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Effects of Septal Lesions and Chlordiazepoxide (*Librium*) on Avoidance Behavior in Rats[1]

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FELDMAN, R. S., B. R. KAADA AND T. LANGFELDT. *Effects of septal lesions and chlordiazepoxide (Librium) on avoidance behavior in rats.* PHARMAC. BIOCHEM. BEHAV. 1(4) 379–387, 1973.–The combined effects of septal lesions and chlordiazepoxide (CDP) were observed during 5 consecutive procedures involving active avoidance, and passive avoidance during approach–avoidance conflict. The Maier paradigm on a Lashley jumping apparatus was used. The studies led to the following results and conclusions. Septal lesions had no effect on response latency in an active avoidance test. Septal lesions reduced latencies during conflict and learning tests when negative incentives were salient features. Adding CDP reduced latencies further. During extinction tests when negative incentives were withdrawn, response latency for the controls declined to that of the septal-lesioned rats. (Thus, the disinhibitory effects of septal-lesioned rats.) Thus, the disinhibitory effects of septal lesions that become manifest during passive avoidance tests, are enhanced by CDP. This suggests that the septum is not a significant site for CDP action.

Septal lesions Chlordiazepoxide

A NUMBER of experiments have been performed investigating the effects of chlordiazepoxide (CDP) on stereotyped behavior [2, 3, 4, 5, 6, 7]. These studies utilized the Maier paradigm which is a two-stage procedure using a Lashley jumping apparatus. In the first stage a two-choice problem is insoluble; half the responses to any cue (Bright vs. Dark) or position (Left vs. Right) are rewarded and half are punished. During rewarded responses the animal jumps against an unlocked window and gains access to a food station or escape from grid shock. During punished responses the animal jumps against a locked window, suffers a bump and falls to a net 1.0 M below. This sequence of trials (usually 10 trials per day, 16 days) causes increases in response latency and stereotyped responses to a position. In the second stage the problem is made soluble usually by having the Dark window correct, but most animals (approximately 85%) persistently make the position response adopted in the previous stage and fail to solve the problem within the limit of 200 trials. These rats are designated as fixated. However, there is ample evidence that the fixated rats discriminate between the correct and incorrect window. They typically make fewer abortive jumps and jump with significantly shorter latencies to the correct window.

It is resonable to view fixated behavior as an inhibitory deficit of performance. That is, the fixated rats persist in the nonadaptive (or perhaps more accurately, the less adaptive) position response because they are unable to inhibit this ongoing response even though it is consistently punished on 50% of the trials in the presence of a clearly discriminated cue. We shall refer to this effect as frustration-induced disinhibition.

Using psychotropic drugs to probe the mechanism of fixated behavior it was found that rats injected with benzodiazepines, chlordiazepoxide (CDP) and diazepam, became behaviorally depressed and ataxic, yet showed a decrease in response latency during the insoluble problem stage and without receiving the drug during the soluble problem phase showed a highly significant increase in solutions [2,3].

In a subsequent study [8] it was reported that rats showed a latency decline if CDP were given only during the soluble problem state. However, there were, if anything, fewer solutions than expected. Other studies [3] have shown that if rats that are already fixated are given CDP ½ hr before testing there is also a marked latency decrease though, again, solutions rarely occurred. These decreased latency effects are manifestations of the drug's disinhibitory property, a property that has been frequently demonstrated in other studies with the same and related substances [1, 6, 15, 16]. Thus, CDP causes a disinhibition which results in lower latency in situations that are somewhat stressful, but fails to yield problem solving since disinhibition favors the occurrence of incorrect responses.

It is known that selective lesions among limbic structures also yields response disinhibition. It was previously observed [9] that electrical stimulation of the anterior limbic

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field, the subcallosal-septal area, produced suppression of spontaneous somatomotor movement, motor afterdischarges and movements induced by cortical stimulation, and mono- and polysynaptic spinal reflexes, as well as suppression of a number of autonomic activities. With these observations as a point of departure the inhibitory and facilitory area of limbic cortex and septum was systematically investigated with respect to active and passive avoidance [10, 13, 14]. It was found that inhibitory functions were diminished by subcallosal septal lesions in cats and rats.

Noting that septal lesions and CDP administration both yield disinhibition of responses that are under aversive control, it would be of considerable interest to investigate the combined effects of septal lesions and CDP. Earlier studies have done this to some extent. It was reported [18] that CDP significantly decreased electrically evoked septal afterdischarges. It was later found that the frequency of the spontaneous EEG in cats was significantly increased by benzodiazepines, and this increase was attributed to depression of septal disinhibition [19]. These findings suggested that the septal area might be a significant focus for CDP effects it not a primary action site.

The present study investigated the single and combined effects of septal lesions and CDP administration on rats that were challenged in a Maier paradigm experiment. That is, after the rats were trained to jump they were given an insoluble and a soluble problem. The first objective of this study was to compare the lesion and drug effects in an attempt to illuminate the mechanism of disinhibition as it applies to behavior fixations. Second, this study used the lesion and the drug to establish parallels between Lashley apparatus experiments and other experimental paradigms such as one- and two-way avoidance problems.

Therefore, all of the above tests of the effects of septal lesions on responses in the Maier paradigm were also done when CDP was added to one group of septal-lesioned rats. The simple rationale for this was to test the hypothesis that the septal area is an important site of action for CDP and that if this site were destroyed, then CDP effects would be diminished.

After the training period and the insoluble and the soluble problem, two more tests were done. The first of these was to determine the effects of CDP on septal-lesioned and control rats using each rat as its own no-drug control. This is a more sensitive method of testing the effects of CDP on septal-lesioned rats. Finally, all fixated rats were subjected to a kind of extinction procedure wherein both windows were to remain unlocked as testing continued. The rational for this test was that of removing the inhibition generated by the locked window to see if that would cause the situation to revert to a simple one-way avoidance problem. The prediction was that the difference between septal-lesioned and normal rats should now substantially disappear since disinhibition was no longer a factor in the situation.

