Receptors & Clinical Investigation 2014; 1: e106. doi: 10.14800/rci.106; © 2014 by Parthasarathi Rajagopalan, *et al.*
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BRIEF REPORT

Dual, Postsynaptic 5-HT$_{2B}$ Antagonist and 5-HT$_{1A}$ Agonist Approach to the Treatment of METH/MDMA Addiction and Related Behavioral Disorders. Part 2. Proof of Concept.

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Received: March 4, 2014
Published online: May 6, 2014

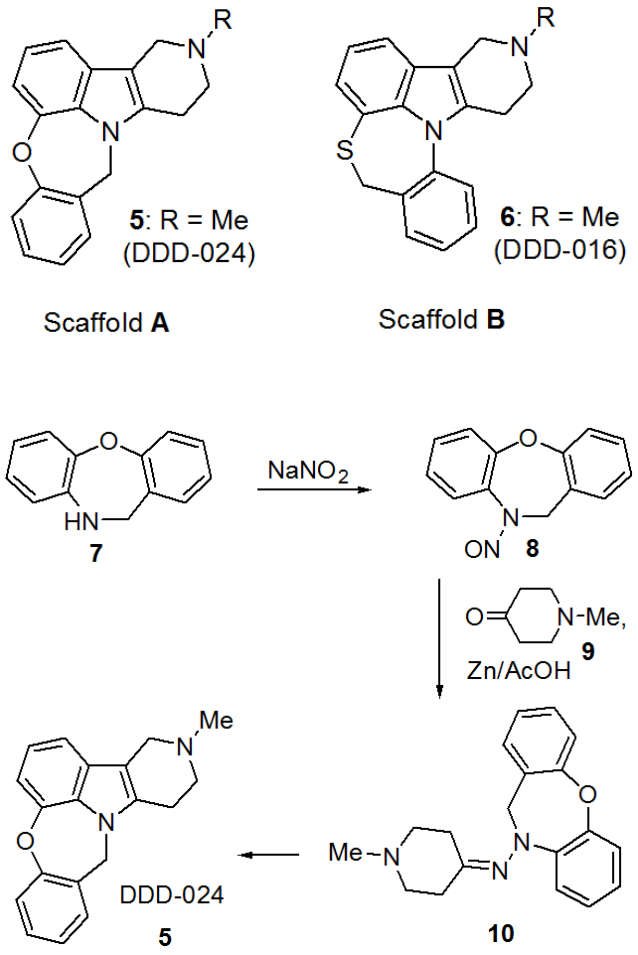
The synthesis, *in vitro*, and preliminary *in vivo* pharmacology of DDD-024, a novel, pentacyclic compound, is described. It exhibits high affinity to the 5-HT$_{1A}$ and good affinity to the 5-HT$_{2B}$ receptors and is an agonist of the former and antagonist of the latter receptors, a profile which meets with the requirements delineated in our hypothesis advanced in Part 1 under the section 'Hypothesis' in this issue. Further, *in vivo*, it was quite active in METH-seeking behavior test in rats, MDMA-induced hyperlocomotion test in mice, and Porsolt's forced swim test in rats for antidepressant activity thereby providing convincing proof of concept for our hypothesis.

Keywords: DD-024; 5-HT$_{1A}$; 5-HT$_{2B}$

To cite this article: Parthasarathi Rajagopalan, et al. Postsynaptic 5-HT$_{2B}$ Antagonist and 5-HT$_{1A}$ Agonist Approach to the Treatment of METH/MDMA Addiction and Related Problems. Part 2. Proof of Concept. Receptor Clin Invest 2014; 1: e106. doi: 10.14800/rci.106.

In our continuing effort directed toward the discovery of small molecules for the treatment of CNS disorders, we had recently synthesized DDD-024 (**5**), a prototype of a new pentacyclic scaffold **A**. Surprisingly, DDD-024 displayed receptor binding properties remarkably different from those of our previously reported potential pentacyclic antipsychotic DDD-016 (**6**), a prototype of scaffold **B**[1] isomeric to A, indicating that the position of the phenyl ring in the seven-membered heterocyclic ring, besides the hetero atoms, in these scaffolds seem to strongly influence the binding profiles of **5** and **6**.

The synthesis of **5** starting with the known dibenzo[*b*,*e*]1,4-oxazepine (**7**)[2] is outlined in Scheme 1. The nitroso derivative **8** of **7** was reduced with zinc and acetic acid in the presence of 1-methyl-4-piperidone (**9**) to yield the hydrazone (**10**) which underwent Fischer indole cyclization *in situ* to furnish **5**. The overall yield of the product was low because of the competing denitrosation of **8** to the parent amine **7**. However, the redeeming feature here is that the recovered **7** can be recycled at least a couple of times to obtain more of **5**. DDD-024 (**5**) was evaluated in a high-throughput screening (HTS) panel consisting of over 40 transmembrane receptors, ion



Scheme 1. Synthesis of DDD-024.

channels, and monamine transporters[3]. This panel was chosen to determine the binding affinities of DDD-024 not only of the target 5-HT_{2B} and HT_{1A} receptors but also of the off-target receptors and transporters as well to determine its selectivity and specificity profile. The K_i's of the binding affinities of DDD-024 are listed in Table 1.

