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Rapid Recovery of Self-Stimulation from Depression Produced by the Atypical Neuroleptic Risperidone is not Prevented by 5-HT_2 Receptor Stimulation

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GROTTICK, A. J., A. M. J. MONTGOMERY AND L. J. HERBERG. *Rapid recovery of self-stimulation from depression produced by the atypical neuroleptic risperidone is not prevented by 5-HT_2 receptor stimulation.* PHARMACOL BIOCHEM BEHAV **58**(4) 1045–1049, 1997.—Behavioral effects of the antipsychotic drug risperidone were tested in rats responding for variable-interval stimulation of the ventral tegmental area (VTA). Risperidone (0–0.9 mg/kg) produced a dose-dependent depression of responding in the 60 min after injection. Self-stimulation tests delayed for 30 or 120 min after injection showed that inhibition of responding by risperidone was limited in duration, with response rates recovering to pre-injection levels in a time-dependent manner. Recovery occurred regardless of opportunity to engage in self-stimulation, and was virtually complete at a time when receptor occupancy has been shown to be almost undiminished. The atypical properties of risperidone have been ascribed to its potent antagonist activity at 5-HT_2 receptors; however, spontaneous recovery from the effects of risperidone was not prevented by simultaneous administration of a selective 5-HT_2 agonist (DOI), even though DOI when given alone produced a 50–70% reduction in response rates. These results show that the inhibitory effect of risperidone on operant performance may be self-limiting in a manner that is not accounted for by its pharmacokinetic properties nor by its antagonist activity at central 5-HT_2 receptors. © 1997 Elsevier Science Inc.

Alpha$_2$ adrenoceptors Atypical neuroleptics DOI Risperidone Schizophrenia Self-stimulation 5-HT_2 receptors

NEUROLEPTIC agents in the management of schizophrenia have been classified as typical or atypical on the basis of differing biochemical and clinical effects (11). Atypical compounds are reported to be effective in treating both positive and negative symptoms of schizophrenia in patients refractory to conventional treatments, and to be relatively free from extrapyramidal side effects (9). Atypical antipsychotics differ also in their pharmacological activity: typical neuroleptics tend to be preferentially active as antagonists at dopamine (DA) receptors, especially the D2 subtype, whereas atypical agents show greater affinity for other sites that are thought to contribute to their special beneficial effects (12). The antipsychotic clozapine, for example, has a particularly high 5-HT_2A/D2 receptor binding ratio (13), leading to the suggestion that an affinity 10-fold higher for 5-HT_2 than for D2 receptors may be a key property of atypical antischizophrenic drugs (13).

Typical and atypical antischizophrenic drugs have also been distinguished by animal behavioral procedures: typical neuroleptic drugs characteristically lead to progressive declines in responding within sessions in both appetitively and aversively motivated tasks, including intracranial self-stimulation (ICSS) (5), whereas atypical neuroleptics fail to do (18,19), and performance in the case of ICSS, in rats treated with risperidone (7), may even revert to normal levels at a time when receptor occupancy is still virtually complete (23).

The apparently self-limited depressant activity of atypical neuroleptics suggests an interesting possibility: that they contain their own antidote to the incremental inhibitory activity

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seen with typical neuroleptics. This could account for the absence of within-session response decrements in various behavioral tasks (18,19) and, taken further, for the partial recovery of responding for ICSS (7). Responding for ICSS has proved a useful measure of the behavioral effects of antischizophrenic drugs (20,26) and has the particular advantage in the present instance of not being subject to satiation, enabling drug effects to be studied as they develop over time. Drug-induced changes in self-stimulation rate at ventral tegmental sites, as in the present study, have been shown to measure reward-unrelated performance factors (1), reflecting the activity of pyramidal and extrapyramidal motor mechanisms, rather than mesolimbic reinforcement processes. The former are especially implicated in the unwanted motor side effects of typical neuroleptics (26).

