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DIFFERENTIAL INTERACTIONS OF DIMETHYLTRYPTAMINE (DMT) WITH 5-HT_{1A} AND 5-HT_2 RECEPTORS

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Abstract—The interactions of the indolealkylamine *N,N*-dimethyltryptamine (DMT) with 5-hydroxytryptamine$_{1A}$ (5-HT_{1A}) and 5-HT_2 receptors in rat brain were analyzed using radioligand binding techniques and biochemical functional assays. The affinity of DMT for 5-HT_{1A} sites labeled by [^{3}H]-8-hydroxy-2-(di-*n*-propylamino)tetralin ([^{3}H]-8-OH-DPAT) was decreased in the presence of 10^{-4} M GTP, suggesting agonist activity of DMT at this receptor. Adenylate cyclase studies in rat hippocampi showed that DMT inhibited forskolin-stimulated cyclase activity, a 5-HT_{1A} agonist effect. DMT displayed full agonist activity with an EC_{50} of 4×10^{-6} M in the cyclase assay. In contrast to the agonist actions of DMT at 5-HT_{1A} receptors, DMT appeared to have antagonistic properties at 5-HT_2 receptors. The ability of DMT to compete for [^{3}H]-ketanserin-labeled 5-HT_2 receptors was not affected by the presence of 10^{-4} M GTP, suggesting antagonist activity of DMT at 5-HT_2 receptors. In addition, DMT antagonized 5-HT_2-receptor-mediated phosphatidylinositol (PI) turnover in rat cortex at concentrations above 10^{-7} M, with 70% of the 5-HT-induced PI response inhibited at 10^{-4} M DMT. Micromolar concentrations of DMT produced a slight PI stimulation that was not blocked by the 5-HT_2 antagonist ketanserin. These studies suggest that DMT has opposing actions on 5-HT receptor subtypes, displaying agonist activity at 5-HT_{1A} receptors and antagonist activity at 5-HT_2 receptors.

The presence of multiple 5-hydroxytryptamine (5-HT) receptors in the central nervous system is now well established [1, 2]. In general, tryptamine derivatives such as 5-HT display agonist effects at all known 5-HT receptor subtypes [3–5]. On the other hand, ergot derivatives such as methysergide, metergoline, lisuride and *d*-lysergic acid diethylamide (*d*-LSD) have been reported to display differential effects (i.e. agonism versus antagonism) at various 5-HT receptor subtypes [3, 4, 6–8]. For example, *d*-LSD has been shown to be a full agonist at central 5-HT_{1A} receptors linked to inhibition of adenylate cyclase [9], while it has been reported to display no direct agonist effects [7, 10] or only weak partial agonist activity [11] at 5-HT_2 receptors. In cortical electrophysiological studies, *d*-LSD inhibits 5-HT_2-mediated neuronal depolarizations produced by 5-HT, whereas *d*-LSD alone does not produce a depolarization, suggesting a 5-HT_2 antagonist action of *d*-LSD [12].

In contrast to *d*-LSD, the effects of the indolealkylamine *N,N*-dimethyltryptamine (DMT) on second-messenger systems linked to 5-HT receptors have not been analyzed extensively to date. Such information may be important since DMT and *d*-LSD share the ability to induce hallucinations yet belong to different chemical classes. Therefore, the purpose of the present study was to determine the agonist versus antagonist properties of DMT at central 5-HT_{1A} and 5-HT_2 receptors using receptor binding techniques and biochemical functional assays.

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MATERIALS AND METHODS

Radioligand studies. Receptor binding assays were performed as described previously [13, 14]. Briefly, adult rat brains were obtained immediately following decapitation and the frontal cortex of each was dissected as needed. Tissues were homogenized in 20 vol. of 50 mM Tris–HCl buffer (pH 7.7 at 25°) using a Brinkmann Polytron and then centrifuged in an IEC B20A centrifuge at 49,000 *g* for 10 min. The supernatant was discarded and the pellet resuspended in the same volume of Tris–HCl buffer and incubated at 37° for 10 min prior to a second centrifugation at 49,000 *g* for 10 min. The final pellet was resuspended in 70 vol. of Tris–HCl buffer containing 10 μM pargyline, 4 mM calcium chloride and 0.1% ascorbic acid. The suspensions were used immediately in the binding assay.

