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Early Postnatal X-irradiation of the Hippocampus and Discrimination Learning in Adult Rats

Russell A. Gazzara and Joseph Altman
Laboratory of Developmental Neurobiology
Department of Biological Sciences
Purdue University

Rats with X-irradiation-produced degranulation of the hippocampal dentate gyrus were trained in the acquisition and reversal of simultaneous visual and tactile discriminations in a T-maze. These experiments employed the same treatment, apparatus, and procedure but varied in task difficulty. In the brightness and roughness discriminations, the irradiated rats were not handicapped in acquiring or reversing discriminations of low or low-moderate task difficulty. However, these rats were handicapped in acquiring and reversing discriminations of moderate and high task difficulty. In a Black/White discrimination, in which the stimuli were restricted to the goal-arm walls, the irradiated rats were handicapped in the acquisition (low task difficulty) and reversal (moderate task difficulty) phases of the task. These results suggest that the irradiated rats were not handicapped when the noticeability of the stimuli was high, irrespective of modality used, but were handicapped when the noticeability of the stimuli was low. In addition, these results are consistent with the hypothesis that rats with hippocampal damage are inattentive due to hyperactivity.

Several studies have shown that rats with hippocampal lesions are not handicapped in acquiring easy simultaneous visual discriminations (e.g., Kimble, 1963; Kimble & Kimble, 1970; Silveira & Kimble, 1968; control *M* trials = 31, 51, and 49.3, respectively) but are handicapped in acquiring a more difficult successive visual discrimination (Kimble, 1963; control *M* = 55 trials). However, other studies have shown that rats with hippocampal lesions are handicapped in acquiring easy simultaneous visual discriminations (Duncan & Duncan, 1971; Niki, 1962; control *Mdn* trials = 38 and 30, respectively) but are not handicapped in acquiring a difficult simultaneous visual discrimination (Stevens & Cowey, 1974; control *Mdn* = 82.5 errors). Experiments dealing with discrimination reversal learning have generally demonstrated learning deficits in rats with hippocampal lesions (e.g., Greene, 1971; Jarrard, 1976; Kimble & Kimble, 1965; Silveira & Kimble, 1968), but an absence of deficit in reversal learning has also been found (e.g., Stevens, 1973). These conflicting results may be due to several factors, including differences in lesion technique and testing procedure.

Studies investigating discrimination learning in rats with hippocampal lesions generally employ either ablation or electrolytic lesions. These techniques lead to great variations in the amount and location of hippocampal tissue damaged. This makes the interpretation of available results difficult because variations in the size (Coscina & Lash, 1969; Douglas & Isaacson, 1964; Green, Beatty, & Schwartzbaum, 1967; Rabe & Haddad, 1968; Snyder & Isaacson, 1965) or the location (Hughes, 1965; Nadel, 1968; Stevens & Cowey, 1973) of hippocampal lesions produce different behavioral effects. In the seven experiments reported here, the

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Requests for reprints should be sent to Russell A. Gazzara, who is now at 58-242 NPI, University of California, 760 Westwood Plaza, Los Angeles, California 90024

acquisition and reversal of simultaneous discriminations (varied from easy to difficult in both visual and tactile sensory modalities) were investigated in rats in which a single reproducible technique for producing hippocampal damage was used. Focal X-irradiation of the hippocampus of rats between Postnatal Day 2 and Postnatal Day 15 prevents the formation of 85% of the normally occurring granule cells of the dentate gyrus (Bayer & Altman, 1975; Gerbrandt, Rose, Wheeler, & Lynch, 1978; Hirsh, Holt, & Mosseri, 1978). This reduction in dentate granule cells has been shown to produce behavioral effects similar to those found after hippocampal lesions (Bayer, Brunner, Hine, & Altman, 1973; Haggbloom, Brunner, & Bayer, 1974). In addition, results from our laboratory suggest that virtually no recovery of function takes place in rats X-irradiated during infancy (Brunner & Altman, 1974). To facilitate comparisons among the seven experiments, we used the same testing apparatus (T-maze) and training procedure throughout.

In the first three experiments, we examined the ability of irradiated rats to learn the acquisition and reversal of visual brightness discriminations which were varied in difficulty from low (Experiment 1A, Bright/Dark) to moderate (Experiment 1B, Bright/Dim #1) to high (Experiment 1C, Bright/Dim #2) as defined by the mean number of trials required by control rats to learn the acquisition phase of the discriminations (low, 66.3–72.5 trials; moderate, 95.3–96.5 trials; high, 173.3–300.0 trials). In the second three experiments, we examined the ability of irradiated rats to learn the acquisition and reversal of tactile roughness discriminations which were varied in difficulty from low (Experiment 2A, Rough #1/Smooth) to moderate (Experiment 2B, Rough #2/Smooth) to high (Experiment 2C, Rough #3/Rough #4). Finally, we examined the ability of irradiated rats to learn the acquisition and reversal of a Black/White discrimination in which the discriminanda were restricted to the goal-arm walls (Experiment 3).

