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Diazepam Impairs Acquisition But Not Performance in the Morris Water Maze

ROBERT K. McNAMARA AND RONALD W. SKELTON

University of Victoria, Department of Psychology, P.O. Box 3050, Victoria, B.C., Canada V8W 3P5

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McNAMARA, R. K. AND R. W. SKELTON. *Diazepam impairs acquisition but not performance in the Morris water maze.* PHARMACOL BIOCHEM BEHAV **38**(3) 651–658, 1991.—Diazepam is known to produce anterograde amnesia in both humans and animals. The present investigation sought to determine if this impairment is a direct result of diazepam's interference with mnemonic processes or a result of deficits in performance or retrieval. Diazepam (3 mg/kg) was administered prior to training in the Morris water maze either before or after the rats had acquired the location of a submerged escape platform. Diazepam was found to impair acquisition but not retrieval of spatial information and this impairment was not due to the sedative, hypothermic or state-dependent learning effects of diazepam. These results replicate previous findings in the Morris water maze and provide new evidence that this deficit is primarily mnemonic in nature.

Diazepam Learning Memory Anterograde amnesia Sedation Hypothermia Anxiety Retention Morris water maze Consolidation

SINCE their synthesis by Hoffmann-La Roche, chlordiazepoxide (Librium®; 1955) and its more potent analog diazepam (Valium®; 1959), have come to dominate the market for anxiolytic/sedative drugs (2). Soon after their release onto the market, benzodiazepines (BZD) were discovered to have amnesic properties (11,24). Since these early observations, a large literature has accumulated which effectively replicates and characterises BZD-induced memory impairment (5, 27, 41). The common finding among these studies is that BZDs impair acquisition while sparing short-term memory and retrieval processes. In fact, BZDs may actually produce a significant retrograde facilitation of memory (3,18).

Although the amnesic effects of BZDs may be considered a positive side-effect in presurgical situations, problems may arise because 1) BZDs are so widely prescribed and are typically taken while patients are engaged in their daily activities, 2) little tolerance develops to the amnesic effects (15, 16, 37), 3) subjects may not be subjectively aware of their memory impairment, even after doing poorly on memory tasks (19), 4) the amnesia can last up to 14 hours after BZD administration (15), 5) the use of BZDs in a psychiatric context (i.e., to combat phobias) may hinder habituation or the acquisition of appropriate coping skills (17). Therefore, it would be valuable to determine the nature of this impairment more precisely.

It is not yet clear whether BZDs impair learning and memory processes directly or as a secondary result of performance deficits. Two alternative explanations are sedation and state-dependent learning. Sedation is a state of myorelaxation or drowsiness that may impair sensory and motor processes but not memory per se. This notion has been both supported (14, 26, 38) and rejected (9,15). State-dependent learning is a phenomenon in which accurate recall only occurs when the subject is in the same drug state during both acquisition and recall (35). State-dependent learning may account for some of the effects of BZDs (23,37), but cannot account for all of BZD's amnesic effects. For example, memory for information that is acquired and retrieved under the same drug state is impaired, whereas memory for information acquired in a nondrug state and recalled in a drug state is as good as, or better than, placebo controls (18). In sum, it is not clear whether BZDs exert their amnesic effects through sedation, state-dependent learning or a disruption of mnemonic processes.

One approach to this problem would be to study the effects of BZDs in animals. BZDs have been found to impair learning and memory in animals (5, 7, 43), but many of these investigations failed to dissociate learning impairments from performance deficits. For example, BZDs may impair the acquisition of a passive avoidance task but this cannot be unambiguously attributed to impairments of learning and memory because BZDs increase pain thresholds (22,46) thereby attenuating the significance of the shock (43). Alternatively, the impairment of passive-avoidance may have occurred through state-dependent learning processes (36). BZDs also impair successive discriminations, but, again, this cannot be unambiguously attributed to memory deficits. In this paradigm, BZD-treated rats overrespond to the disinhibitory properties of BZDs rather than impairments of learning and memory per se (6). In a third popular paradigm, the appetitively motivated radial arm maze (34), BZDs have inconsistent effects, both impairing performance (21,45) or having no effect (20). Thus the nature of BZD-induced memory impairment has not been clearly delineated in either animals or man.

