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Synthesis and in vitro binding studies of piperazine-alkyl-naphthamides: Impact of homology and sulphonamide/carboxamide bioisosteric replacement on the affinity for 5-HT$_{1A}$, α_{2A}, D4.2, D3 and D2L receptors

Mélissa Résimont, Jean-François Liégeois *

Laboratory of Medicinal Chemistry and C.I.R.M., University of Liège, Avenue de l'Hôpital, 1 (B36), B-4000 Liège 1, Belgium

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ABSTRACT

A series of carboxamide and sulphonamide alkyl(ethyl to hexyl)piperazine analogues were prepared and tested for their affinity to bind to a range of receptors potentially involved in psychiatric disorders. These chemical modifications led us to explore the impact of homology and bioisosteric replacement of the amide group. All of these compounds possessed a high affinity for 5-HT$_{1A}$ receptors, irrespective of the size of the linker, the carboxamide derivative with a pentyl linker had the highest affinity for α_{2A} receptor sites and also a high affinity for 5-HT$_{1A}$ and D3 receptors. The sulphonamide analogue with a hexyl linker possessed a high affinity for 5-HT$_{1A}$, D4.2 and D3 receptors.

The multiple receptor strategy remains a valuable approach for developing CNS drugs[1,2] as a complex circuitry exists in vivo as demonstrated by dopamine release studies.[3,4] Conversely, it is clear that a pharmacological tool must be selective for a specific target to prevent problems interpreting both pharmacological and psychopharmacological data. For example, WAY-100,635 is only relatively selective for 5-HT$_{1A}$ receptors, as it also displays a reasonable affinity to interact with D4 receptors.[5] Recently, we showed that some piperazinyl-ethyl carboxamide derivatives (Fig. 1) possessed a significant affinity for both 5-HT$_{1A}$ and D4.2 receptors and found that some chemical modulations tended to increase selectivity.[6]

In the present work, we were interested in extending this series by exploring two concepts of medicinal chemistry namely homology and bioisosteric replacement. Homology in this study is represented by modifying the size of the linker between the amide moiety and the basic heterocycle from an ethyl up to a hexyl chain (Fig. 1). The linker size is limited by the expected low solubility related to an increased lipophilicity. Bioisosterism in the present work concerns the replacement of a carboxamide group possessing a sp^2 hybridization and thus a trigonal planar configuration by a sulphonamide moiety with a sp^3 hybridization that presents a tetrahedral configuration. The preliminary biological evaluation was done by measuring the in vitro binding affinity of these molecules for receptors that are potentially targets for the development of putative antipsychotic drugs (e.g., 5-HT$_{1A}$, α_{2A} or D4.2 receptors).

The target compounds were synthesized by reaction of the appropriate primary amine with the appropriate acyl chloride (2-naphthoyl or 2-naphthalenesulfonyl) as mentioned in Scheme 1. The crude amines were obtained following a Gabriel procedure using the appropriate N-substituted phthalimides which were synthesized by reaction of 1-(2-methoxy-phenyl) piperazine with *N*-(ω-bromoalkyl)phthalimide in the presence of potassium carbonate. The *N*-arylpiperazine derivative was obtained from commercial sources, while *N*-(ω-bromoalkyl)phthalimide analogues were purchased or prepared in-house following classical methods and further characterised. All intermediates were used after purification and crystallization followed by a classical analytical characterisation (e.g., ^{1}H NMR, elemental analysis). Target compounds were mainly isolated as a base and further characterised.

In vitro binding experiments were conducted on membrane preparations isolated from cells transfected and expressing cloned human or rat receptors. The radioligands used were [^{3}H]-8-OH-DPAT (~0.25 nM), [^{3}H]-MK912 (~0.7 nM), [^{3}H]-YM-09151-2 (~0.2 nM), [^{3}H]-spiperone for 5-HT$_{1A}$, α_{2A}, D4.2 and D3 and D2L receptors, respectively. Experimental procedures for filtration on GF/C glass microfibre filters and radioactivity counting were as previously described[6] and are summarised in Table 1, the biological results are reported in Table 2.