To assess the effect of degranulation of the dentate gyrus on activity level, we tested most of the irradiated and control rats in an open field.

General Method

Subjects

Litters of laboratory-bred Long-Evans rats were culled to six males on the day after birth and randomly assigned to either the X-irradiated or the control group. The litters were weaned at 21 days of age, housed in colony cages, and fed laboratory chow and water ad lib until the start of experimental food deprivation. The colony room was maintained on a 12:12 hr light/dark cycle and had a mean temperature of 22 °C. Prior to discrimination training, some of the rats were observed in an open field; the rest were experimentally naive. Rats were 65–95 days of age at the start of open-field testing and 90–120 days of age at the start of discrimination training.

Irradiation

A detailed description of the X-irradiation procedure used in this study has been published elsewhere (Bayer & Peters, 1977). Briefly, X-rays were delivered at a rate of 46 R/min from a General Electric Maxitron 300 KV X-ray machine through 1.5 mm of additional copper filtration. Prior to X-ray exposure the rat pups were immobilized in Lucite holders and placed under a protective lead shield. An adjustable slit in the lead shield restricted X-ray exposure to that part of the head containing the hippocampus. The irradiated rats were exposed to 200 R X-rays on Days 2 and 3, and to 150 R on Days 5, 7, 9, 11, 13, and 15. The control rats were restrained in the same manner as the irradiated rats but were not exposed to X-rays.

Apparatus

Open field. The open-field apparatus measured 54 × 54 × 28 cm and was constructed from unpainted wood on three walls and clear Lucite on the front wall. The black Lucite floor was divided into twenty-five 10-cm squares. Illumination was provided by a 40-W incandescent bulb positioned 60 cm above the center of the open field. The apparatus was located in a sound-attenuating room, and the rats were observed from behind a one-way mirror.

Discrimination training. The testing apparatus used in all discrimination experiments was a T-maze. Masonite guillotine doors divided the maze into a start box, a straight alley, and two goal arms. The walls and ceiling of the entire maze, and the floors of the start box and straight alley, were constructed of double-layered clear Lucite for the insertion of discriminanda. In all experiments, neutral-gray paper was inserted into the walls, and black paper was inserted into the floors, of the start box and straight alley. The removable floors of the goal arms consisted of either dull-black anodized aluminum or white Lucite plates. Plastic food cups were placed at the ends of the goal arms. The food cup in the correct goal arm contained food pellets, and the food cup in the incorrect goal arm was empty. Each goal arm contained a 90° bend that hid the food cup from view until the goal-arm door was passed. Holes were drilled in the end walls of the goal arms to permit

placement of food pellets outside the walls to control for food odor. The walls of the testing room were covered with black cloth to reduce extramaze visual cues.

Procedure

Open field. Each rat was placed in the center of the open field and observed for 6 min on 3 consecutive days. The number of squares entered was recorded. After each rat was tested, the open field was wiped with a damp sponge and dried with paper towels.

Discrimination training. Three weeks before the start of discrimination training, each rat was moved to an individual cage, food deprived to 80% (± 10 g) of its ad lib weight, and maintained at this level for the remainder of the experiment. Each rat was handled daily for approximately 5 min during the food deprivation period.

Pretraining was begun 4 days before the experiment. On the first 2 days, each rat was fed ten 45-mg Noyes food pellets in its home cage, in addition to its normal ration of laboratory chow. On the third day, five food pellets in a small plastic cup were placed in each goal arm, and each rat was given 10 min to explore the maze with all doors removed. On the fourth day, each rat received six forced-choice pretraining trials in the sequence RRLRLL, each run being rewarded with five food pellets.

Training was begun on the fifth day after the start of pretraining. Each testing squad consisted of three control and three irradiated rats. Three of the rats were rewarded for responding to one discriminandum; the other three, for responding to the other discriminandum. A trial consisted of placing the rat in the start box, opening the start box door after the rat oriented to it, and closing the goal-arm door after the rat passed through it. On correct trials the rat was allowed to eat the reward of five food pellets before being returned to its holding cage. On incorrect trials the rat was confined in the goal arm for 20 sec. A noncorrection procedure was used. Water was available at all times in the