One paradigm which may help to resolve this controversy is the Morris water maze (32,33). This task appears well suited to the examination of BZD-induced amnesia because several different measures of *learning* and *performance* can be measured simultaneously and over long periods of testing. Previous investigations have demonstrated that both chlordiazepoxide (30) and diazepam (29) impair place-learning in the Morris water maze. However, because the Morris water task uses cold water (18–26 ± 1°C) to motivate the animal, it is possible that the pre-

viously observed acquisition impairments were due to BZD-induced hypothermia, rather than a direct effect of BZDs on memory processes (40,47). Additionally, it is possible that the observed place-learning deficit resulted from impairments of retention, recall, or sedation. The present study sought to determine if BZDs impair memory directly or as a secondary result of impaired performance.

METHOD

Animals

Twenty-five Long Evans male rats (Charles-River, Quebec) weighing 300–550 g served as subjects. They were housed individually in the animal vivarium where lights were maintained on a 12:12-h light-dark cycle. All tests were conducted during the light portion of the cycle. Food and water were available ad lib.

Apparatus

The Morris water maze consisted of a circular pool (diameter: 150 cm, height: 45 cm) with a featureless white inner surface. The pool was filled to a height of 25 cm with 22°C (±1°C) water, in which 1500 ml of powdered skim milk was dissolved. The hidden escape platform was a clear Plexiglas stand (13 × 13 cm) submerged 3 cm below the water surface so that it was invisible at water level. The visible platform was a black stand (13 × 13 cm) that protruded 5 cm above the surface of the water.

Drugs and Group Assignment

At the beginning of the experiment, rats were randomly divided into five treatment groups. The first group (Diazepam; Hoffmann-La Roche Inc.; n = 5) was administered diazepam throughout both acquisition and reversal phases (see the Procedure section). The second group (Saline; n = 5) received an equivalent volume of saline for both phases of testing and served as the placebo control. The third group (diazepam-saline, DS; n = 5) received diazepam for the first phase (acquisition) and saline for the second phase (platform reversal). The fourth group (saline-diazepam, SD; n = 5) received saline first and then diazepam. The fifth group (Switch; n = 5) received saline until they acquired the platform location, at which point they were switched to diazepam (3 mg/kg) for the remainder of the acquisition phase, and also for the reversal phase. Diazepam was always injected IP in a dose of 3 mg/kg and saline (0.9%) was delivered in an equivalent volume of 0.6 ml/kg. All injections were administered in the pool room 30 min prior to testing. Control experiments have found that equivalent volumes of the commercial diazepam vehicle (propylene glycol, ethanol, buffer) does not have any significant effects on maze performance (unpublished observations).

Procedure

The experiment was divided into an initial acquisition phase (I) which consisted of training all groups to acquisition criterion, plus a probe and cue test, retraining and a drug reversal probe; and a reversal phase (II) which included training all groups to criterion performance under a reversed drug condition with a reversed platform location and a second probe test.

Phase I: Initial acquisition. During initial acquisition, the hidden escape platform was located in the center of the northwest quadrant. All groups were given four trials each day and tested until an acquisition criterion was reached (mean group distance under 250 cm over two consecutive days). For each trial the rat was placed in the water facing the pool at one of four randomly determined starting locations (north, south, east, or west pole). During each trial, the rat's swim path, drawn on a map of the pool and measured with a map-reading device, and escape latency, measured with a stopwatch to a tenth of a second, were recorded. Once the rat located the platform, it was permitted to remain on it for 15 seconds, and the occurrence of a rear was recorded. The rat was removed from the pool if it did not locate the platform within 60 seconds. The rat was placed on the platform for 15 s at the end of the final trial if it did not locate the platform at least once on the previous four trials. After each trial, the rat was returned to a waiting cage positioned 90 cm under a 250 W brooding lamp (for warmth) and allowed to remain there for the 5-min intertrial interval. The rat's core body temperature was measured rectally three times daily: prior to drug administration (predrug), thirty minutes after drug administration (preswim), and immediately following the last trial (postswim). Preswim temperature change (T_c) was calculated relative to the predrug temperature, postswim temperature (T_c) change was calculated relative to the preswim temperature.

After acquisition was complete, a probe trial was given to assess the strength and accuracy of initial acquisition. Rats were required to swim in the pool without the escape platform for 60 s. All rats were released from the same starting location and the distance spent in each quadrant was recorded. Following this single trial, a cue task was given to assess any sensorimotor deficits induced by diazepam. Rats were required to navigate to a visible platform located in a different quadrant on each trial; swim path lengths and escape latencies were recorded.

For the next two days, rats were retrained to the original platform location to compensate for interference produced by the probe and cue tests. This consisted of eight additional trials (4 trials/day) with the hidden platform replaced in the old location. After reacquisition, the DS and SD groups were given the appropriate drug reversal and a second probe trial was given.