Results and Discussion

DDD-024 displays high affinity to 5-HT_{1A} and good affinity to 5-HT_{2B}, the two target receptors, moderate affinities to 5-HT_{1D}, 5-HT_{2C}, 5-HT_7, σ_2, and lower affinities to 5-HT_{1B}, 5-HT_{2A}, and 5-HT_6 receptors. It should be noted that none of the receptors mentioned above other than the 5-HT_{1A} and 5-HT_{2B} subtypes plays any significant role in the mechanisms of action of METH or MDMA. DDD-024 does not bind to any of the dopamine receptors which is a very important factor in the development of a medication for METH/MDMA addiction because such an agent would be free of undesirable dopaminergic side effects. DDD-024 exhibits no affinity to the dopamine transporter (DAT) or to the serotonin transporter (SERT) and only moderate affinity to the norepinephrine transporter (NET). Most importantly, in the functional assay DDD-024 displayed **agonist** activity at the 5-HT_{1A} and **antagonist** activity at the 5-HT_{2B} receptors (see Table 2). It is pertinent to mention here that because DDD-024 is a 5-HT_{2B} antagonist, any medication derived from it would be free of the potential cardiotoxicity associated with 5-HT_{2B} agonists.

DDD-024 was also subjected to the following three key preliminary in vivo tests: METH-seeking behaviour in rats, MDMA-induced hyperlocomotion in mice, and antidepressant activity (Porsolt's forced swim test)[4,5] in rats to evaluate its usefulness in the treatment of METH/MDMA addictions and associated behavioural problems. DDD-024 was highly active at 10 mg/kg i.p. in blocking meth seeking paradigm in rats trained to self-administer METH; it completely blocked the effect of MDMA-induced hyperlocomotion in mice at 20 mg/kg i.p.; and it was as active as imipramine in the antidepressant (Porsolt) test in rats (both compounds tested at 10 mg/kg p.o.). The first two crucial results can only be due to its postsynaptic 5-HT_{2B} antagonist activity and the third only to its potent postsynaptic 5-HT_{1A} agonist activity. These three paradigms, therefore, provide convincing **proof of concept** for our hypothesis. Of the few CNS tests in which the 5-HT_{1A} agonists are active, we chose to evaluate DDD-024 in the antidepressant test to begin with because depression is a very common and chronic behavioural problem among the METH/MDMA addicts. Since DDD-024 does not bind to SERT or DAT, the in vivo anti-METH/MDMA and antidepressant effects elicited by it can only be explained on the basis of its **direct post-synaptic activity** at the relevant regions of the brain as discussed earlier. This is particularly important in so far as the 5-HT_{1A} receptors are concerned because presynaptically they have the opposite effect of facilitating behavioural problems resulting from METH/MDMA abuse.

In conclusion, we are gratified to report that DDD-024 with its unique pentacyclic structure has not only provided the proof of concept for our hypothesis but that it is also an exciting lead for the development of a much needed medication for the treatment METH/MDMA addition and related behavioural disorders.

Acknowledgements

We thank Prof. Luc Maroteaux (University of Pierre and Marie Curie, Paris, France) for the MDMA-induced

Table 1. Receptor Binding Data (Ki ± SEM) nM of DDD-024 (5)

Receptor	K_i	Receptor	K_i
5-HT_{1A}	32 ± 4	α_{1D}	5033 ± 755
5-HT_{1B}	738 ± 110	α_{2A}	1459 ± 219
5-HT_{1D}	385 ± 44	α_{2B}	> 10^4
5-HT_{1E}	> 10^4	α_{2C}	507
5-HT_{2A}	927 ± 143	β_1	> 10^4
5-HT_{2B}	195 ± 29	β_2	> 10^4
5-HT_{2C}	248 ± 23	β_3	> 10^4
5-HT_3	> 10^4	δ	> 10^4
5-HT_4	> 10^4	κ	> 10^4
5-HT_{5A}	1905 ± 286	μ	> 10^4
5-HT_6	589 ± 88	σ_1	1815 ± 272
5-HT_7	258 ± 40	σ_2	310 ± 50
D_1	> 10^4	M_1	> 10^4
D_2	> 10^4	M_2	> 10^4
D_3	> 10^4	M_3	> 10^4
D_4	> 10^4	M_4	> 10^4
D_5	> 10^4	M_5	> 10^4
H_1	> 10^4	GABA A	> 10^4
H_2	> 10^4	NMDA	> 10^4
H_3	> 10^4	DAT	> 10^4
H_4	> 10^4	NET	650 ± 97
α_{1A}	5863 ± 879	SERT	> 10^4
α_{1B}	> 10^4		

Table 2. Functional Assay of DDD-024.

	% Inhibition of Control (Agonist Activity)[a]	% Inhibition of Control (Antagonist Activity)[b]
5-HT_{1A}	89	-23
5-HT_{2B}	25	89

[a]Agonist control: Serotonin and 5-OH-DPAT.
[b]Antagonist control: WAY100635 and SB306553.

hyperlocomotion test, Prof. Ronald E. See (University of South Carolina, Columbia, SC) for the meth seeking behaviour test. We also thank National Institute of Mental Health (NIMH) Psychoactive Drug Screening Program (PDSP) for the receptor binding studies.

Conflict of Interest

The authors declare that there is no conflict of interest.

References

1. Rawls SM, Shah A, Ayoub G, Raffa RB. 5-HT1A-like receptor activation inhibits abstinence-induced methamphetamine withdrawal in planarians. Neurosci Lett 2010; 484: 113-117.
2. Margolis BJ, Swidorski JJ, Rogers BN. An efficient assembly of heterobenzazepine ring system utilizing an intramolecular palladium-catalyzed cycloamination. J Org Chem 2003; 68:644-647.
3. The binding studies were conducted by the Psychoactive Drug Screening Program (PDSP) of University of North Carolina Chapel Hill sponsored by the National Institute of Mental Health (NIMH). National Institues of Health, USA.
4. Porsolt RD, Bertin A, Jaffre M.Animal model of depression. Biomedicine 1979; 30:139-140.
5. Petit-Demouliere B, Chenu F, Bouri M. Forced swimming test in mice: a review of antidepressant activity. Pyschopharm 2005; 177:245-255.