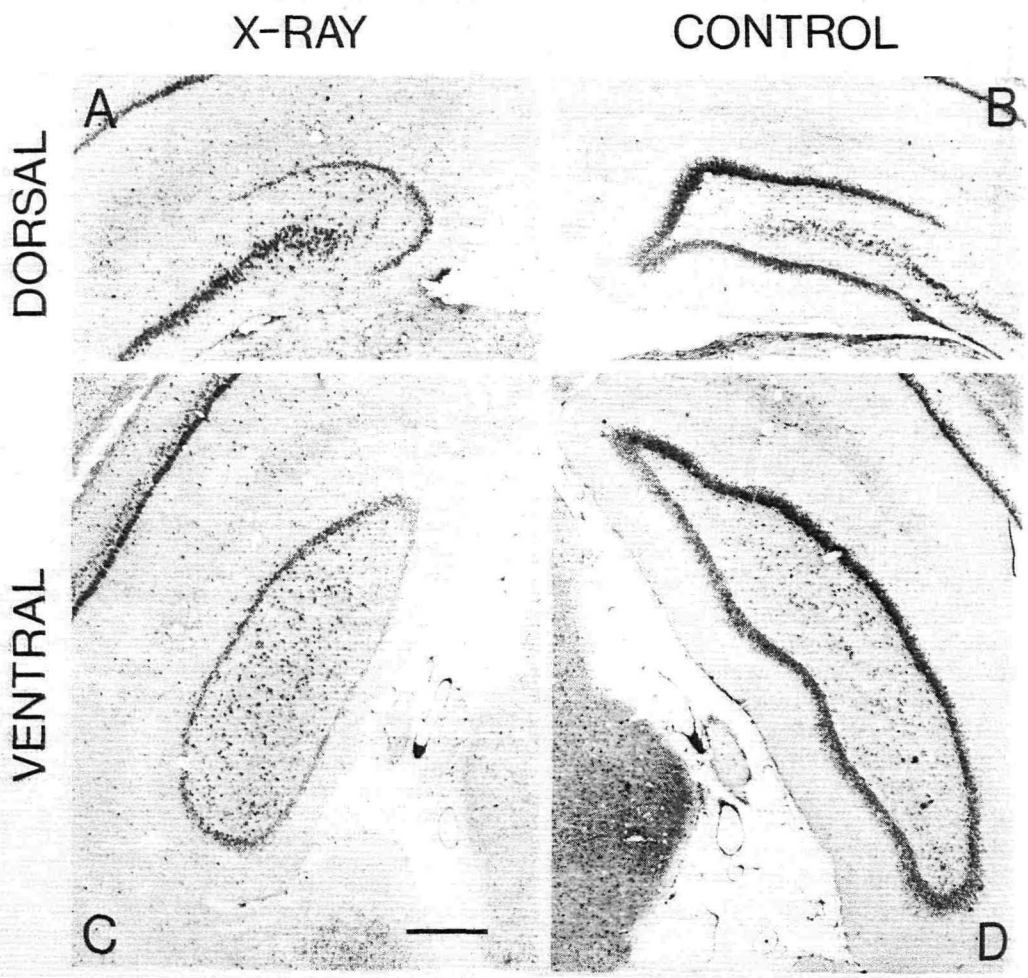

Figure 1. The dentate gyrus of control and irradiated rats at the rostral (A and B) level used for granule cell counts of the dorsal hippocampal formation, and at the caudal (C and D) level used for granule cell counts of the ventral hippocampal formation. (Hematoxylin and eosin. Scale = .5 mm.)

holding cage. Five sequential schedules were used to determine the position of the discriminanda. They had the following characteristics: (a) Each schedule determined the position of the discriminanda for one day's testing, (b) in each schedule the positive stimulus did not appear on the same side more than three times in a row, and (c) in each schedule the positive stimulus appeared an equal number of times on the left and right sides. After all rats in a squad had received one trial, the floors of the goal arms were cleaned to reduce odor cues, and the position of the discriminanda was established for the next trial. This procedure was repeated until each rat had received 20 trials in one day. The intertrial interval was 5–10 min. The testing order within a squad was changed daily. The entire maze was wiped with a damp sponge after the completion of each squad's daily testing.

Learning criterion was defined as 18 correct trials out of 20 in one day. Reversal training was begun on the day after each rat reached criterion. Measures for both acquisition and reversal training were trials to criterion and errors to criterion.

Histology

After completion of behavioral testing, all irradiated rats and a representative sample of control rats were anesthetized with an overdose of sodium pentobarbital and perfused intracardially with 10% neutral-buffered formalin. The brains were removed, postfixed in Bouin's solution for 24 hr, and then taken through several daily changes of formalin. After clearance in formalin the brains were embedded in paraffin, and 6-μm coronal sections were stained with hematoxylin and eosin. Counts of dentate granule cells in the hippocampal formation were made in anatomically matched sections to determine the extent of cell reduction due to X-irradiation. The granule cells of the dorsal hippocampal formation were counted in that section where the stratum pyramidale of dorsal and ventral Ammon's horn intersect (approximately level A2.8 of Pellegrino, Pellegrino, & Cushman, 1979). The granule cells of the ventral hippocampal formation were counted in that section where the distinct stratum of CA_3 pyramidal cells of Ammon's horn first disappears (approximately level A1.0 of Pellegrino et al., 1979).

Figure 1 illustrates the reduction in dentate granule cells produced by the X-irradiation schedule used in this study.

Experiments 1A, 1B, 1C: Brightness Discrimination

Method

Subjects. In Experiment 1A (Bright/Dark), subjects were drawn from nine control and nine irradiated litters, and the results are based on 16 control and 16 irradiated rats. Four rats assigned to this experiment, two control and two irradiated rats, were discarded for refusing to run in the maze. In Experiment 1B (Bright/Dim #1), subjects were drawn from nine control and nine irradiated litters, and the results are based on 17 control and 17 irradiated rats. Two rats assigned to this experiment were not included in the results: One control rat refused to run in the maze, and one irradiated rat was found to have a glioma in the corpus callosum. In Experiment 1C (Bright/Dim #2), subjects were drawn from nine control and nine irradiated litters, and the results are based on 15 control and 16 irradiated rats. Five rats assigned to this experiment were not included in the results: One irradiated and two control rats refused to run in the maze, one control rat died during testing, and one irradiated rat was judged to be blind.

