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Effects of Psychotropic Drugs on Rat Responding in an Operant Paradigm Involving Choice Between Delayed Reinforcers

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CHARRIER, D. AND M. H. THIÉBOT. *Effect of psychotropic drugs on rat responding in an operant paradigm involving choice between delayed reinforcers.* PHARMACOL BIOCHEM BEHAV **54**(1) 149–157, 1996. – Preference for immediate reward, taken as an index of impulsiveness, has been suggested to be under the preferential control of central serotonin (5-HT) function. The present study examined the effects of the acute administration of drugs which directly or indirectly alter 5-HT transmission on tolerance to delay of reward in rats subjected to a procedure of discrete-trial choice in an operant chamber. Different groups of rats were trained to choose between two levers giving access to reinforcers differing in both magnitude and delay: one food pellet, delayed by 0 or 5 s, vs. five pellets delivered after a prereinforcer interval fixed at either 15, 30, 45, or 60 s, depending on the experiments. The learning curves indicated that rats were able to adjust their choice strategy precisely according to various factors: the respective duration of the delays before the small and large rewards, the immediacy of the small reward delivery, and the lengthening of the trials by a postreinforcer delay (or intertrial interval). Whatever the experimental parameters and stage of the learning, an acute administration of drugs able to reduce 5-HT neuronal activity (benzodiazepines; 5-HT_{1A} receptor partial agonists: buspirone and MDL 73005EF) or enhance 5-HT transmission (5-HT reuptake inhibitors: indalpine and zimelidine; 5-HT_{1A} receptor full agonist: 8-OH-DPAT) failed significantly to alter choice strategy (decreased or increased preference for the large but delayed reward, respectively), as they did in other situations such as a T-maze procedure. Only *d*-amphetamine (0.5 mg/kg), on one occasion, significantly reduced preference for the larger reward. The choice strategy was also insensitive to acute changes in experimental parameters such as a reduction in delay or increase in the magnitude of the large reinforcement. These results indicate that the present operant paradigm is not sensitive to acute modifications in the internal state of the animals and in the reward contingencies, and therefore is not useful to evaluate tolerance to delay and variations in impulsiveness in rats.

5-HT_{1A} Receptor agonists Amphetamine Benzodiazepines Delay of reward Operant behaviour Rat Serotonin reuptake inhibitors Waiting capacity

NUMEROUS behavioural studies have provided evidence over the last 30 years that animals are sensitive to delay of reward. This applies to various species including rats, which are able to adjust choice strategies according to amounts and delays of reinforcement (1,17,24,25,27). More recently, it has been suggested that serotonergic mechanisms may play a significant role in the ability of animals to tolerate a delay before the delivery of an expected reward. Such tolerance to delay has been proposed as an index of self-control or impulsiveness (1,17,25). Evidence consistent with this suggestion has been obtained using a variety of experimental procedures involving delayed reinforcement. One of them consisted of discrete trials in a T-maze, where rats had to choose between immediate access to a small food reward and delayed access to a larger reward. The choice strategy of trained rats typically depended on the waiting period imposed in the arm giving access to the larger reward. A variety of compounds reducing 5-HT function—*para*-chlorophenylalanine (pCPA), which blocked 5-HT synthesis; 5-HT_{1A} receptor partial agonists; and benzodiazepines (which, among other neurobiologic effects, re-

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duced 5-HT neuronal activity)—shifted rats' choice toward the immediate reward. Serotonin reuptake inhibitors, on the contrary, increased the number of selections of the large but delayed reward (4,40,41). Another procedure, the "adjusting-delay" paradigm (27) was also designed to directly investigate the subject's choice between delayed reinforcers. The rat made repeated choices in a two-lever operant chamber between a small quantity of food delivered after a short delay and a larger reward delivered after a variable delay, the length of which was determined by the subject's previous choice. Consistent with the data reported above, the indifference point (the delay to the larger reward that rendered the two reinforcers equally effective) was shortened in rats subjected to lesion of 5-HT neurones in the raphe nuclei by the specific neurotoxin 5,7-dihydroxytryptamine (5,7-DHT) (45). However, no further pharmacologic investigations have been performed as yet. Taken together, these results suggest that 5-HT processes are important in maintaining the effectiveness of positive reinforcers whose delivery is delayed after the emission of the response.

