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In vitro pharmacology of aripiprazole, its metabolite and experimental dopamine partial agonists at human dopamine D_2 and D_3 receptors

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ABSTRACT

Aripiprazole is the first dopamine D_2/D_3 receptor partial agonist successfully developed and ultimately approved for treatment of a broad spectrum of psychiatric and neurological disorders. Aripiprazole's dopamine D_2 and serotonin 5-HT_{1A} receptor partial agonist activities have been postulated to confer clinical efficacy without marked sedation, and a relatively favorable overall side-effect profile. Using aripiprazole's unique profile as a benchmark for new dopamine partial agonist development may facilitate discovery of new antipsychotics. We conducted an in vitro comparative analysis between aripiprazole, and its human metabolite OPC-14857 (7-(4-[4-(2,3-dichlorophenyl)-1-piperazinyl)butoxy)-2(1H)-quinolinone)); RGH-188 (trans-1-[4-[2-[4-(2,3-dichlorophenyl)piperazine-1-yl]ethyl]cyclohexyl]-3,3-dimethylurea), and its metabolite didesmethyl-RGH-188 (DDM-RGH-188); as well as bifeprunox, sarizotan, N-desmethylclozapine (NDMC; clozapine metabolite), and SDZ 208-912 (*N*-[(8α)-2-chloro-6-methylergolin-8-yl]-2,2-dimethylpropanamide). In vitro pharmacological assessment included inhibition of forskolin-stimulated cAMP accumulation and the reversal of dopamine-induced inhibition in clonal Chinese hamster ovary cell lines expressing D_{2S}, D_{2L}, D_3 Ser-9 and D_3 Gly-9 for human dopamine receptors. All test compounds behaved as dopamine D_2/D_3 receptor partial agonists. Aripiprazole's intrinsic activity at dopamine D_{2S} and D_{2L} receptors was similar to that of OPC-14857 and RGH-188; lower than that of dopamine and bifeprunox; and higher than that of DDM-RGH-188, SDZ 208-912, sarizotan, and NDMC. Aripiprazole's intrinsic activity at dopamine D_3 Ser-9 and D_3 Gly-9 receptors was similar to that of OPC-14857 and sarizotan; lower than that of dopamine, bifeprunox, RGH-188 and DDM-RGH-188; and higher than that of SDZ 208-912 and NDMC. A consolidated assessment of these findings may help defining the most appropriate magnitude of intrinsic activity at dopamine D_2/D_3 receptors for clinical efficacy and safety.

1. Introduction

Aripiprazole is a dopamine D_2/D_3 and serotonin 5-HT_{1A} receptor partial agonist which is approved in the US for treatment of irritability associated with autistic disorder in pediatric patients, schizophrenia and bipolar disorder (adult and pediatric patients); and, as an adjunctive treatment for adults with major depressive disorder (Findling et al., 2008, 2009; Keck et al., 2003; Marcus et al., 2008, 2009; Potkin et al., 2003).

To date, dopamine D_2/D_3 receptor partial agonists RGH-188 (trans-1-[4-[2-[4-(2,3-dichlorophenyl)piperazine-1-yl]ethyl]cyclohexyl]-3,3-dimethylurea); bifeprunox; sarizotan; clozapine metabolite, N-desmethylclozapine (NDMC); and SDZ 208-912 (*N*-[(8α)-2-chloro-6-methylergolin-8-yl]-2,2-dimethylpropanamide) have been assessed for the treatment of patients with schizophrenia (Bardin et al., 2006; Benkert et al., 1995; Burstein et al., 2005; Casey et al., 2008; Kiss et al., 2010). However, most have failed in development, potentially due to a lack of precedent with which to guide target parameters for optimal intrinsic activity. In clinical studies, bifeprunox, which has higher intrinsic activity at dopamine D_2/D_3 receptors than aripiprazole (Tadori et al., 2007, 2008), demonstrated a favorable tolerability and safety profile with relatively low potential for Parkinsonism and prolactin elevation. However, the clinical efficacy profile for bifeprunox was not sufficient to warrant regulatory approval (Casey et al., 2008; Newman-Tancredi, 2010). On the other hand, SDZ 208-912, which has lower intrinsic activity than aripiprazole (Tadori et al., 2007, 2008), demonstrated an efficacy and tolerability profile similar to haloperidol (Benkert et al., 1995). However, it was shown to decrease prolactin secretion (Duval et al., 1993). To date, functionally selective dopamine D_3 receptor antagonists are not available for clinical use. Hence, the role of the dopamine D_3 receptor in human psychopathology is not well characterized.

