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Pharmacological Validation of Ritanserin and Risperidone in the Drug Discrimination Test Procedure in the Rat

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ABSTRACT

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The results presented here indicate that 0.16 mg/kg LSD, 2.50 mg/kg 8-OHDPAT, 1.25 mg/kg d-amphetamine, 10.00 mg/kg cocaine, 40.00 mg/kg chlordiazepoxide, 2.50 mg/kg xylazine, and 0.04 mg/kg fentanyl can be used as discriminative stimuli in a two-lever drug discrimination test procedure in the rat. The central 5-HT_2 antagonist ritanserin and the 5-HT_2 and catecholamine (CA)-antagonist risperidone were tested for stimulus generalization with, and possible antagonism of, the discriminative stimulus properties of the various training drugs. With both drugs at all doses tested, no stimulus generalization was observed with any of the training drugs. Ritanserin completely blocked the discriminative stimulus properties of LSD at 40.00 mg/kg but was, at doses up to 40.00 mg/kg, unable to block the discriminative stimulus properties of any of the other training drugs. Risperidone completely antagonized the stimulus properties of LSD and d-amphetamine, partially blocked cocaine, and possessed minor effects on 8-OHDPAT and fentanyl. Whereas ritanserin was almost without any effects or response rate, risperidone mostly reduced response rate at doses starting between 0.16 and 0.63 mg/kg. However, the complete antagonism of the LSD and d-amphetamine was observed without effects on response rate. Globally, these results confirm ritanserin to be a selective 5-HT_2 antagonist without any effects on conditioned behaviour. Risperidone was found to be a potent 5-HT_2 and DA antagonist, affecting con-

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ditioned behaviour by interfering with response rate and with the response-reinforcement contingency.

Key words: drug discrimination learning, LSD, 8-OHDPAT, d-amphetamine, cocaine, chlordiazepoxide, xylazine, fentanyl

INTRODUCTION

Ritanserin has been characterized as a potent, selective, and long-acting 5-HT_2 antagonist. The selective 5-HT_2 antagonist properties were measured both in vivo [Awouters et al., 1988] and in vitro [Leysen et al., 1985; Van Nueten et al., 1986]. Biochemically, a >50-fold difference was observed between the K_i-values of ritanserin for 5-HT_2 binding sites (K_i: 0.2 nM) and for H_1 binding sites (K_i: 15.0 nM), the second lowest measured K_i-value. For interactions with 5-HT_{1A} or 5-HT_{1B} binding sites ratios >1,000 were reported [Leysen, 1988; Leysen et al., 1985, 1987]. In in vivo tests, the selectivity of ritanserin for 5-HT_2 mediated effects was even more pronounced. Whereas the lowest ED_{50}s for peripheral (reversal of tryptamine-induced cyanosis) and central 5-HT_2 (antagonism of tryptamine forepaw treading and body coarse tremor) antagonism were 0.0070 and 0.037 mg/kg, respectively, no activity was observed at doses up to 10.00 mg/kg in tests measuring interactions with other neurotransmitters, including tests for 5-HT_1 activity [Awouters et al., 1988; Meert et al., 1987]. Thus, in in vivo experiments, a >250-fold selectivity with regards to interactions with 5-HT_2, as compared to other neurotransmitters, was observed. In additional testing, ritanserin was found to possess anxiety-, stress-, and depression-reducing properties [Meert, 1986; Meert and Janssen, 1989; Colpaert et al., 1985; Critchley and Handley, 1987; Marek et al., 1989]. Furthermore, ritanserin was observed to increase slow wave sleep (S.W.S.), both in animals [Dugovic and Wauquier, 1987] and man [Declerck et al., 1987; Idzikowski et al., 1986]. Clinically, across various disorders, ritanserin decreased fatigue and increased energy levels, an activity recognized as an antiasthenic effect. Furthermore, improvements of depressed mood and anxiety were observed, probably as a consequence of better coping mechanisms. Combinations of these elements prompted investigators to ascribe thymosthenic properties to the therapeutic actions of ritanserin in man. Therefore, ritanserin was described as an original thymosthenic with a regulatory impact on S.W.S. [Reyntjens et al., 1986; Janssen, 1988].

Risperidone, in contrast to ritanserin, was found to be a potent 5-HT_2 and catecholamine antagonist [Janssen et al., 1988]. In terms of 5-HT_2 antagonism in vivo, risperidone was even more potent than ritanserin with the lowest ED_{50}s for peripheral and central 5-HT_2 antagonism being 0.0011 and 0.014 mg/kg, respectively. Peripheral and central dopamine D_2 antagonism was already measured at 0.0057 and 0.016 mg/kg. Antagonism of noradrenaline and clonidine were measured at doses of 0.074 and 0.67 mg/kg. Also biochemically, risperidone revealed a very high binding affinity for 5-HT_2 binding sites (K_i-value of 0.16 nM) [Leysen et al., 1988]. Risperidone also had a high affinity for catecholaminergic binding sites (K_i-values of 0.81, 3.13, 2.23, and 7.54 nM for adrenergic-α_1, dopamine D_2, histamine H_1, and adrenergic-α_2 binding sites). Due to its potent 5-HT_2 and catecholamine antagonist properties, risperidone was observed to be a very potent 5-HTP and LSD antagonist [Meert et al., 1988, 1989]. Furthermore, risperidone was found active in other tests used to screen for antipsychotics [Janssen et al., 1988]. Clinically, risperidone was demonstrated to be effective against both the positive and negative symptoms of schizophrenia [Janssen, 1987; Roose et al., 1989].

In order to further characterize the in vivo pharmacological profile of ritanserin and risperidone, we tested both drugs in a two-lever drug discrimination test procedure in the rat. The training drugs used in this study included the hallucinogenic LSD, the putative 5-HT1A agonist 8-OHDPAT, the stimulants d-amphetamine and cocaine, the benzodiazepine chlordiazepoxide, the α_2-adrenoceptor agonist xylazine, and the opiate fentanyl. Experiments on generalization with and on antagonism of the discriminative stimulus properties of these

training drugs were carried out. Some attention was also given to the discrimination training of the different training drugs.

MATERIALS AND METHODS

Animals

Ninety-four male Wistar rats weighing 240 ± 20 g at the beginning of the experiment were used. At testing the body weight varied among the animals between 300 and 400 g. The animals were housed individually in standard living cages. All housing and testing took place in a continuously illuminated and air-conditioned room (temperature: 21 ± 1°C; relative humidity: 65 ± 5 %). Tap water was freely available. Access to dry powdered standard laboratory food was limited (see below).

Apparatus

Six test cages (Coulbourn Instruments®) fitted with a house light and two levers were programmed by solid-state logic modules. Between the two levers, a food pellet receptacle was mounted 2 cm above the floor of the cages. The cages were placed in a light- and sound-attenuating outer box.

Procedure

The drug discrimination procedure has been described in detail elsewhere [Meert et al., 1989]. Daily discrimination training started after habituation and initial shaping to lever press for food on a fixed ratio 10 (FR = 10) schedule. At a fixed time before being placed in the test cage, the rats were injected with either the training drug or physiological saline. Depending on whether they were injected with drug or saline, they obtained food by pressing either the drug lever (DL) or the saline lever (SL), respectively. After every 10th press (FR-10) on the correct lever, a 45 mg food pellet was delivered by a food dispenser. Responses on the incorrect lever (i.e., the SL after the training drug or the DL after saline) had no consequences. The lever assignments were DL: left, SL: right in about one-half of the animals and SL: left, DL: right in the other half. These assignments remained unchanged throughout the study. At the beginning of each session, the FRF-value was noted. This is the sum of the total number of responses on both levers until ten responses are made on the appropriate lever. Fifteen minutes after the rat was placed in the test chamber, the session was terminated, and all responses on both levers were recorded. The response rate (i.e., the total sum of the responses on both the DL and SL during the 15 min session) and the percentage responding on the selected lever (i.e., the ratio of the number of responses on the appropriate lever to the response rate) were calculated After the session, the animal was removed to its living cage. Two hours later, it was allowed to feed freely for 1 hour. On weekends, no sessions were run, and the animals were given free access to food between 10 a.m. and 12 noon.

Every week, each rat was run once daily on 5 consecutive days. Daily training drug (D) or saline (S) injections were given according to two monthly alternating sequences, i.e., 1) D-S-S-D-S, S-D-D-S-S, S-D-S-D-D, D-S-D-S-D and 2) S-D-D-S-S, D-S-D-S-D, D-S-S-D-D, S-D-S-D-S. Rats whose sequential numbers were odd were run according to one sequence, whereas even-numbered animals were run according to the alternative sequence. Discrimination training proceeded individually for each rat until ten consecutive sessions occurred in which an FRF-value ≤ 14 was obtained. Animals reaching this criterion were used for testing. For all the different training drugs, almost every animal entering a particular drug discrimination training achieved the criterion. No differences among the different-training drugs were observed.

Test sessions were run on Fridays only, and the training procedure was continued on the remaining days. On test days, the animal was given the treatment being studied and was put in the operant chamber at a specified time after the treatment. It was then noted on which of

the two levers the animal first made a total of ten responses. This lever is referred to as the selected lever. Once this lever selection was established, the rat obtained a first food pellet, and subsequent reinforcement was contingent upon pressing (FR-10) the selected lever. Testing was postponed to the next test day if the FRF-value exceeded 14 on any of the 3 most recent training days. In addition, test data were discarded, and the test condition was later retested if the FRF-value during testing exceeded 14.

Before being used in tests, the animals were given 1 week of habituation to a double treatment condition. That is, before every administration of saline or the training drug, the animals were always given an additional subcutaneous injection of saline 60 min prior to the test. The double treatment on training days was continued for the duration of the experiments.

