Advanced Review

Structure–activity relationships of serotonin 5-HT_{2A} agonists

David E. Nichols*

Of the 14 known types of serotonin receptors one of the most extensively studied is the 5-HT_{2A} (5-hydroxytryptamine) receptor. In the brain, this receptor plays a key role in regulation of cortical function and cognition, appears to be the principal target for the hallucinogenic/psychedelic drugs such as lysergic acid diethylamide (LSD), and also is a target for the newest atypical antipsychotic agents, which are antagonists or inverse agonists at this site. Among the structure–activity relationships that are known for this receptor type, there are three chemical classes of agonists: tryptamines, ergolines, and phenethylamines. Important structural features are identified for agonist activity and some of these agonists have features in common. In addition to effects at the receptor will be the focus, these drugs are also hallucinogenic (psychedelic) agents, and much of the SAR was developed on the basis of effects in humans, before modern pharmacological techniques were available. A certain amount of our knowledge therefore relies on those human studies.

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INTRODUCTION

Over the past half-century, substances now pharmacologically classified as agonists at the serotonin 5-HT_{2A} (5-hydroxytryptamine) receptor have been of considerable interest. Prior to knowledge of the molecular pharmacology of these molecules, it was recognized that they had powerful effects on the human psyche, with drugs such as mescaline and lysergic acid diethylamide (LSD) called hallucinogens or psychedelics. Thus, much of the early work to understand the structure–activity relationships of these drugs was motivated by attempts to understand how these substances worked, and which molecular features were required to produce a psychedelic effect in man. Over the years, as modern pharmacology techniques developed, these studies went in more molecular directions. Our understanding has expanded of the roles played by the 5-HT_{2A} receptor in normal brain function, so that studies of 5-HT_{2A} receptor structure-activity relationships (SAR) today take on greater significance, both from a theoretical and practical perspective.

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There are three main chemical types that are agonists at this receptor: the tryptamines, ergolines related to LSD, which can be considered to be rigidified tryptamines, and the phenethylamines. These are illustrated in Figure 1.

Unfortunately, the data for serotonin receptor affinity and functional potency are fairly sparse for many of these molecules. Early work primarily involved behavioral studies or experiments with a variety of smooth muscle assays (e.g., rat fundus, rat uterus, sheep umbilical artery strips). Although we now know that some of those assays reflect agonist activity at 5-HT_{2A} receptors, one can only infer that those results paralleled the activity at this receptor. Nonetheless, in many cases it is necessary to rely on behavioral or smooth muscle data in order to provide a more complete understanding of the structure–activity relationships of 5-HT_{2A} agonists. Therefore, reports from early studies that bear on a consideration of structure–activity relationships will largely focus on behavioral or, in some cases, human hallucinogenic activity. In more recent years receptor binding and functional data have been available, and the discussion focuses on those data, and largely ignores behavioral data.

The discussion begins with the tryptamines because they are the chemotypes that most closely

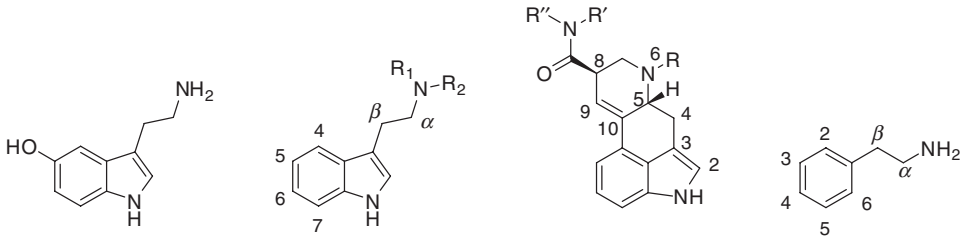


FIGURE 1 | The structure of the neurotransmitter serotonin and the three basic chemotypes of serotonin 5-HT_{2A} (5-hydroxytryptamine) agonists.

resemble the natural neurotransmitter serotonin (5-HT). Consideration is then given to the ergolines, which can be considered to be rigidified tryptamines. These are perhaps the least studied, with the exception of LSD itself. The paucity of structure–activity data for ergolines principally results from the synthetic difficulty attendant to chemical transformations of the ergolines. Finally, the phenethylamines, which are the most extensively explored, are briefly reviewed.

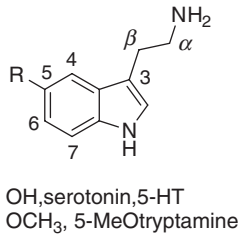


FIGURE 2 | Ring positions for substituted tryptamines.

TRYPTAMINES

Although simple tryptamines are structurally related to the endogenous transmitter serotonin not much molecular modification can actually be carried out on this class of molecule that allows retention of agonist activity. Although a number of simple tryptamines, largely *N*, *N*-substituent variations, have been administered to humans,[1] their effects at the receptor level remain mostly unknown.

Ring Substituents

The 5-hydroxy group of serotonin stands out as a key feature of the molecule. *O*-methylation gives 5-methoxytryptamine, which has high *in vivo* agonist activity at the 5-HT_{2A} receptor, as well as at all other serotonin receptor subtypes. Neither serotonin nor 5-methoxytryptamine has *in vivo* activity if administered orally to humans, presumed to be because of rapid side chain deamination by monoamine oxidase A in the liver. The affinities of 5-HT and 5-methoxytryptamine for the rat 5-HT_{2A} receptor are identical.[2,3] For tryptamines, in general, 5-HT2 agonist activity is generally enhanced by substitution with an oxygen atom at the 4- or 5-position. For example, the K_i of *N*, *N*-dimethyltryptamine in rat brain cortical homogenate has been reported as 75 nM.[4] In that same study, adding a 5-methoxy increased the affinity to 14 nM. Similarly, 4-OH-N,N-dimethyltryptamine (psilocin) had a reported affinity of 6 nM (Figure 2).

It has been reported that 6-fluoro-*N*, *N*-diethyltryptamine (6-F-DET) lacked activity as a hallucinogen in humans.[5] A more recent study showed that it did not possess LSD- or 1-(2,5-dimethoxy-4-iodopheny)-2-aminopropane (DOI)-like activity in a drug discrimination study in rats trained to discriminate these drugs from saline.[6] Although 6-F-DET has affinity for the rat 5-HT_{2A} receptor that is virtually identical to DET, its EC50 in the phosphoinositide (PI) turnover assay (40 μM) was markedly reduced from that of DET itself (5.4 μM), and 6-F-DET had only 63% intrinsic activity at a concentration of 100 μM. This loss of efficacy and potency likely explains the absence of significant DET-like activity in man, despite having a comparable receptor affinity.

In the study by Blair et al.,[6] the effect of ring fluorination also was studied for four other tryptamines, where comparisons were made between 6- and 7-F-psilocin and 4- and 6-fluoro-5-methoxyDMT, **1**, **2**, **3**, and **4**, respectively (Figure 3) with their non-fluorinated counterparts. Fluorination of psilocin in either the 6- or 7- position had identical effects on affinity at the rat 5-HT_{2A} receptor, reducing it by about one-half compared with psilocin itself. Fluorination of 5-MeO-DMT in either the 4- or 6-position had no significant effect on intrinsic activity, but the EC50 values for the fluorinated compounds were increased from 2.4 μM for 5-MeO-DMT to 7.9 and 18.1 μM for the 6-fluoro and 4-fluoro congeners, **3** and **4**, respectively. Fluorination generally had little effect on affinity at the rat 5-HT_{2C} receptor, but surprisingly, had marked effects on 5-HT_{1A} receptor affinity.

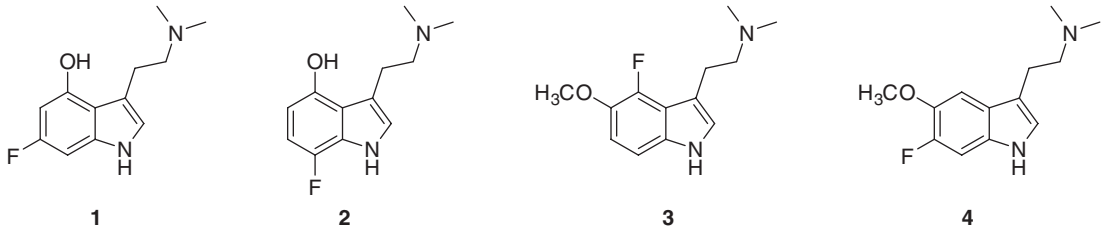


FIGURE 3 | Structures of 6-fluoropsilocin **1**, 7-fluoropsilocin **2**, 4-fluoro-5-methoxy-DMT **3**, and 6-fluoro-5-methoxy-DMT **4**.

The 4-fluoro-5-methoxy-DMT compound (**3**) had affinity at the human 5-HT$_{1A}$ receptor of 0.23 nM, a nearly 10-fold increase over 5-MeO-DMT itself (1.7 nM). This substitution pattern was later exploited to create a 5-HT$_{1A}$ ligand by replacing the *N*,*N*-dimethyl substituents with a pyrrolidyl moiety[4] to afford a molecule **5**, Figure 4, with exceptionally high 5-HT$_{1A}$ receptor affinity and *in vivo* potency in the drug discrimination assay in rats trained to discriminate the 5-HT$_{1A}$ agonist LY293284 from saline.[7]

Early work with benzo[*b*]thiophenes **6** and 3-indenalkylamines **7** (Figure 5) demonstrated that for compounds lacking ring substituents, the ability to act as agonists in the rat fundus was about the same as for the tryptamines themselves.[8] That is, the indole NH was not essential to activate the 5-HT2 receptor in the rat fundus. No modern studies have been carried out to assess affinity at 5-HT$_{2A}$ receptors.

Replacing the phenyl portion of DMT with a thiophene (Figure 6) to afford thienylpyrroles was anticipated to lead to bioisosteric molecules that

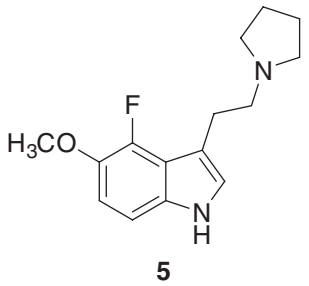


FIGURE 4 | The structure of a 4-fluoro-5-methoxy-substituted tryptamine with potent 5-HT$_{1A}$ (5-hydroxytryptamine) receptor agonist activity.

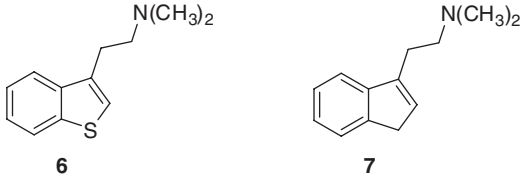


FIGURE 5 | Potential bioisosteres of tryptamines by replacing N(1) with sulfur or CH_2.

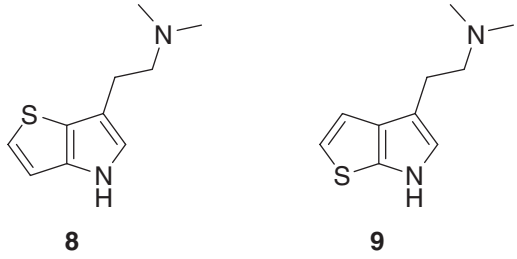


FIGURE 6 | Thieno[3,2-*b*]- and thieno[2,3-*b*]pyrrole analogs of DMT.

possessed DMT-like activity. Thus, Blair et al.[9] reported the synthesis and biological activity, for thieno[3,2-*b*]- and thieno[2,3-*b*]pyrrole analogs **8** and **9**, respectively, of DMT. In radioligand competition experiments, both isosteres had lower affinity than DMT, with the [2,3-*b*] isomer **9** having greatest affinity (106 vs 65 nM for DMT). Both isomers also had somewhat higher affinities than DMT at the 5-HT$_{1A}$ receptor. Affinities at the rat 5-HT$_{2C}$ and human 5-HT$_{1A}$ receptors were increased for both thienopyrroles. Although DMT substituted in a drug discrimination assay in rats trained to discriminate LSD from saline, neither of the thienopyrrole isosteres substituted, nor did they substitute in rats trained to discriminate DOI from saline. With both training drugs, the [3,2-*b*] isomer **8** gave the greatest degree of partial substitution, and it might be speculated that a hydrogen bond donor in the receptor might be weakly able to engage the sulfur atom in the thienyl ring when it was projecting toward the edge of the molecule that normally bears the oxygen atom in serotonin. Both of the thiophene isosteres substituted in rats trained to discriminate the 5-HT$_{1A}$ agonist LY293284, with **8** being about twice the potency of **9**.

