ELSEVIER

European Journal of Pharmacology 351 (1998) 273–286

ejp

Dopamine D_2 and D_4 receptor ligands: relation to antipsychotic action

Julie M. Wilson [a,c], Suparna Sanyal [a,c], Hubert H.M. Van Tol [a,b,c,d,*]

[a] *Laboratory for Molecular Neurobiology,3 Clarke Institute of Psychiatry, 250 College Street, Toronto, Ontario, Canada M5T 1R8*
[b] *Department of Psychiatry, University of Toronto, Toronto, Canada*
[c] *Department of Pharmacology, University of Toronto, Toronto, Canada*
[d] *Institute of Medical Sciences, University of Toronto, Toronto, Canada*

Received 16 April 1998; accepted 17 April 1998

Abstract

Since the discovery that the antipsychotic action of phenothiazines was mediated by dopamine D_2 receptors, the dopamine system has been scrutinized for schizophrenia related abnormalities. The focus has been to create neuroleptics with improved antipsychotic profiles and reduced side effects. With the identification of multiple dopamine receptor subtypes, the hypotheses regarding the role of dopamine in schizophrenia and antipsychotic action of neuroleptics have been refined. Even after the molecular identification of newer dopamine D_2-like receptor subtypes (D_3 and D_4), the dopamine D_2 receptor is still considered the predominant site for antipsychotic action. However, there has been much debate concerning the modulatory role of other dopamine receptor sites in the mechanism of action of antipsychotic drugs. Specifically, the dopamine D_4 receptor has received much attention in this regard, since the atypical antipsychotic agent, clozapine, preferentially blocks this receptor subtype as compared with dopamine D_2 and D_3 receptors. In this review we will highlight some of the observations and arguments regarding the involvement of the dopamine D_2 and D_4 receptor sites in the therapeutic efficacy of antipsychotic medication.

Keywords: Antipsychotic; Clozapine; Schizophrenia; Dopamine D_4 receptor antagonist; Neuroleptic loose; Dopamine receptor; Extrapyramidal symptom; Dopamine D_2-like receptor

1. Introduction

For decades, clinicians and scientists have sought the ideal agent for the pharmacological treatment of schizophrenia. Efforts have been hampered by the complexity of this neuropsychiatric disorder, which manifests itself in a vast array of symptoms. However, no specific characteristics are focal to a diagnosis of schizophrenia and no single symptom is consistently present in all patients. Consequently, the diagnosis of schizophrenia as a single disorder, or as a variety of different disorders has been discussed, but is not yet resolved (Cardno and Farmer, 1995; Tsuang and Farrone, 1995). In general, schizophrenia is associated with alterations in cognitive and emotional functioning. The diverse symptoms of the disorder have been grouped according to positive and negative symptoms. The positive symptoms of schizophrenia include delusions, hallucinations, psychosis and paranoia; the negative symptoms include loss of motivation, blunting of emotions, loss of energy and slowed speech. Cognitive deficits include reductions in working and semantic memory, attention and verbal fluency. For the purposes of this review we will concentrate predominantly on pharmacological agents which mediate antipsychotic activity.

Although the underlying causes of schizophrenia remain unknown, much attention has been focused on the brain dopamine system. This is in large part due to the fact that the first antipsychotic agents, the phenothiazines, which were effective in reducing the positive symptoms of schizophrenia, were shown to have three-dimensional configurations that could be superimposed on the three-dimensional structure of dopamine (Feinberg and Snyder, 1975). Furthermore, since dopaminergic agents could induce a schizophrenia-like psychosis (Randrup and Munkvad, 1965; Snyder, 1973; Lieberman et al., 1987), and these effects could be inhibited by dopamine D_2 receptor antagonists (Seeman and Lee, 1975), it was proposed that the positive symptoms of schizophrenia were related to enhanced dopaminergic neurotransmission. Additionally, the clinical potency of different classes of antipsychotic drugs,

* Corresponding author. Tel.: +1-416-979-4661; fax: +1-416-979-4663; e- mail: hubert.van.tol@utoronto.ca

0014-2999/98/$19.00

despite different chemical structures, correlated well with their affinities for the dopamine D_2 receptor (Seeman and Lee, 1975; Creese et al., 1976; Seeman et al., 1976; Seeman, 1992), suggesting that this receptor was a common target of clinical action. Together with the observation of increased dopamine D_2-like sites in brains of schizophrenic patients (see below) these observations have resulted in the formulation of the dopamine hypothesis of schizophrenia (Seeman, 1987). Blockade of limbic dopamine D_2 receptors has been proposed as a critical component in mediating the positive antipsychotic effects, whereas excessive blockade of dopamine D_2 receptors in striatum is believed to be associated with the frequently reported extrapyramidal side effects (for review see Deutch et al., 1991). Blockade of dopamine D_2 receptors in pituitary is associated with hyperprolactinaemia (Meltzer et al., 1989). More recently, with the identification of additional dopamine D_2-like receptor subtypes (D_3 and D_4), a possible role for blockade of these receptors in mediating an antipsychotic effect has warranted further investigation.

The main dopaminergic pathways in the brain are the nigrostriatal, mesolimbic, mesocortical and tuberoinfundibular pathways. The nigrostriatal dopamine neurones project from the substantia nigra (A9) to the striatum and are involved primarily in control of motor activity. Mesolimbic dopamine neurones project from the ventral tegmental area (A10) and retrorubral field (A8) to the nucleus accumbens, olfactory tubercle, septum and amygdala. Mesocortical dopaminergic cells project from A10 and A8 groups to the prefrontal cortex, entorhinal, piriform and anterior cingulate cortices. The mesocorticolimbic dopamine system plays an important role in the regulation of emotional and cognitive function, and is thought to provide modulation of efferents from other brain areas that might be disturbed in schizophrenia (Mogenson et al., 1988; Csernansky et al., 1991; Amalric and Koob, 1993). It has been suggested that the negative symptoms of schizophrenia might arise from a dopaminergic deficit in the prefrontal cortex (hypofrontality), whereas the positive symptoms might be related to hyperdopaminergic activity in the mesolimbic dopamine neurones (Davis et al., 1991).

2. Dopamine receptor subtypes

Currently there are 5 known dopamine receptor subtypes that have been categorised according to structural, functional and pharmacological characteristics. These receptor classes include 2 main types of receptors, dopamine D_1-like and D_2-like. The dopamine D_1-like receptors (D_1 and D_5) activate adenylyl cyclase, whereas the dopamine D_2-like receptors can inhibit adenylyl cyclase and can also activate various other pathways. The first dopamine receptor to be cloned was the dopamine D_2 receptor which was cloned from rat brain based on its homology with the previously cloned β-adrenoceptor (Bunzow et al., 1988). Shortly after the initial cloning of the dopamine D_2 receptor, it was determined that alternative splicing of the receptor gene resulted in the expression of 2 protein isoforms, dopamine D_2(short) and D_2(long) which differ in length by 29 amino acids in the third cytoplasmic loop (Dal Toso et al., 1989; Giros et al., 1989; Grandy et al., 1989; Monsma et al., 1989). Additional dopamine D_2-like receptors, the dopamine D_3 and D_4 receptors, were subsequently cloned based on their homology to the dopamine D_2 receptor (Sokoloff et al., 1990; Van Tol et al., 1991). Similarly the dopamine D_5 receptor was cloned (Grandy et al., 1991; Sunahara et al., 1991; Tiberi et al., 1991) based on its homology with the dopamine D_1 receptor (Dearry et al., 1990; Monsma et al., 1990; Sunahara et al., 1990; Zhou et al., 1990).

3. Antipsychotic agents

3.1. Typical antipsychotic agents

Traditional antipsychotic agents belonged to the phenothiazine class. Examples include chlorpromazine, thioridazine and fluphenazine, which consist of a phenothiazine nucleus with minor molecular substitutions. Other classes of antipsychotic agents include butyrophenone and benzamide derivatives (for example, haloperidol and sulpiride) which have pharmacological characteristics similar to those of the phenothiazines.

The phenothiazine class of antipsychotic agents, introduced during the 1950s, markedly improved the positive symptoms of schizophrenia and reduced the average duration of hospitalisation. Continued treatment significantly reduced the probability of reappearance of psychotic symptoms and rehospitalisation (Kane and Lieberman, 1987). However, these antipsychotic agents had no effect on cognitive functioning and were not therapeutically effective in alleviating the negative symptoms of schizophrenia. Indeed, some classic antipsychotics might actually exacerbate the negative symptoms of the disorder, although it is not yet clear whether this is a drug effect or part of the disease process (Raleigh, 1996). In addition, a significant proportion of schizophrenic patients with positive symptoms is refractory (5–25%) or become intolerant (5–10%) to treatment with classical neuroleptics (Brenner et al., 1990). A major drawback with the classical antipsychotic drugs is the emergence of unwanted side-effects involving extrapyramidal dysfunction. The most common side effects include Parkinsonian-like symptoms (tremor, rigidity and bradykinesia), akathesia, dystonia and tardive dyskinesia [for review see Ref. (Raleigh, 1996)].

3.2. Atypical antipsychotic agents

The search for more effective compounds with lower incidence of extrapyramidal side effects resulted in the

discovery of clozapine, an antipsychotic agent with several clinical advantages over the classical antipsychotics. Thus, clinically, clozapine was highly effective not only in alleviating positive symptoms of schizophrenia, but also had some beneficial effects on negative symptoms and improving cognitive deficits. One of the major benefits of clozapine treatment was its ability to mediate these effects with a low propensity to induce extrapyramidal side effects and no tardive dyskinesia. A further benefit of clozapine was its ability to prevent psychosis in some patients who were either refractory or intolerant to the effects of classical neuroleptics (Kane et al., 1988a,b). Additionally, clozapine, unlike classical neuroleptics, only produced a mild or transient increase in prolactin secretion (Meltzer et al., 1989).

However, despite these beneficial effects, the clinical use of clozapine has been limited since it is known to cause agranulocytosis in a small percentage (1–2%) of patients (Kane and Freeman, 1994). The search has therefore continued for additional antipsychotic agents that have the beneficial properties of clozapine, but which lack its unwanted side effects. However, none of the presently available agents appear to display the full clinical spectrum of clozapine.

4. Dopamine D_2-like receptors as targets for antipsychotic drugs

4.1. Pharmacological profile of antipsychotics at dopamine D_2-like receptors

As described previously, the clinical potency of antipsychotic drugs correlates well with their affinities for the dopamine D_2 receptor (Seeman and Lee, 1975; Creese et al., 1976; Seeman et al., 1976; Seeman, 1992), suggesting that this receptor was a common target of clinical action. However, with the identification of additional dopamine D_2-like receptor subtypes (D_3 and D_4), the question has arisen as to the relative contribution of these receptors to antipsychotic action and to the possible modulatory role in the action of other antipsychotic agents. Only limited information is available concerning selective D_3 receptor antagonists. Nafadotride, an antagonist with higher affinity at the dopamine D_3 receptor (0.3 nM) than the dopamine D_2 receptor ($\sim$ 3 nM), increased spontaneous locomotion in rats (Sautel et al., 1995). At higher doses, nafadotride, like haloperidol, produced catalepsy (Sautel et al., 1995), likely through blockade of dopamine D_2 receptors (Levant and Vansell, 1997). Additionally, other dopamine D_3 receptor antagonists were found to be ineffective in the conditioned avoidance response in rats (Millan et al., 1997), or reduced amphetamine-induced locomotor activity (Corbin et al., 1998). The experimental animal studies are as yet insufficient to predict the antipsychotic potential of the dopamine D_3 receptor antagonists. For the purposes of the remainder of the present review, we will focus predominantly on the dopamine D_2 and D_4 receptors.