Phase II: Platform reversal. To assess the effects of drug reversal on acquisition, all rats were required to learn the location of the hidden platform placed in the quadrant diagonally opposite to the previous location (reversed). To assess proactive interference, the distance spent in the old quadrant was also measured. After each group had reached the acquisition criterion (mean group distance under 250 cm over 2 consecutive days), a final probe trial was given.

Data Analysis

Over both phases of testing, group differences in escape latency, swim path length, heading error over the first 12 cm of the swim path, swim speed, T_c, and the probability of rearing at least once while on the platform were assessed using analysis of variance (ANOVA) with repeated measures. Post hoc comparisons were assessed using Tukey's (HSD) method.

RESULTS

Phase I: Initial Acquisition

The distance required by the Diazepam, Saline, and Switch groups to locate the submerged escape platform over the 14 days of testing are shown in Fig. 1. The Saline and Switch groups rapidly acquired the platform location, reaching asymptotic performance by the fourth day of testing. The Diazepam group was impaired on all three measures of performance. Overall, their swim path lengths were significantly longer, $F(2,97) = 207.4$, $p<0.001$, and over training, decreased slower than those of the saline-treated group, $F(26,1261) = 4.7$, $p<0.001$. Escape latencies (a more ambiguous measure of performance) showed a sim-

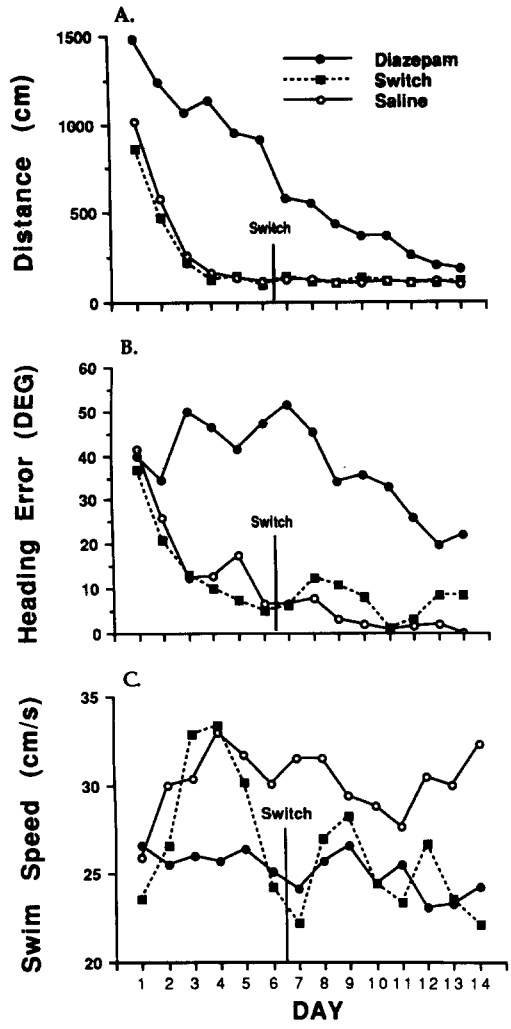

FIG. 1. Effects of diazepam on (A) the distance taken to locate the escape platform, (B) heading errors and (C) swim speed. The preadministration of diazepam resulted in greater distances required to find the platform, larger heading errors and reductions in swim speed. Note that when the Switch group was switched from saline to diazepam on day 7, neither the distance nor heading errors increased substantially despite a sustained reduction of swim speed.

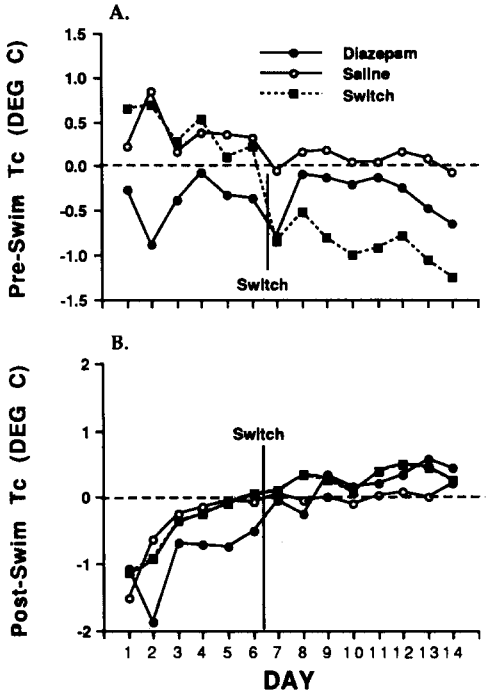

FIG. 2. Effects of diazepam on (A) preswim T_c (preswim − predrug) and (B) postswim T_c (postswim − preswim) during initial acquisition. Note: 1) the consistently lower preswim T_c over course of testing, 2) the reduction in preswim T_c when the Switch group is *switched* to diazepam on day 7, and 3) the increases in postswim T_c in all three groups.