The present study aimed first to characterise the reported inhibition of self-stimulation by risperidone, and its apparently spontaneous reversal over time (7). We then examined the possible contribution of 5-HT_2 receptor antagonism to this phenomenon, investigating whether the reversal could be prevented by simultaneous administration of the 5-HT_{2A} agonist, (±)-2,5-dimethoxy-4-iodoamphetamine (DOI).

METHOD

Subjects

Male PVG hooded rats (Bantin & Kingman, Hull, UK) weighing 220–270 g were implanted, under halothane anaesthesia, with twisted bipolar stainless steel electrodes of 0.25 mm nominal diameter (Plastics One Inc., Roanoke, VA), aimed at the ventral tegmental area (VTA). Electrode coordinates relative to bregma (16) were −5.5 mm posterior, 1.0 mm lateral, and 8.5 mm below the surface of the skull. At the end of the experiment the brains were fixed with 10% formal saline and the anatomical location of electrode placements was verified on 10× enlarged photographs of 50-μm unstained frozen sections.

Self-stimulation

On recovery from surgery the rats were trained to operate a lever to obtain a 0.5-s, 50-Hz sinewave constant-current reinforcing stimulus available at randomly varied intervals of 10 s mean duration (VI 10 s). Stimulus intensities were fixed at the lowest value that would just maintain uninterrupted responding in repeated preliminary trials with current intensities descending in 1-decilog steps. Response rates, which ranged between approximately 10 and 40 bar presses/min, were printed out automatically at 10-min intervals and expressed as a percentage of preinjection baseline rates. Test sessions were preceded by a warm-up period of approximately 15 min. Postinjection rates were timed from the first lever-press after injection.

Drugs

Risperidone (a gift from Janssen Pharmaceutica, Beerse, Belgium) was dissolved in distilled water with 15–20 drops of 85% lactic acid, and the pH adjusted to 6.0 with 0.1-M NaOH. (±)-2,5-Dimethoxy-4-iodoamphetamine (DOI) was dissolved in distilled water. Solutions were administered subcutaneously in a volume of 1.0 ml/kg.

Procedure

Experiments were based on a fully repeated measures design. Subjects were pseudorandomly assigned to treatment conditions and were allowed at least 2 drug-free days between tests. Dose–response relationships in the hour after injection were determined in all animals for five doses of risperidone (0, 0.03, 0.10, 0.30, and 0.9 mg/kg) [dose–response curves not shown; see (7)] and four doses of DOI (0, 0.24, 0.80, and 2.40 mg/kg.

Changes over time were examined for two doses of risperidone (0.2 and 0.9 mg/kg). Pre-test time-out intervals of 30 or 120 min, during which the rats were returned to their home cages, were followed by 90 min of responding. These time-out intervals allowed performance to be assessed over a period of 210 min, and distinguished between changes due to the passage of time and changes that depended on lever-pressing activity by the rat. The lower dose of risperidone (0.2 mg/kg) was not tested with the longer time-out interval because preliminary studies showed that responding after this dose returned to control levels within 120 min following injection.

Risperidone/DOI interaction (0.9 mg/kg vs. 0.8 mg/kg) was examined over a 4-h session comprising periods of responding and time-outs. Following a warm-up period, the rats were injected with either risperidone or vehicle, or DOI or vehicle, or with both risperidone and DOI, and returned to their home cages for 30 min. Recording continued for 30 min, followed by a time-out of 120 min, and a further 60-min period of responding.

Statistical Analysis

Data from all experiments except the dose–response studies were analyzed by 30-min intervals and at individual 10-min time points, enabling the detection of changes in response rate that occurred during intervening periods of nonresponding. All data were initially analyzed by ANOVA followed by tests of simple main effects where appropriate.