The compounds tested for their possible use as a training drug in the drug discrimination test procedure included LSD (0.16 mg/kg, IP, t-15 min), 8-OHDPAT (2.50 mg/kg, IP, t-15 min), d-amphetamine (1.25 mg/kg, SC, t-30 min), cocaine (10.00 mg/kg, IP, t-15 min), chlordiazepoxide (40.00 mg/kg, SC, t-30 min), xylazine (2.50 mg/kg, IP, t-30 min), and fentanyl (0.04 mg/kg, SC, t-30 min). The ED_{50} for generalization of each training drug with the corresponding training condition of that drug was determined (e.g., ED_{50} for LSD in the 0.16 mg/kg LSD-saline condition).

Ritanserin and risperidone were examined in two sets of experiments that were designed to determine their possible generalization with and antagonism of the training drug. To test for a generalization to the training drug, either ritanserin, risperidone, or the vehicles were injected subcutaneously (SC) at t-60 min before testing, followed by an injection with saline given according to the treatment conditions of the training drug. To test for an antagonism, ritanserin, risperidone, or the solvents were injected SC at 60 min before testing. This injection was, at the appropriate time, followed by a treatment with the training drug. In some experiments on the antagonism of the training drug, pretreatment times of 120 and 240 min were used (see "Results" section). The doses of ritanserin and of risperidone that were tested were selected on the basis of preliminary experiments. Each test condition was tested in 5 rats. The rats were randomly selected to participate in the tests on antagonist or agonist drug effects. Once a rat was selected for a particular experiment, all doses and the vehicle solution were tested within this rat.

Drugs

Chlordiazepoxide hydrochloride, cocaine hydrochloride, fentanyl citrate, d-lysergic acid diethylamide tartrate (LSD), 8-OHDPAT, and xylazine hydrochloride were dissolved in water. Risperidone and ritanserin up to 1 mg/ml were dissolved in 2 equivalents (Eq) tartaric acid; 4 mg/ml ritanserin was dissolved in 2 Eq lactic acid and 20% propylene glycol. Because no differences were observed between the two vehicles of ritanserin, only the results with 2 Eq tartaric acid are reported. The doses of ritanserin and risperidone were selected from the geometrical series 0.0025, 0.01, ... , 0.63, 2.50 mg/kg. Occasionally, additional doses from the geometrical series 0.00125, 0.0050, . . . , 1.25, 5.00 mg/kg were used. All doses of drugs, saline, or vehicle were administered in a volume of 1 ml/100 g body weight.

Statistics

The Wilcoxon matched-pairs signed-ranks test (Siegel, 1956; two-tailed) was used throughout in order to evaluate differences between drug and vehicle treatments. ED_{50}s and 95% confidence limits were calculated according to Finney's iterative method [Finney, 1971].

RESULTS

A summary of the duration of training of the seven training drugs is given in Table 1 (upper panel) and Figure 1. The average ($\pm$ 1 SEM) duration of training for 15 rats to discriminate 0.16 mg/kg LSD from saline, and thus to reach the criterion of 10 $\times$ FRF $\leq$ 14,

TABLE 1. Summary of the Results Obtained With the Different Training Drugs in the Drug Discrimination Studies*

Training conditions						
Drug	Dose (mg/kg)	Route	-t	No. of rats	Sessions to 10 × FRF ≤ 14: Mean ± 1 SEM	ED_{50} (95% confidence limits) generalization training drug(mg/kg)
LSD	0.16	IP	15	15	27.33 ± 2.78	0.26 (0.16–0.41)
8-OHDPAT	2.50	IP	15	12	37.33 ± 4.89	0.51 (0.34–0.76)
d-amphetamine	1.25	SC	30	15	22.60 ± 2.02	0.51 (0.34–0.76)
Cocaine	10.00	IP	15	7	33.86 ± 3.50	1.55 (1.030–2.31)
Chlordiazepoxide	40.00	SC	30	15	29.47 ± 3.99	4.086 (2.53–6.59)
Xylazine	2.50	IP	30	15	37.60 ± 6.57	0.77 (0.52–1.16)
Fentanyl	0.04	SC	30	15	32.33 ± 3.24	0.021 (0.014–0.032)

	Dose			Response rate: mean ± 1 SEM	
Drug	(mg/kg)	Route	-t	Drug sessions	Saline sessions
LSD	0.16	IP	15	1,041.02 ± 81.46	1,425.09 ± 86.59
8-OHDPAT	2.50	IP	12	434.66 ± 42.40	1,264.45 ± 52.79
d-amphetamine	1.25	SC	30	555.23 ± 62.62	1,365.08 ± 67.56
Cocaine	10.00	IP	15	838.24 ± 89.87	1,361.47 ± 142.94
Chlordiazepoxide	40.00	SC	30	1,400.11 ± 72.36	1,454.61 ± 82.20
Xylazine	2.50	IP	30	510.39 ± 42.17	1,349.72 ± 115.02
Fentanyl	0.04	SC	30	691.83 ± 99.70	1,453.83 ± 104.25

*The table represents the different training conditions, the No. of rats trained at each condition, the average (± 1 SEM) No. of sessions required to reach the training criterion of 10 FRF-values ≤ 14, the ED_{50}s (based on data of 5 rats) for stimulus generalization of the training drug with the corresponding training drug condition (upper panel), and the average response rates in drug and saline sessions (lower panel).

was 27.33 (± 2.78) sessions. For 2.50 mg/kg 8-OHDPAT, the average duration of training in 12 rats was 37.33 (± 4.89) sessions; for 1.25 mg/kg d-amphetamine in 15 animals it was 22.60 (± 2.02), for 10.00 mg/kg cocaine in 7 rats it was 33.86 (± 3.50), and for 40.00 mg/kg chlordiazepoxide, 2.50 mg/kg xylazine, and 0.04 mg/kg fentanyl in 15 animals it was 29.47 (± 3.99), 37.60 (± 6.57), and 32.33 (± 3.24) sessions, respectively. Subdivided into drug and saline sessions, the LSD-trained animals reached on the average FRF-values < 14 from session 6 on in the drug sessions and from session 10 on in the saline sessions. At all further drug and saline sessions, mean FRF-values < 14 were noted. For 2.50 mg/kg 8-OHDPAT, the first mean FRF-value < 14 was observed on day 7 for the drug sessions and on day 6 for the saline sessions. During further training, FRF-values < 14 were always measured in the drug and saline sessions starting from session 10. For 1.25 mg/kg d-amphetamine, FRF-values < 14 started from session 6, both in drug and saline sessions. For 10.00 mg/kg cocaine, the first mean value < 14 was noted on the fifth drug and ninth saline session, although consistent values below 14 were only found from drug session 12 and saline session 15. For 40.00 mg/kg chlordiazepoxide, an FRF value < 14 was first noted in drug session 6 and saline session 7 but only from saline session 14 onwards did average FRF-values remain below 14. In the drug sessions of rats trained to discriminate 2.50 mg/kg xylazine from saline, the first mean FRF-value < 14 was observed at session 10, and they were continuously below this level from session 13 onwards. The only exception was seen at session 19, where an average FRF of 14.40 (± 1.80) was obtained. During saline sessions, a continuous FRF-value below the criterion was observed starting from session 11. For 0.04 mg/kg fentanyl, FRF-values < 14 were measured from drug and saline session 7, although it was only from drug session 17 onwards that average FRF-values remained below 14.

After training, average response rates were calculated for each training drug in both the