METHOD

Animals

Fifty-two albino rats of Möll-Wistar strain were used in this study. All were approximately 90 days old at the beginning of training. However, 7 rats did not survive surgery, 2 died during one of the later procedures, and 6 septal-lesioned rats were discarded from any analysis of results because necropsy showed that their septal area was more than 50% intact.

It should be noted, also, that some animals that were used for one experimental test could not be used for a subsequent test. For example, a rat that was fixated to the left might jump normally when the correct window appeared on the left but abortively (e.g., dive to the net without touching the window) when the incorrect window appeared on the left. This rat could be used in an analysis of latency scores during the soluble problem, but this rat could not be used in the subsequent extinction tests. In the extinction tests both windows were correct but because this rat did not jump against the formerly incorrect window it obviously could not discover that it was now unlocked. Therefore this test would be meaningless for this animal.

Apparatus

A semi-automatically controlled Lashley jumping stand similar to the one previously illustrated [4] was used. It consists of a pair of 15 cm sq. translucent Plexiglas windows set side by side and separated by a nose piece 5 cm wide and extending 7 cm toward the jumping grid. These windows could be independently locked and illuminated. A jumping grid 11.5 x 20 cm was centered before the windows; the leading edge of the jumping grid was 22 cm from the windows. There was a cloth net 1.0 M below the windows into which the rat fell if it jumped against a locked window. An electronic shock source and scrambling device produced a 0.35 mA shock on the jumping grid to force a response; and the windows were transilluminated by 25 W bulbs.

Procedure

Training. All rats were trained to respond on the apparatus by a method of approximation. At first the jumping grid was placed directly between the two open windows. The windows were transilluminated one at a time, and the illumination was alternated after 2 trials. The animals during this stage of training were food deprived for 23 hr and a small cup of wet lab chow mash was available when they stepped from the grid through the windows. On alternate trials the rats were gently guided to the side opposite that of the previous response to encourage responding to both windows.

On subsequent days the jumping grid was drawn back about 2–3 cm from the windows until the rats were jumping 22 cm through the open windows. Then the windows were gradually closed over a period of three days. After that all rats were readily jumping through the closed but unlocked windows. The animals received 10 trials per day during these training procedures, 5 to the right and 5 to the left side, half of these were to Dark and half to Bright. The rats were highly motivated to jump for the food reward obtainable behaind the windows so little prodding or grid shock was necessary to induce jumping. At the end of the 10 trial sessions the animals were returned to their home cage and given 40 g of lab chow. Drinking water was always available.

Surgical procedures. After jumping training was completed all rats were anesthetized with Nembutal and fixed in a stereotaxic device. After the scalp was retracted, bilateral burr holes were drilled 1.5 mm anterior to the bregma and about 0.75 mm from the midline. A monopolar lesioning electrode was then inserted bilaterally to a depth of about 6.0 mm. A ground electrode was connected to the cut skin. After a lesioning current of 2.0 mA was applied for 30 sec, the electrode was withdrawn, the scalp was sutured, and a 0.2 cc prophylactic dose of penicillin was injected into the

rat's left thigh.

The control animals were treated in the same way except that after burr holes were drilled in their skulls no lesioning electrode was inserted.

All rats were then given a 10 day rest period in their home cages.

Retraining. Next, the rats were retrained for jumping. First they were tested for their ability to jump the 22 cm to completely closed windows. Some rats within a few trials were jumping as well as they did before surgery. Other rats had to be retrained by moving the jumping platform a few cm forward, opening the windows, and prodding the rats to jump by nudging them or applying brief pulses of weak grid shock. After a few successful responses the platform was moved back a few cm or so and a few more trials were given and so on. This was done until the animals were again jumping 22 cm to closed windows. Most animals with septal lesions showed startle responses to touching, were distracted by tail tapping and nudging, and they jerked when toggle switches clicked on and off, but all were able to resume efficient jumping within the 2 or 3 daily sessions of 10–20 trials each. When all animals had resumed jumping efficiently the experimental procedures began.

Active avoidance testing. For this test 12 control, 13 septal-lesioned, and 12 spetal-lesioned rats that were to be given CDP in the next stage were given 10 trials per day for 4 days. The illuminated window was switched from side to side in a fixed random order [7], and both windows were unlocked. A continuous 0.35 mA grid shock was applied to the rat if it did not respond within 30 sec. No food was available behind the windows during this test nor any subsequent test. The trials followed one after the other as soon as the direction and latency of response was recorded, and the apparatus was set for the next trial. An average of 10 sec elapsed between trials. After testing, the animals were returned to their home cage and given an approximate ration of food pellets.

Insoluble problem testing. In this test all rats were given 10 trials per day of an insoluble problem. The problem was insoluble in that one of the pair of windows was locked in a set random sequence [7] so that a consistent response to a position (Left–Right) or to a brightness (Bright–Dark) was punished on 50% of trials. This procedure continued for 8 consecutive days, i.e., 80 trials. In most other experiments 160 trials of the insoluble problem were given and this usually produced a high proportion of fixations, approximately 85%. In this study only 80 trials were given to provide for the possibility that septal lesions would contribute an increment toward more fixations. A former study [20] showed that along with 80 insoluble problem trials lesions in frontal cortex in rats did add a significant proportion of fixations. This addition certainly would have been obscured had 160 trials been given.

Also, 12 of 25 septal rats in this group were given an IP injection of CDP (15 mg/kg) ½ hr before daily tests. As beofre, response latency was recorded for all trials, and grid shock forced a response if the rat did not respond within 30 sec.

Soluble problem testing. During this stage all rats (12 controls, and 25 septal-lesioned) were presented with a soluble problem. That is, one of the windows, usually Dark, was always unlocked and a response to it led to escape from the jumping grid. Grid shock occurred after 30 sec. The animal was considered to have solved the problem if it responded to the correct window without making more than one error in 3 consecutive days of testing (30 trials). Testing in this stage stopped if the rats did not meet the learning criterion within 20 days (200 trials). These rats were designated as fixated. No drug was given during this stage. The animals that solved the problem were removed from any additional tests.

