Application of the Bivalent Ligand Approach to the Design of Novel Dimeric Serotonin Reuptake Inhibitors

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Chemical modulation of the monoamine neurotransmitter systems involving dopamine (DA), serotonin (5-HT), and norepinephrine (NE) provides an important means to control certain neurological disorders such as depression,[1] anxiety,[2] alcoholism,[3] chronic pain,[4] eating disorders,[5] and obsessive compulsive disorders.[6] For example, a major pharmaceutical approach to the treatment of depression has come about through the development of agents that interfere with the primary mechanism of removal of 5-HT or NE from the synapse.[7] As the result, the selective 5-HT reuptake inhibitors (SSRIs) such as fluoxetine (Prozac)[8] and paroxetine (Paxil),[9] among others, have been developed for the treatment of depression and related psychological disorders. Despite these recent clinical developments, a detailed understanding of the structural factors that govern the potency and selectivity of ligands at the specific monoamine transporters is still evolving. During our efforts to discover ligands of possible use as medications,[10] we discovered a rather interesting aspect of the 5-HT transporter (SERT) structure activity relationships (SAR), namely that significant selectivity and potency can be achieved through the use of a bivalent ligand approach (Figure 1).

The rationale for employing the bivalent ligand approach stems from the possibility that dimeric structures may be capable of bridging independent recognition sites on the transporters resulting in a thermodynamically more favorable binding interaction than the monovalent binding of two molecules.[11] As such proximal binding sites are likely to differ in their location for the three monoaminergic transporters the length of the linker connecting the two binding moieties could thus provide a means to fine-tune transporter selectivity profiles. Empirical support for such a possibility stems from the enhanced potency and selectivity reported for both bivalent narcotic antagonists containing the naltrexamine pharmacophore[12] and for serotonin-based bivalent 5-$HT_{1B/1D}$ agonists.[13]

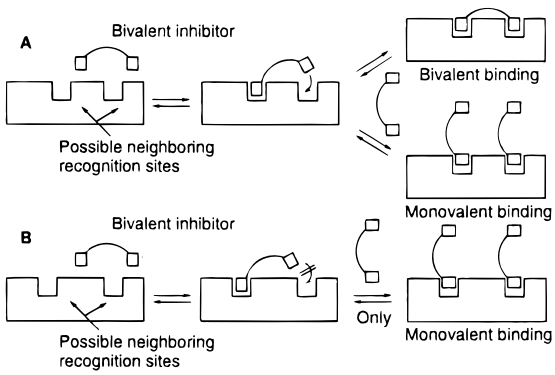

Figure 1. (A) Model for the binding of a bivalent ligand of appropriate chain length to neighboring recognition sites. Given the proper linker geometry, the bivalent binding is favored. (B) Model binding of bivalent neighboring recognition sites with a bivalent ligand with an inadequate chain length. Only univalent binding is possible.[12b]

We have recently reported on the chemistry and pharmacology of some 3,4-disubstituted piperidine-based ligands that show reasonable potency at the dopamine transporter (DAT).[14] On the basis of these and other studies[15] it had become apparent to us that 3,4-disubstituted piperidines are structurally related to drugs such as femoxetine and paroxetine that exhibit high potency and selectivity for the SERT.[16] One of these piperidines was therefore chosen as the starting monomer for the assembly of bivalent ligands that were postulated to exhibit potent and selective SERT activity.

Briefly, piperidine-based ligands **4**−**16** were prepared in optically pure form from arecoline (**1**)[14] (Scheme 1) and were evaluated for their ability to inhibit high affinity uptake of DA, 5-HT, and NE using rat synaptosomal nerve endings.[17] The uptake data expressed as K_i values and the selectivity profile (ratio of K_i values) for these compounds are provided in Table 1. In general, all dimers (**7**−**16**) exhibit substantially higher potency at the SERT than their monomeric counterparts. As is apparent from Table 1, the SERT and the DAT potency increase of the bivalent ligand **11** over the monovalent ligand **3** is greater than a factor of 2. However, the NET activity of compound **11** is slightly decreased, as the bivalent inhibitor may undergo univalent binding at high concentrations (Figure 1B). Therefore, the presence of a five-methylene spacer in the linking chain of **11** appears to favor bivalent binding at the SERT and the DAT (Figure 1, model A). Specifically, the potency of piperidine **11**, a bivalent analogue of the parent piperidine **3**, has improved by a factor of greater than