In order to understand the involvement of various receptor subtypes in mediating the antipsychotic effects of agents, and to determine why atypical antipsychotic drugs elicit low levels of parkinsonism, it is necessary to obtain accurate inhibition constants (K_i) or dissociation constants (K_d) for antipsychotic drugs at the various receptors. However, despite standard experimental conditions, different K values for particular neuroleptics have been reported (see Table 1). These differences appear to arise from the use of different radioligands for the various receptors. For example, labeling of the dopamine D_2 receptor with [^{3}H]nemonapride, [^{3}H]spiperone or [^{3}H]raclopride has yielded variable K_i values for clozapine (187 nM, 137 nM and 59 nM, respectively; (Seeman and Van Tol, 1995)). Similarly for the dopamine D_4 receptor, the K_i values of clozapine for displacement of [^{3}H]nemonapride and [^{3}H]spiperone were 38 nM and 22 nM, respectively, while [^{3}H]clozapine bound to dopamine D_4 receptors with a K_d of 1.6 nM (Seeman and Van Tol,

Table 1
Affinities for typical and atypical neuroleptics at dopamine and serotonin 5-HT_{2A} receptors

	K_i values (nM)					
	Ref.	D_2	D_3^a	D_4^a	D_1	5-HT_{2A}
Typical antipsychotic						
Chlorpromazine	b	1.5	1.5	37		
	c	0.66		1.15		3.5
Haloperidol	b	1		5.1		
	c	0.35		0.84		25
	d	0.82	7.3	2.5		28
Fluphenazine	b	0.5		46		
	c	0.32		50		80
Atypical Antipsychotics						
Clozapine	b	138		9		
	c	44		1.6		11
	d	36	160	22	53	51
	e	190	280	40		
	f	60	83			
Remoxipride	b	300		3690		
	c	30		2800		3100
	d	2000			> 41,000	23,000
	e	125	970			
Seroquel	c	78		3000		2500
Melperone	c	88		410		280
Olanzapine	c	3.7		2		5.8
	d	2.1	2.0	17	10	1.9
	e	31	49	28		
Risperidone	c	0.3		0.25		0.14
	d	0.44	2.8	13	21	0.39
	e	5.9	14	16		
	f	1.7	6.7			

K_i values (nM) at native receptors are presented, unless otherwise indicated (a, cloned D_3 and D_4 receptors). Data are from b) (Van Tol et al., 1991), c) (Seeman et al., 1996) and d), e), f) (Arnt and Skarsfeldt, 1998) (Lundbeck, Janssen and Astra, respectively). The radioligand independent affinity constants are from reference b.

1995). The differences in K_i values display a linear correlation with the membrane/buffer partition coefficients of the radioligands. Extrapolation of the K_i values for neuroleptics to a zero membrane/buffer partition coefficient gives a value that is in close agreement with its K_d value obtained through saturation binding and Scatchard analysis. This method for determining the K_i values eliminates the variability in binding affinity resulting from the use of different radioligands in competition binding analysis (Seeman and Van Tol, 1995; Seeman and Tallerico, 1998).

Most antipsychotic drugs display a high correlation between their clinical potency and their affinity at the dopamine D_2 receptor, suggesting this site as a critical target for mediating therapeutic effects. However, for clozapine, the clinical potency correlated better with its affinity for the dopamine D_4 receptor, which led to the dopamine D_4 receptor being proposed as the primary target for mediating clozapine's therapeutic effects. Further, it has been postulated that the polymorphic variants of the dopamine D_4 receptor may be responsible for the variable therapeutic response to clozapine, exemplified by the existence of a clozapine treatment-refractory schizophrenic population. However, although clozapine displays small differences in affinity for the polymorphic variants of the human dopamine D_4 receptor (Asghari et al., 1994), such differences are unlikely to be related to clozapine response since the therapeutic dose range of clozapine would effectively block all polymorphic variants of the dopamine D_4 receptor. In addition, genetic studies have failed to demonstrate a positive correlation between dopamine D_4 polymorphisms and clozapine response (Shaikh et al., 1993; Rao et al., 1994).

4.2. Regional distribution

The psychotic symptoms of schizophrenia are thought to be mediated through enhanced dopaminergic transmission in the limbic system, whereas the extrapyramidal side effects are thought to be due to decreased striatal dopamine function (Davis et al., 1991). In the brain, expression of dopamine D_2 receptors is highest in striatum, nucleus accumbens, olfactory tubercle and substantia nigra (Meador-Woodruff et al., 1989; Mengod et al., 1989; Najlerahim et al., 1989; Weiner and Brann, 1989; Mansour et al., 1990). The patterns of dopamine D_3 and D_4 receptor expression are more restricted to the mesolimbic system. For the D_3 receptor, the highest concentrations are found in the nucleus accumbens, whereas dopamine D_4 receptor mRNA is more abundant in frontal cortex, amygdala, thalamus, hypothalamus, pituitary with lower levels in hippocampus (Joyce and Meador-Woodruff, 1997). Overall, it appears that the dopamine D_2 receptors are more dominantly expressed in areas associated with motor control, while dopamine D_3 and D_4 receptors are more exclusively located in areas where the dopamine system is thought to serve a role in modulating emotion and cognition. However, in absolute terms, dopamine D_2 receptors are also present at prominent levels in extra-striatal brain areas, allowing for the possibility that the dopamine D_2 receptor might also play a role in cognitive and emotional aspects of antipsychotic treatment.

4.3. Differences in dopamine D_2-like receptor densities in schizophrenic brain

Much of the debate on a putative role of dopamine D_2 and D_4 receptors in antipsychotic action of neuroleptics was sparked by various reports on increased levels of these receptors in post-mortem (Seeman et al., 1987) and in vivo (Wong et al., 1986) studies of patients with schizophrenia. Additional in vivo imaging studies using [^{11}C]*N*-methylspiperone supported the observation of increased dopamine D_2-like sites in the brains of patients with psychosis (Tune et al., 1993; Pearlson et al., 1995; Tune et al., 1996; Wong et al., 1997, but see, Nordstrom et al., 1995), although this could not be confirmed when [^{11}C]raclopride was used in positron emission tomography (PET) studies (Farde et al., 1990; Hietala et al., 1994).

In order to evaluate the underlying cause for such differences in binding density, 3 different radioligands, [^{3}H]spiperone, [^{3}H]nemonapride and [^{3}H]raclopride were used to study dopamine D_2-like receptor densities in post-mortem caudate nucleus and putamen of schizophrenic patients and control subjects (Seeman et al., 1993). While increased dopamine D_2-like receptor densities were observed in schizophrenic brain using [^{3}H]spiperone and [^{3}H]nemonapride, such differences were not detected with [^{3}H]raclopride. These data are in agreement with the PET studies. Since [^{3}H]raclopride binds to dopamine D_2 and D_3 receptors, but not to dopamine D_4 receptors, and [^{3}H]nemonapride binds to all 3 dopamine D_2-like receptors, it was argued that the increase was mediated by a dopamine D_4-like receptor. Similar observations were made by others (Murray et al., 1995; Sumiyoshi et al., 1995; Marzella et al., 1997), although failures to see such a difference have also been reported (Lahti et al., 1996).

Receptor distribution studies with dopamine D_4-specific antisera and ligands did not support the presence of significant levels of dopamine D_4 receptors in caudate nucleus and putamen (Mrzljak et al., 1996; Ariano et al., 1997; Primus et al., 1997). However, it has been reported that the raclopride insensitive dopamine D_2-like binding site can be displaced by the dopamine D_4 receptor specific antagonist L-745,870 (3-[{4-(4-chlorophenyl)piperazin-1-yl)]methyl}-1*H*-pyrrolo[2,3b]pyridine) (Tarazi et al., 1997a,b). In order to resolve the controversial issue regarding the nature of these sites, a novel non-selective dopamine receptor ligand ([^{3}H]SDZ GLC 756; ([^{3}H-1-methyl][−]-(3*R*, 4a*R*, 10a*R*[*trans*])-1,2,3,4,4a,5,10,10a-octahydro-6-hydroxy-1-methyl-3-[(2-pyridyl-thio)-methyl]benzo[*g*]quinoline · HCl) was discovered that enabled the detection of a

dopaminergic receptor site that was markedly up-regulated in schizophrenic brain (Seeman et al., 1997b). This radioligand can detect all dopaminergic receptors with the exception of D_3 sites. In saturation binding analysis of schizophrenic brain tissue, this compound revealed an elevated number of dopaminergic sites that were not blocked by an excess of unlabeled raclopride (D_2, D_3) and SCH23390 ((*R*)-(+)-8-chloro-2,3,4,5-tetrahydro-3-methyl-5-phenyl-1*H*-3-benzezepine-7-ol) (D_1, D_5), but was completely eliminated by the addition of guanine nucleotides (Seeman et al., 1997b). Nevertheless, in binding studies on heterologously expressed cloned dopamine D_2 receptors, raclopride could completely displace [^{3}H]SDZ GLC 756 binding. In vitro autoradiographic studies with this compound revealed the presence of these raclopride-insensitive dopamine D_2-like sites in the basal ganglia of rat and mouse brain (see Fig. 1). However, these sites could not be detected in transgenic D_2-deficient mice (Baik et al., 1995) using similar autoradiographic analysis (Seeman et al., 1997b). These data indicate that the elevated number of dopamine D_2 sites detected in schizophrenic brain by SDZ GLC 765 are likely derived from the dopamine D_2 receptor gene. This is in agreement with the observation that [^{3}H]SDZ GLC 756 binding to these dopamine D_2-like sites is guanine nucleotide sensitive, since SDZ GLC 765 is an agonist at the dopamine D_2 receptor (Markstein et al., 1996). Furthermore, dopamine displayed a significantly higher affinity for this site than norepinephrine or serotonin (Fig. 2) supporting the notion that the observed sites are very likely dopaminergic. However, in contrast to the cloned dopamine D_2 receptor, these elevated sites in the brain of schizophrenic patients and rat striatum appear to display a poor affinity for the dopamine D_2 receptor antagonists, raclopride and S-sulpiride (Figs. 1 and 2). Taking all these observations into account we