ilar pattern of results and statistical significance (data not shown). Heading errors provided an even clearer picture of diazepam's effects (see Fig. 1B); the Diazepam group made greater errors overall, $F(2,97)=68.0$, $p<0.001$, and showed less improvement with training than the Saline group, $F(26,1261)=3.1$, $p<0.001$. Post hoc comparisons revealed that the Diazepam group had longer escape latencies ($p<0.01$), longer swim paths ($p<0.01$) and greater heading errors ($p<0.01$) than the Saline group. Perhaps the most significant observation was that diazepam did not impair performance of a previously learned escape response. When the Switch group was switched from saline to diazepam on Day 7, they showed no increase in path length or any other impairment of place-learning (Fig. 1A).

Diazepam also reduced swim speeds but the pattern of those results suggested that these performance factors could not account for the changes in platform localization. Figure 1C shows that the swim speeds of the Diazepam group were slower overall, $F(2,97)=42.7$, $p<0.001$, and did not recover over training as did the Saline group, $F(26,1261)=2.5$, $p<0.001$. Swim speeds in the Switch group were slower after the switch to diazepam ($p<0.05$), but were overall quite variable. Although it is not at all clear why the Switch group began swimming slowly before being switched to diazepam, it is important to recognize that this change did not affect swim path lengths, and that overall, these results show that diazepam's effect on swim speed had little or nothing to do with diazepam's effect on the rats' ability to locate the platform.

Diazepam lowered body temperature over all 14 days (Fig. 2A), but only exacerbated the swim-induced hypothermia for the first 6 days (Fig. 2B). A repeated measures ANOVA revealed significant effects of drug and days and significant interaction for both preswim temperature change and postswim temperature change ($p<0.01$). Post hoc analysis showed that the Diazepam group had greater preswim hypothermia than the Saline group ($p<0.01$), but did not have significantly greater hypothermia after swimming ($p>0.05$).

The probe trial revealed that the Diazepam group had not acquired the ability to locate the platform using spatial cues, despite having acquired the ability to navigate to the submerged platform during acquisition. The Saline and Switch groups swam significantly more than chance distances (25%) in the correct quadrant of the pool ($p<0.01$), whereas the Diazepam group spent only chance level distances there ($27\pm5.2\%$) (see Fig. 3A). These results confirm the observations from the acquisition phase, namely, that diazepam impairs acquisition, but not retention of the platform location. Swim speeds were comparable for all groups (Fig. 3B), and only the Saline group showed a significant ($p<0.05$) but very slight (-0.32 ± 0.07) hypothermia from swimming (Fig. 3C).

