

Structure–Activity Relationships of Phenylalkylamines as Agonist Ligands for 5-HT_{2A} Receptors

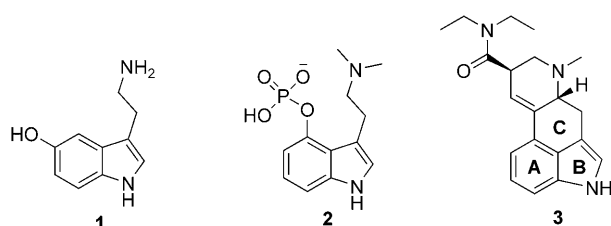
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Agonist activation of central 5-HT_{2A} receptors results in diverse effects, such as hallucinations and changes of consciousness. Recent findings indicate that activation of the 5-HT_{2A} receptor also leads to interesting physiological responses, possibly holding therapeutic value. Selective agonists are needed to study the full therapeutic potential of this receptor. 5-HT_{2A} ligands with agonist profiles are primarily derived from phenylalkylamines, indolealkylamines, and certain piperazines. Of these, phenylalkylamines,

most notably substituted phenylisopropylamines, are considered the most selective agonists for 5-HT₂ receptors. This review summarizes the structure–activity relationships (SAR) of phenylalkylamines as agonist ligands for 5-HT_{2A} receptors. Selectivity is a central theme, as is selectivity for the 5-HT_{2A} receptor and for its specific signaling pathways. SAR data from receptor affinity studies, functional assays, behavioral drug discrimination as well as human studies are discussed.

Introduction

Wide-ranging physiological processes are mediated through the serotonin (5-hydroxytryptamine, 5-HT, **1**) system. 5-HT and



its receptors are scattered throughout the body. Dysfunction has been implicated in cardiovascular and digestive disorders as well as numerous psychiatric disorders.^[1] Pharmacological manipulation of the 5-HT system is believed to have therapeutic potential, and therefore the subject of intense research.^[2] There are seven distinct families of 5-HT receptors (5-HT_{1–7}), and there is molecular and functional evidence for the existence of at least 14 different mammalian subtypes.^[3] With the exception of the 5-HT₃ receptor, a ligand-gated ion channel, all 5-HT receptors are G protein-coupled receptors (GPCRs).^[3,4] The 5-HT₂ receptor family has three known subtypes, 5-HT_{2A}, 5-HT_{2B} and 5-HT_{2C}^[4,5] with ~46–50% sequence identity.^[11] Moreover, the transmembrane domains of the 5-HT_{2A} and 5-HT_{2C} receptors share 80% sequence identity, suggesting similar pharmacological profiles.^[6]

In the central nervous system (CNS), 5-HT_{2A} receptors are primarily found in cortical and forebrain areas, various brainstem nuclei, and the hippocampus.^[7] The cellular localization of 5-HT_{2A} receptors is primarily on the dendrites^[8,9] of cortical pyramidal glutamatergic projection neurons,^[10,11] local GABAergic interneurons,^[12] and on cholinergic neurons.^[13–15] A proportion of 5-HT_{2A} receptors is believed to be located presynaptically on,

most probably, monoamine axons.^[9] Glial 5-HT_{2A} receptors have been identified also.^[9,16] Peripherally, the 5-HT_{2A} receptor is found in platelets, vascular smooth muscle cells, and ocular tissue.^[17–19] In contrast, the 5-HT_{2B} receptor is primarily found in the periphery, such as the rat stomach fundus, and canine lungs and smooth muscles.^[3,4,20] Furthermore 5-HT_{2B} receptors are found in the hearts of primates and rats.^[21] The murine 5-HT_{2B} receptor is expressed in the stomach, intestine, pulmonary smooth muscles, myocardium, and the brain, most notably cerebellar Purkinje cells.^[22] Distribution of the 5-HT_{2C} receptor is primarily limited to the CNS.^[1] 5-HT_{2C} mRNA is broadly distributed throughout numerous brain regions; this receptor subtype is believed to be the principal 5-HT receptor in the brain.^[23]

Physiological Roles and Therapeutic Potential of 5-HT₂ Receptors

Given their distribution pattern, 5-HT₂ receptors have diverse physiological roles. Central 5-HT_{2A} receptors modulate GABAergic and glutamergic neurotransmission.^[17] Activation of 5-HT_{2A} receptors stimulates the secretion of various hormones.^[24] 5-HT_{2A} receptors play a physiological role in working memory,^[25] the regulation of cognitive states, and associative learning.^[26] Moreover, 5-HT_{2A} receptors influence neuronal plasticity

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through brain-derived neurotrophic factor (BDNF)-mediated processes.^[27] Intra-ocular pressure (IOP) is regulated by 5-HT_{2A} receptors.^[18,19] Peripheral 5-HT_{2A} receptors mediate diverse processes such as vasoconstriction and platelet aggregation.^[1,28] 5-HT_{2B} receptors mediate neural tube morphogenesis in the embryo,^[29] and are important in the development of the cardiovascular system.^[30,31] The 5-HT_{2C} receptor is involved in diverse processes such as locomotor activity, angiogenesis, and neuro-endocrine functions.^[17] Moreover, the 5-HT_{2C} receptor regulates various aspects of feeding and food intake,^[32] and is implicated in sexual dysfunction in males.^[33,34] Both 5-HT_{2A} and 5-HT_{2B} receptors were found to mediate liver regeneration in a partial hepatectomy model.^[35]

Drugs that target 5-HT₂ receptors are used in the treatment of various psychiatric disorders, including depression,^[36] anxiety,^[37–39] obsessive-compulsive disorders,^[40] and schizophrenia.^[41,42] Many antipsychotic drugs are 5-HT_{2A} receptor inverse agonists or antagonists.^[43,44] Activation of the 5-HT_{2A} receptor by agonist psilocybin (**2**) produces schizophrenia-like psychotic symptoms in humans, which are significantly reduced by selective 5-HT_{2A} antagonists.^[45] Psychotomimetic effects of hallucinogenic drugs such as psilocybin (**2**), *d*-lysergic acid *N,N*-diethylamide (LSD, **3**), and mescaline (**4**) are primarily mediated by 5-HT_{2A} receptors.^[46–49] Mystical-type experiences of sustained personal meaning have been reported to result from the use of psilocybin in a controlled setting.^[50] Elucidation on how these experiences arise in the brain could potentially have therapeutic possibilities.^[50] The reduction in IOP by 5-HT_{2A} agonists has recently been recognized as an efficient treatment for ocular hypertension and glaucoma.^[18,51,52] Agonist activation of 5-HT_{2A} and 5-HT_{2B} receptors results in liver regeneration,^[35] however clinical efficacy in liver regeneration following transplantation remains to be demonstrated.^[53,54] 5-HT_{2B} receptor antagonists can be used to treat anxiety disorders, however, their application is limited because of the role this receptor plays in embryogenesis.^[17] Drugs targeting both 5-HT_{2B} and 5-HT_{2C} receptors can be used to treat migraines.^[42,55] Drugs modulating 5-HT_{2C} activity are applicable in the treatment of obesity, erectile dysfunction and anxiety disorders.^[17,32]

5-HT₂ Receptor Agonist Ligands

Ligands for 5-HT₂ receptors belong to structurally diverse chemical classes, most notably indolealkylamines, phenylalkylamines, arylpiperazines, alkylpiperidines, alkylpiperazines, polycyclic/tricyclic agents, among others.^[56] 5-HT₂ receptor subtype ligands have been developed.^[17,57,58] Agonist activation of 5-HT₂ receptors has been the subject of recent studies, indicating a growing interest in the therapeutic potential underlying activation of this class of receptors.^[18,32,35,36,38,40,50] 5-HT₂ receptor agonists generally show little subtype selectivity, however, some selective agonists have been designed for 5-HT_{2B} and 5-HT_{2C} receptors.^[32,59,60] The identification of agonists with 5-HT_{2A} receptor subtype selectivity lags behind, and the need for their development has been acknowledged.^[3,4,17] Agonists selective for the 5-HT_{2A} receptor and its associated signaling pathways

are needed to research the full therapeutic potential of this receptor.

5-HT_{2A} receptor agonists and partial agonists are primarily indolealkylamines and phenylalkylamines.^[61] Certain piperazines have also been classified as agonists.^[62] Based on their structures, the indolealkylamines can be subdivided into tryptamines, ergolines and β -carboline, and the phenylalkylamine class includes phenethylamines and phenylisopropylamines (amphetamines). Indolealkylamines generally show little subtype selectivity, binding multiple 5-HT receptor subclasses.^[63] The most selective agonists for 5-HT₂ receptor subtypes are found in the phenylalkylamine class, most notably the substituted phenylisopropylamines. The structure–activity relationships (SAR) have been extensively studied generating a vast number of phenylalkylamines with 5-HT_{2A} binding potential and functional activity.^[64–67] The purpose of this review is to summarize the SAR of the phenylalkylamines as agonist ligands for the 5-HT_{2A} receptor, focusing on subtype and function selectivity. Signaling pathways, pharmacological methods, site-directed mutagenesis and molecular modeling studies relevant for 5-HT_{2A} receptor research are described.

5-HT_{2A} Receptor Signaling and Functional Selectivity

The concepts used in receptor pharmacology are constantly revised to fully describe the complexity of GPCR signaling.^[68,69] Ligands are believed to induce conformational changes in the GPCR resulting in differential activation of the associated signal transduction pathways.^[70,71] Moreover, cellular conditions are recognized as an important factor in determining drug action, and the classical concept of “intrinsic efficacy” is no longer supported.^[72] Detailed knowledge of downstream signaling pathways coupled to 5-HT_{2A} receptors is needed to study the functional activity of these ligands. As a pleiotropic GPCR, the 5-HT_{2A} receptor can couple to different G proteins, and has the ability to show a broad array of responses, such as internalization and desensitization.^[73,74] The ligand-induced differential activation of downstream signaling pathways has been given various names, such as “agonist-directed trafficking of receptor stimulus”, “protean agonism” and “ligand-biased efficacy”,^[70,71,75,76] however, it has been suggested that “functional selectivity” is the most suitable term to refer to this concept.^[71]

5-HT_{2A} receptors coupled to heterotrimeric GTP binding proteins regulate a variety of cell responses (Figure 1). The 5-HT_{2A} receptor activates phospholipase C (PLC) and phospholipase A₂ (PLA₂), and is involved in various other signaling cascades.^[77–80] PLC- β is activated by the 5-HT_{2A} receptor mainly through coupling with G $\alpha_{q/11}$, resulting in the release of inositol-1,4,5-triphosphate (IP₃) and 1,2-diacylglycerol (DAG) through lipid hydrolysis of phosphatidylinositol-4,5-bisphosphate (PIP₂).^[81,82] IP₃ is responsible for Ca²⁺ release from intracellular stores, whereas DAG is involved in protein kinase C (PKC) activation.^[83–86] Conversion of DAG to endocannabinoid 2-arachidonoylglycerol (2-AG) by DAG lipase (DGL)^[87,88] is known to occur following 5-HT_{2A} receptor activation in NIH 3T3 cells,^[89] adding another level of complexity.