Binding assays for drug displacement studies consisted of 0.1 mL [^{3}H]-ketanserin (final concentration of 0.4 to 0.8 nM) or 0.1 mL [^{3}H]-8-hydroxy-2-(di-*n*-propylamino)tetralin ([^{3}H]-8-OH-DPAT) (final concentration of 0.2 to 0.4 nM), 0.1 mL buffer or displacing drug, 0.1 mL buffer or GTP (final concentration 10^{-4} M) and 0.7 mL tissue suspension. Following incubation at 25° for 30 min, the assays were filtered rapidly under vacuum through Whatman GF/B filters with two 5-mL washes using 50 mM Tris–HCl buffer. Radioactivity was measured by liquid scintillation spectroscopy in 5 mL of Aquasol (New England Nuclear, Boston, MA) at approximately 54% efficiency. Specific binding was defined using 1 μM cinanserin for [^{3}H]-ketanserin studies and 10 μM 5-HT for [^{3}H]-8-OH-DPAT studies. The K_i value of DMT in the presence

of GTP was calculated using a K_D value for [^{3}H]-8-OH-DPAT of 2.7 nM, whereas a K_D value of 0.75 nM was used to calculate the K_i value of DMT in the absence of GTP [14, 15].

Adenylate cyclase assay. Adenylate cyclase activity was measured in rat hippocampal tissue as described previously [16]. Frozen rat brains were thawed and hippocampi removed and homogenized in 2.5 mL of tissue medium [17]. The homogenate was then centrifuged at 39,000 *g* for 10 min. The pellet was resuspended in tissue medium, centrifuged, and then resuspended in 9 vol. of tissue medium. The membrane homogenate (50 μL) was added to 125 μL of assay medium along with 10 μM forskolin (final concentration) and various concentrations of the drug to be tested. Assay medium, drug and membrane homogenate were preincubated for 5 min at 30° prior to the addition of [^{32}P]-adenosine-5-triphosphate ([^{32}P]-ATP). The reaction proceeded at 30° for 5 min and then was stopped by the addition of 100 μL stopping solution (2% sodium lauryl sulfate, 45 mM ATP and 1.3 mM cAMP) and incubated at 37° for 5 min. [^{3}H]-cAMP (8000 cpm) was added to each sample to determine recovery. The added [^{32}P]-ATP and resulting [^{32}P]-cAMP were separated first on Dowex, and then on alumina columns.

Phosphatidylinositol turnover. The methods used in this study to determine phosphatidylinositol (PI) turnover in cortical slices were modified slightly from previously described methods [18]. Cortices were dissected from Sprague–Dawley rats (100–200 g) and cut into 350 × 350 μm slices using a Brinkmann McIlwain Tissue Chopper. Slices were then incubated at 37° for 45 min in a shaking water bath with modified Krebs solution and bubbled O_2/CO_2 (95%/5%). Aliquots (50 μl) of slices were transferred to 20-mL scintillation vials along with 1 μCi of [^{3}H]-myoinositol, 6 mM lithium chloride and 10 μM pargyline in modified Krebs buffer. Next, vials were gassed, capped and incubated again for 45 min at 37°. Antagonists then were added if indicated and incubated for 15 min. 5-HT (10^{-5} M) or various concentrations of DMT were then added, and the vials were again gassed, capped and incubated at 37° for 45 min. A chloroform/methanol mixture was added to each vial to stop the reaction. Distilled H_2O and $CHCl_3$ were added to each vial to separate the solution into phases. The vials were then vortexed and set aside to allow phase separation.

Dowex ion-exchange columns (200–400 mesh) were used for the extraction. The aqueous phase from each vial was transferred to the columns. Two washes were performed with 5 mM unlabeled myoinositol. When the washes were complete, collection tubes were set up and the radiolabeled inositol phosphates were eluted with 1 N ammonium formate in 0.1 N formic acid. From each tube, the eluate was transferred to scintillation vials along with 10 mL of Aquasol scintillation fluid. The vials were then placed in a liquid scintillation counter. The amount of radioactivity was considered a measure of PI turnover.

Drugs. Drugs were obtained from the following sources: [^{3}H]-ketanserin (61.8 Ci/mmol; New England Nuclear); [^{3}H]-8-OH-DPAT (129 Ci/mmol; New England Nuclear); [^{3}H]-myoinositol (15 Ci/mmol; American Radiolabeled Chemicals Inc., St. Louis, MO); [^{32}P]-ATP (10–50 Ci/mmol; Amersham International, U.K.); [^{3}H]-cAMP (31.2 Ci/mmol; New England Nuclear); cinanserin (E.R. Squibb & Sons, Inc., Princeton, NJ); ketanserin (Janssen Pharmaceutical, Piscataway, NJ); (−)-pindolol (Sandoz, East Hanover, NJ); and all other drugs (Sigma Chemical Co., St. Louis, MO).