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(1) Broekkamp, C. L. E.; Leysen, D.; Peeters, B. W. M. M.; Pinder, R. M. *J. Med. Chem.* **1995**, *38*, 4615−4633.
(2) Frances, A.; Manning, D.; Marin, D.; Kocsis, J.; McKinney, K.; Hall, W.; Klein, M. *Psychopharamacol. Suppl.* **1992**, *106*, S82−S86.
(3) Kranzler, H. R.; Amine, H.; Modesto-Lowe, V.; Oncken, C. *Pharmacol. Treat. Drug Alcohol Depend.* **1999**, *22*, 401−423. Schaffer, A.; Naranjo, C. A. *Drugs* **1998**, *56*, 571−585.
(4) Sullivan, M. J.; Reesor, K.; Mikail, S.; Fisher, R. *Pain* **1993**, *52*, 294.
(5) (a) Peterson, C. B.; Mitchell, J. E. *J. Clin. Psychiatry* **1999**, *55*, 685−697. (b) Kaye, W. H. *Psychopharmacol. Bull.* **1997**, *33*, 335−344.
(6) Brody, A. L.; Saxena, S.; Schwartz, J. M.; Stoessel, P. W.; Maidment, K.; Phelps, M. E.; Baxter, L. R., Jr. *Psychiatr. Res.* **1998**, *84*, 1−6.
(7) (a) Briner, K.; Dodel, R. C. *Curr. Pharm. Design* **1998**, *4*, 291−302. (b) Owens, M. J.; Morgan, N.; Plott, S. J.; Nemeroff, C. B. *J. Pharmacol. Exp. Ther.* **1997**, *283*, 1305−1322. (c) Mongeau, R.; Blier, P.; de Montigny, C. *Brain Res. Rev.* **1997**, *23*, 145−195.
(8) (a) Wong, D. T.; Bymaster, F. P.; Engleman, E. A. *Life Sci.* **1995**, *57*, 411-441. (b) Robertson, D. W.; Krushinski, J. H.; Fuller, R. W.; Leander, J. D. *J. Med. Chem.* **1988**, *31*, 1412−1417.
(9) Dechant, K. L.; Clissold, S. P. *Drugs* **1991**, *41*, 225−253.
(10) Smith, M. P.; Hoepping, A.; Johnson, K. M.; Trzcinska, M.; Kozikowski, A. P. *DDT* **1999**, *4*, 322−332.
(11) (a) Shuker, S. B.; Hajduk, P. J.; Meadows, R. P.; Fesik, S. W. *Science* **1996**, *274*, 1531−1534. (b) Jencks, W. P. *Proc. Natl. Acad. Sci. U.S.A.* **1981**, *78*, 4046-4050. (c) Mammen, M.; Choi, S. -K.; Whitesides, G. M. *Angew. Chem. Int. Ed.* **1998**, *37*, 2754-2794.
(12) (a) Erez, M.; Takemori, A. I.; Portoghese, P. S. *J. Med. Chem.* **1982**, *25*, 847−849. (b) Portoghese, P. S. *J. Med. Chem.* **1992**, *35*, 1927−1937.
(13) (a) Halazy, S.; Perez, M.; Fourrier, C.; Pallard, I.; Pauwels, P. J.; Palmier, C.; Gareth, W. J.; Valentine, J.-P.; Bonnafous, R.; Martinez, J. *J. Med. Chem.* **1996**, *39*, 4920−4927. (b) LeBoulluec, K. L.; Mattson, R. J.; Mahle, C. D.; McGovern, R. T.; Nowak, H. P.; Gentile, A. J. *Bioorg. Med. Chem. Lett.* **1995**, *5*, 123−126. (c) Halazy, S.; Perez, M.; Fourrier, C.; Pallard, I.; Pauwels, P. J.; Palmier, C.; John, G. W.; Valentin, J-. P.; Bonnafous, R.; Martinez, J. *J. Med. Chem.* **1996**, *39*, 4920−4927.
(14) (a) Kozikowski, A. P.; Araldi, G. L.; Boja, J.; Meil, W. M.; Johnson, K. M.; Flippen-Anderson, J. L.; George, C.; Saiah, E. *J. Med. Chem.* **1998**, *41*, 1962−1969. (b) Tamiz, A. P.; Zhang, J.; Fliuppen-Anderson, J. L.; Zhang, M.; Johnson, K. M.; Deschaux, O.; Tella, S.; Kozikowski, A. P. *J. Med. Chem.* **2000**, *43*, 1215−1222.
(15) Smith, M. P.; Johnson, K. M.; Zhang, M.; Flippen-Anderson, J. L.; Kozikowski, A. P. *J. Am. Chem. Soc.* **1998**, *120*, 9072−9073.
(16) Engelstoft, M.; Hansen, J. B. *Acta Chem. Scand.* **1996**, *50*, 164−169.
(17) Wang, S.; Sakamuri, S.; Enyedy, I.; Kozikowski, A. P.; Deschaux, O.; Bandyopadhyay, B. C.; Tella, S. R.; Zaman, W. A.; Johnson, K. M. *J. Med. Chem.* **2000**, *43*, 351−360.

