patterns. Furthermore, the data in Table 1 indicate a relatively high degree of stereoselectivity in the cyclopropane scaffold with the (1*S*,2*R*) configuration giving superior affinity for the NK_3 receptor. Therefore the (1*S*,2*R*) isomer was chosen for further SAR studies.

The aryl group directly bond to the cyclopropane ring is most sterically restricted and we speculated that substituents in this part of the scaffold would likely effect receptor affinity. Therefore our next design iteration was devoted to vary the substitution pattern of this aromatic ring. (Table 2).[10] In this study, it was found that the original 3,4-dichloro substitution pattern was superior. While compounds (e.g., **7**, **8** and **9**) with either neutral or electron-withdrawing substituents maintain NK_3 receptor affinity, more polar substituents such

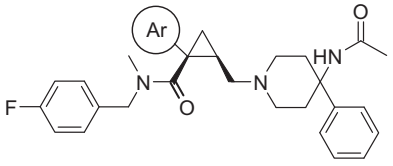
Table 1
SAR of *N*-benzyl amide analogues



#	Stereo-chemistry	Ar	hNK$_3$R K_i (nM)[a]
1a	(1*S*, 2*R*)	C_6H_5	5.9
1b	(1*R*, 2*S*)	C_6H_5	83
7	*rac*	2-Cl-C_6H_4	20
8	*rac*	2-F-C_6H_4	18
9	*rac*	2-Me-C_6H_4	5.3
10	*rac*	2-OMe-C_6H_4	14
11	*rac*	3-F-C_6H_4	12
12	*rac*	3-Cl-C_6H_4	24
13	*rac*	3-OMe-C_6H_4	33
14	*rac*	3-CF_3-C_6H_4	50
15	(1*S*, 2*R*)	4-F-C_6H_4	5.8
16	*rac*	4-Cl-C_6H_4	11
17	*rac*	4-Me-C_6H_4	7.5
18	*rac*	4-OMe-C_6H_4	15
19	*rac*	4-CF_3-C_6H_4	4.6
20	*rac*	4-SMe-C_6H_4	28
21	*rac*	2,3-Cl_2-C_6H_3	24
22	*rac*	2,4-Cl_2-C_6H_3	16
23	*rac*	3,4-Cl_2-C_6H_3	13
24	*rac*	3,5-Cl_2-C_6H_3	23
25	*rac*	2,4-OMe_2-C_6H_3	5.9
26	*rac*	3,4-OMe_2-C_6H_3	67
27	*rac*	3,5-OMe_2-C_6H_3	32
28	*rac*	2-Thiophenyl	70
29	*rac*	3-Thiophenyl	14

[a] Displacement of ^{3}H-labelled Lu AE93103 from the cloned hNK$_3$ receptor expressed in BHK cells. Data are means (n = 2 or more).

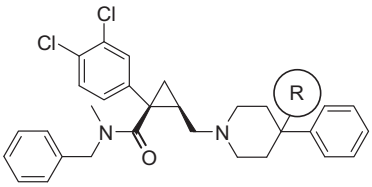
Table 2
SAR of aromatic ring on cyclopropane core

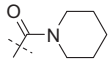
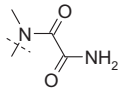


#	Ar	hNK$_3$R K_i (nM)[a]
30	4-F-C_6H_4	67
31	4-Cl-C_6H_4	36
32	4-Me-C_6H_4	37
33	4-OMe-C_6H_4	350
34	4-CF_3-C_6H_4	1000
35	4-CN-C_6H_4	270
36	3-F-C_6H_4	140
37	3-Cl-C_6H_4	32
38	3-Me-C_6H_4	40
39	3-OMe-C_6H_4	56
40	3-CF_3-C_6H_4	210
41	3-CN-C_6H_4	1900
42	3,4-F_2-C_6H_3	47
43	3,4-Cl_2-C_6H_3	5.8
44	3,5-Cl_2-C_6H_3	90
45	3,4-Me-C_6H_3	7.3
46	3,4-OCH_2O-C_6H_3	140
47	2-Thiphenyl	93
48	3-Thiophenyl	110

[a] Displacement of ^{3}H-labelled Lu AE93103 from the cloned hNK$_3$ receptor expressed in BHK cells. Data are means (n = 2 or more).