Other potential bioisosteres of tryptamines would include replacing the indole N with an oxygen atom to give benzo[*b*]furans (Figure 7). The dimethylamino compound **10** and the racemic α-methyl congener **11** both had about one-sixth the affinity of their indole congeners, measured using displacement of [^{125}I]DOI from rat frontal cortical homogenate.[10] This result parallels the findings

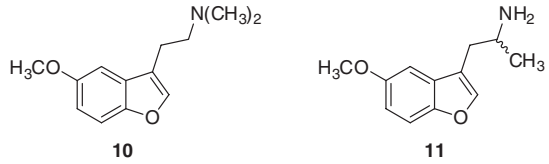


FIGURE 7 | Benzofuran bioisosteres of tryptamines.

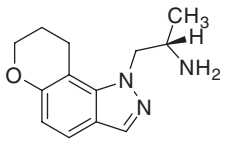
by McKenna et al.,[4] who compared *N*-methyl-*N*-isopropyltryptamine with its benzofuran isostere in its ability to displace [^{125}I]-*R*-DOI from rat cortical homogenate. In that report, the tryptamine had an IC50 of 38 nM whereas the benzofuran IC50 was 500 nM, 13-fold lower affinity.

An interesting variation on ring substitutions was the discovery of indazole ligands with high 5-HT_{2A} agonist activity.[11,12] In particular, AL-38022A **12** was developed as a highly potent 5-HT_{2A} agonist with effects against glaucoma (Figure 8). It was a full agonist at all of the 5-HT2 receptor subtypes, with an EC50 in the range 0.5–2.2 nM for several functional responses.[13] It produced full substitution in a drug discrimination assay in rats trained to discriminate the hallucinogen 1-(2,5-dimethoxy-4-methylphenyl-2-aminopropane (DOM) from saline, with an ED50 of 0.05 mg/kg. It also produced full substitution in monkeys trained to discriminate DOM from saline, with an ED50 of 0.04 mg/kg, and comparable to the potent 5-$HT_{2A/2C}$ agonist DOI.

N-Alkylation

Another obvious area for molecular modification is the side chain amino group, alkylating it to produce a variety of secondary or tertiary amines. There are extensive data published for effects of a number of *N*-substituted tryptamines in humans,[1] but generally there are only scant data available for actual receptor affinities/potencies.

The most systematic study of receptor effects of tryptamine *N*-alkylation was reported by McKenna et al.[4] In that study, several series of *N*-alkylated tryptamines were examined, either with no ring substituents, or a 5-methoxy or 4-hydroxy substituent. Displacement of [^{125}I]DOI from rat cortical homogenate was used to measure 5-HT_{2A} receptor affinity. Highest affinities (4–30 nM) were observed with *N*,*N*-dimethyl, *N*,*N*-diethyl, *N*-methyl-*N*-isopropyl, and *N*,*N*-diisopropyl substituents. Even 4-OH-*N*,*N*-di(sec)butyltryptamine had a reported the affinity of 39 nM, but the affinity of 4-OH-*N*,*N*-diisobutyltryptamine dropped to 260 nM. When the dialkyl groups were tethered into a heterocyclic ring, an *N*-pyrrolidyl had affinity similar to *N*,*N*-dimethyltryptamine (110 vs 75 nM, respectively), but the affinity of the *N*-piperidyl dropped to 760 nM. Although no receptor data are reported for it, 5-methoxy-*N*,*N*-diallyltryptamine (5-MeO-DALT) has recently appeared on the street as a new 'legal high'.



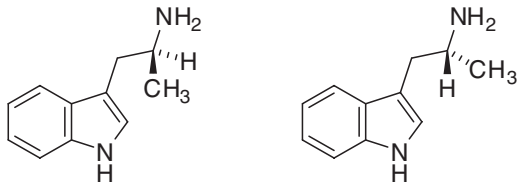
12, AL-38022A

FIGURE 8 | An indazole analog of a tryptamine with potent 5-HT_{2A} (5-hydroxytryptamine) agonist activity.

Side Chain Alkylation

Addition of an α-methyl to the side chain of tryptamines generally renders them orally active, by inhibiting their deamination by monoamine oxidase (MAO). This feature also creates a stereo center in the molecule, and the enantiomers generally have differing biological activities. Racemic α-methyltryptamine itself had affinity at the human 5-HT_{2A}, 5-HT_{2B}, and 5-HT_{2C} receptors of 164, 58, and 30 nM, respectively.[14] The affinities of *R*- and *S*-α-methyltryptamine, Figure 9, ***R***-13 and ***S***-13, respectively, in rat cortical homogenate using [^{125}I]DOI displacement were reported as 130 and 46 nM, respectively.[4]

For 5-HT_{2A} agonist activity, the enantiomer with the *S*-(+)-configuration is most active for molecules with a 5-OH or 5-OCH_3 substituent.[15] This *in vitro* observation translates into *in vivo* human hallucinogenic activity, where (*S*)-(+)-5-methoxy-α-methyltryptamine has an effective human hallucinogenic dosage of about 2.4 mg, whereas 3.0 mg of the *R* isomer failed to produce a significant effect.[16] Furthermore, the *S*-(+)- enantiomer has affinity comparable to the non-alkylated tryptamine, whereas the (−) isomer is less potent. That is, a tryptamine



S-(+)-AMT **(*S*)-13** *R*-(-)-AMT **(*R*)-13**

FIGURE 9 | The structures of *S*-(+)- and *R*-(−)-α-methyltryptamine.

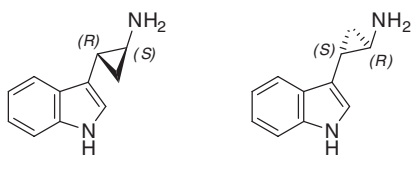
and the *S* enantiomer of its α-methyl congener have comparable affinities, whereas *R*-α-methyltryptamine is less potent. The affinities of *R*- and *S*-5-MeO-alpha-methyltryptamine at the rat 5-HT$_{2A}$ receptor labeled by [^{125}I]DOI were reported as 47 and 2 nM, respectively.[3] This stereochemical preference appears to be reversed at the rat 5-HT$_{1B}$ receptor.[15]

Extending the α-methyl to α-ethyl leads to a compound known as Monase, which was marketed as an antidepressant until 1962. It appeared in Germany in 1986 as a 'designer drug' that was associated with one death.[17] It has been reported to have 'neurotoxic' properties similar to 3,4-methylenedioxymethamphetamine (MDMA)[18] and, not surprisingly, has been described as having MDMA-like psychopharmacology in humans,[19,20] but there are no data on its activity at the 5-HT2 receptors.

A logical extension of this work was a study of *trans*-2-(indol-3-yl)cyclopropylamines **14**.[14] The (1*R*,2*S*)-(−)- enantiomer of *trans*-2-(indol-3-yl)cyclopropylamine had highest affinity at human 5-HT$_{2A}$ and 5-HT$_{2B}$ sites, but surprisingly, the (1*S*,2*R*)-(+)- isomer had higher affinity at the 5-HT$_{2C}$ receptor. Ring substituents 4-OMe, 5-OMe, and 5-F generally increased affinity over the unsubstituted compound, but unfortunately, the difficulty of synthesis and extreme chemical instability of the compounds precluded preparation of the enantiomeric substituted compounds (Figure 10).

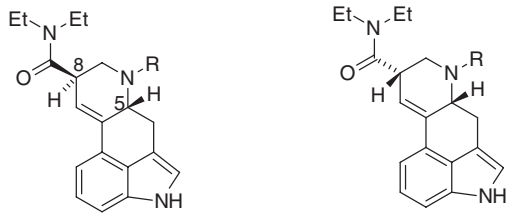
ERGOLINES

Ergolines are tetracyclic molecules, ultimately derived from alkaloids produced by the ergot fungus. The most important one, from the perspective of 5-HT$_{2A}$ agonists, is LSD, also referred to as LSD-25. LSD is the most potent of the psychedelic agents in humans, although its affinity and functional potency at the human 5-HT$_{2A}$ receptor are fairly unremarkable compared with simpler compounds such as DOI. Numerous clinical studies of LSD and certain of its congeners were performed in the 1950s and 1960s. Those studies have been reviewed in some



1S,2R-(+)-**14** *1R,2S*-(−)-**14**

FIGURE 10 | Enantiomeric-*trans*-cyclopropane analogs of tryptamines.



5*R*,8*R*-LSD 5*R*,8*S*-isoLSD

FIGURE 11 | *N*, *N*-diethyllysergamide (LSD) and inactive epimeric iso-LSD.

detail,[21–24] and little substantive new information has been published since then, with a few exceptions that are discussed below.

Both carbons 5 and 8 are chiral, and it is only ergolines with the 5*R*,8*R*-configuration, as illustrated in Figure 1, which have biological activity. That isomer is dextrorotatory, so LSD is referred to as (+)-LSD or *d*-LSD. Early receptor binding studies by Bennett and Snyder[25] demonstrated that (+)-LSD had nanomolar affinity for [^{3}H]LSD-labeled sites in rat cortex, whereas its enantiomer, 5*S*,8*S*-(−)-LSD, had 2500-fold lower affinity. The 8-position readily epimerizes to provide (+)-isolysergic acid diethylamide, which has about 30-fold lower affinity and is inactive as an hallucinogen. This transformation is facile and occurs under slightly acidic pH (Figure 11).

Because of its complex structure, only a few modifications of LSD have been carried out, and those involved alterations of the amide function, reduction of the 2,3- or 9,10-double bonds, substitutions on the indole nitrogen or at the 2-position, and changes in the alkyl group on the basic nitrogen atom. Unfortunately, very few of these changes have been studied using modern molecular pharmacology methods, and only some of them have been assessed in human psychopharmacology.

Halogenation at the 2-position of LSD as in 2-bromo-LSD (BOL-148) or 2-iodo-LSD leads to molecules that are 5-HT$_{2A}$ antagonists. Although virtually no work has been done with BOL-148 since the early 1970s, it was demonstrated early on that it could block the effects of LSD in humans.[26] [^{125}I]2-Iodo-LSD has found application as a radioactive label for 5-HT2 family receptors.[27–30]

Reduction of the 9,10-double bond of LSD abolishes hallucinogenic activity.[31,32] The reason(s) for the loss of activity are not clear, nor has there ever been a comparison of receptor activities of dihydro-LSD with those of LSD. Reduction of the 9,10 olefinic bond of LSD gives a molecule that still maintains relative planarity, like LSD. Although a correlation has been reported between hallucinogenic activity of

tryptamines and the orientation of one of the nodes in the highest occupied π-like orbital,[33] this correlation failed for LSD, and the author of this study stated, 'The 9,10 double bond in LSD must fulfill some role that is not modeled in this work.'

Reduction of the 2,3-bond of the indole nucleus leads to a compound reported to have about one eighth the psychoactivity of LSD.[34] This compound had a delayed onset of action relative to LSD, and the authors speculated that 'a metabolic change to a more active substance' might contribute to the difference. As 2,3-dihydroindoles can be fairly readily aromatized to indoles, it still seems possible that such an oxidative transformation might take place in the body.

Extension of the N(6)-methyl group of LSD to longer alkyl groups[35] gives compounds that are more potent than LSD *in vivo* in rodent behavior and which in some cases have potency comparable to, or slightly greater than LSD in humans.[1] It remains unknown what effect extension of the *N*-alkyl group has on serotonin receptor affinity and potency.

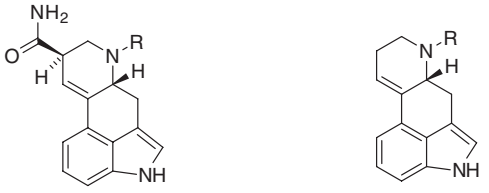
Studies of the Amide Portion of Lysergic Acid Derivatives

The simplest ergoline with human psychoactive properties, and presumably 5-HT_{2A} agonist activity, is lysergic acid amide (**15**, ergine), which was reported by Hofmann and Tscherter[36] to be the active component in *Rivea corymbosa* seeds, used by the Aztecs in various magical potions and ointments (Figure 12). Surprisingly, if the C(8) amide substituent is removed completely to give 8-descarboxy lysergic acid **16**, the compound is reported to produce a behavioral profile in mice 'remarkably similar to that shown by LSD'.[37]

With respect to other lysergic acid amides, it is noteworthy that the *in vivo* potency of LSD is exquisitely sensitive to the presence and nature of the *N*,*N*-diethylamide moiety. It has been known for more than half a century that any change, however slight, results in about one order of magnitude loss in potency. Clearly, this loss of effect cannot simply be related to hydrophobicity, and is probably not a function of metabolic liability in the body. Rather, it was hypothesized that the receptor(s) might have a specific stereochemically defined and sterically constrained region that accommodated the diethylamide moiety.