Latency tests with CDP. For the next six weeks the rats that were fixated and did not show abortive jumping responses were subject to alternated testing under CDP and no-drug conditions. This is the test in which the rats served as their own controls during drug vs. no-drug tests. Specifically, the soluble problem was continued and 8 nonlesioned controls and 16 septal-lesioned rats were injected ½ hr before daily testing. The rats were tested with drug for two days, then with saline for two days, and then given no tests for 3 days. For the first two weeks 15 mg/kg of CDP was given but this produced severe behavioral depression and ataxia so for the next 4 weeks 10 mg/kg was used. Grid shock forced a response after 30 sec. Response latency was recorded for every trial.

Extinction tests. Only 7 controls and 15 septal-lesioned rats completed this procedure (1 rat in each group died during this test). All animals were given 10 consecutive trials for 20 days during which both windows were unlocked. Response latency was recorded for each response. No drug was given; grid shock came on after 30 sec, but most responses occurred before this happened.

Histology. When the animals completed all procedures or if they were removed earlier from further testing, they were sacrificed with an overdose of Nembutal and perfused with saline and 10% Formalin. The brains were removed, fixed in Formalin and embedded in paraffin. They were then serially sectioned in a frontal plane at 15 μ. Every twentieth section was mounted and stained by the Nissl method. The lesion size was determined by projecting and drawing each mounted section. Two independent observers visually estimated the extent of the lesion. Comparisons were made against normal brain sections.

RESULTS

Histology

Figure 1 shows reconstructions of the smallest acceptable and the largest septal lesion, A and B, about 50% and 100% respectively. For all other rats used in this study the lesion had an extent between these two extremes. Six operated rats were found to have septal lesions that left more than 50% of the septal nuclei intact. Even though these animals had completed most, if not all, of the procedures, they were discarded and not used in any data analysis. In no result was lesion size of any significance once rats with lesions below 50% were discarded from the analysis. Also, estimates were made of the amount of damage done to the fornix. The amount of damage ranged from nothing to almost total bilateral destruction. Specifically, 10 animals showed no damage; 9 showed partial damage on one side; 4 showed partial damage on both sides, and 2 showed large or total damage on both sides. These findings, however, could not be reliably related to any behavioral result.

Active Avoidance Testing

The left side of Fig. 2 shows the latency of response during the 4 days of active avoidance testing for the control