RESULTS

Effect of 10^{-4} M GTP on DMT competition for 5-HT$_{1A}$ and 5-HT$_2$ binding sites labeled by [^{3}H]-8-OH-DPAT and [^{3}H]-ketanserin in rat frontal cortex. Drug competition studies using DMT were performed with [^{3}H]-8-OH-DPAT (K_D = 0.75 nM in the absence of GTP and 2.7 nM in the presence of GTP) [14, 15] or [^{3}H]-ketanserin (K_D = 0.65 nM) [19] in the absence or presence of 10^{-4} M GTP. This concentration of nucleotide has been reported to decrease the potency of agonists, but not antagonists, for 5-HT receptor binding sites [14, 20]. As shown in Fig. 1A, 10^{-4} M GTP significantly decreased the ability of DMT to compete for [^{3}H]-8-OH-DPAT-labeled 5-HT$_{1A}$ receptor binding sites. In the absence of GTP, the apparent affinity constant (K_i) of DMT was 130 ± 6 nM, whereas in the presence of 10^{-4} M GTP the K_i was 464 ± 10 nM. Thus, the addition of 10^{-4} M GTP significantly decreased the affinity of DMT for [^{3}H]-8-OH-DPAT-labeled 5-HT$_{1A}$ binding sites ($P < 0.001$; Student's *t*-test).

In marked contrast, DMT competition for [^{3}H]-ketanserin-labeled 5-HT$_2$ receptor binding sites was essentially the same in the absence or presence of 10^{-4} M GTP (Fig. 1B). DMT displayed a K_i value versus [^{3}H]-ketanserin binding of 440 ± 20 nM in the absence of GTP and 455 ± 10 nM in the presence of 10^{-4} M GTP. These values were not significantly different ($P > 0.05$; Student's *t*-test).

Effects of DMT and 8-OH-DPAT on forskolin-stimulated adenylate cyclase. In rat hippocampi, 10 μM forskolin caused an approximately 7-fold increase in adenylate cyclase activity. In our system, the 5-HT$_{1A}$ selective agonist 8-OH-DPAT produced a 26 ± 3% inhibition of cyclase activity at 10^{-5} M (N = 5), a value similar to that obtained by other investigators [9, 21]. The effect of DMT on forskolin-stimulated adenylate cyclase activity in rat hippocampi is shown in Fig. 2. DMT caused a concentration-dependent decrease of forskolin-stimulated adenylate cyclase with a maximum inhibition of 30 ± 4% occurring at 10^{-4} M DMT. The EC_{50} of DMT was approximately 4×10^{-6} M.

In addition, experiments were performed with increasing concentrations of DMT in the presence of 10^{-6} M (−)-pindolol, a potent 5-HT$_{1A}$ receptor antagonist which has been reported to inhibit the effect of 8-OH-DPAT on forskolin-stimulated adenylate cyclase [16]. As shown in Fig. 2, 10^{-6} M (−)-pindolol blocked the DMT-induced inhibition of adenylate cyclase activity. At 10^{-4} M DMT and 10^{-6} M (−)-pindolol, cyclase activity was 97 ± 3% of the baseline forskolin signal.

Effects of DMT on PI turnover and 5-HT-stimulated PI turnover in rat cortex. Rat cortical

