Behavioral dissociations between C57BL/6 and DBA/2 mice on learning and memory tasks: A hippocampal-dysfunction hypothesis

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We recently demonstrated that C57BL/6 (C57) mice are capable of spatial learning in the Morris water task whereas DBA/2 (DBA) mice are impaired. There may be a number of reasons why the DBA mice are impaired in the Morris task. The main objective of this study was to understand the nature of the DBA impairment. First, we showed that DBA mice do not have a general learning impairment, since they can solve two different discrimination problems. Using a water-filled plus maze and a modified version of the Morris task, we showed that DBA mice can (1) see distal cues, (2) attend to the extramaze environment, and (3) learn the place of a hidden platform if training allows them to utilize only part of the distal environment. Probe data indicate, however, that in contrast to C57 mice, DBA mice do not form a complete representation of the extramaze environment. Final experiments demonstrate that DBA mice are also impaired on a task that requires conditional learning but perform as well as C57 mice on a response-tendency task and a nonconditional learning task. The results of these and other studies suggest that learning and memory processes in DBA mice may be disrupted as a result of impaired hippocampal function.

Studies have shown that the inbred strains of mice, C57BL/6 (C57) and DBA/2 (DBA), perform differently on learning and memory tasks. Performance differences between these two strains appear to be task dependent. DBA mice perform better than C57 mice on one- (Weinberger, Koob, & Martinez, 1992) or two-way active avoidance tasks (Schwegler & Lipp, 1981, 1983; Schwegler, Lipp, Van der Loos, & Buselmaier, 1981). However, in most other tasks, DBA mice are impaired relative to the C57 strain. For example, C57 mice perform better on radial-arm mazes (Amassari-Teule & Caprioli, 1985; Rossi-Arnaud, Fagioli, & Ammassari-Teule, 1991), a spontaneous alternation task (Bertholet & Crusio, 1991), a spatial open-field task (Rouillet & Lassalle, 1990), and on a water escape task (Schopke, Wolfer, Lipp, & Leisinger-Trigona, 1991; Wolfer, Lipp, Leisinger-Trigona, & Hausheer-Zarmakupi, 1987). Since the learning and memory processes of C57 and DBA mice appear to be different, these strains can be a unique and useful tool for understanding the neurobiology of learning and memory. Although performance differences between C57 and DBA mice suggest that a general learning deficit in one of the strains is unlikely, the exact nature of the learning difference(s) is unclear.

The Morris water task (Morris, 1981) has become one of the most widely used tasks to study spatial learning in rodents. In one form of the task, spatial cues are irrele-

vant for solution, since the escape platform is (1) visible and (2) typically located in random locations. To solve this task, a subject only needs to see the platform and swim to it for escape. In the other conventional form of the task, the escape platform is located below the surface of the water in a fixed location. For a subject to find the platform, it must learn the multiple spatial relationships between extramaze cues and the relationships of distal cues to the platform.

Using the Morris task, we recently showed that C57 and DBA mice were capable of solving the "nonspatial" form of the task—that is, they were able to learn to navigate to a visible platform. C57 mice were also capable of using distal (i.e., spatial) cues to locate a fixed hidden platform. DBA mice, however, were impaired on the hidden-platform task (Upchurch & Wehner, 1988a, 1988b, 1989).

In the present study, we used a number of other water tasks to try and understand the nature of the DBA impairment. These tasks were chosen because they all involve the same motor skills and are likely to be equally motivating. With these different tasks, we were able to ask a number of questions. First, we determined if discrimination processes were different in C57 and DBA mice, since most learning and memory tasks require the capacity to discriminate visual cues. Second, there are a number of behavioral problems that could produce impairments on the Morris task that are not spatial learning problems per se. To test other alternatives, C57 and DBA mice were tested on a modified version of the Morris task. Finally, another set of experiments was designed to determine if DBA mice are impaired on tasks that require conditional/relational learning processes. Conditional

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learning was tested because it has not been evaluated previously in inbred strains of mice, it is intrinsic to spatial-learning tasks, and lesions to certain brain regions disrupt spatial learning in the Morris task and impair conditional learning (see Gray & McNaughton, 1983, for review; also see Freeman & Stanton, 1991).

Our results suggest that DBA mice have a selective impairment in a learning and memory process that results in poor performance on tasks that require spatial and/or conditional information. Given the behavioral and neurobiological differences between the two strains, we hypothesize that the impaired hippocampal function in DBA mice may underlie the performance differences between these inbred strains of mice.

EXPERIMENT 1

To study visual discrimination learning in C57 and DBA mice, we adapted a water version of the Lashley jump stand developed by Rudy and Castro (1987). In this task, two stimuli are suspended from a partition at one end of an aquarium. The stimuli can vary along any visual dimension (i.e., brightness, pattern, shape, etc.). In addition, the position of the stimuli varies from trial to trial such that the position of the stimuli is irrelevant for solution. The task requires a subject to attend to the proper stimulus dimension (e.g., brightness), to discriminate the difference between the stimuli, and to learn that the escape platform is always located behind only one of the stimuli. Therefore, if DBA mice are poor at spatial tasks because of an inability to discriminate between simple visual cues, they should also be impaired on this task.

Method

Subjects. The subjects were 17 C57BL/6Jbg and 18 DBA/2Jbg male mice. Eight different litters contributed subjects to the four groups. The mice were obtained originally from Jackson Laboratory but now are bred at the Institute for Behavioral Genetics, University of Colorado, Boulder, CO. The mice were 60–120 days of age and maintained on a 12:12-h light:dark cycle with lights on at 0700 h. They were housed in groups of 5 and were tested between 1000 and 1600 h.

Apparatus. The apparatus consisted of an aquarium (45.5 × 22 × 42 cm) that contained a partition holding two stimuli plates, an escape platform, and an observation platform (for a schematic, see Rudy & Castro, 1987, Figure 1). The discriminanda were metal plates (9.2 × 11.5 cm) that were suspended from the partition. There were four different stimuli: black, white, black and white horizontal stripes, and black and white vertical stripes. There were four white (1.5 cm wide) and three black (1 cm wide) stripes on the horizontal and vertical stimuli plates; the only difference was in their orientation. The partition, a clear Plexiglas panel (37 × .5 cm), had a suspending rod that held the discriminanda so that the bottoms were 1.5 cm above the water level and 15 cm from the back of the aquarium. The aquarium was wrapped in gray plastic.

An adjustable Plexiglas escape platform (8.8 × 8.8 cm) was located behind one of the discriminanda with its top 1 cm below the surface of the water. The observation platform (8.8 × 6.4 cm) was located opposite the partition with its top protruding 3 cm above the water. The water was 24 cm deep and was rendered opaque with nontoxic white Crayola powdered paint. The water temperature was maintained at 28°C.