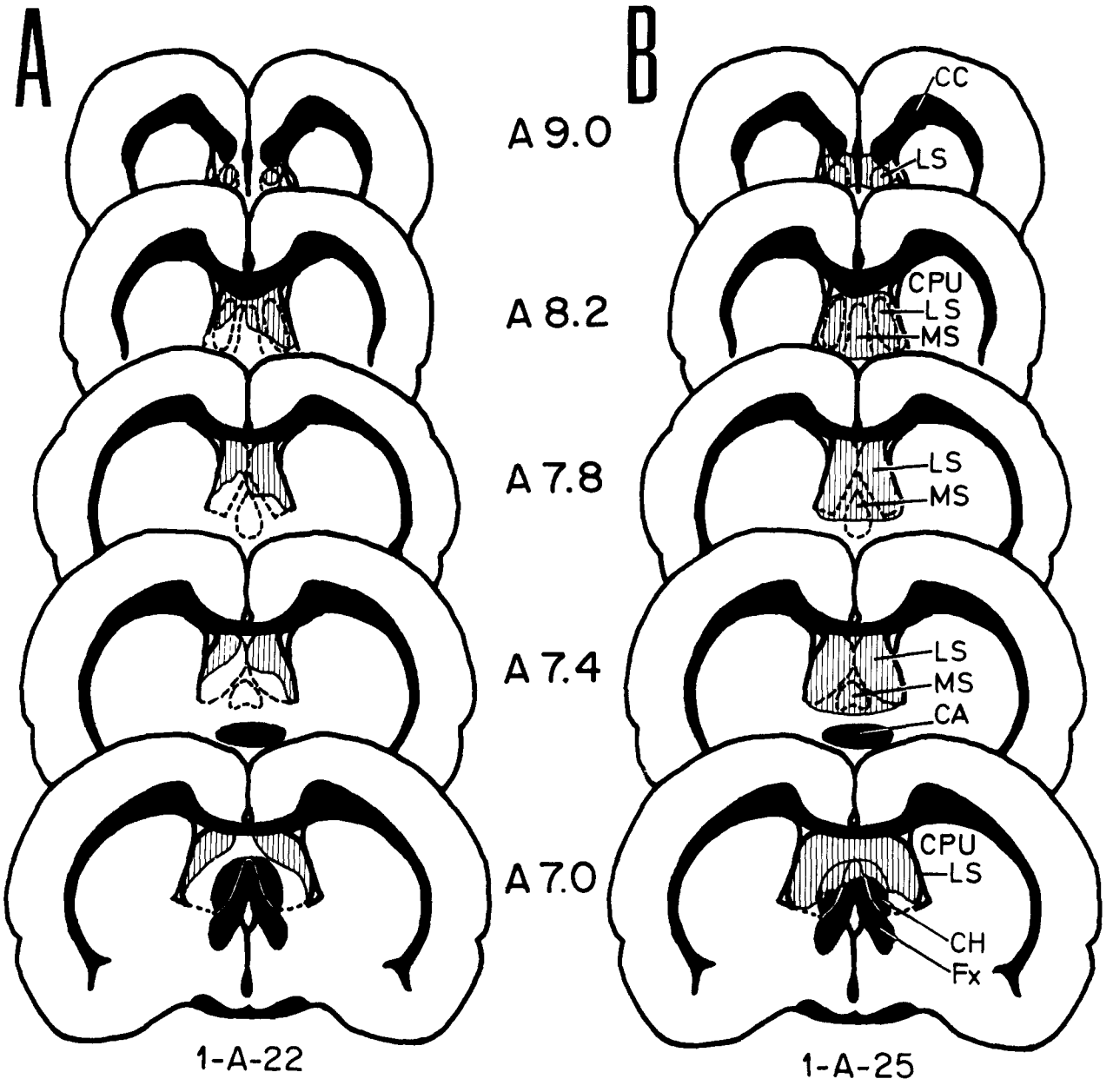

FIG. 1. Restruction of bilateral septal lesion (vertical hatching) in two rats showing in (A) the smallest acceptable lesion (approximately 50%), and in (B) almost 100% destruction of the septal nuclei. All other lesions had an extent between these two limits. CA: commisura anterior; CC: corpus callosum; CH: commisura hippocampi; CPU: nucleus caudatus/putamen; Fx: fornix; LS: nucleus laterali septi; MS: nucleus medialis septi.

and lesioned rats. The results showed that the average group differences were not significant. However, the latency for the controls after a decline on the second day increased on the third and fourth day; while the latency for the lesioned rats declined and remained lower. This group difference was significant (analysis of variance: Trials effect $F = 15.19$; $p<0.001$; Groups x Trials interaction $F = 3.21$, $p<0.01$).

Comparing the average lesion size for the two septal groups it was found that the lesion alone and lesion + CDP groups had 70% and 80% damage respectively; this difference was not significant.

Insoluable Problem Testing

The right side of Fig. 2 shows the response latencies of the three groups during the eight days of insoluble problem testing. It is seen that there is little overlap among the groups with the septal groups consistently lower. An analysis of variance showed that during this stage there is a group effect ($F = 4.65$, $p<0.025$), and there is a trials effect ($F = 2.51$, $p<0.025$), probably reflecting a gradual increase in latency over trials. Between groups it was found that the difference between the control and lesion alone group just missed the 0.05 level of significance ($F = 4.10$;

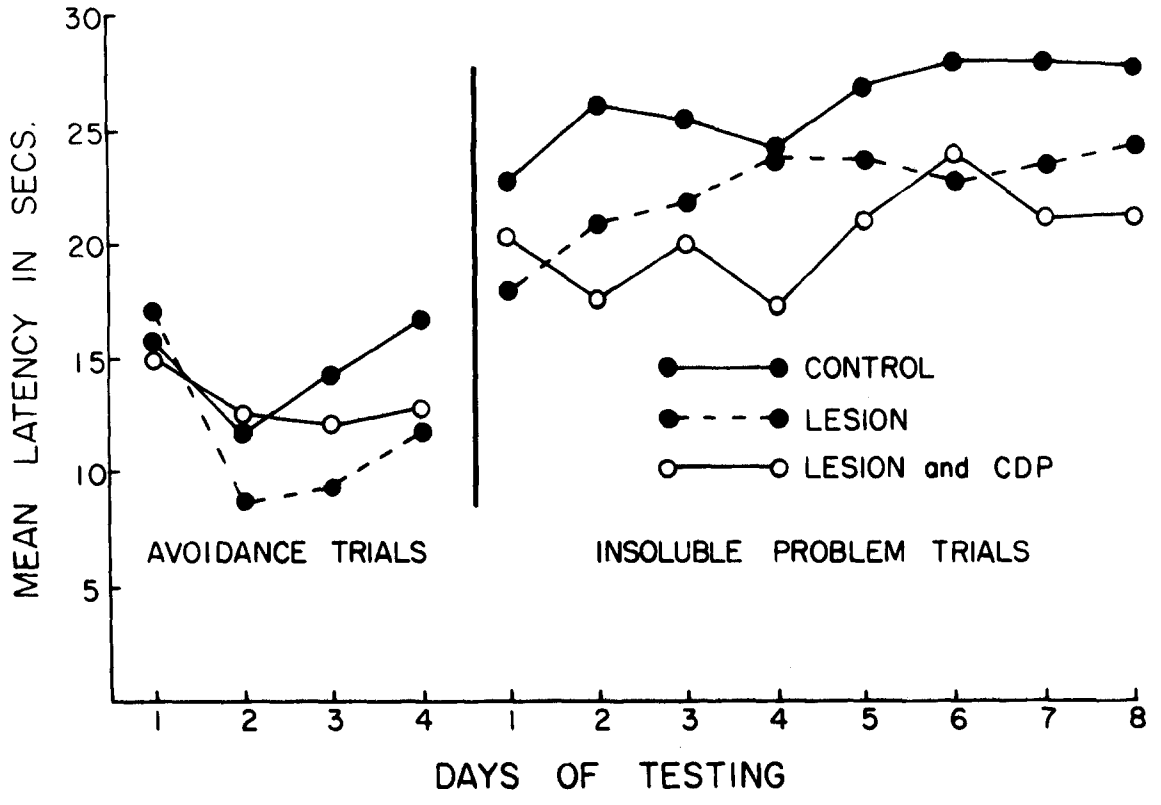

FIG. 2. Response latency during the avoidance and insoluble problem tests. Chlordiazepoxide was administered only during the insoluble problem test.

$0.05<p<0.10$), while the difference between the controls and the lesion and CDP group was highly significant ($F = 9.03$; $p<0.01$). Comparing the two lesion groups, it is seen that the lesion and CDP is, as expected, lower than the lesion alone group but the difference is not significant.

Soluble Problem Testing

In this stage two kinds of data are considered. First, it was found that some rats among both controls and septal-lesioned rats could solve the problem; 2 of 12 controls, 3 of 13 of the lesion alone, and 4 of 12 in the lesion and CDP groups reached the learning criterion. These differences were not significant, nor was there any significant difference with respect to lesion size for solvers and nonsolvers; 77% for solvers, 71% for nonsolvers.