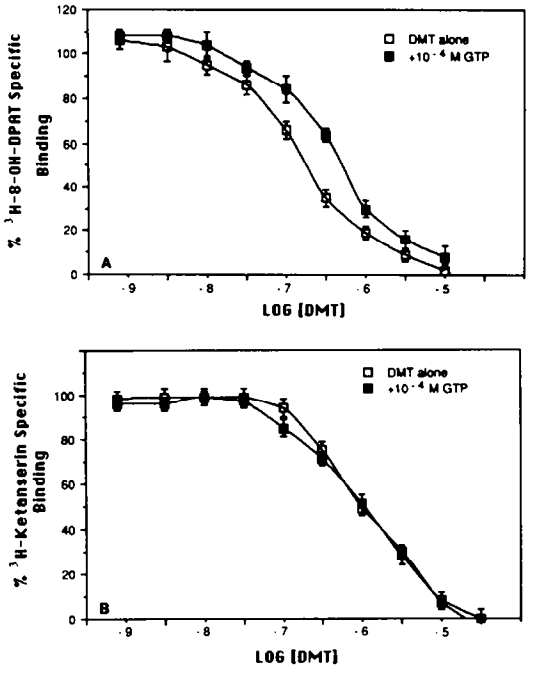

Fig. 1. Effect of 10^{-4} M GTP on DMT competition for 5-HT$_{1A}$ receptors labeled with [^{3}H]-8-OH-DPAT and 5-HT$_2$ receptors labeled with [^{3}H]-ketanserin in rat cortex. (A) Specific binding of [^{3}H]-8-OH-DPAT was defined as the excess over blanks taken in the presence of 10^{-5} M 5-HT, with a final assay concentration of 0.2 to 0.4 nM [^{3}H]-8-OH-DPAT. In the presence of 10^{-4} M GTP, total specific [^{3}H]-8-OH-DPAT binding was reduced by 75%, averaging 880 ± 150 cpm in the absence and 200 ± 60 cpm in the presence of GTP. The K_i value of DMT in the presence of GTP was calculated using a K_D value of 2.7 nM, whereas a K_D value of 0.75 nM was used to calculate the K_i value of DMT in the absence of GTP [14, 15]. The presence of GTP significantly decreased the affinity of DMT for 5-HT$_{1A}$ cites ($P < 0.001$, Student's t-test). (B) Specific [^{3}H]-ketanserin binding was defined as the excess over blanks taken in the presence of 1 μM cinanserin with a final assay concentration of 0.4 to 0.8 nM [^{3}H]-ketanserin. GTP (10^{-4} M) did not affect total specific [^{3}H]-ketanserin binding, which averaged 2900 ± 200 cpm in the absence and 3000 ± 200 cpm in the presence of GTP. Results are the means ± SEM of three experiments performed in triplicate.

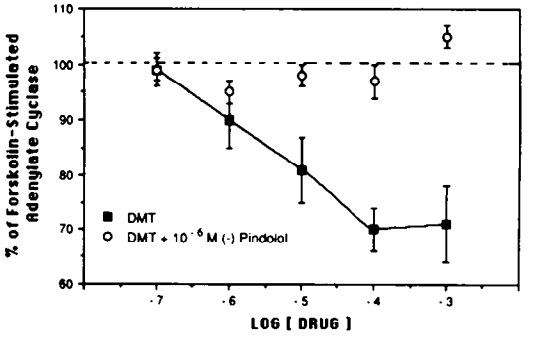

Fig. 2. Effect of increasing concentrations of DMT on forskolin-stimulated adenylate cyclase in rat hippocampus. The effect of increasing concentrations of DMT on forskolin-stimulated adenylate cyclase was tested in the absence and presence of 10^{-6} M (−)-pindolol. Data shown are the means ± SEM of 3–8 experiments, each performed in triplicate. Baseline values for adenylate cyclase: 2600 ± 400 cpm.

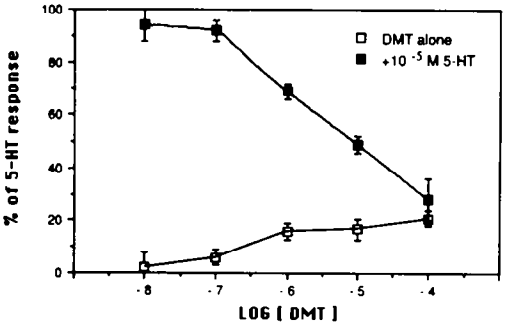

Fig. 3. Effect of increasing concentration of DMT on PI turnover in the absence and presence of 10^{-5} M 5-HT. The 5-HT (10^{-5} M) PI signal averaged 60 ± 4% above baseline values (1600 ± 100 cpm) and is represented as 100% response. Results are expressed as a percentage of this 5-HT response. Data shown are the means ± SEM of 3–4 experiments, each performed in triplicate.

slices were incubated for 45 min in the presence of various concentrations of DMT (Fig. 3). In the control condition, PI stimulation produced by 10^{-5} M 5-HT averaged 60 ± 4% above baseline levels. DMT began to stimulate PI turnover at approximately 10^{-7} M with and EC_{50} value of 3×10^{-6} M. DMT induced a maximal stimulation of 12 ± 1% above baseline at 10^{-6} M (i.e. 20% of the 10^{-5} M 5-HT signal).