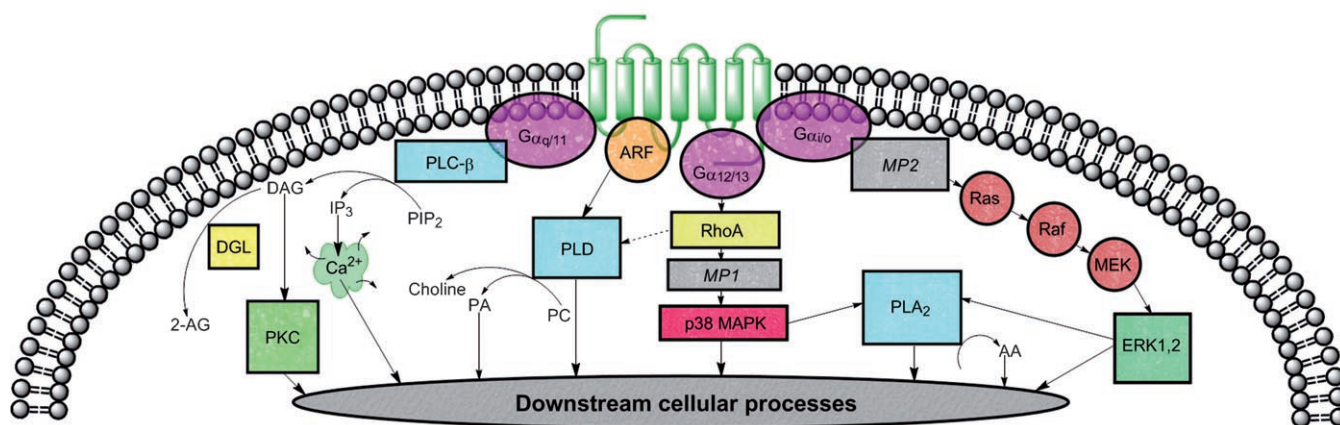


Figure 1. Graphical representation of the known 5-HT_{2A} signaling pathways. The 5-HT_{2A} receptor couples to various downstream effectors enabling diverse cellular responses following receptor activation. Some of the mediating proteins (MP) are omitted for clarity. Mediating proteins (MP1) in the Gα_{12/13}-RhoA-p38 pathway most probably are PKN, MEKK, MKK3/6 and MKK4.^[89] Shc, Grb2 and SOS are the proteins mediating (MP2) the Ras-Raf-MEK1,2-ERK1,2 pathway. Receptor regulatory pathways (e.g. phosphorylation; internalization; desensitization) following agonist activation, are not shown. Note: the localization of proteins and messengers in this Figure does not represent their localization in a functional cell.

The 5-HT_{2A} receptor is responsible for PLA₂ activation and subsequent arachidonic acid (AA) release through two parallel signaling cascades.^[90,91] The Ras-Raf-MEK-ERK signaling pathway is activated through Gα_{i/o}, leading to phosphorylation of cPLA₂ by ERK1,2. The other cascade involves a Gα_{12/13}-RhoA-p38 pathway, which results in p38 mitogen-activated protein kinase (MAPK)-mediated phosphorylation of PLA₂.^[91] The 5-HT_{2A} receptor can also couple with monomeric G-protein ADP-ribosylation factors (ARF) resulting in phospholipase D (PLD) activity.^[92] Rho proteins (e.g. RhoA) are involved in PLD activation also, in which PKC may function as a modulator.^[93] PLD promotion, notably through ARF1, leads to hydrolysis of phosphatidylcholine (PC) to yield phosphatidic acid (PA) and choline.^[94,95] PA is involved in regulation of numerous downstream cellular processes.

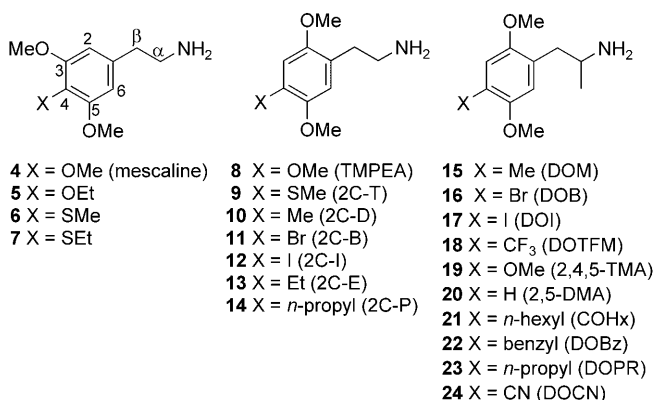
As a consequence of the acute pharmacological responses just described, 5-HT_{2A} receptor activation results in altered gene expression. Activation of 5-HT₂ receptors by known agonist DOI (17) has been found to induce expression of immedi-

patterns.^[96–98] The expression of Fos in the rat cortex elicited by DOI is mediated through 5-HT_{2A} receptors; treatment with a selective 5-HT_{2A} antagonist completely blocked the response while treatment with a 5-HT_{2C} antagonist did not influence expression levels.^[99] The genomic response to LSD (3), a hallucinogenic ergoline with a broad pharmacological profile,^[100,101] has also been studied.^[102,103] More recent work reported the use of transcriptome fingerprints elicited by 5-HT_{2A} ligands as a tool to distinguish between hallucinogenic and non-hallucinogenic agonist effects; results indicated that *c-fos* expression was a response to general 5-HT_{2A} receptor activation, while induction of *erg-1*, *erg-2* and *period-1* resulted from activation by behaviorally active agonists.^[104] This is hypothesized to result from the ability of ligands to differentially regulate intracellular signaling pathways.^[105]

Pharmacological Methods

The effects of structural modifications to ligands can be evaluated by several pharmacological methods, of which binding affinity studies and functional assays are most relevant for the SAR of 5-HT_{2A} receptors. Binding affinities can be determined by radioligand competition assays using rat brain homogenate or cloned human receptors.^[106] Several antagonist and agonist radioligands, with known binding properties, are available for competition assays at the 5-HT_{2A} receptor.^[107–109] Agonists and antagonists display different affinities for various receptor states,^[110] an observation accounted for by receptor theories.^[70,111–113]

Functional assays measure the physiological response elicited by a drug in target cells or tissues.^[114] Efficacy can be defined as the extent to which a ligand causes the receptor to change its behavior towards the cell.^[74] Several functional responses can be used to study the activation of 5-HT₂ receptors in isolated tissue.^[28] Historically, smooth muscle contraction in isolated vasculature was used as a model for receptor activation. Recent studies have used intracellular signals from down-



ate early genes *c-fos*, *ngf1c*, *tis1* and *arc* in various rat brain regions, whereas antagonists were able to block the expression

stream pathways, such as those described above, to measure receptor activation.^[71]

In vivo 5-HT-targeting drug evaluation has largely been carried out in rodents, where 5-HT mimetics cause a state of behavioral excitation, described as serotonin syndrome.^[28] The behavioral effects of phenylalkylamines have been investigated in drug discrimination (DD) studies.^[115] Rats are trained to discriminate between injections of the training drug and saline in a two-lever DD task.^[116] Substitution of the training drug, or stimulus generalization, suggests that a drug with effects similar to the training drug has been administered.^[61] The potency of a drug, and the effective dose (ED) can also be established in this paradigm. Drug-elicited behaviors, such as the head-twitch response, are studied as well.^[115] A fair number of phenylalkylamines have been examined in humans, and dosage data is available.^[117] Numerous factors unrelated to the study of receptor-ligand interactions can influence the in vivo effects of a drug, hence the direct comparison of receptor binding data with in vivo data is tentative.

Site-Directed Mutagenesis and Molecular Modeling

Site-directed mutagenesis studies have been carried out to identify the structural requirements for interactions in ligand-receptor complexes,^[118] and these studies have been reviewed in depth.^[62,82,119] Briefly, a protonated amine moiety is believed to anchor the ligand in the transmembrane helix 3 (TMH3) domain through an ionic interaction with the carboxylate group of Asp 155 (3.32).^[120] Phe340 (6.52) is essential for agonist binding and recognition. It is involved in stabilizing the aromatic ring of ligands, and mutations at this residue cause a dramatic decrease in agonist affinity and efficacy.^[121–123] Positions 5.42, 5.43, and 5.46 of transmembrane helix 5 (TMH5) play important roles in ligand recognition and specificity.^[118] The Ser 159 (3.36) residue interacts with the ligand through hydrogen bonding; this interaction may account for the differing functional effects of structurally similar indolealkylamines.^[124] Other serine residues have been found to influence agonist binding as well.^[125–127]

Three-dimensional templates based on a bacteriorhodopsin model were able to explain some of the results from mutagenesis studies.^[62,128,129] Comparative molecular modeling of ligand-GPCR interactions was greatly facilitated by the elucidation of the bovine rhodopsin crystal structure,^[130] and subsequent refinements.^[131–133] The key role of Asp 155 (3.32) as a terminal amine anchor is in agreement with previous mutagenesis data.^[134] Molecular modeling of the 5-HT_{2A} receptor, together with detailed site-directed mutagenesis studies, suggests a hydrophobic binding pocket surrounding the ligand, with aromatic residues Trp 151 (3.28), Phe 243 (5.47), Phe 244 (5.48), Trp 336 (6.48), Phe 339 (6.51), Phe 340 (6.52), Trp 367 (7.40), and Tyr 370 (7.43).^[135,136] Polar moieties within agonists may interact with Ser 159 (3.36), Thr 160 (3.37), Ser 239 (5.43), Ser 242 (5.46), and Asn 343 (6.55) from the 5-HT_{2A} binding site.^[124,137] Docking studies using a human 5-HT_{2A} receptor homology-based

model, created from an in silico activated bovine rhodopsin crystal structure, confirmed these interactions.^[137]

Different binding orientations of DOM (15) and 5-HT (1) have been suggested using a rat 5-HT_{2A} receptor model based on the frog rhodopsin projection map.^[135] The molecular mechanism of partial agonism and the relative efficacy of 5-HT_{2A} receptor ligands are determined by the ligand interaction with Ser 159 (3.36) and Ser 242 (5.46).^[138] It was concluded that specific interactions in TMH3 and TMH5 are responsible for the varying degrees of receptor activation. Recently, Ser 239 (5.43) was shown to be more critical for agonist binding and function than Ser 242 (5.46) within TMH5.^[139] The oxygen atom at the 5-position of phenylalkylamines forms a hydrogen bond with Ser 239 (5.43). Ser 242 (5.46) is believed to act as a hydrogen bond acceptor, interacting with N(1)H of the indole ring in indolealkylamines, but not phenylalkylamines.