Another type of timing procedure is exemplified by the interresponse time greater than t (IRT $> t$) schedules (previously known as differential reinforcement of low rate of responding, or DRL schedules), which require the animal to refrain from lever pressing for a specified period of time to receive reward. As distinct from the situations described above, which arrange a delay between the response and the reinforcement, in IRT $> t$ schedules, the behaviour itself constitutes the event to be timed. Despite this important difference, and the fact that they do not involve a choice strategy, IRT $> t$ procedures have been considered suitable to appreciate rats' capacities to wait, especially when the time requirement is long (31–33,37,46). Performance was improved by systemic administration of a variety of serotonin (5-HT) reuptake inhibitors: 5-hydroxytryptophan (a 5-HT precursor) and 5-HT_{1A} receptor full agonists such as 8-OH-DPAT and flesinoxan, which induced a coherent shift of the IRT distribution toward longer durations, thus reducing the premature nonrewarded responses (26,29,30,33,36,43). On the other hand, benzodiazepines, several 5-HT_{1A} receptor partial agonists, local infusions of 8-OH-DPAT into the median raphe nucleus, and 5,7-DHT lesions of ascending 5-HT pathways had the opposite effect and impaired IRT $> t$ performance by increasing premature burst responses or by disrupting the IRT distribution (12,30,35,46). All of these results indicate that tolerance to delay (i.e., self-control or impulsiveness) could be evaluated in animals and constituted a psychological dimension sensitive to pharmacologic manipulations of 5-HT transmission or lesions of 5-HT neurones.

The present study aimed to investigate whether similar behavioural and pharmacologic results could be obtained using another operant schedule expected to be less time- and animal-consuming, and to permit more extensive studies than would be possible in discrete-choice maze procedures and the adjusting delay operant schedule. Its major advantage would be to allow more precise insight into the neurobiologic mechanisms involved in self-control and impulsiveness and the effect of drugs thereon. For this purpose, rats were subjected, in an operant chamber, to a discrete-trial choice between two levers giving access to reinforcers differing in both magnitude and delay, according to a paradigm as close as possible to that found in the T-maze test (40). The sensitivity of the procedure to various psychotropic drugs was assessed by investigating the ability of selective 5-HT reuptake inhibitors, 5-HT_{1A} receptor ligands, and/or benzodiazepines to modify the rats' choice strategy acutely.

METHOD

Subjects

The experiments were carried out on male Wistar rats (Centre d'Elevage R. Janvier, Le Genest, France) weighing 120 ± 10 g at the beginning of the training and 200–250 g at the time of the drug sessions. They were housed eight per cage under standard laboratory conditions (12 L : 12 D cycle, room temperature of 21 ± 1°C) with water freely available in the home cage. Rats were maintained at 80–85% of their free feeding weight by a daily schedule of food restriction (13 g of standard chow per day per rat), established 1 week before the beginning of the training and maintained until the end of the experiments. Rats were subjected to daily saline injections (IP or SC) over a period of at least 1 week before receiving the drug under study.

Apparatus

The experiments were conducted in two standard ventilated, sound-attenuated operant chambers (Campden Instruments Ltd., Leicester, UK). Each chamber was fitted with an automatic magazine delivering food pellets (45 mg; Campden Instruments). A tray located between two motor-driven retractable levers was closed with a flap which the rat had to push with its nose to gain access to the pellets. The chambers were continuously illuminated (excepted where otherwise mentioned) with one house light (24 V, 3 W) located in the middle of the ceiling. The operant schedules were automatically controlled by electromechanical switching and timing devices (Campden Instruments) connected to a printer for automatic data collection.

Experimental Procedure

General training procedure. All experiments were conducted daily, 5 days a week. Rats were initially trained to press both levers to obtain food pellets according to a continuous schedule of reinforcement for five (20-min) daily sessions. Thereafter, they underwent pretraining sessions which consisted of 20 consecutive trials, which started with the insertion of the two levers into the chamber and ended 5 s after the delivery of the food pellets. A response on either lever resulted in the immediate withdrawal of the two levers. One lever (whose position, left or right, was counterbalanced across the rats) was always associated with the delivery of five pellets (larger reward), whereas the other one gave access to one pellet (smaller reward). Depending on the experiment, both reinforcers were delivered either immediately or after a 5-s delay. After four to five sessions, the choice strategy stabilised, and >80% of the responses were performed on the lever associated with the larger reward. Rats were then subjected to training sessions (20 trials), during which the responses on the lever associated with the larger reward initiated a long prereinforcer delay followed by the delivery of five pellets. The responses on the other lever had the same consequence as during the pretraining. Five seconds after food delivery, the next trial was initiated by the presentation of the two levers (Fig. 1).