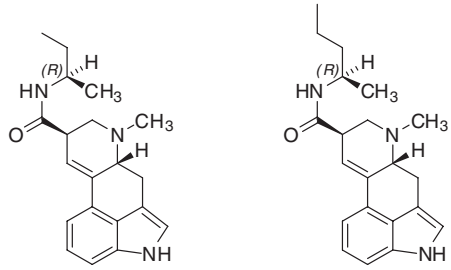
Strong evidence that this amide-binding region might be very specific was provided by the finding that lysergamides of *R*- and *S*-2-aminobutane gave lysergamides that differed in their pharmacological properties.[38] The amide with the *R* configuration in the amine **17** was essentially equipotent to LSD in a drug discrimination assay in rats trained to discriminate LSD from saline. The lysergamide from the *S*-amine had only about one fourth the potency. In radioligand displacement studies, using [^{125}I]DOI in rat frontal cortical homogenate, the lysergamides with the *R*- and *S*-2-aminobutane amide had K_i values of 2.6 and 7.8 nM, respectively, which correlated with their *in vivo* drug discrimination potencies (Figure 13).

This approach was extended by the examination of a series of chiral 2-aminoalkane amides of lysergic acid, where the alkyl group was extended from butyl to heptyl.[39] In that study, [^{3}H]ketanserin displacement from rat frontal cortex homogenate was used to measure 5-HT_{2A} receptor affinity. In every case, the lysergamide with the *R* configuration in the amide secondary alkyl group had higher affinity than the one with the *S* configuration. Affinity dropped off as the chain length increased, with the *R*-heptyl amide having a K_i of only 80 nM. Interestingly, extending the length of the 2-alkyl groups of the amide markedly increased the 5-HT_{1A} receptor affinity, with the *R*-hexyl substituent having a K_i of 0.32 nM! This finding indicates that the 5-HT_{1A} receptor has greater tolerance for bulk attached to the amide than does the 5-HT_{2A} receptor. The only compounds tested in functional assays were the pentyl isomers **18**, where each isomer was a full agonist in the phosphoinositide hydrolysis assay, but the *S* isomer was about 17-fold less potent (see Table 1 for comparisons of various amide-substituted lysergamides).



15 16

FIGURE 12 | The structures of lysergic acid amide **15** (ergine) and 8-descarboxy lysergic acid **16**.



R-17 18

FIGURE 13 | The structures of lysergamides of *R*-2-butyl **17** and *R*-2-pentylamine **18**.

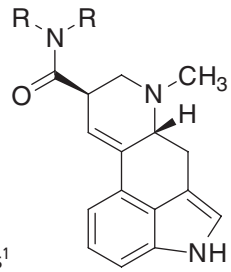


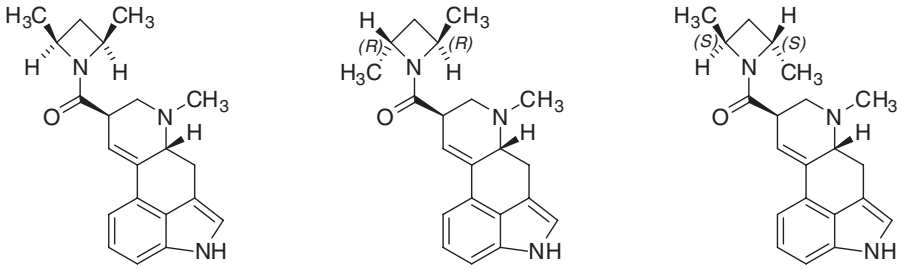
TABLE 1 | 5-HT_{2A} Receptor Affinity and Functional Effects of Selected Lysergamides[1]

Compound Number	Amide *R*,*R* Groups	5-HT_{2A}[2]	5-HT_{2C}[3]	5-HT_{1A}[4]	IP3 (EC50)
	N,*N*-diethyl (LSD)	2.1 ± 0.03	2.3 ± 0.2	1.1 ± 0.3	8.4 ± 1.6 (22.4 ± 2.7%)
***R*-17**	H,*R-sec-butyl*	2.6 ± 0.4		2.0 ± 0.2	
***S*-17**	H,*S-sec-butyl*	7.8 ± 0.2		4.6 ± 0.3	
***R*-18**	H,*R-sec-pentyl*	4.5 ± 0.5		0.6 ± 0.1	5.4 ± 2.1 (100%)
***S*-18**	H,*S-sec-pentyl*	34 ± 2		8 ± 1	91 ± 25 (100%)
***R*-19**	H,*R-sec-hexyl*	16 ± 2		0.32	
***S*-19**	H,*S-sec hexyl*	55 ± 7		4.9 ± 0.3	
***R*-20**	H,*R-sec-heptyl*	80 ± 9		3.3 ± 0.4	
***S*-20**	H,*S-sec-heptyl*	360 ± 20		14 ± 2	
21	H,*3-aminopentane*	8 ± 0.2		2.1 ± 0.3	
22	*cis*-2,3-Dimethylazetidine	7.9 ± 0.85	23 ± 2.9	1.1 ± 0.12	69 ± 8.1 (30%)
23	*R*, *R-trans*-2,3-dimethylazetidine	21 ± 4	130 ± 11	6.8 ± 0.26	102 ± 25 (36%)
24	*S*, *S-trans*-2,3-dimethylazetidine	8.3 ± 1.7	6.5 ± 0.15	0.45 ± 01	19 ± 4.5% (43%)
25	$-(CH_2)_5-$	12.2 ± 0.2	6.1 ± 0.5	0.66 ± 0.08	140 ± 26 (33.7 ± 5.1%)
26	2,5-Dimethylpyrrolidyl	27 ± 1	11.2 ± 0.5	24 ± 4.8	18.2 ± 3.6 (36.5 ± 3.7%)
27	$-(CH_2)_6-$	2.6 ± 0.1	2.3 ± 0.1	0.45 ± 0.09	26 ± 1.9 (46.3 ± 6.4%)
28	$-(CH_2)_2O(CH_2)_2-$	16.2 ± 1.8	51 ± 2.0	2.6 ± 0.3	173 ± 35 (31.8 ± 2.8%)
29	*N*-methyl-*N*-isopropyl	3.2 ± 0.1	7.4 ± 0.4	2.1 ± 0.4	18.2 ± 3.6 (36 ± 2.1%)
30	*N*-ethyl-*N*-2,2,2-trifluoroethyl	1.6 ± 0.03	1.8 ± 0.2	1.1 ± 0.2	18.6 ± 3.4 (31.8 ± 2.8%)
31	*N*-ethyl-*N*-2-methoxyethyl	7.1 ± 0.4	7.8 ± 0.5	4.0 ± 0.9	30.3 ± 6.7 (29.6 ± 4.5%)

[1]Data from Parrish.[42]
[2][^{125}I]DOI, human 5-HT_{2A}.
[3][^{125}I]DOI, human 5-HT_{2C}.
[4][^{3}H]-8-OH-DPAT human 5-HT_{1A}.

In drug discrimination tests in rats trained to discriminate LSD from saline, substitution occurred with the *R*-pentyl lysergamide **18**, but not with the *S*-pentyl, the hexyl, or heptyl compounds. These *in vivo* results parallel the affinities observed at the rat 5-HT_{2A} receptor.

The hypothesis that the receptor might have a specific region that was optimally complementary to the *N*, *N*-diethylamide was finally tested by the synthesis of conformationally-constrained 2,3-dimethylazetidine amides of lysergic acid. These dimethylazetidines can exist in three isomeric forms: a 2,3-*cis*-meso isomer **22**, *R*,*R-trans* **23** and *S*,*S-trans* **24** isomers. The lysergic acid amide of each of these was prepared and tested, and in drug discrimination experiments in rats trained to recognize the effects of LSD, the *S*, *S-trans*-azetidide **24** gave the lysergamide that was most similar to LSD in potency.[40] As can be seen in Table 1, a comparison of the affinities and 5-HT_{2A} potencies of LSD with each of the three azetidide congeners also revealed that the *S*, *S* congener **24** had a profile most nearly comparable to LSD. The *R*, *R* compound **23** had 2- to 3-fold lower affinity at the 5-HT_{2A} receptor and a 50- to 60-fold lower affinity at the 5-HT_{2C} receptor. The *cis* compound **22** principally differed from the *S*, *S* isomer in that it had about fourfold lower affinity at the 5-HT_{2C} receptor. The *S*, *S* isomer was about one-half the potency of LSD for the activation of phosphoinositide hydrolysis, whereas the *R*, *R* and *cis* compounds were 8- to 12-fold less potent (Figure 14).



22 23 24

FIGURE 14 | Lysergamides from *cis* **22**, *S,S-trans* **23**, and *R,R-trans* **24** dimethylazetidines.

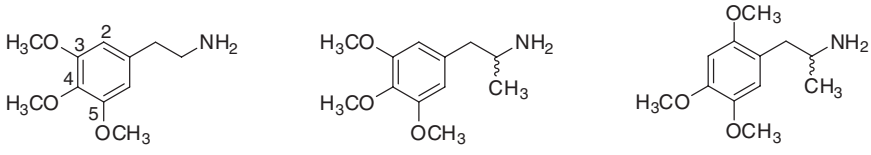
More recently, we have developed an in-silico-agonist-activated model of the 5-HT_{2A} receptor. Docking, molecular dynamics, and minimization of LSD in the receptor revealed that the diethyl groups nestle into a region that is bounded by a number of receptor residues.[41] Thus, as conjectured, the receptor appears to have evolved so that it is apparently specifically complementary to the diethyl group on LSD. When LSD was docked into the receptor, following molecular dynamics (MD) and minimization, the ethyl groups of the amide adopt conformations anticipated from the studies of the 2,3-dimethylazetidide compounds.

PHENETHYLAMINES AND CONGENERS

The phenethylamines have been the most widely explored class of 5-HT_{2A} agonist. To complement the discussion here the reader is encouraged to read an earlier review on this topic,[43] as well as a more recent extensive review on phenethylamine 5-HT_{2A} agonists.[44]

The prototype for this class is mescaline **32**, a simple trimethoxy phenethylamine that was first isolated from the peyote cactus, *Lophophora williamsii*. Like all known 5-HT_{2A} agonists it is hallucinogenic in man, but has very low potency, a typical oral dose of the sulfate being in the range 250–400 mg. The earliest structure modification of mescaline was to introduce a methyl group onto the α-side chain carbon, leading to a compound known as trimethoxyamphetamiine (TMA) **33**,[45,46] to provide the first of a very large class now generically referred to as 'substituted amphetamines'. Between 1964 and 1969 Alexander Shulgin carried out a series of experiments where the ring substituents were varied to establish that the most potent hallucinogenic amphetamines had a 2,4,5-ring substitution pattern,[47] with the simplest member named TMA-2 (**34**). Additional studies have been summarized by Shulgin in 1978.[48] Although no receptor affinities or potencies are reported, the 1991 compendium by the Shulgins[16] lists dosages and effects for a large number of substituted phenethylamines, and it can be inferred that these potencies must reflect, at least in part, the 5-HT_{2A}-receptor-activating properties of those molecules (Figure 15).

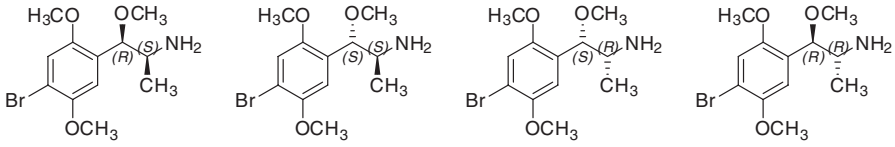
The development of an asymmetric synthesis allowed the facile preparation of the enantiomers of numerous substituted amphetamines.[49] Aldous et al.[50] also reported a method for resolution of the enantiomers through the recrystallization of *N*-benzyloxycarbonyl-L-phenalanine-*p*-nitrophenyl esters. Unfortunately, these developments preceded the modern molecular biology era, and affinity and potency at actual receptors could not be reported at that time. Some of the assays, however, were highly correlated with *in vivo* hallucinogenic potencies in humans, which today we know are mediated by the activation of serotonin 5-HT_{2A} receptors. Thus, one



Mescaline TMA TMA-2

32 33 34

FIGURE 15 | Structures of mescaline, the α-methyl derivative of mescaline, TMA, and its isomeric 2,4,5-substituted analog, TMA-2.



(1*S*,2*R*) K_i 17.4 nM **35**

(1*S*,2*S*) K_i 6 nM **36**

(1*R*,2*S*) K_i 0.8 nM **37**

(1*R*,2*R*) K_i 0.3 nM **38**

FIGURE 16 | Serotonin 5-HT$_{2A}$ (5-hydroxytryptamine) receptor affinities for stereoisomeric β-methoxy DOB analogs.

can infer that much of the structure–activity data for hallucinogenic agents could be interpreted as reflecting the activity at this receptor.