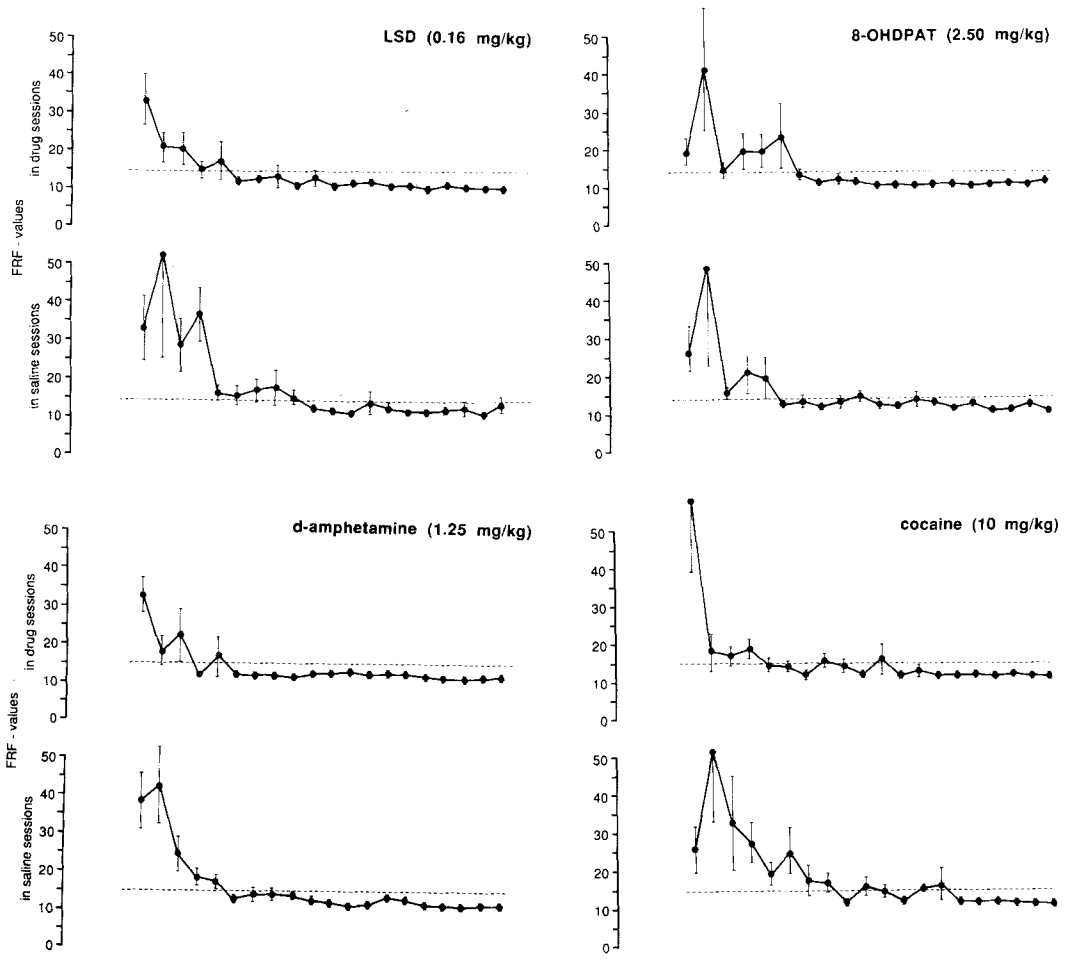


Fig. 1

drug and the saline sessions, using response rates from the first 5 successive drug and saline sessions, respectively (Table 1; lower panel). In terms of average response rate, no differences [Mann-Whitney U-test, two-tailed, $P > .05$; Siegel, 1956] were observed in the saline sessions of the different training drugs with the highest and lowest mean percentages being 1,454.61 (± 82.20) (chlordiazepoxide) and 1,264.45 (± 52.79) responses (8-OHDPAT), respectively. In the drug sessions, the average response rates differed considerably among the training drugs with average rates varying between 434.66 (± 42.40) (8-OHDPAT) and 1,400.11 (± 72.36) (chlordiazepoxide) responses. The relative order of response rates during the drug sessions was: chlordiazepoxide > LSD > cocaine > fentanyl > d-amphetamine > xylazine > 8-OHDPAT. As compared to the corresponding saline sessions, significantly lower ($P < .05$) rates were observed during all drug sessions except for chlordiazepoxide. The percentage

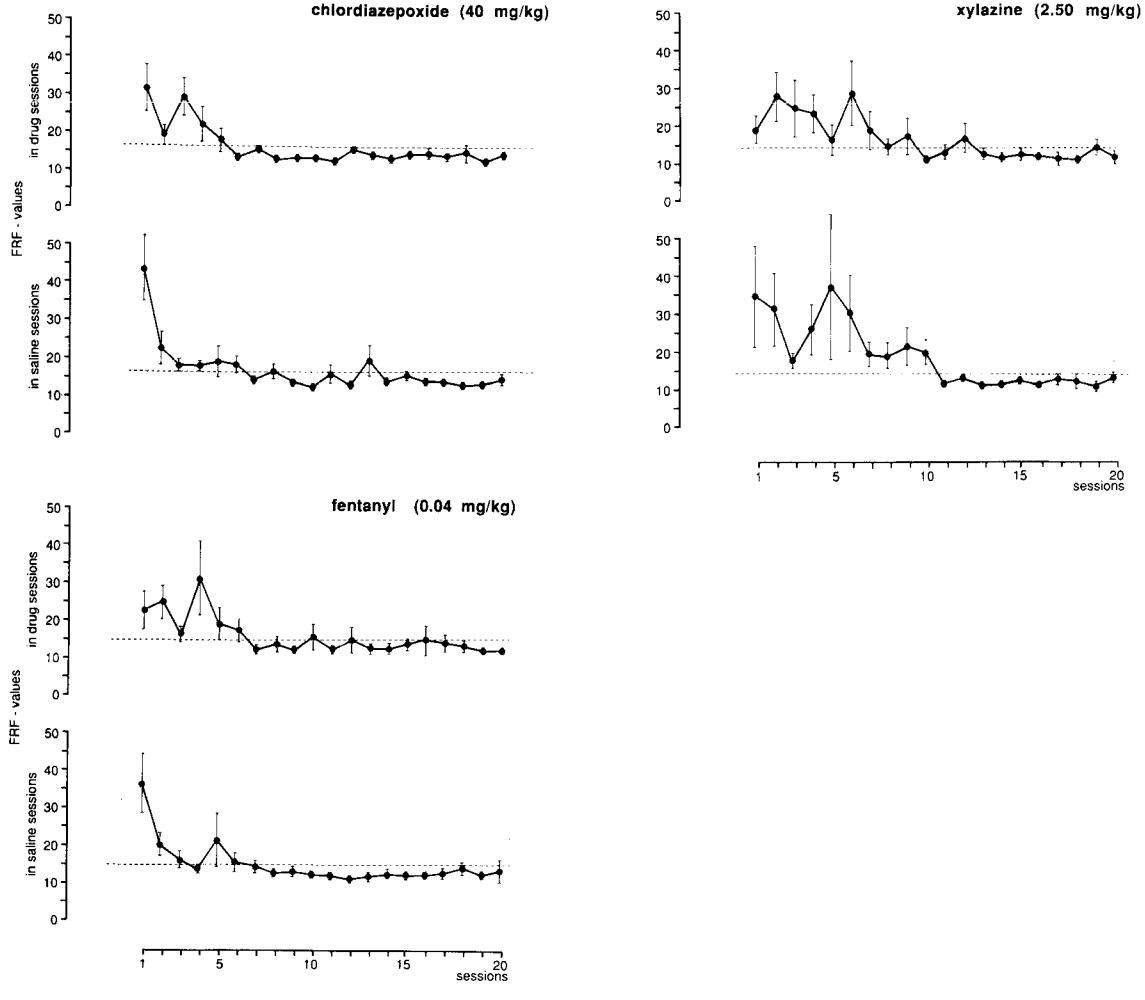


Fig. 1. Training results of rats learning to discriminate 0.16 mg/kg LSD, 2.50 mg/kg 8-OHDPAT, 1.25 mg/kg d-amphetamine, 10 mg/kg cocaine, 40 mg/kg chlordiazepoxide, 2.50 mg/kg xylazine, and 0.04 mg/kg fentanyl from saline in a two-lever drug discrimination test procedure. The figure represents the mean (± 1 SEM) FRF-value in drug and saline sessions in function of the duration of training, based on 15 rats. LSD was injected IP 15 min prior to test and 45 min after a SC treatment with saline. The horizontal dotted line represents the FRF-value = 14.

reductions in drug versus saline sessions were 26.95, 65.62, 59.33, 38.45, 62.19, and 52.41% for LSD, 8-OHDPAT, d-amphetamine, cocaine, xylazine, and fentanyl, respectively.

For each training drug, the ED_{50} (and 95% confidence limits) of the training drug for stimulus generalization with the training condition was determined (Table 1, right upper panel). The ED_{50} of LSD in 0.16 mg/kg trained animals was 0.26 (0.16–0.41) mg/kg. For 2.50 mg/kg 8-OHDPAT and 1.25 mg/kg d-amphetamine it was 0.51 (0.34–0.76) mg/kg. For 10.00 mg/kg cocaine, 40.00 mg/kg chlordiazepoxide, 2.50 mg/kg xylazine, and 0.04 mg/kg fentanyl the corresponding ED_{50}s were 1.55 (1.030–2.31), 4.086 (2.53–6.59), 0.77 (0.52–1.16), and 0.021 (0.014–0.032) mg/kg, respectively.

Ritanserin was tested for stimulus generalization with LSD, 8-OHDPAT, d-amphet amine, cocaine, chlordiazepoxide, xylazine, and fentanyl. At doses ranging from 0.16 to 40.00 mg/kg ritanserin, no single animal selected the drug lever in any of the tests (Fig. 2). The accuracy with which ritanserin-treated rats selected the saline lever was also reflected in the average percentages of responding on the selected (in casu saline) lever and in the mean FRF-values for lever selection (Table 2). Mean percentages of responding on the saline lever $\leq$ 97.5% or FRF-values $>$ 11.00 were not measured at any time except for 10.00 mg/kg ritanserin in the fentanyl-trained rats, where the average FRF-value was 11.20 ($\pm$ 0.73). Response rate, expressed as a percentage of the rate obtained in the last saline session and compared to the vehicle treatment, was significantly reduced ($P < .05$) with 0.63 mg/kg ritanserin in fentanyl-trained rats and with 40.00 mg/kg ritanserin in chlordiazepoxide- and xylazine-treated animals (Fig. 2).

Ritanserin antagonized the discriminative stimulus properties of LSD in a dose-related manner, while having no effect on 8-OHDPAT, d-amphetamine, xylazine, fentanyl, and cocaine (Fig. 3). As was observed during the generalization experiments, the average FRF-values were $\leq$11.00 and the percentage responding on the selected lever was $>$97.5% except for 0.63 and 2.50 mg/kg ritanserin at 8-OHDPAT and 2.50, 10.00, and 40.00 mg/kg at fentanyl (Table 3). Response rates, expressed as a percentage of the rate during the last drug session and compared to vehicle controls, were reduced at 2.5, 10.00, and 40.00 mg/kg ritanserin in the experiments on xylazine antagonism.