Behavioural manipulations. We performed two experiments to assess whether an acute variation in some experimental parameters (shorter prereinforcer delay or increased magnitude of the larger reward) would result in a shift in rats' strategy of choice. For this purpose, two independent groups

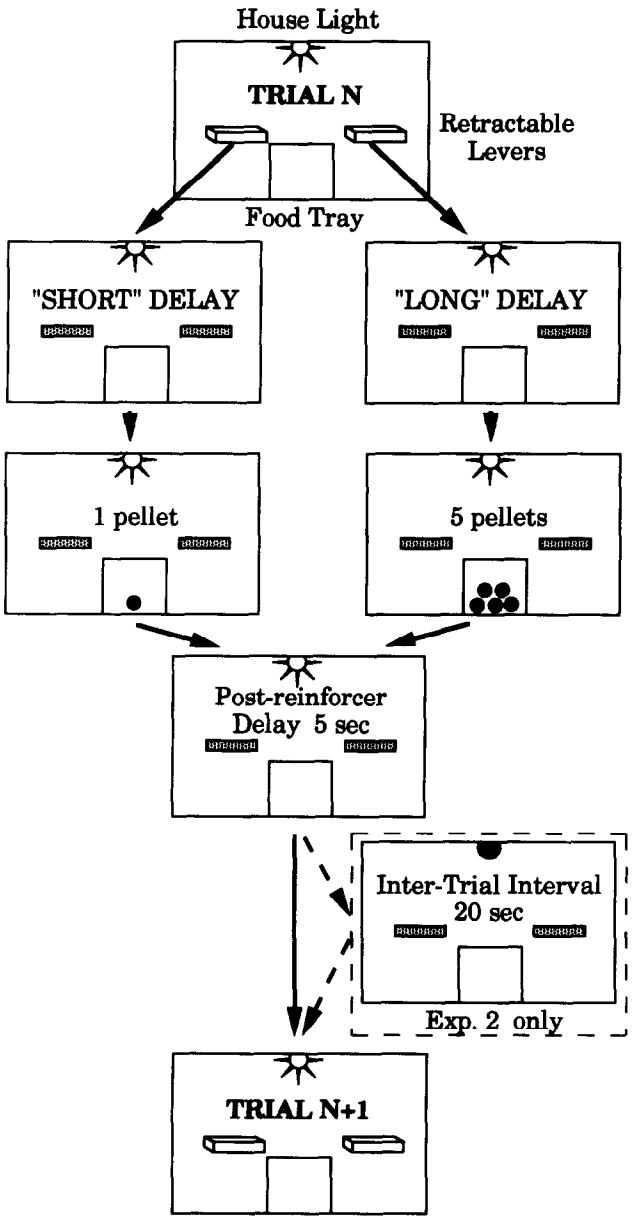

FIG. 1. Schematic diagram of the sequence of events that could occur on a choice trial, depending on which lever was pressed. Each box represents one stage within a trial. The short and long prereinforcer delays depended on the experiment: Experiment 1A: 0 s vs. 15 or 30 s; Experiment 1B: 5 s vs. 30, 45, or 60 s; Experiment 2: 5 s vs. 30 s with a 20-s intertrial interval (house light off).

of rats were trained to choose between five pellets delayed by either 30 s (group A) or 45 s (group B) and one pellet delayed by 5 s, as described previously. After 13 training sessions, the rats were subjected to a single test session. For rats of group A (divided into two subgroups), the long delay associated with the larger reward was reduced from 30 s to either 15 s (for one subgroup) or 5 s (for the other subgroup). For group B, the responses on the lever associated with the larger reward initiated the usual 45-s delay before the delivery of eight pellets (as distinct from five pellets during the training sessions). The parameters of food delivery associated with the other lever were not modified (one pellet after a 5-s delay). No drug was given before these test sessions.

Pharmacologic manipulations. The initial stages of the training were conducted as described earlier.

Experiment 1. During pretraining sessions, both reinforcers were delivered either immediately (Experiment 1A) or after a 5-s delay (Experiment 1B). During training sessions (20 trials), the responses on the lever associated with the larger reward initiated a prereinforcer delay fixed at 15, 30, or 45 s (depending on the group and on the experiment, 1A or 1B), followed by the delivery of five pellets. The responses on the other lever had the same consequence as during the pretraining [i.e., delivery of one pellet either immediately (Experiment 1A) or after a 5-s delay (Experiment 1B)]. Five seconds after food delivery, the next trial was initiated by the presentation of the two levers. The delays and the magnitude of the rewards were not subjected to further modification during training and drug sessions.

Drug sessions. The strategy of drug administration was chosen according to whether a drug-induced decrease or increase in tolerance to delay was expected. This was achieved in different groups of rats subjected to different contrasts in delay to reward and tested after a variable number of training sessions. For this purpose, rats were divided into groups matched according to the frequency of selection of the lever associated to the large but delayed reward during the last training session and were given the compound under study or its vehicle. Drug sessions lasted until 20 trials were completed or 30 min had elapsed, whichever was first. In fact, the second eventuality occurred with only a few rats. Animals were subjected to no more than four drug sessions, and drug treatments were administered at intervals of at least 7 days (for details, see Results).