Classic Phenylalkylamines

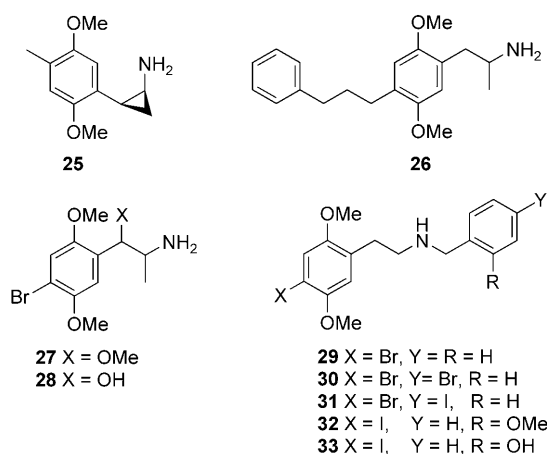
Phenylalkylamines 4–24 were used in the early SAR studies. Research focused on the nature of substituent X, *para* to the alkylamine side chain. In the case of 3,4,5-substituted phenethylamine, escaline (5) is 5–8 times more potent in humans than mescaline (4).^[140] [¹²⁵I]DOI (17) radioligand competition studies using cloned human 5-HT_{2A} receptors further confirm this finding; the affinity of 5 (K_i = 216 nM) exceeded the affinity of 4 (K_i = 551 nM) for 5-HT_{2A} receptors.^[141] It has been noted that homologation beyond *n*-propoxy leads to diminished hallucinogenic activity of this series in humans.^[142] Replacing the alkoxy substituents at the 4-position with alkylthio substituents leads to a further increase in potency. The potency of 4-thio-mescaline (6) is an order of magnitude greater than the parent compound 4, and 4-thioescaline (7) is three times more potent than 5.^[142,143]

The same pattern is seen in 2,4,5-substituted phenethylamines. For example, replacement of the 4-methoxy group in TMPEA (8) by a 4-methylthio group (2C-T, 9) leads to a large increase in potency in humans.^[117] Halogen and alkyl *para*-substituted phenethylamines, including 2C-D (10), 2C-B (11) and 2C-I (12), are generally the most potent compounds of this series.^[117,144,145] Homologation of the 4-alkyl group of 2C-D (10) results in more potent agents such as the 4-ethyl (13, 2C-E), and 4-*n*-propyl (14, 2C-P) analogues.^[145]

α -Methylation of the phenethylamines to their corresponding phenylisopropylamines leads to the most potent compounds, such as DOM (15), DOB (16), and DOI (17).^[146–148] A significant increase in potency was observed from compound 10 (ED_{50} = 5.6 μ mol kg⁻¹) to ($\pm$)-15 (ED_{50} = 1.8 μ mol kg⁻¹) in DD studies,^[149] and in human clinical studies.^[117,144] However, an affinity study using rat brain [³H]ketanserin labeled 5-HT₂ sites showed that the affinity of compound 10 (K_i = 110 nM) is approximately equal to the affinity of its phenylisopropylamine analogue ($\pm$)-15 (K_i = 100 nM).^[150] Similarly, the binding affinity of compound 18 (DOTFM, K_i = 1.5 nM) was close to that of its phenethylamine congener (K_i = 1.1 nM).^[151] A number of hypotheses account for the discrepancy between the binding affinities and in vivo potency of these compounds. The α -methyl

group may increase the general lipophilicity of the molecule, enhancing CNS distribution.^[65] Alternatively, the α -methyl group possibly contributes to metabolism inhibition by deamination. A pharmacological mechanism also contributes to the difference in potencies; the α -methyl group increases activity at 5-HT_{2A} receptors while having no effect at 5-HT_{2C} sites.^[151] Comparison of phenethylamines with their ($\pm$)-phenylisopropylamine counterparts clearly demonstrated similar binding affinities yet significant differences in their ability to activate second messenger systems.^[152]

Higher α -alkyl homologues of phenylalkylamines, or α -di-alkyl substituted analogues showed little or no activity.^[153–156] Incorporation of a cyclopropyl ring in the side chain (e.g. **25**) re-



tained some activity, however, potency is diminished when compared with its phenylisopropylamine counterpart (**15**).^[157,158] Docking studies of compound **25** using an activated 5-HT_{2A} receptor homology-based model confirmed the role of Asp155(3.32) in anchoring the protonated amine.^[137] The 2-methoxy group forms a hydrogen bond with Ser159(3.36) and Thr160(3.37) while the 5-methoxy group interacts with Ser239(5.43). Moreover, the *para* substituent projects into a lipophilic pocket formed by Ile206(4.56), Leu215(4.65) and Gly238(5.42), suggesting both a lipophilic and steric interaction.^[137]

Early phenylalkylamine SAR studies concerned with substitution patterns revealed that a 2,4,5-trisubstitution pattern on the aromatic ring leads to optimal activity in humans.^[159,160] This was clearly shown for the trimethoxyphenylisopropylamine (TMA) analogues, of which 2,4,5-TMA (**19**) was the most potent, followed by 2,4,6- and 2,3,6-TMA, while the least potent were the remaining three trisubstitution configurations.^[159] A DD study using rats trained to discriminate DOM (**15**) from saline showed a similar pattern of potency (3,4,5 < 2,4,6 < 2,4,5).^[161] This general pattern is in line with the relative affinity of these analogues for the 5-HT_{2A} receptor. Compound **19** (K_i = 1250 nM) showed greater affinity than 3,4,5-TMA (K_i = 16500 nM) for the [³H]ketanserin labeled 5-HT_{2A} receptor in rat brain homogenate.^[162] In contrast, only two of the dimethoxy derivatives, 2,5-DMA (**20**) and 2,4-DMA, were found to substitute for DOM (**15**).^[161] 2,5-DMA (**20**),^[117] the most

potent dimethoxy derivative, binds with a fivefold lower affinity (K_i = 5200 nM) in comparison with compound **19**.^[162]

Studies on the homologation of phenylisopropylamines at the 4-position found a decrease of *in vivo* hallucinogenic potency beyond *n*-propyl.^[163] Yet later work using [³H]ketanserin as a radioligand for 5-HT₂ receptors in rat brain homogenate revealed significantly increased binding affinities with the more lipophilic derivatives; *n*-hexyl and *n*-octyl derivatives showed the highest binding affinity. After *in vitro* testing, these derivatives were found to act as 5-HT₂ receptor antagonists.^[67] Using [¹²⁵I]DOI labeled human receptor data, the highest binding affinity at 5-HT_{2A} receptors was found for the 4-*n*-hexyl analogue DOHx (**21**, K_i = 0.1 nM), followed by the 4-benzyl analogue DOBz (**22**, K_i = 0.4 nM), DOB (**16**, K_i = 0.6 nM), DOI (**17**, K_i = 0.7 nM), and the 4-*n*-propyl analogue DOPR (**23**, K_i = 0.9 nM).^[63]

Continuing this line of reasoning, the 4-(3-phenylpropyl) derivative **26** was expected to have an antagonist profile.^[67] However, later work using cloned rat 5-HT_{2A} receptors, and [³H]ketanserin as a radioligand, showed a binding affinity of K_i = 30 nM for **26**, comparable to the affinity of DOB (**16**, K_i = 32 nM), and revealed a partial agonist character in a phosphoinositide (PI) hydrolysis assay.^[164] Affinity studies on analogues of compound **26** revealed that the 2,5-dimethoxy substitution pattern is not optimal for 5-HT_{2A} receptor affinity of 4-(3-phenylpropyl)-substituted compounds. Monomethoxy and other dimethoxy derivatives evaluated in the same assay had better affinities than compound **26**, the 3,5-dimethoxy-substituted analogue was a partial agonist with 7.5-fold increased affinity (K_i = 4 nM), and 10-fold increased selectivity for the 5-HT_{2A} receptor. Various substitution patterns based on compound **26** have been synthesized and are mainly antagonistic in character.^[164,165]

Polar substituents at the 4 position generally show little affinity in the phenylalkylamine series.^[67] Addition of a β -methyl group to the side chain has been found to reduce *in vivo* activity, and this applies to β -hydroxy and β -keto groups as well.^[166,167] Interestingly, certain β -methoxy-substituted phenethylamines retain some potency in humans.^[117,145] The poor ability of β -oxygenated agents to cross the blood-brain barrier and enter the CNS is thought to account for the reduced *in vivo* activity.^[168] Addition of polar substituents on the β -position of DOB (**16**) leads to an overall reduction in lipophilicity of the molecule. Evaluation of (1*R*,2*R*)-**27** revealed a high affinity for [¹²⁵I]DOI labeled 5-HT_{2A} binding sites (K_i = 0.3 nM), and a full agonist character in a Ca²⁺-mobilization assay (EC_{50} = 0.13 μ M; 93% of 5-HT stimulation). The parent compound *R*-(–)-DOB (**16**) had a comparable affinity (K_i = 0.2 nM), and was a potent partial agonist in the same assay (EC_{50} = 0.02 μ M; 51% of 5-HT stimulation). The more polar β -hydroxy derivative (1*R*,2*R*)-**28** was tested in a DD assay with DOM-trained rats, and substituted fully for the training drug (ED_{50} = 4.3 μ mol kg^{–1}) at a potency 17-fold less than *R*-(–)-DOB (**16**, ED_{50} = 0.25 μ mol kg^{–1}). Both compounds (1*R*,2*R*)-**27** and (1*R*,2*R*)-**28** have diminished ability to enter the CNS,^[168] which is therapeutically interesting in treating ocular hypertension and glaucoma.^[18]

The stereochemical properties of the phenylisopropylamines have also been investigated. An early study revealed that the *R*-(−) isomer of 3,4-DMA was the more potent enantiomer.^[169] The publication of a convenient synthesis^[170] enabled other phenylisopropylamines to be studied; the *R*-(−) isomers of **15**–**17** were more potent than the *S*-(+) isomers.^[171] Binding studies using [³H]DOB (**16**) as a radioligand indicated *R*-(−)-**16** (K_i = 0.39 nM) as the highest affinity enantiomer, followed by (±)-**16** (K_i = 0.79 nM), and *S*-(+)-**16** (K_i = 2.3 nM).^[172] Similarly, a DD assay in the same study found *R*-(−)-**16** to be ~10 times more potent than *S*-(+)-**16**. The same pattern was seen for compound **17**, where the *R*-(−) isomer is 2–3 times as potent as the *S*-(+) enantiomer.^[109, 173] More recently, a study found *R*-(−) isomers to be more potent and more efficacious at stimulating human 5-HT_{2A} receptor-mediated PLC activation.^[152] Clearly the *R*-(−) isomers constitute the eutomeric series.