Although the *R*-(−) enantiomers of hallucinogenic amphetamines are most potent in humans, and are more potent in activating the human 5-HT$_{2A}$ receptor, in dog peripheral vasculature the *S*-(+)-isomers are more potent in producing smooth muscle contraction.[51]

β-Oxygenation

Glennon et al.[52] have studied the effect of β-oxygenation on the 5-HT$_{2A}$ agonist properties of 1-(2,5-dimethoxy-4-bromophenyl)-2-aminopropane (DOB) (Figure 16). The four stereoisomers of the β-oxygenated compounds were studied, either with a β-OH or a β-OCH$_3$. As shown in Figure 16, the 1*R*,2*R* stereoisomer **38** had the highest affinity.

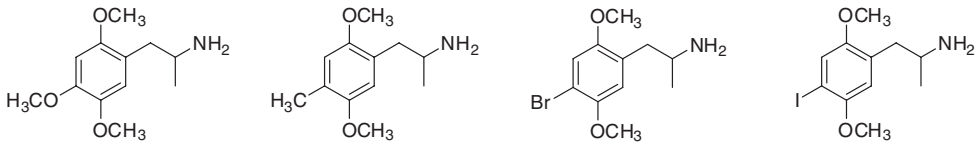
The affinities clearly correlate with the stereochemistry at the α-carbon, as the *R* stereochemistry has highest affinity in the β-unsubstituted amphetamines. With respect to efficacy in a cell-based calcium mobilization assay, the 1*R*,2*R* stereoisomer **38** was a full agonist (93% efficacy), whereas the other isomers were partial agonists, with efficacies varying from 31 to 54%. The corresponding β-hydroxy compounds were less potent and less efficacious, but the 1*R*,2*R*-β-hydroxy analog still fully substituted in a two-lever drug discrimination task in rats trained to discriminate DOM from saline. It is perhaps not surprising, therefore, that an earlier report of analogous β-oxygenated compounds described hallucinogen-like effects in man.[53]

Ring Substituents

Following the pioneering work of Shulgin establishing that the 2,4,5- substitution pattern was optimal for hallucinogenic activity in the substituted amphetamines, extensive work ensued to establish the range of substitution that could be tolerated. It should be noted, however, that 2,4,5-trimethoxyphenethylamine, an isomer of mescaline, lacks mescaline-like effects in man.[48] In general, 2,5-dimethoxy substituents provide optimal activity and receptor affinity, although an early study suggests that the 2-methoxy, but not the 5-methoxy, may be replaced by an OH group.[54]

An early review of hallucinogenic potency in humans first summarized the SAR of various ring substituents and orientations in substituted amphetamines.[47] A relatively hydrophobic substituent at the 4-position in either 2,4,5- or 3,4,5-substituted molecules gives the most potent compounds. The initial observation of this effect was the potency of the 4-methyl compound, DOM (**39** "STP"), compared to its 2,4,5-trimethoxy **34** (TMA-2) substituted congener, where DOM was about 10 times more potent than TMA-2 (Figure 17).

A dramatic example of this substitution pattern is exemplified by the series of three dimethoxy-monoethoxy amphetamines. Only the 2,5-dimethoxy-4-ethoxy compound (MEM) had good activity, whereas 2,4-dimethoxy-5-ethoxy and 2-ethoxy-4,5-dimethoxy substituted compounds did not.[55,56] A variety of alkyl groups have been shown to give active compounds, including the short alkyl groups ethyl and propyl, as



TMA-2 **34** DOM **39** DOB **40** DOI **41**

FIGURE 17 | Potent-substituted amphetamine-type hallucinogenic 5-HT$_{2A}$ (5-hydroxytryptamine) receptor agonists with different 4-substituents.

well as the halogens Cl, Br, and I, and a variety of alkylthio[50,57,58] substituents. The substituent that so far affords a compound with the highest potency is the 4-trifluoromethyl moiety.[59]

The [^{125}I]-labeled 4-Iodo congener **41** (DOI) as well as its phenethylamine counterpart, 2C-I, have been used as radioligands to label the 5-HT$_{2A/2C}$ receptors.[3,27,60] The [^{131}I]-labeled compound also was briefly studied as a potential imaging agent,[61] as were the [^{82}]Br and [^{77}]Br isotopically-labeled versions of DOB **40**, which had suggested uses as brain-scanning agents.[62]

A 4-alkylthio substituent also gives very active compounds. Jacob et al.[63] individually replaced the methoxy groups of 2,4,5-trimethoxyamphetamine with methylthio groups. Using the rabbit hyperthermia assay, it was demonstrated that the 2,5-dimethoxy-4-methylthio compound[58] had about one-half the potency of DOM. Replacing the 2- or 5-methoxy groups with methylthio groups afforded compounds that were nearly inert, compared to DOM. These experiments clearly showed that in the phenethylamines the oxygen atom at the 2- and 5-positions was a requisite for high agonist activity. It might be noted that the 5-methylthio analog was nearly twice the potency of the 2-methylthio compound, indicating that there is some greater degree of tolerance at the 5-position, but nonetheless, anything other than an oxygen atom at the 2- and 5-positions is quite deleterious to activity. The 2- and 5-methoxy groups of DOM and DOEt also were individually replaced with methylthio groups, and again, the resulting thio analogs suffered a dramatic loss of potency, as assessed in human self-experiments.[64]

When a similar approach was employed with mescaline analogs somewhat different results were obtained. It was found that replacing the 3-methoxy with a methylthio (**42**) gave a compound that was rated in human self-experiments to be about sixfold more potent than mescaline itself.[65] Replacing the 4-methoxy of mescaline (**43**) gave a compound that was estimated to be about 12-fold more potent than mescaline. In this case, therefore, the activity increased when either the 3- or the 4-methoxy is replaced by methylthio, suggesting a less critical role for the 3-methoxy in mescaline analogs than for the 2-methoxy in the 2,4,5-substitution series.

Relatively hydrophobic 4-substituents probably play several roles in the biological activity of these molecules. First of all, they provide compounds with improved pharmacokinetic properties. That is, alkyl groups, halogens, and alkylthio groups increase the overall hydrophobicity of the molecules so that they partition better into the central nervous system (CNS).[66] This importance also is evident in 3,4,5-substituted mescaline analogs bearing more hydrophobic substituents in the 4-position.[67] A later study identified a steric limitation to the size of the 4-substituent, suggesting that an alkyl group of only about three carbon atoms was tolerated before the activity dropped off.[68] By contrast, polar 4-substituents such as OH, NH_2, and COOH gave compounds with very low affinity (K_i > 25,000 nM).[69] The latter study also examined compounds with very long lipophilic 4-substituents such as *n*-hexyl and *n*-octyl, which had high affinities at [^{3}H]ketanserin-labeled sites. Although smaller substituents gave agonist molecules, preliminary studies with these latter compounds suggested that they were 5-HT2 *antagonists*.

A comparison of two isomeric 4-butyl groups in this series (Figure 18) revealed that 2,5-dimethoxy-4-isobutylamphetamine **44** retained significant activity in a drug discrimination task, in rats trained to discriminate LSD from saline, whereas the 2-butyl homolog was about one third less potent than the isobutyl and also failed to produce full substitution in the rats. Asymmetric synthesis of the two separate 2-butyl isomers, with either *R* or *S* stereochemistry in the 2-butyl group, was then carried out. Assessment of receptor affinities by displacement of [^{125}I]DOI from rat frontal cortical homogenate revealed identical K_i values of 7.8 nM.[70] Drug discrimination tests in LSD-trained rats showed, however, that the *R* isomer (**45**) was slightly more potent than the *S* (**46**) (ED50 of 3.1 vs 4.8 μmol, respectively) (Figure 19). This difference could reflect a slight difference in functional potency or intrinsic activity, but those were not examined. In general, however, the conclusion is that there is no chiral discrimination by the receptor in this region, and that branching in the 4-alkyl group, *per se*, is detrimental to the activity.

Very large bulky groups at the 4-position, such as the *tert*-butyl, lead to inactive compounds,[16,71–74] although the 4-isopropyl compound DOIPr is reported to retain good human activity.[16] Not surprisingly, therefore, aryl groups attached at the 4-position gave antagonists, generally with low affinity.[75] Interestingly, however, when a 3-phenylpropyl substituent was introduced at this position, the compound proved to be a weak partial agonist.[76]

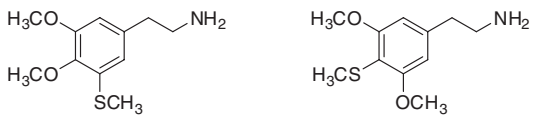
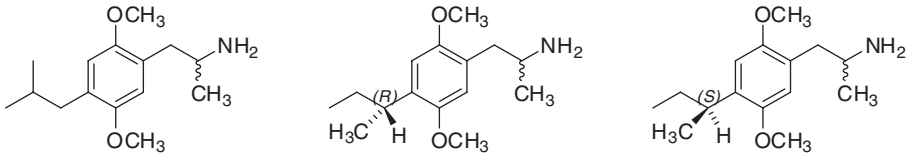


FIGURE 18 | Sulfur analogs of mescaline.



44 45 46

FIGURE 19 | Potential 5-HT$_{2A}$ (5-hydroxytryptamine) receptor agonists with an isomeric 4-butyl ring substituent.

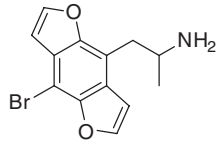
The key to retaining agonist activity seems to be retention of the 2,5-dioxygenation pattern, coupled with a hydrophobic 4-substituent that meets certain size and hydrophobicity criteria. Nevertheless, there are a number of structural modifications of this basic pharmacophore that lead to compounds with high affinity at the 5-HT$_{2A}$ receptor, but which are antagonists. For example, removal of the 5-methoxy group, and moving the alkyl substituent from the 4- to the 5-position, gave a high affinity antagonist.[77]

The correlation between lipophilicity of the 4-substituent as well as limitations on the length and bulk of the substituent are consistent with the presence of a complementary hydrophobic region within the 5-HT$_{2A}$ receptor. Although the location of this putative hydrophobic region has not yet been elucidated, it is evident from simulated docking studies[78,79] that it must lie somewhere within transmembrane helices 5 and/or 6. Although indoles engage Ser242 within the human 5-HT$_{2A}$ receptor, the dimethoxy-substituted phenethylamines do not.[80] The bicyclic indole nucleus of serotonin also is larger than the phenyl ring of the phenethylamines, and it seems possible that the hydrophobic 4-substituent of the phenethylamines acts as a sort of wedge or spacer to fill more fully the binding cavity that has evolved to accommodate the relatively larger indole nucleus of serotonin.

Constrained Methoxy Mimics—Benzofuran Analogs

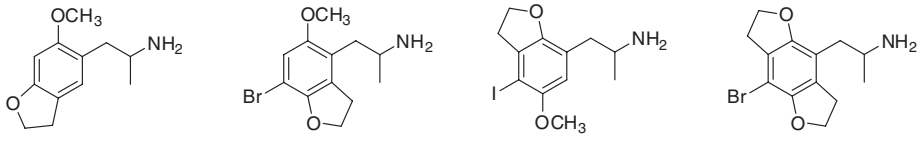
In the author's laboratory we had reasoned that the aromatic methoxy groups might be serving as hydrogen bond acceptors for some polar residues within the receptor site. If that hypothesis was true, there should be a dependence on the orientation of the unshared electron pairs on the oxygen atoms. Early studies quickly demonstrated that when the 5-methoxy group was constrained into a dihydrofuran, the orientation of the ring was crucial, consistent with the notion of a specific orientation of the oxygen unshared electrons. When the 5-methoxy of DOM (**39**) was 'tethered' to the 4-position, as in **47**, the activity was reduced nearly 20-fold compared to DOM in drug discrimination tasks.[81] By contrast, when the 5-methoxy was tethered to the 6-position, compound **48** was as at least as potent as DOM.[82] These studies seemed to indicate clearly that the electrons of the methoxy oxygen should be oriented in a particular direction for optimal receptor interaction. Affinity for the [^{125}I]DOI-labeled receptor in rat prefrontal cortex paralleled these findings, with an affinity for **47** of 488 nM and for **48** of 3.1 nM (Figure 20).

A similar approach to tethering of the 2-methoxy into a dihydrofuran also led to a very potent compound (**49**),[83] but when both the 2- and 5-methoxy functions were incorporated into dihydrofuran rings, the result was a series of exceptionally potent 5-HT$_{2A/2C}$ agonists exemplified by **50**,[83,84] which had an affinity of 0.48 nM for the cloned human 5-HT$_{2A}$ receptor. Aromatization of the dihydrofuran rings to afford **51** led to even further enhancement of affinity and potency (Figure 21).[85]



51

FIGURE 21 | An extremely potent rigid-substituted amphetamine analog.