Risperidone, tested at doses ranging from 0.0025 to 0.63 mg/kg, produced no stimulus generalization at all with either LSD, 8-OHDPAT, d-amphetamine, cocaine, chlordiaze-poxide, xylazine, or fentanyl (Fig. 4). Except for 0.16 mg/kg risperidone in 8-OHDPAT-trained rats and for 0.63 mg/kg risperidone in all training conditions, the mean percentages responding on the selected (saline) lever were $>$97.5%. With 0.63 mg/kg risperidone, the average percentages responding varied between 77.70% ($\pm$ 3.50) (cocaine) and 90.91% ($\pm$ 3.97) (fentanyl). In all the generalization tests, average FRF-values $\leq$ 11.00 were obtained (Table 4). As compared to the vehicle treatment, response rate reductions were observed with 0.16 mg/kg risperidone in 8-OHDPAT-, cocaine-, and chlordiazepoxide-trained rats. With 0.63 mg/kg risperidone, there was a significant reduction in response rate in all tests on stimulus generalization (Fig. 4).

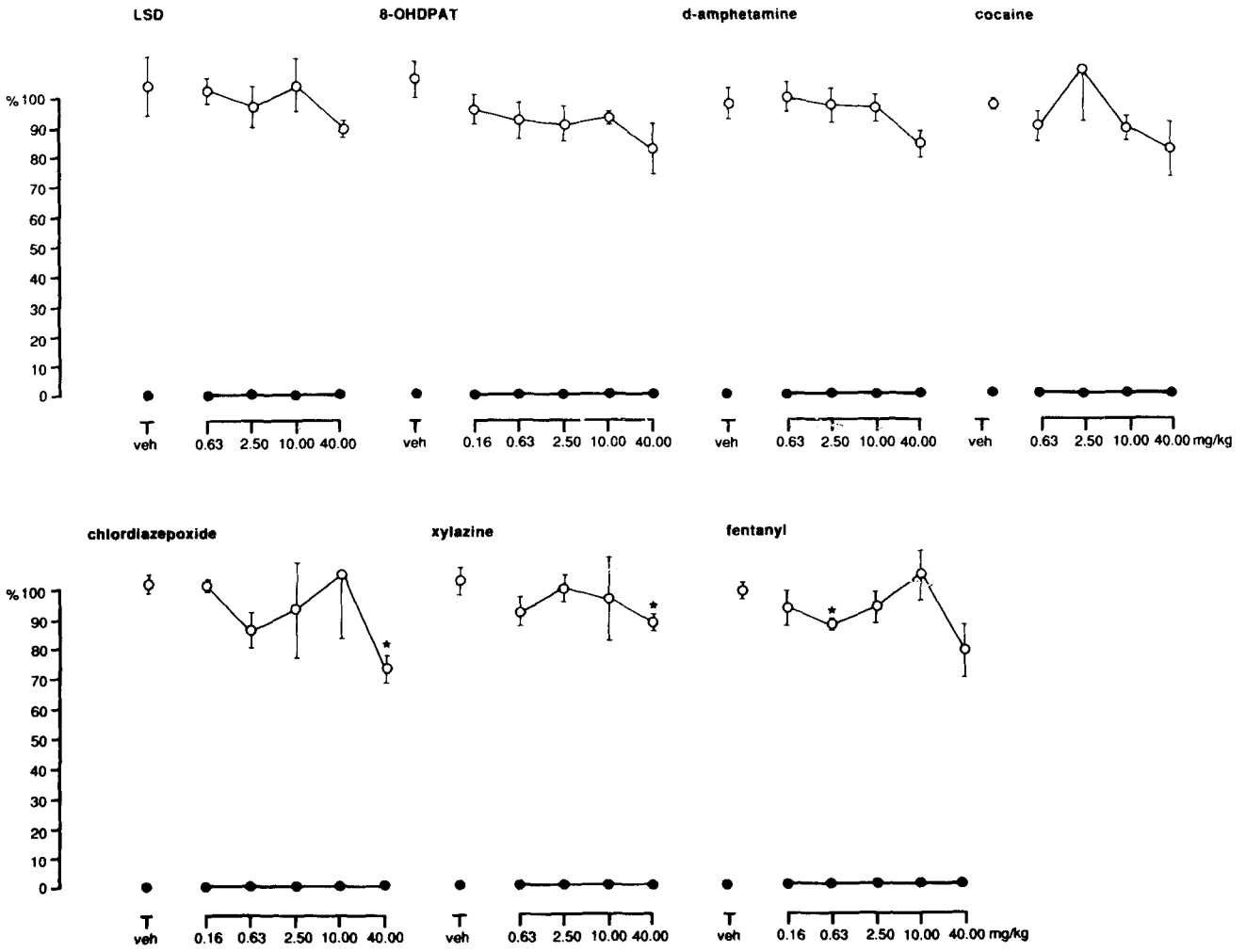
Risperidone was tested for its ability to antagonize the discriminative stimulus properties of LSD after a pretreatment period of 1 and 4 hours. At both periods of time, a dose-related antagonism of LSD was observed, reaching a 100% antagonism at 0.63 mg/kg (Fig. 5). However, whereas at the 1 hour condition, doses $\geq$ 0.16 mg/kg reduced response rate, no such effects were apparent after 4 hours. In 8-OHDPAT-trained rats, 1 out of 5 rats selected the saline lever at 0.63 mg/kg risperidone, producing an antagonism of 20%. On rats trained to discriminate 2.50 mg/kg xylazine from saline, no antagonism was observed with risperidone at doses up to 0.63 mg/kg. At this dose, almost a complete inhibition of response rate was observed. At a dose of 0.63 mg/kg risperidone, given Sc 60 min before testing, risperidone blocked the discriminative stimulus properties of fentanyl in 60% of the rats. However, at this dose a rate reduction of more than 80% was present. After a 2 hour pretreatment, no antag-onism of the discriminative stimulus properties of fentanyl was present anymore although

response rate reductions were still present (Fig. 5). Risperidone also antagonized the discriminative stimulus properties of d-amphetamine. After 1 hour of pretreatment, an antagonism of 80% was observed at the dose of 0.63 mg/kg risperidone (Fig. 6). To completely block the discriminative effects of d-amphetamine after 2 hours, a dose of 1.25 mg/kg risperidone was needed. No significant reductions in response rate were observed in these d-amphetamine experiments with risperidone although there was, especially at the highest tested doses, a tendency for a reduction in response rate. In contrast to the antagonism of the d-amphetamine cue, risperidone was unable to block the discriminative stimulus properties of cocaine after a 1 hour pretreatment period. At doses up to 0.63 mg/kg risperidone, and this in spite of a severe rate reduction, no antagonism of cocaine was found. After 2 hours of pretreatment, 0.63 and 2.50 mg/kg risperidone antagonized cocaine with, respectively, 20% and 40% antagonism. Here also, strong rate-reducing effects were apparent.

In all antagonism studies, risperidone was observed to produce FRF-values $\leq$ 11.00 except at 0.63 mg/kg risperidone in LSD- (1 hour) and 8-OHDPAT-trained rats. In these two conditions, the average FRF-values were 12.50 ($\pm$ 1.50) and 11.25 ($\pm$ 0.63), respectively (Tables 5, 6). In terms of the percentage responding on the selected lever, values $>$ 97.50% were measured in the experiments on LSD antagonism with the exception of 0.16 and 0.63 mg/kg risperidone after 1 hour. Here the mean values were 89.01% ($\pm$ 4.63) and 85.91% ($\pm$ 13.11). In the experiments on the antagonism of 8-OHDPAT, the average percent responding on the selected lever varied from 98.56% ($\pm$ 1.34) at 0.01 mg/kg risperidone to 83.31% ($\pm$ 15.65) at 0.63 mg/kg, with a vehicle control value of 93.72% ($\pm$ 5.75). For d-amphetamine the percentage responding on the selected lever ranged from 78.90% ($\pm$ 9.36) (0.63 mg/kg risperidone at 2 hours) to 99.87% ($\pm$ 0.06) (0.01 mg/kg risperidone at 1 hour). With regards to cocaine and xylazine values $>$ 97.00% were always obtained. The only exception was observed with 2.50 mg/kg risperidone after a pretreatment period of 2 hours with a mean percentage responding of 95.74% ($\pm$ 2.60). For fentanyl, the percentage responding on the selected lever varied between 99.71% ($\pm$ 0.18) (0.01 mg/kg risperidone at 1 hour) and 57.33% ($\pm$ 21.62) (2.50 mg/kg risperidone at 2 hours).

DISCUSSION

The results on the training data presented here indicate that LSD (0.16 mg/kg, IP, t-15 min), 8-OHDPAT (2.50 mg/kg, IP, t-15 min), d-amphetamine (1.25 mg/kg, SC, t-30 min), cocaine (10.00 mg/kg, IP, t-15 min), chlordiazepoxide (40.00 mg/kg, SC, t-30 min), xylazine (2.50 mg/kg, IP, t-30 min) and fentanyl (0.04 mg/kg, SC, t-30 min) can be used as discriminative stimuli in a drug discrimination test procedure in the rat. The relative duration of training of these drugs, expressed as the average number of sessions to reach the criterion of 10 successive FRF-values $\leq$ 14, was d-amphetamine $<$ LSD $<$ chlordiazepoxide $<$ fentanyl $<$ cocaine $<$ 8-OHDPAT $<$ xylazine. In terms of average response rates after training, no differences were observed between the different training drugs in the saline sessions. However, the number of responses during the drug sessions considerably varied among the training drugs and except for 40.00 mg/kg chlordiazepoxide, significant lower rates were obtained in the drug sessions as compared to the corresponding saline sessions. The relative order of rate reduction during the drug sessions was 8-OHDPAT $>$ xylazine $>$ d-amphetamine $>$ fentanyl $>$ cocaine $>$ LSD $>$ chlordiazepoxide. For all training drugs, generalization gradients with the training conditions could be obtained after a subcutaneous treatment at 60 min before testing. The corresponding ED_{50}s were 0.26 mg/kg for LSD, 0.51 mg/kg for 8-OHDPAT and d-amphetamine, 1.55 mg/kg for cocaine, 4.086 mg/kg for chlordiazepoxide, 0.77 mg/kg for xylazine, and 0.021 mg/kg for fentanyl. The ratios of the ED_{50}s versus the training dose of the different training drugs were 1.63, 0.20, 0.41, 0.16, 0.10, 0.31, and 0.53, respectively. As a consequence, to produce a 50% stimulus generalization, 1.6 times the training dose of LSD was needed, whereas for chlordiazepoxide only a tenth of the training dose was sufficient.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0.16 0.63 2.50 10.00 40.00
veh
0.63 2.50 10.00 40.00
veh
0.63 2.50 10.00 40.00 mg/kg
chlordiazepoxide
xylazine
fentanyl
%100
90
80
70
60
50
40
30
20
10
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0.16 0.63 2.50 10.00 40.00
veh
0.63 2.50 10.00 40.00
veh
0.16 0.63 2.50 10.00 40.00 mg/kg

