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The Effects of Quipazine and Fluoxetine on Extinction of a Previously-Reinforced Operant Response in Rats

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BENINGER, R. J. *The effects of quipazine and fluoxetine on extinction of a previously-reinforced operant response in rats.* PHARMACOL BIOCHEM BEHAV 21(4) 533–537, 1984.—Previous studies have shown that treatments that reduce serotonergic neurotransmission lead to enhanced responding during extinction. To evaluate the generality of this effect, the present study examined the effects of the serotonin agonists, quipazine and fluoxetine on responding in extinction. In Experiment 1, 72 rats were trained to lever press on a continuous reinforcement schedule for 5 30-min sessions. Four sessions of extinction followed; 30-min prior to each, 3 groups (n=16) received quipazine (0, 1.0, 5.0 mg/kg) and 3 groups (n=8) received fluoxetine (0, 1.0, 5.0 mg/kg). The 5.0 mg/kg dose of quipazine resulted in a significant reduction in responding on day 1; the lower dose of quipazine and both doses of fluoxetine were without significant effect. In Experiment 2, 3 similarly trained groups (n=8) received either saline or quipazine (5.0 mg/kg) prior to each extinction session; additionally, one quipazine group was injected twice with the 5.0 mg/kg dose in its home cage several days before the beginning of extinction. The results of the drug-naive quipazine group replicated those of that group from Experiment 1 whereas the drug-experienced group showed no significant effect of quipazine in extinction. The results suggested that prior drug experience could modify the effects of quipazine on behaviour. Apart from this drug novelty effect the lack of significant effect of either quipazine or fluoxetine suggested that the effects of manipulations believed to increase and decrease serotonin functioning on responding in extinction may not be symmetrical. These results may be understood with reference to the hypothesis that serotonin plays a role in tuning out or reducing responsiveness to nonreinforced or irrelevant stimuli.

Quipazine Fluoxetine Extinction Serotonin

SEROTONIN is a monoamine in the brain that has been localized in cell bodies of midbrain raphe nuclei that project extensively to forebrain regions [1]. There has been considerable interest in serotonin's possible roles in behavioral processes [16]. The results of some studies have led to the suggestion that serotonin may play a role in the "tuning-out" of nonreinforced or irrelevant stimuli [3,23].

The "tuning-out" hypothesis was based on the observation that animals treated with the serotonin synthesis inhibitor, parachlorophenylalanine (PCPA) failed to show latent inhibition [23]. PCPA treatments resulted in a slowing of habituation to an auditory stimulus [4,6] and an increase in "reactivity" to novel stimuli [6] providing further support for this point of view.

One procedure that would seem to provide a rigorous test of the tuning-out hypothesis is the withholding of reinforcement for a previously trained operant response. In this situation animals normally show an extinction curve, responding less and less with repeated exposures to stimuli previously associated with reinforcement. Consistent with this suggestion it was found that animals treated with PCPA had significantly reduced rates of extinction [3].

Many studies of serotonin function have utilized procedures that reduce brain levels of this monoamine [3, 4, 6, 16, 23]. The purpose of the present study was to investigate the effects of pharmacological agents thought to act as serotonin agonists. Thus, the effects on extinction of a previously-reinforced lever press operant, of the direct acting serotonin agonist, quipazine and the indirect acting (uptake blocker) agonist, fluoxetine [8] were tested in Experiment 1.

Quipazine was injected in doses of 1.0 and 5.0 mg/kg. Previous biochemical studies have shown that doses as low as 2.5 mg/kg produce significant reductions in brain levels of the serotonin metabolite, 5-hydroxyindoleacetic acid [10,12]. Behavioral studies have reported significant effects with doses ranging from 1.0 to 5.0 mg/kg [2, 5, 13]. The doses used here were selected to ensure both behaviorally and biochemically relevant doses. Note that these two doses of quipazine have been reported to be without significant effects on dopamine metabolites in the striatum [18,21]; the results of some drug discrimination studies, however, suggest the possibility that even low doses of quipazine may have a dopaminergic action [22].

Fluoxetine also was injected in doses of 1.0 and 5.0 mg/kg. Doses as low as 2.5 mg/kg have been reported to produce significant reductions in central 5-hydroxyindoleacetic acid levels [9,20] and doses ranging from 2.5 to 10.0 mg/kg recently were reported to impair rats' acquisition of a conditioned avoidance response [15]. Therefore, as was the case for quipazine, both behaviorally and biochemically relevant doses were assured by those selected.

According to the tuning-out hypothesis, these drugs might

result in less responding in extinction. Results revealed that quipazine produced a decrease in nonreinforced responding during the first test session whereas fluoxetine had no significant effect. Experiment 2 was carried out to test the hypothesis that the effects of quipazine were due to the novelty of the drug state; this variable has been shown by others to influence drug effects [11,14].