To determine the effect of DMT on 5-HT-stimulated PI turnover, DMT concentration–response curves were analyzed in the presence of 10^{-5} M 5-HT. As shown in Fig. 3, micromolar concentrations of DMT decreased 5-HT-stimulated PI turnover. In the absence of DMT, 5-HT (10^{-5} M) stimulated PI turnover to 60 ± 4% above baseline. DMT concentration-dependently antagonized 5-HT-stimulated PI turnover at drug concentrations above 10^{-7} M, with 70 ± 8% of the 5-HT-induced PI response inhibited at a DMT concentration of 10^{-4} M.

Effects of 10^{-6} M ketanserin on PI turnover stimulated by 10^{-5} M concentrations of 5-HT and DMT. An attempt was made to determine if the stimulating effect of 5-HT and DMT on PI turnover is mediated by 5-HT$_2$ receptors. The selective 5-HT$_2$ antagonist ketanserin (10^{-6} M) was added 15 min before the addition of either 10^{-5} M 5-HT or 10^{-5} M DMT to the assay system (Fig. 4). The PI response obtained with 10^{-5} M 5-HT averaged 50 ± 3% above baseline values. In the presence of 10^{-6} M ketanserin and 10^{-5} M 5-HT, PI turnover was reduced significantly to only 4.5 ± 3% above baseline values ($P < 0.001$; Student's t-test). By contrast, the PI response to 10^{-5} M DMT was essentially the same

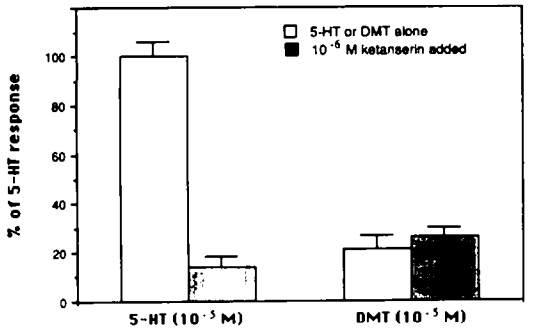

Fig. 4. Effects of 10^{-6} M ketanserin on 5-HT and DMT stimulation of PI turnover in rat cortex. Inositol phosphate levels were measured in the absence and presence of 10^{-6} M ketanserin. The 5-HT (10^{-5} M) PI signal averaged 50 ± 3% above baseline values (1600 ± 100 cpm) and is represented as 100% response. The effect of 10^{-5} M DMT is shown as a percentage of the 10^{-5} M 5-HT response. Data shown are the means ± SEM of 3–4 experiments, each performed in triplicate. Ketanserin significantly reduced the 5-HT response ($P < 0.001$; Student's t-test).

in the absence (11 ± 2%) and presence (13 ± 2%) of 10^{-6} M ketanserin ($P > 0.05$; Student's t-test). Preliminary studies with ketanserin alone did not show a PI turnover above baseline levels. Therefore, the relatively low level of PI stimulation induced by 10^{-5} M DMT (200 ± 10 cpm above baseline) does not appear to be 5-HT_2 receptor mediated since it was not blocked by 10^{-6} M ketanserin.

DISCUSSION

The major finding of the present study was that the indolealkylamine hallucinogen DMT appeared to have opposing actions on 5-HT_{1A} and 5-HT_2 receptor subtypes in rat brain. The guanyl nucleotide GTP (10^{-4} M) significantly decreased the potency of DMT in competing for 5-HT_{1A} receptors (from a K_i value of 130 nM to 464 nM), a finding which has been reported, but not proven, to indicate agonist activity at this site [14, 20]. In addition, agonist activity of DMT at 5-HT_{1A} receptors was demonstrated directly by the ability of this agent to concentration-dependently inhibit forskolin-stimulated adenylate cyclase activity in rat hippocampus, an effect mediated via 5-HT_{1A} receptors [4, 9]. DMT appears to be a full agonist since it was equally as efficacious as the selective 5-HT_{1A} agonist 8-OH-DPAT in inhibiting cyclase activity. The 5-HT_{1A} antagonist (−)-pindolol blocked the DMT inhibition of cyclase production. In addition, previous studies have demonstrated agonist activity of DMT at 5-HT_{1A} receptors. For example, DMT enhances the acoustic startle response in rats [22], a 5-HT_{1A} agonist effect [23]. Furthermore, DMT has been shown to inhibit firing of raphe neurons [24], an action also linked to the activation of 5-HT_{1A} receptors [25].