RESULTS

Short-term Effect of Risperidone

As previously reported (7), risperidone depressed self-stimulation responding in a dose-dependent manner during the 60 min after injection, $F(4, 28) = 106.7$, $p < 0.001$, with an ED_{50} of approximately 0.2 mg/kg. Figure 1 plots the time course of this effect and shows that all doses larger than 30 μg/

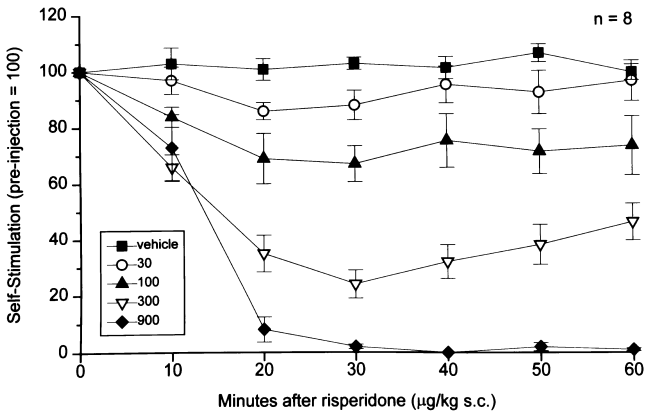

FIG. 1. Mean self-stimulation response rates (±SE) recorded at 10-min intervals in the 1 h following injection of risperidone or vehicle. Scores are expressed as a percentage of the preinjection rate. Vertical bars represent standard errors of the mean.

kg produced a significant effect within 20 min of injection, persisting throughout the 60 min of recording.

Risperidone Time Course

Self-stimulation tests prolonged to 3.5 h showed that depression of responding was only temporary (Fig. 2). Both doses of risperidone that were tested over this period produced an initial depression of responding [for 0.2 mg/kg, $F(1, 81) = 32.24$, $p < 0.01$; for 0.9 mg/kg, $F(1, 162) = 74.43$, $p < 0.01$, in the first 10-min of self-stimulation] that continued for the next 20 min (i.e., from 30–60 min postinjection). However, by the end of the next 10-min period, the effect of the 0.2-mg/kg dose was significantly less than in the previous 10 min, $F(1, 144) = 5.02$, $p < 0.05$. This recovery gained momentum during the rest of the session, so that by 90 min after injection the effect of the 0.2-mg/kg dose did not differ from that of vehicle, $F(1, 81) = 3.29$, NS. A similar pattern was seen for the 0.9-mg/kg dose, although over a longer time course: after 2 h, responding had recovered significantly compared to 20 min earlier, $F(1, 306) = 11.11$, $p < 0.01$, and recovery continued until the rate of responding was no longer significantly less than after vehicle [at 200 min: $F(1, 162) = 0.94$, NS]. In contrast, rats injected with doses of conventional neuroleptics that were similarly effective at 30 min (haloperidol 0.075 mg/kg, $n = 9$, or chlorpromazine 1.0 mg/kg, $n = 9$) and tested for self-stimulation in an identical manner, showed further slowing, or negligible recovery, especially during time-out periods, in the 3-h period after injection: respectively from lows of 28% ± 5.1 and 32% ± 8.6 after haloperidol and chlorpromazine, to ensuing scores of 27% ± 5.0 and 45% ± 8.6, measured 190 min after injection in the first 10 min after a 1-h time-out. This nonincremental pattern is consistent with several previous self-stimulation studies [results not shown; see (20,26) for review].

A further finding was that initial response rates of risperidone-treated rats placed in the Skinner box after a 2-h time-out were not significantly different from the response rates of rats that had spent the preceding 90 min engaged in self-stimulation after a time-out of only 30 min, $F(1, 306) = 1.59$, NS. The initial response rates of the long (120-min) pretest interval group were moreover significantly higher than the initial rates of the short (30-min) pretest interval group, $F(1, 306) = 7.07$, $p < 0.01$.