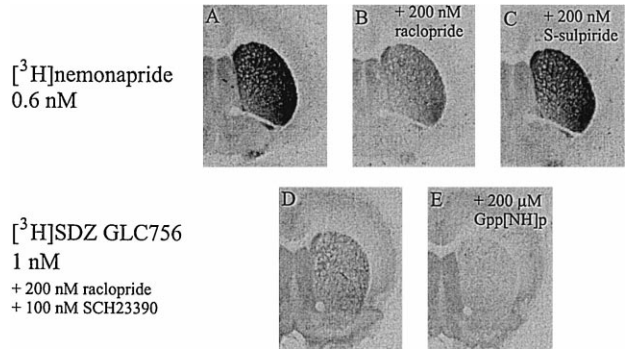

Fig. 1. In vitro autoradiography of raclopride-insensitive D_2-like binding sites in striatum of rat brain (courtesy of Dr. J. N. Nobrega, Univ. of Toronto). The upper panel shows the binding of 0.6 nM [^{3}H]nemonapride (A), in the presence of 200 nM raclopride (B), or 200 nM S-sulpiride (C). The lower panel depicts [^{3}H]SDZ GLC 756 binding sites in the presence of excess raclopride (200 nM) and SCH23390 (100 nM) (D); binding to this site in the striatum can be displaced by the addition of 200 μM Gpp[NH]p (E).

speculate about the existence of a novel dopamine D_2-like receptor site in striatum that shares pharmacological characteristics of the dopamine D_4 receptor, in that it has low affinities for raclopride and S-sulpiride. However, this site is unlikely to represent the dopamine D_4 receptor, since SDZ GLC 756 binding to the dopamine D_4 receptor is not sensitive to guanine nucleotides and behaves like a full antagonist in functional studies at the cloned dopamine D_4 receptor (unpublished data, SS and HVT). Furthermore, it was determined that bromocriptine displays a 10-fold higher affinity than clozapine for this site, whereas at the cloned dopamine D_4 receptor these 2 ligands display the reverse rank order of potency. It can be argued that the raclopride-insensitive binding site detected with [^{3}H]SDZ GLC 756 is likely the same as that detected with [^{3}H]nemonapride, since both sites display similar subregional distribution and low affinity for S-sulpiride. Since [^{3}H]nemonapride binding at the raclopride-insensitive site can be displaced by the σ-receptor ligand PPAP (*R*(−)-*N*-(3-phenyl-*n*-propyl)-1-phenyl-2-aminopropane hydrochloride), it has been suggested that it might represent a σ-type binding site (Helmeste et al., 1997). However, this seems unlikely, since the affinity of nemonapride for σ receptor sites as determined using [^{3}H](+)3-PPP (3-(3-hydroxyphenyl)-*N*-*n*-propylpiridene) and [^{3}H](+) pentazocine (80 nM and 3000 nM, respectively) (Tam and Cook, 1984; DeHaven-Hudkins et al., 1992) is far lower than the affinity of [^{3}H]nemonapride for the raclopride-insensitive binding site (K_d less than 1 nM). Furthermore, in the autoradiographic studies [^{3}H]nemonapride is used at concentrations of $\sim$600 pM (Fig. 1), a concentration that is unlikely to detect the aforementioned σ receptor sites. The absence of raclopride-insensitive binding sites in the dopamine D_2-deficient mice as well as the observation that the regional distribution profile of this compound was very similar to that of the dopamine D_2 receptor, suggests that this novel site might be derived from the dopamine D_2 receptor gene. This notion is further supported by the observation that the raclopride-insensitive dopamine D_2 site can be detected in heterologous expression experiments with the cloned dopamine D_2 receptor (personal communication, Dr. P. Seeman).

Studies involving the measurement of mRNA levels for both the dopamine D_2 and D_4 receptors in caudate nucleus and putamen of schizophrenic patients failed to show the presence of significant levels of dopamine D_4 mRNA or increased levels of dopamine D_2 mRNA (Meador-Woodruff et al., 1997; Stefanis et al., 1998). While unaltered dopamine D_2 mRNA levels appear inconsistent with the aforementioned increase in dopamine D_2 receptor binding sites, it should be considered that receptor densities can be strongly modulated by various post-transcriptional processes. Meanwhile, reduced levels of dopamine D_4 mRNA, as well as D_3 mRNA, were detected in the orbito-frontal cortex of schizophrenic patients (Meador-Woodruff et al., 1997), although increased densities of

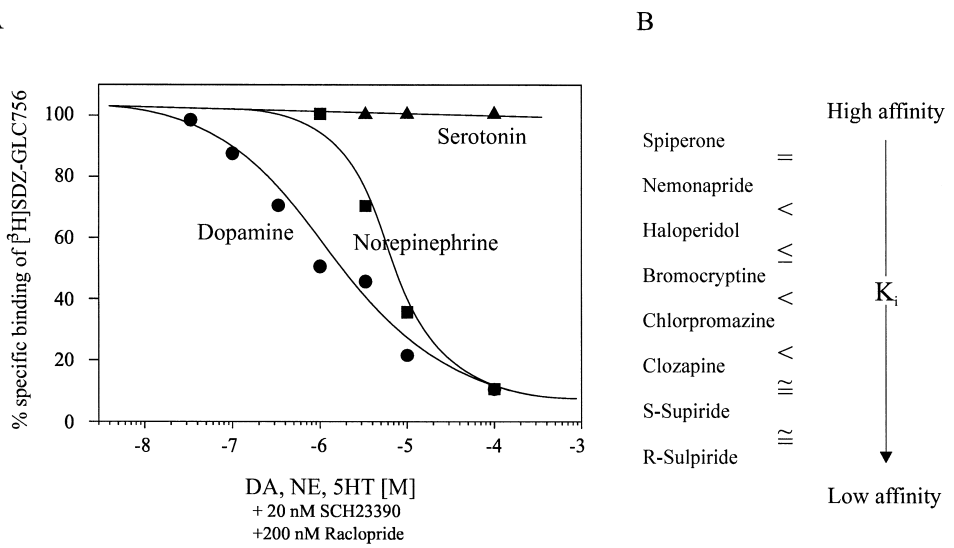

Fig. 2. Pharmacological profile of the raclopride-insensitive dopamine D_2-like binding sites in striatum. The figure illustrates competition binding analysis of [^{3}H]SDZ GLC 756 (600 pM) with increasing concentrations of cold ligands (dopamine, norepinephrine and serotonin) in the presence of 20 nM SCH23390 and 200 nM raclopride (A). The relative rank order of potency for known D_2 receptor ligands is listed next to the figure (B).

dopamine D_4 mRNA in the frontal cortex of schizophrenic patients have also been reported (Stefanis et al., 1998). Since the reported changes in dopamine D_4 mRNA levels in frontal cortex are conflicting, and no information is currently available concerning the levels of dopamine D_4 receptor protein in this brain area of schizophrenic patients, further interpretation of these results is difficult.

Other noteworthy observations with respect to the nature of dopamine D_2-like sites in schizophrenia is that the density of dopamine D_2-like sites in normal brain measured by [^{3}H]raclopride are higher than that measured by [^{3}H]spiperone, but in schizophrenic brain the density of [^{3}H]spiperone binding sites is higher than [^{3}H]raclopride binding sites (Seeman et al., 1993). Similar differences in densities measured by these 2 ligands occur in PET studies (Farde et al., 1990; Tune et al., 1993; Farde et al., 1995; Nordstrom et al., 1995; Pearlson et al., 1995). Additionally, there have been observations suggesting that spiperone and raclopride may label distinct sites (Bischoff and Gunst, 1997). Nevertheless, the discrepancies observed using differential binding protocols raises the question as to what is really being measured and highlights the difficulty in interpreting the data correctly, but does not undermine the interpretation that dopamine D_2-like sites are elevated in brain of patients with schizophrenia. It is unlikely however, that the differential binding is caused by up-regulation in D_3 receptors in drug-naive schizophrenic patients (Gurevich et al., 1997) since both spiperone and raclopride readily bind to D_3 receptors. Moreover, antipsychotic treatment apparently normalizes D_3 receptor levels in schizophrenic patients (Gurevich et al., 1997), whereas elevated levels of [^{3}H]spiperone binding sites were detected in several neuroleptic-treated schizophrenic patients (Seeman et al., 1993).

Taking all of the above into account it is not unreasonable to assume that all reports on up-regulated dopamine D_2-like sites in caudate nucleus and putamen of patients with schizophrenia involve a raclopride-insensitive dopamine D_2 receptor isoform. Confirmation of the true nature of this site, as well as its possible role in schizophrenia and/or antipsychotic treatment remain to be established. Although the levels of this site can be up-regulated by neuroleptic treatment (Schoots et al., 1995), the up-regulation in brains of patients with schizophrenia is, at least in part, not neuroleptic induced (Seeman et al., 1993, 1997b).

4.4. Receptor occupancy

In vivo imaging studies have demonstrated that the beneficial effects of antipsychotic agents are achieved at about 70% occupancy of dopamine D_2 receptors in the basal ganglia, whereas extrapyramidal side effects become apparent at about 80% occupancy (Farde and Nordstrom, 1993). Thus, the therapeutic window is so narrow that it is extremely rare for patients receiving traditional neuroleptics not to exhibit extrapyramidal side effects (Casey, 1995). The high rate of occurrence of unwanted side effects might contribute to the low level of patient compliance and high incidence of re-emergence of psychotic symptoms.

At therapeutic doses of clozapine, more than 75% of dopamine D_4 receptors are occupied (Seeman, 1995), whereas less than 50% of dopamine D_2 receptors are occupied (Farde et al., 1992; Seeman, 1995). Thus, it was hypothesised that the low occupancy at dopamine D_2 receptors was insufficient to produce either the antipsychotic effect or extrapyramidal side effects of clozapine

(Farde et al., 1992; Pilowsky et al., 1992). It was also suggested that the antipsychotic effects of clozapine were mediated through blockade of dopamine D_4 rather than D_2 receptors. However, this notion has recently been revisited based on the dependency of ligand affinity on the radioligand used (Seeman and Tallerico, 1998). In this regard, dopamine D_2 occupancy using [^{11}C]raclopride was determined to be 48% (Farde et al., 1994), whereas with [^{18}F]methylspiperone occupancy varied between 0 and 22% (Karbe et al., 1991). However, as with in vitro experiments, the apparent in vivo occupancy of the antipsychotic agent is dependent upon the tissue/buffer partition co-efficient of the radioligand. When the data is extrapolated to a tissue/buffer partition co-efficient of zero, which mimics the clinical situation in which no radioligand is present, the extrapolated dopamine D_2 receptor occupancy by clozapine is approximately 85% (Seeman and Tallerico, 1998). This raises the question as to why clozapine is free from extrapyramidal side effects. This could be explained by the affinity of clozapine relative to that of dopamine for the dopamine D_2 receptor. Since clozapine and other atypical neuroleptics have relatively low affinity for the dopamine D_2 receptor, it is more likely that endogenous dopamine will displace the drugs from the receptor. Consequently, atypical antipsychotic agents have been termed 'loose' neuroleptics (see Section 4.1). The displacement of low affinity neuroleptics by dopamine will be more pronounced in areas with high levels of endogenous dopamine, like the striatum (thereby alleviating extrapyramidal symptoms), but less in extra-striatal areas preserving antipsychotic efficacy (Seeman et al., 1996, 1997a).

