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Comparison of the Effects of Buspirone and Chlordiazepoxide on Successive Discrimination

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PANICKAR, K. S. AND N. McNAUGHTON. *Comparison of the effects of buspirone and chlordiazepoxide on successive discrimination.* PHARMACOL BIOCHEM BEHAV **39**(2) 275–278, 1991.—Buspirone is a novel anxiolytic which does not share the muscle relaxant, anticonvulsant and sedative properties of classical anxiolytics such as the benzodiazepines. It has variable effects in conflict tasks based on shock which normally show consistent effects with classical anxiolytics. The present experiment investigated the effects of buspirone on successive discrimination, a conflict task employing omission of reward rather than shock. Buspirone (3.3, 1.1 and 0.3 mg/kg, IP) and chlordiazepoxide (5 and 20 mg/kg, IP) were administered to separate groups of rats throughout acquisition of a visual successive discrimination. Chlordiazepoxide released nonrewarded responding in a dose-related fashion. The effects of buspirone were qualitatively similar in releasing response suppression but were both less in magnitude and less clearly related to dose. The experiment shows that the action of buspirone in successive discrimination tasks does not depend on the use of shock but, rather, appears to be a genuine failure to fully release behavioural inhibition.

Successive discrimination Buspirone Chlordiazepoxide Anxiolytic Nonreward

BUSPIRONE is a 5HT1A agonist (2, 4, 10, 22, 28) which is clinically effective as an anxiolytic but lacks the anticonvulsant, sedative and muscle relaxant properties of drugs such as the benzodiazepines (12, 32, 33). In conflict tasks based on punishment there are reports that buspirone has effects like those of classical anxiolytics (17, 23, 24). However, some studies employing punished lever-pressing in the rat have failed to get a strong effect with buspirone (1,7). Studies employing punished drinking have also shown that the effects of buspirone are not always like those of benzodiazepines (17, 31, 37). One possible reason for the variable effects reported is that, as well as reducing behavioural inhibition, buspirone interacts with the perceived intensity of shock or some other nonspecific effect of shock.

Anxiolytics, especially benzodiazepines, have been shown to increase responding suppressed by omission of reward in an identical manner to that suppressed by shock (27,30). We, therefore, decided to test the effect of buspirone on a successive discrimination task which is formally similar to the classical Geller-Seifter (9) assay for anxiolytics, but which employs nonreward rather than shock to inhibit responding.

In the Geller-Seifter schedule, rats are trained to lever press on a random interval schedule for reward, on which signalled intrusion periods are superimposed, where every lever press is both rewarded and shocked. The successive discrimination task we used is procedurally similar to Geller-Seifter—the only difference being the employment of nonreward rather than shock during the signalled intrusion periods. This removes the potentially confounding factor of changes in reaction to shock from the assessment of changes in behavioural inhibition. It should also be noted that response release from shock suppression could well be viewed as part of a general motoric disinhibition rather than reflecting a relief of anxiety (14).

Buspirone is also relatively ineffective in a variety of animal models of anxiety (20, 21, 23, 25). As an animal model of anxiety, successive discrimination is well grounded in psychological theory. Other models have been mainly developed for their sensitivity to benzodiazepine ligands (8) and hence might not tap the same underlying psychological processes. Thus the failure of buspirone to act like a classical anxiolytic could be due to an interaction with shock in the case of previous experiments with successive discrimination and to a lack of involvement of true anxiety in the other tests. If this were the case one would predict that buspirone would not have similar effects to classical anxiolytic drugs on responding suppressed by reward omission. Perhaps a more likely possibility is that the various animal models including Geller-Seifter assess approximately the same underlying processes. If this is the case buspirone should have the same effects on responding suppressed by reward omission as other anxiolytics.

METHOD

Subjects

Subjects were 30 experimentally naive Sprague-Dawley rats weighing between 350–450 g. All animals were housed 4 to a cage and maintained on a 23-hour food deprivation schedule. Water was available ad lib except in the testing chambers. Light was via an external window and the housing and laboratory

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temperature were maintained at 22°C.

Injections

All animals were injected intraperitoneally in a volume of 1 ml/kg body weight. Three different doses of buspirone HCl (3.3 mg, 1.1 mg, 0.3 mg/kg) and two doses of chlordiazepoxide (20 mg, 5 mg/kg) dissolved in 0.9% w/v saline were administered to separate experimental groups, while the control animals received an equivalent volume of saline. Since higher doses of buspirone (30 and 10 mg/kg) produced a total loss of responding in earlier operant experiments done in the laboratory, they were not included in this experiment. Testing commenced 20 minutes after injection.

Apparatus

Fourteen Campden Instrument operant boxes (24.5 × 22.5 × 23 cm) with grid floors were used to train and test all subjects. Each box was fitted with a food hopper and two retractable levers. For the present experiments only one of the retractable levers was extended into the chamber throughout the session. Illumination was provided by a 2.8 W houselight. The experiments were controlled and data collected by a BBC Microcomputer.

