

# Expert Opinion

1. Introduction
2. Before the discovery of dopamine receptors
3. Therapeutic concentrations of antipsychotics
4. Actions of antipsychotic drugs on neurons
5. The dopamine D<sub>1</sub> receptor
6. Discovery of the antipsychotic dopamine D<sub>2</sub> receptor
7. Nomenclature of dopamine receptors
8. Therapeutic concentrations of all antipsychotics occupy 60 – 75% of D<sub>2</sub>
9. 'Fast-off-D<sub>2</sub>' theory of atypical antipsychotic action
10. Are serotonin receptors a clinical target for atypical antipsychotics?
14. Additional evidence for D<sub>2</sub> as the therapeutic target for antipsychotics
15. No evidence for D<sub>3</sub> as an antipsychotic target
16. No evidence for D<sub>4</sub> as an antipsychotic target
17. D<sub>2</sub><sup>High</sup> as the antipsychotic target
18. Non-dopamine receptor targets
19. Expert opinion and future outlook

For reprint orders,  
please contact:  
[ben.fisher@informa.com](mailto:ben.fisher@informa.com)

**informa**  
healthcare

Central & Peripheral Nervous Systems

## Targeting the dopamine D<sub>2</sub> receptor in schizophrenia

Philip Seeman

*University of Toronto, Pharmacology and Psychiatry Departments, Faculty of Medicine,  
Medical Science Building, Room 4344, 1 King's College Circle, Toronto, M5S 1A8, Canada*

After a 12-year search for the antipsychotic receptor, the binding site was discovered and labelled by [<sup>3</sup>H]haloperidol in 1975. Of the various neurotransmitters, dopamine was the most potent in inhibiting the binding of [<sup>3</sup>H]haloperidol, indicating that the antipsychotic receptor was a dopamine receptor, now named the dopamine D<sub>2</sub> receptor, a major targeting site in schizophrenia. All antipsychotic drugs, including traditional and newer antipsychotics, either bind to D<sub>2</sub> in direct relation to their clinical potencies or hinder normal dopamine neurotransmission, as in the case of partial dopamine agonists. In fact, the antipsychotic concentrations found in the plasma water of treated patients closely match the predicted therapeutic absolute concentrations, adjusted for the 60 – 75% D<sub>2</sub> occupancy needed for clinical efficacy. Antipsychotics that elicit low or no Parkinsonism or prolactinaemia are loosely attached to D<sub>2</sub> and rapidly dissociate from D<sub>2</sub>, whereas those eliciting Parkinsonism stay tightly attached to D<sub>2</sub> for many hours. Because animal models of psychosis (amfetamine sensitisation, brain lesions) all show a marked elevation in the number of high-affinity states of D<sub>2</sub>, the antipsychotics are thought to specifically target these D<sub>2</sub><sup>High</sup> states in psychosis in general and schizophrenia in particular.

**Keywords:** antipsychotic, brain imaging, domperidone, dopamine receptor, psychosis, schizophrenia, supersensitivity

*Expert Opin. Ther. Targets (2006) 10(4):515-531*

### 1. Introduction

The discovery of dopamine receptors is closely associated with the discovery of antipsychotic drugs. The research in this field started with the synthesis of antihistamines after World War II, particularly with H Laborit using such compounds to enhance surgical analgesia. In patients receiving these various antihistamines, Laborit noticed a 'euphoric quietude', and that the patients were 'calm and somnolent, with a relaxed and detached expression.' Of this series of Rhône Poulenc compounds, RP4560, now known as chlorpromazine, was the most clinically acceptable.

Chlorpromazine was soon tested for various medical illnesses. Although Sigwald and Bouttier [1] were the first to use chlorpromazine as the sole drug for a psychotic patient, they did not report their findings until 1953, after a 1952 report by Delay *et al.* [2] that chlorpromazine alleviated hallucinations and stopped internal 'voices' in eight patients. An important feature of the action of chlorpromazine was that it was effective within three days. This relatively fast improvement, especially during the first week of antipsychotic treatment, has been observed many times, as summarised by Agid *et al.* [3].

The clinical success of chlorpromazine fostered the search to locate the therapeutic target and mode of action of chlorpromazine. The assumption then, as now, was that finding such a therapeutic target would open the avenue to uncovering the biochemical cause of psychosis and possibly schizophrenia.

## 2. Before the discovery of dopamine receptors

In searching for the therapeutic target and mechanism of action of chlorpromazine in the 1960s and 1970s, many types of physiological and biochemical experiments were carried out. A variety of possible therapeutic targets were explored for the mode of action of chlorpromazine, including its action on mitochondrial enzymes, sodium–potassium-ATPase and related enzymes, as well as its membrane-stabilising action, such as its strong potency to inhibit membrane action potentials, and to stabilise cellular and subcellular membranes from releasing their contents [4]. It also became clear in 1963 that all antipsychotics were surface active, readily explaining their hydrophobic affinity for biomembranes. Some of these non-receptor findings, such as the potent surface activities of the antipsychotics, showed an astonishingly excellent correlation with clinical antipsychotic potencies [5].

Because high doses of chlorpromazine and other antipsychotics (or 'neuroleptics', as they were then called) also elicited Parkinsonism as an unwanted side effect, basic scientists soon focused on the antipsychotic action on brain dopamine pathways. The reason for examining the brain dopamine pathway was based on the finding by Ehringer and Hornykiewicz [6] that the Parkinsonism of Parkinson's disease was associated with a major loss of brain dopamine. It was thought, therefore, that the unwanted side effect of chlorpromazine-induced Parkinsonism, as well as the antipsychotic action itself, might arise by antipsychotics interfering with dopamine neurotransmission. The working hypothesis was that if the brain target or targets for antipsychotics could be found, then it was possible that these targets could be overactive or underactive in psychosis or schizophrenia.

## 3. Therapeutic concentrations of antipsychotics

However, these early experiments in the 1960s revealed that the active concentrations *in vitro* of the antipsychotics were generally very high, between 20 nM and 100 nM [4]. Such concentrations, however, were far in excess of the nanomolar concentrations (e.g., 1 – 2 nM for haloperidol) that exist in the plasma water and in the spinal fluid in patients being successfully treated with these medications, as determined by plasma haloperidol measurements where the amount of haloperidol bound by the plasma proteins (92%) was taken into account [7]. It should be noted that an antipsychotic drug can exist in either a positively charged form or in a neutral uncharged form. The neutral form readily permeates cell membranes, and its concentration in the aqueous phase in the plasma is expected to be identical to the aqueous concentration of the antipsychotic in the spinal fluid in contact with the neurons.

## 4. Actions of antipsychotic drugs on neurons

Indirect evidence for the existence of distinct dopamine receptors on neurons and their sensitivity to antipsychotics came from *in vitro* and *in vivo* experiments, showing that dopamine agonists can excite or inhibit neurons in the nigrostriatal dopamine pathway. Moreover, other workers showed that direct application of dopamine on neurons also stimulated or inhibited snail neurons [8], and that haloperidol or fluphenazine could block these actions [9]. Here too, however, the antipsychotic concentrations used were far higher than those found in the plasma water or spinal fluid in patients; in fact, the concentrations used would be lethal to humans.

Additional work *in vivo* found that chlorpromazine and haloperidol increased the turnover of adrenaline and dopamine, as shown by the increased production of normetanephrine and methoxytyramine, respectively. To explain the increased production of these metabolites, it was suggested that 'the most likely [mechanism] appears to be that chlorpromazine and haloperidol block monoaminergic receptors in brain; as is well known, they block the effects of accumulated 5-hydroxytryptamine' [10]. In other words, these authors proposed that antipsychotics might block all three types of receptors for noradrenaline, dopamine and serotonin, but they did not identify which receptor was selectively blocked or how to identify or test any of these receptors directly *in vitro*. This study [10] in 1963 by Carlsson and Lindqvist is often mistakenly cited as discovering 'the dopamine receptor' and that antipsychotics selectively acted on this receptor. However, in 1964 N-E Andén, a student of A Carlsson, had a different view, and proposed that 'chlorpromazine and haloperidol delays the elimination of the (metabolites)' [11]. Moreover, seven years later, Andén *et al.* [12] reported that antipsychotics increased the turnover of both dopamine and noradrenaline, but they could not show that the antipsychotics were selective in blocking dopamine; for example, chlorpromazine enhanced the turnover of noradrenaline and dopamine equally. Therefore, it remained for *in vitro* radio-receptor assays to detect the dopamine receptor directly and to demonstrate antipsychotic selectivity for the dopamine receptor.

## 5. The dopamine D<sub>1</sub> receptor

With the advent of assays for adenylyl cyclase in the 1960s, it was found that dopamine stimulated adenylyl cyclase in the superior cervical ganglion [13]. This receptor was later named the dopamine D<sub>1</sub> receptor, selectively labelled by [<sup>3</sup>H]SCH 23390, and subsequently cloned by three research groups in 1990.

The dissociation constants at D<sub>1</sub> for the antipsychotics are given in Table 1. There is no correlation between the antipsychotic clinical doses and the dissociation constants of the antipsychotic antagonists at D<sub>1</sub>, as illustrated in Figure 1.

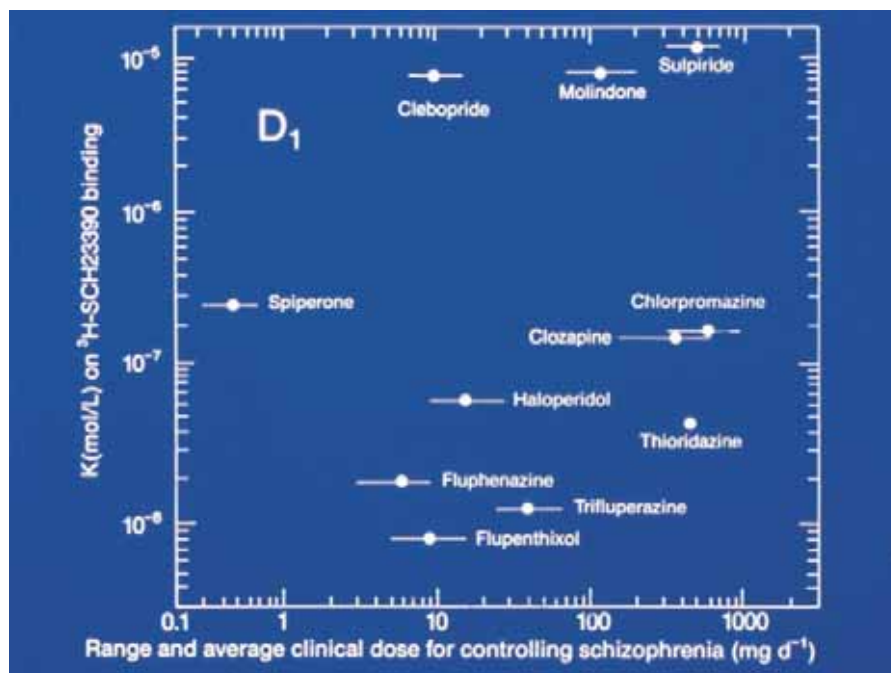


Figure 1. There is no correlation between the clinical antipsychotic doses and the antipsychotic dissociation constants (or concentrations) that inhibit the binding of a  $D_1$  ligand ( $[^3H]$ SCH23390) at dopamine  $D_1$  receptors in homogenised striatal tissue. The high concentrations inhibiting the  $D_1$  receptor are far higher than those found clinically in the plasma water or spinal fluid.