4.5. Dopamine D_2 vs. D_4 receptor blockade in antipsychotic action

Irrespective as to whether schizophrenic patients display detectable changes in any of the dopaminergic receptor sites, most, if not all, antipsychotic agents appear to mediate their antipsychotic effect through blockade of dopamine D_2-like sites. This was originally hypothesized following the observation that a tight correlation existed between the clinical potency of various antipsychotics and their affinity for dopamine D_2 receptors (Creese et al., 1976; Seeman et al., 1976). With the discovery of newer dopamine D_2-like receptor subtypes, similar correlation studies were performed using the affinity of various antipsychotic drugs for the dopamine D_3 and D_4 receptors (Seeman, 1992; Seeman and Van Tol, 1995). The findings in these studies still implicated the dopamine D_2 receptor as the predominant target for antipsychotic action. An apparent exception in those analyses was clozapine, since the reported levels of this drug in free plasma water, at clinically effective doses, were about 10-fold lower than those predicted to act at the dopamine D_2 receptor. Taking into account that clozapine displays a unique clinical profile (Tamminga and Gerlach, 1987), with relatively high affinities for several receptors, it has been postulated that other receptors, most notably 5-HT_{2A} and dopamine D_4, may be dominant targets for mediating the antipsychotic actions of this drug (for review and references therein see Seeman et al., 1997c). However, more recent data on levels of clozapine in free plasma water and spinal fluid of patients with clinically effective doses of this drug (Nordin et al., 1995), suggest that dopamine D_2 receptor occupancy is actually higher (70–80%) and would be sufficient to explain its antipsychotic efficacy (Seeman and Tallerico, 1998).

4.6. Novel antipsychotic agents

To obtain more conclusive insight into the contribution of individual receptor subtypes in antipsychotic treatment, the development of specific antagonists for the different receptors is desirable. With the cloning of a large variety of receptor subtypes and the ability to pharmacologically analyse these receptors, new highly specific drugs have been developed.

4.6.1. Dopamine D_2 vs. D_4 receptors

Although the therapeutic action of most antipsychotic agents can be explained through blockade of the dopamine D_2 receptor, few of the clinically used neuroleptics are dopamine D_2 receptor-specific. Some of the more potent and selective dopamine D_2 receptor antagonists, like raclopride and sulpiride, also block D_3 receptor sites. Remoxipride is probably the most selective dopamine D_2 receptor antagonist used to date; however, this neuroleptic has a relatively low affinity for the dopamine D_2 receptor. Considering that endogenous dopamine can potentially modulate the blockade of dopamine D_2 receptors by low affinity neuroleptics, it is difficult to assess the extent to which the therapeutic response of these loose antipsychotics correlates with the antagonism of dopamine D_2 receptors. Although the creation of a dopamine D_2 receptor-deficient mouse (Baik et al., 1995) has shown a clear association between blockade of dopamine D_2 receptors and motor side effects, another strain of dopamine D_2 receptor-deficient mice (Kelly et al., 1997) displayed a much less severe motor impairment. These data suggest that differences in motor side effects mediated by neuroleptics may also be modulated by the genetic background of the individual. Moreover, clinical studies examining the motor side effects of neuroleptics might suffer from the same stratification problems often encountered in genetic association studies. Therefore, much of the existing data on the beneficial profiles of certain neuroleptics with regards to motor side effects should be interpreted with caution since experimental groups should be closely matched using genetic criteria.

Considering the selectivity of remoxipride for the dopamine D_2 receptor, and the fact that this drug is an effective antipsychotic agent, it is evident that dopamine D_2 receptor blockade is sufficient to mediate antipsychotic

action. Nevertheless, it seems that the relative affinity of the antipsychotic agent at the dopamine D_2 receptor correlates with the severity of motor side effects (Seeman and Tallerico, 1998). Thus, tightly bound neuroleptics with high dopamine D_2 receptor affinity (K_i less than 6 nM) are associated with higher liability for extrapyramidal side effects, through blockade of striatal dopamine D_2 receptors, whereas the loosely bound agents with lower affinity at the dopamine D_2 receptor (range 30 to 88 nM; Table 1) display lower liability for extrapyramidal dysfunction (for reviews, see Seeman et al., 1996; Arnt and Skarsfeldt, 1998). The differences in affinity at the dopamine D_2 receptor might therefore account for the different extrapyramidal dysfunction profiles of typical and atypical neuroleptics. However, at high concentrations most atypical antipsychotic agents will also elicit extrapyramidal side effects. An exception to this is observed with clozapine, which even at very high concentrations does not elicit extrapyramidal side effects in humans or catalepsy in the experimental animal (for review see Arnt and Skarsfeldt, 1998).

An additional dopamine D_2 receptor-mediated side effect frequently observed in patients receiving typical neuroleptic treatment is hyperprolactinaemia. Under normal conditions, prolactin release in the pituitary is negatively modulated by the tuberoinfundibular dopamine system. Consequently, blockade of dopamine D_2 receptors in the pituitary by typical antipsychotic agents results in increased prolactin release. Similar findings of hyperprolactinaemia have been reported in dopamine D_2 receptor-deficient mice (Kelly et al., 1997). These data demonstrate the involvement of the dopamine D_2 receptor in the inhibition of prolactin release. In contrast to typical antipsychotic agents, the atypical antipsychotic clozapine is not associated with chronic hyperprolactinaemia. Consequently, a site different from the dopamine D_2 receptor was proposed to mediate clozapine's antipsychotic activity (Meltzer et al., 1989). However, both remoxipride and clozapine can cause short-lived increases in prolactin secretion (Gudelsky et al., 1987; Von Bahr et al., 1991) suggesting that low dopamine D_2 receptor affinity may be sufficient to account for clozapine's low propensity to produce hyperprolactinaemia, although modulatory effects might also be mediated through other receptor sites (Meltzer and Gudelsky, 1992).

Clozapine has been used as a prototype for the development of effective new antipsychotic agents with fewer side effects. The unique aspects of clozapine's pharmacological profile compared with typical neuroleptics include its high affinity at the dopamine D_4 receptor. This receptor was believed to be a likely candidate in mediating clozapine's antipsychotic effects and low extrapyramidal liability. To test this hypothesis, novel highly selective dopamine D_4 receptor antagonists have been synthesized. These selective dopamine D_4 receptor antagonists include CP-293,019 (Pfizer (Zorn et al., 1996)), PD 167021 (Parke-Davis, (Corbin and Heffner, 1997)), NRA-0045 (Okuyama et al., 1997a,b), L-745,870 (Merck (Patel et al., 1997)), NGD 94-1 (Neurogen (Tallman et al., 1997)), U101,387G (Upjohn (Merchant et al., 1996)) and RO 61-6270 (Hoffmann La Roche (Hartman et al., 1996)). The reported K_i values for these compounds at the dopamine D_2, D_4 and 5-HT_{2A} receptors, where available, are summarised in Table 2.

Several of these agents have been tested for efficacy in experimental animal models which have been proposed as predictive screening tests for antipsychotic activity in humans. These tests include apomorphine- or amphetamine-induced hyperactivity and reversal of apomorphine-induced disruption of pre-pulse inhibition. Further behavioral tests in the experimental animal are proposed to be indicative as to whether a drug might induce extrapyramidal dysfunction in humans. These include the ability of a drug to reverse stereotypy, or to induce catalepsy. Indeed, typical antipsychotic agents such as haloperidol have been shown to inhibit both amphetamine-induced hyperactivity and stereotypy, whereas atypical drugs, such as clozapine, inhibit only amphetamine-induced hyperactivity. Thus, atypical neuroleptics with low clinical liability for extrapyramidal dysfunction have little effect on amphetamine-induced stereotyped behavior in experimental animals (for review see Arnt and Skarsfeldt, 1998).

Of the novel dopamine D_4 receptor selective compounds currently available, CP-293,019, PD 167021 and NRA-0045 reversed the hyperlocomotion mediated by the

Table 2
Affinities of novel D_4 receptor selective antagonists at dopamine D_2, D_4 and serotonin 5-HT2A receptors

Compound	Company	K_i values (nM)			Reference
		D_2	D_4	5-HT_{2A}	
CP-293,019	Pfizer	>3 μM	3.3 nM		Zorn et al., 1996
RO 61-6270	Hoffmann La Roche	>5 μM	5 nM	>5 μM	Hartman et al., 1996
PD 167021	Parke-Davis		$\surd$		Corbin and Heffner, 1997
NRA-0045		$\sim$200 nM	2.5 nM	1.9 nM	Okuyama et al., 1997
L-745,870	Merck	960 nM	0.43 nM	>200 nM	Patel et al., 1997
NGD 94-1	Neurogen	>2 μM	3.6 nM		Tallman et al., 1997
U101,387G	Upjohn	>5 μM	10.1 nM	>1.6 μM	Merchant et al., 1996

K_i values (nM) are included when available in the references cited. Otherwise, $\surd$ indicates a reported selectivity for the D_4 receptor, although K_i values are not available.

dopaminergic agents, amphetamine or apomorphine (Zorn et al., 1996; Corbin and Heffner, 1997; Majchrzak et al., 1997; Okuyama et al., 1997a,b; Mansbach et al., 1998). Additionally, U101,387G reversed amphetamine- induced behavioral sensitization (Feldpausch et al., 1996; Garimella et al., 1997), but had no effect on either amphetamine- or apomorphine-induced hyperactivity (Merchant et al., 1996; Stone et al., 1996). Similarly, L-745,870, which displays approximately 2000-fold selectivity for the dopamine D_4 (0.43 nM) over D_2 (960 nM) receptor, and low affinity for the 5-HT_{2A} (> 200 nM; (Patel et al., 1997)), was shown to be ineffective in reversing apomorphine-induced hyperactivity (Bristow et al., 1997). Thus, the experimental animal literature is somewhat contradictory concerning the effects of dopamine D_4 receptor blockade in behavioral models of hyperlocomotion.

Other behavioral tests that are predictive of antipsychotic activity in humans have provided more consistent results. CP-293,019, NRA-0045, U101,387G and PD 167,021 were shown to significantly reverse apomorphine-induced disruption of pre-pulse inhibition (Zorn et al., 1996; Corbin and Heffner, 1997; Okuyama et al., 1997; Mansbach et al., 1998), suggesting that these agents might be therapeutically effective in relieving some symptoms related to schizophrenia. In addition, CP-293,019 and NRA-0045 did not reduce apomorphine- or methamphetamine-induced sereotypy (Zorn et al., 1996; Okuyama et al., 1997) or catalepsy (Zorn et al., 1996; Okuyama et al., 1997a,b). These data suggest that CP-293,019 and NRA 0045 would display low liability for extrapyramidal dysfunction. However, none of these compounds have yet been tested in clinical trials.