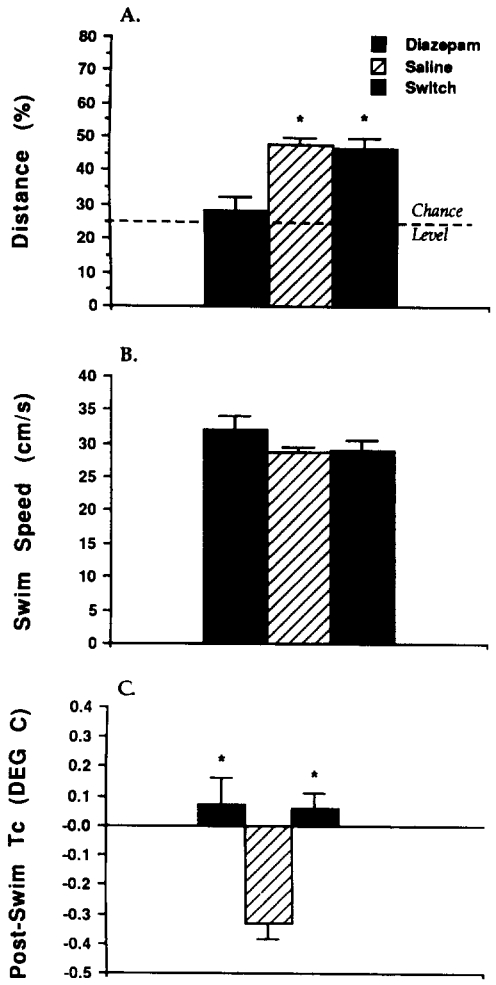

FIG. 3. Effects of diazepam on (A) the distance spent in the correct quadrant, (B) swim speed, and (C) postswim T_c (preswim − postswim) during the first probe trial. Both the Saline and Switch groups, but not the Diazepam group, demonstrated an 'above chance' preference for the quadrant that previously contained the escape platform. Also, none of the group's swim speeds differed during the probe trial. Finally, only the Saline group's postswim T_c was reduced. Data expressed as mean ± S.E.M. *$p<0.01$ compared to chance level (25%) in (A) and to Saline T_c in (C).

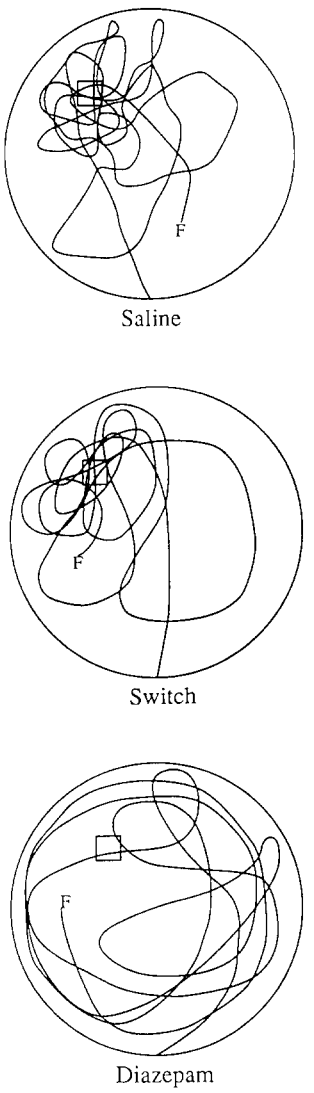

FIG. 4. Swim paths during the first probe trial from rats closest to their group mean. Note that the Switch group rat, despite having an elongated and circuitous swim path, still spent the majority of time in the correct quadrant. The 'F' denotes where the rat was removed from the pool after the trial was finished.

There was a very clear difference in the topography of the swim paths of rats treated with diazepam. Figure 4 shows the actual swim paths taken during the probe trial by rats closest to their group mean. These paths reveal the typical pattern of their respective groups: saline-treated rats showed a concentrated search in the correct quadrant, often using sharp turns and small tight loops; diazepam-treated rats swam all over the pool using slow turns and large gradual loops; rats switched from saline to diazepam also used slow turns but were able to concentrate their search in the correct quadrant.

None of the groups were impaired when required to navigate to the black visible platform, $F(2,97)=1.73$, $p=0.18$. This result suggests that the diazepam-treated animal can learn to swim to a single visual cue to escape the cold water, and can coordinate their behaviour to reach and climb onto the platform.

During the drug reversal probe trial, the Saline group showed a significant reduction of the distance spent in the correct quadrant relative to the previous probe trial ($p<0.01$). This suggests that the previous probe trial and the cue task disrupted retention of the original platform location and that no conclusions can be made regarding the state-dependent learning phenomenon from this test.

Phase II: Platform Reversal

When the platform position was reversed to the quadrant opposite that used for initial training, and half of the rats in the Saline and Diazepam groups were reversed to the opposite drug condition, the pattern of results replicated those observed in acquisition, namely, diazepam impaired acquisition of the new platform location. Figure 5A shows that all groups receiving diazepam (Diazepam, SD, Switch) had significantly longer swim path lengths, $F(4,95)=19.0$, $p<0.001$, and slower acquisition, $F(28,665)=2.7$, $p<0.001$. Post hoc tests revealed that all three groups were

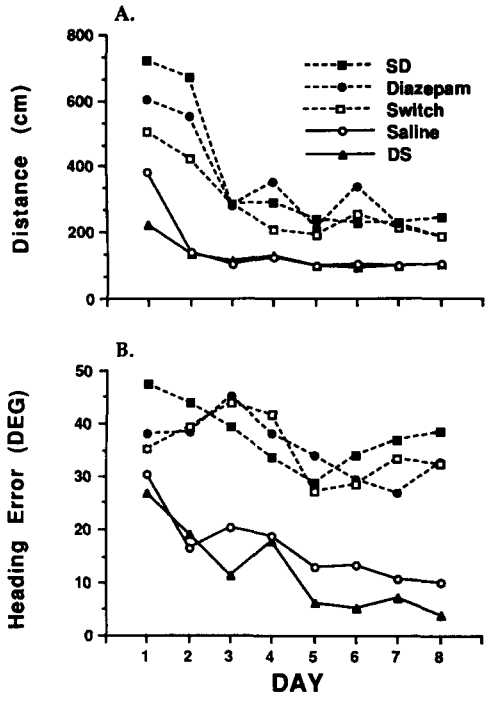

FIG. 5. An illustration of (A) the distance required to locate the hidden escape platform and (B) heading errors during reversal acquisition. Note that only those groups receiving diazepam (SD, Diazepam, Switch) have greater distances and heading errors.