In contrast to the agonist activity at 5-HT_{1A} receptors, DMT appears to have primarily antagonistic properties at 5-HT_2 receptors. In radioligand binding studies, GTP fails to alter the ability of DMT to compete for 5-HT_2 receptors in rat brain, suggesting a lack of agonist activity at this receptor [20]. Antagonist activity of DMT at 5-HT_2 receptors is also suggested by the effects of this drug on the PI turnover system in rat cortex. DMT inhibited 5-HT-stimulated PI turnover at concentrations above 10^{-7} M and exhibited a 70 ± 8% inhibition of the 5-HT response at 10^{-4} M DMT. Micromolar concentrations of DMT alone produced a slight PI stimulation which was not blocked by the 5-HT_2 antagonist ketanserin, suggesting that DMT activation of this biochemical pathway is not via 5-HT_2 receptors.

The ability of DMT to antagonize 5-HT_2-mediated effects also has been reported in other systems. DMT inhibits 5-HT-induced excitation of CNS neurons [26], a putative 5-HT_2-mediated effect [27–29]. Furthermore, application of DMT alone to cat brainstem neurons produces no excitatory effects. DMT also has been shown to inhibit 5-HT-induced platelet shape change, a 5-HT_2-mediated effect [30, 31]. DMT alone displayed weak agonist activity in this system. In addition to this platelet effect, DMT has been shown to display weak partial agonist activity in two other peripheral tissues which contain 5-HT_2 receptors, the rabbit aorta and rat jugular vein [32]. Therefore, DMT may possess very weak partial agonist activity at 5-HT_2 receptors. In the peripheral studies, however, 5-HT_2 antagonists were not tested for their ability to block the agonist effect of DMT. The PI stimulation produced by DMT in the present study was not affected by the 5-HT_2 antagonist ketanserin, suggesting that DMT stimulation of PI turnover is not via the 5-HT_2 receptor.

The data indicating that DMT both stimulated 5-HT_{1A} receptors and antagonized 5-HT_2 receptors are similar to the results obtained with the structurally related indolealkylamine hallucinogen, d-LSD. Similar to the agonist actions of DMT at 5-HT_{1A} receptors, d-LSD also inhibits raphe firing [24], enhances rat acoustic startle responses [33], and potently inhibits forskolin-stimulated adenylate cyclase production [9]. Likewise, d-LSD antagonizes 5-HT-stimulated PI turnover at 5-HT_2 receptors and has also been shown to be an antagonist in numerous other 5-HT_2-mediated systems [7]. In addition, GTP fails to affect the binding of [^{3}H]-LSD to 5-HT_2 receptors yet significantly decreases the B_{max} and the affinity of [^{3}H]-LSD binding to the 5-HT_{1A} receptor [8]. The findings are analogous to the DMT results presented in this report.

The ability of a drug to stimulate one subtype of 5-HT receptor and to antagonise another has been reported for other compounds as well. For instance, the piperazines m-trifluoromethylphenylpiperazine (TFMPP) and m-chlorophenylpiperazine (MCPP) antagonize 5-HT_2-mediated PI turnover in cortex, yet both drugs stimulate 5-HT_{1C}-mediated PI turnover in the choroid plexus [34]. Behavioral studies suggest that lisuride, TFMPP, imipramine and dihydroergosine inhibit 5-HT_2-mediated head twitch and induce the 5-HT_{1A} receptor-mediated behavioral syndrome in rats [6, 35].

To our knowledge, previously analyzed tryptamine

derivatives have displayed only agonist activity in biochemical studies of 5-HT receptor subtypes. For example, the tryptamine derivates 5-HT, 5-methoxytryptamine, 5-carboxyamidotryptamine, α-methyl-5-HT and 1-methyl-5-HT have been shown to be agonists at 5-HT_{1A}, 5-HT_{1C} and/or 5-HT_2 receptors [3–5]. However, the second-messenger effects of dimethyltryptamine derivatives have been studied only at 5-HT_{1A} and 5-HT_{1C} receptors. Similar to the tryptamine derivates mentioned above, which display full agonist activity at 5-HT_{1A} and 5-HT_{1C} receptors, the dimethyltryptamine derivatives 5-methoxy-DMT and bufotenine behave as full agonists at 5-HT_{1A} receptors [4]. These dimethyltryptamines, however, produce only 50% of the full agonist response at 5-HT_{1C} receptors linked to PI turnover [5]. In contrast to the full agonist and partial agonist actions of dimethyltryptamines at 5-HT_{1A} and 5-HT_{1C} receptors, respectively, the present study found DMT to display antagonistic activity at 5-HT_2 receptors. Future structure–activity studies may elucidate the molecular properties underlying the agonist, partial agonist and antagonist activities of dimethyltryptamines at 5-HT receptor subtypes.