Adapted from [14] with permission.

These data suggested that  $D_1$  was not the major or common target for antipsychotics. Equally important, moreover, is the fact that the antipsychotic molarities at  $D_1$  are between 10 nM and 10,000 nM, far in excess of the therapeutic concentrations in the spinal fluid of treated patients.

In addition to the lack of targeting  $D_1$  receptors by clinical doses of the common antipsychotics,  $D_1$ -selective antagonists have not been found effective as antipsychotics.

## 6. Discovery of the antipsychotic dopamine $D_2$ receptor

In 1974 and 1975, in order to detect and discover the dopamine receptors on which the antipsychotics presumably acted, it was essential to label a receptor with a ligand, such as radioactive haloperidol, having an affinity (or dissociation constant) of  $\sim 1$  nM, because, as indicated above, this was the haloperidol therapeutic concentration found in the spinal fluid or plasma water of treated patients. For this to occur, the specific activity of  $[^3H]$ haloperidol would have to be at least 10 Ci/mmol. Although  $[^3H]$ haloperidol donated in 1971 by Janssen Pharmaceutica only had a low specific activity of 0.07 Ci/mmol, I.R.E. Belgique custom synthesised  $[^3H]$ haloperidol (10.5 Ci/mmol) for Seeman's laboratory by June 1974.

Specific binding of this new  $[^3H]$ haloperidol to brain striatal tissue was readily detected in 1975 [15]. It was soon found that all the antipsychotics inhibited the binding of  $[^3H]$ haloperidol in direct relation to their clinical potencies [16], as shown in Figure 2.

The data in Figure 2, therefore, indicated that the 'antipsychotic receptor' had finally been successfully discovered. Equally important, of the endogenous compounds tested, dopamine was the most potent in inhibiting the binding of  $[^3H]$ haloperidol, indicating that the antipsychotic receptor was actually a dopamine receptor.

## 7. Nomenclature of dopamine receptors

The antipsychotic dopamine receptor labelled by  $[^3H]$ haloperidol was later termed the  $D_2$  receptor [17]. It is important to note that the data for the binding of  $[^3H]$ haloperidol identifying the antipsychotic receptor [15] was very different from the pattern of  $[^3H]$ dopamine binding described by Burt *et al.* [18] and Snyder *et al.* [19]. For example, the binding of  $[^3H]$ haloperidol was inhibited by  $\sim 5000$  nM dopamine, whereas that of  $[^3H]$ dopamine was inhibited by  $\sim 3$  nM dopamine. For several years, this latter  $[^3H]$ dopamine binding site was termed the ' $D_3$  site' [20], a term which is not to be confused with the later discovery of the dopamine  $D_3$  receptor [21].

**Table 1.** *K* values (dissociation constants, nM)

Human clone	M <sub>1</sub>	D <sub>1</sub>	D <sub>2</sub>	D <sub>2</sub>	D <sub>2</sub>	D <sub>3</sub>	D <sub>4</sub>	D <sub>4</sub>
<b>[<sup>3</sup>H] ligand used:</b>	<b>QNB</b>	<b>Sch.</b>		<b>Raclo.</b>	<b>Spip.</b>	<b>Raclo.</b>		<b>Spip.</b>
[ <sup>3</sup> H]Amisulpride			1.7	1.8	4.6			
Amoxapine	49		–	21	56			5.5
Aripiprazole				1.8				
Butaclamol-(+)				0.14	0.9			70
Chlorpromazine	3.4	17		1.2	4.6	1.4		9.6
[ <sup>3</sup> H]Chlorpromazine			0.77				1.2	–
Clozapine	9.5	90	–	75	180	190	–	22
[ <sup>3</sup> H]Clozapine			51				2	–
Clozapine-iso	14.4	120		15	60	21		21
Clozapine-desmethyl	17	73		180	300			120
Cyproheptadine				24				
Epidepride				0.04	0.06			
Flupentixol- <i>cis</i>		1.8		0.38	0.7	0.7		15
Flupentixol- <i>trans</i>				151				
Fluphenazine		2.6		0.55	1.2	0.17		30
Haloperidol	794	55		0.74	2.7	8.8		2
[ <sup>3</sup> H]Haloperidol			0.4				0.85	–
Iloperidone (HP873)	109	5.6		5.4	10	20	–	9.6
Loxapine	117	18		9.2	22.7	7.2		8
Loxapine-iso	203	16		22	86	18		11
Melperone		148		152	375	315		720
Molindone		1558		4.9	15	44		3900
Olanzapine	2.1	9.2		7.4	21	14		15
[ <sup>3</sup> H]Olanzapine			2.7				1.6	–
Perphenazine		4.5		0.27	0.47	0.23	–	32
Pimozide	470			1.4	0.95			
Prochlorperazine		7.7		1.7	4			89
Quetiapine	135	290		140	680	240		2000
[ <sup>3</sup> H]Quetiapine			104					–
Raclopride			–	1.6	7.1	2.9		2400
[ <sup>3</sup> H]Raclopride			1.9			1.6		–
Remoxipride	> 10 μM	4900		67	800	960		2400
Risperidone	> 10 μM	42		1.09	4	3.5		4.4
Risperidone-9-OH				1.6				
Sertindole	400	22	3	1.9	6.5	3	–	11
[ <sup>3</sup> H]Sertindole			1.2				0.85	–

Dissociation constants (*K* values) for antipsychotics at cloned dopamine D<sub>1</sub>, D<sub>2</sub>, D<sub>3</sub> and D<sub>4</sub> receptors, and at the cholinergic muscarinic M<sub>1</sub> receptor. Because [<sup>3</sup>H]raclopride is more loosely bound to D<sub>2</sub> than [<sup>3</sup>H]spiperone, the antipsychotic *K* values are consistently lower at D<sub>2</sub> when measured with [<sup>3</sup>H]raclopride. Data from [30]. Blank boxes indicate not done. QNB: [<sup>3</sup>H]Quinuclidinylbenzilate (cholinergic muscarinic ligand); Raclo.: [<sup>3</sup>H]Raclopride; Sch.: [<sup>3</sup>H]Schering 23390; Spip.: [<sup>3</sup>H]Spiperone.

Table 1. *K* values (dissociation constants, nM) (continued)

Human clone	M <sub>1</sub>	D <sub>1</sub>	D <sub>2</sub>	D <sub>2</sub>	D <sub>2</sub>	D <sub>3</sub>	D <sub>4</sub>	D <sub>4</sub>
Spiperone			–	0.02	0.06		–	
[ <sup>3</sup> H]Spiperone			0.07			0.32	0.09	–
Sulpiride-S				9.9	8	10		1000
Trifluoperazine		2.9		1.4	3.8	0.7		39
Ziprasidone	265	9		2.7	6	1.5		8

Dissociation constants (*K* values) for antipsychotics at cloned dopamine D<sub>1</sub>, D<sub>2</sub>, D<sub>3</sub> and D<sub>4</sub> receptors, and at the cholinergic muscarinic M<sub>1</sub> receptor.

Because [<sup>3</sup>H]raclopride is more loosely bound to D<sub>2</sub> than [<sup>3</sup>H]spiperone, the antipsychotic *K* values are consistently lower at D<sub>2</sub> when measured with [<sup>3</sup>H]raclopride.

Data from [30]. Blank boxes indicate not done. QNB: [<sup>3</sup>H]Quinuclidinylbenzilate (cholinergic muscarinic ligand); Racio.: [<sup>3</sup>H]Raclopride; Sch.: [<sup>3</sup>H]Schering 23390;

Spip.: [<sup>3</sup>H]Spiperone.

Now that the various dopamine receptors have been cloned, the D<sub>1</sub>-like group consists of D<sub>1</sub> and D<sub>5</sub>, whereas the D<sub>2</sub>-like group consists of D<sub>2</sub>, D<sub>3</sub> and D<sub>4</sub>. The D<sub>2</sub> receptor has three forms, D<sub>2</sub><sup>Short</sup>, D<sub>2</sub><sup>Long</sup>, and D<sub>2</sub><sup>Longer</sup>.

## 8. Therapeutic concentrations of all antipsychotics occupy 60 – 75% of D<sub>2</sub>

It is now known that therapeutic levels of all the antipsychotic drugs occupy 60 – 80% of brain D<sub>2</sub> receptors, as shown by Farde *et al.* [22] and confirmed by others [23] (see review in [24]). This range of 60 – 80% occupancy of D<sub>2</sub> for the therapeutic effect is not precise and rigid but is numerically derived from various series of patients and from many studies. In general, minimal D<sub>2</sub> occupancy for an antipsychotic effect would be 60 – 65%, although there are examples of patients responding favourably with lower D<sub>2</sub> occupancies (see later). Optimal antipsychotic action may generally require 65 – 70% D<sub>2</sub> occupancy. The side effects of hyperprolactinaemia and Parkinsonism generally arise above 78% and 80 – 82%, respectively, but here too, there are considerable variations from patient to patient. D<sub>2</sub> occupancies with aripiprazole can attain apparent D<sub>2</sub> occupancies exceeding 90%, even in the absence of Parkinsonism. However, in the case of aripiprazole, a partial agonist at D<sub>2</sub>, the high D<sub>2</sub> occupancies may be associated with some internalisation of D<sub>2</sub> into the cytoplasm of the neurons, because dopamine agonists are known to cause D<sub>2</sub> receptor internalisation.

A current active area of clinical research on dopamine receptors is to measure the occupancy of D<sub>2</sub> receptors in both the striatum and outside the striatum in individuals taking antipsychotic medications. Some researchers find that the same D<sub>2</sub> occupancy occurs in both striatal and limbic regions, while others find a higher occupancy in the limbic regions [25,26]. Interestingly, Kessler *et al.* have found that olanzapine-treated patients have a significantly lower D<sub>2</sub> occupancy of 40.2% in the combined region of the substantia nigra and the ventral tegmental area, in contrast to the D<sub>2</sub> occupancy of 59.3% in the striatum of haloperidol-treated patients [27].

Of the three D<sub>2</sub>-like receptors (D<sub>2</sub>, D<sub>3</sub> and D<sub>4</sub>), only the D<sub>2</sub> receptor itself is blocked by antipsychotic drugs in direct

relation to their clinical antipsychotic potencies, as illustrated in Figure 2. No other receptor, including any other dopamine receptor, or any of the serotonin receptors or glutamate receptors, exhibit such a correlation.