Experiment 2. To assess whether clearly interspaced trials might overcome possible changes in postreinforcement pausing as a function of the reinforcement magnitude and/or improve the rats' ability to distinguish schedule contingencies, the procedure was modified by introducing an intertrial interval (ITI). For this purpose, rats trained as described before (Experiment 1B) were required to choose between levers associated with either five pellets delivered after a 30-s delay or one pellet delivered after 5 s, during 20-trial sessions. At the end of each trial (i.e., 5 s after food delivery), the house light was extinguished for a 20-s ITI (i.e., a postreinforcement delay), during which the levers remained retracted. At the end of the ITI, the next trial began with the illumination of the house light and the presentation of the levers. Drug sessions, whose internal organisation did not differ from the training sessions, were conducted as described before. They lasted until 20 trials were completed.

Experiment 3. The possibility of rapid perceptual learning in the course of the training procedure, resulting in a fixation of the choice strategy, was assessed in rats of an additional group, trained to choose between five pellets delayed by 60-s and one pellet after 5 s. After three training sessions, the rats were divided into two groups according to the frequency with which they chose the lever associated with the larger reward, either $>$ or $<$ 50% of the trials, during the last training session. According to the expected drug effects, rats of the first group ($>$50%) were given diazepam (2 mg/kg) or saline; rats of the second group ($<$50%) received indalpine (4 mg/kg) or saline before a single 20-trial drug-session performed 24 h later.

Drugs

d-Amphetamine-SO_4 (Sigma, St. Louis, MO), buspirone-HCl (Bristol Myers, Paris-La Défense, France), MDL 73005EF [8-(2-[2,3-dihydro-1,4-benzodioxin-2-yl-methyl-amino]ethyl)-8-azaspiro (4,5) decane-7,9-dione methyl sulfonate] (Merrell Dow, Strasbourg, France), 8-OH-DPAT-HBr [8-hydroxy-2-(di-*n*-propylamino)tetralin] (Research Biochemicals International, South Matick, MA), and zimelidine-2HCl × H_2O (Astra, Södertälje, Sweden) were dissolved in saline (NaCl 0.9%). Diazepam base (Hoffmann-La Roche, Basel, Switzerland) and indalpine base (Pharmuka, R.P.R.-Vitry, France) were suspended in acacia gum in saline. Drugs or their vehicle were administered IP (*d*-amphetamine, buspirone, diazepam, indalpine, and zimelidine) or SC (8-OH-DPAT and MDL 73005EF) in a volume of 0.5 ml/100 g body wt., 30 min before the drug session (60 min for 8-OH-DPAT and zimelidine). As appropriate, the doses are expressed as the salt or the base.

Statistical Analysis

For each rat, the number of choices of the lever associated with the large but delayed reward was plotted as a percentage of the total trials and the drug session duration was recorded. Statistical comparisons of the group means were performed using one-way analysis of variance (ANOVA). When appropriate, planned pairwise comparisons between treated and control groups were made using one-tailed Dunnett's *t*-test or unpaired Student's *t*-test. Within-group comparisons (training vs. test condition) were analysed using one-tailed paired Student's *t*-test.

RESULTS

Behavioural Manipulations

Rats of independent groups learned to choose between one pellet delivered after 5 s and five pellets delayed by either 30 s (groups A, $n = 16$ in each group) or 45 s (group B; $n = 30$). During the 13th training session, the frequency of choice of the large but delayed reward was 35–45%. The reduction of the delay before the delivery of the larger reward from 30 to 15 or 5 s (groups A) or the increase in magnitude of the larger reward from five to eight pellets (group B) did not significantly alter the choice strategy (paired Student's *t*-tests = 0.29, 1.37, and 1.60, respectively) (Fig. 2).

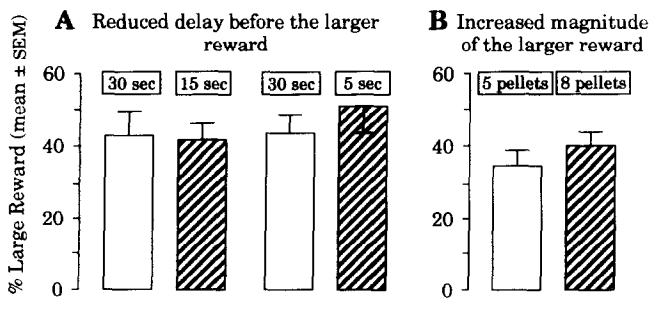

FIG. 2. Effects of an acute change in reward contingencies on the frequency of choice of the lever associated with the large but delayed reward, in independent groups of rats daily subjected to 20-trial sessions. (A) Decrease from 30 to 15 or 5 s of the delay before the large reward ($n = 16$ in each group). (B) Increase from five to eight pellets of the large reward magnitude ($n = 30$). (□) last training session; (▨) test session.