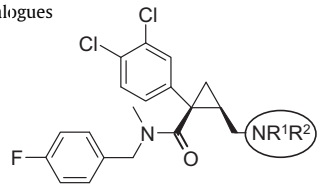
Table 3
SAR of phenyl piperidine analogues

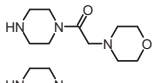
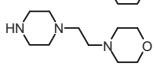
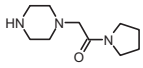
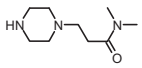
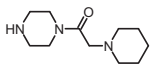
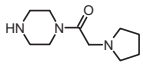
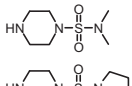
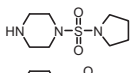
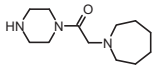
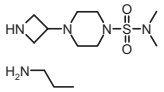


#	R	hNK$_3$R K_i[a] (nM)	Intrinsic clearance[b] (L/min)	Caco-2 Papp[c] ($\times 10^{-6}$ cm/s)	Caco-2 efflux ratio[c]
1a		5.9	3.5	5.6	2.6
49		4.2	19	0.8	12
50		6.1	>20	0.3	17
51		1.5	6.3	0.4	>100
52		2.8	>20	NA	NA
53		5.6	6.8	0.6	42
54		12	2	0.6	85
55		3.6	6.9	1.3	13
56		7.8	15	0.1	120

[a] Displacement of ^{3}H-labelled Lu AE93103 from the cloned hNK$_3$ receptor expressed in BHK cells. Data are means ($n = 2$ or more).
[b] See Ref. 9.
[c] See Ref. 10.

Table 4
SAR of piperazine analogues



#	NR^{1}R^2	hNK$_3$R K_i[a] (nM)	Intrinsic clearance[b] (L/min)	Caco-2 Papp[c] ($\times 10^{-6}$ cm/s)	Caco-2 efflux ratio[c]
57		3.3	16	35	1.5
58		3.7	4.3	NA	NA
59		1.8	12	27	1.3
60		17	8.5	28	1.1
61		7.3	10	2.9	4.2
62		12	5.6	5.7	3.5
63		7.6	>20	25	1.5
64		5.8	>20	NA	NA
65		8.7	15	NA	NA
66		1.9	12	NA	NA
67		150	0.9	5.4	2.7
68		140	0.9	11	1.5

[a] Displacement of ^{3}H-labelled Lu AE93103 from the cloned hNK$_3$ receptor expressed in BHK cells. Data are means ($n = 2$ or more).
[b] See Ref. 9.
[c] See Ref. 10.

as a cyano group, resulted in compounds (e.g., **35** and **41**) with lower affinity.

However, in contrast to binding, the ADME profile of compound **1a** was found to be poor. The intrinsic clearance in human liver microsomes[11] was high relative to the human liver-blood flow (1.4 L/min). Furthermore, a high efflux ratio and low permeability were found in the Caco-2 assay[12] (Table 3). These results are indications of poor systemic exposure and the results from the Caco-2 assay also imply poor blood–brain barrier penetration. The first attempt to improve the ADME profile of the lead series, was to vary the amide functionality located in the phenyl-piperidine-fragment. As shown in Table 3, several non-basic nitrogen containing functional groups were tolerated at the 4-position of the piperidine ring. However, only in one case the clearance was improved; a urea group rather than an acetamide group gave an intrinsic clearance of 2 L/min, but for this compound an even higher efflux ratio in the Caco-2 assay was found (**54**, Table 3).