Seocnd, of the rats in both groups (10 controls and 18 septal-lesioned rats) that failed to solve the problem and persisted in position responding during the 200 trial test, it was seen that the rats in both groups soon learned to respond faster to the correct than to the incorrect window. This is shown in latency data in Fig. 3. Moreover, the septal-lesioned rats alone and the septal-lesioned rats that had received CDP during the previous stage show an overall lower latency than the controls. The curves for these latter two groups are combined because their treatment during this stage was the same and the results were virtually identical. The analysis of variance yielded a significant Group effect showing that the combined latency scores of correct and incorrect responses were significantly lower for the septal-lesioned rats ($F = 10.2$; $p<0.005$).

Latency Testing with CDP

Figure 4 shows the effects of CDP on the latency of response for these control and septal-lesioned rats that served as their own controls. It can be seen that during the first 2 weeks at the higher drug dose (15 mg/kg) latencies are somewhat higher on drug trials but during the last 4 weeks the disinhibitory effects of the lower dose (10 mg/kg) of CDP is manifested in the definite drop in latencies during drug trials. This effect is clearly seen in the control rats for correct and incorrect responses, and this result confirms the significant results obtained in previous studies [3, 6, 8]. For the septal-lesioned rats, there was no drug-induced drop in latency for correct responses, probably because these rats were already responding as fast as they could. For incorrect responses, the combined drug scores were compared with the no-drug scores for each week. Student's t for matched samples ($df = 14$) showed that the drug scores were significantly lower than the no-drug score for each of the last 4 weeks (see Table 1). Thus CDP disinhibition does occur in septal-lesioned rats.

Extinction Test

During this test both windows (Bright and Dark) were unlocked and testing continued. All rats that completed this test consistently performed their fixated response. Figure 5 shows that within a few days of testing the latency of response to the formerly incorrect window showed a decline and toward the end of the 20 day test period there is almost no difference in latency between responses to the two windows. Comparing the first with the last day of the

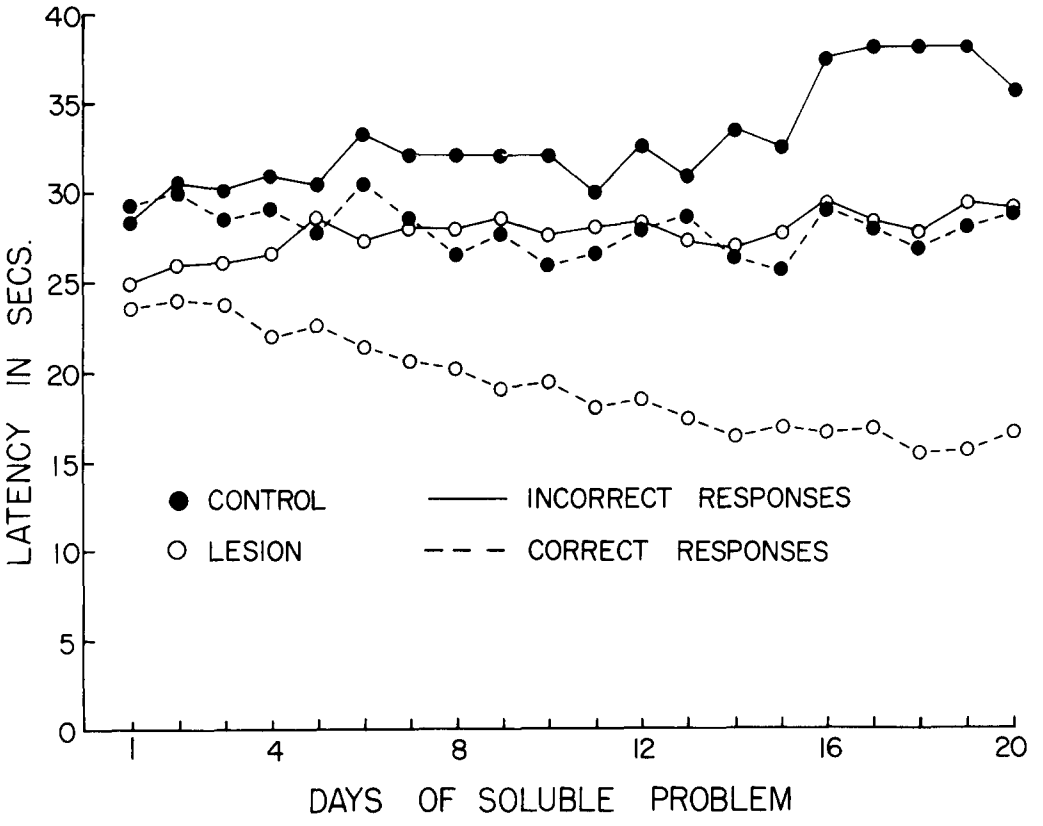

FIG. 3. Latency of response during the soluble problem for septal-lesioned and control animals. These rats discriminated between the correct and incorrect response but they fixated a position response and failed to solve the problem.

test, Wilcoxon's one-tailed, matched-pairs signed-ranks test showed that for the controls the difference on the last day was significantly less than that for the first ($p<0.05$) and for the septal-lesioned animals the same was true ($p<0.01$). It can also be seen that the latency for correct responses for the septal-lesioned rats remained almost constant at approximately 18–20 sec for the 20 days, and that all other latencies declined toward that level. This suggests that this value represents a floor level for these procedures. The analysis of variance showed that there was a significant Group effect ($F = 8.63$; $p<0.01$) and a significant Trials effect ($F = 7.09$; $p<0.001$). But there was no Group x Trials interaction ($F = 0.99$). This suggests that the septal-lesioned rats were, on the average, faster than the controls, that response latency for the formerly incorrect window declined significantly, but that the rate of decline was not significantly different between the septal-lesioned and control groups.

