J. Pharm. Pharmacol. 1997, 49: 206–208
Received May 21, 1996
Accepted July 4, 1996

Antagonistic Effects of OPC-14597, a Novel Antipsychotic Drug, on Quinpirole- and (−)-Sulpiride-induced Changes in Evoked Dopamine Release in Rat Striatal Slices

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Abstract

The effects of a newly synthesized quinolinone derivative, 7-{4-(4-(2,3-dichlorophenyl)-1-piperazinyl) butoxy}-3,4-dihydro-2-(1*H*)-quinolinone (OPC-14597), a novel antipsychotic drug, on electrically evoked dopamine release in rat striatal slices were investigated.

OPC-14597 (0·1–10 μM) had no effect on the dopamine release evoked in the striatal slices. The decrease induced by quinpirole, a dopamine receptor agonist, in evoked dopamine release was attenuated by superfusion with OPC-14597 (1 and 10 μM) which by itself had no effect on evoked dopamine release. The increase induced by (−)-sulpiride, a dopamine receptor antagonist, in evoked dopamine release was, moreover, also attenuated by 1 and 10 μM OPC-14597.

These findings indicate that OPC-14597 antagonizes both dopamine agonist- and antagonist-induced changes in evoked dopamine release in striatal slices in rats.

The behavioural, neurochemical and pharmacological properties of a novel antipsychotic drug, 7-{4-(4(2,3-dichlorophenyl)-1-piperazinyl) butoxy}-3,4-dihydro-2-(1*H*)-quinolinone (OPC-14597), have already been reported (Kikuchi et al 1995; Semba et al 1995). In a behavioural study, OPC-14597 was shown to antagonize apomorphine-induced stereotypy and hyperlocomotion (Kikuchi et al 1995). The systemic administration of OPC-14597 at low doses (2·5–10 mg kg^{-1}) increased rat striatal extracellular 3,4-dihydroxyphenylacetic acid and homovanillic acid levels (Semba et al 1995). Although these properties of OPC-14597 are similar to those of other conventional antipsychotic drugs, OPC-14597 has no cataleptogenic activity and a high dose (40 mg kg^{-1}) of the drug has no effect on striatal extracellular 3,4-dihydroxyphenylacetic acid and homovanillic acid levels (Semba et al 1995). It has, moreover, been shown that OPC-14597 inhibits γ-butyrolactone-induced increases in the rate of dopamine synthesis in mouse forebrain and antagonizes increases induced in extracellular dopamine levels by the dopamine autoreceptor antagonist, (+)-AJ-76. OPC-14597 seems to act both as a dopamine agonist and as a dopamine antagonist. A recent clinical study showed that OPC-14597 alleviated both positive and negative symptoms of schizophrenia without causing any overt extrapyramidal side effects or any increase in plasma prolactin concentrations (Toru et al 1994).

In this study, to clarify the effect of OPC-14597 on release-modulating dopamine autoreceptors, we investigated the effects of OPC-14597 on dopamine agonist- or antagonist-induced changes, or both, in electrically evoked dopamine release in rat striatal slices.

Materials and Methods

Male Wistar rats, 250–300 g, were housed in a light-, humidity- and temperature-controlled environment for more than 7 days before the experiments. They were killed by decapitation, between 0900 and 1000 h. The brains were rapidly removed and three 0·3-mm thick slices were prepared, in ice-cold Kreb's solution oxygenated with 95% O_2 and 5% CO_2, with a Micro Slicer (Dohan EM. Co.) at A9760 to A 7630, according to the atlas of Konig & Klippel (1963). One of the slices was used for control experiments (perfusion without drugs) and two were used for perfusion with drugs. The striatal tissue in the slices was punched out with a metal tube (3 mm, i.d.). One striatal slice was placed in a chamber made from a teflon tube in which platinum electrodes were mounted to stimulate the slice. The slice was then superfused with Kreb's solution and oxygenated with 95% O_2 and 5% CO_2, at a flow rate of 0·7 mL min^{-1}, at 37°C. The composition of the Kreb's solution (mM) was: NaCl 118·0; KCl 4·9; $NaHCO_3$ 25·0; NaH_2PO_4 1·25; $CaCl_2$ 1·25; $MgCl_2$ 1·18; and glucose 11·0, together with nomifensine (3 μM). After 30-min superfusion, electrical field stimulation was performed with platinum spiral electrodes installed at the end of the chamber. The stimuli were 20 mA rectangular pulses of 2-ms duration, with a frequency of 1 Hz, applied for 2 min, 30 min (S1) and 62 min (S2) after the beginning of superfusion. Various concentrations of OPC-14597 with or without quinpirole (2 μM), a dopamine receptor agonist, were added to the superfusion medium 15 min before S2. In another experiment, 25 min before S2 (−)-sulpiride (1 μM), a dopamine receptor antagonist, was added to the superfusion medium 10 min before superfusion with various concentrations of OPC-14597. The superfusate was collected in tubes, as the 7-min fraction. Dopamine released into the superfusate was adsorbed on alumina, eluted with acetic acid (0·1 M; 300 μL), and quantified by high-performance liquid chromatography with electrochemical detection (HPLC-ECD) (Yamada et al 1993). The evoked dopamine release during S1 and S2 was calculated by subtracting the spontaneous release from the total release. The spontaneous release during each stimulation, assessed from the sample collected during the 7-min period

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preceding the stimulation, was expressed as spl and sp2, respectively.