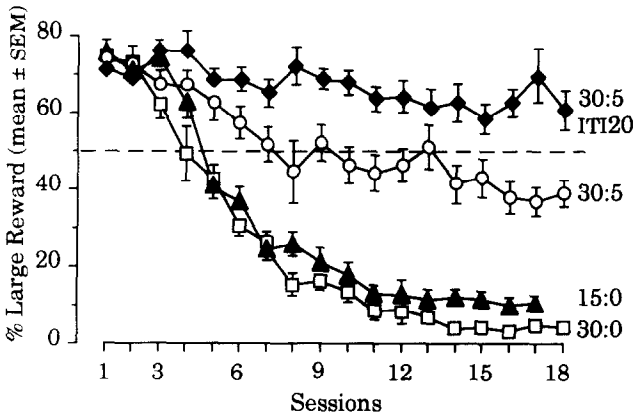

FIG. 3. Examples of the evolution of the frequency of choice (mean % ± SEM) of the lever associated to the large but delayed reward in four independent groups of rats daily subjected to 20-trial sessions. ▲-▲, choice between five pellets after 15 s and one pellet immediately (15 : 0); □-□, choice between five pellets after 30 s and one pellet immediately (30 : 0); ○-○, choice between five pellets after 30 s and one pellet after 5 s (30 : 5); ◆-◆, choice between five pellets after 30-s delay and one pellet after 5 s; trials included a 20-s intertrial interval (30 : 5; ITI 20) (n = 30 to 32 rats per group).

Pharmacologic Manipulations

Experiment 1A: choice between five pellets after a 15- or 30-s delay and one pellet immediately; no ITI. When the delay was fixed at 30 s, the frequency of choice of the large but delayed reward was 70–80% during the first session and progressively decreased to reach 4–8% from the 12th session onward. When the delay was fixed at 15 s, the preference for the larger reward progressively decreased from 70%–80% to about 12% during the 12th session (Fig. 3).

Diazepam (2–4 mg/kg) administered before the fourth session after the introduction of the delay, when the frequency of choice of the large but delayed reward was still relatively high under control conditions, did not significantly modify the choice strategy whether the delay was fixed at 15 s [$F(2, 21) = 1.13$] or 30 s [$F(2, 21) = 0.99$] (Table 1). The drug session durations were 9–11 min (delay 15 s) and 12–15 min (delay 30 s).

Indalpine, administered before the 12th session, when the frequency of choice of the large but delayed reward was low, did not significantly alter the choice strategy, whether the delay was fixed at 15 s [indalpine 0.5–4 mg/kg: $F(4, 52) = 0.41$] or 30 s [indalpine 2–4 mg/kg: $F(2, 24) = 0.12$] (Table 1). The drug session duration was 5–6 min, whatever the delay.

Experiment 1B: choice between five pellets after a 30- or 45-s delay and one pellet after 5 s, no ITI. When the contrast between the reinforcers was one pellet delayed by 5 s vs. five pellets delivered after 30 s, the frequency of choice of the larger reward was 70–80% during the first session and slowly decreased to reach about 40–50% from the eighth session onward (Fig. 3). When the long delay was fixed at 45 s, the preference for the larger reward progressively decreased from 65% during the first session to 20% from the 14th session onward (not shown).

Diazepam (2–4 mg/kg), administered when the rats still preferred the large but delayed reward (i.e., before the fourth session after the introduction of the long delay), did not significantly modify the choice strategy whether the large reward

TABLE 1

CHOICE BETWEEN ONE PELLET IMMEDIATELY AND FIVE PELLETS AFTER 15 OR 30 s

	mg/kg	*n*	Delay Before Larger Reward	No. of Sessions*	% Choice of Larger Reward
Diazepam	0	10	15 s	4	62 ± 3
(30 min, IP)	2	8			72 ± 4
	4	8			63 ± 7
Indalpine	0	19	15 s	12	15 ± 3
(30 min, IP)	0.5	10			11 ± 4
	1	10			9 ± 3
	2	9			12 ± 5
	4	9			12 ± 4
Diazepam	0	8	30 s	4	49 ± 10
(30 min, IP)	2	8			56 ± 7
	4	8			48 ± 5
Indalpine	0	9	30 s	12	8 ± 3
(30 min, IP)	2	9			8 ± 2
	4	9			7 ± 3

Data represent effects of diazepam and indalpine on the frequency (mean % ± SEM) of choice of the large but delayed reward, during a 20-trial session.