Although this well-established finding in Figure 2 may be criticised as simply a relation between the D<sub>2</sub>-blocking concentrations and the clinical doses at which extrapyramidal signs first appear, it is important to note that the absolute concentrations of antipsychotics which block D<sub>2</sub> receptors in the brain are precisely identical to the therapeutic concentrations found in the spinal fluid or plasma water of patients whose psychotic symptoms are successfully controlled by antipsychotics. These two sets of identical concentrations are here presented for the first time in Table 2 and Figure 3, as detailed in the following paragraphs with many references to the literature.

In fact, the therapeutic concentration in the plasma water or the spinal fluid of any antipsychotic can be reasonably predicted from the dissociation constant, or *K* value, at D<sub>2</sub> and by knowing that 60 – 75% of the D<sub>2</sub> receptors need to be occupied for optimum clinical antipsychotic action (D<sub>2</sub> occupancies > 78 or 79% are usually associated with hyperprolactinaemia or Parkinsonian signs [28]).

The predicted therapeutic concentrations of antipsychotics are worked out in Table 2, knowing only the *K* value and the fact that 60 – 75% occupancy is therapeutic. The antipsychotic concentrations found in the plasma water of patients is in good agreement with the predicted concentrations, as shown in Table 2 and in Figure 3.

The relations in Figures 2 and 3 remain a cornerstone of the dopamine hypothesis of psychosis or schizophrenia, still the major contender for an explanatory theory of schizophrenia causation.

The data in Table 2 and Figure 3 have not been corrected for the effect of endogenous dopamine that competes with the antipsychotic for D<sub>2</sub>. Endogenous dopamine acts to raise the antipsychotic dose needed to block D<sub>2</sub>, as detailed elsewhere [30]. It should also be noted that the interaction between the drug, the ligand, the endogenous dopamine, and the receptor, all complicate the interpretation of data for the dissociation constants of the various drugs for the receptor. Nevertheless, despite such

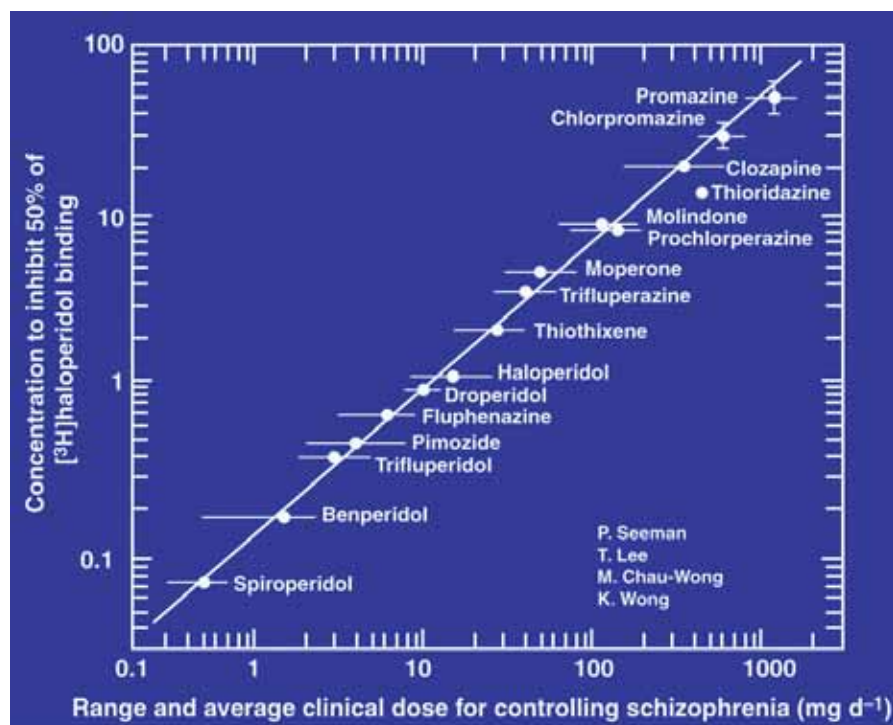


Figure 2. The clinical antipsychotic doses correlate with the concentrations that 50% inhibit the specific binding of [<sup>3</sup>H]haloperidol in homogenised caudate nucleus tissue (calf). These concentrations are similar to those found in the plasma water or spinal fluid in antipsychotic-treated patients.

Redrawn from data in [15,16], with permission.

complications, and despite variations in the *in vitro* composition of buffers and salts between different laboratories, the dissociation constants of various antipsychotics at the D<sub>2</sub> receptor are now in general agreement.

### 9. 'Fast-off-D<sub>2</sub>' theory of atypical antipsychotic action

As noted above, clinically effective doses of antipsychotic drugs occupy between 60 and 75% of brain dopamine D<sub>2</sub> receptors in patients, as measured by positron emission tomography or single-photon emission tomography in the human striatum.

Clozapine and quetiapine, however, have consistently been apparent exceptions. For example, in patients taking therapeutically effective antipsychotic doses of clozapine, the drug only occupies between 0 and ~ 50% of brain dopamine D<sub>2</sub> receptors 6 – 12 h after the oral dose, as measured by a variety of radioligands using either positron tomography or single photon tomography (reviewed in [24,29,30]).

However, the apparently low occupancy of D<sub>2</sub> by clozapine and quetiapine is readily explained by the fact that these two antipsychotics rapidly dissociate from D<sub>2</sub> [24,30]. This also holds for remoxipride and amisulpride (see Table 3 and the reference therein), two atypical drugs not used clinically in

Canada. For example, [<sup>3</sup>H]clozapine, [<sup>3</sup>H]quetiapine, [<sup>3</sup>H]remoxipride and [<sup>3</sup>H]amisulpride dissociate from human cloned D<sub>2</sub> receptors at least one hundred times faster than [<sup>3</sup>H]haloperidol or [<sup>3</sup>H]chlorpromazine.

Using methods similar to those reported earlier for the human cloned dopamine D<sub>2</sub><sup>Long</sup> receptor in CHO cells [24], and using drug concentrations found in the spinal fluid of patients, the times for 50% dissociation from D<sub>2</sub> are in Table 3.

These *in vitro* data in Table 3 match those found clinically for clozapine, quetiapine and haloperidol in schizophrenia patients and volunteers. For example, it has been found by positron emission tomography (PET), using [<sup>11</sup>C]raclopride, that the human brain (striatum) occupancy of D<sub>2</sub> by quetiapine and clozapine rapidly falls off within 24 h, in contrast to that for haloperidol which maintains its D<sub>2</sub> occupancy constant over 24 h. The maximum occupancy of D<sub>2</sub> by either clozapine or quetiapine occurs at 2 h and is in the therapeutic range of 65 – 72% (summarised in [30]). The D<sub>2</sub> occupancy by clozapine falls off by 50% in about 18 h, whereas that for quetiapine falls off by 50% in about 8 h (summarised in [30]).

Thus, the rapid release of clozapine and quetiapine from D<sub>2</sub> and their replacement by endogenous dopamine would readily account for the low D<sub>2</sub> receptor occupancy shown by

**Table 2. Therapeutic concentrations of antipsychotic drugs in the spinal fluid or plasma water.**

Drug	Average therapeutic concentration (ng/ml plasma)	% of drug free in plasma	Therapeutic free concentration in plasma water (nM)	Dissociation constant ( $K_d$ ) in $D_2^{Long}$ (nM) [30]	Concentration (nM) in spinal fluid to occupy 60 – 75% of $D_2 = K_d \times (1.5 - 3)$
Aripiprazole	240 – 440 [37]	~1%	5 – 10	1.8	2.7 – 5.4
Chlorpromazine	22 – 134 [48-50]	2.1% [51,52]	4.6 – 8.8	1.2	1.8 – 3.6
Clozapine	162 – 504 [32,34]	8.2% [33,101]	40 – 126	75	113 – 225
Clozapine-demethyl	90 – 277*	9.7% [101]	28 – 86	180	
Clozapine-like range:			40 – 170†		
Flupentixol-cis	2.2 [53]	35.5% [54]	1.4 – 2.2	0.38	0.6 – 1.1
Haloperidol	**	8.5%**	1 – 3**	0.74	1.1 – 2.2
Olanzapine	23 – 120 [29,34]	7% [31]	5 – 24	7.4	11 – 22
Perphenazine	6.5 nM	9**	0.4 – 0.8	0.27	0.4 – 0.8
Quetiapine	300 – 800 [36]§	17% [35]	130 – 354	140	210 – 420
Raclopride	41 – 100 nM**	4.8%**	2 – 4.8	1.6 – 1.9	2.6 – 5
Remoxipride	3600 nM**	5.5%**	198	67	100 – 201
Risperidone	11 [47]	10% [46]	2.6	1.1	1.7 – 3.3
9-OH-risperidone	24 [47]	22.6% [46]	12¶	1.6	
Thioridazine	**	0.15%**	3.4 – 3.7	1.1	1.7 – 3.3
Trifluoperazine	18 – 97 [55]	1.8% [56]	0.8 – 4	1.4	2.1 – 4.2

The antipsychotic therapeutic concentrations in the plasma water (or spinal fluid) can be predicted from the dissociation constants,  $K_i$  or  $K_d$  (in column 5), and adjusted for the therapeutic  $D_2$  occupancy of 60 – 75% (as shown at the top of column 6). The observed therapeutic concentrations in the plasma water of patients maintained on antipsychotics were derived from the observed values (usually in ng/ml plasma), corrected for % drug bound to plasma proteins, and are shown in column 4.

\*Clozapine-demethyl concentration in plasma is  $55 \pm 7\%$  that of clozapine [38-44], except for the value of 135% [45]. †Net clozapine-like concentration is sum of clozapine plus half that for demethyl-clozapine, because demethyl-clozapine has about half the affinity for  $D_2$ . §At 1 – 2 hour peak. ¶Net risperidone-like activity is sum of risperidone concentration and 9-OH-risperidone, where the latter is reduced by 0.69 because of lower affinity for  $D_2$ . \*\*References in [32].

these atypical antipsychotics, especially because the brain scans are taken 6 – 12 h after the last oral dose in patients on maintenance doses.

It is important to emphasise that the rapid release of clozapine and quetiapine is a molecular event which occurs quickly regardless of the clinical dose used. In other words, even though high doses of clozapine and quetiapine may be used in the patient, these drugs continue to go on and off the  $D_2$  receptor rapidly, allowing extensive and frequent access of endogenous dopamine to the receptor.

Hence, it appears that some antipsychotics such as clozapine and quetiapine occupy  $D_2$  receptors transiently during the day. As just mentioned, PET imaging of patients with schizophrenia reveals that the  $D_2$  receptor occupancies by clozapine and quetiapine wear off quickly after an oral dose, and patients may show no occupancy whatsoever within 48 h of the last dose, in contrast to typical antipsychotics which may continue to occupy  $D_2$  receptors for days. This may explain why psychotic relapses of patients on clozapine and quetiapine occur soon after withdrawal of the antipsychotic (reviewed in [24]), much earlier than after withdrawal of conventional antipsychotic drugs, such as haloperidol or chlorpromazine.