As seen with α -alkyl or β -alkyl homologues of phenylalkylamines, side chain modifications greatly influence activity at 5-HT_{2A} receptors. *N*-alkylation or *N,N*-dialkylation proved detrimental to the potency of most phenylalkylamines.^[172, 174, 175] An exception was found in a series of *N*-benzyl substituted phenylalkylamines,^[176] affinity studies comparing 2C-B (**11**) with its *N*-benzylated analogues (**29**–**31**) revealed an approximate two-fold increase in binding affinity for [¹²⁵I]DOI labeled 5-HT_{2A} receptors. Moreover compounds **29**–**31** showed increased subtype selectivity (> 100 fold) for 5-HT_{2A} over 5-HT_{2C} receptors, however the question whether **29**–**31** act as agonists or antagonists has not been addressed.^[176] Another study showed that a series of related *N*-benzyl analogues act as potent partial agonists in vascular in vitro models.^[177–179] The *N*-(2-methoxybenzyl) analogue **32** (25I-NBOMe) of 2C-I (**12**) was an extremely potent partial 5-HT_{2A} agonist (EC_{50} = 0.0813 nM; 30% of 5-HT stimulation) compared with DOI (**17**, EC_{50} = 7.41 nM; 68% of 5-HT stimulation) in the same model.^[180] A recent study evaluated a series of *N*-benzyl analogues in cell-based assays and expanded previous findings.^[181] The affinity of **32** (K_i = 0.087 nM) for cloned [¹²⁵I]DOI labeled rat 5-HT_{2A} receptors surpassed the affinity of both 2C-I (**12**, K_i = 0.62 nM), and DOI (**17**, K_i = 0.58 nM). In a functional PI hydrolysis assay using cloned human 5-HT_{2A} receptors, **32** (EC_{50} = 0.44 nM; 81% of 5-HT stimulation) exhibited an agonist character and potency well above its parent compound 2C-I (**12**, EC_{50} = 2.54 nM; 82% of 5-HT stimulation), while the *N*-(2-hydroxybenzyl) analogue **33** showed an even greater potency (EC_{50} = 0.19 nM; 86% of 5-HT stimulation). The *N*-substituted phenylalkylamines tested showed a modest 5-HT_{2A} versus 5-HT_{2C} subtype selectivity. Virtual docking of several *N*-benzyl phenylalkylamines to an activated human 5-HT_{2A} receptor homology model confirmed the previously identified interaction between the phenylalkylamine pharmacophore and Phe 340(6.52).^[137, 181] The *N*-benzyl moiety was predicted to interact with Phe 339(6.51) through a π - π interaction; site-directed mutagenesis experiments supported this prediction.^[181] This study shows *N*-benzyl phenylalkylamines to be a promising new group of 5-HT_{2A} receptor ligands with high affinity and potency.

Phenylalkylamines generally lack significant selectivity for the 5-HT_{2A} receptor over the other two subtypes. This has

been demonstrated using [¹²⁵I]DOI for 5-HT_{2A} and 5-HT_{2C}, and [³H]5-HT for 5-HT_{2B} as radioligands at cloned human receptor sites.^[63] Comparison of the binding affinities of 17 classical phenylisopropylamines showed strong correlations at the three receptor subtypes. A near unity slope was found for 5-HT_{2A} and 5-HT_{2C} receptor affinity correlations, revealing a close correspondence. The only exception was the 4-cyano analogue **24** (DOCN), with a 22-fold higher affinity for 5-HT_{2A} (K_i = 45.7 nM) over 5-HT_{2C} (K_i = 1011 nM).^[63]

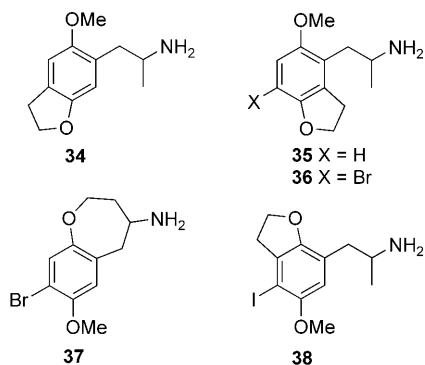
Recently, phenylalkylamines were evaluated for their functional selectivity towards PLC- and PLA₂-mediated signaling pathways.^[182] PLC-mediated inositol phosphate (IP) accumulation and PLA₂-mediated AA release was measured using CHO-K1 cells expressing human 5-HT_{2A} and 5-HT_{2C} receptors. Compared with their phenethylamine counterparts, phenylisopropylamines tested in this study showed an overall greater relative efficacy for both pathways at 5-HT_{2A} receptors. DOI (**17**) was found to be functionally selective for PLA₂ (relative efficacy ~65%) over the PLC pathway (relative efficacy ~45%). Also selective for the PLA₂ pathway was 2,5-dimethoxy-4-nitrophenethylamine (2C-N), which did not activate the PLC cascade. On the other hand, 2,5-DMA (**20**) was found to be fully selective for the PLC pathway, not activating PLA₂ at all. In the same assay, some of the other phenylalkylamines, including mescaline (**4**), 2C-I (**12**), DOM (**15**), showed no selectivity for either pathway.

In summary, the 2,5-dimethoxy-substituted phenylisopropylamines are generally the most potent of the classical phenylalkylamine 5-HT₂ receptor probes. Agonist activity increases as the nature of the 4-substituent varies from H < OR < SR < R < X, with R = alkyl, and X = halogen, while an antagonistic profile emerges with higher 4-alkyl homologues (e.g. *n*-hexyl). Hydrophobic or electron-withdrawing substituents lead to increased 5-HT₂ binding affinity. This led to a maximum with 4-*n*-hexyl- followed by 4-benzyl- and 4-bromo-substituted agents at 5-HT_{2A} receptor sites. Phenylisopropylamine ligands displayed little 5-HT₂ receptor subtype selectivity. The highly potent *N*-benzyl phenethylamines recently developed provide new SAR data and are potential selective 5-HT_{2A} receptor ligands. The differential activation of signal transduction pathways by various phenylalkylamines confirms the concept of functional selectivity, allowing the design of agonists with significant preference for a single 5-HT_{2A} receptor-coupled signaling cascade.

Novel Rigid Phenylalkylamines

Mutagenesis and molecular modeling studies can be used to design ligands with extremely high affinity or the ability to activate a specific signaling pathway. Furthermore, the steric requirements of the 5-HT_{2A} receptor binding pocket can be probed through SAR studies of rigidified ligands. Incorporation of methoxy substituents into rigid ring structures in phenylalkylamine has been used to investigate the active binding orientation of these substituents; restricting the ligand flexibility of DOM (**15**) through the synthesis of 2,3-dihydrobenzofuran analogues showed that the LSD-like activity was greatly re-

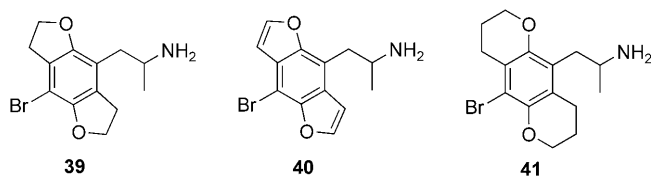
duced when the oxygen lone pair of the 5-methoxy group was directed *syn* to the alkylamine chain (dihydrofuran **34**).^[183] Another possible explanation for the diminished LSD-like activity



is unfavorable steric interactions between the ring structure and the binding site.

Orienting the oxygen lone pair *anti* to the side chain gave compound **35** and the 7-bromo analogue **36**.^[184] DD tests showed full substitution for both compounds in rats trained to discriminate LSD from saline, with the 7-bromo analogue **36** ($ED_{50}=0.57 \mu\text{mol kg}^{-1}$) more potent than DOM (**15**, $ED_{50}=0.89 \mu\text{mol kg}^{-1}$) and equipotent to DOB (**16**). Binding affinity and energy calculations confirmed that these rigid analogues model the active binding conformation of DOM (**15**).^[184] Using a tetrahydro-1-benzoxepin scaffold, the oxygen lone pair of the 2-methoxy group of DOB was oriented *anti* to the side chain, giving agents with low affinity for 5-HT_{2A} sites (e.g. **37**).^[185] These agents were around 15 times less potent than their parent compounds, suggesting that the *anti* orientation of the lone electron pairs could not be accommodated by the agonist binding site. This led to the design and synthesis of pharmacologically active compound **38**, in which the oxygen lone pair is *syn* to the alkylamine side chain.^[186]

The rigid analogue approach was used to probe the location of hydrogen bonds between phenylalkylamine ligands and the 5-HT_{2A} receptor. The results of these studies led to the synthesis and evaluation of compounds with both aromatic methoxy groups incorporated into dihydrofuran rings.^[186] The most potent ligand in this series was compound **39** ($ED_{50}=0.061 \mu\text{mol kg}^{-1}$), which was more potent than its parent



compound DOB (**16**, $ED_{50}=1.12 \mu\text{mol kg}^{-1}$) in rats trained to discriminate LSD from saline. The binding affinity of analogue **39** ($K_i=18 \text{ nM}$) was close to the affinity of DOB (**16**, $K_i=22 \text{ nM}$) at [³H]ketanserin-labeled 5-HT_{2A} receptor sites from rat frontal

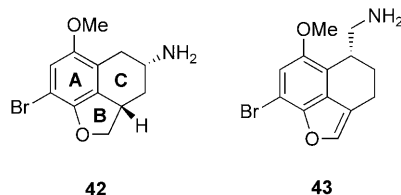
cortex homogenate. At cloned human receptor sites labeled with [¹²⁵I]DOI, compound **39** showed subnanomolar affinity for 5-HT_{2A} ($K_i=0.48 \text{ nM}$) and 5-HT_{2C} ($K_i=0.30 \text{ nM}$) receptors.^[186] The fully aromatic analogue **40** has the highest affinity for [¹²⁵I]DOI-labeled human 5-HT_{2A} ($K_i=0.04 \text{ nM}$) and 5-HT_{2C} ($K_i=0.02 \text{ nM}$) receptors.^[187] Furthermore, a DD study showed that derivative **40** ($ED_{50}=22 \text{ nmol kg}^{-1}$) was the first alkylamine derivative to surpass LSD ($ED_{50}=40 \text{ nmol kg}^{-1}$) in potency. Recently, benzodifuran analogues with decreased lipophilicity were synthesized and studied for their ability to lower IOP to treat ocular hypertension and glaucoma; while this has led to potent ligands, little subtype selectivity over the 5-HT_{2C} receptor has been achieved.^[188]