METHOD

Subjects

Ninety-six experimentally naive male Wistar albino rats weighing from 250 to 300 g were maintained at 80% of these free-feeding weights throughout the experiment by daily feeding with measured rations of standard laboratory chow. Rats were individually housed in stainless steel wire cages located in a climatically controlled colony room kept on a 12 hr light (0700–1900 hrs)/dark cycle. Water always was available in the home cage.

Apparatus

Three similar test chambers (23×20×19 cm high), constructed of Plexiglas sides and top, aluminum plate end walls and a grid floor, were outfitted with a lever (5.0 cm wide) located to the left of centre on one of the end walls at a height of 5.0 cm. A feeder cup was located to the right of the lever at a height of 2.5 cm. Each chamber was housed in a styrofoam-insulated laminated wooden box, ventilated by a small fan that also provided constant masking noise and illuminated by a 6 watt bulb. Environmental stimulus events and data collection were controlled by solid state switching and timing devices (BRS/LVE) located in an adjacent room.

Procedure

Experiment 1. The purpose of this experiment was to examine the effects of quipazine and fluoxetine on extinction of a previously reinforced operant response. Seventy-two rats were tested in three squads of 24, one squad completing testing before the next began. Each squad was tested three rats at a time, one in each chamber, at the same time each day. Lever press acquisition training was carried out over a 5 day period, each rat receiving 30 min of exposure to the chamber each day during which each lever press produced the delivery of one 45 mg food pellet (Bioserv No. 0021). Several food pellets also were placed in the feeder cup prior to these sessions and most rats learned to lever press. Those that did not underwent response shaping. During the next 5 days each rat received a daily 30-min session of training on continuous reinforcement (CRF); i.e., each lever press produced a food pellet. This was followed by two nontest days in the home cage and then 4 30-min test sessions with reinforcement no longer occurring (extinction).

Before the first extinction session, rats in each squad were randomly assigned to one of three drug dose groups (n=8). IP injections were made 30 min prior to each session. Groups in the first two squads received saline or quipazine maleate (Polysciences) in doses of 1.0 or 5.0 mg/kg and groups in the third received saline or fluoxetine (Lilly), also in doses of 1.0 and 5.0 mg/kg. The dependent variable was total number of responses per session.

Experiment 2. This study was done to test the possibility that the decrease in responding during the first extinction session observed in the rats receiving 5.0 mg/kg of quipazine was related to the novelty of the drug-produced stimuli. Twenty-four rats underwent lever press acquisition and CRF training as described above. Immediately following the fifth CRF session, the rats were assigned randomly to one of 3 groups (n=8). One group received two injections of quipazine (5.0 mg/kg) in their home cage whereas the remaining groups received saline. Injections occurred in the late afternoon following the fifth CRF session and the next morning.

Extinction sessions began 4 days following the last session of CRF. Thus, pretreatment injections of quipazine or saline occurred approximately 4 and 3 days prior to testing. One of the groups that received pretreatment with saline also received saline 30 min prior to extinction sessions. The remaining two groups received quipazine (5.0 mg/kg) before extinction; the 8 rats that received vehicle following CRF were drug naive whereas those previously receiving quipazine were drug experienced.

RESULTS

The distribution of respone rates in extinction tends to be positively skewed, a small number of rats showing extraordinarily high rates in one or more sessions. This phenomenon is illustrated in Fig. 1, which shows frequency distributions of response rates (responses per session) in class intervals of 100 for each group on each test day in Experiments 1 and 2. In the group receiving saline as a control for quipazine in Experiment 1, for example, one rat responded over 600 times in extinction session 2 and a different rat responded at this high rate in session 4; this rate was not produced by these or any other rats in this group on any test day. Because the assumptions of parametric statistics would be violated by these anomolies, Kruskal-Wallis analysis of variance by ranks and Mann Whitney U tests [7] were used for data analyses.

Experiment 1. The results of the first two squads receiving quipazine were combined and their median rates of responding per session for the last day of CRF (baseline) and for the 4 drug test days are shown in the upper panel of Fig. 2. Inspection of the baseline rates shows that the groups were similar and analysis revealed no significant differences ($p>0.05$). All groups underwent extinction, comparisons between baseline and test session 4 being significant in each case ($p<0.01$). Analysis of groups during test session 1 also revealed differences ($p<0.001$). The slightly higher rate of the 1.0 mg/kg group was not significant but the rate of the high dose group was significantly lower than that of the other two groups ($p<0.002$). There were no significant differences among groups on test days 2 or 4 but group differences were found on day 3 ($p<0.02$), the response rate of the 5.0 mg/kg group being marginally lower than that of the other 2 groups ($p<0.05$). Thus, the 1.0 mg/kg dose of quipazine had no significant effect on responding in extinction whereas the 5.0 mg/kg dose produced a dramatic decrease in responding on the first day, a small decrease on the third, and had little effect on the remaining 2 days.