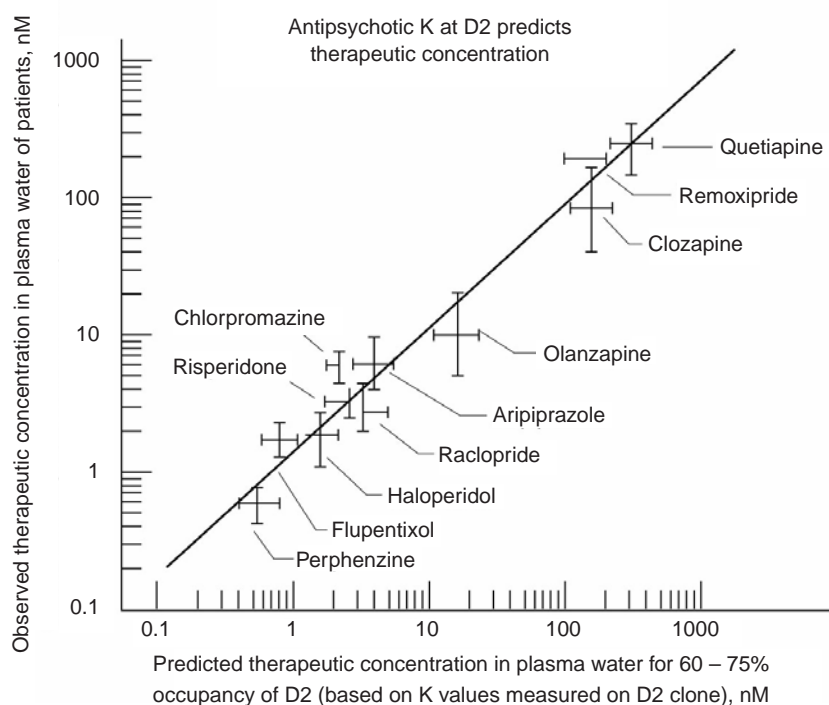
The data for the rapidly dissociating antipsychotics (amoxapine, aripiprazole, clozapine, perlapine, quetiapine, remoxipride and paliperidone) are compatible with the low or absent amount of extrapyramidal signs (EPS) in patients. The extent of risperidone-associated EPS may depend on the proportions of risperidone and its metabolite, paliperidone, in the patient. Olanzapine has a slow off-rate from  $D_2$ , compatible with its dose-dependent incidence of EPS; however, the potent anticholinergic action of olanzapine (its dissociation constant of 2.1 nM matches that of benztropine, Cogentin®, at the muscarinic receptor) provides an effective anti-EPS mechanism.

Atypical antipsychotics, therefore, are helpful to patients by transiently occupying  $D_2$  and then rapidly dissociating to allow dopamine neurotransmission, as illustrated in Figure 4. This keeps prolactin levels normal, spares cognition and obviates EPS.

#### 9.1 Clinical implications of the 'Fast-off- $D_2$ ' theory of atypical antipsychotic action

As outlined above, the 'fast-off- $D_2$ ' theory of atypical antipsychotic action is that the atypicals have low affinities for  $D_2$ , and are loosely bound to, and rapidly released from,  $D_2$ .





**Figure 3.** The observed therapeutic antipsychotic concentrations in the spinal fluid or in the plasma water in treated patients are essentially identical to the predicted antipsychotic concentrations that occupy ~ 60 – 75% of the D<sub>2</sub> receptors *in vitro*. The concentrations in the plasma water were obtained by correcting for the amount bound to the plasma proteins. For example, the concentrations to occupy 75% of D<sub>2</sub> were calculated as being three times higher than the dissociation constant at D<sub>2</sub> (Table 1).

A critical aspect of the theory is that the atypical antipsychotics bind more loosely to D<sub>2</sub> than does dopamine itself, whereas the traditional, typical antipsychotics bind more tightly than dopamine, the dissociation constant for dopamine being 1.7 nM at the functional high-affinity state of D<sub>2</sub>, as listed in Table 3.

Table 3 illustrates a general demarcation between typicals and atypicals. That is, the typical antipsychotics generally have *K* values lower than the *K* for dopamine (at the high-affinity state of the D<sub>2</sub> receptor), whereas the atypicals have *K* values higher than that for dopamine.

Although risperidone appears to be an exception to this generalisation, risperidone is the weakest 'atypical' antipsychotic, eliciting dose-dependent extrapyramidal signs in 60 – 70% of patients taking ≥ 6 mg per day. In the temporal cortex of humans, risperidone remains attached to D<sub>2</sub> for a very long time, requiring at least 60 h for its occupancy to fall by 50% [58].

Clearly, the separation between typicals and atypicals in Table 3 is not sharp and precise because antipsychotic drugs with *K* values between 2 nM and 10 nM, including loxapine, often reveal dose-dependent extrapyramidal signs. Thus, the demarcation between typical and atypical antipsychotics is not a sharp divide but rather a continuous one. Antipsychotics

become increasingly more atypical as their binding to the D<sub>2</sub> receptor becomes more loose and they are released more quickly.

Although almost all atypical antipsychotics have loose binding, with dissociation constants looser than 1.8 nM, they can still elicit dose-dependent Parkinsonism. For example, olanzapine, with a dissociation constant of 7.4 nM, is known to be associated with a dose-dependent incidence of extrapyramidal signs in some patients, especially at higher doses. If the binding is extremely 'loose,' as with clozapine, remoxipride, quetiapine, and melperone, essentially no EPS occurs.

Drugs that are too 'loose' or have far too low an affinity for D<sub>2</sub> receptors cease to exhibit any antipsychotic activity at all. Moreover, although the degree of occupancy of atypicals at D<sub>2</sub> receptors has a direct influence on EPS, the potent anticholinergic action of olanzapine and clozapine provides an additional anti-EPS mechanism. Olanzapine at 20 mg/day, for example, occupies up to 79% of the cholinergic muscarinic receptors in humans [59]. Furthermore, in the case of thioridazine, its anticholinergic properties keep it relatively free of eliciting EPS.

Table 4 summarises a few clinical distinctions between the typical antipsychotics which are tightly bound to D<sub>2</sub> and the



**Table 3. Fast-off-D<sub>2</sub> antipsychotics generally elicit no or low Parkinsonism.**

	Time for 50% offset from D <sub>2</sub> [57]	K <sub>i</sub> or K <sub>d</sub> (nM)	Parkinsonism
Remoxipride (5 nM)	13 s	67	Absent
Clozapine (200 nM)	15 s	75	Absent
Quetiapine (200 nM)	16 s	140	Absent or low
Perlapine (140 nM)	24 s	138	Absent or low
S-(-)-Amisulpride (4 nM)	42 s	1.7*	Absent or low
Aripiprazole (10 nM)	52 s	1.8 <sup>†</sup>	Low
9-Hydroxy-risperidone (2 nM) <sup>§</sup>	60 s	1.6	Low
Amoxapine (40 nM)	66 s	21	Low
Loxapine (20 nM)	16 min	9.2	Dose-dependent
Olanzapine (5 nM)	17 min	7.4	Dose-dependent
Dopamine at D <sub>2</sub> <sup>High</sup> state		1.7	
Raclopride (4 nM)	23 min	1.6	Dose-dependent
Risperidone (2 nM)	27 min	1.09	Dose-dependent
Chlorpromazine (1.5 nM)	30 min	1.2	Common
Haloperidol (2 nM)	38 min	0.4 – 0.74	Common, dose-dependent

The times for 50% offset from the cloned D<sub>2</sub><sup>Long</sup> receptor are shown in column 2. The K<sub>i</sub> or K<sub>d</sub> values are from **Table 1**. The K<sub>i</sub> values are the dissociation constants or inhibition concentrations which inhibited the binding of the [<sup>3</sup>H]ligand by 50%, corrected for the concentration of the [<sup>3</sup>H]ligand; the K<sub>d</sub> values are the dissociation constants obtained by means of saturation data, using the titrated form of the drug, as indicated. The antipsychotics with K values lower than that for dopamine (at D<sub>2</sub><sup>High</sup>) desorb slowly from D<sub>2</sub> and are consistently associated with Parkinsonism. Antipsychotics with K values > 20 nM dissociate quickly and elicit little or no Parkinsonism. Antipsychotics with intermediate values (loxapine and olanzapine) elicit dose-dependent Parkinsonism. Data from [30,57]. \*Amisulpride has low and slow permeation into brain. <sup>†</sup>Aripiprazole is a partial agonist. <sup>§</sup>Paliperidone.

atypical antipsychotics which are loosely bound to D<sub>2</sub>. The required antipsychotic dose (in milligrams) will be low for tightly bound drugs, but high for loosely bound drugs. The typicals, being tightly bound to D<sub>2</sub>, will elicit EPS and elevated prolactin, whereas the atypicals, being loosely bound and rapidly released from D<sub>2</sub>, will not elicit these side effects, or at least to a markedly lesser extent. Finally, because the typicals remain attached to D<sub>2</sub> and readily accumulate in brain tissue, the typicals will eventually lead to tardive dyskinesia. The atypicals, however, are much less fat-soluble, and because they are readily released from D<sub>2</sub> and from the brain tissue, the risk of causing tardive dyskinesia is much reduced or absent.

### 9.2 'Fast-off-D<sub>2</sub>' theory predicts low antipsychotic dose treatment of L-DOPA psychosis

The treatment of patients with psychosis in Parkinson's disease (as a consequence of L-DOPA treatment) is best done with a very loose binding antipsychotic, such as clozapine or quetiapine, to allow dopamine neurotransmission required for motor function to continue.

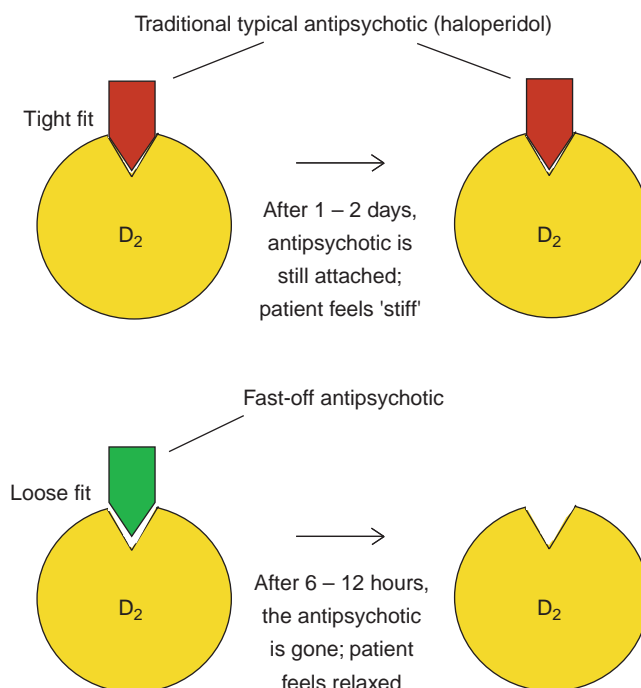
It is well known in neurology that L-DOPA psychosis in a Parkinson's diseased patient is best treated with a dose of clozapine which is ~ 10% the dose normally used for psychosis in schizophrenia. The 'fast-off-D<sub>2</sub>' hypothesis readily and quantitatively predicts this. In Parkinson's disease, where 90 – 95% of the dopamine content is absent, the

synaptic release of dopamine is depressed by over 90%, effectively reducing the amount of endogenous dopamine that would otherwise compete with the administered antipsychotic, as previously analysed [30].