DISCUSSION

The results of the active avoidance test showed that septal-lesioned rats performing a well practiced avoidance response are not significantly different from controls with respect to overall latency. However, in this test the septals were different in one respect from the controls; namely, the increase in latency on the third and the fourth day was not as evident for the septal-lesioned animals. Our best explanation for this finding is that during original training and post-surgery retraining the rats were hungry and had food available after a response. During the active avoidance test the sole motivation to respond was avoidance of the grid shock. Practice effects and continued anticipation of food would account for the drop in latency on the second day for controls and lesioned rats, and for the controls nonreward would account for the latency rise during the third and fourth days. For the septals, the disinhibitory effect of the lesion would possibly eliminate this latency rise.

During insoluble problem trials, when an element of punishment and fear was introduced there was a noticeable latency difference between septal-operated and control rats and the addition of CDP to septal-lesioned rats increased the difference. These findings would seem to rule against the possibility that the septum is a significant center for CDP effects since damage to this structure obviously did not hinder the disinhibitory CDP effect.

One additional finding in the insoluble problem test was that there was no decline in latency over days for the CDP treated septal-lesioned animals. The latency decline has been a consistent feature of repeated CDP and diazepam tests in our studies [4]. Consistent with other studies [16] our explanation of the latency decline was that it was due to tolerance of the depressant effect of the drug. The depressant effect of the benzodiazepines in rats has been traced to a reduced turnover rate of norepinephrine (NE) in the midbrain-hindbrain region [21]. After 4–6 doses, however, the turnover rate returned to normal, thus accounting

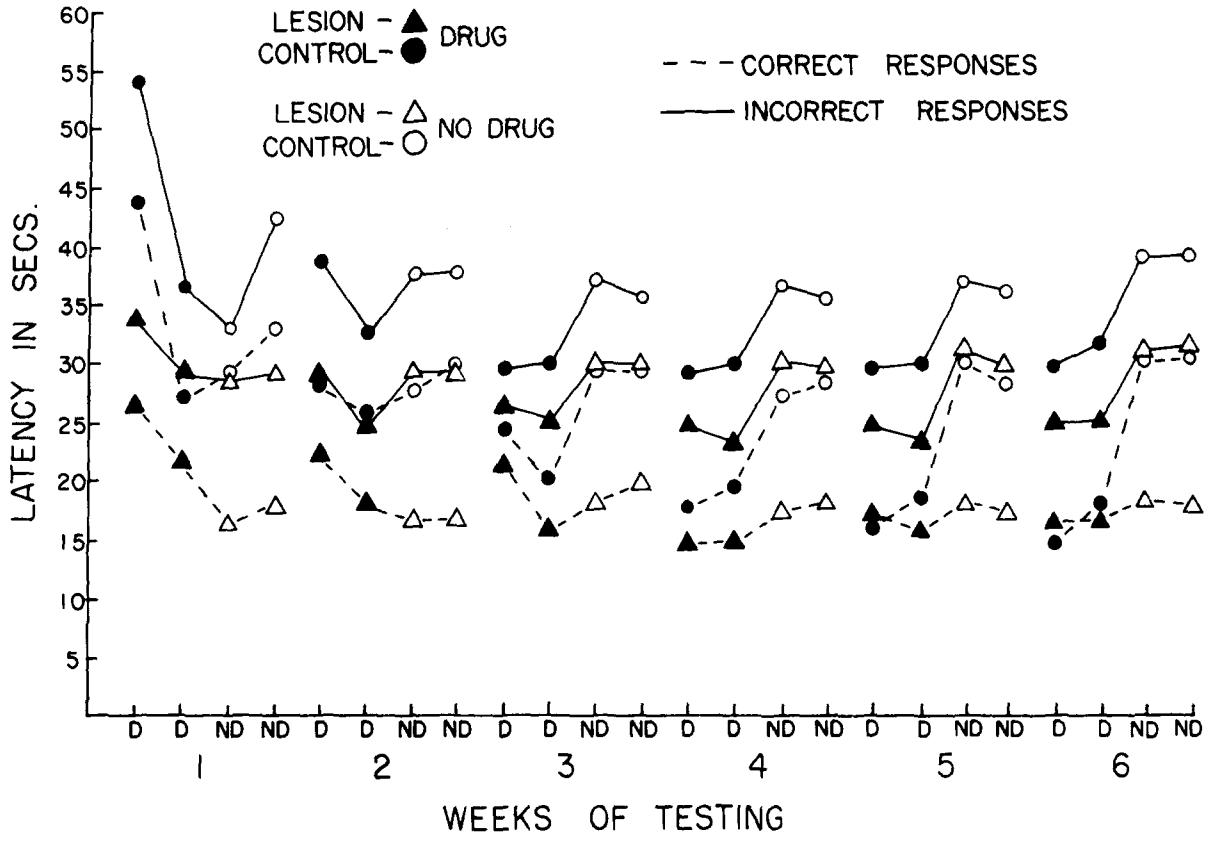

FIG. 4. Response latency for septal-lesioned and control rats during drug-on (D) and drug-off (ND) tests. During the first 2 weeks the drug dose was 15mg/kg, while during the last 4 weeks it was 10 mg/kg. During the last 4 weeks on drug days (solid points) latencies are generally lower for both groups.

TABLE 1

STATISTICAL COMPARISONS BETWEEN DRUG AND NO-DRUG TESTS FOR EACH OF THE LAST 4 WEEKS (INCORRECT RESPONSES, SEPTAL-LESIONED RATS ONLY)

Week of Testing	*t*	*p*
3	2.26	0.025
4	2.83	0.01
5	2.37	0.025
6	2.79	0.01

for the relative rapid tolerance of the depressant effect. If septal lesions were to lower NE levels in that part of the brain it would explain the absence of tolerance. Septal lesions have been found to cause a small (8% of control) and insignificant drop in whole brain levels of NE [12] but the change in the midbrain-hindbrain region was not ascertained.