47 48 49 50

FIGURE 20 | Phenethylamines with methoxy groups constrained into dihydrofuran moieties.

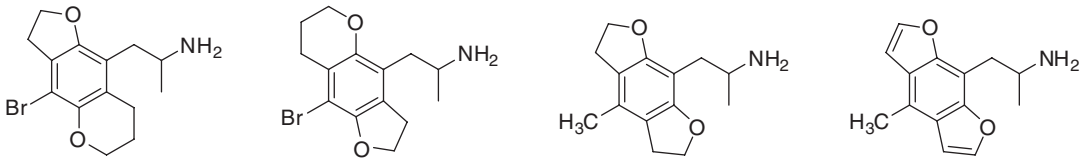


FIGURE 22 | Ring-expanded molecules with 'rigidified methoxy groups'.

Finally, hybrid benzofuran and benzopyran molecules were designed and tested to determine whether the 2- or 5-methoxy groups were more sterically restricted within the receptor.[86] The compound with the dihydrofuran replacing the 2-methoxy (**52**) had slightly higher 5-HT_{2A} affinity than did **53** (3.6 vs 5.3 nM, respectively); **52** also was about fourfold more potent than **53** for inducing PI turnover. In rats trained to discriminate LSD, parallel results were observed, where the **52** was about three times more potent than **53** (Figure 22). The results were explained on the basis of docking into a homology model of the 5-HT_{2A} receptor, which places the 2-methoxy downward toward the bottom of the receptor, with the 5-methoxy projecting toward the extracellular space. Thus, it was reasoned, there may be less steric tolerance for modifications of the 2-methoxy because of its location projected down into the ligand binding site.

A similar strategy applied to 2,6-dimethoxy-4-methylamphetamine likewise resulted in significant potency enhancement in the dihydrofuran **54**, with a further increase in the fully aromatic **55**.[87] Despite the presumed loss of hydrogen bond strength for a furan versus a dihydrofuran or methoxy group, these compounds obviously bound well to the receptor and it may be that the fully aromatic planar tricyclic difurano analogs present a larger planar aromatic hydrophobic face within the receptor that adds to binding energy through van der Waals or pi stacking interactions.

Surprisingly, however, when this strategy was applied to mescaline analogs, activity of the tethered compounds was reduced. The mono furanyl compound **56** lost efficacy and mescaline-like potency in a rat behavioral model, and the difuranyl compound **57** suffered a further decrease in the activity.[88] It was speculated that for 3,4,5-substituted compounds, perhaps the methoxy groups needed to be freely rotating in order to achieve the active binding orientation. In any event, these divergent results support the idea that the binding pose of 2,4,5-substituted compounds differs from that of 3,4,5-substituted compounds (Figure 23).

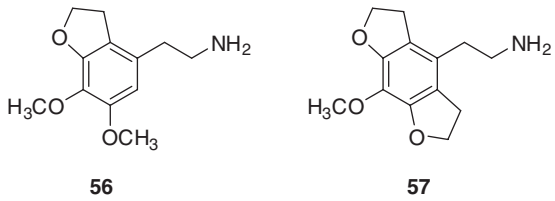


FIGURE 23 | Mescaline analogs with constrained methoxy groups.

The discovery that DOI could reduce intraocular pressure[89] led to efforts to identify novel 5-HT_{2A} agonists that might be useful to treat glaucoma, but lacking the side effects that are typical of hallucinogenic agents. As a result, a number of benzodifurans were evaluated with alkoxymethyl and oxadiazole methyl substituents that had high 5-HT_{2A} agonist activity, with the goal of reducing lipophilicity so as to minimize penetration into the CNS. Several of these compounds had high potency with reduced lipophilicity, and are exemplified in Figure 24.[90]

Effect of α-Alkylation

Study of the effect of the α-methyl group in phenethylamines has shown that *racemic* amphetamines have approximately the same affinity at the human 5-HT_{2A} receptor as do the unmethylated phenethylamines.[60,91–93] In a study by Parrish et al.[91] the EC50s for stimulating phosphoinositide hydrolysis were nearly identical for phenethylamines and amphetamines, but the intrinsic activity was higher for the racemic amphetamines. For the amphetamine enantiomers, however, the affinity, potency, and intrinsic activity were significantly higher for the *R*-(−)

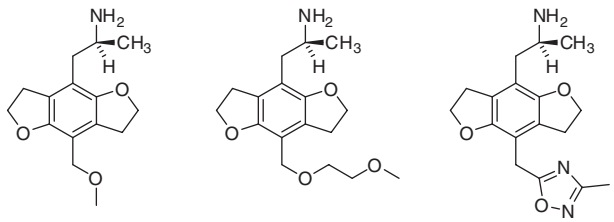
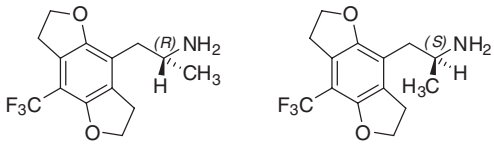


FIGURE 24 | Rigid difurano compounds with extended 4-substituents.



***R*-58** ***S*-58**

FIGURE 25 | Enantiomers of potent difurano 4-trifluoromethyl analogs.

enantiomer than for the *S*-(+) antipode. For DOB and DOI, the difference in EC50 was approximately fourfold higher for the *R* isomer than for the *S* isomer. With a CF_3 as the 4-substituent, however, and the 2- and 5-methoxy groups constrained into dihydrofuran rings [i.e., 'TFMFly' (*R*- or *S*-**58**)] (Figure 25), although the difference in *affinity* for the enantiomers was more than 40-fold, the difference in EC50 was less than twofold. The *R* enantiomers were all about 50% more potent than the *S* enantiomers in the PI turnover assay.

Parrish et al.[91] also examined these compounds at the rat 5-HT$_{2A}$ receptor, and although there were some minor differences, the overall trends were essentially the same. On the basis of molecular modeling studies, the authors speculated that in the *R* enantiomers, the α-methyl group interacted with Phe340$^{(6.52)}$ through van der Waals interactions, as a possible explanation for the higher potency and intrinsic activity of these isomers. With respect to utility as a radioligand, the α-methyl compound had no advantage over the simpler phenethylamines. That is, for use as a radioligand to label 5-HT$_{2A/2C}$ receptors, [^{125}I]2C-I gives results comparable to [^{125}I]DOI.[3]

Incorporation of the α-methyl into a cyclopropane ring gives substituted 2-phenylcyclopropylamines with high potency, both *in vitro* and *in vivo*. *In vivo* potency for the *cis*- and *trans*-cyclopropane analogs of mescaline was first reported by Cooper and Walters.[94,95] These workers found that the *trans* compound **59** produced effects in rodents that qualitatively resembled mescaline, with a potency somewhat greater than mescaline and a slightly shorter duration of action (Figure 26).

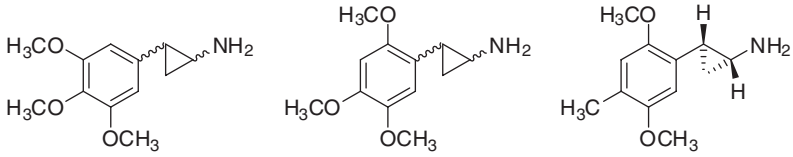
Aldous et al.[50] subsequently explored several cyclopropane analogs of substituted amphetamines. Although it was not then possible to measure receptor effects, production of hyperthermia in rabbits as well as producing changes in cat encephaolgram (EEG) were taken as indicators of possible hallucinogenic action, which we now know is correlated with actions at the 5-HT$_{2A}$ receptor. In particular, the *trans*-2,4,5-trimethoxy **60** and *trans*-2,5-dimethoxy-4-methyl compounds **61** had hallucinogen-like activity, with about 20 and 35%, respectively, of the activity of DOM.

A subsequent study of the enantiomers of the *trans*-cyclopropane analog of DOM, ((−)-**61** trans-2-(2,5-dimethoxy-4-methylphenyl)cyclopropylamine (DMCPA)), showed that in three behavioral responses DMCPA had activity nearly comparable to that produced by DOM.[96] The 1*R*,2*S*-(−)- isomer of DMCPA **61** was most potent, with the (+)-antipode being nearly inert. In the rabbit hyperthermia assay, both racemic and (−) DMCPA produced a robust response, with the effect of the (+) isomer not significantly different from saline.[97] Using [^{125}I]2C-I to label the 5-HT$_{2A/2C}$ receptor in rat cortical homogenate, the affinities of the 1*R*,2*S* and 1*S*,2*R* enantiomers were 2.2 and 21.6 nM, respectively. In the same study, the K_i of (*R*)-DOI was virtually identical (2.6 nM) to (−)-DMCPA, suggesting that the incorporation of the α-methyl into a cyclopropane ring increases affinity for this receptor.

Expansion of the cyclopropane ring to a cyclobutane, however, led to a dramatic 50- to 75-fold loss of *in vivo* activity,[98] and methyl group substituents on the cyclopropane ring were likewise not tolerated.[99] Coupled with a variety of other studies that examined conformationally-constrained analogs of phenethylamine type 5-HT$_{2A}$ agonists, one can conclude that the orthosteric binding site within the receptor is very sterically restricted around the side chain, and might best be viewed as a sort of slot into which the ligand must fit.

ATTEMPTS TO IDENTIFY THE 'ACTIVE' CONFORMATION OF THE PHENETHYLAMINES

The high activity of *trans*-2-phenylcyclopropylamines clearly indicated that the side chain of phenethylamines must exist in a *trans*-extended conformation. Early studies with 2-aminotetralin and indan derivatives had indicated that the side chain probably could not lie in the plane of the aromatic ring.[100–102] Additional evidence for the nature of the active conformation came with virtual docking of mescaline into a homology model of an in silico 'activated' model of the 5-HT$_{2A}$ receptor.[103] Upon docking, it was observed that tethering the side chain back to the ring to afford an aminomethyl compound would provide a compound that closely mimicked the virtually docked conformation of mescaline. The modeling further predicted the active absolute configuration of that molecule would be *R*, based on virtual docking of both enantiomers. Synthesis and resolution confirmed this prediction, with the *R* enantiomer **62** having a K_i at the cloned human 5-HT$_{2A}$ receptor of 70 nM, whereas the less active *S* enantiomer had a



59 60 (1*R*,2*S*)-(–)-**61**

FIGURE 26 | 2-Phenylcyclopropylamine analogs of phenethylamine agonists.

K_i of 1120 nM.[104] The *R* enantiomer had an EC50 of 3200 nM for activating IP3 accumulation, whereas the *S* enantiomer had an EC50 >50,000 nM. The *R* enantiomer also had efficacy in this pathway virtually identical to mescaline (Figure 27).

Applying this strategy to a compound with the more potent 2,4,5-trisubstitution pattern led to molecule **63**, which had slightly increased but modest affinity (47 nM) for the human 5-HT_{2A} receptor. Contracting the five-membered ring to a cyclobutene, however, gave highly potent **64**, (3-bromo-2,5-dimethoxybicyclo[4.2.0]octa-1,3,5-trien-7-yl)methanamine, known as TCB-2.[105] Virtual docking of this molecule to a homology model of the receptor also had predicted that the *R* enantiomer would be most active. Indeed, pharmacological evaluation revealed that the *R* benzocyclobutene **64** had a K_i of 0.26 nM, while the *S* isomer had a K_i of only 42 nM. The EC50 for IP3 accumulation with the *R* enantiomer was 18 nM, which had an intrinsic activity of 97%, making it a full agonist in this signaling pathway. The *S* enantiomer was much less potent, with an EC50 of only 460 nM.