Thus, although a daily dose of 500 mg clozapine might be suitable for treating schizophrenia psychosis, a dose of 50 mg (or less) would be more than adequate to treat L-DOPA psychosis. It is important to note that this analysis [30] holds for competition between endogenous dopamine and a loosely bound antipsychotic. A tightly bound antipsychotic, such as haloperidol, would not readily permit endogenous dopamine to replace it competitively.

### 9.3 'Fast-off-D<sub>2</sub>' theory predicts difference between clozapine and isoclozapine

As reported by Kapur *et al.* [60], the single most powerful predictor of atypicality is the low affinity to, and fast dissociation from, the D<sub>2</sub> receptor, and not high affinity to any other receptor. This hypothesis is supported by their findings that clozapine and isoclozapine have identical potencies on many cloned receptors, including muscarinic M<sub>1</sub>, dopamine D<sub>1</sub>, dopamine D<sub>4</sub>, serotonin-1A, and serotonin-2A receptors, but differ fivefold in their potency only on D<sub>2</sub> receptors. Thus, in several tests of atypicality (early activation of certain genes, catalepsy in animals and prolactin elevation), clozapine behaves like an



**Figure 4.** Antipsychotics that are tightly bound to D<sub>2</sub> markedly reduce dopamine neurotransmission and elicit Parkinsonism and prolactinaemia. Antipsychotics that are loosely bound to D<sub>2</sub> desorb quickly from D<sub>2</sub> and elicit little or no Parkinsonism or prolactinaemia.

**Table 4.** Aspects of 'tight' and 'loose' antipsychotic binding at dopamine D<sub>2</sub> receptors.

	'Tight'	'Loose'
Dose	Low	High
Extrapyramidal signs	Yes	No
Prolactin	High	Normal
Tardive dyskinesia	High risk	Low risk

atypical antipsychotic. Isoclozapine, however, behaves like a conventional antipsychotic.

## 10. Are serotonin receptors a clinical target for atypical antipsychotics?

Although it has often been suggested that the blockade of serotonin-2A receptors may contribute to antipsychotic action and may alleviate the Parkinsonism caused by D<sub>2</sub> blockade [62], the following findings do not support this principle.

### 10.1 Remoxipride does not block 5-HT<sub>2</sub> receptors

Remoxipride is a highly effective atypical antipsychotic drug with no extrapyramidal signs in humans, as well as no catalepsy in rats at doses 30 times higher than the antihyperactivity

dose [61] and no hyperprolactinaemia, yet does not block serotonin-2A receptors [30,32]. For example, the dissociation constant of remoxipride at serotonin-2A receptors is at least 100-fold higher than that at the therapeutic D<sub>2</sub> receptor [30,32], indicating that remoxipride does not occupy serotonin-2A receptors at clinically therapeutic doses. As indicated in Table 3, remoxipride has the highest fast-off-D<sub>2</sub> rate of all antipsychotics.

### 10.2 Amisulpride does not block 5-HT<sub>2</sub> receptors

Amisulpride is a highly effective antipsychotic which is atypical and which does not occupy any serotonin-2A receptors in humans at doses up to 1200 mg per day [63]. However, because amisulpride has a very low permeability across the blood-brain barrier, it must be given at very high doses; 600 – 800 mg/day for antipsychotic action. Therefore, despite the rapid dissociation of 42 seconds, amisulpride from D<sub>2</sub> receptors (Table 3) of the anterior pituitary gland, the pituitary is outside the blood-brain barrier and is fully exposed to the massive concentration of amisulpride in the plasma, thereby resulting in a maintained block of D<sub>2</sub> with hyperprolactinaemia [64]. However, despite the elevated prolactin, amisulpride has no effect on serotonin receptors but provides good antipsychotic action without Parkinsonism. It is the rapid dissociation of 42 seconds (Table 3) which assists in minimising or preventing Parkinsonism, in keeping with the fast-off basis for clozapine and quetiapine.

### 10.3 Serotonin receptor blockade enhances catalepsy

Instead of reducing catalepsy, as predicted by the serotonin-blockade hypothesis, selective serotonin-2A receptor blockade with the drug M100,907 enhances the catalepsy of submaximal doses of the D<sub>2</sub> block by raclopride [65].

### 10.4 Antipsychotic catalepsy doses are not related to D<sub>2</sub>/5-HT<sub>2</sub> ratio

There is no correlation between the antipsychotic catalepsy doses and the ratio of the antipsychotic dissociation constants at D<sub>2</sub> and at serotonin-2A receptors [66,67].

### 10.5 Antipsychotic D<sub>2</sub>/5-HT<sub>2</sub> ratio does not identify atypical antipsychotics

Using the ratio of antipsychotic dissociation constants obtained in this laboratory on human cloned D<sub>2</sub> and serotonin-2A receptors (Table 1 and [30]), there is no obvious demarcation between typical and atypical antipsychotics.

### 10.6 Serotonin receptor block does not alleviate extrapyramidal signs

A high degree of serotonin-2A receptor occupancy (95%) by risperidone (6 mg/day) does not prevent EPS in a series of patients [68]. Moreover, although loxapine and olanzapine have 5-HT<sub>2A</sub>/D<sub>2</sub> receptor affinity ratios similar to that of clozapine, Kalkman *et al.* found that clozapine inhibited the cataleptic response to loxapine or olanzapine. These results provide a strong argument against the idea that 5-HT<sub>2A</sub> blockade is the explanation for the low EPS profile of clozapine [69].

### 10.7 Serotonin receptor block does not alter threshold for D<sub>2</sub> block

Using [<sup>11</sup>C]raclopride for imaging brain D<sub>2</sub> receptors and [<sup>11</sup>C]setoperone for imaging brain serotonin-2A receptors, Kapur *et al.* [23] have found that the high occupancy of serotonin-2A receptors by olanzapine or by risperidone did not alter the D<sub>2</sub> occupancy required for the antipsychotic effect or the D<sub>2</sub> occupancy at which extrapyramidal signs occur. The threshold doses for antipsychotic action consistently occupy 65% of brain D<sub>2</sub> receptors in patients, and the threshold doses for extrapyramidal signs consistently occupy 80% of brain D<sub>2</sub> receptors in patients, whether or not the serotonin-2A receptors were occupied.

It is not clear, therefore, what clinical benefit, if any, is provided by the blockade of serotonin receptors. Although low doses of cyproheptadine have been used [70] to block serotonin-2A receptors and supplement antipsychotic administration, it should be noted that cyproheptadine has a D<sub>2</sub> blocking action. It has a *K* value of 24 nM at D<sub>2</sub> receptors, compared with 75 nM for clozapine and 21 nM for amoxapine (Table 1). Amoxapine, although marketed as an antidepressant, has antipsychotic properties.

### 10.8 Chlorpromazine blocks 5-HT<sub>2</sub> receptors but elicits Parkinsonism

Chlorpromazine, the first typical antipsychotic, at 500 mg/day blocks 65% of serotonin-2A receptors. This 'high level of serotonin-2A block suggests that the distinct clinical profiles of chlorpromazine and clozapine are unrelated to serotonin-2A receptor blockade' [63].

### 10.9 Block of serotonin receptors not needed for antipsychotic action

It has been reported that the block of serotonin-2A receptors 'is not a prerequisite for the antipsychotic effect' [68,69]. In fact, full block of serotonin-2A receptors occurs at subtherapeutic doses of risperidone, olanzapine and clozapine, indicating that serotonin-2A blockade has little or no antipsychotic action (but see [70]).

### 13.10 Agonism of serotonin-1A receptors reduces catalepsy

Although it has long been known that the stimulation of serotonin-1A receptors can alleviate catalepsy in animals caused by D<sub>2</sub> blockade [71,72], only recently have drugs been found which simultaneously block D<sub>2</sub>, but stimulate serotonin-1A receptors [73].

## 14. Additional evidence for D<sub>2</sub> as the therapeutic target for antipsychotics

The D<sub>2</sub> receptor continues to be the main therapeutic target for antipsychotic action, whether it is targeted by antagonists or partial agonists, such as aripiprazole (Figure 2). In fact, as stated by Su *et al.* [74], 'no drug has yet been identified with antipsychotic action without a significant affinity for the D<sub>2</sub> receptor'.

In addition, over and above the pharmacological role of D<sub>2</sub> as an antipsychotic target, there are findings indicating that various aspects of D<sub>2</sub> contribute to psychosis in general, or schizophrenia, in particular.

For example, Hirvonen *et al.* [75] have found that the number of D<sub>2</sub> receptors is significantly elevated in healthy identical co-twins of individuals with schizophrenia. These findings suggest that an elevation of D<sub>2</sub> may be a necessary requirement for schizophrenia, but may not be sufficient to elicit the syndrome.

Furthermore, the D<sub>2</sub> receptor has a polymorphism at position 311 where the serine is replaced by cysteine. In schizophrenia, it has been found in 27 samples, comprising 3707 patients and 5363 control subjects, that the serine311cysteine polymorphism was significantly associated with schizophrenia [76].

Although measurements on nonmedicated schizophrenia patients with [<sup>11</sup>C]methylspiperone revealed elevations in striatal D<sub>2</sub> density, whereas measurements with [<sup>11</sup>C]raclopride did not show such elevations (reviewed in [30]), the number of D<sub>2</sub> receptors in the caudate-putamen is significantly elevated in untreated schizophrenia [77].

## 15. No evidence for D<sub>3</sub> as an antipsychotic target

A detailed analysis along the lines shown in Table 2 and Figure 2, using the *K* values for D<sub>3</sub> in Table 1, shows that D<sub>3</sub> is occupied by some, but not all antipsychotics, as is the case with D<sub>2</sub>. This analysis, for example, readily shows that D<sub>3</sub> is not extensively occupied at clinical concentrations of remoxipride, clozapine, spiperone, quetiapine, molindone, melperone and haloperidol [67].

Although BP 897 is a partial agonist at D<sub>3</sub> with a selectivity for D<sub>3</sub> of ~ 100-fold higher than that for D<sub>2</sub>, BP 897 (at 10 mg/day) did not appear effective against schizophrenia symptoms [78]. Other drugs moderately selective for D<sub>3</sub>, such as S33138 and A437,203, are currently being tested on schizophrenia patients. The highly D<sub>3</sub>-selective drug, FAUC 365, has not yet been tested in this disease. It is possible that the D<sub>3</sub>-selective compounds may be helpful in treating drug abuse [79-82].