Procedure. Prior to the first trial, a subject was trained to climb onto the escape platform by placing its forepaws against the top and gently pushing itself onto the platform. This procedure was used until a subject readily climbed onto the platform. After pretraining, a subject was given its first trial. The stimuli were put into position, and the subject was placed on the observation platform facing the direction of the escape platform. Approximately 15 sec later, the observation platform was removed, and the subject was placed into the water facing the stimuli. The subject was allowed to swim until it climbed onto the escape platform or until it swam behind the incorrect stimulus plate. If the subject swam to the correct stimulus and climbed onto the platform, it was removed from behind the plate and placed on the observation platform. If the subject swam to the incorrect stimulus, it was trapped for 10 sec and then placed on the observation platform. The intertrial interval was approximately 30 sec, during which the subject remained on the observation platform. While on the observation platform, the experimenter kept the subject's head directed towards the stimuli by lightly tapping on the subject's nose.

For an individual subject, the escape platform was always located behind the same stimulus plate. Each plate was located equally often on either side of the partition. The schedule of alternation was such that the same stimulus plate could never be on the same side for more than two consecutive trials. Eight C57 and 9 DBA mice were trained on the brightness (B/W) problem. Nine C57 and 9 DBA mice were trained on the pattern (H/V) problem. On each problem, half of the subjects were trained with the escape platform behind the black or vertical plate (B+/W−; V+/H−), and the other half were trained with the platform behind the white or horizontal plate (B−/W+; V−/H+).

On Day 1 of training, a subject was given two blocks of 8 trials separated by 1 h. On Day 2 and on subsequent days, a subject was given two blocks of 12 trials separated by 1 h. Each subject was trained until it learned to swim to the correct stimulus plate on 7 of 8 trials within a block of trials, or until 136 trials were given. If a subject never reached criterion, it was given a value of 137.

Results and Discussion

C57 and DBA mice learned to solve both the brightness and pattern discrimination problems (Figure 1). In addition, there was no difference in the number of trials required to reach criterion between the two problems. A two-way analysis of variance (ANOVA) showed the main effect of strain [$F(1,31) = .129, p > .05$], problem [$F(1,31) = 1.266, p > .05$], and the strain × problem interaction [$F(1,31) = .121, p > .05$] were not significantly different.

These data suggest that C57 and DBA mice have the processes necessary for solving simultaneous brightness and pattern discrimination tasks. More importantly, they show that C57 and DBA mouse performance does not differ (see Henderson, 1972). Thus, it is unlikely that DBA mice are impaired on some tasks because of an inability to discriminate among visual stimuli presented simultaneously.

EXPERIMENT 2A

As described above, animals locate a hidden platform by learning relationships between distal (spatial) cues and the escape platform in the place-learning version of the Morris water task. C57 mice are able to solve the place-learning task, whereas DBA mice are impaired (Upchurch

Figure 1. The mean number of trials to reach criterion for C57 and DBA mice on simultaneous brightness (black/white) and pattern (horizontal/vertical) discrimination problems. The bars indicate the SEM.

& Wehner, 1988a, 1988b). Assuming an animal can learn to navigate to a visible platform, there are at least four reasons why it might be impaired on the place-learning task. First, an animal may simply not be able to see stimuli at long distances. Second, even if an animal can see distal cues, it may not attend to them. Third, although an animal may attend to distal cues, it may be unable to make a simple association between even a subset of the distal environment and the escape platform. Finally, an animal may be capable of learning to associate a set of distal cues and the place of the platform but may be unable to learn the multiple relationships among distal cues, and the relationships between distal cues and the escape platform. In the standard hidden-platform version of the Morris water task (i.e., an animal is trained to locate a hidden platform in a fixed location from *multiple* start sites), an animal must have all of these skills to locate the escape platform from any place in the pool. It is important to note that these considerations are relevant for many other spatial tasks in which DBA mice have been found to be impaired (Schopke et al., 1991; Schwegler, Crusio, & Brust, 1990; Schwegler, Crusio, Lipp, & Heimrich, 1988; Wolfer et al., 1987).

In Experiment 2A, we designed a task to determine if DBA mice can see and attend to distal cues. C57 and DBA mice were trained to find a hidden platform in a water-filled plus (+) maze. There are two important aspects of

this task. First, the platform is always located in a fixed position with respect to extramaze cues. Second, an animal is started from one of the three arms that does not contain the platform. Therefore, this task has both a non-spatial and a spatial solution. An animal could solve the task by learning to navigate toward the set of distal cues on the wall directly behind the platform without learning its spatial location. Alternatively, it could learn the spatial location of the platform in the room. Regardless of how an animal solves the task, the minimum requirement is that it must be able to see and attend to distal cues.

Method

Subjects. The subjects were 14 C57BL/6J and 15 DBA/2J male mice from six different litters.

Apparatus. The apparatus was a water-filled + maze constructed from 7 mm thick clear Plexiglas. Each arm was 21.5 cm long, 10.5 cm wide, and 34.5 cm deep. A height-adjustable escape platform (8.8 × 8.8 cm) could be placed at the end of any arm of the maze. The top of the platform was 1 cm below the surface of the water. The water was approximately 15 cm deep and was maintained at 28°C. White nontoxic Crayola powdered paint was added to the water to obscure vision of the platform. The maze was in a room (235 × 195 cm) that had several objects in it, such as a table, black door, and shelves.

Procedures. Prior to the first trial, a subject was placed on the escape platform that was located at the end of one arm. Fifteen seconds later, the subject was placed in the water with its forepaws against the platform and allowed three practice climbs. For any given subject, the escape platform was always located in the same arm with respect to extramaze cues, but its location varied between subjects. On each trial, a subject was placed, facing the center of the +, in one of the three arms that did not contain the platform. The start arm was pseudorandomly chosen, with the restriction that each arm was used in a block of three trials. The subject was allowed to swim into any of the arms. If it chose the correct arm, the subject was allowed to climb onto the platform and then was removed from the maze and placed in a holding tub. If it chose an incorrect arm, it was trapped in that arm 10 sec and then removed from the maze and placed in a holding tub. Each subject was given 12 trials on Day 1, 18 trials on Day 2, and 30 trials on all subsequent days until a criterion of 9 out of 10 correct choices was reached on a given day. The intertrial interval was approximately 2 to 5 min, during which the other subjects were given their trials. This subject rotation occurred among 4 to 6 subjects.

Results and Discussion

Both C57 and DBA mice learned to locate the escape platform in the correct arm (Figure 2). The mean number of trials to reach criterion was not significantly different between the two strains [$t(27) = .233, p > .05$]. These results demonstrate that DBA mice can see and attend to extramaze cues to locate a hidden platform. These data do not necessarily demonstrate that DBA mice learned the place of a hidden platform by navigating toward the set of distal cues directly behind the platform. It could be that allowing DBA mice to locate a hidden platform by approaching only a set of distal cues actually promoted the learning of the relationship among the entire distal environment. This possibility was addressed by using a different task in Experiment 2B.