The effects of fluoxetine on responding in extinction are shown in the middle panel of Fig. 2. As was the case with quipazine, there were no differences among groups in baseline rates and all groups underwent extinction ($p<0.01$). The group receiving 1.0 mg/kg of fluoxetine, like that quipazine dose, showed an insignificant increase in responding on day 1 but groups did not differ significantly on any test day.

Experiment 2. The results are shown in the lower panel of

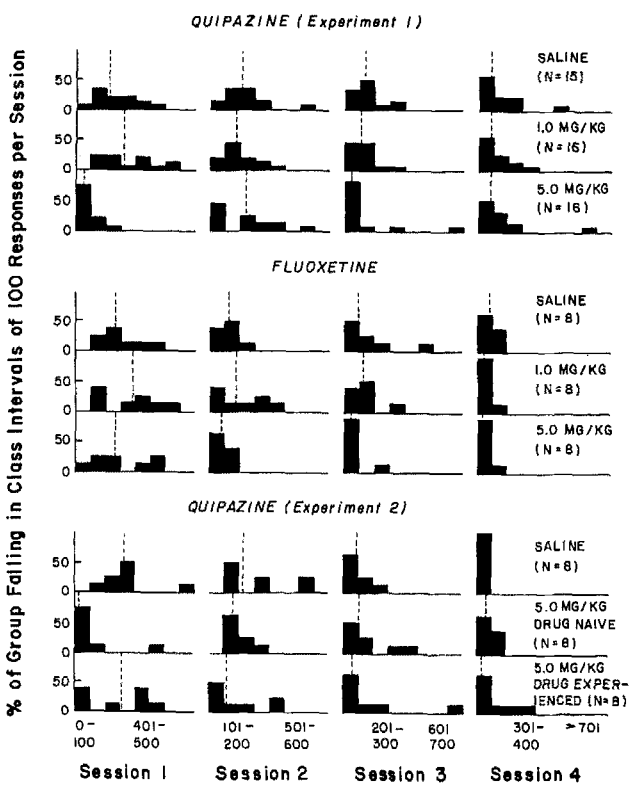

FIG. 1. Percent of animals from each group in Experiments 1 and 2 falling into class intervals of 100 responses per session during extinction sessions 1–4. The broken vertical line rising from each set of histograms shows the median value for that distribution. Drugs were administered in the doses indicated 30 min prior to each session.

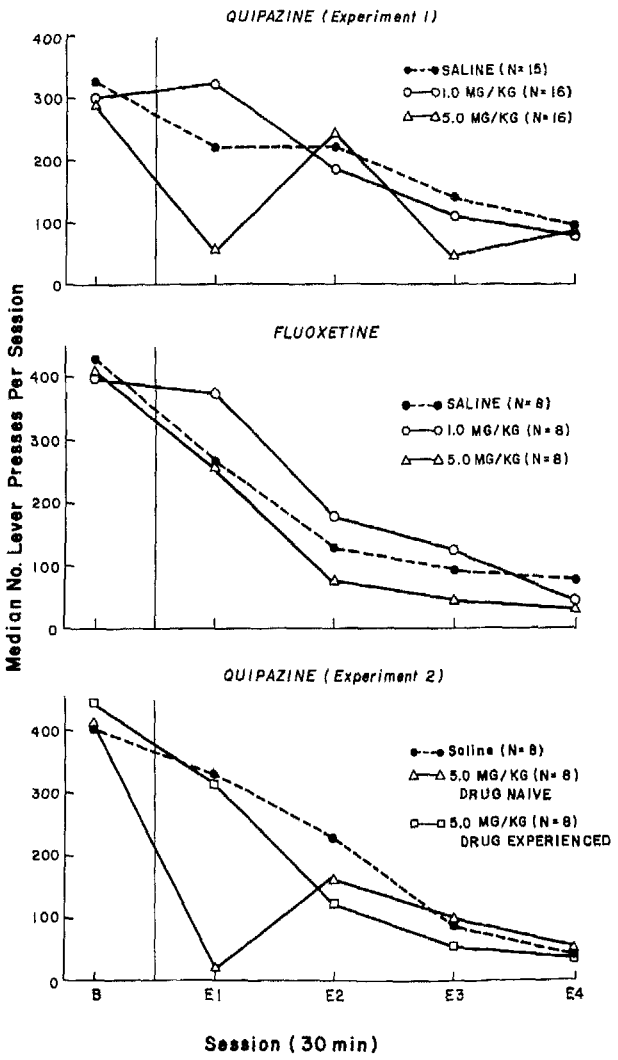

FIG. 2. Median number of lever presses for each group in Experiments 1 and 2 during the fifth training session of continuous reinforcement (B) and during the 4 extinction sessions (E1–4). Drugs were administered in the doses indicated 30 min prior to each extinction session.