All rats were observed in an open field before discrimination training except for two irradiated rats (Experiment 1A) and one control rat (Experiment 1C).

Procedure. The visual discriminanda were provided by light boxes attached to the underside of the table supporting the T-maze, and white translucent Lucite plates were used as the floors of the goal arms. Light was produced by two 20-W fluorescent tubes located under either side of the choice point and two 15-W fluorescent tubes located under the food cups. The light was diffused by passage through several layers of white Lucite, the light intensity was regulated by a General Electric light dimmer and by passage through several layers of white paper. Luminance levels were measured by a United Detector Technology light meter (Model 111C-11CP) calibrated at 560 nm. In Experiment 1A (Bright/Dark), the luminance of the Bright stimulus, measured at the choice point, ranged from 11.16 to 12.63 cd/m^2, the luminance of the Dark stimulus, measured with the other goal arm lit, was .07 cd/m^2. In Experiment 1B (Bright/Dim #1), the luminance of the Bright stimulus ranged from 7.90 to 9.52 cd/m^2, the luminance of the Dim #1 stimulus ranged from .10 to .12 cd/m^2. In Experiment 1C (Bright/Dim #2), the luminance of the Bright stimulus ranged from 11.16 to 12.63 cd/m^2, the luminance of the Dim #2 stimulus ranged from .78 to .93 cd/m^2. White paper was inserted into the walls of the goal arms to reflect the light entering through the floors. Both goal arms were lit with the Bright stimulus during pretraining. The overhead room lights were off during these experiments.

Results and Discussion

Anatomical. The mean (± *SE*) percentage reductions in granule cells of the dorsal dentate gyrus in irradiated rats were 85.0 ± .6, Experiment 1A; 85.7 ± .4, Experiment 1B; and 87.7 ± .6, Experiment 1C. These reductions are comparable with those found in previous studies from our laboratory (Bayer et al., 1973; Brunner, Haggbloom, & Gazzara, 1974; Haggbloom et al., 1974) and by others (Gerbrandt et al., 1978; Hirsh et al., 1978). The mean (± *SE*) percentage reductions in granule cells of the ventral dentate gyrus in irradiated rats were 75.8 ± .8, Experiment 1A; 76.9 ± .5, Experiment 1B; and 80.4 ± .8, Experiment 1C. The smaller reduction in granule cells of the

Table 1
Mean (± SE) Trials and Errors to Criterion in Visual Brightness Discriminations

Group	*n*	Acquisition		Reversal	
		Trials	Errors	Trials	Errors
Experiment 1A (Bright/Dark)					
Control	16	72.5 ± 5.4	21.6 ± 2.8	151.2 ± 8.6	76.8 ± 5.0
X-ray	16	77.5 ± 6.8	25.1 ± 3.4	160.0 ± 8.6	82.4 ± 6.5
Experiment 1B (Bright/Dim #1)					
Control	17	96.5 ± 8.1	34.1 ± 4.3	184.7 ± 12.7	93.8 ± 6.4
X-ray	17	110.6 ± 8.6	40.9 ± 5.3	227.1 ± 15.0[a]	117.4 ± 8.4[b]
Experiment 1C (Bright/Dim #2)					
Control	15	173.3 ± 11.3	65.0 ± 5.0	297.3 ± 20.1	146.7 ± 10.4
X-ray	16	223.8 ± 18.6[c]	93.6 ± 9.2[d]	377.5 ± 22.2[e]	203.4 ± 12.7[f]

[a] $F(1, 32) = 4.67$, $p = .036$
[b] $F(1, 32) = 4.99$, $p = .031$
[c] $F(1, 29) = 5.17$, $p = .029$.
[d] $F(1, 29) = 7.18$, $p = .012$.
[e] $F(1, 29) = 7.11$, $p = .012$
[f] $F(1, 29) = 11.74$, $p = .002$

ventral dentate gyrus has two possible causes. First, the granule cells of the ventral dentate gyrus begin to form earlier than those of the dorsal dentate gyrus (Bayer, 1980); thus a lesser percentage of the ventral granule cells may be affected by the X-irradiation schedule used in this study. Second, placement of the lead shielding, used to protect other radiosensitive portions of the brain (e.g., the cerebellum), may have inadvertently reduced the amount of X-ray exposure received by the caudal portion of the hippocampus.

Behavioral. Since some of the rats trained in the brightness discrimination tasks were not tested in the open field, the open-field data were collapsed across experiments. A one-way analysis of variance revealed that the irradiated rats entered significantly more squares (and thus were more active) than the control rats (Control $M \pm SE = 237.0 \pm 11.2$; X-ray $M \pm SE = 325.0 \pm 23.3$), $F(1, 92) = 11.60$, $p = .001$.