The most significant finding is that all compounds interacted with 5-HT$_{1A}$ receptors irrespective of the size of the linker or the

* Corresponding author. Tel.: +32 43 66 43 77; fax: +32 43 66 43 62.
E-mail address: JF.Liegeois@ulg.ac.be (J.-F. Liégeois).

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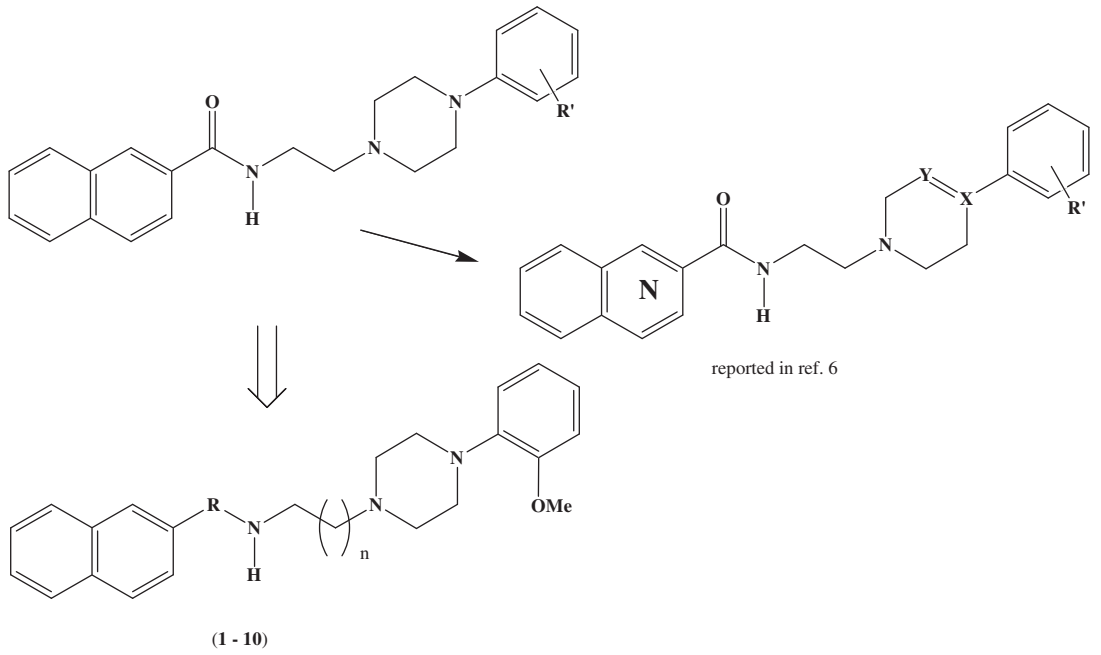
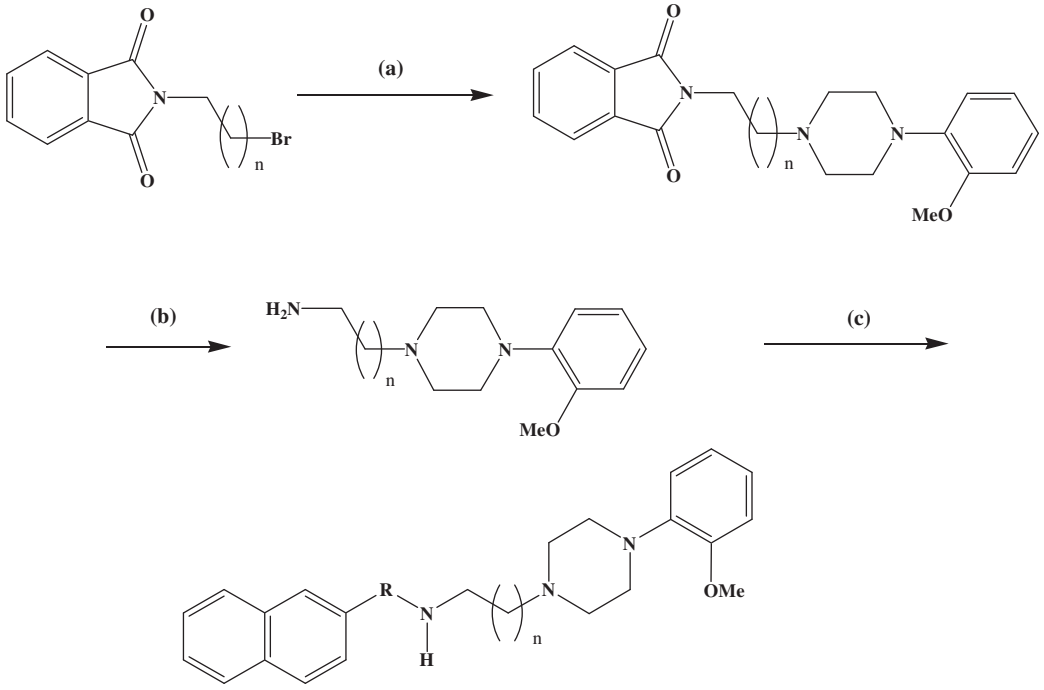