DOI Dose–response

All doses of DOI that were tested led to depressed responding (Fig. 3), $F(3, 21) = 47.5$, $p < 0.001$, with the time taken to reach maximal effect being dose dependent. During the second half of the session only the 0.24-mg/kg and 2.4-mg/kg doses differed from each other, $F(1, 42) = 9.40$, $p < 0.01$.

Risperidone/DOI Interaction

Risperidone (0.9 mg/kg) produced an almost total initial suppression of responding (97% depression) during the first 10-min period after injection, while DOI alone (0.8 mg/kg) also had an appreciable effect during this period (57%), so

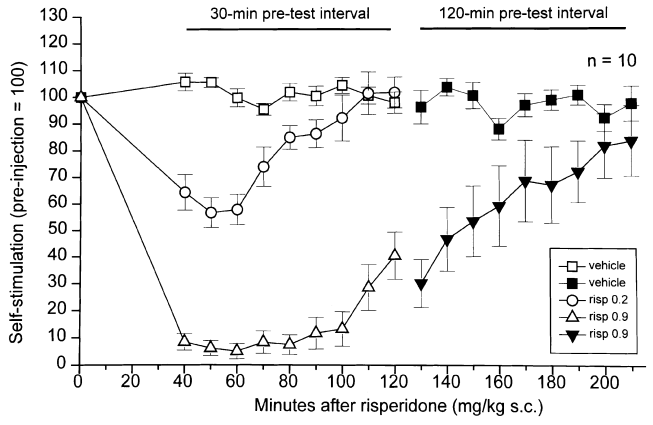

FIG. 2. The effects of two doses of risperidone (0.2, 0.9 mg/kg) on responding for ICSS over a period of 3.5 h. The data were derived from two separate test sessions with different pretest intervals (30 min, open symbols; 120 min, closed symbols). Points refer to mean response rates (±SE) as a percentage of preinjection rates.

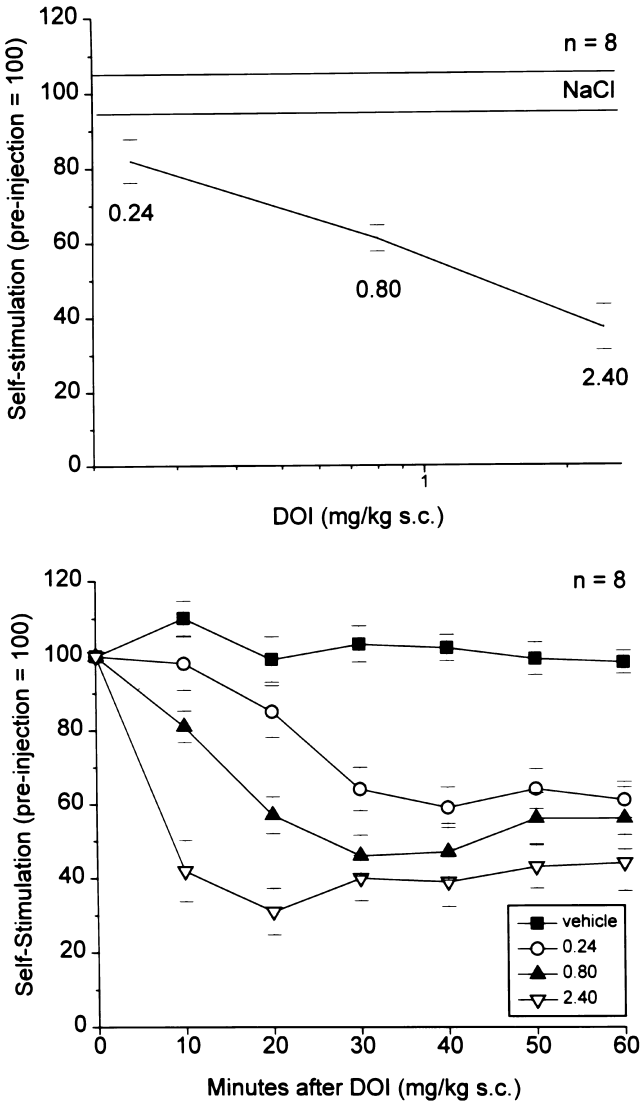

FIG. 3. Dose–response for 30 min after injection (top panel) and time–response (bottom panel) for four doses of DOI. Points show mean rates (±SE) as a percentage of preinjection rates. Paired horizontal bars indicate mean response rate (±SE) after injection of NaCl.