Using clonal Chinese hamster ovary (CHO) cell lines expressing low and high densities of human dopamine D_{2L} and D_{2S} receptors (hD_{2L}-Low, hD_{2L}-High, hD_{2S}-Low and hD_{2S}-High, respectively), we demonstrated that maximal agonist effects of partial agonists depended on variation in receptor expression level, and the use of

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Table 1
Affinities of compounds for human dopamine D_{2L}, D_{2S}, D_3 Ser-9 and D_3 Gly-9 receptors.

	hD_{2L}	hD_{2S}	hD_3 Ser-9	hD_3 Gly-9
	K_i (nM)	K_i (nM)	K_i (nM)	K_i (nM)
SDZ 208-912	0.360 ± 0.057	0.349 ± 0.047	0.186 ± 0.016	0.190 ± 0.015
DDM-RGH-188	0.529 ± 0.032	0.404 ± 0.029	0.112 ± 0.008	0.123 ± 0.007
RGH-188	0.706 ± 0.038	0.671 ± 0.074	0.260 ± 0.016	0.275 ± 0.010
Bifeprunox	0.710 ± 0.108	0.705 ± 0.174	0.364 ± 0.024	0.336 ± 0.024
Sarizotan	2.01 ± 0.21	1.46 ± 0.21	2.00 ± 0.23	1.92 ± 0.16
Aripiprazole	2.24 ± 0.56	1.98 ± 0.38	7.36 ± 0.61	5.88 ± 0.57
OPC-14857	2.25 ± 0.45	1.72 ± 0.35	5.15 ± 0.32	3.77 ± 0.12
NDMC	122 ± 6	113 ± 20	256 ± 34	290 ± 3
Dopamine	1196 ± 81	1628 ± 18	36.7 ± 2.0	34.4 ± 1.7

Values of K_i were derived from the experiments with [^{3}H]raclopride on membranes from hD_{2L}-High and hD_{2S}-High, and [^{3}H]7-OH-DPAT on membranes from hD_3-Ser-9 and hD_3-Gly-9. All values are means ± S.E.M. of three experiments carried out in duplicate.

hD_{2S}-Low, hD_{2L}-High and hD_{2S}-High was suitable to investigate their agonist effects at dopamine D_2 receptors (Tadori et al., 2007).

Dopamine D_3 receptors contain a serine to glycine substitution in the N-terminal domain of the receptor (Ser9Gly), a polymorphism which has been inconclusively correlated with diminished tolerability and response to antipsychotics in patients with schizophrenia (Arranz and Leon, 2007; Fathalli et al., 2008). Using clonal CHO cell lines expressing low and high densities of human dopamine D_3 Ser-9 and Gly-9 receptors, we demonstrated that the agonist potency of dopamine was little dependent on receptor expression levels, and the maximal response of dopamine and partial agonists was dependent on receptor expression levels, and the use of cells expressing high densities of these receptors was suitable to investigate agonist effects of partial agonists (Tadori et al., 2008).

In order to better understand the pharmacological properties of aripiprazole metabolite (OPC-14857), RGH-188, RGH-188 metabolite (DDM-RGH-188), sarizotan, and clozapine metabolite (NDMC), we have profiled each compound and herein compared their pharmacology with that of aripiprazole, bifeprunox and SDZ 208-912 at dopamine D_2 and D_3 receptors.