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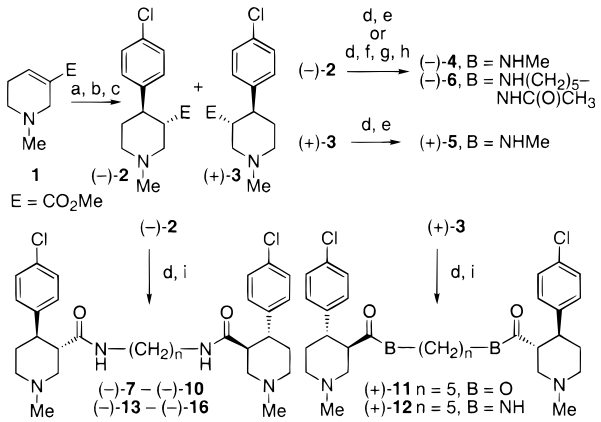
Table 1. Activity of Bivalent Inhibitors at the Monoamine Transporters, $K_i \pm$ SE (nM)

compd	B	spacer	isomer	[^{3}H]DA[b] uptake K_i (nM)	[^{3}H]NE[b] uptake K_i (nM)	[^{3}H]5−HT[b] uptake K_i (nM)	selectivity DAT/SERT	selectivity NET/SERT
2	OMe	−	(−)-*trans*	2890 ± 250	242 ± 3.0	3600 ± 410	0.80	0.067
3	OMe	−	(+)-*trans*	228 ± 30	90 ± 5.2	5880 ± 440	0.039	0.015
4	NHMe	−	(−)-*trans*	>70 000	3110 ± 530	>10 000	>7	0.3
5	NHMe	−	(+)-*trans*	>9000	4380 ± 1100	>53 000	>0.16	>0.083
6	*c*	−	(−)-*trans*	>70 000	>12 000	1340 ± 110	>52	>9.0
7	NH	$-(CH_2)_2-$	(−)-*trans*	1340 ± 190	473 ±138	258 ± 42	5.2	1.8
8	NH	$-(CH_2)_3-$	(−)-*trans*	5090 ± 90	373 ± 55	342 ± 6.0	15	1.1
9	NH	$-(CH_2)_4-$	(−)-*trans*	1510 ± 20	247 ± 1.2	14 ± 1.9	108	18
10	NH	$-(CH_2)_5-$	(−)-*trans*	1960 ± 200	393 ± 6.7	1.2 ± 0.1	1633	328
11	O	$-(CH_2)_5-$	(+)-*trans*	56 ± 4.7	182 ± 8.0	25 ± 5.4	2.2	7.3
12	NH	$-(CH_2)_5-$	(+)-*trans*	39 ± 4.3	158 ± 15	7.0 ± 0.6	5.6	23
13	NH	$-(CH_2)_6-$	(−)-*trans*	3030 ± 540	850 ± 45	11 ± 1.6	275	77
14	NH	$-(CH_2)_7-$	(−)-*trans*	1390 ± 20	727 ± 18	3.9 ± 0.2	356	186
15	NH	$-(CH_2)_8-$	(−)-*trans*	3184 ± 213	1037 ± 62	2.1 ± 0.1	1516	493
16	NH	$-(CH_2)_9-$	(−)-*trans*	1510 ± 15	1089 ± 276	28.9 ± 4.3	52	38

[a] All compounds exhibit spectral data in agreement with the assigned structures (see Supporting Information), and gave satisfactory C, H, N analyses. [b] Data are mean ± standard error of at least three experiments as described in ref 17. [c] Compound **6** is a monomer with B = NH−$(CH_2)_5$−C(O)NHCH$_3$.