different from the saline groups ($p<0.01$). The DS group (receiving saline) was no different from the Saline group, but was significantly different from the Diazepam group ($p<0.01$). Heading errors (Fig. 5B) and escape latencies (not shown) showed a similar pattern of results and statistical significances. It is worth noting that the DS group displayed the fastest acquisition, indicating that this group had: little or no residual effect of the previous 18 days' treatment with diazepam; little impairment of procedural learning during the first phase; little or no proactive interference from training to the old platform location under diazepam. In contrast, the Switch group and the SD group were as impaired as the Diazepam group, despite having mastered all the procedural components of the task during original acquisition under saline.

During the probe trial that followed reversal training, the groups treated with diazepam (Diazepam, SD, Switch) failed to show a preference for the correct quadrant, whereas those treated with saline (Saline, DS) showed a clear preference ($p<0.01$; see Fig. 6A). None of the groups differed on swim speeds or postswim T_c (Fig. 6B, C). Interestingly, the DS group showed no evidence of residual impairment from previous diazepam treatment, spending more time in the correct quadrant than any other group.

The percentage of distance spent by each group in the previously correct quadrant is illustrated in Fig. 7. All of the groups except for the DS group displayed proactive interference during the initial testing with the reversed location. All of the groups except for the SD group showed a gradual decline in the distance spent in the previously correct quadrant. A repeated measures ANOVA showed that the groups differed, $F(4,20)=27.7$, $p<0.001$, and changed over days, $F(7,140)=15.4$, $p<0.001$, but that the change over days did not vary significantly between groups, $F(28,140)=1.2$, $p=0.3$. Post hoc analysis revealed that relative to the Saline group, the Switch and the SD groups spent

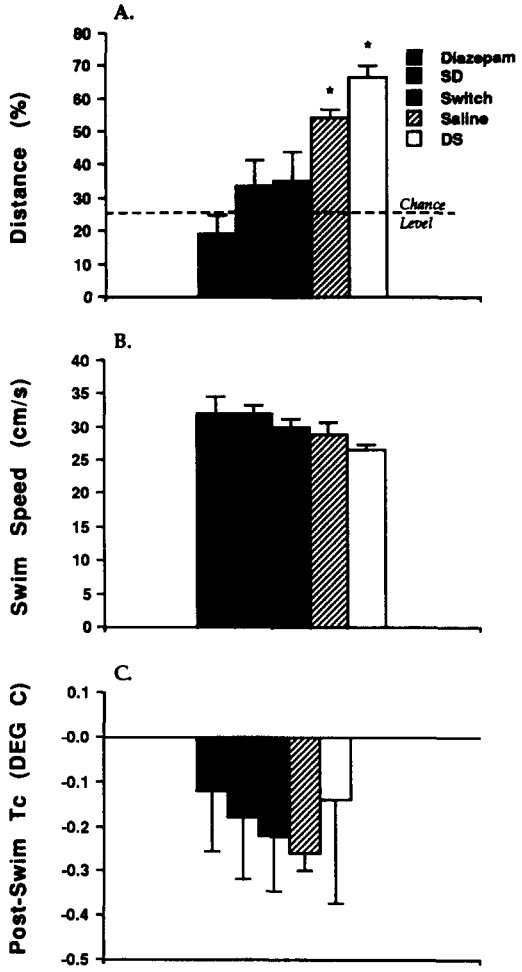

FIG. 6. (A) The percentage distance in the correct quadrant, (B) swim speed, and (C) postswim T_c (preswim − postswim) during the final probe trial. Note that the saline-treated groups (Saline and DS), but not the groups receiving diazepam (Diazepam, SD, Switch), demonstrate a preference for the correct quadrant, despite comparable swim speeds and postswim T_cs. *$p<0.01$ compared to chance level (25%).