TABLE 2. Percentage Responding on the Selected Lever (Upper Panel) and FRF-Values (Lower Panel) of Rats Treated With Ritanserin in the Generalization Experiments†

Generalization compound tested	Vehicle	Dose of ritanserin (in mg/kg) 0.16	0.63	2.50	10.00	40.00
LSD	99.98 ± 0.02	—	99.95 ± 0.05	99.22 ± 0.78	99.97 ± 0.03	100.00 ± 0.00
8-OHDPAT	99.98 ± 0.02	99.98 ± 0.02	99.97 ± 0.02	99.98 ± 0.02	99.97 ± 0.02	99.87 ± 0.06
d-amphetamine	99.94 ± 0.04	—	99.97 ± 0.03	100.00 ± 0.00	99.99 ± 0.01	99.98 ± 0.02
Cocaine	99.99 ± 0.01	—	99.81 ± 0.19	99.89 ± 0.05	99.93 ± 0.04	99.88 ± 0.03
Chlordiaze-poxide	99.96 ± 0.04	99.91 ± 0.06	99.76 ± 0.19	99.82 ± 0.10	99.72 ± 0.21	99.78 ± 0.13
Xylazine	99.81 ± 0.19	—	99.99 ± 0.01	99.83 ± 0.14	97.97 ± 1.99	99.19 ± 0.82
Fentanyl	99.88 ± 0.09	99.89 ± 0.09	99.96 ± 0.03	99.95 ± 0.03	99.82 ± 0.12	98.78 ± 0.67
LSD	10.00 ± 0.00	—	10.00 ± 0.00	10.20 ± 0.20	10.00 ± 0.00	10.00 ± 0.00
8-OHDPAT	10.20 ± 0.20	10.20 ± 0.20	10.20 ± 0.20	10.20 ± 0.20	10.00 ± 0.00	10.20 ± 0.20
d-amphetamine	10.20 ± 0.20	—	10.00 ± 0.00	10.00 ± 0.00	10.00 ± 0.00	10.00 ± 0.00
Cocaine	10.20 ± 0.20	—	10.60 ± 0.60	11.00 ± 0.45	10.40 ± 0.24	10.00 ± 0.00
Chlordiaze-poxide	10.40 ± 0.40	10.40 ± 0.40	10.80 ± 0.58	10.80 ± 0.49	10.40 ± 0.24	10.80 ± 0.37
Xylazine	10.00 ± 0.00	—	10.20 ± 0.20	10.40 ± 0.40	10.20 ± 0.20	10.00 ± 0.00
Fentanyl	10.60 ± 0.24	11.20 ± 0.80	10.40 ± 0.24	10.80 ± 0.49	11.20 ± 0.73	10.20 ± 0.20

†Given are the different doses of ritanserin in function of the different training compounds. Each data set represents the average (± 1 SEM) value of 5 rats. Differences from vehicle conditions were evaluated using the Wilcoxon test (two-tailed; $P < .05$).

Ritanserin and risperidone, tested for stimulus generalization after a subcutaneous treatment at 60 min before testing with doses ranging from 0.16 to 40.00 mg/kg and from 0.0025 to 0.63 mg/kg, respectively, produced no stimulus generalization at all with either LSD, 8-OHDPAT, d-amphetamine, cocaine, chlordiazepoxide, xylazine, or fentanyl. Because in the drug discrimination test procedure only drugs producing analogous subjective effects as the training drug reveal a stimulus generalization with the training drug [Colpaert and Slangen, 1982], the present results indicate ritanserin and risperidone to be devoid of the subjective effects analogous to LSD, 8-OHPAT, d-amphetamine, cocaine, chlordiazepoxide, xylazine, and fentanyl. Thus, both ritanserin and risperidone possess no intrinsic hallucinogenic [Glennon and Rosecrans, 1982; Glennon et al., 1983], no serotonin 5-HT_{1A}-agonist [Glennon, 1986; Tricklebanck et al., 1987], no stimulatory and/or dopaminergic [Nielsen et al., 1989; Colpaert, 1986; Colpaert et al., 1978a,b], no benzodiazepine-like [Colpaert et al., 1976a; Sanger and Zivkoviv, 1987], no α_2-adrenoceptor agonist [Colpaert and Janssen, 1985] nor central opiate-like [Colpaert, 1978; Colpaert and Janssen, 1986] effects. The lack of stimulus generalization with ritanserin and risperidone to any of the training conditions was also re-

Fig. 2. Effects of ritanserin in generalization experiments in rats (n = 5) trained to discriminate either LSD, 8-OHDPAT, d-amphetamine, cocaine, chlordiazepoxide, xylazine, or fentanyl from saline in a drug discrimination test procedure. Ritanserin was injected SC 60 min before test. A saline injection was dependent on the training conditions, given 15 or 30 min before testing. Abscissa: doses of ritanserin in mg/kg. The ordinate expresses the percentage of rats selecting the DL (bold line) and the percentage of response rate (open circles). Animals not selecting the DL selected the SL. Response rate (mean ± 1 SEM) expresses the number of responses emitted in the test session as a percentage of the responses emitted in the most recently preceding saline session. One asterisk indicates $P < .05$ (two-tailed, Wilcoxon test) for the difference between the test result and the vehicle result.

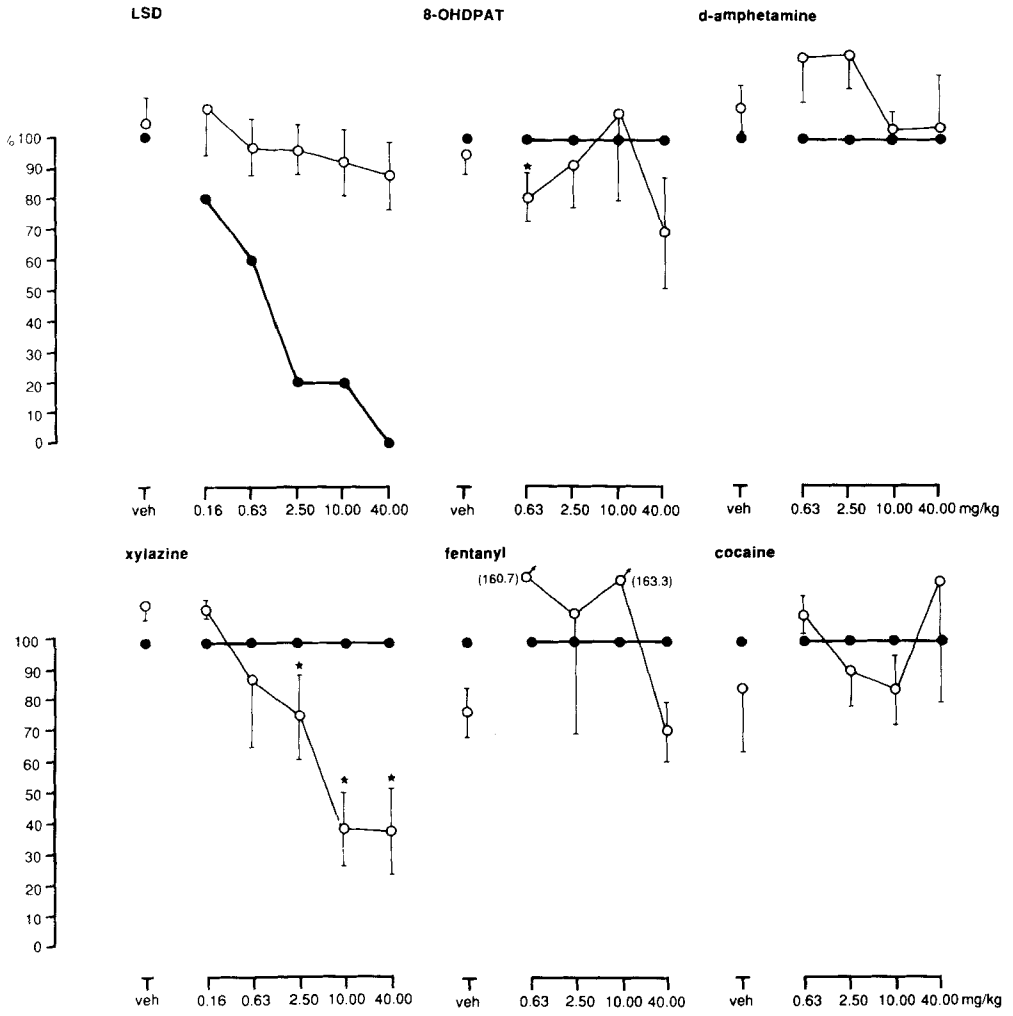


Fig. 3. Ritanserin in experiments on the antagonism of the discriminative stimulus properties of LSD, 8-OHDPAT, d-amphetamine, cocaine, chlordiazepoxide, xylazine, and fentanyl in a drug discrimination test procedure. Ritanserin was injected SC 60 min before test. This treatment was followed, at the appropriate time, by the training drug treatment. The ordinate expresses the percentage of rats selecting the DL (bold line) and the percentage of response rate (open circles). Animals not selecting the DL selected the SL. Response rate (mean ± SEM) expresses the number of responses emitted in the test session as a percentage of the responses emitted in the most recently preceding drug session. One asterisk indicates $P < .05$ (two-tailed, Wilcoxon test) for the difference between the test result and the vehicle result.