Figure 2. The mean number of trials for C57 and DBA mice to reach criterion on the plus-maze task. The bars indicate the SEM.

EXPERIMENT 2B

The results of the previous experiments indicate that it is unlikely that DBA mice are impaired on the hidden-platform version of the Morris task (or other spatial tasks) due to impaired visual or attentional processes (see Experiment 1). These data suggest that DBA mice must be impaired because either they cannot learn even a simple association between the place of the platform and a specific set of distal cues or they cannot learn the multiple relationships among distal cues and the relationships of those cues and the platform. It is important to note that the ability to learn the place of a hidden platform using only a specific set of distal cues is *not* spatial learning per se. "True" spatial learning requires knowledge of the multiple relationships between extramaze cues and relationships of those distal cues to the platform.

Hippocampally lesioned rats are impaired on the hidden platform version of the Morris water task (Morris, Garrud, Rawlins, & O'Keefe, 1982; Sutherland, Kolb, & Whishaw, 1982). Eichenbaum, Stewart, and Morris (1990), however, showed that if hippocampally damaged animals (caused by transection of the fornix) were released from the same location on every trial, they could learn the place of the hidden platform. However, in probe trials, they are impaired in locating the platform if they are released from new locations, whereas control rats are not impaired (also see Morris, 1981, Rudy & Paylor, 1988, and Sutherland & Rudy, 1988). A possible reason for these findings is that lesioned animals may learn a simple relationship between limited distal cues and the platform when they are allowed to use the same navigation path repeatedly.

In this experiment, we used a modified version of the Eichenbaum et al. (1990) procedure to determine if C57 and DBA mice can learn the place of a hidden platform when allowed to use only a set of distal cues. The nature of the C57 and DBA representations in this task was determined by using probe trials.

Method

Subjects. The subjects were 8 C57BL/6J and 8 DBA/2J male mice derived from at least four litters.

Apparatus. The training apparatus was a circular, galvanized steel pool, 115 cm in diameter and 60 cm deep. The water level was 25 cm. Water temperature was maintained at 26°C. White and blue nontoxic Crayola paint was added to make the water opaque and to match the color of the pool walls in order to reduce the contrast between the water and the pool wall. The pool was located in the west end of a laboratory room that was 3.96 × 2.84 × 2.51 m. The pool was at least 35 cm away from the nearest extramaze cue. The room contained a number of extramaze cues that were determined to be visible to an animal swimming in the pool. Extramaze cues included a counter that extended the length of the room, a set of shelves, video equipment, black pieces of cardboard attached on one wall, and an experimenter who remained 85 cm from the pool.

The visible platform was 11.5 × 11.5 cm in diameter and 24.5 cm tall. Attached to the platform was a white sail from a child's toy sailboat. The sail was 13.5 cm tall and 7.7 cm wide at the bottom. The top of the platform was 1 cm below the surface of the water. The hidden platform was the same as the visible platform except that the sail was removed.

The behavior of each animal was recorded by a video camera suspended over the pool.

Procedure. Pretraining. Prior to the first trial, each subject was placed on the visible platform, where it remained for 15 sec. The animal was then placed in the water and allowed to swim for 15 sec. It was then placed on the platform for 10 sec and given three practice climbs. The subject was given its first trial 20 sec later.

General training. Each trial was started by removing the animal from the platform and placing it (facing the pool wall) along the edge of the pool. In contrast to standard training procedures, an animal was started from the same place on every trial. The starting location was at a 45° angle from the escape platform. The platform remained in the same location on every trial. During a trial, a subject was allowed 60 sec to locate the platform. If the subject did not reach the platform in the allotted time, it was placed there by the experimenter. The subject was then given 20–25 sec on the platform before being placed in the pool for another trial. A subject was given 4 trials in succession before being put into a holding tub while another subject was trained. This type of rotation occurred among two animals of each strain. Animals were given 12 trials a day.

Phase 1 training. Phase 1 was made up of three types of trials. During Phase 1A, an animal was given 20 trials with the visible platform. During Phase 1B, each animal was given 20 trials in which the platform was either visible (V) or hidden (H). These trials were intermixed in the following schedule: V, H, V, H, V, H, V, V, V, H, H, V, H, V, H, H, V, H, V, H. In Phase 1C, an animal was given 20 trials with a hidden platform. Since the animals were given 12 trials a day for 5 days, some of the days had two different parts of Phase 1 training.

Immediately after the last trial of Phase 1C, each animal was given a probe trial with the platform removed. During this probe trial, a subject was released from the same start location and allowed to search the pool for 60 sec.

Phase 2 training. Phase 2 began on the next day. During this phase of training, the subjects were first given four hidden-platform

trials starting from the same location as that of Phase 1. Each subject was then given another set of four trials. During three of these trials, the animals were started from new locations. For comparison, each animal was also given a trial at the original training site location. Finally, animals were given another four trials at the original start site.

Immediately following the last trial, each animal was given another probe trial. During this probe trial, however, each subject was started either 135° or 225° away from the original start location. Each subject was allowed 60 sec to search the pool during the probe trial.

Dependent variables. During the training phases, escape latencies to locate the platform were recorded. From the probe trial, four measures were obtained to characterize each subject's search pattern: quadrant search time, platform crossings, latency to cross the training site, and heading error. To obtain the search time measure, the pool was conceptually divided into four equal quadrants, and the amount of time the subject spent in each quadrant was determined. Platform crossings were obtained by counting the number of times a subject crossed the place in the training quadrant where the platform had been located during training. For comparison, the number of times a subject crossed the equivalent locations in each of the other three quadrants was determined. Latency to cross was obtained by measuring the time it took a subject to cross the spot where the platform initially had been located during the probe. Heading error was obtained by noting the direction in which the subject's head was pointed once it was approximately 15 cm away from the wall. The distance (in degrees) that this path would take the subject from the platform was then calculated.

Results

Phase 1A. Initially, DBA mice took longer to locate the visible platform compared with the C57 mice, but by the end of Phase 1A, both strains were swimming directly to the platform in less than 10 sec (Figure 3A). A one-way ANOVA with repeated measures revealed no significant main effect of strain [$F(1,13) = 3.114, p = .101$]. There was a main effect of trial number [$F(19,247) = 8.519, p < .0001$], which indicates that, overall, the latency to locate the platform decreased with training. Finally, there was a significant strain $\times$ trial number interaction [$F(19,247) = 1.923, p = .0131$]. Simple effects analysis of the interaction showed that DBA mice had significantly longer escape latencies on trial numbers 1 and 3 but that C57 had longer latencies on trial number 4.