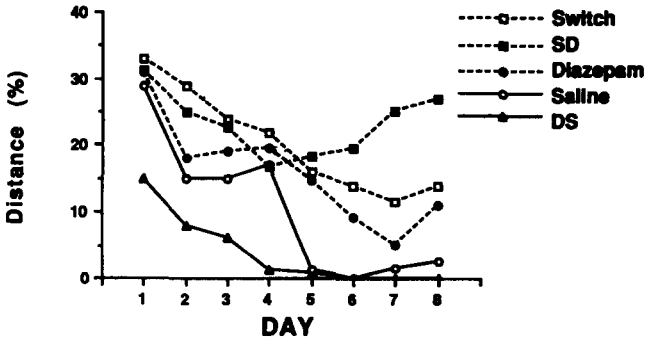

FIG. 7. An illustration of the percentage distance spent in the previously correct quadrant during the reversal phase. Note that the DS group learned quickly while the SD group remained at chance levels throughout testing.

a greater percentage of distance in the old quadrant ($p<0.01$), while the DS group spent a lower percentage of distance in the old quadrant ($p<0.05$). The greatest perseveration to the previously correct quadrant was shown by the SD group which had been originally trained under saline and then reversed to diazepam.

Performance variables were also affected by diazepam during the reversal phase (data not shown). Diazepam-treated rats again swam slower under diazepam relative to the saline-treated control, $F(4,95)=23.9$, $p<0.001$. Those groups receiving diazepam (Diazepam, SD, Switch), as well as the DS group, had slower swim speeds relative to the Saline group ($p<0.05$). The effects on body temperature in the reversal phase were similar to those observed in the acquisition phase. Namely, diazepam reduced preswim body temperatures, $F(4,20)=37.8$, $p<0.001$, but did not exacerbate the swim-induced hypothermia (data not shown). Post hoc tests showed that diazepam-treated rats were different from saline-treated rats before swimming ($p<0.01$), but not after ($p>0.05$).

The effect of diazepam on the probability of rearing while on the platform was minimal and failed to reach statistical significance. However, it might be noted that both the Saline and Diazepam groups showed a dishabituation of rearing when the platform was reversed, but only the Saline group showed a full habituation of rearing by the final day of the reversal phase.

DISCUSSION

The present study found that diazepam-treated rats have severe place-learning deficits as revealed by longer swim paths, slower escape latencies and larger heading errors relative to saline-treated controls. Although diazepam-treated rats also had slower swim speeds and lower core body temperatures throughout acquisition, these could not account for the deficits in place learning. The group switched from saline to diazepam demonstrated total savings, despite reductions of core body temperature and swim speed. Diazepam did not impair acquisition of a visible platform task. When the location of the submerged platform was reversed, only those rats receiving diazepam demonstrated impaired acquisition. The present impairment of acquisition replicates previous findings in the Morris water maze (29,30) and further suggests that this deficit is primarily mnemonic and specific to encoding processes. The following discussion will focus on each of these principle findings individually and in relation to current knowledge of BZD effects.

Diazepam has sedative properties which have two consequences: attentional/perceptual impairments and myorelaxation. In the present study, the place-learning deficit cannot be attributed to the myorelaxation effects of diazepam for three reasons: 1) the Diazepam group swam consistently slower than the Saline group, even as the group reached criterion levels, 2) the Switch group did not show impaired maze performance in spite of a reduction in swim speed, and 3) the Diazepam group did not swim slower during the probe trials. These results suggest that the place-learning deficit produced by diazepam cannot be attributed to myorelaxation side-effects. Neither could the deficits have been due to perceptual/attentional factors. BZD-induced memory impairments have been attributed to deficits of attention in both humans (38) and animals (45). However, in the present study, the Switch group, while receiving diazepam, was still able to navigate to the hidden platform. Because the distance taken to locate the platform did not increase when diazepam was administered, it is likely that the Switch group continued to use the same efficient strategy (42). These findings suggest that the diazepam-treated rat can use, perceive and attend to distal spatial cues to locate the platform.

There were several clear indications that diazepam did not interfere with acquisition of the procedural components of this task. First, the Diazepam group achieved criterion performance levels in initial acquisition, even though the subsequent probe trial showed that they had little knowledge of the platform's location. Second, the Diazepam group did not show an impairment when trained to the visible platform. Third, acquisition of the reversed platform location by the DS group was as fast as the Saline group, despite having been trained under diazepam for all of Phase I. Lastly, the Switch and the SD groups were as impaired as the Diazepam group during reversal training, despite having acquired all procedural components of the task under saline during Phase I. Thus the deficit in learning produced by diazepam was specific to the spatial components of the task.