One-way analyses of variance were performed on trials and errors to criterion in the acquisition and reversal phases of the discrimination tasks. In Experiment 1A (Bright/Dark), there was no significant difference between control and irradiated rats in either trials to acquisition (Control $M = 72.5$; X-ray $M = 77.5$) or trials to reversal (Control $M = 151.2$; X-ray $M = 160.0$). In Experiment 1B (Bright/Dim #1), there was no significant difference between control and irradiated rats in trials to acquisition (Control $M = 96.5$; X-ray $M = 110.6$), but the irradiated rats required significantly more trials to reverse the task than did their respective controls (Control $M = 184.7$; X-ray $M = 227.1$). In Experiment 1C (Bright/Dim #2), the irradiated rats required significantly more trials than the control rats to learn both the acquisition (Control $M = 173.3$; X-ray $M = 223.8$) and the reversal (Control $M = 297.3$; X-ray $M = 377.5$) phases of the task. In all three experiments, the analyses of errors to criterion produced the same results as did the analyses of trials to criterion (Table 1).

Experiments 1A, 1B, and 1C were identical in terms of magnitude of granule cell reduction, testing apparatus, testing procedures, and sensory modality. The only aspect that varied in the experiments was level of task difficulty. In brightness discriminations, the irradiated rats showed no acquisition or reversal deficits when task difficulty was low, a reversal deficit when task difficulty was moderate, and both an acquisition and a reversal deficit when task difficulty was high.

Since these experiments were conducted

in the visual modality, it was of interest to determine whether this task-difficulty effect was restricted to this modality or was of a more general nature. Accordingly, three experiments were designed to test the ability of irradiated rats to learn tactile discriminations of progressively increasing task difficulty. To facilitate comparisons, we made an attempt to match the task difficulty of the acquisition of tactile discriminations with that of the brightness discriminations.

Experiments 2A, 2B, 2C: Roughness Discrimination

These experiments were designed to determine whether irradiated rats would demonstrate deficits in learning a tactile roughness discrimination as task difficulty was progressively increased from low (Experiment 2A, Rough #1/Smooth) to moderate (Experiment 2B, Rough #2/Smooth) to high (Experiment 2C, Rough #3/Rough #4).

Method

Subjects In Experiment 2A (Rough #1/Smooth), subjects were drawn from 11 control and 11 irradiated litters, and the results are based on 19 control and 18 irradiated rats. Five rats assigned to this experiment were not included in the results One control and one irradiated rat refused to run in the maze, one irradiated rat could not be tamed by handling, one irradiated rat died before it could be perfused, and one control rat had an abnormally low granule cell count In Experiment 2B (Rough #2/Smooth), subjects were drawn from nine control and nine irradiated litters, and the results are based on 17 control and 18 irradiated rats One control rat was excluded from the experiment due to a middle ear infection In Experiment 2C (Rough #3/Rough #4), subjects were drawn from nine control and nine irradiated litters, and the results are based on 16 control and 17 irradiated rats Three rats assigned to this experiment were not included in the results, one control and one irradiated rat because of illness and one control rat for refusing to run in the maze

The rats in Experiment 2A were experimentally naive at the start of training The rats in Experiment 2B (except for three irradiated and three control rats) and Experiment 2C were observed in an open field prior to discrimination training

Procedure The tactile discriminanda were dull-black anodized aluminum plates They were used as the floors of the goal arms of the T-maze, replacing the white Lucite plates used in the brightness discriminations In Experiment 2A (Rough #1/Smooth), the Rough #1 stimulus was milled to a pattern of raised squares (1.6 × 1.6 × 1.6 mm), separated by 3 2 mm, covering the entire surface; the Smooth stimulus was unmilled. In Experiment 2B (Rough #2/Smooth), the Rough #2 stimulus was milled to a pattern of parallel ridges 1.6 mm high and 4.6 mm wide, separated by 1.6 mm, aligned along the long dimension of the plate (modified from Finger & Frommer, 1968), the Smooth stimulus was unmilled In Experiment 2C (Rough #3/Rough #4), the Rough #3 stimulus was milled to a pattern of parallel ridges 1.6 mm high and 1 6 mm wide, separated by 4 6 mm, aligned along the long dimension of the plate; the ridges of the Rough #4 stimulus were 1.6 mm high and 4 0 mm wide and were separated by 4 0 mm (both modified from Finger & Frommer, 1968). The Smooth stimulus was present in both goal arms during pretraining in all experiments Neutral-gray paper was inserted into the walls of the goal arms The maze was illuminated from above by a bank of four 40-W fluorescent tubes shone through several layers of white paper and diffusers to reduce light intensity and evenly distribute the light, the luminance level was 71 4 cd/m^2

Results and Discussion

Anatomical. The mean (± *SE*) percentage reductions in granule cells of the dorsal dentate gyrus in irradiated rats were 85.8 ± .7, Experiment 2A; 86.2 ± .6, Experiment 2B; and 83.3 ± .5, Experiment 2C. The mean (± *SE*) percentage reductions in granule cells of the ventral dentate gyrus in irradiated rats were 73.8 ± 1.0, Experiment 2A; 76.0 ± .8, Experiment 2B; and 74.1 ± .9, Experiment 2C. These reductions are similar to those found in Experiments 1A, 1B, and 1C. The reductions in granule cell population of four irradiated rats in Experiment 2A had to be estimated because of tissue damage incurred during histological processing; they were judged to be comparable with those reported in Experiment 2A.