When the benzocyclobutene enantiomers were tested in the two-lever drug discrimination assay in rats trained to discriminate either LSD or DOI from saline, the *R* enantiomer had potency comparable to each of the training drugs. In LSD-trained rats LSD had an ED50 of 38 nmol/kg, while the *R* enantiomer of the benzocyclobutene had an ED50 of 24 nmol/kg. No substitution was observed for the *S* enantiomer at doses up to 250 nmol/kg. These results strongly support the hypothesis that the side chain of the phenethylamine 5-HT_{2A} agonists/hallucinogens binds to the receptor in a conformation displaced out of the ring plane. Virtual docking studies of **64** into a homology model of the activated 5-HT_{2A} receptor,[105]

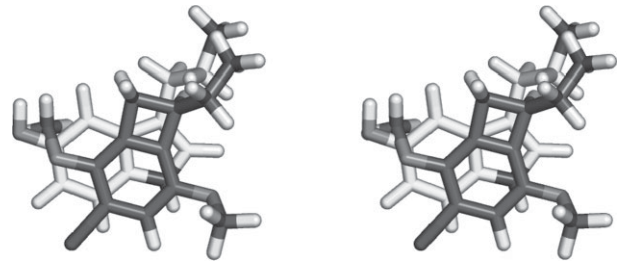
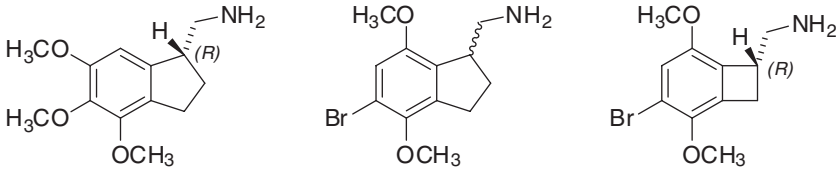


FIGURE 28 | Stereopair (cross-eyed) views of structures of serotonin (light gray) and **64** (dark gray) virtually docked into a homology model of the human 5-HT_{2A} (5-hydroxytryptamine) receptor. The extracellular face of the receptor faces upward. The 5-hydroxy and 5-methoxy moieties of serotonin and **64**, respectively, are to the left and are proposed to accept a hydrogen bond from Ser239.[80] The charged amino groups are at the top right, and engage Asp155 through an ionic salt bridge.

and a comparison with serotonin docked into the same aligned structure, illustrates the possible binding modes of the two different chemotypes of agonist ligands (Figure 28).

Effect of N-Alkylation

In contrast to the tryptamines, the phenethylamines generally cannot tolerate simple *N*-substitution, even with small groups such as methyl or ethyl (Table 2). Quite remarkably, however, whereas simple and short alkyl groups lead to inactive compounds, the addition of an *N*-benzyl affords compounds with remarkable affinity and potency.[106] Further, an oxygen atom on the ortho position of the *N*-benzyl group enhances the activity even further. This enhancement only occurred in compounds lacking the α-methyl in the side chain, i.e., phenethylamines. When an α-methyl was introduced, affinity dropped off by about 20-fold, indicating that whereas the α-methyl



62 63 64

FIGURE 27 | Constrained phenethylamines with activity at 5-HT_{2A} (5-hydroxytryptamine) receptors.

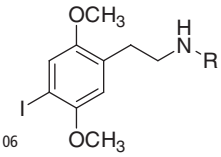
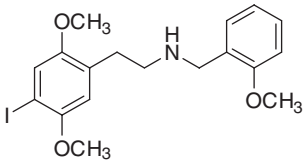


TABLE 2 | Effects of Phenethylamine *N*-Substituents on Affinity at Several Serotonin Receptor Subtypes[106]

R Substituent	Human 5-HT_{2A}	Rat 5-HT_{2A}	Human 5-HT_{2C}	Rat 5-HT_{2C}	Human 5-HT_{1A}
–H	0.73 ± 0.06	0.65 ± 0.07	1.82 ± 0.20	1.22 ± 0.03	123 ± 24
–CH_3	1907 ± 254	1286 ± 64	nd	206 ± 34	247 ± 23
–*n*-Pr	1295 ± 151	734 ± 30	nd	656 ± 127	879 ± 64
–Benzyl	0.25 ± 0.05	0.31 ± 0.03	1.08 ± 0.24	1.15 ± 0.90	2205 ± 106
–BOMe[1]	0.044 ± 0.006	0.09 ± 0.010	0.43 ± 0.08	0.13 ± 0.02	1696 ± 311

[1] *N*-(ortho-methoxybenzyl).



65

FIGURE 29 | An example of a potent *N*-benzylphenethylamine.

enhanced potency in simple phenethylamines, it was deleterious when present in an *N*-benzylphenethylamine. The most well-studied of these compounds is 2CINBOMe **65** (Figure 29).

The extremely high affinity of the *N*-benzyl compounds has made them useful both as radioligands for receptor binding studies,[107] and as [^{11}C]-labeled PET ligands for *in vivo* imaging studies.[108] It appears that an aryl group with a hydrogen bond *acceptor*, such as an ether, gives highest activity, as a comparison between the ortho-OH- and an ortho-OCH_3- substituted compounds clearly shows. Additional examples of modification to the *N*-benzyl moiety are illustrated in Tables 3 and 4. On the basis of the number of very potent-substituted phenethylamines known, there are likely a number of quite potent compounds yet to be identified by the simple addition of an *N*-benzyl moiety.

A number of similar *N*-alkyated molecules in this series may have potential as [^{3}H] radioligands for *in vitro* studies, or as [^{11}C]-labeled derivatives for possible use as positron emission tomography

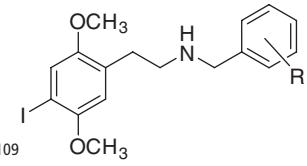


TABLE 3 | Effect of the Benzyl Group Substitution on Affinity of *N*-Benzylphenethylamines[109]

R	h5-HT_{2A}	r5-HT_{2A}	h5-HT_{2C}	r5-HT_{2C}	h5-HT_{1A}
2-OCH_3	0.044 ± 0.006	0.09 ± 0.01	0.43 ± 0.08	0.13 ± 0.02	1696 ± 311
2-OH	0.061 ± 0.012	0.12 ± 0.02	0.13 ± 0.01	0.21 ± 0.02	2749 ± 210
2-CN	nd	276 ± 65	23.2 ± 4.1	nd	nd
2-$CONH_2$	1.18 ± 0.22	0.84 ± 0.1	nd	0.73 ± 0.09	nd
2-CH_2OH	0.79 ± 0.05	0.44 ± 0.03	nd	0.43 ± 0.01	nd
2-CF_3	1.31 ± 0.15	nd	nd	nd	nd
4-CF_3	205 ± 44	nd	nd	nd	nd
2-F	0.26 ± 0.05	0.28 ± 0.04	2.36 ± 0.41	0.85 ± 0.11	3803 ± 170
4-F	37.3 ± 6.0	nd	nd	nd	nd
2,3-OCH_2O	0.049 ± 0.008	0.193 ± 0.022	1.7 ± 0.23	0.41 ± 0.07	971 ± 69
3,4-OCH_2O	0.69 ± 0.05	nd	nd	nd	nd
2-OH-4,5-OCH_2O	0.82 ± 0.17	nd	nd	nd	nd

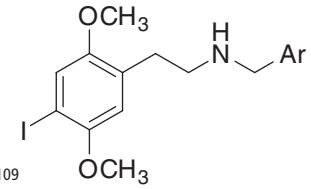


TABLE 4 | Effect of Other *N*-Aryl Groups on Phenethylamines[109]

Ar	h5-HT_{2A}	r5-HT_{2A}	h5-HT_{2C}	r5-HT_{2C}	h5-HT_{1A}
2-Furyl	nd	0.78 ± 0.12	nd	0.99 ± 0.1	nd
2-Thienyl	1.02 ± 0.09	0.45 ± 0.09	nd	0.59 ± 0.06	nd
2-Pyridyl	nd	3.45 ± 0.7	nd	5.81 ± 1.14	nd
3-Indolyl	nd	2.67 ± 0.49	nd	11.8 ± 1.7	nd
1-Naphthyl	nd	1.07 ± 0.11	nd	14.0 ± 1.5	nd
2-Naphthyl	4.83 ± 0.55	3.74 ± 0.52	38.9 ± 7.6	176 ± 30	1137 ± 152
2,3-Dihydrobenzofuran-7-yl	0.026 ± 0.006	nd	1.03 ± 0.15	nd	nd
1-OH-2-naphthyl	0.45 ± 0.06	nd	nd	nd	nd
3-OH-2-naphthyl	664 ± 18	nd	nd	nd	nd
2-OH-1-naphthyl	3552 ± 3207	nd	nd	nd	nd

(PET) tracers for central 5-HT_{2A} receptors. The first such molecule to be studied for these uses was **65**, discussed earlier.[107,108] More recently, a number of related [^{11}C]-labeled molecules, including analogs with aromatic Cl, CF_3, and especially Br substituents, have been reported to have excellent potential as PET ligands.[110]

RECEPTOR MODELS

As a member of the family A, G protein coupled receptors (GPCRs), there has been a recent work to develop an in silico-activated model of the human 5-HT_{2A} receptor. The earliest model was proposed by Chambers and Nichols,[78] developed from the crystal structure of bovine rhodopsin. The *cis*-retinal chromophore was isomerized in silico to the all-*trans* form of retinal, followed by rigid-body dynamics. Construction of a homology model of the 5-HT_{2A} receptor from this in silico-activated rhodopsin template, and virtual docking with various 5-HT_{2A} agonist ligands, allowed the formulation of a number of hypothesis about the functional topography of the ligand binding site, which were subsequently validated by site-directed mutagenesis and ligand testing.[80,106] In addition, virtual docking studies allowed the design of new agonist ligands as well as the prediction of their more active enantiomers.[104,105] More recently, Isberg et al.[79] have developed an in silico-activated model of the 5-HT_{2A} receptor starting with the published crystal structure of the β-2-adrenergic receptor. Continued refinements of this, and other G protein coupled receptors, will ultimately allow a greater understanding of the molecular features that lead to receptor binding and activation. This knowledge, in turn, should result in the ability to carry out structure-based ligand design or true *de novo* drug discovery for the particular receptor.

CONCLUSION

The structure–activity relationships of serotonin 5-HT_{2A} receptor agonists are now fairly well developed for three major chemotypes of ligands. Much of the work was initially driven by an attempt to explain the activity of hallucinogenic (psychedelic) agents in man. Hallucinogens profoundly affect consciousness, and obviously, wherever the hallucinogens exerted their effects in the brain was of considerable importance. Such an understanding certainly had philosophical implications, but gained practical importance after it was shown that the 5-HT_{2A} receptor was a target for atypical antipsychotic agents,[111] and was also involved in working memory processes.[112] Early attempts were made to identify structural similarities between phenethylamines and tryptamines that might account for the ability of phenethylamines to interact with the 5-HT_{2A} receptor. But, as this review has shown, there is little similarity between them other than that the two series are arylethylamines, each with specific molecular features that allow receptor recognition. Intriguing questions remain, however. For example, why is LSD such a potent hallucinogen when it has rather unremarkable *in vitro* agonist properties?

Further, the variation in activity of substituted lysergic acid amides suggests that the receptor is very sensitive to the nature of the amide in a way that cannot yet be explained. Finally, for 5-HT_{2A} agonist molecules that have been studied in man, the psychopharmacology can vary widely. Recently, it has been realized that receptors can couple to multiple effectors, and that different agonists can produce different biochemical endpoints. Thus, it is likely that specific agonists with particular substitution patterns may be able selectively to activate a subset of effectors, a phenomenon now known as functional selectivity.[113] Such actions may partially account for the different CNS effects of particular '5-HT_{2A} agonists', but complicate the understanding of agonist pharmacology, leading to a virtually open-ended question to correlate psychopharmacology with agonist activity.