In a later study, conformationally restricted analogues were evaluated for their functional activity at 5-HT_{2A} receptors using a PI hydrolysis assay.^[189] In this assay, parent compound ($\pm$)-DOB (**15**, $EC_{50}=72 \text{ nM}$; 79% of maximal 5-HT stimulation) was less potent than *R*(-)-**39** ($EC_{50}=8.38 \text{ nM}$; 80% of maximal 5-HT stimulation) while the most potent ligand was *R*(-)-**40** ($EC_{50}=2.7 \text{ nM}$; 93% of 5-HT stimulation). As with the original phenylalkylamines, the *R*(-) enantiomer of these conformationally restricted analogues generally displayed increased activity and binding affinity. [³H]DOB labeled 5-HT_{2A} and [¹²⁵I]DOI labeled 5-HT_{2C} cloned rat receptors further confirmed this finding.^[189] None of the agents studied showed significant subtype selectivity; the CF₃ analogue of *R*(-)-**39** was found to be the most 5-HT_{2A} subtype selective, showing approximately 2.5 times higher affinity for the 5-HT_{2A} receptor over the 5-HT_{2C} receptor. The 5-HT_{2A} receptor is able to accommodate larger hexahydrobenzodipyrans analogues such as **41**, although these analogues show decreased functional activity.^[190] Various other analogues, designed using the same line of reasoning, have been synthesized and evaluated, and none of them surpass the properties of compound **40**.^[141,191] A recent study looking at differences in hydrogen bond accepting capabilities, aromaticity, and lipophilicity between compounds **40** and **39** found that lipophilicity best explains the greater in vitro and in vivo potency of **40**.^[192] Other ligands were included in the same study, but **40** was found the most lipophilic, and potent agonist.^[192]

Clearly, some of the conformationally constrained ligands effectively map the active binding conformation of the 2,5-dimethoxy phenylalkylamine pharmacophore within the 5-HT_{2A} binding pocket. Initial elucidation of the binding modes and structural analogy between 5-HT (**1**), LSD (**3**) and phenylalkylamine hallucinogens, suggested a correlation between the A ring of LSD (**3**) and the aromatic phenylalkylamine ring.^[193] A DD study looking at the behavioral effects of α -methyl and *N*-methyl substituents of indolealkylamine and phenylalkylamine hallucinogens gave consistent data.^[194] Another hypothesis proposed that the aromatic phenylalkylamine ring and the pyrrole ring B of LSD (**3**) are analogous.^[195] Both hypotheses were initially supported by SAR studies on stereochemical selectivity, and rigid DOM (**15**) analogues.^[157,196,197]

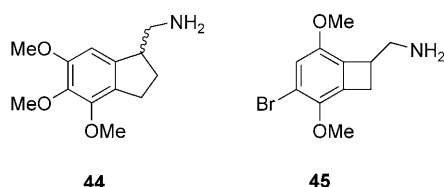
Substituted tetrahydronaphthofurans have recently been synthesized as phenylalkylamine–ergoline composite molecules designed to mimic the A, B and C ring structure of ergo-

lines.^[198] Pharmacological evaluation of compound **42** revealed binding affinities at [¹²⁵I]DOI-labeled cloned human 5-HT_{2A} (K_i = 13.0 nM), and 5-HT_{2C} (K_i = 5.96 nM) receptors, which is 20–30-



fold less than the affinity of **39** for these sites. Moreover, these compounds lacked LSD-like behavioral effects in a DD model. Based on these results, further work on structural similarities between ergolines and phenylalkylamines has ceased.^[198,199] Investigation into the 2-aminoalkyl side chain orientation led to the synthesis of analogue **43**, with a nonplanar side chain. The affinity of compound **43** (K_i = 2.6 nM), for [¹²⁵I]DOI-labeled cloned rat 5-HT_{2A} receptors, was close that of DOB (**16**, K_i = 2.2 nM) and LSD (**3**, K_i = 3.5 nM). In a functional assay measuring IP₃ accumulation, compound **43** was shown to be a partial agonist (EC_{50} = 120 nM; 33 % of 5-HT stimulation), and it displayed partial substitution in LSD and DOI-trained rats.^[199] One conclusion is that the side chain of ligand **43** might not possess the optimal dihedral angle for full receptor activation.

Extending previous work on indanalkylamine analogues of DOM (**15**),^[197] a conformationally constrained 1-aminomethylindan analogue (**44**) of mescaline (**4**) was recently designed^[200]



using the aforementioned in silico activated 5-HT_{2A} homology model.^[137] Analogue **44** showed a threefold increase in affinity (K_i = 130 nM), and a twofold increase in stimulating IP₃ accumulation (EC_{50} = 6100 nM) compared to mescaline (**4**, K_i = 360 nM; EC_{50} = 11 300 nM). The *R*-(–)-**44** enantiomer showed a twofold increase in affinity (K_i = 69 nM) and potency (EC_{50} = 3200 nM) over the racemate (±)-**44**. Further testing showed both compounds (±)-**44** and *R*-(–)-**44** are comparable to the parent compound; both fully substituted for LSD (**3**) in a DD assay, and their stereoselective profile was in line with docking experiments. The 5-HT_{2A} receptor binding site can accommodate the out-of-plane conformation of the alkylamine side chain. Subsequent work to elucidate the active binding orientation of the side chain used rigid benzocycloalkyl-1-methylamines analogues of 2C-B (**11**).^[201] Ligand *R*-(–)-**45** showed a threefold higher affinity (K_i = 0.26 nM) for [¹²⁵I]DOI-labeled cloned human 5-HT_{2A} receptor sites than its parent compound 2C-B (**11**, K_i = 0.88 nM). In a DD assay using LSD trained rats, enantiomer *R*-(–)-**45** (ED_{50} = 24 nmol kg^{–1}) surpassed DOI (**17**, ED_{50} = 270 nmol kg^{–1}) and LSD (**3**, ED_{50} = 38 nmol kg^{–1}). In rats trained to discriminate DOI (**17**) from saline, LSD (**3**, ED_{50} = 15 nmol kg^{–1}) was found to be more potent than *R*-(–)-**45** (ED_{50} = 24 nmol kg^{–1}). These results, supported by receptor docking simulations, suggest that when bound to the receptor, the side chain lies in a perpendicular plane relative to the aromatic ring.^[201]

Evaluation of the functional activity of *R*-(–)-**45** in NIH 3T3 cells expressing rat 5-HT_{2A} receptors, revealed a 65-fold selectivity for IP₃ accumulation (EC_{50} = 18 nM; 97% of 5-HT stimulation) over AA (EC_{50} = 1180 nM), and 2-AG (EC_{50} = 1120 nM) production,^[201] clearly showing functional selectivity for the PLC pathway over the PLA₂ cascade. However, activation of the PLC signaling pathway alone does not account for the stimulus effects of hallucinogens;^[202] PLA₂ stimulation by a ligand correlates better with hallucinogenic properties.^[90] Although compound **45** fully substitutes the training drug in DD studies, hallucinogenic effects in humans have not been confirmed. Similar phenylalkylamines with significant functional selectivity for a single pathway may prove to be nonhallucinogenic selective 5-HT_{2A} receptor agonists. In addition to PLC and PLA₂ signaling, the role of PLD signaling in the behavioral effects of phenylalkylamines remains to be elucidated.^[46,182] While the 5-HT_{2A}-coupled signaling pathways are complex, and a significant amount of research into the functional selectivity of ligands remains to be done, preliminary results show that it might be possible to develop phenylalkylamine agonists selective for the 5-HT_{2A} subtype, and for a specific signaling pathway.

Conclusions and Future Directions

A large number of phenylalkylamine agonists for the 5-HT_{2A} receptor are currently recognized. New ligands have been designed through careful SAR evaluation of the phenylalkylamine pharmacophore, facilitated by comparative molecular modeling of ligand–receptor complexes, and data from site-directed mutagenesis studies. The rigid analogue approach proved useful in probing the binding site, elucidating optimal binding conformations, and has led to the synthesis of ligands with exceptional pharmacological profiles. As yet, none of the ligands designed show significant selectivity for the 5-HT_{2A} receptor subtype, confirming the similarity with the 5-HT_{2C} binding pocket. Selectivity towards specific signaling pathways adds another level of complexity.

5-HT_{2A} receptor agonists are important research tools for neuroscience and other disciplines, essential for the study of neurochemistry, neurobiology and neurophysiology. Moreover, subtype and functionally selective ligands may hold valuable therapeutic potential. Functionally selective, signal pathway specific ligands that are nonhallucinogenic in nature, might eventually be used to augment pharmacological psychiatric therapies for depression^[36] and anxiety.^[38,39] In cognitive pharmacology, these agents might prove useful in memory,^[25] learning,^[26] and BDNF-mediated processes.^[27] In the periphery, ligands with reduced ability to enter the CNS may be used to study the cardiovascular system,^[80] and represent a novel ther-

apy for glaucoma and ocular hypertension.^[18] With the recognition of DOI-mediated liver regeneration,^[35] the study of ligands with an agonist profile at 5-HT_{2A} receptors is far from over.

Keywords: 5-HT_{2A} receptors • drug design • phenylalkylamines • serotonin • structure–activity relationships