We speculated that the efflux issue of the compound series might derive from the phenyl ring in the piperidine fragment. This prompted us to explore the SAR and ADME properties of these molecules with a selection of amines that did not carry aromatic substituents. As shown in Table 4, high affinity for the NK$_3$ receptor was maintained with a series of *N*-substituted piperazines. More

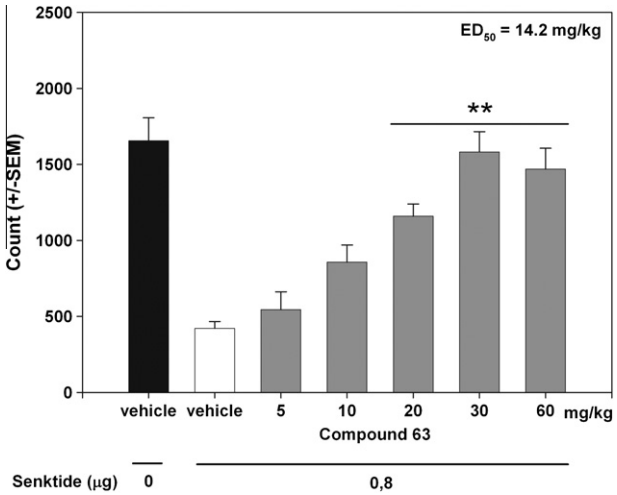


Figure 2. Senktide induced hypoactivity (gerbils). Compound **63** was administered sc, 60 min prior to senktide. Senktide was administered icv, directly before behavioural analysis was started. Data shown are the mean activity counts over a period of 30 min after senktide administration. **p <0.01 versus vehicle/senktide treatment. $n = 8$–16 per group.

importantly, the Caco-2 permeability and efflux were significantly improved and the efflux ratio approached unity (Papp >10 and efflux ratio <2 are indicative of good permeability, and that no efflux is involved). However, the stability of the compounds in human liver microsomes was not improved.

With the aim to improve the metabolic stability, a broader variation of amine substituents NR_2 on the structure shown in Table 4 was also performed. The best results from that variation came from small aliphatic amines such as compounds **67** and **68** (Table 4). These compounds have a low intrinsic clearance and a relatively small Caco-2 efflux ratio. However, the affinity for the NK_3 receptor could not be maintained.

We selected compound **63** for in vivo PK studies in rat. Intrinsic clearance in rat microsomes was 210 mL/min and correlated well with high in vivo clearance of 108 mL/min/kg. Oral bioavailability was <5% which is most likely explained by first-pass metabolism, since the Caco-2 data suggested high permeability and low efflux ratio. Compound **63** was also selected for in vivo efficacy in the senktide-induced hypoactivity assay in gerbils. The selection of this particular compound was based on its low Caco-2 efflux ratio and high permeability leading to expectation of good CNS penetration as well as on the high NK_3 receptor affinity. Compound **63** was also found to display selectivity towards the NK_1 and NK_2 receptors (K_i = 340 nM and K_i = 75 nM, respectively).[8] In this model, central administration of the selective NK_3 receptor agonist senktide induces a reduction of explorative behavior (hypoactivity), which lasts approximately 30 min.[13] The reversal of this functional read out of NK_3 receptor agonism indicates central mediated in vivo activity of the test compound. Compound **63** was administered sc in order to avoid expected first-pass metabolism, 60 min prior to senktide administration and fully reversed senktide-induced hypoactivity with an ED_{50} = 14.2 mg/kg (Fig. 2). Subsequent to the behavioral test, that is, 90 min after drug administration, four animals of each dose group were sacrificed for plasma and brain sampling. Compound **63** was found to enter well into the brain, with an average brain/plasma ratio of 1.9.

In this Letter, we have described the discovery of a series of trisubstituted cyclopropanes as novel non-peptidic NK_3 receptor antagonists. Extensive SAR analysis and targeted optimization of the ADME profile resulted in compounds with good blood–brain barrier penetration as verified by in vivo activity in the senktide-induced hypoactivity model. However, to advance this compound series further, an improvement in metabolic stability needs to be obtained.