Scheme 1[a]



[a] (a) 4-ClPhMgBr, ether, −10 °C; (b) dibenzoyl-D-tartaric acid, MeOH or dibenzoyl-L-tartaric acid, MeOH; (c) NaOMe, MeOH; (d) HCl (6 N), reflux; $(COCl)_2$, CH_2Cl_2; (e) methylamine, TEA, CH_2Cl_2 (f) FMOC−NH−$(CH_2)_5$−NH_2·HCl, TEA, CH_2Cl_2; (g) DMF, TEA, 12 h, then acetic anhydride; (h) HCl/ether (1.0 M); (i) diamine or diol, TEA, CH_2Cl_2.

4-fold at the DAT and 230-fold at the SERT, while the NET potency is decreased by a factor of 2. Replacement of the ester groups in the linking chain of **11** with amide groups gave **12** and resulted in another 3-fold increase of potency at the SERT with a DAT K_i of 39 nM. Interestingly, **10**, the enantiomer of piperidine **12** is yet 6-fold more potent at the SERT and has a K_i of 1.2 nM. Piperidine **10** no longer exhibits high potency at the DAT while the NET activity is reduced 2-fold. Removal of one of the piperidine moieties from **10** gave **6** which is >1000-fold less potent at the SERT. Piperidine **10** has selectivity for the SERT over the DAT and the NET of 1633 and 328, respectively. We have thus succeeded in using the length of the linking chain (Figure 2), and the absolute stereochemistry of the piperidine unit to control monoamine transporter selectivity.

In conclusion, The present results support the possible existence of dual binding sites at the SERT, where an enhanced potency of greater than 8000-fold is achieved by bridging two neighboring recognition sites via a linking chain of five methylene units. While the proximal sites for binding may be located within a single entity of the SERT, the possibility exists that the SERT is oligomeric[18] and that the piperidine dimer may bridge sites located on two different SERT units. Distinguishing between these possibilities is difficult. Hill plot analysis of the inhibition data revealed that

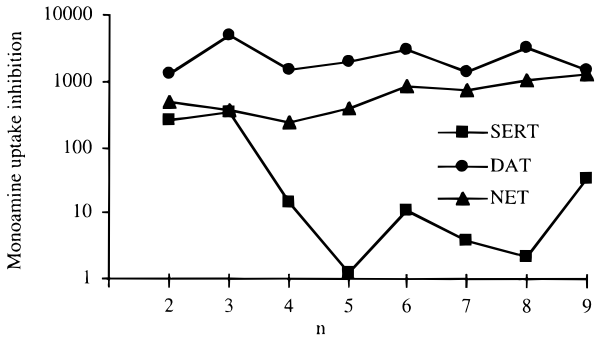


Figure 2. Monoamine reuptake inhibitory activity (K_i, nM) of the (−)-ligands as a function of the number of methylene units (n) in the linking chain.

the Hill coefficients for the dimers were not substantially different from unity. For example, for **10**, $n = 0.97 \pm 0.11$. This suggests that the two postulated binding sites do not interact in either a positive or negative manner in binding the dimer headgroups. It further suggests that these binding sites must behave independently and that they have similar affinity for the dimer headgroups. Another possibility is that one end of the dimer binds to its recognition site in a manner that is supported by nonspecific binding of the other end of the dimer to a hydrophobic region located either in the surrounding membrane or in the SERT protein itself. However, preliminary data with piperidine (−)-**6** suggest that the binding of both ends of the dimer have specific structural requirements. Final resolution of these issues awaits further SAR studies involving nonsymmetrical analogues. Nevertheless, the SAR developed for these bivalent piperidines indicates that this series of molecules may readily be tailored to have dual activity at the DAT/SERT or at the SERT exclusively. The present results have important implications for the design of a new generation of SSRIs of possible use in the treatment of a wide range of neurological disorders.

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Supporting Information Available: Experimental details and 300 MHz ^{1}H NMR spectra for all the compounds described in Scheme 1 (PDF). This material is available free of charge via the Internet at http://pubs.acs.org.

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(18) Kilic, F.; Rudnick, G. *Proc. Natl. Acad. Sci. U.S.A.* **2000**, in press.