REFERENCES

1. Shulgin AT, Shulgin A. *TIHKAL. The Continuation*. Berkeley, CA: Transform Press; 1997.
2. Gupta SK, Diez E, Heasley LE, Osawa S, Johnson GL. A G protein mutant that inhibits thrombin and purinergic receptor activation of phospholipase A2. *Science* 1990, 249:662–666.
3. Johnson MP, Mathis CA, Shulgin AT, Hoffman AJ, Nichols DE. [125I]-2-(2,5-dimethoxy-4-iodophenyl)aminoethane ([125I]-2C-I) as a label for the 5-HT2 receptor in rat frontal cortex. *Pharmacol Biochem Behav* 1990, 35:211–217.
4. McKenna DJ, Repke DB, Lo L, Peroutka SJ. Differential interactions of indolealkylamines with 5-hydroxytryptamine receptor subtypes. *Neuropharmacology* 1990, 29:193–198.
5. Kalir A, Szara S. Synthesis and pharmacological activity of fluorinated tryptamine derivatives. *J Med Chem* 1963, 6:716–719.
6. Blair JB, Kurrasch-Orbaugh D, Marona-Lewicka D, Cumbay MG, Watts VJ, Barker EL, Nichols DE. Effect of ring fluorination on the pharmacology of hallucinogenic tryptamines. *J Med Chem* 2000, 43:4701–4710.
7. Laban U, Kurrasch-Orbaugh D, Marona-Lewicka D, Nichols DE. A novel fluorinated tryptamine with highly potent serotonin 5-HT1A receptor agonist properties. *Bioorg Med Chem Lett* 2001, 11:793–795.
8. Winter JC, Gessner PK, Godse DD. Synthesis of some 3-indenealkylamines. Comparison of the biological activity of 3-indenealkylamines and 3-benzo[b]thiophenealkylamines with their tryptamine isosteres. *J Med Chem* 1967, 10:856–858.
9. Blair JB, Marona-Lewicka D, Kanthasamy A, Lucaites VL, Nelson DL, Nichols DE. Thieno[3,2-b]- and thieno[2,3-b]pyrrole bioisosteric analogues of the hallucinogen and serotonin agonist N,N-dimethyltryptamine. *J Med Chem* 1999, 42:1106–1111.
10. Tomaszewski Z, Johnson MP, Huang X, Nichols DE. Benzofuran bioisosteres of hallucinogenic tryptamines. *J Med Chem* 1992, 35:2061–2064.
11. May JA, Chen HH, Rusinko A, Lynch VM, Sharif NA, McLaughlin MA. A novel and selective 5-HT2 receptor agonist with ocular hypotensive activity: (S)-(+)-1-(2-aminopropyl)-8,9-dihydropyrano [3,2-e] indole. *J Med Chem* 2003, 46:4188–4195.
12. May JA, Dantanarayana AP, Zinke PW, McLaughlin MA, Sharif NA. 1-((S)-2-aminopropyl)-1H-indazol-6-ol: a potent peripherally acting 5-HT2 receptor agonist with ocular hypotensive activity. *J Med Chem* 2006, 49:318–328.
13. May JA, Sharif NA, Chen HH, Liao JC, Kelly CR, Glennon RA, Young R, Li JX, Rice KC, France CP. Pharmacological properties and discriminative stimulus effects of a novel and selective 5-HT2 receptor agonist AL-38022A [(S)-2-(8,9-dihydro-7H-pyrano[2,3-g]indazol-1-yl)-1-methylethylamine]. *Pharmacol Biochem Behav* 2009, 91:307–314.
14. Vangveravong S, Kanthasamy A, Lucaites VL, Nelson DL, Nichols DE. Synthesis and serotonin receptor affinities of a series of trans-2-(indol-3-yl)cyclopropylamine derivatives. *J Med Chem* 1998, 41:4995–5001.
15. Nichols DE, Lloyd DH, Johnson MP, Hoffman AJ. Synthesis and serotonin receptor affinities of a series of enantiomers of α-methyltryptamines: evidence for the binding conformation of tryptamines at serotonin 5-HT1B receptors. *J Med Chem* 1988, 31:1406–1412.
16. Shulgin A, Shulgin A. *PIHKAL A Chemical Love Story*. Berkeley, CA: Transform Press; 1991.
17. Daldrup T, Heller C, Matthiesen U, Honus S, Bresges A, Haarhoff K. Etryptamine, a new designer drug with a fatal effect. *Z Rechtsmed* 1986, 97:61–68.
18. Huang XM, Johnson MP, Nichols DE. Reduction in brain serotonin markers by α-ethyltryptamine (Monase). *Eur J Pharmacol* 1991, 200:187–190.
19. Krebs KM, Geyer MA. Behavioral characterization of α-ethyltryptamine, a tryptamine derivative with MDMA-like properties in rats. *Psychopharmacology (Berl)* 1993, 113:284–287.
20. Schechter MD. MDMA-like stimulus effects of hallucinogens in male Fawn-Hooded rats. *Pharmacol Biochem Behav* 1998, 59:265–270.

21. Brimblecombe RW, Pinder RM. *Hallucinogenic Agents*. Bristol: Wright-Scientechnica; 1975.
22. Nichols DE. Hallucinogens. In: Verderame M, ed. *Handbook of CNS Agents and Local Anesthetics*. Boca Raton: CRC Press Inc.; 1986, 303–325.
23. Shulgin AT. Chemistry of psychotomimetics. In: Hoffmeister F, Stille G, eds. *Handbook of Experimental Pharmacology*. New York: Springer-Verlag; 1982, 3–29.
24. Siva Sankar DV. *LSD: A Total Study*. Westbury, NY: PJD Publications; 1975.
25. Bennett JP Jr, Snyder, SH. Serotonin and lysergic acid diethylamide binding in rat brain membranes: relationship to postsynaptic serotonin receptors. *Mol Pharmacol* 1976, 12:373–389.
26. Ginzel KH, Mayer-Gross W. Prevention ofpsychological effects of d-lysergic acid diethylamide (LSD 25) by its 2-brom derivative (BOL 148). *Nature* 1956, 178:210.
27. McKenna DJ, Nazarali AJ, Hoffman AJ, Nichols DE, Mathis CA, Saavedra JM. Common receptors for hallucinogens in rat brain: a comparative autoradiographic study using [125I]LSD and [125I]DOI, a new psychotomimetic radioligand. *Brain Res* 1989, 476:45–56.
28. Nakada MT, Wieczorek CM, Rainbow TC. Localization and characterization by quantitative autoradiography of [125I]LSD binding sites in rat brain. *Neurosci Lett* 1984, 49:13–18.
29. Watts SW, Gackenheimer SL, Gehlert DR, Cohen ML. Autoradiographic comparison of [125I]LSD-labeled 5-HT2A receptor distribution in rat and guinea pig brain. *Neurochem Int* 1994, 24:565–574.
30. Hartig PR, Kadan MJ, Evans MJ, Krohn AM. 125I-LSD: a high sensitivity ligand for serotonin receptors. *Eur J Pharmacol* 1983, 89:321–322.
31. Hofmann A. Psychotomimetic agents. In: Burger A, ed. *Drugs Affecting the Central Nervous System*. London: Edward Arnold; 1968, 169.
32. Stoll A, Hofmann A. Amide der sterreoisomeren lysergsauren und dihydro-lysergsauren. *Helv Chim Acta* 1955, 38:421–433.
33. Clare BW. A novel quantum theoretic QSAR for hallucinogenic tryptamines: a major factor is the orientation of [pi] orbital nodes. *J Mol Struct: THEOCHEM* 2004, 712:143–148.
34. Gorodetzky CW, Isbell H. A comparison of 2,3-dihydro-lysergic acid diethylamide with LSD-25. *Psychopharmacologia* 1964, 6:229–233.
35. Hoffman AJ, Nichols DE. Synthesis and LSD-like discriminative stimulus properties in a series of N(6)-alkyl norlysergic acid N,N-diethylamide derivatives. *J Med Chem* 1985, 28:1252–1255.
36. Hofmann A. Teonanacatl and Ololuiqui, two ancient magic drugs of Mexico. *Bull Narcotics* 1971, XXIII:3–14.
37. Bach NJ, Hall DA, Kornfeld EC. Descarboxylysergic acid (9,10-didehydro-6-methylergoline). *J Med Chem* 1974, 17:312–314.
38. Oberlender R, Pfaff RC, Johnson MP, Huang XM, Nichols DE. Stereoselective LSD-like activity in d-lysergic acid amides of (R)- and (S)-2-aminobutane. *J Med Chem* 1992, 35:203–211.
39. Monte AP, Marona-Lewicka D, Kanthasamy A, Sanders-Bush E, Nichols DE. Stereoselective LSD-like activity in a series of d-lysergic acid amides of (R)- and (S)-2-aminoalkanes. *J Med Chem* 1995, 38:958–966.
40. Nichols DE, Frescas S, Marona-Lewicka D, Kurrasch-Orbaugh DM. Lysergamides of isomeric 2,4-dimethylazetidines map the binding orientation of the diethylamide moiety in the potent hallucinogenic agent N,N-diethyllysergamide (LSD). *J Med Chem* 2002, 45:4344–4349.
41. Juncosa JI, Organic synthesis combined with molecular modeling: a powerful approach to map the functional topography of dopamine and serotonin receptors. 2011. Purdue University.
42. Parrish JC. Toward a molecular understanding of hallucinogen action. 2006. Purdue University.
43. Nichols DE. Structure-activity relationships of phenethylamine hallucinogens. *J Pharm Sci* 1981, 70:839–849.
44. Blaazer AR, Smid P, Kruse CG. Structure-activity relationships of phenylalkylamines as agonist ligands for 5-HT(2A) receptors. *Chem Med Chem* 2008, 3:1299–1309.
45. Hey P. The synthesis of a new homolog of mescaline. *Quart J Pharm Pharmacol* 1947, 20:129–134.
46. Peretz DI, Smythies JR, Gibson WC. A new hallucinogen: 3,4,5-trimethoxyphenyl-β-aminopropane (with notes on a stroboscopic phenomenon). *J Ment Sci* 1955, 101:317–329.
47. Shulgin AT, Sargent T, Naranjo C. Structure–activity relationships of one-ring psychotomimetics. *Nature* 1969, 221:537–541.
48. Shulgin AT. Psychotomimetic drugs: structure-activity relationships. In: Iversen LL, Iversen SD, Snyder SH, eds. *Handbook of Psychopharmacology*. New York: Plenum Press; 1978, 243–333.
49. Nichols DE, Barfknecht CF, Rusterholz DB, Benington F, Morin RD. Asymmetric synthesis of psychotomimetic phenylisopropylamines. *J Med Chem* 1973, 16:480–483.
50. Aldous FA, Barrass BC, Brewster K, Buxton DA, Green DM, Pinder RM, Rich P, Skeels M, Tutt KJ. Structure-activity relationships in psychotomimetic phenylalkylamines. *J Med Chem* 1974, 17:1100–1111.
51. Cheng HC, Long JP, Nichols DE, Barfknecht CF. Effects of psychotomimetics on vascular strips: studies of methoxylated amphetamines and optical isomers

of 2,5-dimethoxy-4-methylamphetamine and 2,5-dimethoxy-4-bromoamphetamine. *J Pharmacol Exp Ther* 1974, 188:114–123.