flected in the nearly perfect FRF-values (≤11.00) and percentages of responding on the selected saline lever (>97%). Only at ≥0.63 mg/kg risperidone, were percentages of responding on the SL < 90.00% observed. Thus, in spite of the fact that the animals started to make a correct lever selection (FRF-value ≤ 11.00) and earned food pellet reinforcement, they made frequent responses on the alternative (in casu DL), non food-reinforced, lever. A sedative-effect-mediated interference with stimulus control is not likely to account for these effects

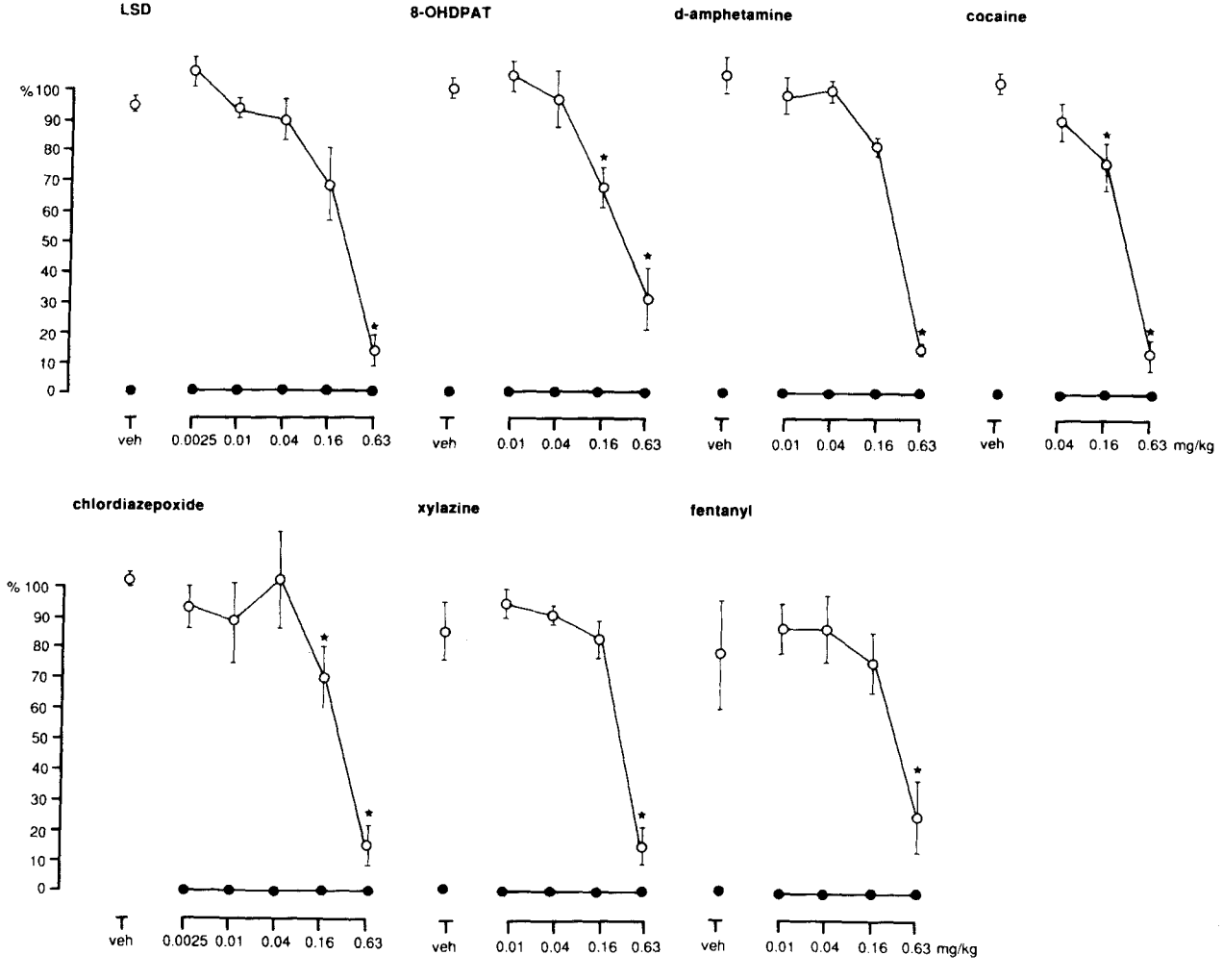
TABLE 3. Percentage Responding on the Selected Lever (Upper Panel) and FRF-Values (Lower Panel) of Rats Treated With Ritanserin in the Antagonism Experiments (See Also Table 2).

Antagonism compound tested	Vehicle	Dose of ritanserin (in mg/kg) 0.16	0.63	2.50	10.00	40.00
LSD	99.85 ± 0.08	100.00 ± 0.00	98.12 ± 1.84	97.46 ± 2.52	99.97 ± 0.03	99.53 ± 0.40
8-OHDPAT	100.00 ± 0.00	—	94.43 ± 5.20	96.82 ± 2.70	98.75 ± 0.89	98.28 ± 1.72
d-amphetamine	98.26 ± 1.74	—	97.88 ± 1.16	99.33 ± 0.48	99.76 ± 0.16	99.58 ± 0.25
Cocaine	99.05 ± 0.95	—	98.44 ± 1.56	99.13 ± 0.87	98.88 ± 1.08	99.87 ± 0.13
Xylazine	99.53 ± 0.32	99.91 ± 0.09	97.99 ± 1.12	97.77 ± 1.31	98.87 ± 0.92	100.00 ± 0.00
Fentanyl	98.50 ± 0.53	—	98.25 ± 0.91	93.05 ± 4.12	95.71 ± 3.89	84.96 ± 4.52*
LSD	10.60 ± 0.60	10.00 ± 0.00	10.00 ± 0.00	10.00 ± 0.00	10.20 ± 0.20	10.40 ± 0.40
8-OHDPAT	10.00 ± 0.00	—	10.20 ± 0.20	10.00 ± 0.00	10.20 ± 0.20	10.00 ± 0.00
d-amphetamine	10.00 ± 0.00	—	10.40 ± 0.40	10.00 ± 0.00	10.00 ± 0.00	10.00 ± 0.00
Cocaine	10.00 ± 0.00	—	10.00 ± 0.00	10.00 ± 0.00	10.20 ± 0.20	10.00 ± 0.00
Xylazine	10.20 ± 0.20	10.00 ± 0.00	11.00 ± 1.00	10.80 ± 0.80	10.60 ± 0.60	10.00 ± 0.00
Fentanyl	10.20 ± 0.20	—	10.40 ± 0.24	10.00 ± 0.00	10.00 ± 0.00	10.20 ± 0.20

since sedation will result in a total reduction of response rate without affecting lever selection. More likely, it seems that the food pellets have lost their reinforcing properties, resulting in a disruption of the response-reinforcement contingency. The shift in lever responding was clearly present within a test session. As time progressed, the percentage responses emitted on the alternative lever increased. Therefore, risperidone has an interference with the reinforcing stimulus control rather than with the discriminative stimulus control of responding. An analogous effect has already been described for the neuroleptic haloperidol in drug discrimination procedures in the rat [Colpaert et al., 1978a,b]. The disruption of the response-reinforcement contingency observed with haloperidol and risperidone might be due to an increase in reinforcement threshold. Stimulantia such as d-amphetamine and cocaine, which lead to a dopamine overstimulation, are reported to decrease the reinforcement threshold [Koob and Bloom, 1988]. One might suggest that compounds with a well-pronounced dopamine antagonism would introduce an opposite effect.

In terms of response rate in the generalization experiments, ritanserin was without any effect on responding in the different generalization experiments up to 40.00 mg/kg. At 40 mg/kg, only in two out of the seven generalization tests was a small reduction in response rate measured. Therefore, it might be concluded that at doses up to 40.00 mg/kg, ritanserin possesses no intrinsic disruptive effects on responding in a food-reinforced conditioned behaviour. Risperidone, on the contrary, started to reduce response rate at a dose of 0.16 mg/kg. At 0.63 mg/kg risperidone, a significant reduction was present in all experiments on stimulus generalization. Therefore, these results indicate that even at doses that almost completely disrupt response rate, risperidone revealed no stimulus generalization at all with any of the training drugs.