As mentioned, it seemed possible that the BZD-induced acquisition impairment observed here and previously could have been due to hypothermia. Several studies have demonstrated that diazepam can induce hypothermia [present results, (4, 44, 47)], and hypothermia alone can induce both retrograde (39) and anterograde amnesia (40). Cold water is used to motivate the rat in the Morris water maze. Therefore, the combination of both drug-induced and environmentally induced hypothermia might have been sufficient to impair memory processes. However, three results from the present study argue against this interpretation: 1) the Diazepam group's body temperature did not decrease during swimming more than that of controls, 2) the Diazepam group was impaired but not hypothermic on all three probe trials, and 3) the average change in the body temperature for the groups receiving diazepam was -0.35°C (± 0.1) prior to swimming and -0.21°C (± 0.2) after swimming. A total drop of 0.56°C in body temperature is not as severe as those previously found to induce amnesia, which typically exceed 5°C below normothermia (40). These results suggest that hypothermia did not produce the observed anterograde amnesia.

Previously, the amnesic effects of BZD have been attributed to state-dependent learning (36), but this interpretation does not suit the present findings. Here, rats administered diazepam during both acquisition and retrieval (Diazepam group) were impaired, whereas rats trained under saline and switched to diazepam (Switch group) were not. Furthermore, the group which displayed the most proactive interference in the reversal phase was the one that had been trained under saline and then reversed to diazepam. These results are opposite to what would have been predicted by the state-dependent learning hypothesis (35).

Diazepam has also been shown to reduce experimental indices of anxiety (13). Anxiety has typically been viewed as an impediment to learning and memory [e.g., (10)]. The present results suggest that anxiety is necessary for spatial learning. It was noted incidentally that saline-treated, but not diazepam-treated, rats consistently vocalized when handled suggesting that the diazepam-treated rats were less 'emotional' (1). However, diazepam did not impair acquisition of the visible platform task suggesting that the cue task does not require anxiety for either learning or performance. With the hidden platform task, it appeared that anxiety was not required for performance of the task, only for its acquisition. Further experiments will explore the relationship between anxiety, learning and memory in this task.

Diazepam may have reduced the rate of acquisition by attenuating the aversiveness of the cold water used for motivation but this possibility seems unlikely. In the present study, the Diazepam group swam slower over the course of testing, which would suggest reduced motivation. BZDs have been found to increase opioid activity (46), pain thresholds (22), and the occurrence of punished behaviour (13). Further, naloxone, an opioid antagonist, has been found to facilitate acquisition in the Morris water maze (8). However, BZDs impair place learning in the Morris water maze across a range of water temperatures [19°C (29), 22°C

(present study), and 26°C (30)]. Finally, BZDs also impair appetitively motivated spatial memory tasks (45). These results suggest that the place learning deficit induced by diazepam does not depend on either aversive motivation or diazepam-induced increases in opioid activity.

Although the diazepam groups eventually learned to swim to the submerged platform in both the initial acquisition and reversal phases, it was clear that these rats never really acquired its spatial location. This was demonstrated in the probe trials, in which the diazepam-treated rats swam randomly about the pool, failing to concentrate their search in the correct quadrant. Indeed, it appeared that diazepam produced a total anterograde amnesia and the rats adopted alternative strategies to locate the platform (42). This may have included a 'taxis' strategy, such as swimming towards or away from a single cue, or a 'praxis' strategy, such as swimming in a particular pattern (e.g., sequence of loops). Support for the latter strategy comes from the finding that diazepam-treated rats typically swam in a circular pattern until eventually bumping into the platform. This 'praxis' strategy can be seen in the illustrative swim paths drawn from the first probe trial (Fig. 4). These findings suggest that diazepam produced a severe and persisting anterograde amnesia which necessitated the adoption of a response-based search strategy.

In summary, the present study found that diazepam produces a selective and reversible anterograde amnesia in the Morris water maze. This impairment does not appear to be the result of diazepam-induced myorelaxation, perceptual/attentional impairments, hypothermia, retention/retrieval impairments, or state-dependent learning. Further, diazepam-treated rats appear to adopt nonmnemonic strategies to compensate for their impairment. Overall, these findings replicate prior human and animal investigations and suggests that BZDs play an important role in memory modulation and consolidation. Finally, these results, along with previous findings (29,30), suggest that the Morris water maze can serve as a useful model for dissociating the learning and performance effects of pharmaceuticals as well as an important tool for testing potential nonamnesic, anxiolytic agents.

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