Values are expressed as ng dopamine (mg protein)$^{-1}$/7-min fraction or as the S2/S1 ratio (mean ± s.e.m.). Statistical comparisons were performed by analysis of variance then Scheffé's test.

Results

The spontaneous release of dopamine was 0·22 ± 0·01 ng mg^{-1} protein/7-min fraction (mean ± s.e.m., n = 16, where n represents the number of slices taken from eight animals). Electrical stimulation induced a significant increase in dopamine release, 7 times greater than the spontaneous release (1·67 ± 0·24 ng (mg protein)$^{-1}$ fraction^{-1}). The control S2/S1 value was 1·29 ± 0·08. OPC-14597, even at a concentration higher than 10 μM, had no effect on either the evoked (Table 1) or spontaneous (data not shown) dopamine release. The dopamine receptor agonist, quinpirole (2 μM) reduced the evoked dopamine release in striatal slices by 44% (S2/S1 ratio, 0·72 ± 0·08, $F = 3·49$, $P < 0·05$ compared with the S2/S1 ratio of the control group). When slices were superfused with 1 or 10 μM OPC-14597, which by itself had no effect on evoked dopamine release, the quinpirole-induced decrease in evoked dopamine release was significantly attenuated in a concentration-dependent manner (1·25 ± 0·14 for 1 μM OPC-14597, $F = 3·58$, $P < 0·05$; 1·40 ± 0·10 for 10 μM OPC-14597, $F = 6·09$, $P < 0·01$, compared with the S2/S1 ratio of the quinpirole group, Table 2). In contrast, the dopamine receptor antagonist, (−)-sulpiride (2 μM), enhanced the evoked dopamine release by 135% (S2/S1 ratio, 1·74 ± 0·12, $F = 4·23$, $P < 0·05$, compared with the S2/S1 ratios of the control group). The (−)-sulpiride-induced increase in evoked dopamine release was significantly attenuated, in a concentration–dependent manner, by superfusion with OPC-14597 (1·20 ± 0·15 for 1 μM OPC-14597, $F = 3·13$, $P < 0·05$; 0·99 ± 0·09 for 10 μM OPC-14597, $F = 6·18$, $P < 0·01$, compared with the S2/S1 ratio of the (−)-sulpiride group, Table 3).

Table 1. Effects of OPC-14597 on electrically evoked dopamine release in rat striatal slices.

Drug released	Concn (μM)	n	Ratio of amounts of dopamine released before and after addition of drugs
Control	0	16	1·29 ± 0·08
(−)-Sulpiride	1	8	1·74 ± 0·12*
Quinpirole	2	8	0·72 ± 0·08*
OPC-14597	0·1	8	1·21 ± 0·12
OPC-14597	1	8	1·23 ± 0·06
OPC-14597	10	8	1·31 ± 0·07

*$P < 0·01$ compared with the S2/S1 ratio of the control group (analysis of variance then Scheffé's test).

Discussion

It is well established that dopamine D_2 or D_3 agonists reduce the evoked dopamine release from dopamine nerve terminals by activating release-modulating dopamine autoreceptors (Farnebo & Humberger 1971; Arbilla & Langer 1981; Starke et al 1983; Parker & Cubeddu 1985; Lane & Blaha 1986; Yamada et al 1991; 1994). OPC-14597 is considered to have a high affinity for D_2 and D_3 receptor binding (Sibley et al 1994) and to inhibit the reserpine- and γ-butyrolactone-induced increases in tyrosine hydroxylase activity in mouse and rat brain, these increases being completely antagonized by haloperidol (Kikuchi et al 1995). These results suggest that OPC-14597 acts as a dopamine autoreceptor agonist at dopamine nerve terminals. Despite these dopamine agonistic properties

Table 2. Antagonistic effect of OPC-14597 on quinpirole-induced reduction in evoked dopamine release in rat striatal slices.