In terms of an antagonism of the discriminative stimulus properties of the different training drugs, ritanserin had no effects on 8-OHPAT, d-amphetamine, cocaine, chlordiazepoxide, xylazine, and on fentanyl. In any animal tested at different doses of ritanserin, no antagonism of the discriminative stimulus properties of one of these training drugs was observed. For the LSD-cue, a dose-related antagonism was observed, with a complete blockade at 40.00 mg/kg ritanserin. The way of responding of the ritanserin-treated rats in the different antagonism studies was nearly perfect since most FRF-values were <11.00 and the percentage responding on the selected lever was $>97.00\%$. As was observed in the generalization experiments, ritanserin had no intrinsic effects on response rate except in the experiments on xylazine-trained rats. When given in combination with xylazine, ritanserin started to reduce


LSD
8-OHDPAT
d-amphetamine
cocaine
% 100
90
80
70
60
50
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veh
0.0025 0.01 0.04 0.16 0.63
0.01 0.04 0.16 0.63
0.04 0.16 0.63 mg/kg
chlordiazepoxide
xylazine
fentanyl
0.01 0.04 0.16 0.63 mg/kg

TABLE 4. Percentage Responding on the Selected Lever (Upper Panel) and FRF-Values (Lower Panel) of Rats Treated With Risperidone in the Generalization Experiments (See Also Table 2)

Generalization compound tested	Vehicle	Dose of risperidone (in mg/kg)				
		0.0025	0.01	0.04	0.16	0.63
LSD	99.97 ± 0.03	100.0 ± 0.00	99.90 ± 0.05	99.92 ± 0.04	99.05 ± 0.90	79.26 ± 8.15
8-OHDPAT	99.94 ± 0.06	—	99.70 ± 0.25	99.53 ± 0.44	95.19 ± 2.84	87.93 ± 5.55*
d-amphetamine	99.98 ± 0.02	—	99.99 ± 0.01	98.86 ± 1.10	99.86 ± 0.09	89.08 ± 4.83*
Cocaine	99.96 ± 0.04	—	—	100.00 ± 0.00	99.42 ± 0.24	77.70 ± 3.50*
Chlordiazepoxide	99.32 ± 0.02	99.89 ± 0.05	99.85 ± 0.09	99.96 ± 0.02	97.76 ± 1.28	89.39 ± 6.04
Xylazine	99.98 ± 0.02	—	99.95 ± 0.02	99.70 ± 0.14	99.21 ± 0.46	84.94 ± 4.18*
Fentanyl	99.45 ± 0.48	—	99.80 ± 0.14	99.85 ± 0.08	99.44 ± 0.34	90.91 ± 3.97*
LSD	10.20 ± 0.20	10.00 ± 0.00	10.40 ± 0.24	10.80 ± 0.37	10.40 ± 0.24	10.20 ± 0.20
8-OHDPAT	10.00 ± 0.00	—	10.60 ± 0.40	10.00 ± 0.00	10.40 ± 0.24	10.80 ± 0.80
d-amphetamine	10.40 ± 0.40	—	10.00 ± 0.00	10.00 ± 0.00	10.00 ± 0.00	10.20 ± 0.20
Cocaine	10.00 ± 0.00	—	—	10.00 ± 0.00	10.00 ± 0.00	10.00 ± 0.00
Chlordiazepoxide	10.60 ± 0.24	11.00 ± 0.63	11.00 ± 0.55	10.40 ± 0.24	10.80 ± 0.20	11.00 ± 0.77
Xylazine	10.20 ± 0.20	—	10.40 ± 0.24	11.40 ± 0.40	10.40 ± 0.40	10.80 ± 0.49
Fentanyl	10.20 ± 0.20	—	10.80 ± 0.37	10.60 ± 0.40	10.00 ± 0.00	10.00 ± 0.00

response rate in a dose-related manner at a dose of 2.50 mg/kg. It thus seems that a serotonin 5-HT_2 antagonism combined with an α_2-adrenoceptor agonism clearly affects rate of conditioned responding.

Risperidone antagonized the discriminative stimulus properties of 0.16 mg/kg LSD in a dose-related manner, reaching a complete antagonism of LSD with 0.63 mg/kg risperidone after a pretreatment period of both 1 and 4 hours. Risperidone was without any effect on 2.50 mg/kg xylazine, and only one out of five rats revealed an antagonism of 2.50 mg/kg 8-OHDPAT at 0.63 mg/kg risperidone, the highest dose tested. Risperidone, furthermore, antagonized the discriminative stimulus properties of 1.25 mg/kg d-amphetamine. A complete antagonism of d-amphetamine was found with 1.25 mg/kg risperidone, given 2 hours before testing. Risperidone was unable to block the discriminative stimulus properties of 10.00 mg/kg cocaine after 1 hour pretreatment and reached only a 40% antagonism with 2.50 mg/kg risperidone after 2 hours. For fentanyl, a 60% antagonism was observed at 0.63 mg/kg risperidone after a 1 hour pretreatment. However, at this dose, pronounced rate-depressive effects were present. Furthermore, after 2 hours of pretreatment and in spite of strong rate-reducing effects, no antagonism of the fentanyl cue was observed with doses of risperidone $\leq$ 2.50 mg/kg. Therefore, the antagonism of fentanyl with risperidone is very time limited. The inability of risperidone to interact with a narcotic analgesic mechanism was also demonstrated by the lack of a risperidone effect in the tail withdrawal test procedure (TWR) by itself [Janssen et al., 1988] or in combination with fentanyl, where risperidone was unable to decrease or increase the analgetic activity of fentanyl (personal observations). Thus, some minor differences seem to exist between the interactions of risperidone with the analgesic properties of fentanyl, as measured in the TWR, and with the narcotic cuing effects of this drug in the drug discrimination test procedure.

In almost all antagonism studies with risperidone, FRF-values $\leq$ 11.00 were observed, confirming the accuracy of the lever selection. Also, in terms of the percentage responding on

Fig. 4. Effects of risperidone in the different generalization experiments. See also legend to Figure 2.

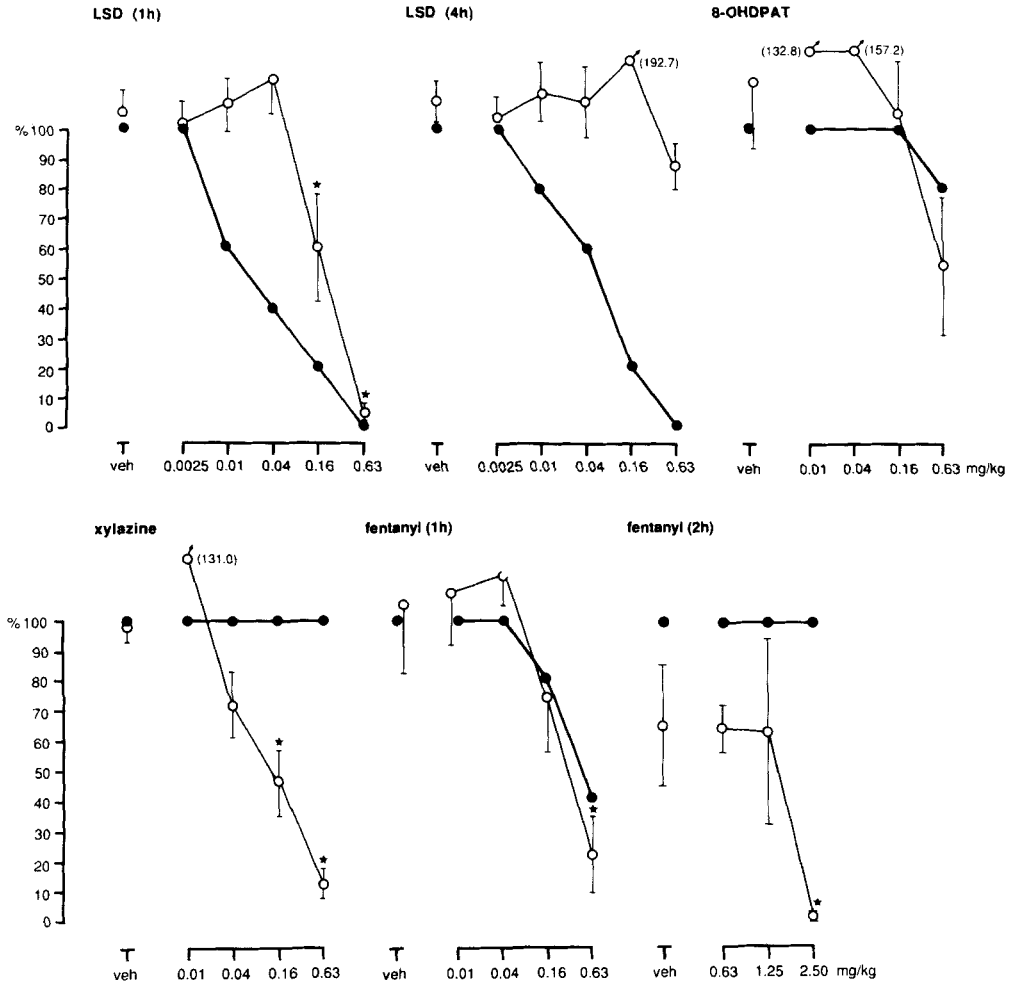

Fig. 5. Effects of risperidone in different antagonism experiments. The preinjection times of risperidone are given between dots. See also legend to Figure 3.

the selected lever, values mostly > 97.00% were found except at the highest doses tested. At these doses, as was observed in the studies on stimulus generalization, risperidone-treated animals started to respond on the non-reinforced alternative lever. As a consequence, there was a drop in the average percentage responding on the selected lever. Rate reducing effects were measured on LSD, xylazine, and cocaine (1 hour) antagonism starting from the dose of 0.16 mg/kg. In the fentanyl experiment and the cocaine antagonism after 2 hours, doses between 0.63 and 2.50 mg/kg risperidone started to decrease response rate. At doses $\geq$ 0.63 mg/kg risperidone, no rate reducing effects were observed during the experiments on the antagonism of 8-OHDPAT, d-amphetamine, and LSD after 4 hours, although for 8-OHDPAT there was a clear tendency for rate reduction. It thus seems that some compounds, and especially d-amphetamine, can overcome the rate-reducing effects of risperidone normally observed after 1 and 2 hours pretreatment. Furthermore, as demonstrated in the LSD experiment after 4 hours, the rate-reducing effects of risperidone seem to disappear over time, leaving the LSD antagonist properties intact [Meert et al., 1989].