Phase 1B. Both strains of mice continued to swim directly to the platform when it was visible. When the platform was hidden, however, both C57 and DBA mice took longer to escape the water (Figure 3B). C57 mice appeared to improve over the 10 hidden platform trials of Phase 1B. The performance of DBA mice, however, appeared to be quite variable. On some trials, DBA mice appeared better than C57 mice, but on other trials, their latencies were longer. A two-way ANOVA (strain and

Figure 3. The mean time to locate the escape platform for C57 and DBA mice during visible platform (Figure 3A), visible and hidden platform (Figure 3B), and hidden platform (Figure 3C) trials of Experiment 2B (see text for details of the training protocol). The bars indicate the SEM.

platform type as between factors) with repeated measures revealed a main effect of strain [$F(1,26) = 5.839, p = .023$], suggesting that overall C57 mice performed better than DBA mice. There was also a main effect of platform type [$F(1,26) = 52.259, p < .0001$], which indicates that both strains of mice performed better on trials in which the platform was visible than when it was hidden. The main effect of trial number and all interactions were not statistically significant ($ps > .05$).

Phase 1C. Figure 3C shows the performance on the last 20 trials of hidden-platform training. There are two important points to be emphasized. First, C57 mice had lower escape latencies than did DBA mice. Second, even though the DBA mice had longer latencies, their performance improved such that by the end of training they were navigating to the platform in about 10 sec. A one-way ANOVA with repeated measures revealed a main effect of strain [$F(1,13) = 8.923, p = .0105$]. The main ef-

fect of trial number and the strain $\times$ trial number interaction were not significant ($ps > .05$).

Phase 1 Probe. Although C57 mice appeared to spend more time in the correct quadrant compared to DBA mice, both strains selectively searched the place where the platform was located (Figure 4). A one-way ANOVA with repeated measures revealed no main effect of strain [$F(1,13) = .031, p > .05$]. There was a main effect of quadrant [$F(3,39) = 20.848, p < .0001$] and a significant strain $\times$ quadrant interaction [$F(3,39) = 4.542, p = .008$]. Post hoc analysis on the main effect of quadrant (Newman-Keuls, $p < .05$) showed that, overall, animals spent significantly more time in the training quadrant than in the other quadrants that did not differ from each other. Simple effects analysis on the strain $\times$ quadrant interaction showed that C57s spent significantly more time in the training quadrant than did DBA mice ($p = .007$) but that each strain spent more time in the training quadrant

Figure 4. Performance on the probe trial for C57 and DBA mice when they were started from the same site as that of every trial during Phase 1 training. Mean search time, platform crossing, heading error, and latency to cross training site are presented (see text for a description of how these measures were obtained). The bars indicate the SEM.

than in the other quadrants ($ps < .03$). Time spent in the other quadrants did not differ ($ps > .05$).

As with the search time measure, C57 mice crossed the training site more often than did DBA mice. However, both strains crossed the training site more often than they crossed equivalent sites in the other quadrants. A one-way ANOVA with repeated measures showed that the main effect of strain was not significant [$F(1,13) = 1.065$, $p = .3209$], but there was a significant main effect of quadrant [$F(3,39) = 20.591$, $p < .0001$] and strain $\times$ quadrant interaction [$F(3,39) = 4.854$, $p = .0058$]. Post hoc analysis on the main effect of quadrant (Newman-Keuls, $p < .05$) showed that, overall, animals crossed the training site significantly more often than they crossed equivalent sites in the other quadrants. Simple effects analysis on the strain $\times$ quadrant interaction showed that C57 mice crossed the training site more often than did DBA mice ($p = .001$) but that each strain crossed the training site more often than they crossed sites in the other quadrants ($ps < .03$). Platform crossings in the other quadrants did not differ ($ps > .05$).

Both strains of mice initially headed toward the place where the platform was located. The difference between the heading error scores for C57 and DBA mice was not significantly different [$t(13) = .294$, $p > .05$]. Although, in general, DBA mice took longer to locate the hidden platform during Phase 1C training, the latency to cross the training site the first time during the probe was not statistically different between the two strains of mice [$t(13) = -1.177$, $p > .05$].

Phase 2. Performance on the four reinstatement trials was similar to that at the end of Phase 1 training (data not shown). However, the two strains performed differently during the block of trials in which they started from new locations. During this block of four trials, each subject started from the training site once and from new locations on the other three trials. As with the latency to cross measure during Probe 1, the time it took C57 and DBA mice to locate the platform when they started from the original site was not statistically different [$t(13) = 1.617$, $p > .05$]. However, DBA mice took longer to locate the escape platform than did C57 mice when they started from new locations (top panel of Figure 5). The difference between the DBA and C57 mouse performance when the mice started from new sites was significant [$t(13) = 2.77$, $p < .05$].

When the C57 mice were given a probe trial starting from a new location, they still selectively searched the place where the platform had been located. DBA mice, however, performed worse than C57 mice and, more importantly, *did not selectively search* the place where the platform had been located (Figure 5). A one-way ANOVA on the quadrant search time and platform crossing measures confirmed these observations. An analysis of the quadrant search time measure showed no significant main effect of strain ($p > .05$), but there was a significant main effect of quadrant [$F(3,39) = 23.047$, $p <$

$.0001$] and a significant strain $\times$ quadrant interaction [$F(3,39) = 8.083$, $p = .0003$]. Post hoc analysis on the main effect of quadrant (Newman-Keuls, $p < .05$) showed that, overall, animals spent significantly more time in the training quadrant than in the other quadrants. Simple effects analysis on the strain $\times$ quadrant interaction showed that C57 spent significantly more time in the training quadrant than did DBA mice ($p = .007$) and C57 mice spent more time in the training quadrant than in the other quadrants ($ps < .001$). DBA mice, however, did not spend more time in the training quadrant than in the other quadrants ($p > .05$).

Similarly, C57 mice crossed the correct place where the platform had been located more often than they crossed equivalent sites in the other three quadrants. DBA mice did not cross the training site more often than they crossed the sites in the other three quadrants. A one-way ANOVA with repeated measures revealed a significant main effect of strain [$F(1,13) = 13.319$, $p = .0029$], quadrant [$F(3,39) = 31.629$, $p < .0001$], and strain $\times$ quadrant interaction [$F(3,39) = 12.665$, $p < .0001$]. Post hoc analysis on the main effect of quadrant (Newman-Keuls, $p < .05$) showed that, overall, animals crossed the platform site in the training quadrant more often than they crossed the sites in the other quadrants. Simple effects analysis on the strain $\times$ quadrant interaction showed that C57 mice crossed the training site more often than they crossed the other sites ($p < .0001$). DBA mice, however, did not cross the training site more often than they crossed the sites in the other quadrants ($p > .05$).

In addition, C57 mice had significantly smaller heading-error scores than did DBA mice [$t(13) = -3.763$, $p < .003$]. C57 mice also crossed the training site for the first time during the probe faster than did the DBA mice. This difference was nearly significant [$t(13) = -2.11$, $p = .054$].