Currently, L-745,870 is the only selective dopamine D_4 receptor antagonist to have been tested in both behavioral experimental animal studies and in clinical trials. However, results from experimental animal studies have been conflicting. While one study reported that L-745,870 was effective in reversing apomorphine-induced disruption of pre-pulse inhibition (Mansbach et al., 1998), another study reported that L-745,870 was ineffective (Bristow et al., 1997). This latter study also reported that L-745,870 was ineffective in reversing amphetamine-induced hyperactivity and apomorphine-induced stereotypy in mice (Bristow et al., 1997). In one clinical trial, L-745,870 was ineffective as an antipsychotic for treatment of neuroleptic responsive inpatients with acute schizophrenia (Kramer et al., 1997). From these data it would appear that dopamine D_4 blockade by itself is not sufficient to alleviate positive symptomatology of schizophrenia and supports the aforementioned apparent requirement for dopamine D_2 receptor blockade to inhibit psychosis. Nevertheless, since several other selective dopamine D_4 receptor antagonists have effects similar to those of clozapine in animal behavioral models, dopamine D_4 receptor blockade might contribute to the atypical profile of certain antipsychotics.

There is currently no information available to distinguish why, on a receptor basis, these dopamine D_4 receptor antagonists might display conflicting results in the behavioral paradigms. It is conceivable that these agents might display differential affinity at an, as yet, unidentified receptor subtype. In addition, it still remains to be determined whether the other selective dopamine D_4 receptor antagonists, which had a better predictive antipsychotic profile than L-745,870 in behavioral paradigms, will be effective in clinical trials.

Further studies examining the role of the dopamine D_4 receptor suggest that it might play a role in modulating motor function. For example, in the dopamine D_4 receptor-deficient mouse (Rubinstein et al., 1997), apomorphine induced locomotor activity was measured following administration of low doses of clozapine, which would selectively block dopamine D_4 receptors, but not dopamine D_2 receptors. At low doses of clozapine, locomotor activity was attenuated in the wild-type mouse, but no effect on locomotor activity was detected in the dopamine D_4 receptor-deficient mouse. At higher doses of clozapine, locomotor activity was diminished to an equal extent in both wild-type and dopamine D_4 receptor-deficient mice, likely through blockade of striatal dopamine D_2 receptors. These data suggest that in the wild-type mouse, the dopamine D_4 receptor plays a role in modulating locomotor activity. Moreover, the dopamine D_4 knockout study suggests that the dopamine D_4 receptor modulates dopamine synthesis and turnover in striatum. In this regard, an elevated dopamine/DOPAC (dihydrophenylacetic acid) ratio was observed in dorsal striatum of dopamine D_4 deficient mice. Similarly, selective dopamine D_4 receptor antagonists have been found to increase dopamine release in the striatum (Wright et al., 1996; Youngren et al., 1997, but see Holland et al., 1996). Together, these studies suggest that antipsychotic agents with higher affinity at dopamine D_4 than D_2 receptors, will demonstrate lower liability for extrapyramidal side effects, possibly through increased synaptic dopamine levels which could displace the drug from striatal dopamine D_2 receptors (see Fig. 3). This phenomenon is more likely to apply to 'loose' dopamine D_2 receptor antagonists.

Some selective D_4 receptor antagonists (CP 293,019 and RO 61-6270) mediate increased c-*fos* expression in prefrontal cortex and nucleus accumbens, but not in striatum in rat brain (Hartman et al., 1996; Holland et al., 1996). A similar result was obtained following administration of clozapine (Holland et al., 1996). Since c-*fos* is an indicator of neuronal activity, elevated c-*fos* expression in frontal cortex and nucleus accumbens infers regional specificity of action in brain areas implicated in antipsychotic action. The c-*fos* induction pattern of highly selective dopamine D_4 receptor antagonists, which mimics that of clozapine, but not of typical antipsychotics (Robertson and Fibiger, 1992), might also be predictive of efficacy against negative symptoms of schizophrenia and low propensity for extrapyramidal dysfunction.

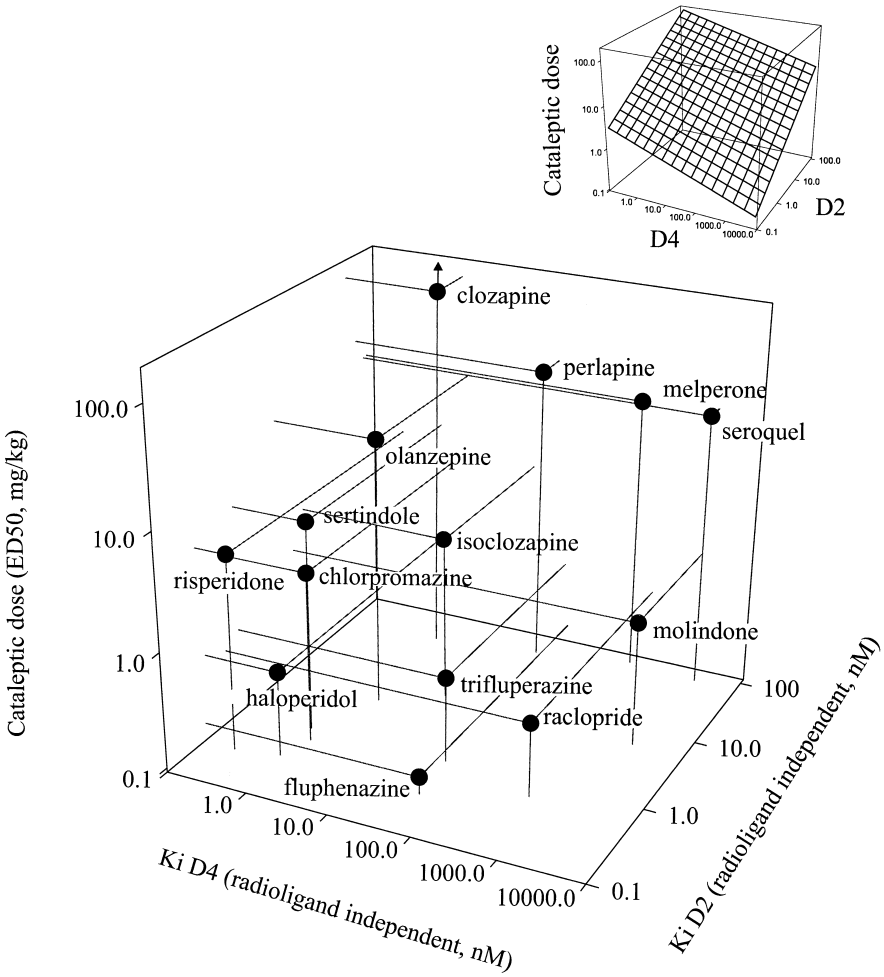

Fig. 3. Three dimensional plot of the ligand independent affinities [nM] of various antipsychotic drugs to the dopamine D_2 and D_4 receptors versus the half maximal dose (ED50, mg/kg) required to produce catalepsy in rats. The maximal dose to induce catalepsy for clozapine could not be determined, which is indicated with an arrow. The insert at the top right shows the plane on which most of the drugs would fall, reflecting the relative contribution of dopamine D_4 receptor affinity and change in cataleptic index. The data shown are taken from Seeman et al., 1997a.

The recent observation that olanzapine- and loxapine-induced, but not haloperidol-induced, catalepsy can be reversed by co-administration of clozapine (Kalkman et al., 1997) suggests that an additional receptor site might contribute to clozapine's reduced liability for motor side-effects. Since olanzepine, loxapine and clozapine display a near identical pharmacological profile, blocking the various serotonin, dopamine and muscarinic sites at equivalent potencies (Roth et al., 1994; Bymaster et al., 1996) this observation suggests the existence of an additional, as yet, unidentified target for clozapine's anti-cataleptic effect and unique atypical profile.

5. Conclusion

In conclusion, the preponderance of evidence supports the hypothesis that reduction in positive symptomatology of schizophrenia by antipsychotic agents is mediated almost exclusively through blockade of dopamine D_2 receptors. The liability for antipsychotic agents to produce extrapyramidal side-effects is also tightly correlated with dopamine D_2 receptor blockade, although the likelihood and/or severity of motor dysfunction appears to be diminished for low affinity dopamine D_2 receptor blockers. While exclusive blockade at the dopamine D_4 receptor may not be sufficient for antipsychotic action, it is tempting to speculate that in combination with loose dopamine D_2 receptor blockade, it may display an improved profile to classic typical neuroleptics.

Acknowledgements

We would like to extend our sincere gratitude to Dr. Philip Seeman and Dr. Jose Nobrega for their helpful comments in preparing this manuscript. This work is supported by the Medical Research Council of Canada and the Ontario Mental Health Foundation. Hubert H.M. Van Tol is a Career Scientist of the Ontario Ministry of Health.

References

Amalric, M., Koob, G.F., 1993. Functionally selective neurochemical afferents and efferents of the mesocorticolimbic and nigrostriatal dopamine system. Prog. Brain Res. 99, 209–226.

Ariano, M.A., Wang, J., Noblett, K.L., Larson, E.R., Sibley, D.R., 1997. Cellular distribution of the rat D4 dopamine receptor protein in the CNS using anti-receptor antisera. Brain Res. 752, 26–34.

Arnt, J., Skarsfeldt, T., 1998. Do novel antipsychotics have similar pharmacological characteristics? A review of the evidence. Neuropsychopharmacology 18, 63–101.

Asghari, V., Schoots, O., van Kats, S., Ohara, K., Jovanovic, V., Guan, H.C., Bunzow, J.R., Petronis, A., Van Tol, H.H.M., 1994. Dopamine D_4 receptor repeat: analysis of different native and mutant forms of the human and rat genes. Mol. Pharmacol. 46, 364–373.

Baik, J.H., Picetti, R., Saiardi, A., Thiriet, G., Dierich, A., Depaulis, A., Le Meur, M., Borrelli, E., 1995. Parkinsonian-like locomotor impairment in mice lacking dopamine D_2 receptors. Nature 377, 424–428.

Bischoff, S., Gunst, F., 1997. Distinct binding patterns of [^{3}H]raclopride and [^{3}H]spiperone at dopamine D_2 receptors in vivo in rat brain. Implications for pet studies. J. Recept. Signal Transduction Res. 17, 419–431.

Brenner, H.D., Dencker, S.J., Goldstein, M.J., Hubbard, J.W., Keegan, D.L., Kruger, G., Kulhanek, F., Liberman, R.P., Malm, U., Midha, K.K., 1990. Defining treatment refractoriness in schizophrenia. Schizophr. Bull. 16, 551–561.

Bristow, L.J., Collinson, N., Cook, G.P., Curtis, N., Freedman, S.B., Kulagowski, J.J., Leeson, P.D., Patel, S., Ragan, C.I., Ridgill, M., Saywell, K.L., Tricklebank, M.D., 1997. L-745,870, a subtype selective dopamine D4 receptor antagonist, does not exhibit a neuroleptic-like profile in rodent behavioral tests. J. Pharmacol. Exp. Ther. 283, 1256–1263.