Pretraining

After 2 weeks of 23-hour food deprivation, the rats were magazine trained using a noncontingent Random Time 30 s (RT 30) schedule. On this schedule all intervals between 0 and 60 seconds had an equal probability of occurrence. The computer selected an interval using a random number generator and then delivered a 45 mg reward pellet (Camden Instruments). A new interval was then selected for the next delivery. The lever was retracted from the box throughout magazine training. All subjects received a single daily session for 2 days.

The RT 30 schedule was then discontinued and the retractable lever extended into the box. Food pellets were now available on a continuous reinforcement schedule contingent on lever pressing. On the first day, wet mash was smeared on the lever. Each session lasted for 30 minutes and all subjects were given one session per day for 6 days.

Successive Discrimination Training and Drug Treatment

On the basis of the total number of responses during the continuous reinforcement schedule, the rats were assigned to different dose groups (5 rats per group) in a fashion which also counterbalanced testing chamber and time of testing as far as possible. They were then placed on a Random Interval 30 s schedule (RI 30, as for RT 30 but with reward delivery contingent on lever pressing). These sessions lasted for 45 minutes and subjects were given a single daily session.

Successive Discrimination Experiment

The rats were placed on RI 30 for 9 days before any drugs were administered. On the 10th day each subject received drug at half the nominal dose for its group to allow a gradual transition from no-drug to drug state. From the next day onwards they received the full dose of the drug. By day 15 response rates had stabilized and so on the 16th day of RI 30 a visual stimulus (three 2.8 W lights) was superimposed on the RI 30, with the light coming on for 60 seconds 9 times during the 45-minute session. Subjects were tested for 14 days after the inclusion of the visual stimulus before the nonreward contingency was introduced. This was to ensure that there were no effects of the stimulus itself on responding. On day 30 and for the remainder of the experiment the visual stimulus signalled that reward was not available.

Data Collection and Analysis

The computer recorded the number of lever presses made by the rat in the 60 s prior to the introduction of the stimulus (Pre-CS) and the 60 s while the stimulus was present (CS). These were cumulated separately over a session and constituted the raw data for analysis. The data were submitted to a square root transform [$X' = \text{Sqrt}(X + 0.5)$] to achieve normality of distribution (38). They were then submitted to analysis of variance. All effects involving treatment and days were assessed for the presence of orthogonal linear, quadratic and cubic polynomial components (29). The data for chlordiazepoxide and buspirone were analysed separately, with the control data repeated as the zero point for the two separate dose-response curves to allow extraction of the linear, quadratic and cubic trends. The linear trend extracted by this method is identical to the slope of a linear regression fitted to the relevant means and the higher order trends represent symmetrical curves with an increasing number of inflections as the order of polynomial increases.

RESULTS

Chlordiazepoxide at 5 mg/kg and 20 mg/kg increased Pre-CS responding relative to controls. In a similar fashion, the two lower doses of buspirone increased responding (Fig. 1A) whereas the highest dose of 3.3 mg/kg buspirone produced a decrease in responding.

Over the 20 days of testing, chlordiazepoxide clearly, and dose dependently, impaired response suppression, as assessed by the difference between CS and Pre-CS scores [discrimination × chlordiazepoxide dose × daypairs, dev × lin × lin, $F(1,270) = 14.4$, $p<0.001$, Fig. 1B]. The two higher doses of buspirone also impaired response suppression but to a less obvious extent than chlordiazepoxide [discrimination × buspirone dose × daypairs, dev × lin × lin, $F(1,360) = 5.5$, $p<0.05$].

Inspection of the data suggested that by the 10th day of testing control performance had reached asymptote and that this could have obscured, statistically, buspirone's effects. So a post hoc reanalysis limited to the first 5 daypairs was done in order to assess the effects of the drugs purely on acquisition of response suppression. The straight lines plotted in Fig. 1B represent the linear components from this second analysis.

Student-Newman-Keuls testing on the slope coefficients showed a significant difference between 20 mg/kg chlordiazepoxide and 5 mg/kg chlordiazepoxide ($q = 4.7$, $P = 2$, $n = 123$, $p<0.001$) and between 5 mg/kg chlordiazepoxide and controls ($q = 5.4$, $P = 2$, $n = 123$, $p<0.001$). In the case of buspirone, controls were significantly different from all doses of buspirone (3.3 mg/kg: $q = 3.66$, $P = 4$, $n = 164$, $p<0.05$; 1.1 mg/kg: $q = 3.45$, $P = 3$, $n = 164$, $p<0.05$; 0.3 mg/kg: $q = 3.66$, $P = 3$, $n = 164$, $p<0.05$) but the different doses of buspirone did not differ significantly from each other.

DISCUSSION

Chlordiazepoxide impaired successive discrimination by releasing response suppression in a dose-related fashion as has been reported previously (3, 18, 34).