Behavioral. Since the rats trained in Experiment 2A and three irradiated and three control rats trained in Experiment 2B were not tested in the open field, the open-field data were collapsed across Experiments 2B and 2C. A one-way analysis of variance revealed that the irradiated rats entered significantly more squares than did the control rats (Control $M \pm SE = 216.2 \pm 11.8$; X-ray $M \pm SE = 289.8 \pm 23.6$), $F(1, 60) = 7.48$, $p = .008$.

One control and four irradiated rats in Experiment 2C (Rough #3/Rough #4) failed to reach acquisition criterion after 780 trials. They were each assigned an acquisition score of 780 trials. Since the rats that did not complete acquisition could not be

trained on the reversal task, the reversal data from Experiment 2C were not analyzed. However, the data from these rats were included in the analysis of acquisition in Experiment 2C.

One-way analyses of variance were performed on trials and errors to criterion on the acquisition and reversal phases of the discrimination tasks. In Experiment 2A (Rough #1/Smooth), there was no significant difference between control and irradiated rats in either trials to acquisition (Control M = 66.3; X-ray M = 72.2) or trials to reversal (Control M = 104.2; X-ray M = 96.7). Similarly, in Experiment 2B (Rough #2/Smooth), there was no significant difference between control and irradiated rats in either trials to acquisition (Control M = 95.3; X-ray M = 102.2) or trials to reversal (Control M = 148.2; X-ray M = 180.0). In Experiment 2C (Rough #3/Rough #4), the irradiated rats required significantly more trials than the control rats to learn the acquisition phase of the task (Control M = 300.0; X-ray M = 474.1); reversal could not be analyzed. In all three experiments, the analyses of errors to criterion produced the same results as did the analyses of trials to criterion (Table 2).

Six experiments investigated the acquisition of brightness (Experiments 1A, 1B, 1C) and roughness (Experiments 2A, 2B, 2C) discriminations in hippocampally X-irradiated rats. Each experiment was initially assigned to a task-difficulty category according to the mean number of trials required by the control rats to acquire the discrimination. To determine whether these empirical categories actually represent different levels of task difficulty, we performed a one-way analysis of variance on the trials-to-acquisition scores of the control rats. A significant task-difficulty effect was found, $F(5, 94) = 30.63$, $p < .001$. Further analysis revealed that the six experiments fell into three task-difficulty subgroups (Duncan's multiple-range test, $p < .05$), designated low, moderate, and high (Figure 2A). In terms of these redefined task-difficulty categories, the irradiated rats were not handicapped in acquiring discriminations of low task difficulty, but they were handicapped in acquiring discriminations of moderate and high task difficulty.

Reversal learning in the brightness and roughness discrimination tasks (except Experiment 2C, Rough #3/Rough #4) was analyzed in the same manner. A one-way analysis of variance performed on the trials-to-reversal scores of the control rats revealed a significant task-difficulty effect, $F(4, 79) = 37.70$, $p < .001$. Further analysis revealed that the five experiments fell into four task-difficulty subgroups (Duncan's multiple-range test, $p < .05$), designated low, low-moderate, moderate, and high (Figure

Table 2
Mean (± SE) Trials and Errors to Criterion in Tactile Roughness Discriminations

		Acquisition		Reversal	
Group	n	Trials	Errors	Trials	Errors
Experiment 2A (Rough #1/Smooth)					
Control	19	66.3 ± 2.7	17.5 ± 1.0	104.2 ± 3.9	47.4 ± 2.1
X-ray	18	72.2 ± 5.4	20.0 ± 2.2	96.7 ± 4.6	41.3 ± 2.6
Experiment 2B (Rough #2/Smooth)					
Control	17	95.3 ± 5.6	30.8 ± 2.2	148.2 ± 10.2	66.5 ± 5.4
X-ray	18	102.2 ± 6.8	34.4 ± 2.9	180.0 ± 12.4	77.8 ± 6.2
Experiment 2C (Rough #3/Rough #4)					
Control	16	300.0 ± 37.8	130.0 ± 20.0	429.3 ± 35.2	190.2 ± 14.2
X-ray	17	474.1 ± 54.7[a]	206.7 ± 25.3[b]	438.5 ± 36.8	192.3 ± 17.9

Note Reversal data for Experiment 2C were not analyzed. Means (± *SE*) are presented for comparison only. Control n = 15, X-ray n = 13