- [1] D. Hoyer, J. P. Hannon, G. R. Martin, *Pharmacol. Biochem. Behav.* **2002**, *71*, 533–554.
- [2] B. J. Jones, T. P. Blackburn, *Pharmacol. Biochem. Behav.* **2002**, *71*, 555–568.
- [3] N. M. Barnes, T. Sharp, *Neuropharmacology* **1999**, *38*, 1083–1152.
- [4] D. Hoyer, D. E. Clarke, J. R. Fozard, P. R. Hartig, G. R. Martin, E. J. Mylecharane, P. R. Saxena, P. P. Humphrey, *Pharmacol. Rev.* **1994**, *46*, 157–203.
- [5] G. Baxter, G. Kennett, F. Blaney, T. Blackburn, *Trends Pharmacol. Sci.* **1995**, *16*, 105–110.
- [6] F. G. Boess, I. L. Martin, *Neuropharmacology* **1994**, *33*, 275–317.
- [7] J. F. López-Giménez, G. Mengod, J. M. Palacios, M. T. Vilaró, *Naunyn-Schmiedeberg's Arch. Pharmacol.* **1997**, *356*, 446–454.
- [8] V. Cornea-Hébert, K. C. Watkins, B. L. Roth, W. K. Kroeze, P. Gaudreau, N. Leclerc, L. Descarries, *Neuroscience* **2002**, *113*, 23–35.
- [9] L. A. Miner, J. R. Backstrom, E. Sanders-Bush, S. R. Sesack, *Neuroscience* **2003**, *116*, 107–117.
- [10] M. Amargos-Bosch, A. Bortolozzi, M. V. Puig, J. Serrats, A. Adell, P. Celada, M. Toth, G. Mengod, F. Artigas, *Cereb. Cortex* **2004**, *14*, 281–299.
- [11] N. Santana, A. Bortolozzi, J. Serrats, G. Mengod, F. Artigas, *Cereb. Cortex* **2004**, *14*, 1100–1109.
- [12] P. W. Burnet, S. L. Eastwood, K. Lacey, P. J. Harrison, *Brain Res.* **1995**, *676*, 157–168.
- [13] D. A. Morilak, R. D. Ciaranello, *Brain Res.* **1993**, *627*, 49–54.
- [14] D. A. Morilak, S. J. Garlow, R. D. Ciaranello, *Neuroscience* **1993**, *54*, 701–717.
- [15] M. Lüttgen, S. Ove Ögren, B. Meister, *Brain Res.* **2004**, *1010*, 156–165.
- [16] T. Xu, S. C. Pandey, *Brain Res. Bull.* **2000**, *51*, 499–505.
- [17] J. E. Leysen, *Curr. Drug Targets: CNS Neurol. Disord.* **2004**, *3*, 11–26.
- [18] J. A. May, M. A. McLaughlin, N. A. Sharif, M. R. Hellberg, T. R. Dean, *J. Pharmacol. Exp. Ther.* **2003**, *306*, 301–309.
- [19] N. A. Sharif, C. R. Kelly, J. Y. Crider, T. L. Davis, *J. Ocul. Pharmacol. Ther.* **2006**, *22*, 389–401.
- [20] P. Bonaventure, D. Nepomuceno, K. Miller, J. Chen, C. Kuei, F. Kamme, D. T. Tran, T. W. Lovenberg, C. Liu, *Eur. J. Pharmacol.* **2005**, *513*, 181–192.
- [21] C. S. Elangbam, R. M. Lightfoot, L. W. Yoon, D. R. Creech, R. S. Geske, C. W. Crumley, L. D. Gates, H. G. Wall, *J. Histochem. Cytochem.* **2005**, *53*, 671–677.
- [22] D. S. Choi, L. Maroteaux, *FEBS Lett.* **1996**, *391*, 45–51.
- [23] M. Pompeiano, J. M. Palacios, G. Mengod, *Mol. Brain Res.* **1994**, *23*, 163–178.
- [24] L. D. Van de Kar, A. Javed, Y. Zhang, F. Serres, D. K. Raap, T. S. Gray, *J. Neurosci.* **2001**, *21*, 3572–3579.
- [25] G. V. Williams, S. G. Rao, P. S. Goldman-Rakic, *J. Neurosci.* **2002**, *22*, 2843–2854.
- [26] J. A. Harvey, *Learn. Mem.* **2003**, *10*, 355–362.
- [27] V. A. Vaidya, G. J. Marek, G. K. Aghajanian, R. S. Duman, *J. Neurosci.* **1997**, *17*, 2785–2795.
- [28] J. E. Leysen, D. de Chaffoy de Courcelles, F. De Clerck, C. J. Niemegeers, J. M. Van Nueten, *Neuropharmacology* **1984**, *23*, 1493–1501.
- [29] D. S. Choi, S. J. Ward, N. Messaddeq, J. M. Launay, L. Maroteaux, *Development* **1997**, *124*, 1745–1755.
- [30] C. G. Nebigil, D. S. Choi, A. Dierich, P. Hickel, M. Le Meur, N. Messaddeq, J. M. Launay, L. Maroteaux, *Proc. Natl. Acad. Sci. USA* **2000**, *97*, 9508–9513.
- [31] C. G. Nebigil, J. M. Launay, P. Hickel, C. Tournois, L. Maroteaux, *Proc. Natl. Acad. Sci. USA* **2000**, *97*, 2591–2596.
- [32] B. M. Nilsson, *J. Med. Chem.* **2006**, *49*, 4023–4034.
- [33] M. Bancila, D. Verge, O. Rampin, J. R. Backstrom, E. Sanders-Bush, K. E. McKenna, L. Marson, A. Calas, F. Giuliano, *Neuroscience* **1999**, *92*, 1523–1537.
- [34] Y. Kimura, K. Hatanaka, Y. Naitou, K. Maeno, I. Shimada, A. Koakutsu, F. Wanibuchi, T. Yamaguchi, *Eur. J. Pharmacol.* **2004**, *483*, 37–43.
- [35] M. Lesurtel, R. Graf, B. Aleil, D. J. Walther, Y. Tian, W. Jochum, C. Gachet, M. Bader, P. A. Clavien, *Science* **2006**, *312*, 104–107.
- [36] B. A. Nic Dhonnchadha, N. Ripoll, F. Clenet, M. Hascoet, M. Bourin, *Psychopharmacology* **2005**, *179*, 418–429.
- [37] M. J. Millan, *Prog. Neurobiol.* **2003**, *70*, 83–244.
- [38] B. A. Nic Dhonnchadha, M. Bourin, M. Hascoet, *Behav. Brain Res.* **2003**, *140*, 203–214.
- [39] N. V. Weisstaub, M. Zhou, A. Lira, E. Lambe, J. Gonzalez-Maeso, J. P. Hornung, E. Sibille, M. Underwood, S. Itohara, W. T. Dauer, M. S. Ansorge, E. Morelli, J. J. Mann, M. Toth, G. Aghajanian, S. C. Sealfon, R. Hen, J. A. Gingrich, *Science* **2006**, *313*, 536–540.
- [40] F. A. Moreno, C. B. Wiegand, E. K. Taitano, P. L. Delgado, *J. Clin. Psychiatry* **2006**, *67*, 1735–1740.
- [41] G. K. Aghajanian, G. J. Marek, *Brain Res. Rev.* **2000**, *31*, 302–312.
- [42] M. Naughton, J. B. Mulrooney, B. E. Leonard, *Hum. Psychopharmacol.* **2000**, *15*, 397–415.
- [43] D. M. Weiner, E. S. Burstein, N. Nash, G. E. Croston, E. A. Currier, K. E. Vanover, S. C. Harvey, E. Donohue, H. C. Hansen, C. M. Andersson, T. A. Spalding, D. F. Gibson, K. Krebs-Thomson, S. B. Powell, M. A. Geyer, U. Hacksell, M. R. Brann, *J. Pharmacol. Exp. Ther.* **2001**, *299*, 268–276.
- [44] H. Y. Meltzer, *Neuropsychopharmacology* **1999**, *21*, S106–S115.
- [45] F. X. Vollenweider, M. F. Vollenweider-Scherpenhuyzen, A. Babler, H. Vogel, D. Hell, *Neuroreport* **1998**, *9*, 3897–3902.
- [46] D. E. Nichols, *Pharmacol. Ther.* **2004**, *101*, 131–181.
- [47] R. A. Glennon, R. Young, J. A. Rosecrans, *Eur. J. Pharmacol.* **1983**, *91*, 189–196.
- [48] R. A. Glennon, M. Titeler, J. D. McKenney, *Life Sci.* **1984**, *35*, 2505–2511.
- [49] G. K. Aghajanian, G. J. Marek, *Neuropsychopharmacology* **1999**, *21*, S16–S23.
- [50] a) R. R. Griffiths, W. A. Richards, U. McCann, R. Jesse, *Psychopharmacology* **2006**, *187*, 268–283; b) R. R. Griffiths, W. A. Richards, U. McCann, R. Jesse, *Psychopharmacology* **2006**, *187*, 284–292.
- [51] J. A. May, H. H. Chen, A. Rusinko, V. M. Lynch, N. A. Sharif, M. A. McLaughlin, *J. Med. Chem.* **2003**, *46*, 4188–4195.
- [52] J. A. May, A. P. Dantanarayana, P. W. Zinke, M. A. McLaughlin, N. A. Sharif, *J. Med. Chem.* **2006**, *49*, 318–328.
- [53] J. D. Perkins, *Liver Transpl.* **2006**, *12*, 1431–1434.
- [54] G. Jeyabalan, D. A. Geller, *J. Hepatol.* **2006**, *45*, 629–630.
- [55] D. K. Arulmozhi, A. Veeranjanyulu, S. L. Bodhankar, *Vascul. Pharmacol.* **2005**, *43*, 176–187.
- [56] R. B. Westkaemper, R. A. Glennon, *Curr. Top. Med. Chem.* **2002**, *2*, 575–598.
- [57] T. Heinrich, H. Bottcher, H. Prucher, R. Gottschlich, K. A. Ackermann, C. van Amsterdam, *ChemMedChem* **2006**, *1*, 245–255.
- [58] G. Poissonnet, J. G. Parmentier, J. A. Boutin, S. Goldstein, *Mini-Rev. Med. Chem.* **2004**, *4*, 325–330.
- [59] R. H. Porter, K. R. Benwell, H. Lamb, C. S. Malcolm, N. H. Allen, D. F. Revell, D. R. Adams, M. J. Sheardown, *Br. J. Pharmacol.* **1999**, *128*, 13–20.
- [60] G. S. Baxter, *Behav. Brain Res.* **1996**, *73*, 149–152.
- [61] R. A. Glennon, *NIDA Res. Monogr.* **1994**, *146*, 4–32.
- [62] B. L. Roth, E. G. Hyde in *Serotonergic Neurons and 5-HT Receptors in the CNS* (Eds.: H. G. Baumgarten, M. Göthert), Springer, Berlin, **1997**, pp. 367–394.
- [63] D. L. Nelson, V. L. Lucaites, D. B. Wainscott, R. A. Glennon, *Naunyn-Schmiedeberg's Arch. Pharmacol.* **1999**, *359*, 1–6.
- [64] P. Brawley, J. C. Duffield, *Pharmacol. Rev.* **1972**, *24*, 31–66.
- [65] D. E. Nichols, *J. Pharm. Sci.* **1981**, *70*, 839–849.
- [66] R. A. Glennon, M. Dukat, *Pharmacol. Biochem. Behav.* **1991**, *40*, 1009–1017.
- [67] M. R. Seggel, M. Y. Yousif, R. A. Lyon, M. Titeler, B. L. Roth, E. A. Suba, R. A. Glennon, *J. Med. Chem.* **1990**, *33*, 1032–1036.
- [68] T. Kenakin, *Nat. Rev. Drug. Discovery* **2005**, *4*, 919–927.
- [69] C. Liebmann, *Curr. Pharm. Des.* **2004**, *10*, 1937–1958.
- [70] D. M. Perez, S. S. Karnik, *Pharmacol. Rev.* **2005**, *57*, 147–161.