*Drug (or vehicle) injections were performed before the *N*th session after the introduction of the delay associated with the larger reward.

was delivered after 30 s [$F(2, 21) = 0.04$] or 45 s [$F(2, 24) = 0.00$] (Table 2). The drug session duration ranged from 18 to 28 min (delay 30 s) and 17 to 20 min (delay 45 s).

Zimelidine (8–16 mg/kg) administered before the 14th session after the introduction of a 45-s delay, when the frequency of choice of the large but delayed reward was low, did not significantly change the choice strategy [$F(2, 26) = 0.96$] (Table 2). The drug session duration was 9–11 min.

8-OH-DPAT (0.06–0.5 mg/kg), injected before the eighth session after the introduction of a 45-s delay, and MDL 73005EF (1–2 mg/kg), given before the 18th session after the introduction of a 30-s delay, when the large but delayed reward was chosen in only 20–25% of the trials, failed to modify the frequency with which rats chose the large but delayed reward [$F(3, 42) = 0.23$, and $F(2, 18) = 1.05$, respectively] (Table 2). The drug session duration ranged from 11 to 15 min (experiment with 8-OH-DPAT) and 12 to 17 min (experiment with MDL 73005EF).

d-Amphetamine (0.25–1 mg/kg) was administered at different stages of the evolution of the choice strategy after the introduction of a 30-s delay (i.e., before the fourth, eighth, 12th, or 16th session). Only *d*-amphetamine given before the 12th session induced a significant modification of the choice strategy [$F(2, 23) = 3.50$; $p < 0.05$]. This effect was due to a significant reduction of the preference for the lever associated with the larger reward by the dose of 0.5 mg/kg (Dunnett's t-test = 2.39; $p < 0.05$). No significant variation of the choice strategy was observed during the other test sessions [fourth session: $F(2, 21) = 0.74$; eighth session: $F(2, 25) = 0.10$; 16th session: $F(2, 21) = 0.44$] (Table 2). Whatever the stage of training and dose studied, the drug session duration ranged from 11 to 16 min.

Experiment 2: choice between five pellets after a 30-s delay and one pellet after 5 s, 20-s ITI. Only one group of rats was subjected to this experimental schedule. Under control conditions, the frequency of choice of the large reward delayed by 30 s was 74% during the first training session and slowly decreased to about 60% (Fig. 3).

Diazepam (2–4 mg/kg) injected before the fourth and eighth sessions did not significantly alter the preference for the large but delayed reward [$F(2, 20) = 0.21$, and $F(2, 21) = 0.73$, respectively) (Table 3). The drug session duration ranged from 21 to 29 min.

Buspirone (1–2 mg/kg) administered before the 14th session did not significantly modify rats' choice strategy [$F(2, 22) = 2.09$] (Table 3). The drug session duration was 20–26 min.

Experiment 3. Rats trained to choose between one pellet after 5 s and five pellets after 60 s, and selected for high (>50%) or low (<50%) frequency of choice of the larger reward during the third session were given diazepam (2 mg/kg) or indalpine (4 mg/kg), respectively, before the fourth session. Neither treatment significantly altered the choice strategy (unpaired Student's t-test = 1.48 and 1.05, respectively) (Table 4). The drug session duration ranged from 22 to 23 min (experiment with diazepam) and 16 to 18 min (experiment with indalpine).

DISCUSSION

The present study aimed to investigate whether tolerance to delay of reward in rats subjected to discrete-trial choice in an operant procedure was sensitive to the acute administration of various psychotropic drugs, as was the case in the T-maze waiting paradigm (4,40). For this purpose, rats were given the choice between two reinforcers differing in magnitude and delay, associated with two levers of an operant chamber. As previously demonstrated by numerous studies (1,15,17,24, 25,27), rats were able to progressively adjust their choice strategy according to various factors. They exhibited a clear-cut preference for the larger reward when delays of reinforcement were equal. When prereinforcer delays differed, the preference varied according to their respective durations, the immediacy of the small reward delivery (which reduced tolerance to delay), the stage of learning, and also whether trials were lengthened by a postreinforcer delay [i.e., an ITI (which enhanced tolerance to delay)].