Quinpirole concn (μM)	0	2	2	2	2
OPC-14597 concn (μM)	0	0	0·1	1	10
Ratio of amounts of dopamine released before and after addition of drugs	1·25 ± 0·08	0·72 ± 0·08	0·85 ± 0·06	1·25 ± 0·14*	1·40 ± 0·10**

Slices were superfused with 2 μM quinpirole containing various concentrations of OPC-14597 (0–10 μM) 22 min before the second stimulation. Values are expressed as means ± s.e.m. of the S2/S1 ratio (n = 8 for each point, where n represents number of slices taken from four animals). *$P < 0·05$, **$P < 0·01$; analysis of variance then Scheffés test.

Table 3. Antagonistic effect of OPC-14597 on (-)-sulpiride-induced increase in evoked dopamine release in rat striatal slices.

Sulpiride concn (μM)	0	1	1	1	1
OPC-14597 concn (μM)	0	0	0·1	1	10
Ratio of amounts of dopamine released before and after addition of drugs	1·20 ± 0·09	1·74 ± 0·12	1·89 ± 0·12	1·20 ± 0·15*	0·99 ± 0·09**

Slices were superfused with 1 μM (-)-sulpiride containing various concentrations of OPC-14597 (0–10 μM) 22 min before the second stimulation. Values are expressed as means ± s.e.m. of the S2/S1 ratio (n = 8 at each point, where n represents number of slices taken from four animals). *$P < 0·05$, **$P < 0·01$, analysis of variance then Scheffé's test.

of OPC-14597, the drug, by itself, failed to have any effect on evoked dopamine release in the rat striatal slices (Table 1). The presence of nomifensine, a dopamine-uptake inhibitor, in the Kreb's solution, which caused an increase in dopamine levels in synaptic clefts, might result in a tonic activation of dopamine autoreceptors. This might mask a weak dopamine receptor agonistic effect of OPC-14597 under the experimental conditions used in this work. Further study would be necessary to observe the effect of OPC-14597 on the evoked dopamine release under the nomifensine-free conditions. We have, however, reported that apomorphine or quinpirole reduces the evoked dopamine release under the present experimental conditions (Yamada et al 1994). These results indicate that the inhibitory effect of OPC-14597 on the evoked dopamine release is much weaker than those of apomorphine or quinpirole. Despite its small, or absent, inhibitory effect on dopamine release, the (−)-sulpiride-induced increase in evoked dopamine release was completely abolished by superfusion with OPC-14597 (Table 3). This result agrees with the findings of a previous study that increases in extracellular dopamine levels induced by (+)-AJ-76, D_2 and D_3 receptor antagonists in freely moving rats were attenuated by the systemic administration of OPC-14597 (Semba et al 1995). (−)-Sulpiride increases the evoked dopamine release by blocking release-modulating dopamine autoreceptors, this blocking action being antagonized by apomorphine (Yamada et al 1993). These findings and the present results suggest that the agonistic properties of OPC-14597 would be observed under conditions showing low dopaminergic tone, i.e. when there has been treatment with reserpine, γ-butyrolactone or dopamine receptor antagonists. OPC-14597 seems to act as a partial agonist of release-modulating dopamine autoreceptors. The concept of a partial dopamine agonist has been reviewed by Pulvirenti & Koob (1994), who state that partial agonist properties refer to the efficacy of a given receptor ligand that shows high affinity for the receptor, but low intrinsic activity.

The antagonistic effect of OPC-14597 on the quinpirole-induced reduction in evoked dopamine release (Table 2) could also be explained along the same lines. It has been reported that the firing of nucleus accumbens neurons induced by iontophoretically applied glutamate was inhibited by dopamine, SKF 38393 and quinpirole, but not by OPC-14597. The dopamine-, SKF 38393- and quinpirole-induced inhibition of the glutamate-induced firing was, however, also antagonized by the simultaneous application of OPC-14597 (Amano et al 1995). In a behavioural study, OPC-14597 inhibited the apomorphine-induced postsynaptic behavioural changes of stereotypy and hyperlocomotion in mice and rats; OPC-14597 also inhibited rotation in rats with unilateral striatal kainic acid lesions (Kikuchi et al 1995). These reports show that OPC-14597 has postsynaptic D_2 receptor antagonistic activity. In the present study OPC-14597 showed presynaptic D_2 receptor antagonistic activity when slices were superfused with a dopamine agonist. Under conditions of high dopaminergic tone, OPC-14597 could show an antagonistic action at presynaptic dopamine autoreceptors. Irrespective of the pre- and post-synaptic sites, OPC-14597 might have both dopamine-agonistic and -antagonistic properties; these would depend on the degree of concurrent receptor occupancy by the dopamine agonists or antagonists.

Acknowledgements

We thank Mr T. Kikuchi, Third Tokushima Institute of New Drug Research, Otsuka Pharmaceutical Co. Ltd for the generous supply of OPC-14597.

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