- [71] J. D. Urban, W. P. Clarke, M. von Zastrow, D. E. Nichols, B. Kobilka, H. Weinstein, J. A. Javitch, B. L. Roth, A. Christopoulos, P. M. Sexton, K. J. Miller, M. Spedding, R. B. Mailman, *J. Pharmacol. Exp. Ther.* **2007**, 320, 1–13.
- [72] A. Abbas, B. L. Roth, *Proc. Natl. Acad. Sci. USA* **2008**, 105, 831–832.
- [73] D. Van Oekelen, W. H. Luyten, J. E. Leysen, *Life Sci.* **2003**, 72, 2429–2449.
- [74] T. Kenakin, *Nat. Rev. Drug Discovery* **2002**, 1, 103–110.
- [75] T. Kenakin, *Trends Pharmacol. Sci.* **1995**, 16, 232–238.
- [76] S. Galandrin, G. Oligny-Longpre, M. Bouvier, *Trends Pharmacol. Sci.* **2007**, 28, 423–430.
- [77] B. L. Roth, T. Nakaki, D. M. Chuang, E. Costa, *Neuropharmacology* **1984**, 23, 1223–1225.
- [78] C. C. Felder, R. Y. Kanterman, A. L. Ma, J. Axelrod, *Proc. Natl. Acad. Sci. USA* **1990**, 87, 2187–2191.
- [79] K. A. Berg, S. Maayani, J. Goldfarb, C. Scaramellini, P. Leff, W. P. Clarke, *Mol. Pharmacol.* **1998**, 54, 94–104.
- [80] T. Nagatomo, M. Rashid, H. Abul Muntasir, T. Komiyama, *Pharmacol. Ther.* **2004**, 104, 59–81.
- [81] R. L. Williams, *Biochim. Biophys. Acta* **1999**, 1441, 255–267.
- [82] B. L. Roth, D. L. Willins, K. Kristiansen, W. K. Kroeze, *Pharmacol. Ther.* **1998**, 79, 231–257.
- [83] M. J. Berridge, *Nature* **1993**, 361, 315–325.
- [84] M. J. Berridge, M. D. Bootman, H. L. Roderick, *Nat. Rev. Mol. Cell Biol.* **2003**, 4, 517–529.
- [85] Y. Nishizuka, *Science* **1986**, 233, 305–312.
- [86] K. P. Becker, Y. A. Hannun, *Cell. Mol. Life Sci.* **2005**, 62, 1448–1461.
- [87] N. Stella, P. Schweitzer, D. Piomelli, *Nature* **1997**, 388, 773–778.
- [88] D. Piomelli, *Nat. Rev. Neurosci.* **2003**, 4, 873–884.
- [89] J. C. Parrish, D. E. Nichols, *J. Neurochem.* **2006**, 99, 1164–1175.
- [90] D. M. Kurrasch-Orbaugh, V. J. Watts, E. L. Barker, D. E. Nichols, *J. Pharmacol. Exp. Ther.* **2003**, 304, 229–237.
- [91] D. M. Kurrasch-Orbaugh, J. C. Parrish, V. J. Watts, D. E. Nichols, *J. Neurochem.* **2003**, 86, 980–991.
- [92] R. Mitchell, D. McCulloch, E. Lutz, M. Johnson, C. MacKenzie, M. Fennell, G. Fink, W. Zhou, S. C. Sealfon, *Nature* **1998**, 392, 411–414.
- [93] G. M. Jenkins, M. A. Frohman, *Cell. Mol. Life Sci.* **2005**, 62, 2305–2316.
- [94] D. N. Robertson, M. S. Johnson, L. O. Moggach, P. J. Holland, E. M. Lutz, R. Mitchell, *Mol. Pharmacol.* **2003**, 64, 1239–1250.
- [95] M. McDermott, M. J. Wakelam, A. J. Morris, *Biochem. Cell Biol.* **2004**, 82, 225–253.
- [96] R. A. Leslie, J. M. Moorman, A. Coulson, D. G. Grahame-Smith, *Neuroscience* **1993**, 53, 457–463.
- [97] N. Tilakaratne, E. Friedman, *Eur. J. Pharmacol.* **1996**, 307, 211–217.
- [98] Q. Pei, L. Lewis, M. E. Sprakes, E. J. Jones, D. G. Grahame-Smith, T. S. Zetterstrom, *Neuropharmacology* **2000**, 39, 463–470.
- [99] J. L. Scruggs, S. Patel, M. Bubser, A. Y. Deutch, *J. Neurosci.* **2000**, 20, 8846–8852.
- [100] V. L. Arvanov, X. Liang, A. Russo, R. Y. Wang, *Eur. J. Neurosci.* **1999**, 11, 3064–3072.
- [101] D. E. Nichols, S. Frescas, D. Marona-Lewicka, D. M. Kurrasch-Orbaugh, *J. Med. Chem.* **2002**, 45, 4344–4349.
- [102] C. D. Nichols, E. Sanders-Bush, *Neuropsychopharmacology* **2002**, 26, 634–642.
- [103] C. D. Nichols, E. E. Garcia, E. Sanders-Bush, *Mol. Brain Res.* **2003**, 111, 182–188.
- [104] J. González-Maeso, T. Yuen, B. J. Ebersole, E. Wurmbach, A. Lira, M. Zhou, N. Weisstaub, R. Hen, J. A. Gingrich, S. C. Sealfon, *J. Neurosci.* **2003**, 23, 8836–8843.
- [105] J. González-Maeso, N. V. Weisstaub, M. Zhou, P. Chan, L. Ivic, R. Ang, A. Lira, M. Bradley-Moore, Y. Ge, Q. Zhou, S. C. Sealfon, J. A. Gingrich, *Neuron* **2007**, 53, 439–452.
- [106] J. Song, D. Hanniford, C. Doucette, E. Graham, M. F. Poole, A. Ting, B. Sherf, J. Harrington, K. Brunden, A. Stricker-Krongrad, *Assay Drug Dev. Technol.* **2005**, 3, 649–659.
- [107] J. E. Leysen, C. J. Niemegeers, J. M. Van Nueten, P. M. Laduron, *Mol. Pharmacol.* **1982**, 21, 301–314.
- [108] M. Titeler, K. Herrick, R. A. Lyon, J. D. McKenney, R. A. Glennon, *Eur. J. Pharmacol.* **1985**, 117, 145–146.
- [109] M. P. Johnson, A. J. Hoffman, D. E. Nichols, C. A. Mathis, *Neuropharmacology* **1987**, 26, 1803–1806.
- [110] J. F. López-Giménez, M. Villazón, J. Brea, M. I. Loza, J. M. Palacios, G. Mengod, M. T. Vilaró, *Mol. Pharmacol.* **2001**, 60, 690–699.
- [111] R. J. Lefkowitz, S. Cotecchia, P. Samama, T. Costa, *Trends Pharmacol. Sci.* **1993**, 14, 303–307.
- [112] P. Leff, *Trends Pharmacol. Sci.* **1995**, 16, 89–97.
- [113] T. Kenakin, *Trends Pharmacol. Sci.* **2004**, 25, 186–192.
- [114] W. P. Clarke, R. A. Bond, *Trends Pharmacol. Sci.* **1998**, 19, 270–276.
- [115] W. E. Fantegrossi, K. S. Murnane, C. J. Reissig, *Biochem. Pharmacol.* **2008**, 75, 17–33.
- [116] R. A. Glennon, *NIDA Res. Monogr.* **1991**, 116, 25–44.
- [117] A. Shulgin, A. Shulgin, *PIHKAL: A Chemical Love Story*, Transform, Berkeley, **1991**, p. 978.
- [118] A. M. Van Rhee, K. A. Jacobson, *Drug Dev. Res.* **1996**, 37, 1–38.
- [119] K. Kristiansen, *Pharmacol. Ther.* **2004**, 103, 21–80.
- [120] C. D. Wang, T. K. Gallaher, J. C. Shih, *Mol. Pharmacol.* **1993**, 43, 931–940.
- [121] M. S. Choudhary, S. Craig, B. L. Roth, *Mol. Pharmacol.* **1993**, 43, 755–761.
- [122] M. S. Choudhary, N. Sachs, A. Uluer, R. A. Glennon, R. B. Westkaemper, B. L. Roth, *Mol. Pharmacol.* **1995**, 47, 450–457.
- [123] I. Visiers, J. A. Ballesteros, H. Weinstein, *Methods Enzymol.* **2002**, 343, 329–371.
- [124] N. Almula, B. J. Ebersole, D. Zhang, H. Weinstein, S. C. Sealfon, *J. Biol. Chem.* **1996**, 271, 14672–14675.
- [125] M. P. Johnson, R. J. Loncharich, M. Baez, D. L. Nelson, *Mol. Pharmacol.* **1994**, 45, 277–286.
- [126] N. Almula, B. J. Ebersole, J. A. Ballesteros, H. Weinstein, S. C. Sealfon, *Mol. Pharmacol.* **1996**, 50, 34–42.
- [127] S. Trumpp-Kallmeyer, J. Hoflack, A. Bruinvels, M. Hibert, *J. Med. Chem.* **1992**, 35, 3448–3462.
- [128] E. Jacoby, R. Bouhelal, M. Gerspacher, K. Seuwen, *ChemMedChem* **2006**, 1, 761–782.
- [129] R. B. Westkaemper, R. A. Glennon, *NIDA Res. Monogr.* **1994**, 146, 263–283.
- [130] K. Palczewski, T. Kumasaka, T. Hori, C. A. Behnke, H. Motoshima, B. A. Fox, I. Le Trong, D. C. Teller, T. Okada, R. E. Stenkamp, M. Yamamoto, M. Miyano, *Science* **2000**, 289, 739–745.
- [131] D. C. Teller, T. Okada, C. A. Behnke, K. Palczewski, R. E. Stenkamp, *Biochemistry* **2001**, 40, 7761–7772.
- [132] T. Okada, Y. Fujiyoshi, M. Silow, J. Navarro, E. M. Landau, Y. Shichida, *Proc. Natl. Acad. Sci. USA* **2002**, 99, 5982–5987.
- [133] T. Huber, A. V. Botelho, K. Beyer, M. F. Brown, *Biophys. J.* **2004**, 86, 2078–2100.
- [134] K. Kristiansen, W. K. Kroeze, D. L. Willins, E. I. Gelber, J. E. Savage, R. A. Glennon, B. L. Roth, *J. Pharmacol. Exp. Ther.* **2000**, 293, 735–746.
- [135] D. A. Shapiro, K. Kristiansen, W. K. Kroeze, B. L. Roth, *Mol. Pharmacol.* **2000**, 58, 877–886.
- [136] W. K. Kroeze, K. Kristiansen, B. L. Roth, *Curr. Top. Med. Chem.* **2002**, 2, 507–528.
- [137] J. J. Chambers, D. E. Nichols, *J. Comput.-Aided Mol. Des.* **2002**, 16, 511–520.
- [138] B. J. Ebersole, I. Visiers, H. Weinstein, S. C. Sealfon, *Mol. Pharmacol.* **2003**, 63, 36–43.
- [139] M. R. Braden, D. E. Nichols, *Mol. Pharmacol.* **2007**, 72, 1200–1209.
- [140] U. Braun, G. Braun, P. Jacob III, D. E. Nichols, A. T. Shulgin, *NIDA Res. Monogr.* **1978**, 22, 27–37.
- [141] A. P. Monte, S. R. Waldman, D. Marona-Lewicka, D. B. Wainwright, D. L. Nelson, E. Sanders-Bush, D. E. Nichols, *J. Med. Chem.* **1997**, 40, 2997–3008.
- [142] P. Jacob III, A. T. Shulgin, *J. Med. Chem.* **1984**, 27, 881–888.
- [143] P. Jacob III, A. T. Shulgin, *J. Med. Chem.* **1981**, 24, 1348–1353.
- [144] A. T. Shulgin, M. F. Carter, *Psychopharmacol. Commun.* **1975**, 1, 93–98.
- [145] P. Jacob III, A. T. Shulgin, *NIDA Res. Monogr.* **1994**, 146, 74–91.
- [146] A. T. Shulgin, T. Sargent, C. Naranjo, *Pharmacology* **1971**, 5, 103–107.
- [147] C. F. Barfknecht, D. E. Nichols, *J. Med. Chem.* **1971**, 14, 370–372.
- [148] R. A. Glennon, R. Young, F. Benington, R. D. Morin, *J. Med. Chem.* **1982**, 25, 1163–1168.
- [149] R. A. Glennon, *NIDA Res. Monogr.* **1989**, 94, 43–67.
- [150] M. Shannon, G. Battaglia, R. A. Glennon, M. Titeler, *Eur. J. Pharmacol.* **1984**, 102, 23–29.