Bunzow, J.R., Van Tol, H.H.M., Grandy, D.K., Albert, P., Salon, J., Christie, M., Machida, C.A., Neve, K.A., Civelli, O., 1988. Cloning and expression of a rat D_2 dopamine receptor cDNA. Nature 336, 783–787.

Bymaster, F.P., Calligaro, D.O., Falcone, J.F., Marsh, R.D., Moore, N.A., Tye, N.C., Seeman, P., Wong, D.T., 1996. Radioreceptor binding profile of the atypical antipsychotic olanzapine. Neuropsychopharmacology 14, 87–96.

Cardno, A.G., Farmer, A.E., 1995. The case for or against heterogeneity in the etiology of schizophrenia. The genetic evidence. Schizophr. Res. 17, 153–159.

Casey, D.E., 1995. Motor and mental aspects of extrapyramidal syndromes. Int. Clin. Psychopharmacol. 10 (Suppl.), 105–114.

Corbin, A.E., Heffner, T.G., 1997. Effects of dopamine (DA) D4 antagonists on prepulse inhibition of acoustic startle and amphetamine-stimulated locomotion in rats. Soc. Neurosci. Abstr. 939.13.

Corbin, A.E., Pugsley, T.A., Akunne, H.C., Whetzel, S.Z., Zoski, K.T., Georgic, L.M., Nelson, C.B., Wright, J.L., Wise, L.D., Heffner, T.G., 1998. Pharmacological characterization of PD 152255, a novel dimeric benzimidazole dopamine D3 antagonist. Pharmacol. Biochem. Behav. 59, 487–493.

Creese, I., Burt, D.R., Snyder, S.H., 1976. Dopamine receptor binding predicts clinical and pharmacological potencies of antischizophrenic drugs. Science 192, 481–483.

Csernansky, J.G., Murphy, G.M., Faustman, W.O., 1991. Limbic/mesolimbic connections and the pathogenesis of schizophrenia. Biol. Psychiatry 30, 383–400.

Dal Toso, R., Sommer, B., Ewert, M., Herb, A., Pritchett, D.B., Bach, A., Shivers, B.D., Seeburg, P.H., 1989. The dopamine D_2 receptor: two molecular forms generated by alternative splicing. Embo J. 8, 4025–4034.

Davis, K.L., Kahn, R.S., Ko, G., Davidson, M., 1991. Dopamine in schizophrenia: a review and reconceptualization. Am. J. Psychiatry 148, 1474–1486.

Dearry, A., Gingrich, J.A., Falardeau, P., Fremeau, R.T. Jr., Bates, M.D., Caron, M.G., 1990. Molecular cloning and expression of the gene for a human D1 dopamine receptor. Nature 347, 72–76.

DeHaven-Hudkins, D.L., Fleissner, L.C., Ford-Rice, F.Y., 1992. Characterization of the binding of [^{3}H](+)-pentazocine to sigma recognition sites in guinea pig brain. Eur. J. Pharmacol. 227, 371–378.

Deutch, A.Y., Moghaddam, B., Innis, R.B., Krystal, J.H., Aghajanian, G.K., Bunney, B.S., Charney, D.S., 1991. Mechanisms of action of atypical antipsychotic drugs. Implications for novel therapeutic strategies for schizophrenia. Schizophr. Res. 4, 121–156.

Farde, L., Hall, H., Pauli, S., Halldin, C., 1995. Variability in D2-dopamine receptor density and affinity: a PET study with [^{11}C]raclopride in man. Synapse 20, 200–208.

Farde, L., Nordstrom, A.L., 1993. PET examination of central D2 dopamine receptor occupancy in relation to extrapyramidal syndromes in patients being treated with neuroleptic drugs. Psychopharmacol. Ser. 10, 94–100.

Farde, L., Nordstrom, A.L., Nyberg, S., Halldin, C., Sedvall, G., 1994. D1-, D2-, and 5-HT2-receptor occupancy in clozapine-treated patients. J. Clin. Psychiatry 55 (B), 67–69.

Farde, L., Nordstrom, A.L., Wiesel, F.A., Pauli, S., Halldin, C., Sedvall, G., 1992. Positron emission tomographic analysis of central D1 and D2 dopamine receptor occupancy in patients treated with classical neuroleptics and clozapine. Relation to extrapyramidal side effects. Arch. Gen. Psychiatry 49, 538–544.

Farde, L., Wiesel, F.A., Stone-Elander, S., Halldin, C., Nordstrom, A.L., Hall, H., Sedvall, G., 1990. D2 dopamine receptors in neuroleptic-naive schizophrenic patients. A positron emission tomography study with [^{11}C]raclopride. Arch. Gen. Psychiatry 47, 213–219.

Feinberg, A.P., Snyder, S.H., 1975. Phenothiazine drugs: structure-activity relationships explained by a conformation that mimics dopamine. Proc. Natl. Acad. Sci. USA 72, 1899–1903.

Feldpausch, D.L., Needham, L.M., Meng, Z.-H., Stone, M.P., Svensson, K.A., Merchant, K.M., 1996. Dopamine D4-selective antagonist, U-101387G, blocks amphetamine-sensitization in rats: behavioral and biochemical changes. Soc. Neurosci. Abstr. 697.14.

Garimella, B., Wu, H., Fitch, C.S., Essani, K., Merchant, K.M., 1997. Does the dopamine D4 receptor selective antagonis, PNU-101387G, block amphetamine sensitization by enhancing dynorphin neurotransmission? Soc. Neurosci. Abstr. 874.3.

Giros, B., Sokoloff, P., Martres, M.P., Riou, J.F., Emorine, L.J., Schwartz, J.C., 1989. Alternative splicing directs the expression of two D2 dopamine receptor isoforms. Nature 342, 923–926.

Grandy, D.K., Marchionni, M.A., Makam, H., Stofko, R.E., Alfano, M., Frothingham, L., Fischer, J.B., Burke-Howie, K.J., Bunzow, J.R., Server, A.C. et al., 1989. Cloning of the cDNA and gene for a human D2 dopamine receptor. Proc. Natl. Acad. Sci. U.S.A. 86, 9762–9766.

Grandy, D.K., Zhang, Y.A., Bouvier, C., Zhou, Q.Y., Johnson, R.A., Allen, L., Buck, K., Bunzow, J.R., Salon, J., Civelli, O., 1991. Multiple human D5 dopamine receptor genes: a functional receptor and two pseudogenes. Proc. Natl. Acad. Sci. U.S.A. 88, 9175–9179.

Gudelsky, G.A., Koenig, J.I., Simonovic, M., Koyama, T., Ohmori, T., Meltzer, H.Y., 1987. Differential effects of haloperidol, clozapine, and fluperlapine on tuberoinfundibular dopamine neurons and prolactin secretion in the rat. J. Neural Transm. 68, 227–240.

Gurevich, E.V., Bordelon, Y., Shapiro, R.M., Arnold, S.E., Gur, R.E., Joyce, J.N., 1997. Mesolimbic dopamine D3 receptors and use of antipsychotics in patients with schizophrenia. A postmortem study. Arch. Gen. Psychiatry 54, 225–232.

Hartman, D.S., Smeyne, R., Zenner, M.-T., Goepfert, C., Schlaeger, E.-J., Jenck, F., Civelli, O., Godel, T., Riemer. C., 1996. RO 61-6270, a new highly selective dopamine D4 receptor antagonist, induces c-*fos* expression in mouse cortex. Soc. Neurosci. Abstr. 202.1.

Helmeste, D.M., Tang, S.W., Li, M., Fang, H., 1997. Multiple [^{3}H]-nemonapride binding sites in calf brain. Naunyn-Schmiedeberg's Arch. Pharmacol. 356, 17–21.

Hietala, J., Syvalahti, E., Vuorio, K., Nagren, K., Lehikoinen, P., Ruot-

salainen, U., Rakkolainen, V., Lehtinen, V., Wegelius, U., 1994. Striatal D2 dopamine receptor characteristics in neuroleptic-naive schizophrenic patients studied with positron emission tomography. Arch. Gen. Psychiatry 51, 116–123.

Holland, J.P., Costello, D.G., de Wet, J.R., Rollema, H., Sanner, M.A., Zorn, S.G., Seeger, T.F., 1996. CP-293,029: a D4-selective dopamine antagonist produces clozapine-like effects on c-*fos* mRNA and dopamine levels in rat brain. Soc. Neurosci. Abstr. 697.2.

Joyce, J.N., Meador-Woodruff, J.H., 1997. Linking the family of D2 receptors to neuronal circuits in human brain: insights into schizophrenia. Neuropsychopharmacology 16, 375–384.

Kalkman, H.O., Neumann, V., Tricklebank, M.D., 1997. Clozapine inhibits catalepsy induced by olanzapine and loxapine, but prolongs catalepsy induced by SCH 23390 in rats. Naunyn-Schmiedeberg's Arch. Pharmacol. 355, 361–364.

Kane, J., Honigfeld, G., Singer, J., Meltzer, H., 1988a. Clozapine for the treatment-resistant schizophrenic. A double-blind comparison with chlorpromazine. Arch. Gen. Psychiatry 45, 789–796.

Kane, J.M., Freeman, H.L., 1994. Towards more effective antipsychotic treatment. Br. J. Psychiatry 25 (Suppl.), 22–31.

Kane, J.M., Honigfeld, G., Singer, J., Meltzer, H., 1988b. Clozapine in treatment-resistant schizophrenics. Psychopharmacol. Bull. 24, 62–67.

Kane, J.M., Lieberman, J.A., 1987. Maintenance pharmacotherapy in schizophrenia. In: Meltzer, H.Y. (Ed.), Psychopharmacology: The Third Generation of Progress. Raven Press, New York, pp. 1103–1109.

Karbe, H., Wienhard, K., Hamacher, K., Huber, M., Herholz, K., Coenen, H.H., Stocklin, G., Lovenich, A., Heiss, W.D., 1991. Positron emission tomography with [^{18}F]methylspiperone demonstrates D2 dopamine receptor binding differences of clozapine and haloperidol. J. Neural Transm. Gen. Sect. 86, 163–173.

Kelly, M.A., Rubinstein, M., Asa, S.L., Zhang, G., Saez, C., Bunzow, J.R., Allen, R.G., Hnasko, R., Ben-Jonathan, N., Grandy, D.K., Low, M.J., 1997. Pituitary lactotroph hyperplasia and chronic hyperprolactinemia in dopamine D2 receptor-deficient mice. Neuron 19, 103–113.

Kramer, M.S., Last, B., Getson, A., Reines, S.A., 1997. The effects of a selective D4 dopamine receptor antagonist (L-745,870) in acutely psychotic inpatients with schizophrenia: D4 Dopamine Antagonist Group. Arch. Gen. Psychiatry 54, 567–572.

Lahti, R.A., Roberts, R.C., Conley, R.R., Cochrane, E.V., Mutin, A., Tamminga, C.A., 1996. D2-type dopamine receptors in postmortem human brain sections from normal and schizophrenic subjects. Neuroreport 7, 1945–1948.