Figure 1. Chemical modulations in a series of piperazine-alkyl-naphthamide derivatives (**R** = CO or SO_2; **R′** = H, 2-Cl, 3-Cl, 4-Cl, 2-F, 3-F, 4-F, 2-Me, 3-Me, 4-Me, 2-OMe, 3-OMe, 4-OMe, 3-CF_3; *n* = 1–5; **Y**=**X**: CH_2–N or CH=C).



Scheme 1. Reagents and conditions: (a) 1-(2-methoxy-phenyl) piperazine, K_2CO_3, reflux; (b) NH_2NH_2, EtOH, reflux; (c) 2-naphthoyl chloride or 2-naphthalenesulfonyl chloride, Et_3N, EtOAc, rt; *n* = 1–5; R = –CO– or –SO_2–.

nature of the amide group. The high 5-HT_{1A} affinity of the sulphonamide analogue **4** with a propyl linker was also previously reported in the literature in the development of a series of C3 congeners with serotonergic properties.[7] In terms of bioisosterism, carboxamide derivatives are generally more potent on 5-HT_{1A} receptors, except the hexyl sulphonamide analogue **10** which appears to display a higher affinity for 5-HT_{1A} receptors than its carboxamide analogue **9**. Regarding the impact of the linker on the affinity for 5-HT_{1A} receptors it is observed an uneven evolution.

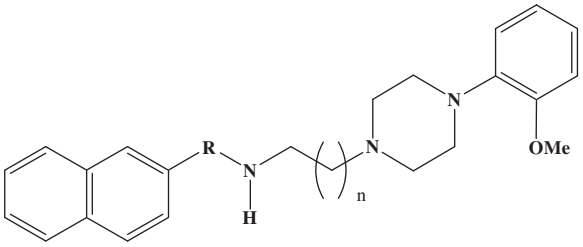
Most of the compounds, except the butyl sulphonamide **6,** presented a lower affinity for D2L receptors when compared with other receptors sites tested. The affinity tended to increase with an increase in linker size. A small linker in this series led to molecules which did not display significant affinity for the D2L receptor. This was similar to the affinity previously observed with piperidine naphthamide derivatives.[8,9]

Carboxamide and sulphonamide alkyl(ethyl)piperazine analogues displayed a high binding affinity for D4.2 receptors, in par-