## 16. No evidence for D<sub>4</sub> as an antipsychotic target

Interestingly, clozapine has a higher affinity at D<sub>4</sub> than at D<sub>2</sub>, as shown in Table 1. Nevertheless, despite the selectivity of clozapine for D<sub>4</sub>, clozapine occupies the necessary 60 – 70% occupancy of brain D<sub>2</sub> receptors at clinical doses (~ 400 mg/day), compatible with the idea that D<sub>2</sub> is the therapeutic target for clozapine, as with all the other antipsychotics. It may be noted that iso-clozapine causes catalepsy, in contrast to clozapine, which does not elicit catalepsy. Both drugs have identical affinity for D<sub>4</sub>, but isoclozapine has higher affinity for D<sub>2</sub> (see Table 2) and, therefore, causes catalepsy.

Although the gene expression of D<sub>4</sub> was found to be elevated in the frontal cortex of schizophrenia tissues, selective D<sub>4</sub> antagonists, such as sonepiprazole and L-745,870, did not have any antipsychotic action.

## 17. D<sub>2</sub><sup>High</sup> as the antipsychotic target

The D<sub>2</sub> receptor, similar to many other G-protein-linked receptors, has a state of high affinity and a state of low affinity for dopamine. The binding of dopamine to D<sub>2</sub> occurs in two concentration ranges. Low nanomolar concentrations of dopamine bind to the high-affinity state of the receptor (D<sub>2</sub><sup>High</sup>) whereas high micromolar concentrations bind to the low-affinity state of the receptor (D<sub>2</sub><sup>Low</sup>).

D<sub>2</sub><sup>High</sup> is the functional state in the anterior pituitary, on which dopamine and other dopamine-like drugs (bromocriptine) act to inhibit the release of prolactin. D<sub>2</sub><sup>High</sup> is presumably also the primary functional form of D<sub>2</sub> in the nervous system, although little work has been done to prove this point.

Although it has been reported that 90% of the D<sub>2</sub> receptors in brain slices are in the D<sub>2</sub><sup>High</sup> state, the proportion of D<sub>2</sub> receptors in the high-affinity state in homogenised striatum

*in vitro* is generally between 15% and 20%. The D<sub>2</sub><sup>High</sup> state quickly converts in a matter of seconds or minutes into the D<sub>2</sub><sup>Low</sup> state by guanine nucleotide.

Extensive reviews by Lieberman *et al.* [83], and by Curran *et al.* [84] show that up to 70% of schizophrenia patients are supersensitive to either methylphenidate or amphetamine at doses which do not affect control humans. These findings are in agreement with the dopamine hypothesis of psychosis or schizophrenia first outlined by J Van Rossum in 1967 [85].

'The hypothesis that neuroleptic drugs may act by blocking dopamine receptors in the brain has been substantiated by preliminary experiments with a few selective and potent neuroleptic drugs. There is an urgent need for a simple isolated tissue that selectively responds to dopamine so that less specific neuroleptic drugs can also be studied and the hypothesis further tested.... When the hypothesis of dopamine blockade by neuroleptic agents can be further substantiated it may have fargoeing consequences for the pathophysiology of schizophrenia. Overstimulation of dopamine receptors could then be part of the aetiology.'

A wide variety of brain alterations in animals (lesions, birth injury by C-section and anoxia, amphetamine or phencyclidine treatment, knockouts of a variety of receptors) all lead to the final common finding of behavioural dopamine supersensitivity and elevated proportions of D<sub>2</sub> receptors in the D<sub>2</sub><sup>High</sup> state in the striatum [86].

For example, repeated administration of amphetamine to animals or humans leads to behavioural dopamine supersensitivity. Although the density of D<sub>2</sub> receptors in the striatum does not change in such animals, it is remarkable that the density of D<sub>2</sub><sup>High</sup> receptors increases dramatically by several-fold [86].

A similar situation occurs in animals that receive hippocampal lesions neonatally. Such animals as adults reveal behavioural dopamine supersensitivity, and the striatum contains a marked increase in the proportion of D<sub>2</sub> receptors in the high-affinity state.

All dopamine-supersensitive animals examined to date reveal elevated D<sub>2</sub><sup>High</sup> receptors. Because antipsychotics such as haloperidol successfully block the symptoms of psychosis found in amphetamine psychosis, phencyclidine psychosis, and brain damage psychosis in humans, and because these conditions in rats are associated with markedly elevated D<sub>2</sub><sup>High</sup> states, it is reasonable to conclude that the main target of antipsychotic action is not only D<sub>2</sub>, but the high-affinity state of D<sub>2</sub>.

Therefore, the molecular control of the high-affinity state of D<sub>2</sub> is emerging as a central problem in this field. At present, there is uncertainty as to whether this high-affinity state of D<sub>2</sub> is controlled through G<sub>o</sub> or one of the G<sub>i</sub> proteins, because this varies from cell to cell.

## 18. Non-dopamine receptor targets

Although the major emphasis of this review has focused on the dopamine D<sub>2</sub> receptor as the main therapeutic target in

treating psychosis in general and schizophrenia in particular, there is a constant search for new targets that may either contribute to the therapeutic action or that may alleviate some of the side effects of  $D_2$  blockade.

For example, a major difficulty emerging with the more recently developed antipsychotics, the so-called atypical antipsychotics such as olanzapine, quetiapine or clozapine, is that they lead to considerable weight gain with a significant risk for diabetes. Although the cause of such weight gain is not known, it is likely related to the blockade of histamine  $H_1$  receptors [87]. Olanzapine, for example, has a potent dissociation constant of 0.087 nM at the histamine  $H_1$  receptor, in contrast to its dissociation constant of 7 – 20 nM at the therapeutic  $D_2$  receptor [30,87]. Currently, there is active research on developing  $D_2$  antagonists that have less affinity for the histamine  $H_1$  receptor in order to minimise the risk of weight gain [88].

Although serotonin-2A receptors do not appear to contribute to either the therapeutic aspect of antipsychotics or significantly alter the side effect profile, as discussed above, there are now at least fourteen types of serotonin receptors that have been identified. Some of these, such as the serotonin-6 and serotonin-7 receptors [90,91], have been suggested to be targets for some of the newer antipsychotics. There is no consensus on this matter, however. Clozapine, for example, has a high affinity for the serotonin-6 and serotonin-7 receptors, with a dissociation constant of 4 – 8 nM at both receptors [89,90], in contrast to the necessary therapeutic concentration of 113 – 225 nM in the patient's spinal fluid or free in the plasma water (Table 2). Clozapine, in other words, has very high affinity for the serotonin-6 and -7 receptors, but the extremely high occupation of these receptors does not appear to be relevant to the therapeutic antipsychotic action. The serotonin-6 and -7 receptors are also unlikely to be a common target for antipsychotics in general because older antipsychotics have very high dissociation constants, such as > 5000 nM for haloperidol, 1250 nM for melperone, 1595 nM for spiperone, and > 5000 nM for trifluoperidol [89].

## 19. Expert opinion and future outlook

Of the five dopamine receptors and their variants, the  $D_2$  receptor and its properties continue to be the most actively investigated because  $D_2$  is the main clinical target for antipsychotics and for the dopamine agonist treatment of Parkinson's disease. The  $D_1$  receptor, however, also has an important clinical role in treating Parkinson's disease because the stimulation

of  $D_1$  synergises with the stimulation of  $D_2$ , possibly via  $D_1/D_2$  heterodimers or cell-cell interactions.

Probably the most central question in determining the basis of psychosis or schizophrenia is to determine the molecular basis of dopamine supersensitivity and to determine which proteins or genes regulate the maintenance of  $D_2$  receptors in their high-affinity state. The discovery of such genes or proteins which foster the conversion of  $D_2$  into more high-affinity states will provide new and possibly more accurate targets for new antipsychotics.

The clinician can apply some of the principles in this review to the treatment of individual patients. Atypical agents, being newer and still protected by patent, are much more expensive than the older drugs. Because they do not elicit EPS and do not elevate prolactin levels does not mean that they are free of serious side effects. One could argue that the side effects associated with some of the atypical drugs (agranulocytosis, obesity, diabetes, ophthalmological problems, cardiovascular problems, sexual problems, obsessive-compulsive symptoms, convulsions, insomnia) are more serious than EPS, high prolactin and even tardive dyskinesia. Low dose, extended dosing regimens of typical drugs may be best suited for a specific patient. Patients known to be non-adherent to regular medication may do better on those drugs that are more tightly bound to the  $D_2$  receptor, where risk of relapse through a short period of non-compliance is a lesser risk.

On the other hand, patients with a history of neuroleptic malignant syndrome or tardive dyskinesia are best treated with drugs that are readily displaced so that, should the syndrome return, the drug is quickly out of their brain. In psychotic patients, high stress levels, accompanied by high endogenous dopamine release, will necessitate higher doses of antipsychotic drug. Periods of low stress will require lower doses. Psychotic patients who may temporarily benefit from high prolactin levels (those who do not want to conceive or, conversely, postpartum women whose milk is insufficient for breastfeeding) may preferentially be prescribed typical antipsychotics. Those with beginning signs of tardive dyskinesia, on the other hand, should be discontinued from the typical antipsychotics and prescribed the newer drugs instead. Knowing how drugs work greatly expands the clinician's repertoire of strategies, allowing optimisation of drug regimens for individualised treatment.

Finally, in order to diagnose and categorise early stages of psychosis, as well as to follow and treat the course of the illness, it is now possible to image the  $D_2^{\text{High}}$  states directly in patients [91,92].