Levant, B., Vansell, N.R., 1997. In vivo occupancy of D2 dopamine receptors by nafadotride. Neuropsychopharmacology 17, 67–71.

Lieberman, J.A., Kane, J.M., Alvir, J., 1987. Provocative tests with psychostimulant drugs in schizophrenia. Psychopharmacology (Berlin) 91, 415–433.

Majchrzak, M.J., Zorn, S.H., Sanner, M.A., Seymour, P.A., 1997. Blockade of the locomotor activity response to apomorphine: a proposed in vivo behavioral assay to detect dopamine D4 receptor antagonists. Soc. Neurosci. Abstr. 165.8.

Mansbach, R.S., Brooks, E.W., Sanner, M.A., Zorn, S.H., 1998. Selective dopamine D4 receptor antagonists reverse apomorphine-induced blockade of prepulse inhibition. Psychopharmacology (Berlin) 135 2, 194–200.

Mansour, A., Meador-Woodruff, J.H., Bunzow, J.R., Civelli, O., Akil, H., Watson, S.J., 1990. Localization of dopamine D2 receptor mRNA and D1 and D2 receptor binding in the rat brain and pituitary: an in situ hybridization- receptor autoradiographic analysis. J. Neurosci. 10, 2587–2600.

Markstein, R., Gull, P., Rudeberg, C., Urwyler, S., Jaton, A.L., Kalkman, H.O., Dixon, A.K., Hoyer, D., 1996. SDZ GLC 756, a novel octahydrobenzo[*g*]quinoline derivative exerts opposing effects on dopamine D1 and D2 receptors. J. Neural Transm. 103, 17–30.

Marzella, P.L., Hill, C., Keks, N., Singh, B., Copolov, D., 1997. The binding of both [^{3}H]nemonapride and [^{3}H]raclopride is increased in schizophrenia. Biol. Psychiatry 42, 648–654.

Meador-Woodruff, J.H., Mansour, A., Bunzow, J.R., Van Tol, H.H.M., Watson, S.J. Jr., Civelli, O., 1989. Distribution of D2 dopamine receptor mRNA in rat brain. Proc. Natl. Acad. Sci. U.S.A. 86, 7625–7628.

Meador-Woodruff, J.H., Haroutunian, V., Powchik, P., Davidson, M., Davis, K.L., Watson, S.J., 1997. Dopamine receptor transcript expression in striatum and prefrontal and occipital cortex. Focal abnormalities in orbitofrontal cortex in schizophrenia. Arch. Gen. Psychiatry 54, 1089–1095.

Meltzer, H.Y., Gudelsky, G.A., 1992. Dopaminergic and serotonergic effects of clozapine. Implications for a unique clinical profile. Arzneimittelforschung 42, 268–272.

Meltzer, H.Y., Koenig, J.I., Nash, J.F., Gudelsky, G.A., 1989. Melperone and clozapine: neuroendocrine effects of atypical neuroleptic drugs. Acta Psychiatr. Scand. 352, 24–29.

Mengod, G., Martinez-Mir, M.I., Vilaro, M.T., Palacios, J.M., 1989. Localization of the mRNA for the dopamine D2 receptor in the rat brain by in situ hybridization histochemistry. Proc. Natl. Acad. Sci. USA 86, 8560–8564.

Merchant, K.M., Gill, G.S., Harris, D.W., Huff, R.M., Eaton, M.J., Lookingland, K., Lutzke, B.S., McCall, R.B., Piercey, M.F., Schreur, P.J., Sethy, V.H., Smith, M.W., Svensson, K.A., Tang, A.H., Vonvoigtlander, P.F., Tenbrink, R.E., 1996. Pharmacological characterization of U-101387, a dopamine D4 receptor selective antagonist. J. Pharmacol. Exp. Ther. 279, 1392–1403.

Millan, M.J., Gressier, H., Brocco, M., 1997. The dopamine D3 receptor antagonist, (+)-S 14297, blocks the cataleptic properties of haloperidol in rats. Eur. J. Pharmacol. 321, R7–9.

Mogenson, G.J., Yang, C.R., Yim, C.Y., 1988. Influence of dopamine on limbic inputs to the nucleus accumbens. Ann. New York Acad Sci. 537, 86–100.

Monsma, Jr., F.J., Mahan, L.C., McVittie, L.D., Gerfen, C.R., Sibley, D.R., 1990. Molecular cloning and expression of a D1 dopamine receptor linked to adenylyl cyclase activation. Proc. Natl. Acad. Sci. USA 87, 6723–6727.

Monsma, Jr., F.J., McVittie, L.D., Gerfen, C.R., Mahan, L.C., Sibley, D.R., 1989. Multiple D2 dopamine receptors produced by alternative RNA splicing. Nature 342, 926–929.

Mrzljak, L., Bergson, C., Pappy, M., Huff, R., Levenson, R., Goldman-Rakic, P.S., 1996. Localization of dopamine D4 receptors in GABAergic neurons of the primate brain. Nature 381, 245–248.

Murray, A.M., Hyde, T.M., Knable, M.B., Herman, M.M., Bigelow, L.B., Carter, J.M., Weinberger, D.R., Kleinman, J.E., 1995. Distribution of putative D4 dopamine receptors in postmortem striatum from patients with schizophrenia. J. Neurosci. 15, 2186–2191.

Najlerahim, A., Barton, A.J., Harrison, P.J., Heffernan, J., Pearson, R.C. 9.Me, 1989. Messenger RNA encoding the D2 dopaminergic receptor detected by in situ hybridization histochemistry in rat brain. FEBS Lett. 255, 335–339.

Nordin, C., Alme, B., Bondesson, U., 1995. CSF and serum concentrations of clozapine and its demethyl metabolite: a pilot study. Psychopharmacology (Berlin) 122, 104–107.

Nordstrom, A.L., Farde, L., Eriksson, L., Halldin, C., 1995. No elevated D2 dopamine receptors in neuroleptic-naive schizophrenic patients revealed by positron emission tomography and [^{11}C]*N*- methylspiperone. Psychiatry Res. 61, 67–83.

Okuyama, S., Chaki, S., Kawashima, N., Suzuki, Y., Ogawa, S., Imagawa, Y., Kawashima, N., Ikeda, Y., Kumagai, T., Nakazato, A., Nagamine, M., Tomisawa, K., 1997a. In vitro and in vivo characterization of the dopamine D_4 receptor, serotonin 5-HT_{2A} receptor and alpha-1 adrenoceptor antagonist (R)-(+)-2-amino-4-(4-fluorophenyl)-5-[1-[4-(4-fluorophenyl)-4-oxopyrrolidin-3-yl]thiazole(NRA0045). J. Pharmacol. Exp. Ther. 282, 56–63.

Okuyama, S., Chaki, S., Kawashima, N., Suzuki, Y., Ogawa, S., Kumagai, T., Nakazato, A., Nagamine, M., Yamaguchi, K., Tomisawa, K.,

1997b. The atypical antipsychotic profile of NRA0045, a novel dopamine D4 and 5-hydroxytryptamine2A receptor antagonist, in rats. Br. J. Pharmacol. 121, 515–525.

Patel, S., Freedman, S., Chapman, K.L., Emms, F., Fletcher, A.E., Knowles, M., Marwood, R., McAllister, G., Myers, J., Curtis, N., Kulagowski, J.J., Leeson, P.D., Ridgill, M., Graham, M., Matheson, S., Rathbone, D., Watt, A.P., Bristow, L.J., Rupniak, N.M., Baskin, E., Lynch, J.J., Ragan, C.I., 1997. Biological profile of L-745,870, a selective antagonist with high affinity for the dopamine D4 receptor. J. Pharmacol. Exp. Ther. 283, 636–647.

Pearlson, G.D., Wong, D.F., Tune, L.E., Ross, C.A., Chase, G.A., Links, J.M., Dannals, R.F., Wilson, A.A., Ravert, H.T., Wagner, H.N. Jr. et al., 1995. In vivo D2 dopamine receptor density in psychotic and nonpsychotic patients with bipolar disorder. Arch. Gen. Psychiatry 52, 471–477.

Pilowsky, L.S., Costa, D.C., Ell, P.J., Murray, R.M., Verhoeff, N.P., Kerwin, R.W., 1992. Clozapine, single photon emission tomography, and the D2 dopamine receptor blockade hypothesis of schizophrenia. Lancet 340, 199–202.

Primus, R.J., Thurkauf, A., Xu, J., Yevich, E., McInerney, S., Shaw, K., Tallman, J.F., Gallagher, D.W., 1997. II. Localization and characterization of dopamine D4 binding sites in rat and human brain by use of the novel, D4 receptor-selective ligand [^{3}H]NGD 94-1. J. Pharmacol. Exp. Ther. 282, 1020–1027.

Raleigh, F., 1996. Use of novel antipsychotic drugs. Pharmacotherapy 16, 160S–165S, discussion 166S–168S.

Randrup, A., Munkvad, I., 1965. Special antagonism of amphetamine-induced abnormal behaviour: inhibition of stereotyped activity with increase of some normal activities. Psychopharmacologia 7, 416–422.

Rao, P.A., Pickar, D., Gejman, P.V., Ram, A., Gershon, E.S., Gelernter, J., 1994. Allelic variation in the D4 dopamine receptor (DRD4) gene does not predict response to clozapine. Arch. Gen. Psychiatry 51, 912–917.

Robertson, G.S., Fibiger, H.C., 1992. Neuroleptics increase c-*fos* expression in the forebrain: contrasting effects of haloperidol and clozapine. Neuroscience 46, 315–328.

Roth, B.L., Craigo, S.C., Choudhary, M.S., Uluer, A., Monsma, F.J. Jr., Shen, Y., Meltzer, H.Y., Sibley, D.R., 1994. Binding of typical and atypical antipsychotic agents to 5- hydroxytryptamine-6 and 5-hydroxytryptamine-7 receptors. J. Pharmacol. Exp. Ther. 268, 1403–1410.

Rubinstein, M., Phillips, T.J., Bunzow, J.R., Falzone, T.L., Dziewczapolski, G., Zhang, G., Fang, Y., Larson, J.L., McDougall, J.A., Chester, J.A. Chest, Saez, C., Pugsley, T.A., Gershanik, O., Low, M.J., Grandy, D.K., 1997. Mice lacking dopamine D4 receptors are supersensitive to ethanol, cocaine, and methamphetamine. Cell 90, 991–1001.

Sautel, F., Griffon, N., Sokoloff, P., Schwartz, J.C., Launay, C., Simon, P., Costentin, J., Schoenfelder, A., Garrido, F., Mann, A. etal et al., 1995. Nafadotride, a potent preferential dopamine D3 receptor antagonist, activates locomotion in rodents. J. Pharmacol. Exp. Ther. 275, 1239–1246.