On the other hand, the disinhibitory effect of the benzodiazepiens has been shown to be related to lower turnover rates of serotonin (5HT) in the midbrain raphé area [21], an area involved in the inhibition of responses that lead to aversive consequences. It has been further shown [12], that septal lesions significantly decreased whole brain levels of 5HT which probably affects the raphé nuclei since they are the major source of 5HT transmission in the brain. Even though Raisman [17] could find no evidence for direct connections from septum to the midbrain in the rat, the fact that septal lesions and CDP have additive disinhibitory effects suggests that septal lesions do influence serotonergic effects of the raphé nuclei. The one irreconcilable difficulty is the suggestion [12] that the lower 5HT level is responsible for increased sensitivity to foot shock and this could easily account for the lower latency in our septal rats. However, according to Stein *et al.* [21] CDP blocks 5HT turnover and releases behavior that has been suppressed by footshock and this suggests quite the opposite effect.

The results of the soluble problem are compatible with the findings of the previous two procedures and strengthen the explanation for them. The overall latency for the septal-lesioned animals are cleary lower than that for the controls and support the concept of septal lesion-induced disinhibition. The latency data also show that the fixated rats discriminated between the correct and incorrect window and that spetal lesions did not interfere with the acquisition or performance of that discrimination. If anything, the latency differences between correct and incorrect responses seem greater than those for the controls. However, the onset time

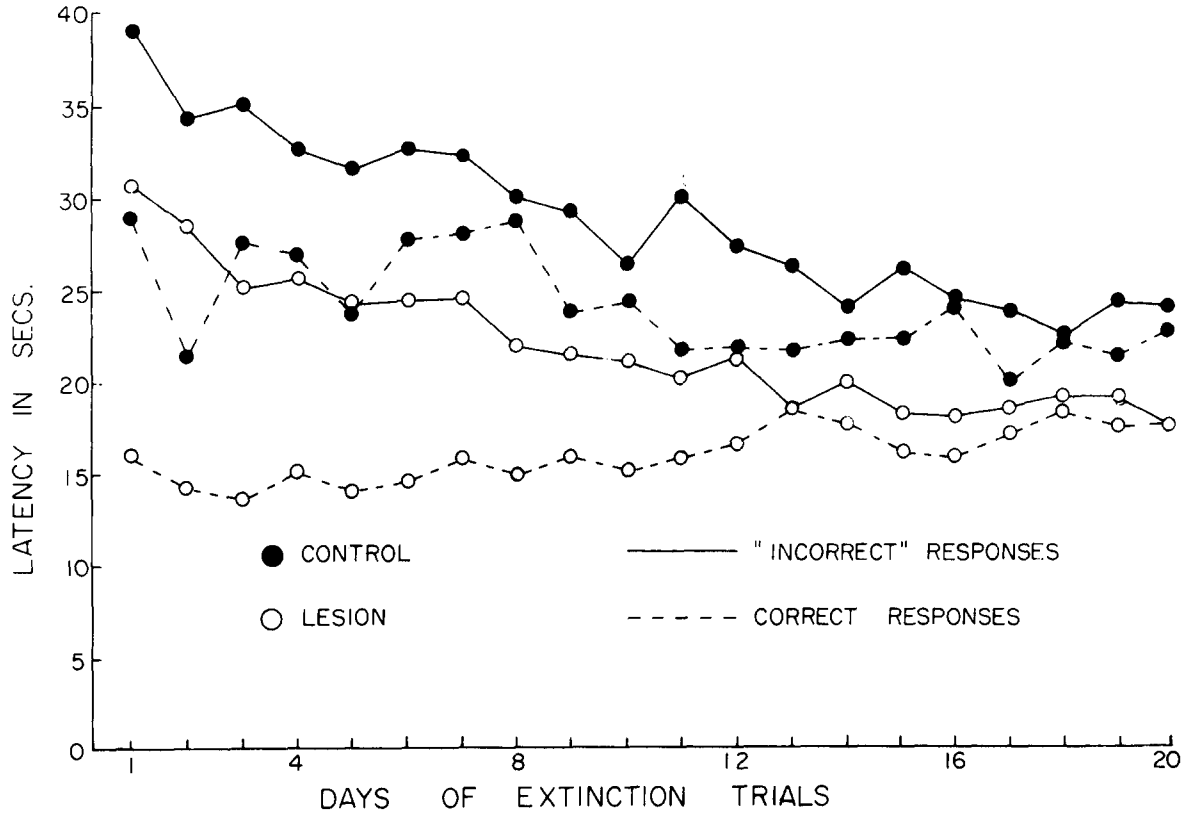

FIG. 5. Response latency during the extinction test. Both windows are unlocked. Latency differences for responses to each window and latency differences between septal-lesioned and control rats diminish.

of grid shock (30 sec) limited for the controls the degree of separation of the latency curves, so that reliable comparisons were not possible.

With respect to the number of solutions that occurred, it was expected that there would not be very many solutions among the septal-lesioned rats because the effects of the lesions would add to the effects of the insoluble problem. Also, while the lesions might cause disinhibition during the insoluble problem, they would continue to do so during the soluable problem and lead to even more incorrect responding. This finding is supported by previous results [5, 8, 11] which show that disinhibitory doses of CDP given during a soluble problem markedly interfered with solutions in Maier paradigm experiments.

The control rats however, did not yield any more solutions after 80 insoluble problem trials than the lesioned rats. A former study [20] using nonoperated controls yielded 6 of 10 (60%) solutions while in this study there were only 2 of 12 (17%). One explanation for the difference might be that the controls in this study had scalp incisions and burr holes drilled in the skull. The discomfort and pain when bumping into locked windows might have raised the level of conflict and fear and increased the incidence of fixations during the soluble problem. Strain differences could also account for the difference in results in the two studies. The former study [20] used Sprague-Dawley rats from the Charles River Breeding Laboratories, Wilmington, Mass.

The next test was a second attempt to increase disinhibitory effects on septal-lesioned animals with CDP. The controls were also treated with CDP and except for the limits imposed by floor effects showed about the same disinhibitory effects as the lesioned rats. Thus the drug exerted its full effect on the lesioned rats and indicated that CDP effects are not mediated in a significant way by septal nuclei.