The effects of buspirone on response suppression were smaller than those of chlordiazepoxide and buspirone's effects showed virtually no change over a 10-fold dose range. It should be noted that these differences between buspirone and chlordiazepoxide in

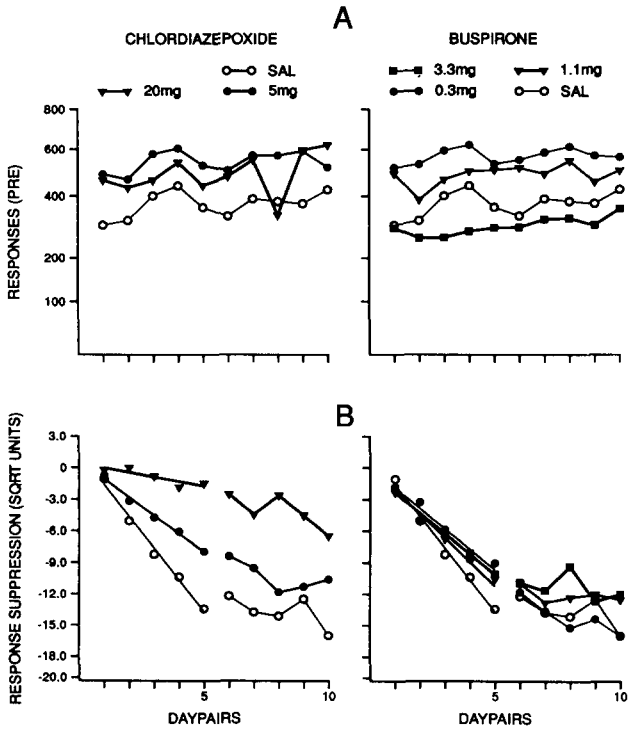

FIG. 1. (A) Effects of chlordiazepoxide and buspirone on responding during acquisition of a successive discrimination task during the 60 seconds preceding (PRE) a visual cue. The nonlinear response axis is the result of square root transform. (B) Response suppression in the same task as assessed by the difference between responding during the 60-second visual stimulus signalling nonreward (POST) and PRE responding. The positive and negative values on the scale show potentiated and suppressed responding, respectively, during stimulus presentation, relative to PRE. The straight lines represent the linear regressions extracted from post hoc analysis of variance of the the first 10 days of testing only (see text). Daypairs indicate the pooling of data of two successive days. The values in (B) are on exactly the same linear scale as (A).

their effects on response suppression cannot be attributed to differences in their effects on Pre-CS responding. Pre-CS responding is similar in all drug groups except 3.3 mg/kg buspirone and response suppression is similar in all drug groups except 20 mg/kg chlordiazepoxide. The effects of 1.1 mg/kg buspirone most clearly approximate those of 5 mg/kg chlordiazepoxide. We have seen the same dose equivalence in other behavioural tests. However, it should be noted that the 3.3 mg/kg dose produced only marginally greater effects and certainly did not approach the effect of the 20 mg/kg dose of chlordiazepoxide. At high doses, therefore, a much greater release of responding is observed with administration of chlordiazepoxide than with buspirone.

There are at least two reasons why high doses of buspirone might produce different effects from chlordiazepoxide. Firstly, buspirone increases locus coeruleus noradrenergic neuronal activity (26,35) whereas benzodiazepines, such as diazepam and chlordiazepoxide, decrease the activity of locus coeruleus neurons (13,26). While depression of the locus coeruleus only reproduces part of the behavioural profile of anxiolytic drugs (19) it does impair successive discrimination. Thus buspirone could impair successive discrimination at low doses but at higher doses this effect could be counteracted by increased activity in the locus coeruleus. (On this hypothesis the primary effect of chlordiazepoxide would be potentiated by accompanying decreased activity in the locus coeruleus.) Secondly, buspirone increases plasma corticosterone levels at doses of 2 mg/kg and above (36). This release might, in some way, counteract the primary effect of the drug. Chlordiazepoxide has been reported to increase plasma corticosterone levels only at relatively high doses [20 mg/kg and 40 mg/kg; (6)]. Doses within 3 to 10 mg/kg do not elevate corticosterone (16).

Since buspirone is equipotent clinically to the benzodiazepines (5, 11, 12, 15), it may be best to view it as having two separate actions. One, which parallels its clinical action and results in similar effects in animals to those of the classical anxiolytics, and a second action, which opposes the first in at least some animal tests, but is irrelevant to its clinical efficacy. Such a two-process account would explain both the nonlinear dose-response curve seen in the present experiments and also the fact that the effectiveness of buspirone varies between experimental paradigms.

At all events, the present results, together with the previously known effects of buspirone, show that its weak effects in successive discrimination paradigms are not restricted to shock-induced suppression. It appears that current animal models may need to be used with considerable care if detection of novel anxiolytics is to be ensured.

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