Discussion

The results from Experiment 2B suggest that DBA mice are capable of learning where a platform is located if they are started from the same place on every trial. DBA mice searched the place where the platform was located when they were started from the same site, indicating they were not locating the hidden platform on the basis of some type of learned swim pattern. However, that DBA mice did not selectively search the correct place when started from a new location suggests that they learned the location of the platform during Phase 1 by utilizing the set of distal cues that could be seen from that specific start location. That is, DBA mice appeared not to have a representation of the entire extramaze environment that is needed to guide their search when they are started from new places. In contrast, C57 mice selectively searched the place where the platform was located independent of their starting location. Thus, C57 mice had a representation of the extramaze environment that could be used to locate the platform from any place in the pool.

Figure 5. The top portion presents the time taken to locate the hidden platform for C57 and DBA mice when they were released from new sites (see text for details). The bottom part of the figure shows the performance on the probe trial when subjects were started from a novel location. The bars indicate the SEM.

Together with the results from Experiment 2A, several alternative explanations can be eliminated that could account for the DBA spatial-learning impairment when they are trained to locate a hidden platform from multiple start locations (Upchurch & Wehner, 1988a, 1988b). It is unlikely that DBA mice are impaired at the spatial-learning version of the Morris task because they cannot see extramaze cues, since they could solve the plus maze task

of Experiment 2A, and they were able to learn the place of the platform in Experiment 2B when started repeatedly from the same site. Similarly, these same data show that DBA mice do attend to extramaze cues. Finally, data from Experiment 2B suggest that they can learn to associate the place of a hidden platform with a set of distal cues but that they do not have a representation that can be used to guide their search when they are started from places

where those distal cues are not readily visible. Data from C57 mice, however, indicate that they have the necessary processes to perform all of these tasks.

EXPERIMENT 3A

In the standard hidden-platform version of the Morris task (Morris, 1981), successful location of the platform using a spatial search strategy is dependent on an animal's ability to learn and remember that the information provided by distal cues is conditional on where that animal is positioned at any time in the pool. Thus, an animal may be impaired not because it has dysfunctional spatial-learning and memory processes, but because it cannot learn to utilize *any* information in a conditional manner. In addition, damage to certain brain regions such as the hippocampus impairs both spatial (Morris et al., 1982; Sutherland et al., 1982) and conditional learning (e.g., Freeman & Stanton, 1991). Since inbred strains of mice have not been tested on conditional learning tasks, this last set of experiments determined if inbred strains of mice can solve conditional tasks and if there are performance differences between C57 and DBA mice.

A water version of a conditional spatial-discrimination task (CSD task) was recently developed to study learning and memory processes in the developing rat (Castro, Paylor, & Rudy, 1987). As the name implies, this task has conditional, spatial, and discrimination components. However, these components can be dissociated with a number of control experiments. The CSD task, therefore, can be used to determine if mice can solve conditional tasks and, more importantly, can address whether the DBA mouse is impaired at conditional tasks independent of spatial processing.

Method

Subjects. The subjects were 6 C57BL/6J and 6 DBA/2J male mice. Three litters contributed subjects to the two groups.

Apparatus. The same + maze used in Experiment 1 was used for the CSD task. One arm of the maze, however, was blocked off with a Plexiglas insert, thus creating a T configuration. The water level was 15 cm deep and maintained at 28°C. An escape platform could be placed at either end of an arm, 1 cm below the surface of the water. Each arm could be blocked with a Plexiglas insert.

Procedures. Prior to the first trial, a subject was placed on the platform positioned at the end of an arm for 15 sec. The subject was then given three practice climbs as described above. The platform was then moved to the other arm, and the same procedure was used. After this pretraining, the subject was ready for its first trial. Each trial consisted of a forced run and a choice run. On the forced run, one arm of the maze was blocked, and the escape platform was placed in the unblocked arm. On the choice run, neither arm was blocked, and the escape platform was placed in the arm that previously had been blocked. The forced run began by placing the subject in the stem and ended when the mouse reached the escape platform. The mouse was allowed to remain on the escape platform for 10 sec. The subject was then placed in a holding tub (< 10 sec) while the maze was prepared for the choice run. On the choice run, the subject was placed in the stem and allowed to swim down either arm. If the subject entered the correct arm, it was allowed to climb onto the escape platform and was then placed in

the holding tub. If the subject chose the incorrect arm, it was trapped for 10 sec before being returned to the holding tub. The arm that was blocked on any one trial was determined randomly, with the provision that the correct choice on the choice run could not be the same arm for more than 2 consecutive trials. Four subjects were rotated through this procedure such that the intertrial interval was approximately 2–3 min. On Day 1, the subjects were given 10 trials. On Day 2, the subjects were given 20 trials. On Day 3, and throughout the rest of training, the subjects were given 30 trials. A subject was trained until a criterion of 9 out of 10 correct responses on the choice run was achieved, or until 240 trials were given. If a subject failed to learn to solve the task within 240 trials, it was given a value of 241. The subjects were trained 5 days a week.

Results and Discussion

Figure 6 shows that C57 mice learned to solve the CSD task. DBA mice, however, were severely impaired. In fact, after 240 trials, none of the DBA mice had reached criterion. The mean number of trials to reach criterion for C57 and DBA mice was significantly different [$t(10) = 5.6003, p < .00025$].

These data show that the CSD task can be used with some strains of mice to study conditional learning and memory processes. More importantly, the CSD task clearly dissociated performance of C57 and DBA mice. These findings alone, however, do not indicate why the DBA mice are impaired on the task. The next two parts of Experiment 3 were devoted to understanding the nature of the CSD impairment.

EXPERIMENT 3B

There are a number of possibilities why C57 mice perform better than DBA mice on the CSD task. The first of these possibilities was tested in Experiment 3B. It could

Figure 6. The mean number of trials to reach criterion for C57 and DBA mice on a conditional spatial discrimination task. The bars indicate the SEM.

be that different inbred strains of mice have different response tendencies that might either facilitate (C57) or compete (DBA) with choosing the correct arm on the choice run (e.g., Castro et al., 1987; Freeman & Stanton, 1991; Green & Stanton, 1989). For example, C57 mice may spontaneously alternate more readily than DBA mice after being forced into one arm of the maze. Alternatively, DBA mice might be poor in the CSD task because they have some strong stimulus bias that makes them predisposed to swim down the arm chosen on the previous forced run. Experiment 3B was designed to determine what response tendencies C57 and DBA mice bring to the CSD task, independent of reinforcement contingencies.

Method

Subjects. The subjects were 6 C57BL/6J and 6 DBA/2J male mice from at least two different litters.

Apparatus. The apparatus was the same as that in Experiment 3A.