[a] $F(1, 31) = 6.70$, $p = .014$

[b] $F(1, 31) = 5.56$, $p = .024$

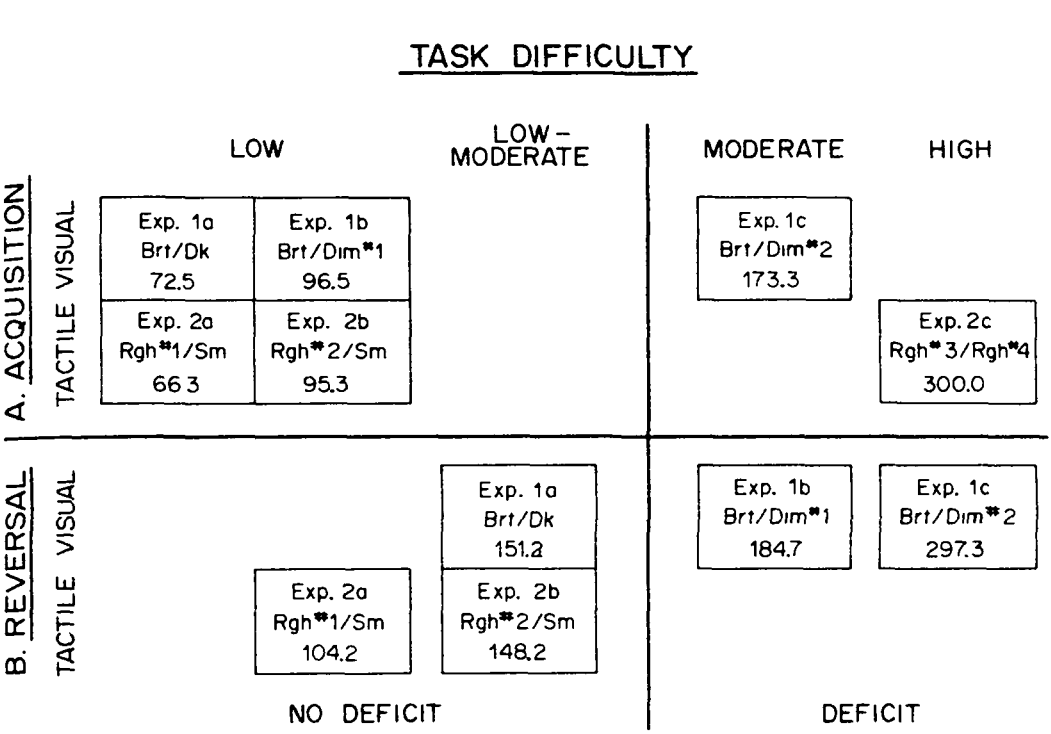

Figure 2 Task-difficulty categories for acquisition (A) and reversal (B) in both the visual brightness and the tactile roughness discriminations (Numbers represent the mean numbers of trials required by the control rats to learn the task "No deficit" and "deficit" refer to the learning ability of the irradiated rats compared with that of the control rats Brt = Bright; Dk = Dark, Rgh = Rough, Sm = Smooth.)

2B). In terms of these categories, the irradiated rats were not handicapped in reversal discriminations of low or low-moderate task difficulty, but they were handicapped in reversal discriminations of moderate and high task difficulty.

Reversal training typically required more trials than acquisition training. Except for Experiment 2A (and Experiment 2C, which was not analyzed), all the experiments fell into higher task-difficulty categories in reversal than in acquisition. (Note shift from left to right when Figures 2A and 2B are compared.) Nevertheless, learning deficits in acquisition and reversal were a function of task difficulty alone, the dividing line occurring somewhere between 151 and 173 trials. A comparison of the roughness tasks with the brightness tasks showed that the control and the irradiated rats required approximately twice as many trials to reverse the brightness discriminations than to learn them originally but that they required only approximately 50% more trials to reverse the roughness discriminations (see Tables 1 and 2). Two possibilities were raised by the faster reversal of the roughness discriminations. First, the sensory modality of the discriminanda is an additional variable affecting the performance of both the control and the irradiated rats. Second, some quality of the stimuli, such as "noticeability," is a significant variable. The rat is forced to walk on the tactile discriminanda, which increases the likelihood that the tactile stimuli will be noticed, but it is not forced to look at the visual stimuli. The higher noticeability of the tactile stimuli may also explain why the irradiated rats were not handicapped in reversing either of the roughness discriminations having low task difficulty in acquisition (Experiments 2A and 2B). In contrast, the lower noticeability of the visual stimuli may explain why the irradiated rats were handicapped in reversing one of the two brightness discriminations having low task difficulty in acquisition (Experiment 1B).

Assuming that the visual stimuli are less noticeable than the tactile stimuli, it may be

possible to reduce the noticeability of the visual stimuli even further. In the brightness discriminations, light was transmitted through the goal-arm floors and was reflected by the walls and ceiling; thus the rats were widely exposed to the visual cue. If, instead, the visual stimuli were restricted to the goal-arm walls, the noticeability of the visual cue might be reduced. Assuming that noticeability is a major variable, the irradiated rats should show a learning deficit in acquisition and reversal when the noticeability of the stimuli is further reduced. In an attempt to test this hypothesis, we chose a visual discrimination having low task difficulty in acquisition (according to the categories in Figure 2A) in which the discriminanda were restricted to the goal-arm walls (Experiment 3).