## Bibliography

- SIGWALD J, BOUTTIER D: 3-Chloro-10-(3'-dimethylaminopropyl)-phenothiazine hydrochloride in current neuro-psychiatry. *Ann. Méd. Interne (Paris)* (1953) **54**(2):150-182.
- DELAY J, DENIKER P, HARL J-M.: Traitement des états d'excitation et d'agitation par une méthode médicamenteuse dérivée de l'hibernothérapie. [Therapeutic method derived from hiberno-therapy in excitation and agitation states.] *Ann. Méd-Psychol. (Paris)* (1952) **110**:267-273.
- AGID O, KAPUR S, ARENOVICH T, ZIPURSKY RB: Delayed-onset hypothesis of antipsychotic action: a hypothesis tested and rejected. *Arch Gen Psychiat* (2003) **60**:1228-1235.
- SEEMAN P: The membrane actions of anesthetics and tranquilizers. *Pharmacol. Rev.* (1972) **24**:583-655.
- SEEMAN P, BIALY HS: The surface activity of tranquilizers. *Biochem. Pharmacol.* (1963) **12**:1181-1191.
- EHRINGER H, HORNYKIEWICZ O: Distribution of noradrenaline and dopamine (3-hydroxytyramine) in the human brain and their behavior in diseases of the extrapyramidal system. *Klin. Wochenschr.* (1960) **38**:1236-1239.
- ZINGALES IA: A gas chromatographic method for the determination of haloperidol in human plasma. *J. Chromatogr.* (1971) **54**:15-24.
- HEISS WD, HOYER J: Dopamine receptor blockade by neuroleptic drugs in *Aplysia* neurones. *Experientia* (1974) **30**:1318-1320.
- STRUYKER BOUDIER HAJ, GIELEN W, COOLS AR, VAN ROSSUM JM: Pharmacological analysis of dopamine-induced inhibition and excitation of neurones of the snail *Helix Aspersa*. *Arch. Int. Pharmacodyn. Ther.* (1974) **209**:324-331.
- CARLSSON A, LINDQVIST M: Effect of chlorpromazine or haloperidol on formation of 3-methoxytyramine and normetanephrine in mouse brain. *Acta Pharmacol. Toxicol. (Copenh.)* (1963) **20**:140-144.
- ANDÉN N-E, ROOS B-E, WERDINIUS B: Effects of chlorpromazine, haloperidol and reserpine on the levels of phenolic acids in rabbit corpus striatum. *Life Sci.* (1964) **3**:149-158.
- ANDÉN N-E, BUTCHER SG, CORRODI H, FUXE K, UNGERSTEDT U: Receptor activity and turnover of dopamine and noradrenaline after neuroleptics. *Eur. J. Pharmacol.* (1970) **11**:303-314.
- KEBABIAN JW, GREENGARD P: Dopamine-sensitive adenyl cyclase: possible role in synaptic transmission. *Science* (1971) **174**:1346-1349.
- SEEMAN P: Dopamine receptors and the dopamine hypothesis of schizophrenia. *Synapse* (1987) **1**:133-152.
- SEEMAN P, CHAU-WONG M, TEDESCO J, WONG K: Brain receptors for antipsychotic drugs and dopamine: direct binding assays. *Proc. Natl. Acad. Sci. USA* (1975) **72**:4376-4380.
- SEEMAN P, LEE T, CHAU-WONG M, WONG K: Antipsychotic drug doses and neuroleptic/dopamine receptors. *Nature (Lond)* (1976) **261**:717-719.
- KEBABIAN JW, CALNE DB: Multiple receptors for dopamine. *Nature* (1979) **277**:93-96.
- BURT DR, CREESE I, SNYDER SH: Properties of [<sup>3</sup>H]haloperidol and [<sup>3</sup>H]dopamine binding associated with dopamine receptors in calf brain membranes. *Mol. Pharmacol.* (1976) **12**:800-812.
- SNYDER SH, CREESE I, BURT DR: The brain's dopamine receptor: labeling with [<sup>3</sup>H]dopamine and [<sup>3</sup>H]haloperidol. *Psychopharmacol. Commun.* (1975) **1**:663-673.
- SEEMAN P: Brain dopamine receptors. *Pharmacol. Rev.* (1980) **32**:229-313.
- SOKOLOFF P, GIROS B, MARTRES MP, BOUTHENET ML, SCHWARTZ JC: Molecular cloning and characterization of a novel dopamine receptor (D<sub>3</sub>) as a target for neuroleptics. *Nature* (1990) **347**:146-151.
- FARDE L, NORDSTROM AL, WIESEL FA *et al.*: Positron emission tomographic analysis of central D<sub>1</sub> and D<sub>2</sub> dopamine receptor occupancy in patients treated with classical neuroleptics and clozapine. Relation to extrapyramidal side effects. *Arch. Gen. Psychiat.* (1992) **49**:538-544.
- KAPUR S, ZIPURSKY RB, REMINGTON G: Comparison of the 5-HT<sub>2</sub> and D<sub>2</sub> receptor occupancy of clozapine, risperidone, and olanzapine in schizophrenia: clinical and theoretical implications. *Am. J. Psychiatry* (1999) **156**:286-293.
- SEEMAN P, TALLERICO T: Rapid release of antipsychotic drugs from dopamine D<sub>2</sub> receptors: an explanation for low receptor occupancy and early clinical relapse upon drug withdrawal of clozapine or quetiapine. *Am. J. Psychiatry* (1999) **156**:876-884.
- STEPHENSON CME, BIGLIANI V, JONES HM *et al.*: Striatal and extra-striatal D<sub>2</sub>/D<sub>3</sub> dopamine receptor occupancy by quetiapine *in vivo*. *Br. J. Psychiat.* (2000) **177**:408-415.
- BRESSAN RA, ERLANDSSON K, JONES HM *et al.*: Is regionally selective D<sub>2</sub>/D<sub>3</sub> dopamine occupancy sufficient for atypical antipsychotic effect? An *in vivo* quantitative [<sup>123</sup>I]epidepride SPET study of amisulpride-treated patients. *Amer. J. Psychiat.* (2003) **160**:1413-1420.
- KESSLER RM, ANSARI MS, RICCARDI P *et al.*: Occupancy of striatal and extrastriatal dopamine D<sub>2</sub>/D<sub>3</sub> receptors by olanzapine and haloperidol. *Neuropsychopharmacology* (2005) **30**:2283-2289.
- TURRONE P, KAPUR S, SEEMAN MV, FLINT AJ: Elevation of prolactin levels by atypical antipsychotics. *Am. J. Psychiatry* (2002) **159**:133-135.
- MAURI MC, STEINHILBER CP, MARINO R *et al.*: Clinical outcome and olanzapine plasma levels in acute schizophrenia. *Eur. Psychiatry* (2005) **20**:55-60.
- SEEMAN P: Atypical antipsychotics: mechanism of action. *Can. J. Psychiatry* (2002) **47**:27-38.
- KASSAHUN K, MATTIUZ E, NYHART JR E *et al.*: Disposition and biotransformation of the antipsychotic agent olanzapine in humans. *Drug Metabol. Dispos.* (1997) **25**:81-93.
- SEEMAN P: Dopamine receptor sequences. Therapeutic levels of neuroleptics occupy D<sub>2</sub> receptors, clozapine occupies D<sub>4</sub>. *Neuropsychopharmacology* (1992) **7**:261-284.
- SCHABER G, STEVENS I, GAERTNER HJ, DIETZ K, BREYER-PFAFF U: Pharmacokinetics of clozapine and its metabolites in psychiatric patient: plasma protein binding and renal clearance. *Br. J. Clin. Pharmacol.* (1998) **46**:453-459.

34. PERRY PJ, LUND BC, SANGER T, BEASLEY C: Olanzapine plasma concentrations and clinical response: Acute phase results of the North American olanzapine trial. *J. Clin. Psychopharmacol.* (2001) **21**:14-20.
35. AICHORN W, MARSTEINER J, WALCH T, ZERNIG G, SARIA A, KEMMLER G: Influence of age, gender, body weight and valproate comedication on quetiapine plasma concentrations. *Int. Clin. Psychopharmacol.* (2006) **21**:81-85.
36. MCCONVILLE BJ, ARVANITIS LA, THYRUM PT *et al.*: Pharmacokinetics, tolerability, and clinical effectiveness of quetiapine fumarate: An open-label trial in adolescents with psychotic disorders. *J. Clin. Psychiat.* (2000) **61**:252-260.
37. MALLIKAARJUN S, SALAZAR DE, BRAMER SL: Pharmacokinetics, tolerability, and safety of aripiprazole following multiple oral dosing in normal healthy volunteers. *J. Clin. Pharmacol.* (2004) **44**:1179-1187.
38. LIN S-K, CHANG W-H, CHUNG M-C, LAM YWE, JANN MW: Disposition of clozapine and demethylclozapine in schizophrenic patients. *J. Clin. Pharmacol.* (1994) **34**:318-324.
39. LIEBERMAN J, JOHNS C, POLLACK S *et al.*: Biochemical effects of clozapine in cerebrospinal fluid of patients with schizophrenia. In: *Advances in Neuropsychiatry and Psychopharmacology* volume I: Schizophrenia Research, CA Tamminga and SC Schulz (Eds), Raven Press, New York, USA (1991):341-349.
40. NORDIN C, ALMÉ B, BONDESSON U: CSF and serum concentrations of clozapine and its demethyl metabolite: a pilot study. *Psychopharmacology* (1995) **122**:104-107.
41. HASEGAWA M, GUTERREZ-ESTEINOU R, WAY L, MELTZER HY: Relationship between clinical efficacy and clozapine concentrations in plasma in schizophrenia: Effect of smoking. *J. Clin. Psychopharmacol.* (1993) **13**:383-390.
42. OLESEN OV, THOMSEN K, JENSEN PN *et al.*: Clozapine serum levels and side effects during steady state treatment of schizophrenic patients; a cross-sectional study. *Psychopharmacology* (1995) **117**:371-378.
43. PERRY PJ, MILLER DD, ARNDT SV, CADORET RJ: Clozapine and norclozapine plasma concentrations and clinical response of treatment-refractory schizophrenic patients. *Am. J. Psychiat.* (1991) **148**:231-235.
44. JANN MW, LIU H-C, WEIN F-C *et al.*: Gender differences in plasma clozapine levels and its metabolites in schizophrenic patients. *Hum. Psychopharmacol.* (1997) **12**:489-495.
45. OLESEN OV, POULSEN B: On-line fully automated determination of clozapine and desmethylclozapine in human serum by solid-phase extraction on exchangeable cartridges and liquid chromatography using a methanol buffer mobile phase on unmodified silica. *J. Chromatog.* (1993) **622**:39-46.
46. MANNENS G, MEULDERMANS W, SNOECK E, HEYKANTS J: Plasma protein binding of risperidone and its distribution in blood. *Psychopharmacology* (1994) **114**:566-572.
47. BALANT-GORGIA AE, GEX-FABRY M, GENET C, BALANT LP: Therapeutic drug monitoring of risperidone using a new, rapid HPLC method: reappraisal of interindividual variability factors. *Ther. Drug Monit.* (1999) **21**:105-115.
48. MACKAY AVF, HEALEY AF, BAKER J: The relationship of plasma chlorpromazine to its 7-hydroxy and sulfoxide metabolites in a large population of chronic schizophrenics. *Br. J. Clin. Pharmacol.* (1974) **1**:425-430.
49. RIVERA-CALIMLIM L, GIFT T, NASRALLAH HA, WYATT RJ, LASAGNA L: Correlation between plasma concentrations of chlorpromazine and clinical response. *Commun. Psychopharmacol.* (1978) **2**:215-222.
50. TOKUNAGA H, KUDO K, IMAMURA T, JITSUFUCHI N, OHTSUKA Y, IKEDA N: Plasma concentrations of antipsychotic drugs in psychiatric inpatients. *Nippon Hoigaku Zasshi* (1997) **51**:417-422.
51. WODE-HELGOEDT B, ALFREDSSON G: Concentrations of chlorpromazine and two of its active metabolites in plasma and cerebrospinal fluid of psychotic patients treated with fixed doses. *Psychopharmacology* (1981) **73**:55-62.
52. ALFREDSON G, SEDVALL G: Protein binding of chlorpromazine in cerebrospinal fluid and serum. *Int. Pharmacopsychiatry* (1980) **15**:261-269.
53. JØRGENSEN A, ANDERSEN J, BJØRNDAL N, DENCKER SJ, LUNDIN L, MALM U: Serum concentrations of cis(Z)flupentixol and prolactin in chronic schizophrenic patients treated with flupentixol and cis(Z)-flupentixol decanoate. *Psychopharmacology* (1982) **77**:58-85.
54. JØRGENSEN A, GOTTFRIES CG: Pharmacokinetic studies on flupentixol and flupentixol decanoate in man using tritium labeled compounds. *Psychopharmacologia* (1972) **27**:1-10.
55. HARRIS PQ, BROWN SJ, FRIEDMAN MJ, BACOPOULOS NG: Plasma drug and homovanillic acid levels in psychotic patients receiving neuroleptics. *Biol. Psychiat.* (1984) **19**:849-860.
56. VERBEECK RK, CARDINAL J-A, HILL AG, MIDHA KK: Binding of phenothiazine neuroleptics to plasma proteins. *Biochem. Pharmacol.* (1983) **32**:2565-2570.
57. SEEMAN P: An update of fast-off dopamine D<sub>2</sub> atypical antipsychotics. *Am. J. Psychiat.* (2005) **162**:1984-1985.
58. TAKANO A, SUHARA T, IKOMA Y *et al.*: Estimation of the time-course of dopamine D<sub>2</sub> receptor occupancy in living human brain from plasma pharmacokinetics of antipsychotics. *Int. J. Neuropsychopharmacol.* (2004) **7**:19-26.
59. RAEDLER TJ, KNABLE MB, JONES DW *et al.*: *In vivo* olanzapine occupancy of muscarinic acetylcholine receptors in patients with schizophrenia. *Neuropsychopharmacology* (2000) **23**:56-68.
60. KAPUR S, SEEMAN P, ZIPURSKY R, REMINGTON GJ: Fast dissociation from the dopamine D<sub>2</sub> receptor (not high affinity at multiple receptors) is the key to 'atypical' antipsychotics. *Schizophrenia Res.* (2001) **49**(1-2 Suppl.):92.
61. ÖGREN, SO, FLORVALL L, HALL H, MAGNUSSON O, ÄNGEBY-MÖLLER K: Neuropharmacological and behavioural properties of remoxipride in the rat. *Acta Psychiat. Scand.* (1990) **82**(Suppl. 358):21-26.
62. MELTZER HY, NASH JF: Effects of antipsychotic drugs on serotonin receptors. *Pharmacol. Rev.* (1991) **43**:587-604.