Table 1
Experimental conditions of in vitro binding procedures

Cloned receptors	Ligand	Non-specific	Incubation buffer	Incubation time and temperature	Filtration and counting
h-5-HT_{1A} (Perkin Elmer 6110501)	[^{3}H]-8-OH-DPAT (0.25 nM)	WB4101 (10 μM)	50 mM Tris–HCl buffer at pH 7.4 + 10 mM $MgSO_4$, 0.5 mM EDTA and 0.1% ascorbic acid	60 min at 27 °C	GF/C 0.3% PEI; Ecoscint A; TRI-CARB 1600TR
h-α_{2A} (Perkin Elmer 6110113)	[^{3}H]-MK912 (0.7 nM)	Metergoline (10 μM)	50 mM Tris–HCl buffer at pH 7.4 + 12.5 mM $MgCl_2$, and 2 mM EDTA	60 min at 27 °C	GF/C 0.3% PEI; Ecoscint A; TRI-CARB 1600TR
h-D4.2 (Sigma D2439)	[^{3}H]-YM-09151-2 (0.2 nM)	Clozapine (10 μM)	50 mM Tris–HCl buffer at pH 7.4 + 5 mM $MgCl_2$, 5 mM KCl, 1.5 mM $CaCl_2$, and 5 mM EDTA	60 min at 27 °C	GF/C 0.3% PEI; Ecoscint A; TRI-CARB 1600TR
r-D3 (Perkin Elmer 6110139)	[^{3}H]-Spiperone (0.5 nM)	Haloperidol (10 μM)	50 mM Tris–HCl buffer at pH 7.4 + 10 mM $MgCl_2$, 1 mM EDTA and 120 mM NaCl	60 min at 27 °C	GF/C 0.3% PEI; Ecoscint A; TRI-CARB 1600TR
h-D2L (RBI D-180)	[^{3}H]-Spiperone (0.2 nM)	Haloperidol (10 μM)	50 mM Tris–HCl buffer at pH 7.4 + 10 mM $MgCl_2$ and 1 mM EDTA	60 min at 27 °C	GF/C 0.3% PEI; Ecoscint A; TRI-CARB 1600TR

Table 2
Affinity of piperazine-alkyl-naphthamide derivatives for human cloned D4.2, 5-HT_{1A}, D3, α_{2A}, and D2L receptors



Compound	R	*n*	D4.2	5-HT_{1A}	D3	α_{2A}	D2L
1	CO	1	2.32 ± 0.66	1.69 ± 0.18	66 ± 4	113 ± 20	890 ± 122
2	SO_2	1	24.4 ± 1.2	5.19 ± 0.49	56 ± 9.9	348 ± 5	44*
3	CO	2	17.8 ± 0.1	4.59 ± 1.07	175 ± 109	71 ± 7.4	46*
4	SO_2	2	112 ± 25	4.64 ± 1.49	58.5 ± 5.0	101 ± 1	132 ± 11
5	CO	3	174 ± 25	2.75 ± 0.25	0.77 ± 0.11	20.2 ± 11.6	30*
6	SO_2	3	62 ± 14	4.1 ± 0.73	27.5 ± 2.1	79 ± 0.3	46.5 ± 6.4
7	CO	4	54 ± 10	11.1 ± 2.5	13 ± 5.7	9.36 ± 4.73	254 ± 35
8	SO_2	4	27.4 ± 2.7	8.53 ± 2.28	12.5 ± 6.4	84 ± 12	438 ± 79
9	CO	5	102 ± 29	6.15 ± 1.92	0.56 ± 0.19	96 ± 7.4	121 ± 6.4
10	SO_2	5	9.62 ± 2.63	2.87 ± 0.52	1.97 ± 0.15	45 ± 15.5	99 ± 17

* K_i (in nM, mean ± SD; $n \geqslant 2$ if unspecified) or percentage of inhibition at 1 μM.

allel to the differing affinity for 5-HT_{1A} receptors. This is most notable for the ethyl carboxamide **1** and hexyl sulphonamide **10** analogues. Carboxamide analogues displayed higher affinity for D4.2 receptors than sulphonamide analogues, when the linker was small (ethyl and propyl). However, the opposite was true for longer chains (butyl to hexyl).