Fig. 2. Baseline response rats did not differ significantly and all groups underwent extinction ($p<0.01$). Group differences during test session 1 were significant ($p<0.05$), the drug naive quipazine group responding significantly less than the control group ($p<0.01$), replicating the results for this group in Experiment 1. The rates of the drug experienced group receiving quipazine, on the other hand, were not significantly different from those of the saline group. Groups did not differ significantly on days 2–4. This, prior experience with quipazine (5.0 mg/kg) abolished the response decrease normally seen in quipazine-treated rats during the first extinction session.

DISCUSSION

The results showed that quipazine, but not fluoxetine, produced a decrease in responding on the first day of extinction of a previously reinforced operant response. This effect was not observed in animals receiving two home cage injections of quipazine three days prior to testing. The finding that prior experience modified the drug effect is consistent with previous reports for other compounds. Thus, it was found that the response depressant effects of the benzodiazepine, oxazepam were absent in animals with a previous history with that compound; interestingly, the antianxiety effect of oxazepam was unaffected by drug history of the same animals [14]. Others have shown that experience with drugs having an unrelated pharmacological action can modify drug effects on behaviour; thus the rate enhancing effects of pentobarbital were observed to be less in monkeys with a history of morphine use [11].

The drug-experience-produced reversal of the depressant effects of 5 mg/kg of quipazine on responding during day one of extinction might be related to state dependent learning effects [17], as quipazine has been found to produce a discriminable drug state [22]. However, both the drug-naive and drug-experienced groups in Experiment 2 underwent operant training while in a drug-free state and received quipazine for the first time in the experimental setting during extinction session one. The influence of the stimulus properties of the drug on responding, therefore, might be expected to be similar for each group. Possibly, experience with the drug resulted in a familiarization with the drug state leading to the observed group difference. As the baseline rates of the two groups did not differ significantly, possible rate-dependent effects [19] of quipazine probably did not produce the ob-

served differences in Experiment 2. Whether the drug-experience effect is attributable to the development of tolerance or other unknown variables cannot be determined from the present data. Nevertheless, the results show that some behavioural effects of quipazine are modified by drug history.

In a previous study it was found that quipazine (2.5 and 5.0 mg/kg) produced no significant effect on locomotor activity of rats in a novel environment but caused an elevation of activity in a familiar environment [2]. In that study, none of the animals had experience with the drug prior to testing. Others have found significant behavioural effects of repeated injections of quipazine [5,13]. These findings would suggest that, at least in some test situations, the significant effects of quipazine cannot simply be attributed to the novelty of the drug state.

Apart from the drug novelty effect, neither quipazine nor fluoxetine significantly affected responding in extinction. The results of previous studies have shown that manipulations that decrease serotonergic neurotransmission result in increased resistance to extinction [3]. These findings raise the possibility that the effects of serotonergic manipulations on extinction may not be bidirectional, a suggestion consistent with previous findings from studies of serotonin's behavioural function. For example, locomotor activity in a novel environment was observed to decrease in rats injected with the receptor blocker, metergoline but failed to change significantly in rats injected with the agonist, quipazine [2]; similarly, it has been found that treatments that reduce forebrain serotonin function have little significant effect on avoidance acquisition whereas those that enhance serotonin activity generally impair avoidance acquisition [15,16].

These nonsymmetrical effects of increased and decreased serotonin neurotransmission on behaviour may be understood with reference to the hypothesis that serotonin may play a role in tuning-out or reducing responsiveness to non-reinforced or irrelevant stimuli. This hypothesis might suggest that serotonin functioning would be high in situations involving novelty or extinction, where stimuli are being tuned-out; serotonin activity might be expected to be low in situations where new response learning was required, e.g., avoidance learning. Treatments that further increased serotonin transmission in novel or extinction situations may have been seen to produce little effect (the present results, [2]) because serotonin activity already was at a high level; on the other hand, decreased serotonin functioning might have been found to lead to decreased locomotor activity in a novel environment because of enhanced freezing [2] and to increased responding in extinction [3], in both cases because of a reduction of the usual tuning-out process. The opposite would be expected in avoidance learning; serotonin activity might be low because tuning-out is not occurring. Accordingly, manipulations that decrease serotonin might have little effect whereas manipulations that enhance serotonin might impair avoidance acquisition because inappropriate tuning-out would occur; as reviewed by Ögren [16], the effects of serotonin manipulations on avoidance learning appear to fit this scheme.

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