TABLE 2

CHOICE BETWEEN ONE PELLET AFTER 5 s AND FIVE PELLETS AFTER 30 OR 45 s

	mg/kg	*n*	Delay Before Larger Reward	No. of Sessions*	% Choice of Larger Reward
Diazepam	0	8	30 s	4	68 ± 5
(30 min, IP)	2	8			69 ± 5
	4	8			69 ± 7
Amphetamine	0	8	30 s	4	80 ± 6
(30 min, IP)	0.25	8			69 ± 9
	0.5	8			69 ± 7
	0	10	30 s	8	57 ± 7
	0.5	9			52 ± 7
	1	9			54 ± 10
	0	9	30 s	12	53 ± 5
	0.5	8			26 ± 8†
	1	9			30 ± 10
	0	8	30 s	16	37 ± 7
	0.5	8			27 ± 9
	1	8			29 ± 8
MDL 73005EF	0	7	30 s	18	20 ± 4
(30 min, SC)	1	7			32 ± 8
	2	7			27 ± 5
Diazepam	0	9	45 s	4	56 ± 8
(30 min, IP)	2	9			56 ± 6
	4	9			56 ± 8
8-OH-DPAT	0	16	45 s	8	25 ± 5
(60 min, SC)	0.06	6			28 ± 6
	0.125	15			21 ± 5
	0.5	9			23 ± 9
Zimelidine	0	10	45 s	14	19 ± 4
(60 min, IP)	8	10			13 ± 3
	16	9			14 ± 4

Data represent effects of diazepam, *d*-amphetamine, MDL 73005EF, 8-OH-DPAT, and zimelidine on the frequency (mean % ± SEM) of choice of the large but delayed reward, during a 20-trial session.

*Drug (or vehicle) injections were performed before the *N*th session after the introduction of the delay associated with the larger reward.

†$p < 0.05$ vs. associated control group (Dunnett's *t*-test after ANOVA).

According to the serotonergic hypothesis of impulse control (20,22,34,37,44) and to previous experimental results obtained in the T-maze test (39,40) and the IRT > *t* procedures (11,12,26,29,35,36,46), drugs which directly or indirectly increase brain 5-HT transmission were expected to enhance preference for the large but delayed reward. Conversely, compounds reducing 5-HT function should lessen tolerance to delay. In fact, in the present paradigm, whatever the experimental parameters, neither the specific 5-HT reuptake inhibitors indalpine and zimelidine nor benzodiazepines increased or decreased preference for the large but delayed reward, respectively, in the course of a single session. The 5-HT_{1A} receptor ligands studied at doses effective in the T-maze test [(41) and unpublished data)] also failed to modify rats' choice strategy whether they were full (8-OH-DPAT) or partial (buspirone and MDL 73005EF) agonists (6,13,42). Accordingly, citalopram and imipramine (specific 5-HT and nonspecific NA/5-HT reuptake inhibitors, respectively) were recently reported not to affect the choice strategy of rats subjected to an operant schedule very similar to the present one (8). Diazepam was also found to be ineffective in reducing rats' tolerance to reward delay in a decision-making procedure which mixed operant responding and maze running (23). Therefore, no evidence was obtained for the present operant schedule to detect the consequences of drug-induced variations in 5-HT function on tolerance to delay in rats. A major factor which may be responsible for such results may relate to the fact that none of the drugs tested induced obvious specific and global changes in 5-HT function. Benzodiazepines may reduce 5-HT neuronal activity, but this is neither their only nor their prominent action (16). On acute administration, the ability of 5-HT uptake inhibitors to enhance extracellular concentrations of 5-HT in projection areas seems to be considerably less than initially assumed (2,18). Finally, it cannot be established whether direct postsynaptic effects or indirect autoreceptor-mediated negative feedback control of 5-HT neurones are most involved in the action of agonists and partial agonists of 5-HT_{1A} receptors [e.g., (11)]. This does not signify that choice strategy in

TABLE 3

CHOICE BETWEEN ONE PELLET AFTER 5 s AND FIVE PELLETS AFTER 30 s DURING 20 TRIALS INTERSPACED BY A 20-s INTERTRIAL INTERVAL

	mg/kg	*n*	Delay Before Larger Reward	No. of Sessions*	% Choice of Larger Reward
Diazepam	0	8	30 s	4	78 ± 5
(30 min, IP)	2	8			74 ± 6
	4	7			80 ± 8
Diazepam	0	8	30 s	8	73 ± 5
(30 min, IP)	2	8			81 ± 5
	4	8			79 ± 5
Buspirone	0	9	30 s	14	62 ± 5
(30 min, IP)	1	8			79 ± 7
	2	8			65 ± 6

Data represent effects of diazepam and buspirone on the frequency (mean % ± SEM) of choice of the large but delayed reward.