that any additive effects of the two were cancelled out by a floor effect (risperidone + DOI yielding 98% depression). Following this period, risperidone + DOI did not differ from risperidone alone at any time point, even though the corresponding depression produced by DOI alone remained between 50 and 70%. By the end of the session, rats injected with risperidone alone or risperidone + DOI, respectively, showed 77 and 69% reversals of the initial suppressions (Fig. 4).

DISCUSSION

The present experiments confirm the transitory nature of the inhibitory effect of risperidone on responding for electrical self-stimulation. Response rates returned to preinjection levels in a time-dependent manner: they were no different from control rates when measured 110 min after the lower dose of risperidone (0.2 mg/kg), even though responding had fallen to 57% of control levels only 60 min earlier. As much as 83% of the initial suppression was reversed over a 3.5-h period.

Recovery that took place during the two pretest time-out intervals showed that it was independent of the opportunity to engage in responding. If recovery were response-dependent one would expect that response rates would not have differed immediately following the long and short pretest time-outs. However, the initial suppressions differed significantly (8% vs. 30% for the 0.9-mg/kg dose). In addition, recovery after 0.9 mg/kg began only after 70 or 80 min of responding in rats subjected to the 30-min pretest delay, but began immediately in the group that had been subject to the 120-min pretest delay. Depression and recovery without the opportunity to engage in responding indicates that risperidone-induced depression is not to any large extent the result of an extinction process, and that recovery was not a learnt coping response.

The recovery from the depressant effect of risperidone is unlikely to have been influenced to any appreciable extent by factors such as changes in its binding to D2 or 5-HT_2 receptors. Receptor studies show that with the doses used, 5-HT_2 and D2 receptor occupancy would reach approximately 75% and 25%, respectively, for the 0.2-mg/kg dose, and 90% and 50% for the 0.9-mg/kg dose of risperidone (21), and dose levels comparable to our higher dose continued to show virtually undiminished binding to frontal cortex and striatal receptors 4 h after parenteral administration in vivo (23).

Suppression of operant responding by antischizophrenic drugs has been explained in terms either of motor or incentive/motivational processes. Although evidence supports both points of view, incremental inhibitory effects are commonly found with both appetitively and aversively motivated, and unconditioned behaviors [for review see (6)]. Microanalysis of these response decrement patterns (RDP's) suggest that they result from a slowing of response termination that cannot be reproduced with reductions in reinforcement magnitude (4). These changes in the structure of responding and in response rates cannot be accounted for by either drug accumulation or elimination (6), and suggest that the underlying cause is performance related. It seems especially significant that atypical antischizophrenic drugs do not produce RDPs (18,19), a finding that could account both for their relative lack of motor side effects in the clinic and in behavioral studies, and for the recovery of responding in risperidone-treated rats.