63. TRICHARD C, PAILLERE-MARTINOT ML, ATTAR-LEVY D, RECASSENS C, MONNET F, MARTINOT JL: Binding of antipsychotic drugs to cortical 5-HT<sub>2A</sub> receptors: a PET study of chlorpromazine, clozapine, and amisulpride in schizophrenic patients. *Am. J. Psychiatry* (1998) **155**:505-508.
64. KAPUR S, LANGLOIS X, VINKEN P, MEGENS AA, DE COSTER R, ANDEWS JS: The differential effects of atypical antipsychotics on prolactin elevation are explained by their differential blood-brain disposition: a pharmacological analysis in rats. *J. Pharmacol. Exp. Ther.* (2002) **302**:1129-1134.
65. WADENBERG M-L G, BROWNING JL, YOUNG KA, HICKS PB: Antagonism at 5-HT<sub>2A</sub> receptors potentiates the effect of haloperidol in a conditioned avoidance response task in rats. *Pharmacol. Biochem. Behav.* (2001) **68**:363-370.
66. SEEMAN P, CORBETT R, VAN TOL HHM: Atypical neuroleptics have low affinity for dopamine D<sub>2</sub> receptors or are selective for D<sub>4</sub>. *Neuropsychopharmacology* (1997) **16**:93-110.
67. SEEMAN P, KAPUR S: Olanzapine binding to dopamine receptors *in vitro* and *in vivo*. In: *Olanzapine (Zyprexa) – A Novel Antipsychotic*. Tran PV, Bymaster F, Tye N, Herrera J, Breier A, Tollefson G (Eds), Eli Lilly & Co., Lippincott Williams & Wilkins, Philadelphia, USA (2000):3-24.
68. NYBERG S, NAKASHIMA Y, NORDSTROM A-L, HALLDIN, FARDE L: Positron emission tomography of *in-vivo* binding characteristics of atypical antipsychotic drugs. Review of D<sub>2</sub> and 5-HT<sub>2</sub> receptor occupancy studies and clinical response. *Br. J. Psychiat.* (1996) **168**:40-44.
69. KALKMAN HO, NEUMANN V, NOZZULAK J, TRICKLEBANK MD: Cataleptogenic effect of subtype selective 5-HT receptor antagonists in the rat. *Eur. J. Pharmacol.* (1998) **343**:201-207.
70. MELTZER HY, LEE MA, RANJAN R, MASON EA, COLA PA: Relapse following clozapine withdrawal: effect of neuroleptic drugs and cyproheptadine. *Psychopharmacology (Berl)* (1996) **124**:176-187.
71. INVERNIZZI RW, CERVO L, SAMANI R: 8-Hydroxy-2-(Di-*N*-propylamino)tetralin, a selective serotonin-1A receptor agonist, blocks haloperidol-induced catalepsy by an action on raphe nuclei medianus and dorsalis. *Neuropharmacology* (1988) **27**:515-518.
72. WADENBERG M-L: Antagonism by 8-OH-DPAT, but not ritanserin, of catalepsy induced by SCH23390 in the rat. *J. Neural Transm.* (1992) **89**:4-9.
73. KLEVEN MS, BARRET-GREVOZ C, SLOT LB, NEWMAN-TANCREDI A: Novel antipsychotic agents with 5-HT(1A) agonist properties: role of 5-HT(1A) receptor activation in attenuation of catalepsy induction in rats. *Neuropharmacology* (2005) **49**:135-143.
74. SU T-P, MALHOTRA AK, HADD K, BREIER A, PICKAR D: D<sub>2</sub> dopamine receptor occupancy: A crossover comparison of risperidone with clozapine therapy in schizophrenic patients. *Arch. Gen. Psychiat.* (1997) **54**:972-973.
75. HIRVONEN J, VAN ERP TG, HUTTUNEN J *et al.*: Increased caudate dopamine D<sub>2</sub> receptor availability as a genetic marker for schizophrenia. *Arch. Gen. Psychiat.* (2005) **62**:371-378.
76. GLATT SJ, JONSSON EG: The Cys allele of the DRD2 Ser311Cys polymorphism has a dominant effect on risk for schizophrenia: evidence from fixed- and random-effects meta-analyses. *Am. J. Med. Genet. B Neuropsychiatr. Genet.* (2006) **141**:149-154.
77. CORRIPIO I, PEREZ V, CATAFAU AM, MENA E, CARRIO I, ALVAREZ E: Striatal D<sub>2</sub> receptor binding as a marker of prognosis and outcome in untreated first-episode psychosis. *Neuroimage* (2006) **29**:662-666.
78. LECRUBIER Y: A partial D<sub>3</sub> receptor agonist in schizophrenia. *Eur. Neuropsychopharmacol.* (2003) **13**:(Suppl. 4):S167-S168.
79. PILLA M, PERACHON S, SAUTEL F *et al.*: Selective inhibition of cocaine-seeking behaviour by a partial dopamine D<sub>3</sub> receptor agonist. *Nature* (1999) **400**:371-375.
80. SOKOLOFF P, LE FOLL B, PERACHON S, BORDET R, RIDRAV S, SCHWARTZ JC: The dopamine D<sub>3</sub> receptor and drug addiction. *Neurotox. Res.* (2001) **3**:433-441.
81. JOYCE JN, MILLAN MJ: Dopamine D<sub>3</sub> receptor antagonists as therapeutic agents. *Drug Discov. Today* (2005) **10**:917-925.
82. HEIDBREDER CA, GARDNER EL, ZHENG-XIONG X *et al.*: The role of central dopamine D<sub>3</sub> receptors in drug addiction: a review of pharmacological evidence. *Brain Res. Rev.* (2005) **49**:77-105.
83. LIEBERMAN JA, KANE JM, ALVIR J: Provocative tests with psychostimulant drugs in schizophrenia. *Psychopharmacology* (1987) **91**:415-433.
84. CURRAN C, BYRAPPA C, McBRIDE A: Stimulant psychosis: systematic review. *Br. J. Psychiat.* (2004) **185**:196-204.
85. BAUMEISTER AA, FRANCIS JL: Historical development of the dopamine hypothesis of schizophrenia. *J. History Neurosci.* (2002) **11**:265-277.
86. SEEMAN P, WEINSHENKER D, QUIRION R *et al.*: Dopamine supersensitivity correlates with D<sub>2</sub><sup>High</sup> states, implying many paths to psychosis. *Proc. Nat. Acad. Sci. USA* (2005) **102**:3513-3518.
87. RICHELSON E, SOUDER T: Binding of antipsychotic drugs to human brain receptors. Focus on newer generation compounds. *Life Sci.* (2000) **68**:29-39.
88. RASMUSSEN K, BENVENGA MJ, BYMASTER FP *et al.*: Preclinical pharmacology of FMPD [6-fluoro-10-[3-(2-methoxyethyl)-4-methyl-piperazin-1-yl]-2-methyl-4*H*-3-thia-4,9-diaza-benzo[*f*]azulene]: A potential novel antipsychotic with lower histamine H<sub>1</sub> receptor affinity than olanzapine. *J. Pharmacol. Exper. Therap.* (2005) **315**:1265-1277.
89. ROTH BL, CRAIGO SC, CHOUDHARY MS *et al.*: Binding of typical and atypical antipsychotic agents to 5-hydroxytryptamine-6 and 5-hydroxytryptamine-7 receptors. *J. Pharmacol. Exper. Therap.* (1994) **268**:1403-1410.
90. PUROHIT A, SMITH C, HERRICK-DAVIS, TEITLER M: Stable expression of constitutively activated mutant h5HT<sub>6</sub> and h5HT<sub>7</sub> serotonin receptors: inverse agonist activity of antipsychotic drugs. *Psychopharmacology* (2005) **179**:461-469.

91. WILSON AA, McCORMICK P, KAPUR S *et al.*: Radiosynthesis and evaluation of [<sup>11</sup>C]-(+)-PHNO as a potential radiotracer for *in vivo* imaging of the dopamine D<sub>2</sub> high affinity state with positron emission tomography (PET). *J. Med. Chem.* (2005) **48**:4153-4160.
92. WILLEIT M, GINOVAR T N, KAPUR S, HOULE S, HUSSEY D, SEEMAN P, WILSON AA: High-affinity states of human brain dopamine D<sub>2/3</sub> receptors imaged by the agonist [<sup>11</sup>C]-(+)-PHNO. *Biol. Psychiat.* (2006) **59**:389-394.

### Website

101. [www.cyprotex.com](http://www.cyprotex.com)  
Cyprotex website (2006).

### Affiliation

Philip Seeman MD, PhD  
Professor, University of Toronto,  
Pharmacology and Psychiatry Departments,  
Faculty of Medicine, Medical Science Building,  
Room 4344, 1 King's College Circle, Toronto,  
M5S 1A8, Canada  
Tel: +1 416 978 4891; +1 416 971 2445;  
E-mail: Philip.Seeman@utoronto.ca