Procedures. As with the CSD task, each trial was made up of a forced run and a choice run. On the forced run, one arm of the T was blocked with a Plexiglas insert so that an animal had to swim down the unblocked arm to escape. The arm that was blocked was randomly chosen, with the exception that the same arm could not be blocked for more than two trials and that both arms were blocked equally often within a block of 10 trials. On the choice run, both arms were open, and an escape platform was placed in both arms. Animals were given 10 trials on Day 1, 20 trials on Day 2, and 30 trials on the subsequent days, until each animal received 100 trials (the number of trials that most [4 out of 6] C57 mice needed to learn the CSD task).

Dependent variables. Two measures were obtained for each subject. First, an animal's *stimulus bias* was determined by dividing training into blocks of 10 trials and calculating the percentage of trials on which the animal chose the same arm on the choice run that it was forced into on the forced run. Note that an animal's tendency to alternate spontaneously is the inverse of its stimulus bias. A subject's *response bias* was also determined. For each block of 10 trials, the arm that a subject chose most frequently was determined. The percentage of trials in which a subject chose that arm served as the response bias measure. This procedure allows for an animal to shift its response bias during training.

Results

The mean stimulus bias and response bias measures for C57 and DBA mice are presented in Figure 7. There are a number of important points in these data. Most importantly, there were no differences between the C57 and DBA mice on either the stimulus or response bias measure. Second, both strains' stimulus bias measures were approximately 50%. As noted above, the inverse of this measure is an animal's tendency to alternate spontaneously. Therefore, both strains tended to alternate on only about 50% of the trials. Finally, on the choice run, both strains tended to return to the same arm that they were forced into on the forced run, and this tendency increased over training. These observations were supported by a one-way ANOVA with repeated measures for both the stimulus bias and response bias data. There was no significant main effect of strain for either the stimulus bias [$F(1,10) = 2.438, p = .1495$] or response bias [$F(1,10) = 2.532, p = .1427$] data. The main effect of trial block

Figure 7. The top panel shows stimulus bias data for C57 and DBA mice on the response tendency task. The bottom panel presents their response bias data. The text describes how these measures were obtained. The bars indicate the SEM.

number was not significant for the stimulus bias data [$F(9,90) = .748, p > .05$] but was significant for the response bias data [$F(9,90) = 3.573, p = .0008$], indicating that both strains' tendency to go back to the same arm on the choice run as that on the forced run increased. Finally, strain $\times$ trial block number interactions for both stimulus bias [$F(9,90) = .715, p > .05$] and response bias [$F(9,90) = .421, p > .05$] data were not significant.

Discussion

These results indicate that the reason C57 mice performed better than DBA mice on the CSD task (Experiment 3A) is probably not because of differences in some predisposed response tendency that facilitated C57 performance or that was incompatible with DBA's ability to solve the CSD task.

It is interesting that C57 and DBA mice's spontaneous alternation rates have been studied previously. Bertholet and Crusio (1991) showed that C57 mice have a higher alternation rate after being forced into an arm of a T maze than do DBA mice. One major difference between the

Bertholet and Crusio (1991) study and the present one is that the maze in the former study was not water-filled. We (Castro et al., 1987) and Means (1988) have shown that animals' performance in a maze is different when they are motivated to escape from water than when they are locating food. When food is the reward, an animal learns to find it more easily if it is not located in the same arm on the choice run as it was on the forced run. The opposite is true during escape from water: An animal more easily learns to locate an escape platform on a choice run if the platform is positioned in the same place as it was during the forced run. Given this, there is reason to believe that C57 and DBA mouse alternation rates may be different during water escape than during food search (Bertholet & Crusio, 1991).

EXPERIMENT 3C

It is unlikely that DBA mice were impaired in the CSD task because they have some response tendency that would compete with their ability to learn the solution of the task. There are, however, a number of other reasons why they could have been impaired. Extramaze cues are always in the same spatial location in the CSD task; therefore, one reason that DBA mice were impaired on the CSD task could be that they were unable to discriminate the two spatial locations of the arms. However, although spatial cues are redundant and may be useful, they are not necessary for solution. It could be possible for an animal to solve the task by simply using a response strategy (e.g., if forced left, then turn right on the choice run). Since this type of strategy is also useful, a second possible reason that DBA mice have difficulty could be that they cannot discriminate between left and right responses. Third, DBA mice might be unable to inhibit the previously reinforced forced-run response on the subsequent choice run. Fourth, they may be unable to discriminate between the forced-run and choice-run components of the CSD problem. Finally, they may have impaired conditional learning processes—that is, they may not be able to use the information from the forced run in the conditional manner that is required to make the correct choice on the choice run.

In Experiment 3C, C57 and DBA mice were required to solve a nonconditional spatial-discrimination task (NCSD). The NCSD task was designed so that the position of the platform on the choice-run was *not conditional* on where it was on the forced run. That is, an animal is forced to either arm on the forced run, but on the choice run, the platform is always located in the same arm. If DBA mice were impaired on the CSD task because they could not learn to discriminate between the two places/responses, to inhibit previously reinforced responses, or to discriminate between forced and choice runs, then they should be impaired on this task.

Method

Subjects. The subjects were 6 C57BL/6J and 6 DBA/2J male mice from three different litters.

Apparatus. The apparatus was the same maze used for the CSD task in Experiment 3A.

Procedures. As in the CSD task, each trial consisted of two components: a forced run and a choice run. Unlike the CSD task, however, the position of the platform on the choice run was *not contingent* on the position of the platform on the forced run. On the forced run, one arm of the maze was blocked, and the escape platform was located in the unblocked arm. On the choice run, both arms were open, but the escape platform was *always* located in the same arm. Note that for this version of the problem, information on the forced run was irrelevant to where the platform was located on the choice run, but the subjects had to determine if it was a forced or a choice run and to learn where to go on the choice run. Half of the subjects were trained with the platform on the choice run in the right arm; the other half were trained with it in the left arm. The subjects were given the same number of trials on each day as that in the CSD task until a criterion of 9 out of 10 correct responses on the choice run was made. Immediately after reaching the criterion, the subjects were placed back into their home cages.

Twenty-four hours after reaching criterion, the subjects were required to make five additional correct responses on choice runs to the same arm that was reinforced during training. Immediately following those five trials, the position of the escape platform on the choice run was reversed to the opposite arm. The subjects were then retrained to a criterion of 9 out of 10 correct responses.

Results and Discussion

All of the subjects learned to solve the task (Figure 8). C57 and DBA mice did not differ on the number of trials to reach criterion during original training, nor did the two strains' performance differ on reversal learning. A one-way ANOVA with a repeated measure revealed no significant main effect of strain [$F(1,10) = .197, p > .05$], trial type [original compared with reversal; $F(1,10) = .001, p > .05$], or strain $\times$ trial type interaction [$F(1,10) = .05, p > .05$].