- [151] D. E. Nichols, S. Frescas, D. Marona-Lewicka, X. Huang, B. L. Roth, G. A. Gudelsky, J. F. Nash, *J. Med. Chem.* **1994**, *37*, 4346–4351.
- [152] J. C. Parrish, M. R. Braden, E. Gundy, D. E. Nichols, *J. Neurochem.* **2005**, *95*, 1575–1584.
- [153] A. T. Shulgin, *Experientia* **1963**, *19*, 127–128.
- [154] R. T. Standridge, H. G. Howell, J. A. Gylys, R. A. Partyka, A. T. Shulgin, *J. Med. Chem.* **1976**, *19*, 1400–1404.
- [155] C. F. Barfknecht, J. F. Caputo, M. B. Tobin, D. C. Dyer, R. T. Standridge, H. G. Howell, W. R. Goodwin, R. A. Partyka, J. A. Gylys, R. L. Cavanagh, *NIDA Res. Monogr.* **1978**, *22*, 16–26.
- [156] H. J. Weintraub, D. E. Nichols, A. Makriyannis, S. W. Fesik, *J. Med. Chem.* **1980**, *23*, 339–341.
- [157] D. E. Nichols, R. Woodard, B. A. Hathaway, M. T. Lowy, K. W. Yom, *J. Med. Chem.* **1979**, *22*, 458–460.
- [158] D. E. Nichols, K. P. Jadhav, R. A. Oberlender, J. E. Zabik, J. F. Bossart, A. Hamada, D. D. Miller, *J. Med. Chem.* **1984**, *27*, 1108–1111.
- [159] A. T. Shulgin, T. Sargent, C. Naranjo, *Nature* **1969**, *221*, 537–541.
- [160] A. T. Shulgin, *Experientia* **1964**, *20*, 366–367.
- [161] R. A. Glennon, R. Young, *Pharmacol. Biochem. Behav.* **1982**, *17*, 603–607.
- [162] R. A. Glennon, R. Raghupathi, P. Bartyzel, M. Teitler, S. Leonhardt, *J. Med. Chem.* **1992**, *35*, 734–740.
- [163] A. T. Shulgin, D. C. Dyer, *J. Med. Chem.* **1975**, *18*, 1201–1204.
- [164] C. S. Dowd, K. Herrick-Davis, C. Egan, A. DuPre, C. Smith, M. Teitler, R. A. Glennon, *J. Med. Chem.* **2000**, *43*, 3074–3084.
- [165] J. B. Rangisetty, M. Dukat, C. S. Dowd, K. Herrick-Davis, A. DuPre, S. Gadepalli, M. Teitler, C. R. Kelley, N. A. Sharif, R. A. Glennon, *J. Med. Chem.* **2001**, *44*, 3283–3291.
- [166] F. A. Aldous, B. C. Barrass, K. Brewster, D. A. Buxton, D. M. Green, R. M. Pinder, P. Rich, M. Skeels, K. J. Tutt, *J. Med. Chem.* **1974**, *17*, 1100–1111.
- [167] R. A. Glennon, S. M. Liebowitz, G. M. Anderson III, *J. Med. Chem.* **1980**, *23*, 294–299.
- [168] R. A. Glennon, M. L. Bondarev, N. Khorana, R. Young, J. A. May, M. R. Hellberg, M. A. McLaughlin, N. A. Sharif, *J. Med. Chem.* **2004**, *47*, 6034–6041.
- [169] C. F. Barfknecht, D. E. Nichols, *J. Med. Chem.* **1972**, *15*, 109–110.
- [170] D. E. Nichols, C. F. Barfknecht, D. B. Rusterholz, F. Benington, R. D. Morin, *J. Med. Chem.* **1973**, *16*, 480–483.
- [171] D. C. Dyer, D. E. Nichols, D. B. Rusterholz, C. F. Barfknecht, *Life Sci.* **1973**, *13*, 885–896.
- [172] R. A. Glennon, M. Titeler, M. R. Seggel, R. A. Lyon, *J. Med. Chem.* **1987**, *30*, 930–932.
- [173] R. A. Glennon, J. D. McKenney, R. A. Lyon, M. Titeler, *J. Med. Chem.* **1986**, *29*, 194–199.
- [174] D. E. Nichols, *Pharmacol. Biochem. Behav.* **1986**, *24*, 335–340.
- [175] R. A. Glennon, S. M. Liebowitz, E. C. Mack, *J. Med. Chem.* **1978**, *21*, 822–825.
- [176] R. A. Glennon, M. Dukat, M. el-Bermawy, H. Law, J. De Los Angeles, M. Teitler, A. King, K. Herrick-Davis, *J. Med. Chem.* **1994**, *37*, 1929–1935.
- [177] H. H. Pertz, A. Rheineck, S. Elz, *Naunyn-Schmiedeberg's Arch. Pharmacol.* **1999**, *359*, R29.
- [178] S. Elz, T. Kläß, R. Heim, U. Warnke, H. H. Pertz, *Naunyn-Schmiedeberg's Arch. Pharmacol.* **2002**, *365*, R29.
- [179] R. Heim, H. H. Pertz, M. Zabel, S. Elz, *Arch. Pharm. Pharm. Med. Chem.* **2002**, *335*, 82.
- [180] R. Heim, PhD thesis, Freien Universität Berlin, Berlin (Germany), **2003**.
- [181] M. R. Braden, J. C. Parrish, J. C. Naylor, D. E. Nichols, *Mol. Pharmacol.* **2006**, *70*, 1956–1964.
- [182] P. R. Moya, K. A. Berg, M. A. Gutierrez-Hernandez, P. Saez-Briones, M. Reyes-Parada, B. K. Cassels, W. P. Clarke, *J. Pharmacol. Exp. Ther.* **2007**, *321*, 1054–1061.
- [183] D. E. Nichols, A. J. Hoffman, R. A. Oberlender, R. M. Riggs, *J. Med. Chem.* **1986**, *29*, 302–304.
- [184] D. E. Nichols, S. E. Snyder, R. Oberlender, M. P. Johnson, X. M. Huang, *J. Med. Chem.* **1991**, *34*, 276–281.
- [185] A. P. Monte, D. Marona-Lewicka, N. V. Cozzi, D. L. Nelson, D. E. Nichols, *Med. Chem. Res.* **1995**, *5*, 651–663.
- [186] A. P. Monte, D. Marona-Lewicka, M. A. Parker, D. B. Wainscott, D. L. Nelson, D. E. Nichols, *J. Med. Chem.* **1996**, *39*, 2953–2961.
- [187] M. A. Parker, D. Marona-Lewicka, V. L. Lucaites, D. L. Nelson, D. E. Nichols, *J. Med. Chem.* **1998**, *41*, 5148–5149.
- [188] Z. Feng, S. Mohapatra, P. G. Klimko, M. R. Hellberg, J. A. May, C. Kelly, G. Williams, M. A. McLaughlin, N. A. Sharif, *Bioorg. Med. Chem. Lett.* **2007**, *17*, 2998–3002.
- [189] J. J. Chambers, D. M. Kurrasch-Orbaugh, M. A. Parker, D. E. Nichols, *J. Med. Chem.* **2001**, *44*, 1003–1010.
- [190] M. S. Whiteside, D. Kurrasch-Orbaugh, D. Marona-Lewicka, D. E. Nichols, A. Monte, *Bioorg. Med. Chem.* **2002**, *10*, 3301–3306.
- [191] J. J. Chambers, D. M. Kurrasch-Orbaugh, D. E. Nichols, *Bioorg. Med. Chem. Lett.* **2002**, *12*, 1997–1999.
- [192] M. A. Parker, D. M. Kurrasch, D. E. Nichols, *Bioorg. Med. Chem.* **2008**, *16*, 4661–4669.
- [193] S. Kang, J. P. Green, *Proc. Natl. Acad. Sci. USA* **1970**, *67*, 62–67.
- [194] R. A. Glennon, R. Young, J. M. Jacyno, *Biochem. Pharmacol.* **1983**, *32*, 1267–1273.
- [195] D. E. Nichols, W. R. Pfister, G. K. Yim, R. J. Cosgrove, *Brain Res. Bull.* **1977**, *2*, 169–171.
- [196] C. F. Barfknecht, D. E. Nichols, D. B. Rusterholz, J. P. Long, J. A. Engelbrecht, J. M. Beaton, R. J. Bradley, D. C. Dyer, *J. Med. Chem.* **1973**, *16*, 804–808.
- [197] D. E. Nichols, C. F. Barfknecht, J. P. Long, R. T. Standridge, H. G. Howell, R. A. Partyka, D. C. Dyer, *J. Med. Chem.* **1974**, *17*, 161–166.
- [198] A. P. Monte, D. Marona-Lewicka, M. M. Lewis, R. B. Mailman, D. B. Wainscott, D. L. Nelson, D. E. Nichols, *J. Med. Chem.* **1998**, *41*, 2134–2145.
- [199] J. J. Chambers, J. C. Parrish, N. H. Jensen, D. M. Kurrasch-Orbaugh, D. Marona-Lewicka, D. E. Nichols, *J. Med. Chem.* **2003**, *46*, 3526–3535.
- [200] T. H. McLean, J. J. Chambers, J. C. Parrish, M. R. Braden, D. Marona-Lewicka, D. Kurrasch-Orbaugh, D. E. Nichols, *J. Med. Chem.* **2006**, *49*, 4269–4274.
- [201] T. H. McLean, J. C. Parrish, M. R. Braden, D. Marona-Lewicka, A. Gallardo-Godoy, D. E. Nichols, *J. Med. Chem.* **2006**, *49*, 5794–5803.
- [202] R. A. Rabin, M. Regina, M. Doat, J. C. Winter, *Pharmacol. Biochem. Behav.* **2002**, *72*, 29–37.

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