In contrast, the affinity for α_{2A} receptors was enhanced when the size of the linker increased. In a series of molecules with an ethyl linker that we recently reported in the literature, the affinity for α_{2A} receptors was effectively weak.[6] The present compounds are unlike the α_{2A}-adrenoceptor ligands recently reported in the literature, because they lack a guanidine or 2-aminoimidazoline moiety.[10] Nevertheless, a number of piperazine-alkyl-naphthamides are shown to possess a significant affinity for the α_{2A} receptor. Compound **7**, with a pentyl moiety, had the highest affinity for this receptor site. Further increase of the size of the linker is unfavourable.

Compound **5** is the D3 receptor partial agonist BP897.[11,12] Therefore, we examined the interaction of compounds **1–10** with D3 receptors. We observed a high affinity for compound **5** at D3 receptors as previously reported, but unlike previous reports[13] compound **5** also presented a similar binding affinity to 5-HT_{1A} receptors. Studies on molecules related to BP897 (compound **5**) have indicated that compounds with an extended and more linear conformation in the aliphatic or aryl spacer are more selective for D3 over D2 receptors.[14] Complete classification of the affinities of these compounds at other receptors has yet to be determined, and the length of the spacer in these studies has been limited to propyl or butyl.[13,14] Similarly, we observed that compounds with a small linker (ethyl and propyl) displayed reduced interaction with D3 receptors, when compared with compounds possessing a longer linker (butyl to hexyl). Moreover, the hexyl analogues **9** and **10** also presented a high affinity for this receptor. Although the difference is low the carboxamide **9** has more affinity than the carboxamide **5**.

In this study we observe that the impact of homology within the linker is different for the examined receptors. A parabolic curve is not observed for chain length against affinity for these receptors

but this relationship is not always found among homologous series.[15]

Although bioisosteric replacement of the carboxamide by a sulphonamide strongly modified the spatial orientation of the side chain as we previously reported,[16] the impact of such modification is differentially observed. In the present work, the presence of this sulphonamide moiety was favourable for some interactions. The success of this chemical modification was varied in the literature. The hypoglycaemic sulfonyl isostere of glybenclamide was found to be more potent,[17] however sulfonyl isosteres of orthopramides did not interact with D2 and 5-HT_2 receptors and lost the prokinetic activity of the reference compounds.[16] The sulphonamide moiety is also frequently found in molecules interacting with a high affinity for 5-HT_7 receptors but usually these molecules also possess a higher 5-HT_{1A} receptor affinity.[7,18–20]

In conclusion, the systematic nature of the present study gives an interesting overview of the impact of these two chemical modifications of piperazine-alkyl-naphthamides on the interactions with a number of receptors implicated in psychiatric disorders. It is clear that further investigation of the quantitative structure–activity relationship (QSAR) of these compounds is required, as all biological data has been reported by one group[6,8,9] this would provide an improved binding profile of such simple molecules in the context of developing pharmacological tools. The biological evaluation will be extended in two directions. Firstly, the determination of their binding affinity for receptors such as 5-HT_{2A} and 5-HT_7 that have been frequently implicated in CNS disorders should be done. Secondly, since bioisosterism or homologation might have important effects on functional behaviours, testing of these compounds in functional assays either in vitro or in electrophysiological bioassays should be informative. In addition, further study of different molecules like pentyl or hexyl analogues with a multi-receptor binding profile should be carried out using behavioural models to detect potential antipsychotic effects. The major drawback detected during in vitro binding investigations is the impact of lipophilicity in terms of solubility for long linker compounds. The preparation of compounds will need an appropriate vehicle, so further chemical developments should be taken into account to limit problems associated with excessive lipophilicity, and reduce possible excessive fat storage when chronically administered. Finally, such data confirmed that several current pharmacological tools are not as selective as previously believed and further research into more selective ligands is required to explore and clarify the physiology of different neuronal systems.