Given the design of the experiment, the pattern of results suggests that DBA mice are able to discriminate the two places/responses, inhibit previously reinforced responses, and discriminate between forced and choice runs. The reversal data indicates that DBA mice can modify

Figure 8. The mean number of trials to reach criterion on the non-conditional spatial discrimination task and the reversal problem for C57 and DBA mice. The bars indicate the SEM.

behavioral responses that were previously reinforced as readily as can C57 mice.

DISCUSSION OF EXPERIMENTS 3A, 3B, AND 3C

Data from this set of experiments show that DBA mice have impaired processes necessary to solve the CSD task. DBA mice did not differ from C57 mice on predisposed response tendencies. Also, they had no difficulty learning to discriminate between the two platform positions, to inhibit reinforced responses, or to determine if it was currently a forced run or a choice run. These results suggest that DBA mice are impaired on the CSD task, not because of the task's spatial component, since they solve the NCSD task, but because they have difficulty using the information acquired on the forced run to guide their response on the choice run.

GENERAL DISCUSSION

In these experiments, we sought to understand better the learning and memory performance differences between C57 and DBA mice. We showed that DBA mice perform as well as do C57 mice on tasks that require the ability to (1) discriminate between stimuli that differ in either brightness or pattern, (2) locate a hidden platform in a plus maze, (3) learn the place where a hidden platform is located in the Morris task when they are trained with procedures that allow them to utilize only part of the entire distal cue environment, and (4) make a correct choice-run response, if the response is not conditional on information acquired on a previous forced run. DBA mice, however, are impaired on tasks that require them to utilize spatial and/or conditional information.

To understand the behavioral differences, the underlying neural substrates that may be different between C57 and DBA mice need to be considered. We believe that DBA mice may be impaired on specific types of learning and memory problems because they have a hippocampus that may be impaired in contrast to the hippocampus of C57 mice. There are two main pieces of evidence to support the notion that DBA mice may suffer from some type of "hippocampal dysfunction."

First, a large body of research indicates that there are both hippocampus-independent and hippocampus dependent learning and memory tasks. Although investigations of performance on tasks by hippocampally lesioned animals have produced a number of theories of hippocampal function (e.g., Cohen & Squire, 1980; DeLong, 1992; Eichenbaum, Otto, & Cohen, 1992; Hirsh, 1974; Mishkin, Malamut, & Bachevalier, 1984; O'Keefe & Nadel, 1978; Olton, Becker, & Handelman, 1979; Rawlins, 1985; Squire, 1987; Sutherland & Rudy, 1989; Teyler & DiScenna, 1986), we are only in a position to describe some tasks that are sensitive to hippocampal dysfunction and their possible relevance to understanding the performance differences between C57 and DBA mice.

One task that clearly depends on hippocampal function is the hidden-platform version of the Morris task (Morris et al., 1982; Sutherland et al., 1982). Although hippocampally lesioned rats can learn to navigate to a visible platform, they are severely impaired at learning the location of a hidden platform using multiple distal cues. Interestingly, fornix-lesioned rats (i.e., those that have a transection of a major source of hippocampal input) can learn the place of a hidden platform when they are trained from the same start location on every trial. However, probe trial data show that fornix-lesioned subjects' representation does not include multiple distal cues, since they are impaired at locating the hidden platform when they are started from new locations (Eichenbaum et al., 1991).

DBA mouse performance in the Morris task parallels that of hippocampally damaged rats. DBA mice can also learn to navigate to a visible platform but are impaired at learning the location of a hidden platform using multiple distal cues (Upchurch & Wehner, 1988a, 1988b). The present study showed that, similar to hippocampally damaged rats (albeit with a fornix transection), DBA mice can learn the place of a hidden platform when they are started from the same start location on every trial. Contrasted to C57 mice, however, they were impaired when they were started from new locations, and they did not selectively search the place where the platform was located on a probe trial when they were started from a new location (see Experiment 2B).

Tasks that require conditional learning and memory processes are also sensitive to hippocampal damage in humans, nonhuman primates, and rodents (Hirsh, 1974, 1980; Mahut, Zola-Morgan, & Moss, 1982; Raffaele & Olton, 1988; Sutherland & Rudy, 1989). Recently, Stanton and colleagues (Freeman & Stanton, 1991; Green & Stanton, 1989) studied the neural basis of the ontogeny of conditional learning by using a discrete-trial delayed alternation task. Their findings are relevant to the present study since the discrete-trial delayed alternation task is quite similar to the CSD task. They showed that fornix-transection impairs the development of the capacity to solve a delayed alternation task but has no effect on the ontogeny of simple position discrimination learning (Freeman & Stanton, 1991). As with hippocampally lesioned rodents, Experiment 3 clearly showed that DBA mice are impaired in a task that involves conditional learning but are capable of learning other "nonconditional" aspects of the task.

Finally, hippocampally lesioned rats are also impaired on other tasks that involve spatial learning such as the radial-arm maze and spontaneous alternation task (see Gray & McNaughton, 1983, for a review). Compared with C57 mice, DBA mice are also impaired in the eight-arm radial maze (Rossi-Arnaud et al., 1991; Schwegler et al., 1990) and spontaneous alternation task (Bertholet & Crusio, 1991).

Not all tasks are sensitive to hippocampal lesions. Tasks that require discrimination processes can be solved by an-

imals without a hippocampus (Berger & Orr, 1983; Douglas, 1967; Douglas & Pribram, 1966; Teitelbaum, 1964; but see Eichenbaum, Cohen, Otto, & Wible, 1991, for hippocampal involvement in odor discrimination). Alvarado and Rudy (1991) have shown that hippocampally lesioned rats can solve simultaneous brightness and pattern discrimination problems in a task identical to that used in Experiment 1. Similar to hippocampally lesioned rodents, DBA mice solve both brightness and pattern discrimination tasks as well as C57 mice.

Interestingly, there are tasks in which hippocampally lesioned animals actually perform better than control subjects. Rats with hippocampal lesions perform better than controls in two-way avoidance tasks (see Gray & McNaughton, 1983). If DBA mouse performance is like that of hippocampally damaged animals, they too should perform better than C57 mice on those same tasks. Schwegler and Lipp (1981), have shown that DBA mice do indeed perform better than C57 mice on two-way avoidance learning tasks. The exact reason for the improved performance by hippocampally lesioned rats and DBA mice is unknown.

Finally, if DBA mice are impaired on some tasks because of hippocampal dysfunction, their performance should be unaffected by a hippocampal lesion when they are tested on hippocampus-dependent tasks. In contrast, if C57 mice perform better than DBA mice because of their having a hippocampus that functions better, then they should be affected by hippocampal lesions. Rossi-Arnaud et al., (1991) tested C57 and DBA mice with hippocampal or amygdala lesions in an eight-arm-radial-maze task. Only the hippocampal-lesion data will be discussed. They showed that C57 mouse performance (as measured by the number of arms chosen prior to first error) was impaired by hippocampal lesions. The number of arms chosen before the first error for DBA mice, however, was not different in hippocampally lesioned subjects.