52. Glennon RA, Bondarev ML, Khorana N, Young R, May JA, Hellberg MR, McLaughlin MA, Sharif NA. β-oxygenated analogues of the 5-HT2A serotonin receptor agonist 1-(4-bromo-2,5-dimethoxyphenyl)-2-aminopropane. *J Med Chem* 2004, 47:6034–6041.
53. Lemaire D, Jacob P III, Shulgin AT. Ring-substituted β-methoxyphenethylamines: a new class of psychotomimetic agents active in man. *J Pharm Pharmacol* 1985, 37:575–577.
54. Glennon RA, Young R, Rosecrans JA. Discriminative stimulus properties of DOM and several molecular modifications. *Pharmacol Biochem Behav* 1982, 16:553–556.
55. Nichols DE, Schooler D, Yeung MC, Oberlender RA, Zabik JE. Unreliability of the rat stomach fundus as a predictor of hallucinogenic activity in substituted phenethylamines. *Life Sci* 1984, 35:1343–1348.
56. Shulgin AT. The ethyl homologs of 2,4,5-trimethoxyphenylisopropylamine. *J Med Chem* 1968, 11:186–187.
57. Shulgin AT, Sargent T, Naranjo C. 4-Bromo-2,5-dimethoxyphenylisopropylamine, a new centrally active amphetamine analog. *Pharmacology* 1971, 5:103–107.
58. Nichols DE, Shulgin AT. Sulfur analogs of psychotomimetic amines. *J Pharm Sci* 1976, 65:1554–1556.
59. Nichols DE, Frescas S, Marona-Lewicka D, Huang X, Roth BL, Gudelsky GA, Nash JF. 1-(2,5-Dimethoxy-4-(trifluoromethyl)phenyl)-2-aminopropane: a potent serotonin 5-HT2A/2C agonist. *J Med Chem* 1994, 37:4346–4351.
60. Johnson MP, Hoffman AJ, Nichols DE, Mathis CA. Binding to the serotonin 5-HT2 receptor by the enantiomers of 125I-DOI. *Neuropharmacology* 1987, 26:1803–1806.
61. Braun U, Shulgin AT, Braun G, Sargent T. Synthesis III and body distribution of several iodine-131 labeled centrally acting drugs. *J Med Chem* 1977, 20:1543–1546.
62. Sargent T III, Kalbhen DA, Shulgin AT, Stauffer H, Kusubov N. A potential new brain-scanning agent: 4-77Br-2,5-dimethoxyphenylisoproplamine (4-Br-DPIA). *J Nucl Med* 1975, 16:243–245.
63. Jacob P III, Anderson G, Meshul CK III, Shulgin AT, Castagnoli N Jr. Monomethylthio analogues of 1-(2,4,5-trimethoxyphenyl)-2-aminopropane. *J Med Chem* 1977, 20:1235–1239.
64. Jacob P III, Shulgin A T. Sulfur analogues of psychotomimetic agents. 2. Analogues of (2,5-dimethoxy-4-methylphenyl)-and (2,5-dimethoxy-4-ethylphenyl)isopropylamine. *J Med Chem* 1983, 26:746–752.
65. Jacob P III, Shulgin AT. Sulfur analogues of psychotomimetic agents. Monothio analogues of mescaline and isomescaline. *J Med Chem* 1981, 24: 1348–1353.
66. Barfknecht CF, Nichols DE, Dunn WJ III. Correlation of psychotomimetic activity of phenethylamines and amphetamines with 1-octanol-water partition coefficients. *J Med Chem* 1975, 18:208–210.
67. Nichols DE, Dyer DC. Lipophilicity and serotonin agonist activity in a series of 4- substituted mescaline analogues. *J Med Chem* 1977, 20:299–301.
68. Nichols DE, Shulgin AT, Dyer DC. Directional lipophilic character in a series of psychotomimetic phenethylamine derivatives. *Life Sci* 1977, 21:569–575.
69. Seggel MR, Yousif MY, Lyon RA, Titeler M, Roth BL, Suba EA, Glennon RA. A structure-affinity study of the binding of 4-substituted analogues of 1-(2,5-dimethoxyphenyl)-2-aminopropane at 5-HT2 serotonin receptors. *J Med Chem* 1990, 33:1032–1036.
70. Oberlender R, Ramachandran PV, Johnson MP, Huang X, Nichols DE. Effect of a chiral 4-alkyl substituent in hallucinogenic amphetamines. *J Med Chem* 1995, 38:3593–3601.
71. Oberlender RA, Kothari PJ, Nichols DE, Zabik JE. Substituent branching in phenethylamine-type hallucinogens: a comparison of 1-[2,5-dimethoxy-4-(2-butyl)phenyl]-2-aminopropane and 1-[2,5-dimethoxy-4-(2-methylpropyl)phenyl]-2-aminopropane. *J Med Chem* 1984, 27:788–792.
72. Glennon RA, Young R, Rosecrans JA. A comparison of the behavioral effects of DOM homologs. *Pharmacol Biochem Behav* 1982, 16:557–559.
73. Glennon RA, Rosecrans JA. Indolealkylamine and phenalkylamine hallucinogens: a brief overview. *Neurosci Biobehav Rev* 1982, 6:489–497.
74. Glennon RA, Doot DL, Young R. DOM and related 2,5-dimethoxy-4-alkylphenylisopropylamines: behavioral and serotonin receptor properties. *Pharmacol Biochem Behav* 1981, 14:287–292.
75. Trachsel D, Nichols DE, Kidd S, Hadorn M, Baumberger F. 4-aryl-substituted 2,5-dimethoxyphenethylamines: synthesis and serotonin 5-HT(2A) receptor affinities. *Chem Biodivers* 2009, 6:692–704.
76. Dowd CS, Herrick-Davis K, Egan C, Dupre A, Smith C, Teitler M, Glennon RA. 1-[4-(3-Phenylalkyl) phenyl]-2-aminopropanes as 5-HT(2A) partial agonists. *J Med Chem* 2000, 43:3074–3084.
77. Rangisetty JB, Dukat M, Dowd CS, Herrick-Davis K, Dupre A, Gadepalli S, Teitler M, Kelley CR, Sharif NA, Glennon RA. 1-[2-methoxy-5-(3-phenylpropyl)]-2-aminopropane unexpectedly shows 5-HT(2A) serotonin receptor affinity and antagonist character. *J Med Chem* 2001, 44:3283–3291.
78. Chambers JJ, Nichols DE. A homology-based model of the human 5-HT2A receptor derived from an in

silico activated G-protein coupled receptor. *J Comput Aided Mol Des* 2002, 16:511–520.

79. Isberg V, Balle T, Sander T, Jorgensen FS, Gloriam DE. G protein- and agonist-bound serotonin 5-HT(2A) receptor model activated by steered molecular dynamics simulations. *J Chem Inf Model* 2011, 51:315–325.
80. Braden MR, Nichols DE. Assessment of the roles of serines 5.43(239) and 5.46(242) for binding and potency of agonist ligands at the human serotonin 5-HT2A receptor. *Mol Pharmacol* 2007, 72:1200–1209.
81. Nichols DE, Hoffman AJ, Oberlender RA, Riggs RM. Synthesis and evaluation of 2,3-dihydrobenzofuran analogues of the hallucinogen 1-(2,5-dimethoxy-4-methylphenyl)-2-aminopropane: drug discrimination studies in rats. *J Med Chem* 1986, 29:302–304.
82. Nichols DE, Snyder SE, Oberlender R, Johnson MP, Huang XM. 2,3-Dihydrobenzofuran analogues of hallucinogenic phenethylamines. *J Med Chem* 1991, 34:276–281.
83. Monte AP, Marona-Lewicka D, Parker MA, Wainscott DB, Nelson DL, Nichols DE. Dihydrobenzofuran analogues of hallucinogens. 3. Models of 4-substituted (2,5-dimethoxyphenyl)alkylamine derivatives with rigidified methoxy groups. *J Med Chem* 1996, 39:2953–2961.
84. Chambers JJ, Kurrasch-Orbaugh DM, Parker MA, Nichols DE. Enantiospecific synthesis and pharmacological evaluation of a series of super-potent, conformationally restricted 5-HT(2A/2C) receptor agonists. *J Med Chem* 2001, 44:1003–1010.
85. Parker MA, Marona-Lewicka D, Lucaites VL, Nelson DL, Nichols DE. A novel (benzodifuranyl) aminoalkane with extremely potent activity at the 5-HT2A receptor. *J Med Chem* 1998, 41:5148–5149.
86. Schultz DM, Prescher JA, Kidd S, Marona-Lewicka D, Nichols DE, Monte A. 'Hybrid' benzofuran-benzopyran congeners as rigid analogs of hallucinogenic phenethylamines. *Bioorg Med Chem* 2008, 16: 6242–6251.
87. Chambers JJ, Kurrasch-Orbaugh DM, Nichols DE. Translocation of the 5-alkoxy substituent of 2,5-dialkoxyarylalkylamines to the 6-position: effects on 5-HT(2A/2C) receptor affinity. *Bioorg Med Chem Lett* 2002, 12:1997–1999.
88. Monte AP, Waldman SR, Marona-Lewicka D, Wainscott DB, Nelson DL, Sanders-Bush E, Nichols DE. Dihydrobenzofuran analogues of hallucinogens. 4. Mescaline derivatives. *J Med Chem* 1997, 40: 2997–3008.
89. May JA, McLaughlin MA, Sharif NA, Hellberg MR, Dean TR. Evaluation of the ocular hypotensive response of serotonin 5-HT1A and 5-HT2 receptor ligands in conscious ocular hypertensive cynomolgus monkeys. *J Pharmacol Exp Ther* 2003, 306:301–309.
90. Feng Z, Mohapatra S, Klimko PG, Hellberg MR, May JA, Kelly C, Williams G, McLaughlin MA, Sharif NA. Novel benzodifuran analogs as potent 5-HT2A receptor agonists with ocular hypotensive activity. *Bioorg Med Chem Lett* 2007, 17:2998–3002.
91. Parrish JC, Braden MR, Gundy E, Nichols DE. Differential phospholipase C activation by phenylalkylamine serotonin 5-HT 2A receptor agonists. *J Neurochem* 2005, 95:1575–1584.
92. Nash JF, Roth BL, Brodkin JD, Nichols DE, Gudelsky GA. Effect of the R(-) and S(+) isomers of MDA and MDMA on phosphatidyl inositol turnover in cultured cells expressing 5-HT2A or 5-HT2C receptors. *Neurosci Lett* 1994, 177:111–115.
93. Glennon RA, Teitler M, Sanders-Bush E. Hallucinogens and serotonergic mechanisms. *NIDA Res Monogr* 1992, 119:131–135.
94. Cooper PD, Walters GC. Stereochemical requirements of the mescaline receptor. *Nature* 1972, 238:96–98.
95. Walters GC, Cooper PD. Alicyclic analogue of mescaline. *Nature* 1968, 218:298–300.
96. Nichols DE, Pfister WR, Yim GK. LSD and phenethylamine hallucinogens: new structural analogy and implications for receptor geometry. *Life Sci* 1978, 22:2165–2170.
97. Nichols DE, Woodard R, Hathaway BA Lowy MT, Yom KW. Resolution and absolute configuration of trans-2-(2,5-dimethoxy-4- methylphenyl) cyclopropylamine, a potent hallucinogen analogue. *J Med Chem* 1979, 22:458–460.
98. Nichols DE, Jadhav KP, Oberlender RA, Zabik JE, Bossart JF, Hamada A, Miller DD. Synthesis and evaluation of substituted 2-phenylcyclobutylamines as analogues of hallucinogenic phenethylamines: lack of LSD-like biological activity. *J Med Chem* 1984, 27:1108–1111.
99. Jacob JN, Nichols DE. Isomeric cyclopropyl ring-methylated homologues of trans-2-(2,5-dimethoxy-4-methylphenyl)cyclopropylamine, an hallucinogen analogue. *J Med Chem* 1982, 25:526–530.
100. Nichols DE, Barfknecht CF, Long JP, Standridge RT, Howell HG, Partyka RA, Dyer DC. Potential psychotomimetics. 2. Rigid analogs of 2,5-dimethoxy-4-methylphenylisopropylamine (DOM, STP). *J Med Chem* 1974, 17:161–166.
101. Coutts RT, Malicky JL. The synthesis of analogs of the hallucinogen 1-(2,5-dimethoxy-4-methylphenyl)-2-aminopropane (DOM). II. Some ring-methoxylated 1-amino-and 2-aminoindanes. *Can J Chem* 1974, 52:381–389.
102. Monte AP, Marona-Lewicka D, Lewis MM, Mailman RB, Wainscott DB, Nelson DL, Nichols DE. Substituted naphthofurans as hallucinogenic phenethylamine-ergoline hybrid molecules with unexpected muscarinic antagonist activity. *J Med Chem* 1998, 41:2134–2145.

103. Chambers JJ, Nichols DE. A Model of the Human 5-HT_{2A} Receptor Based on Constrained Homology. Great Lakes/Central Regional, American Chemical Society Meeting, 2001.
104. McLean TH, Chambers JJ, Parrish JC, Braden MR, Marona-Lewicka D, Kurrasch-Orbaugh D, Nichols DE. C-(4,5,6-trimethoxyindan-1-yl)methanamine: a mescaline analogue designed using a homology model of the 5-HT2A receptor. *J Med Chem* 2006, 49:4269–4274.
105. McLean TH, Parrish JC, Braden MR, Marona-Lewicka D, Gallardo-Godoy A, Nichols DE. 1-Aminomethylbenzocycloalkanes: conformationally restricted hallucinogenic phenethylamine analogues as functionally selective 5-HT2A receptor agonists. *J Med Chem* 2006, 49:5794–5803.
106. Braden MR, Parrish JC, Naylor JC, Nichols DE. Molecular interaction of serotonin 5-HT2A receptor residues Phe339(6.51) and Phe340(6.52) with superpotent N-benzyl phenethylamine agonists. *Mol Pharmacol* 2006, 70:1956–1964.
107. Nichols DE, Frescas SP, Chemel BR, Rehder KS, Zhong D, Lewin AH. High specific activity tritium-labeled N-(2-methoxybenzyl)-2,5-dimethoxy-4-iodophenethylamine (INBMeO): a high-affinity 5-HT2A receptor-selective agonist radioligand. *Bioorg Med Chem* 2008, 16:6116–6123.
108. Ettrup A, Palner M, Gillings N, Santini MA, Hansen M, Kornum BR, Rasmussen LK, Nagren K, Madsen J, Begtrup M, et al. Radiosynthesis and evaluation of 11C-CIMBI-5 as a 5-HT2A receptor agonist radioligand for PET. *J Nucl Med* 2010, 51:1763–1770.
109. Braden MR., Towards a biophysical understanding of hallucinogen action. 2007. Purdue University. 4-10-2007.
110. Ettrup A, Hansen M, Santini MA, Paine J, Gillings N, Palner M, Lehel S, Herth MH, Madsen J, Kristensen J, et al. Radiosynthesis and in vivo evaluation of a series of substituted ^{11}C-phenethylamines as 5-HT_{2A} agonist PET tracers. *Eur J Nucl Med Mol Imaging* 2011, 38:681–693.
111. Meltzer HY. The role of serotonin in schizophrenia and the place of serotonin-dopamine antagonist antipsychotics. *J Clin Psychopharmacol* 1995, 15:2S–3S.
112. Williams GV, Rao SG, Goldman-Rakic PS. The physiological role of 5-HT2A receptors in working memory. *J Neurosci* 2002, 22:2843–2854.
113. Urban JD, Clarke WP, von Zastrow M, Nichols DE, Kobilka B, Weinstein H, Javitch JA, Roth BL, Christopoulos A, Sexton PM, et al. Functional selectivity and classical concepts of quantitative pharmacology. *J Pharmacol Exp Ther* 2007, 320:1–13.