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References and notes

1. Roth, B. L.; Sheffler, D. L.; Kroeze, W. K. *Nat. Rev. Drug Disc.* **2004**, *3*, 353.
2. Marino, M. J.; Knutsen, L. J. S.; Williams, M. *J. Med. Chem.* **2008**, *51*, 1077.
3. Ichikawa, J.; Ishii, I.; Bonaccorso, S.; Fowler, W. L.; O'Laughlin, I. A.; Meltzer, H. Y. *J. Neurochem.* **2001**, *76*, 1521.
4. Hertel, P.; Fagerquist, M. V.; Svensson, T. H. *Science* **1999**, *286*, 105.
5. Chemel, B. R.; Roth, B. L.; Armbruster, B.; Watts, V. J.; Nichols, D. E. *Psychopharmacology (Berlin)* **2006**, *188*, 244.
6. Graulich, A.; Léonard, M.; Résimont, M.; Huang, X.-P.; Roth, B. L.; Liégeois, J.-F. *Aust. J. Chem.* **2010**, *63*, 56.
7. Yoon, J.; Yoo, E.; Kim, J.-Y.; Pae, A. N.; Rhim, H.; Park, W.-K.; Kong, J. Y.; Park Choo, H.-Y. *Bioorg. Med. Chem.* **2008**, *16*, 5405.
8. Carato, P.; Graulich, A.; Jensen, N.; Roth, B. L.; Liégeois, J.-F. *Bioorg. Med. Chem. Lett.* **2007**, *17*, 1565.
9. Carato, P.; Graulich, A.; Jensen, N.; Roth, B. L.; Liégeois, J.-F. *Bioorg. Med. Chem. Lett.* **2007**, *17*, 1570.
10. Rodriguez, F.; Rozas, I.; Ortega, J. E.; Erdozain, A. M.; Meana, J. J.; Calldo, L. F. *J. Med. Chem.* **2009**, *52*, 601.
11. Pilla, M.; Perachon, S.; Sautel, F.; Garrido, F.; Mann, A.; Wermuth, C. G.; Schwartz, J.-C.; Everitt, B. J.; Sokoloff, P. *Nature* **1999**, *400*, 371.
12. Wicke, K.; Garcia-Ladona, J. *Eur. J. Pharmacol.* **2001**, *424*, 85.
13. Campiani, G.; Butini, S.; Trotta, F.; Fattorusso, C.; Catalanotti, B.; Aiello, F.; Gemma, S.; Nacci, V.; Novellino, E.; Ann Stark, J.; Cagnotto, A.; Fumagalli, E.; Carnovali, F.; Cervo, L.; Mennini, T. *J. Med. Chem.* **2003**, *46*, 3822.
14. Hackling, A.; Ghosh, R.; Perachon, S.; Mann, A.; Höltje, H.-D.; Wermuth, C. G.; Schwartz, J.-C.; Sippl, W.; Sokoloff, P.; Stark, H. *J. Med. Chem.* **2003**, *46*, 3883.
15. Wermuth, C. G. The Practice of Medicinal Chemistry. In Wermuth, C. G., Ed.; Academic Press, 2000; p 203.
16. Liégeois, J.-F.; Dive, G.; Dupont, L.; Delarge, J. *Helv. Chim. Acta* **1991**, *74*, 1764.
17. Fournier, J.-P.; Moreau, R. C.; Narcisse, G.; Choay, P. *Eur. J. Med. Chem.* **1982**, *17*, 81.
18. Bojarski, A. J.; Duszyńska, B.; Kolaczkowski, M.; Kowalski, P.; Kowalska, T. *Bioorg. Med. Chem. Lett.* **2004**, *23*, 5863.
19. Medina, R. A.; Sallander, J.; Benhamu, B.; Porras, E.; Campillo, M.; Pardo, L.; Lopez-Rodriguez, M. L. *J. Med. Chem.* **2009**, *52*, 2384.
20. Yoo, E.; Yoon, J.; Pae, A. N.; Rhim, H.; Park, W.-K.; Kong, J. Y.; Park Choo, H.-Y. *Bioorg. Med. Chem.* **2010**, *18*, 1665.