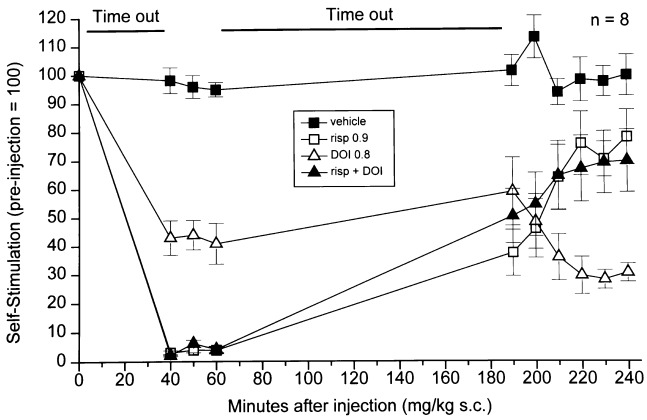

FIG. 4. The effects of risperidone (0.9 mg/kg), DOI (0.8 mg/kg), and their combination, on responding for ICSS over a 210-min period. The rats responded for a total of 90 min during the 210-min period, with the first 30 min of responding being separated from the last 60 min by a time out period of 2 h in which animals were returned to home cages. Points show mean (±SE) response rates as a percentage of preinjection rates.

Pharmacological studies have suggested that a high 5-HT_2:D_2 binding ratio is important for the enhanced antipsychotic effects and reduced motor side effects of atypical antischizophrenic drugs (12,17,22). These conclusions are consistent with results of various behavioral tests, including operant responding for food rewards (19), the consumption of concentrated sucrose solutions (14), and measures of cataleptic immobility [(2,3,8), but see (24,25) for discrepant findings). These results suggest that 5-HT_2 antagonism may act both to counteract the blockade of striatal DA, and to potentiate mesolimbic blockade by antischizophrenic drugs (10). We tested whether 5-HT_2 antagonism may be responsible for recovery in self-stimulation by administering the selective 5-HT_2 agonist DOI in combination with risperidone. DOI at the dose tested was suppressant in its own right, producing a 50% reduction in responding over the session, but the addition of DOI did not affect the rate of recovery, and towards the end of the test session, response rates under the combination were 40% higher than those under DOI alone. These results suggest that other properties of risperidone, for example, its affinity for α_2-adrenoreceptors (15) may be responsible for the spontaneous reversal of its depressant effect on self-stimulation performance.

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REFERENCES

1. Agars, K.; Kokkinidis, L.: Scopolamine increases nonreinforced behavior in an intracranial self-stimulation discrimination paradigm. Pharmacol. Biochem. Behav. 43:657–660; 1992.
2. Balsara, J. J.; Jadhav, J. H.; Chandorka, A. G.: Effects of drugs influencing central serotonergic mechanisms on haloperidol-induced catalepsy. Psychopharmacology (Berlin) 62:67–69; 1979.