Taken together, these data show that there is striking similarity between performance of hippocampally lesioned rodents relative to shams, and DBA mice relative to C57 mice. This parallelism is true not only for tasks in which hippocampal lesions produce impaired performance, but also for tasks in which there is no effect or enhanced performance.

The second reason to believe that hippocampal dysfunction in DBA mice might underlie their poor performance comes from the fact that their hippocampus is morphologically and biochemically different from that of C57 mice (e.g., Ingram & Corfman, 1980). For example, relative to C57 mice, DBA mice appear to have a blunted intra/infrapyramidal mossy fiber projection (iip-MF) system in the CA3 region. Extensive correlational studies with a number of inbred strains of mice, including C57 and DBA mice, have suggested that the extent of the iip-MF system is positively correlated to performance on a radial-arm-maze test of working memory (Crusio, Schwegler, & Lipp, 1987; Schwegler et al., 1990), measures of spatial reference memory in a navigation task (Schopke et al., 1991), and response to novelty in an

open-field task (Crusio, Schwegler, & van Abeelen, 1989; also see Roulet & Lassalle, 1990). A negative correlation has also been observed between the iip-MF system and two-way active avoidance (Schwegler & Lipp, 1981, 1983). Importantly, other measures of performance, such as nonspatial reference memory, showed no significant correlations (Schwegler et al., 1990). Thus, C57 and DBA mice have different iip-MF projection-system morphology, and the iip-MF system appears to contribute to performance in hippocampus-dependent tasks.

We have evaluated protein kinase C (PKC) activity in C57 and DBA mice (Wehner, Sleight, & Upchurch, 1990). A number of recent reports have implicated PKC in learning and memory processes in both invertebrate and mammalian systems (Akers, Lovinger, Colley, Linder, & Routtenberg, 1986; Bank, DeWeer, Kuzirian, Rasmussen, & Alkon, 1988; Bank, LoTurco, & Alkon, 1989; Lester & Alkon, 1991; Olds, Anderson, McPhie, Staten, & Alkon, 1989; Olds et al., 1990; Paylor, Rudy, & Wehner, 1991; Routtenberg, 1991; Scharenberg, Olds, Schreurs, Craig, & Alkon, 1991; Schwartz, Calignano, & Sacktor, 1990). Importantly, PKC has been shown to contribute to performance in the Morris water task in both the mature (Olds et al., 1990; Paylor et al., 1991) and developing rat (Paylor, Morrison, Rudy, Waltrip, & Wehner, 1992).

We have shown that DBA mice have reduced levels of hippocampal (*but not cortical*) PKC activity relative to C57 mice (Wehner et al., 1990). The difference in hippocampal PKC activity and the relationship of this difference to spatial learning performance does not appear to be a fortuitous strain difference, because recombinant inbred strains created by crossing C57 with DBA mice show a highly significant positive correlation between hippocampal PKC activity and spatial learning performance in the Morris task as measured by platform crossings and quadrant search time (Wehner et al., 1990). Furthermore, binding of phorbol esters, one marker of brain PKC, is greater in C57 than in DBA mice, and these differences are restricted to the various subfields of the hippocampus (Paylor, Pauly, & Wehner, 1992). These findings also show that C57 and DBA hippocampal systems are different and that this difference(s) may relate to performance in the Morris task. Whether this relationship between hippocampal PKC and spatial learning performance will generalize to other learning and memory tasks remains to be determined using the recombinant inbred strain approach.

Previous examinations of biochemical regulation of learning differences in C57 and DBA mice have largely focused on the cholinergic system (Amassari-Teule & Caprioli, 1985; Mandel, Ayad, Hermetet, & Ebel, 1974; Pick & Yanai, 1989; Upchurch & Wehner, 1987, 1988a). Although differences in putative cholinergic perikarya, choline acetyltransferase, and acetylcholinesterase activities have been reported in cortical and forebrain regions, and may contribute to performance differences (Albanese, Gozzo, Iacopino, & Altavista, 1985; Mandel et al., 1974; Smolen, Smolen, Wehner, & Collins, 1985), less defini-

tive strain differences have been found in hippocampal cholinergic function. Although C57 male mice were reported to have fewer total hippocampal muscarinic receptors as measured by the binding of ^3H -QNB in membrane preparations (Marks, Patinkin, Artman, Burch, & Collins, 1981) and fewer high affinity binding sites as determined by inhibition of L- ^3H QNB binding (Marks, Romm, & Collins, 1987), localization of ^3H -QNB binding sites by autoradiographic techniques did not reveal any striking distinctions between C57 and DBA hippocampus (unpublished data, this laboratory). M $_1$ muscarinic cholinergic receptors as measured by ^3H -pirenzepine (Marks et al., 1987) or nicotinic receptor numbers (Marks, Romm, Gaffney, & Collins, 1986) are also not different between the two strains. In addition, agonist-induced phosphatidyl inositol (PI) turnover is not different in hippocampal slice preparations from the two strains (unpublished results, this laboratory). These studies suggest that if hippocampal cholinergic function is different between the two strains, it is not apparent at the receptor level or in the PI second messenger system.

Given all of these findings, we believe that there is good reason to believe that DBA mice's poor performance on certain tasks results from their having a hippocampus that is unable to process certain types of information. At this point, however, this is simply a working hypothesis and requires further research. For example, it is important to determine if the mechanisms underlying the spatial impairment are the same or related to the mechanisms contributing to the CSD impairment. That is, are DBA mice impaired on both the Morris and CSD tasks because of some common processes necessary to solve both types of problems, or are they two distinct impairments? We believe that we will be able to address this question by testing the C57 $\times$ DBA recombinant inbred strains on both tasks. In addition, C57 and DBA mice need to be tested on a number of other tasks thought to require hippocampal function, such as transverse and negative patterning problems (Alvarado & Rudy, 1991; Sutherland & Rudy, 1989), and other neurobiological systems (i.e., transmitter receptors and second messengers) need to be examined. PKC is not the only intracellular messenger system implicated in learning and memory. Arachidonic acid, nitric oxide, and Ca^{++} /calmodulin dependent protein kinase II systems have all been implicated as playing a role in learning and memory (e.g., Alkon, 1987; Alkon & Nelson, 1990; Goelet, Castellucci, Schacher, & Kandel, 1986; Greenberg, Castellucci, Bayley, & Schwartz, 1987; Kennedy, 1987; Matzel, Lederhendler, & Alkon, 1990; Schuman & Madison, 1991; Schwartz & Greenberg, 1987; Silva, Paylor, Wehner, & Tonegawa, 1992; Williams, Errington, Lynch, & Bliss, 1989). Strain differences in these systems might also account for performance differences between C57 and DBA mice.

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