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REFERENCES

1. Glennon RA, Central serotonin receptors as targets for drug research. *J Med Chem* **30**: 1–12, 1987.
2. Pierce PA and Peroutka SJ, Hallucinogenic drug interactions with neurotransmitter receptor binding sites in human cortex. *Psychopharmacology (Berlin)* **97**: 118–122, 1989.
3. Kendall DA and Nahorski SR, 5-Hydroxytryptamine-stimulated inositol phospholipid hydrolysis in rat cerebral cortex slices: Pharmacological characterization and effects of antidepressants. *J Pharmacol Exp Ther* **233**: 473–479, 1985.
4. Dumuis A, Sebben M and Bockaert J, Pharmacology of 5-hydroxytryptamine-1A receptors which inhibit cAMP production in hippocampal and cortical neurons in primary culture. *Mol Pharmacol* **33**: 178–186, 1988.
5. Hoyer D, Waeber C, Schoeffter P, Palacios JM and Dravid A, 5-HT_{1C} receptor-mediated stimulation of inositol phosphate production in pig choroid plexus. A pharmacological characterization. *Naunyn Schmiedebergs Arch Pharmacol* **339**: 252–258, 1989.
6. Gerber R, Barbaz BJ, Martin LL, Neale R, Williams M and Liebman JM, Antagonism of L-5-hydroxytryptophan-induced head twitching in rats by lisuride: A mixed 5-hydroxytryptamine agonist–antagonist. *Neurosci Lett* **60**: 207–213, 1985.
7. Pierce PA and Peroutka SJ, Antagonism of 5-hydroxytryptamine$_2$ receptor-mediated phosphatidylinositol turnover by *d*-lysergic acid diethylamide. *J Pharmacol Exp Ther* **247**: 918–925, 1988.
8. Norman AB, Nash DR and Sanberg PR, [^{3}H]Lysergic acid diethylamide (LSD): Differential agonist and antagonist binding properties at 5-HT receptor subtypes in rat brain. *Neurochem Int* **14**: 497–504, 1989.
9. DeVivo M and Maayani S, Characterization of the 5-hydroxytryptamine$_{1A}$ receptor-mediated inhibition of forskolin-stimulated adenylate cyclase activity in guinea pig and rat hippocampal membranes. *J Pharmacol Exp Ther* **238**: 248–253, 1986.
10. Heller WA and Baraban JM, Potent agonist activity of DOB at 5-HT_2 receptors in guinea pig trachea. *Eur J Pharmacol* **138**: 115–117, 1987.
11. Sanders-Bush E, Burris KD and Knoth K, Lysergic acid diethylamide and 2,5-dimethoxy-4-methylamphetamine and partial agonists at serotonin receptors linked to phosphoinositide hydrolysis. *J Pharmacol Exp Ther* **246**: 924–928, 1988.
12. Pierce PA and Peroutka SJ, *d*-LSD differentially affects the dual actions of 5-hydroxytryptamine on cortical neurons. *Neuropharmacology* **29**: 705–712, 1990.
13. Leysen JE, Niemegeers JE, Van Neuten JM and Laduron PM, [^{3}H]Ketanserin (R 41 468), a selective ^{3}H-ligand for serotonin$_2$ receptor binding sites. *Mol Pharmacol* **21**: 301–314, 1982.
14. Schlegel JR and Peroutka SJ, Nucleotide interactions with 5-HT_{1A} binding sites directly labeled by [^{3}H]-8-hydroxy-2-(di-*n*-propylamino)tetralin ([^{3}H]-8-OH-DPAT). *Biochem Pharmacol* **35**: 1943–1949, 1986.
15. Peroutka SJ, Pharmacological differentiation and characterization of 5-HT_{1A}, 5-HT_{1B}, and 5-HT_{1C} binding sites in rat frontal cortex. *J Neurochem* **47**: 529–540, 1986.
16. Salomon Y, Adenylate cyclase assay. In: *Advances in Cyclic Nucleotide Research* (Eds. Brooker G, Greengard P and Robison GA), pp. 1035–1053. Raven Press, New York, 1979.
17. Oksenberg D and Peroutka SJ, Antagonism of 5-HT_{1A} receptor-mediated inhibition of adenylate cyclase activity by pindolol and propranolol isomers. *Biochem Pharmacol* **37**: 3429–3433, 1988.
18. Berridge MJ, Downes PC and Hanley MR, Lithium amplifies agonist-dependent phosphatidylinositol responses in brain and salivary glands. *Biochem J* **206**: 587–595, 1982.
19. Peroutka SJ, Hamik A, Harrington MA, Hoffman AJ, Mathis CA, Pierce PA and Wang SS-H, (*R*)-(−)-[^{77}Br]4-Bromo-2,5-dimethoxyamphetamine labels a novel 5-hydroxytryptamine binding site in brain membranes. *Mol Pharmacol* **34**: 537–542, 1988.
20. Battaglia G, Shannon M and Titeler M, Guanyl nucleotide and divalent cation regulation of cortical S_2 serotonin receptors. *J. Neurochem* **43**: 1213–1219, 1984.
21. Sleight AJ, Marsden CA, Palfreyman MG, Mir AK and Lovenberg W, Chronic MAO A and MAO B inhibition decreases the 5-HT_{1A} receptor-mediated inhibition of forskolin-stimulated adenylate cyclase. *Eur J Pharmacol* **154**: 255–261, 1988.
22. Davis M and Sheard MH, Biphasic dose–response effects of *N-N*-diethyltryptamine on the rat startle reflex. *Pharmacol Biochem Behav* **2**: 827–829, 1974.
23. Nanry KP and Tilson HA, The role of 5-HT_{1A} receptors in the modulation of the acoustic startle reflex in rats. *Psychopharmacology (Berlin)* **97**: 507–513, 1989.
24. Aghajanian GK, Foote WE and Sheard MH, Action of psychotogenic drugs on single midbrain raphe neurons. *J Pharmacol Exp Ther* **171**: 178–187, 1970.
25. Williams JT, Colmers WF and Pan ZZ, Voltage and ligand activated inwardly rectifying currents in rat dorsal raphe neurons *in vitro*. *J Neurosci* **8**: 3499–3506, 1988.
26. Bradley PB and Briggs I, Further studies on the mode of action of psychotomimetic drugs: Antagonism of the excitatory actions of 5-hydroxytryptamine by methylated derivatives of tryptamine. *Br J Pharmacol* **50**: 345–354, 1974.
27. Penington NJ and Reiffenstein RJ, Lack of effect of antagonists on serotonin-induced inhibition in rat hippocampus. *Can J Physiol Pharmacol* **64**: 1413–1418, 1986.