Acknowledgements

Maibritt Glarø, Heidi E. Daugaard, Jacob Bøgesvang, Dorthe Belling, Peter Bregnedal, Bitten Hansen, Mette Larsen, Louise H. Rosendal are acknowledged for their contributions to this work.

References and notes

1. Langlois, X.; Wintmolders, C.; te Riele, P.; Leysen, J. E.; Jurzak, M. *Neuropharmacology* **2001**, *40*, 242.
2. Nagano, M.; Saitow, F.; Haneda, E.; Konishi, S.; Hayashi, M.; Suzuki, H. *Br. J. Pharmacol.* **2006**, *147*, 316.
3. Rigby, M.; O'Donnel, R.; Rupniak, N. M. J. *J. Comp. Neurol.* **2005**, *490*, 335.
4. (a) Albert, J. S.; Potts, W. *Expert Opin. Ther. Patents* **2006**, *16*, 925; (b) Smith, P. W.; Dawson, L. A. *Recent Pat. CNS Drug Discov.* **2008**, *3*, 1; (c) Dawson, L. A.; Smith, P. W. *Curr. Pharm. Des.* **2010**, *16*, 344; (d) Simonsen, K. B.; Juhl, K.; Steiniger-Brach, B.; Nielsen, S. M. *Curr. Opin. Drug Discovery Dev.* **2010**, *13*, 379.
5. Meltzer, H. Y.; Arvanitis, L.; Bauer, D.; Rein, W. *Am. J. Psychiatry* **2004**, *161*, 975.
6. Mesnage, V.; Houeto, J. L.; Bonnet, A. M.; Clavier, I.; Arnulf, I.; Cattelin, F.; Le, F. G.; Damier, P.; Welter, M. L.; Agid, Y. *Clin. Neuropharmacol.* **2004**, *27*, 108.
7. Kronenberg, G.; Berger, P.; Tauber, R. F.; Bandelow, B.; Henkel, V.; Heuser, I. *Pharmacopsychiatry* **2005**, *38*, 24.
8. *NK_1 binding*: Displacement of ^{125}I-labelled SP from the cloned hNK3 receptor. *NK_2 binding*: Displacement of ^{125}I-labelled NKA from the cloned hNK3 receptor.
9. Shuto, S.; Ono, S.; Hase, Y.; Kamiyama, N.; Takada, H.; Yamashita, K.; Matsuda, A. *J. Org. Chem.* **1996**, *61*, 915.
10. The lactone-precursors to **31**–**42**, **44**–**48** (analogues of **4**) were synthesized from (*R*)-(−)-epichlorohydrin and the appropriate benzyl cyanide as described in Ref. 7.
11. The stability of compounds in liver microsomes is determined by the *t½* method, that is, the disappearance of 1 μM drug over time was measured by LCMS. Using 0.5 mg/ml of microsomal protein (liver microsomes from several donors pooled to obtain an average enzyme content) in a NADPH generating system (1.3 mM NADP, 3.3 mM glucose 6-phosphate and 0.4 U/ml glucose 6-phosphate dehydrogenase), 3.3 mM $MgCl_2$, 0.1 M potassium phosphate buffer (pH 7.4), in a total volume of 100 μl, and stopping the incubations at time points 0, 5, 15, 30 and 60 min with 1:1 v/v acetonitrile. *t½* is subsequently scaled to the metabolic competence of a whole liver using 45 mg microsome/g liver, 20 g liver/kg and standard body weight 70 kg.
12. Larsen, M.; Holm, R.; Jensen, K. G.; Sveigaard, C.; Brodin, B.; Nielsen, C. U. *Eur. J. Pharm. Sci.* **2010**, *39*, 68.
13. Jung, M.; Michaud, J. C.; Steinberg, R.; Barnouin, M. C.; Hayar, A.; Mons, G.; Souilhac, J.; Emonds-Alt, X.; Soubrié, P.; Le Fur, G. *Neuroscience* **1996**, *74*, 403.