The final test in this study was the extinction test. The changes in latency obviously indicated that the animals learned that one window was no longer locked. More important was the finding that the overall latency differences between the control and septal-operated rats also declined. These results not only support the findings obtained during the active avoidance test, but show that the response differences between the septal-operated and control rats were the result of the interaction between the septal lesions and the test procedures.

Another point deserves discussion. This concerns what might be called the dissociated disinhibitory effect. In the soluble problem, the fixated septal-lesioned rats show a clear cut acquisition of the Bright-Dark discrimination though their overall latency is significantly lower than that for the controls (see Fig.3). Thus there is a paradox wherein the septal-lesioned animals are disinhibited with respect to overall latency, but not so with respect to discriminating between the incorrect and correct window. Of considerable interest is the fact that CDP-induced disinhibition leads to precisely the same result, i.e., overall lower latency and prompt and pronounced latency separation [8].

This dissociated disinhibitory effect is a common feature as well in frustration-induced disinhibition. That is, position fixated rats clearly discriminate between the correct

and incorrect window by inhibiting responses to the incorrect symbol (e.g., Bright), but do not inhibit responses to the incorrect side.

Finally, fixated rats are frequently found in our experiments that show very little latency separation between correct and incorrect responses. Testing these same animals with CDP frequently yields a very significant latency separation. Why septal lesions, chlordiazepoxide, and frustration lead to these dissociated effects cannot be answered at the present time.

REFERENCES

1. Cook, L. Effects of drugs on operant conditioning. In: *Animal Behavior and Drug Action,* edited by H. Steinberg. London: Churchill, 1964, pp. 23–43.
2. Feldman, R. S. The prevention of fixations with chlordiazepoxide. *J. Neuropsychiat.* **3**: 254–259, 1962.
3. Feldman, R. S. Further studies on assay and testing on fixation-preventing psychotropic drugs. *Psychopharmacologia (Berl.)* **6**: 130–142, 1964.
4. Feldman, R. S. Comparisons of the benzodiazepines with other tranquilizers during conflict and chronic anxiety states. In: *Neuropsychopharmacology,* edited by P. B. Bradley, F. Flugel and P. Hoch. Amsterdam: Elsevier, Vol. 3, 1964, pp. 340–347.
5. Feldman, R. S. The mechanism of fixation prevention and "dissociation" learning with chlordiazepoxide. *Psychopharmacologia (Berl.)* **12**: 384–399, 1968.
6. Feldman, R. S. and K. F. Green. Effects of chlordiazepoxide on fixated behavior in squirrel monkeys. *J. Psychopharmoc.* **1**: 37–45, 1966.
7. Feldman, R. S. and K. F. Green. Antecedents to behavior fixations. *Psychol. Rev.* **74**: 250–271, 1967.
8. Feldman, R. S. and E. Lewis. Response differences of psychotropic drugs in rats during chronic anxiety states. *J. Neuropsychiat.* **3**: Suppl. 1, S 27–41, 1962.
9. Kaada, B. R. Somato-motor, autonomic and electrocorticographic responses to electrical stimulation of 'rhinencephalic' and other structures in primates, cat and dog. *Acta physiol. scand.* **24**: Suppl. 83, 1–258, 1951.
10. Kaada, B. R., E. W. Rasmussen and O. Kveim. Impaired acquisition of passive avoidance behavior by subcallosal, septal, hypothalamic and insular lesions in rats. *J. comp. physiol. Psychol.* **5**: 661–670, 1962.
11. Lewis, E. and R. S. Feldman. The depressive effect of chlordiazepoxide on a negative incentive. *Psychopharmacologia (Berl.)* **6**: 143–150, 1964.
12. Lints, C. E. and J. A. Harvey. Altered sensitivity to footshock and decreased brain content of serotonin following brain lesions in the rat. *J. comp. physiol. Psychol.* **67**: 23–31, 1969.
13. McCleary, R. A. Response specificity in the behavioral effects of limbic system lesions in the cat. *J. comp. physiol. Psychol.* **54**: 605–613, 1961.
14. McCleary, R. A. Response-modulating functions of the limbic system: Initiation and Suppression. In: *Progress in Physiological Psychology,* edited by E. Stellar and J. M. Sprague. New York: Academic Press, Vol. 1. 1966. pp. 209–272.
15. Margules, D. L. and L. Stein. Neuroleptics vs. Tranquilizers: Evidence from animal behavior studies of mode and site of action. In: *Proceedings Fifth International Congress Neuropsychopharmacology,* edited by H. Brill, J. O. Cole, P. Deniker, H. Hippius and P. B. Bradley. New York: Excerpta Med. Found, 1967, pp. 108–120.
16. Margules, D. L. and L. Stein. Increase of "anti-anxiety" activity and tolerance of behavioral depression during chronic administraiton of oxazepam. *Psychopharmacolgia (Berl.)* **13**: 74–80, 1968.
17. Raisman, G. The connections of the septum. *Brain* **89**: 317–348, 1966.
18. Schallek, W. and A. Kuehn. Effects of psychotropic drugs on limbic system of cat. *Proc. Soc. exp. Biol. (N.Y.),* **105**: 115–117, 1960.
19. Schallek, W. and A. Kuehn. Effects of benzodiazepines on spontaneous EEG and arousal responses of cats. *Prog. Brain Res.* **18**: 231–238, 1965.
20. Smith, D. A. and R. S. Feldman. Fixated behavior in rats induced by frontal lesions and conflict. *Physiol. Behav.* **4**: 871–876, 1969.
21. Stein, L., C. D. Wise and B. D. Berger. Antianxiety action of benzodiazepines: Decrease in activity of serotonin neurons in punishment system. In: *The Benzodiazepines,* edited by S. Garattini, E. Mussini, and L. O. Randall. New York: Raven Press, pp. 299–326, 1973.