*Drug (or vehicle) injections were performed before the *N*th session after the introduction of the delay associated with the larger reward.

operant schedules is insensitive to any drug- or lesion-induced modifications in the internal state of the animals. For instance, metergoline (a nonspecific 5-HT receptor antagonist) and diazepam modified response strategy (although unexpectedly by increasing selection of the delayed reward) (7,8). *d*-Amphetamine [at low doses which unlikely to affect 5-HT release (21)] biased choice toward immediate reward [(7), present study]. Lesions of raphe nuclei by 5,7-DHT shortened the indifference point in the adjusting delay paradigm used by Bradshaw and colleagues (45). Thus, the inability of the present schedule to detect drug-induced variations in tolerance to delay and impulse control cannot be generalized to any drug and operant procedure. It is possible that drug sessions must take place in a particular window of sensitivity, as is the case in the T-maze. In fact, preference was unaltered whatever the learning phase, even when animals were still adjusting to the reward contingencies, or in rats exhibiting immediate high or low tolerance to delay. This would indicate that the inability of the drugs to modify choice strategy is unlikely to be accounted for by rigid habits due to repeated training.

In the T-maze, rats usually shifted their choice strategy early during the task acquisition (3,4,40), whereas several sessions were necessary for animals to adjust their strategy to changes in reward contingencies in the present schedule, as in many other operant procedures (8,24,25). In this regard, it is striking to note that a single experimental session was not sufficient for the rats to shift their choice when the magnitude of large reward was enhanced or when the contrast in delays was reduced or even cancelled. This most likely indicates that animals needed more trials in the operant box than in the T-maze to realise that external contingencies had changed and to change their behaviour accordingly. Evenden and Robbins (9) reported data which could be relevant to this point. They showed that in response to a random reinforcement schedule, rats exhibited a win-stay strategy in a two-lever chamber, but a win-shift strategy in a Y-maze. This would imply that in operant procedures, once rewarded rats persevered in that choice, whereas in a maze they have a consistent tendency to sample both possibilities (i.e., to alternate), even when one or both choices have been rewarded. This might be linked to the fact that in operant boxes, there is usually a single source of food, whatever the chosen response lever, whereas changing the response in maze procedures results in sampling different foraging places, a strategy which is prevalent in rodents ethology [see (25)]. Thus, rats' behaviour may be more flexible in the T-maze (especially as both options were rewarded) than in operant chambers. Unbiasing choice toward no preference (i.e., spontaneous alternation) would constitute an alternative

TABLE 4

CHOICE BETWEEN ONE PELLET AFTER 5 s AND FIVE PELLETS AFTER 60 s

	mg/kg	*n*	Delay Before Larger Reward	No. of Sessions*	% Choice of Larger Reward
Diazepam	0	6	60 s	4	71 ± 5
(30 min, IP)	2	7			55 ± 9
Indalpine	0	9	60 s	4	36 ± 6
(30 min, IP)	4	9			27 ± 5

Data represent effects of diazepam or indalpine in rats selected for high (>50%) or low (<50%) frequency of choice of the large but delayed reward, respectively.

*Drug (or vehicle) injection was performed before the fourth session after the introduction of the 60-s delay associated with the larger reward.

mechanism to changes in tolerance to delay in accounting for the effects of drugs in the T-maze. This possibility remains to be tested directly. However, diazepam, indalpine, and zimelidine did not alter rats' strategy under a no-delay condition, and drug effects depended on whether the delay of reward was short or long (4,40). In addition, spontaneous alternation in a Y-maze was reported to be either not modified or reduced by benzodiazepines and lesions of the median raphe (14,19,38). Therefore, a preferential action of drugs on win-shift strategy in the T-maze seems unlikely. Conversely, in operant procedures, a consistent tendency to perseverate in nonrewarded responding (lose-stay strategy) was reported in animals given benzodiazepines and 8-OH-DPAT (5,10), an effect that can offset the consequences of changes in tolerance to delay.

It is also possible that the low sensivity of rats' behaviour to acute variations in test parameters and in the drug state would be accounted for by hidden contingencies inherent to the present paradigm. In particular, it is possible that the delay to reward was not perceived as an uncontrollable negative event discounting the reinforcing value of the food. Indeed, while they were waiting for reward, the rats repeatedly pushed open the flap in front of the tray, and some of these responses were reinforced. Therefore, rats might behave as in chained schedules, in which a sequence of topographically different responses allows control over food delivery [see (28) for a review]. In addition, the click stimulus contingent to opening the tray might have acquired the value of a secondary reinforcer overshadowing the actual value of the primary reinforcers. To conclude, in the present operant schedule, choice strategy varied according to the amount and delay of reward, but no evidence has been obtained for its sensitivity to acute switches in pharmacologic states or reward contingencies. Therefore, the validity of tolerance to delay as an index of impulsivity—or at least to that aspect of impulsivity that is related to delayed gratification—can be questioned. However, since evidence on the sensitivity of tolerance to delay of reward to chronic treatments is scarce and conflicting [(3,46) and unpublished], and although we found a subacute administration (four daily injections) of either diazepam or indalpine to be ineffective (not shown), it remains to investigate the effects of chronic changes in 5-HT function in the present operant paradigm.

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