28. Davies MF, Diesz RA, Prince D and Peroutka SJ, Two distinct effects of 5-hydroxytryptamine on single cortical neurons. *Brain Res* **423**: 347–352, 1987.
29. North RA and Uchimura N, 5-Hydroxytryptamine acts at 5-HT_2 receptors to decrease potassium conductance in rat nucleus accumbens neurones. *J Physiol (Lond)* **417**: 1–12, 1989.
30. Graf M and Pletscher A, Shape change of blood platelets—A model for cerebral 5-hydroxytryptamine receptors? *Br J Pharmacol* **65**: 601–608, 1979.
31. McClue SJ, Brazell C and Stahl SM, Hallucinogenic drugs are partial agonists of the human platelet shape change response: A physiological model of the 5-HT_2 receptor. *Biol Psychiatry* **26**: 297–302, 1989.
32. Leff P, Martin GR and Morse JM, The classification of peripheral 5-HT_2-like receptors using tryptamine agonist and antagonist analogues. *Br J Pharmacol* **89**: 493–499, 1986.
33. Davis M and Sheard MH, Effects of lysergic acid diethylamide (LSD) on habituation and sensitization of the startle response in the rat. *Pharmacol Biochem Behav* **2**: 675–683, 1974.
34. Conn JP and Sanders-Bush E, Relative efficacies of piperazines at the phosphoinositide hydrolysis-linked serotonergic (5-HT-2 and 5-HT-1) receptors. *J Pharmacol Exp Ther* **242**: 552–557, 1987.
35. Pericic D and Manev H, Behavioural evidence for simultaneous dual changes of 5-HT receptor subtypes: Mode of antidepressant action? *Life Sci* **42**: 2593–2601, 1988.