3. Bethany, S.; Neal-Beliveau, J. N.; Lucki, J.; Lucki, I.: Serotonergic involvement in haloperidol-induced catalepsy. J. Pharmacol. Exp. Ther. 265:207–217; 1993.
4. Ettenberg, A.: Dopamine, antipsychotics and rewarded behaviour. Neurosci. Biobehav. Rev. 3:105–111; 1989.
5. Fouriezos, G.; Wise, R. A.: Pimozide-induced extinction of intracranial self-stimulation: Response patterns rule out motor or performance deficits. Brain Res. 103:377–380; 1976.
6. Fowler, S. C.: Neuroleptics produce within-session response decrements: Facts and theories. Drug Dev. Res. 20:101–116; 1990.
7. Herberg, L. J.; Montgomery, A. M. J.; Grottick, A. J.: α_2-Adrenoceptor antagonism may contribute to the atypical properties of risperidone: Experimental support for the Nutt case. J. Psychopharmacol. 9:281–283; 1995.
8. Hicks, P. B.: The effect of serotonergic agents on haloperidol-induced catalepsy. Life Sci. 47:1609–1615; 1990.
9. Kane, J.; Honigfeld, G.; Singer, J.; Meltzer, H. Y.: Clozapine for the treatment-resistant schizophrenic: A double-blind comparison with chlorpromazine. Arch. Gen. Psychiatry 45:789–796; 1988.
10. Kapur, S.: 5-HT_2 antagonism and EPS benefits: Is there a causal connection? Psychopharmacology (Berlin) 124:35–39; 1996.
11. Kinon, B. H.; Lieberman, J. A.: Mechanisms of action of atypical antipsychotic drugs: A critical analysis. Psychopharmacology (Berlin) 124:2–34; 1996.
12. Leysen, J. E.; Janssen, P. M. F.; Schotte, A.; Luyte, W. H. M. L.; Megens, A. A. H. P.: Interaction of antipsychotic drugs with neurotransmitter receptor sites in vitro and in vivo in relation to pharmacological and clinical effects: Role of 5-HT_2 receptors. Psychopharmacology (Berlin) 112:540–554; 1993.
13. Meltzer, H. Y.; Matsubara, S.; Lee, J.-C.: Classification of typical and atypical antipsychotic drugs on the basis of dopamine D1, D2 and serotonin$_2$ pKi values. J. Pharmacol. Exp. Ther. 251:238–246; 1989.
14. Montgomery, A. M. J.; Suri, A.: Potentiation of the effects of raclopride on sucrose consumption by the 5-HT_2 antagonist ritanserin. Psychopharmacology (Berlin) 123:98–102; 1996.
15. Nutt, D. J.: Putting the 'A' in atypical: Does α_2-adrenoceptor antagonism account for the therapeutic advantage of new antipsychotics? J. Psychopharmacol. 8:193–195; 1994.
16. Paxinos, G.; Watson, C.: The rat brain in stereotaxic co-ordinates, 2nd ed. Sydney: Academic Press; 1986.
17. Saller, C. F.; Czupryna, M. J.; Salama, A. I.: 5-HT_2 receptor blockade by ICI 169, 369 and other 5-HT_2 antagonists modulates the effect of D2 dopamine receptor blockade. J. Pharmacol. Exp. Ther. 253:1162–1170; 1990.
18. Sanger, D. J.: Response decrement patterns after antipsychotic and nonantipsychotic drugs. Psychopharmacology (Berlin) 89:98–104; 1986.
19. Sanger, D. J.; Perrault, G.: Effects of typical and atypical antipsychotic drugs on response decrement patterns in rats. J. Pharmacol. Exp. Ther. 272:708–713; 1995.
20. Schaefer, G. J.; Michael, R. P.: Schedule-controlled brain self-stimulation: Has it utility for behavioral pharmacology? Neurosci. Biobehav. Rev. 16:569–583; 1992.
21. Schotte, A.; Janssen, P. F. M.; Gommeren , W.; Luyten, W. H. M. L.; Van Gompel, P.; Le Sage, A. S.; De Loore, K.; Leysen, J. E.: Risperidone compared with new and reference antipsychotic drugs: *In vitro* and *in vivo* receptor binding. Psychopharmacology (Berlin) 124:57–73; 1996.
22. Stockmeier, C. A.; DiCarlo, J.; Zhan, Y.; Thompson, P.; Meltzer, H. Y.: Characterisation of typical and atypical antipsychotic drugs based on in vivo occupancy of serotonin-2 and dopamine-2 receptors. J. Pharmacol. Exp. Ther. 2663:1374–1384; 1993.
23. Sumiyoshi, T.; Kido, H.; Sakamoto, H.; Urasaki, K.; Suzuki, K.; Yamaguchi, N.; Mori, H.; Shiba, K.; Yokogawa, K.: In vivo dopamine-D2 and serotonin-5-HT_2 receptor binding study of risperidone and haloperidol. Pharmacol. Biochem. Behav. 47:553–557; 1994.
24. Vidali, M.; Fregnan, G. B.: Effect of different CNS-active drugs on the catalepsy induced by antipsychotics. Curr. Ther. Res. 25: 544–556; 1979.
25. Wadenberg, M. L.: Antagonism by 8-OH-DPAT, but not ritanserin, of catalepsy induced by SCH 23390 in the rat. J. Neural Trans. [Gen. Section], 89:49–59; 1992.
26. Wauquier, A.: Neuroleptics and self-stimulation. Int. Rev. Neurobiol. 21:335–403; 1970.