Schoots, O., Seeman, P., Guan, H.C., Paterson, A.D., Van Tol, H.H.M., 1995. Long-term haloperidol elevates dopamine D4 receptors by 2-fold in rats. Eur. J. Pharmacol. 289, 67–72.

Seeman, P., 1987. Dopamine receptors and the dopamine hypothesis of schizophrenia. Synapse 1, 133–152.

Seeman, P., 1992. Dopamine receptor sequences. Therapeutic levels of neuroleptics occupy D2 receptors, clozapine occupies D4. Neuropsychopharmacology 7, 261–284.

Seeman, P., 1995. Therapeutic receptor-blocking concentrations of neuroleptics. Int. Clin. Psychopharmacol. 10 (Suppl.), 5–13.

Seeman, P., Bzowej, N.H., Guan, H.C., Bergeron, C., Reynolds, G.P., Bird, E.D., Riederer, P., Jellinger, K., Tourtellotte, W.W., 1987. Human brain D1 and D2 dopamine receptors in schizophrenia: Alzheimer's, Parkinson's, and Huntington's diseases. Neuropsychopharmacology 1, 5–15.

Seeman, P., Corbett, R., Nam, D., Van Tol, H.H.M., 1996. Dopamine and serotonin receptors: amino acid sequences, and clinical role in neuroleptic parkinsonism. Jpn. J. Pharmacol. 71, 187–204.

Seeman, P., Corbett, R., Van Tol, H.H.M., 1997a. Atypical neuroleptics have low affinity for dopamine D2 receptors or are selective for D4 receptors. Neuropsychopharmacology 16, 93–110, discussion 111–135.

Seeman, P., Guan, H.C., Nobrega, J., Jiwa, D., Markstein, R., Balk, J.H., Picetti, R., Borrelli, E., Van Tol, H.H.M., 1997b. Dopamine D2-like sites in schizophrenia, but not in Alzheimer's, Huntington's, or control brains, for [^{3}H]benzquinoline. Synapse 25, 137–146.

Seeman, P., Guan, H.C., Van Tol, H.H.M., 1993. Dopamine D4 receptors elevated in schizophrenia. Nature 365, 441–445.

Seeman, P., Lee, T., 1975. Antipsychotic drugs: direct correlation between clinical potency and presynaptic action on dopamine neurons. Science 188, 1217–1219.

Seeman, P., Lee, T., Chau-Wong, M., Wong, K., 1976. Antipsychotic drug doses and neuroleptic/dopamine receptors. Nature 261, 717–719.

Seeman, P., Tallerico, T., 1998. Antipsychotic drugs which elicit little or no parkinsonism bind more loosely than dopamine to brain D2 receptors, yet occupy high levels of these receptors. Mol. Psychiatry in press.

Seeman, P., Tallerico, T., Corbett, R., Van Tol, H.H.M., Kamboj, R.K., 1997c. Role of dopamine D2, D4 and serotonin(2A) receptors in antipsychotic and anticataleptic action. J. Psychopharmacol. 11, 15–17.

Seeman, P., Van Tol, H.H.M., 1995. Deriving the therapeutic concentrations for clozapine and haloperidol: the apparent dissociation constant of a neuroleptic at the dopamine D2 or D4 receptor varies with the affinity of the competing radioligand. Eur. J. Pharmacol. 291, 59–66.

Shaikh, S., Collier, D., Kerwin, R.W., Pilowsky, L.S., Gill, M., Xu, W.M., Thornton, A., 1993. Dopamine D4 receptor subtypes and response to clozapine. Lancet 341, 116.

Snyder, S.H., 1973. Amphetamine psychosis: a model schizophrenia mediated by catecholamines. Am. J. Psychiatry 130, 61–67.

Sokoloff, P., Giros, B., Martres, M.P., Bouthenet, M.L., Schwartz, J.C., 1990. Molecular cloning and characterization of a novel dopamine receptor (D3) as a target for neuroleptics. Nature 347, 146–151.

Stefanis, N.C., Bresnick, J.N., Kerwin, R.W., Schofield, W.N., McAllister, G., 1998. Elevation of D4 dopamine receptor mRNA in postmortem schizophrenic brain. Brain Res. Mol. Brain Res. 53, 112–119.

Stone, M.P., Waters, N., Green, J.B., Myers, J.E., Lewis, R.A., Cimini, M.G., Svensson, K.A., 1996. Effects of the selective dopamine D4 antagonist U-101387G on locomotor activity and brain monoamine neurochemistry in the rat. Soci. Neurosci. Abstr. 697.17.

Sumiyoshi, T., Stockmeier, C.A., Overholser, J.C., Thompson, P.A., Meltzer, H.Y., 1995. Dopamine D4 receptors and effects of guanine nucleotides on [^{3}H]raclopride binding in postmortem caudate nucleus of subjects with schizophrenia or major depression. Brain Res. 681, 109–116.

Sunahara, R.K., Guan, H.C., BF, O.D., Seeman, P., Laurier, L.G., Ng, G., George, S.R., Torchia, J., Van Tol, H.H.M., Niznik, H.B., 1991. Cloning of the gene for a human dopamine D5 receptor with higher affinity for dopamine than D1. Nature 350, 614–619.

Sunahara, R.K., Niznik, H.B., Weiner, D.M., Stormann, T.M., Brann, M.R., Kennedy, J.L., Gelernter, J.E., Rozmahel, R., Yang, Y.L., Israel, Y. et al., 1990. Human dopamine D1 receptor encoded by an intronless gene on chromosome 5. Nature 347, 80–83.

Tallman, J.F., Primus, R.J., Brodbeck, R., Cornfield, L., Meade, R., Woodruff, K., Ross, P., Thurkauf, A., Gallager, D.W., 1997. INGD 94-1: Identification of a novel, high affinity antagonist at the human dopamine D4 receptor. J. Pharmacol. Exp. Ther. 282, 1011–1019.

Tam, S.W., Cook, L., 1984. Sigma opiates and certain antipsychotic drugs mutually inhibit (+)-[^{3}H] SKF 10,047 and [^{3}H]haloperidol binding in guinea pig brain membranes. Proc. Natl. Acad. Sci. USA 81, 5618–5621.

Tamminga, C.A., Gerlach, J., 1987, New Neuroleptics and experimental antipsychotics in schizophrenia. In: Meltzer, H.Y. (Ed.), Psychopharmacology: The Third Generation of Progress. Raven Press, New York.

Tarazi, F.I., Kula, N.S., Baldessarini, R.J., 1997a. Regional distribution of dopamine D4 receptors in rat forebrain. Neuroreport 8, 3423–3426.

Tarazi, F.I., Yeghiayan, S.K., Baldessarini, R.J., Kula, N.S., Neumeyer, J.L., 1997b. Long-term effects of S(+)*N*-*n*-propylnorapomorphine compared with typical and atypical antipsychotics: differential increases of cerebrocortical D2-like and striatolimbic D4-like dopamine receptors. Neuropsychopharmacology 17, 186–196.

Tiberi, M., Jarvie, K.R., Silvia, C., Falardeau, P., Gingrich, J.A., Godinot, N., Bertrand, L., Yang-Feng, T.L., Fremeau, R.T. Jr., Caron, M.G., 1991. Cloning, molecular characterization, and chromosomal assignment of a gene encoding a second D1 dopamine receptor subtype: differential expression pattern in rat brain compared with the D1A receptor. Proc. Natl. Acad. Sci. USA 88, 7491–7495.

Tsuang, M.T., Farrone, S.V., 1995. The case for heterogeneity in the etiology of schizophrenia. Schizophr. Res. 17, 161–175.

Tune, L., Barta, P., Wong, D., Powers, R.E., Pearlson, G., Tien, A.Y., Wagner, H.N., 1996. Striatal dopamine D2 receptor quantification and superior temporal gyrus: volume determination in 14 chronic schizophrenic subjects. Psychiatry Res. 67, 155–158.

Tune, L.E., Wong, D.F., Pearlson, G., Strauss, M., Young, T., Shaya, E.K., Dannals, R.F., Wilson, A.A., Ravert, H.T., Sapp, J. et al., 1993. Dopamine D2 receptor density estimates in schizophrenia: a positron emission tomography study with ^{11}C *N*-methylspiperone. Psychiatry Res. 49, 219–237.

Van Tol, H.H.M., Bunzow, J.R., Guan, H.C., Sunahara, R.K., Seeman, P., Niznik, H.B., Civelli, O., 1991. Cloning of the gene for a human dopamine D4 receptor with high affinity for the antipsychotic clozapine. Nature 350, 610–614.

Von Bahr, C., Wiesel, F.A., Movin, G., Eneroth, P., Jansson, P., Nilsson, L., Ogenstad, S., 1991. Neuroendocrine responses to single oral doses of remoxipride and sulpiride in healthy female and male volunteers. Psychopharmacology (Berlin) 103, 443–448.

Weiner, D.M., Brann, M.R., 1989. The distribution of a dopamine D2 receptor mRNA in rat brain. FEBS Lett. 253, 207–213.

Wong, D.F., Wagner, H.N. Jr., Tune, L.E., Dannals, R.F., Pearlson, G.D., Links, J.M., Tamminga, C.A., Broussolle, E.P., Ravert, H.T., Wilson, A.A. et al., 1986. Positron emission tomography reveals elevated D2 dopamine receptors in drug-naive schizophrenics. Science 234, 1558–1563.

Wong, W.F., Pearlson, G.D. D.T, Tune, L.E., Young, L.T., Meltzer, C.C., Dannals, R.F., Ravert, H.T., Reith, J., Kuhar, M.J., Gjedde, A., 1997. Quantification of neuroreceptors in the living human brain: IV. Effect of aging and elevations of D2-like receptors in schizophrenia and bipolar illness. J. Cereb. Blood Flow Metab. 17, 331–342.

Wright, J.L., Gregory, T.F., Heffner, T.G., MacKenzie, R.G., Pugsley, T.A., Bander Meulen, S.J., Wise, L.D., 1996. A Novel series of 1-aryloxy-3(4-aryloxypiperidinyl)-2-propanols and potent, selective dopamine D4 receptor antagonists. Soc. Neurosci. Abstr. 189.14.

Youngren, K.D., Jentsch, J.D., Tran, A., Roth, R.H., 1997. Dopamine D4 receptors regulate dopamine synthesis without affecting utilization of release. Soc. Neurosci. Abstr. 271.7.

Zhou, Q.Y., Grandy, D.K., Thambi, L., Kushner, J.A., Van Tol, H.H.M., Cone, R., Pribnow, D., Salon, J., Bunzow, J.R., Civelli, O., 1990. Cloning and expression of human and rat D1 dopamine receptors. Nature 347, 76–80.

Zorn, S.H., Johnson, C.G., Jackson, E.R., Seymour, P., Majchrzak, M., Mansbach, R., Winson, E., de Wet, J.R., Dunaiskis, A., Chappie, T.A., Sanner, M.A., 1996. CP-293,029: A D4 dopamine antagonist with an in vitro and in vivo profile suggestive of an atypical antipsychotic. Soc. Neurosci. Abstr. 697.1.