2. Materials and methods

2.1. Materials

Aripiprazole, OPC-14857, RGH-188, DDM-RGH-188, bifeprunox, SDZ 208-912, sarizotan, and NDMC were synthesized by Otsuka Pharmaceutical Co., Ltd (Tokushima, Japan). Dopamine, haloperidol, butaclamol,

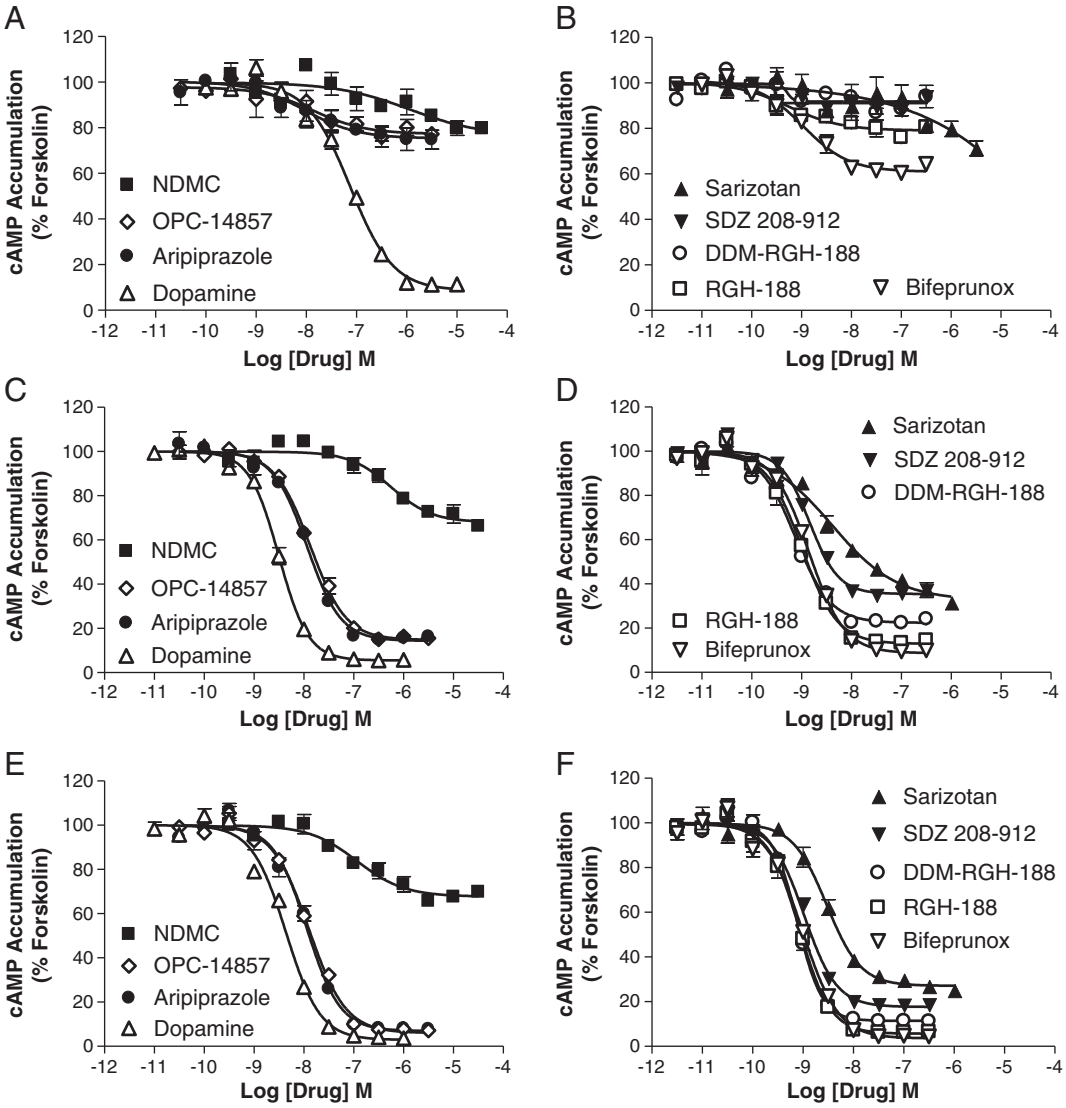

Fig. 1. Concentration–response curves of dopamine, aripiprazole, OPC-4392, NDMC, bifeprunox, RGH-188, DDM-RGH-188, SDZ 208-912 and sarizotan for inhibiting forskolin-stimulated cAMP accumulation in cells: (A, B) hD_{2S}-Low, (C, D) hD_{2L}-High and (E, F) hD_{2S}-High. Cyclic AMP accumulation was normalized to the percentage of forskolin-stimulated cAMP accumulation (set at 100%). Data are means ± S.E.M. of at least three experiments performed in duplicate.