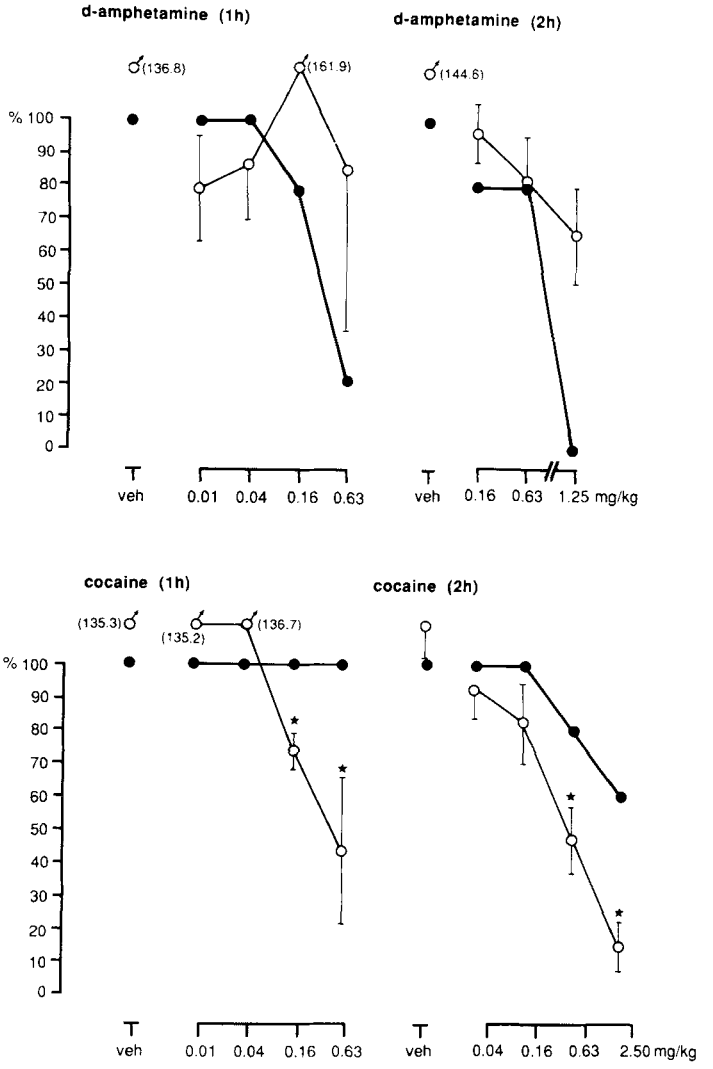

Fig. 6. Effects of risperidone on the antagonism of the d-amphetamine and cocaine cue. The preinjection times of risperidone are given between dots. See also legend to Figure 3.

The results obtained here with ritanserin in the drug discrimination experiments confirm earlier reports indicating ritanserin to be a pure, but relatively weak LSD-antagonist [Colpaert et al., 1985]. Because the discriminative stimulus properties of 0.16 mg/kg LSD in the rat are, besides a catecholaminergic involvement, primarily 5-HT_2 mediated [Meert et al., 1989; Colpaert et al., 1985], and because ritanserin in this study did not interact with compounds acting at serotonergic 5-HT_{1A}, at dopaminergic, at benzodiazepine, at α_2-adrenoceptor, and at opiate systems, these results confirm earlier in vivo data indicating ritanserin to be a selective 5-HT_2-antagonist [Awouters et al., 1988; Meert et al., 1987, 1988]. The results on response rate also confirm ritanserin to be free of sedative and motor disruptive effects and on conditioned responses at doses up to 40.00 mg/kg [Meert and Janssen, 1989; Awouters et al., 1988]. Risperidone was observed to be a potent and pure LSD and amphetamine antagonist at doses

TABLE 5. Percentage Responding on the Selected Lever (Upper Panel) and FRF-Values (Lower Panel) of Rats Treated With Risperidone in the Antagonism Experiments (See Also Table 2)

Antagonism compound tested	Vehicle	Dose of risperidone (in mg/kg) 0.0025	0.01	0.04	0.16	0.63
LSD (1 h)	99.74 ± 0.24	99.67 ± 0.29	99.95 ± 0.03	99.03 ± 0.84	89.01 ± 4.63	85.91 ± 13.11
LSD (4 h)	99.97 ± 0.02	99.78 ± 0.19	99.93 ± 0.05	99.98 ± 0.01	99.10 ± 0.70	99.31 ± 0.36
8-OHDPAT (1 h)	93.72 ± 5.75	—	98.56 ± 1.34	95.65 ± 2.64	94.34 ± 5.26	83.31 ± 15.65
d-amphetamine (1 h)	99.41 ± 0.39	—	99.87 ± 0.06	98.93 ± 0.61	95.11 ± 4.64	98.39 ± 0.90
Cocaine (1 h)	100.00 ± 0.00	—	100.00 ± 0.00	98.48 ± 1.52	99.84 ± 0.10	99.77 ± 0.19
Xylazine (1 h)	98.22 ± 1.32	—	97.74 ± 1.48	97.31 ± 1.45	98.88 ± 0.97	99.97 ± 0.03
Fentanyl (1 h)	94.29 ± 3.77	—	99.71 ± 0.18	94.92 ± 4.24	86.04 ± 9.25	69.39 ± 8.14
LSD (1 h)	10.40 ± 0.24	10.20 ± 0.20	10.20 ± 0.20	10.80 ± 0.37	10.40 ± 0.24	12.50 ± 1.50
LSD (4 h)	10.20 ± 0.20	10.40 ± 0.24	10.40 ± 0.40	10.20 ± 0.20	10.20 ± 0.20	11.00 ± 0.55
8-OHDPAT (1 h)	10.67 ± 0.33	—	10.20 ± 0.20	11.00 ± 0.77	11.00 ± 0.55	11.25 ± 0.63
d-amphetamine (1 h)	10.40 ± 0.40	—	10.00 ± 0.00	10.00 ± 0.00	10.00 ± 0.00	10.00 ± 0.00
Cocaine (1 h)	10.00 ± 0.00	—	10.00 ± 0.00	10.00 ± 0.00	10.00 ± 0.00	10.00 ± 0.00
Xylazine (1 h)	10.60 ± 0.24	—	10.00 ± 0.00	10.20 ± 0.20	10.40 ± 0.24	10.67 ± 0.67
Fentanyl (1 h)	10.80 ± 0.58	—	10.20 ± 0.20	10.20 ± 0.20	10.00 ± 0.00	10.75 ± 0.75

TABLE 6. Percentage Responding on the Selected Lever (Upper Panel) and FRF-Values (Lower Panel) of Rats Treated With Risperidone in the Antagonism Experiments (See Also Table 2)

Antagonism compound tested	Vehicle	Dose of risperidone (in mg/kg) 0.04	0.16	0.63	1.25	2.50
d-amphetamine (2 h)	99.33 ± 0.41	—	95.71 ± 2.92	78.90 ± 9.36	89.40 ± 4.78*	—
Cocaine (2 h)	99.92 ± 0.05	99.71 ± 0.29	100.00 ± 0.00	99.25 ± 0.62	—	95.74 ± 2.60
Fentanyl (2 h)	99.98 ± 0.02	—	—	96.14 ± 2.11	89.28 ± 3.89	57.33 ± 21.62*
d-amphetamine (2 h)	10.40 ± 0.24	—	10.20 ± 0.20	10.80 ± 0.58	10.00 ± 0.00	—
Cocaine (2 h)	10.00 ± 0.00	10.00 ± 0.00	10.00 ± 0.00	10.00 ± 0.00	—	10.00 ± 0.00
Fentanyl (2 h)	10.20 ± 0.20	—	—	10.00 ± 0.00	10.00 ± 0.00	11.00 ± 1.00

that did not affect response rate. Because both 5-HT_2 and catecholaminergic mechanisms are involved in the discriminative stimulus properties of LSD [Meert et al., 1989; Colpaert et al., 1985] and because of the dopamine (especially D_2) involvement in the discriminative stimulus properties of d-amphetamine [Colpaert et al., 1978b, 1976b; Jarbe 1982; Nielsen and Jespen, 1985], the antagonism with risperidone of both the LSD and d-amphetamine cue and the lack of revealing any generalization to both training drugs confirms risperidone to be a potent 5-HT_2 and D_2 antagonist [Janssen et al., 1988; Megens et al., 1989]. The partial antagonism of the discriminative stimulus properties of cocaine with risperidone confirms earlier results indicating that even high doses of DA-antagonists are unable to completely block the 10.00 mg/kg cocaine cue [Cunningham and Appel, 1982; Colpaert, 1986]. These results thus confirm the idea that the discriminative stimulus properties of cocaine only partially depend upon a dopamine stimulatory activity [Colpaert, 1986; Colpaert et al., 1979, 1980; Snoddy and Tessel, 1983]. The inactivity of risperidone at doses up to 0.63 mg/kg on both the xylazine- and the 8-OHDPAT-cue indicates risperidone to have no strong in vivo interactions with α_2-adrenoceptor and serotonin 5-HT_{1A} mechanisms. Also on a central narcotic cue, there was no clearly pronounced effect.

Globally, the results presented here in the drug discrimination test procedure on LSD-, 8-OHDPAT-, d-amphetamine-, cocaine-, chlordiazepoxide-, xylazine-, and fentanyl-trained rats, confirm ritanserin to be a pure 5-HT_2 antagonist without having any intrinsic effect on food-reinforced conditioned responding. Risperidone is observed to be a potent and pure 5-HT_2 and dopamine antagonist with, especially at the highest doses, an impact on conditioned behavioral responding both in terms of reducing response rate and in terms of interference with the reinforcing stimulus control.

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