Experiment 3: Black/White Discrimination

This experiment was designed to determine whether irradiated rats show deficits in learning a visual discrimination of low acquisition task difficulty in which the Black/White stimuli used as the discriminanda are restricted to the goal-arm walls, thus reducing their noticeability.

Method

Subjects Subjects were drawn from nine control and nine irradiated litters, and the results are based on 17 control and 17 irradiated rats. Two rats assigned to this experiment, one control and one irradiated rat, were discarded for refusing to run in the maze Before training in this experiment, 11 control and 11 irradiated rats were observed in an open field, the rest were experimentally naive

Procedure The goal-arm walls of the T-maze contained neutral-gray paper during pretraining, and the black and white papers used as the visual discriminanda during training The floors of the goal arms consisted of smooth black aluminum plates The maze was illuminated as described in Experiments 2A, 2B, 2C

Results and Discussion

Anatomical. The mean (± *SE*) percentage reductions in granule cells of the dentate gyrus in irradiated rats were 85.3 ± .5, dorsal and 78.0 ± .6, ventral. These reductions are similar to those found in Experiments 1A, 1B, 1C, and 2A, 2B, 2C.

Behavioral. Data from the 11 control and 11 irradiated rats tested in the open field were analyzed by a one-way analysis of variance. The irradiated rats entered significantly more squares than did the control rats (Control $M \pm SE$ = 251.4 ± 25.7; X-ray $M \pm SE$ = 364.2 ± 27.0), $F(1, 20) = 9.16$, $p = .007$.

One-way analyses of variance were performed on trials and errors to criterion on the acquisition and reversal phases of the discrimination task. The irradiated rats required significantly more trials to learn both the acquisition (Control M = 96.5; X-ray M = 124.7) and reversal (Control M = 196.5, X-ray M = 264.7) phases of the task. The analysis of errors to criterion produced the same results as did the analysis of trials to criterion (Table 3).

The results of this experiment demonstrated that the irradiated rats were handicapped in learning the acquisition and reversal of a visual discrimination in which the discriminanda were made less noticeable by restricting them to the goal-arm walls. The reversal deficit is consistent with the results of the brightness and roughness discriminations since reversal fell into the moderate task-difficulty category (Control M = 196.5

Table 3
Mean (± SE) *Trials and Errors to Criterion in Black/White Discrimination*

		Acquisition		Reversal	
Group	*n*	Trials	Errors	Trials	Errors
Control	17	96.5 ± 7 9	32.5 ± 3 6	196 5 ± 9.4	108.7 ± 5.3
X-ray	17	124 7 ± 9.9[a]	44 6 ± 4 6[b]	264 7 ± 17.9[c]	146.2 ± 9 1[d]

[a] $F(1, 32) = 4.95$, $p = .031$.
[b] $F(1, 32) = 4\ 39$, $p = .042$.
[c] $F(1, 32) = 11.41$, $p = 002$
[d] $F(1, 32) = 12.38$, $p = .001$.

trials). However, the acquisition deficit is not consistent with these results since acquisition fell into the low task-difficulty category (Control M = 96.5 trials).

General Discussion

Rats with X-ray-induced degranulation of the hippocampal dentate gyrus were trained in brightness and roughness discriminations that varied in task difficulty in terms of the number of trials required by normal rats. The irradiated rats were not handicapped in acquiring or reversing discriminations of low or low-moderate task difficulty; however, they were handicapped in acquiring and reversing discriminations of moderate and high task difficulty. Task difficulty was manipulated by varying the difference between stimuli. High task difficulty was associated with a small difference between the visual or tactile discriminanda; low task difficulty, with a large difference between them. This difference between stimuli may have affected task difficulty in two ways. First, as the difference between stimuli decreased, it may have become more difficult for the rat to differentiate between them. Second, as the difference between stimuli decreased, the likelihood that the rat would attend to the relevant cue also may have decreased. In order for the rat to notice the relevant cue, it must be able to differentiate between stimuli. However, even though the rat is capable of differentiating between the stimuli, it may not attend to them.

If this assumption is correct, the deficits found in the irradiated rats have two possible causes. First, the rats may have had a sensory impairment and thus a greater difficulty in making the discrimination. Second, they may have had normal sensory capabilities but were impaired in attending to the relevant cue. Support for the latter interpretation is provided by a study reported by Truax and Thompson (1969). Rats with hippocampal lesions were trained on an easy size discrimination and were then transferred to four successively more difficult size discriminations. With this procedure, they were not handicapped in acquiring any of the five discriminations, a result suggesting that rats with hippocampal damage are capable of perceiving small differences between stimuli. It is possible that once the hippocampal rats attend to the relevant cue dimension, they continue to attend to it even when the discrimination is made more difficult.

The results of the Black/White discrimination in our study demonstrated that the irradiated rats were handicapped in the acquisition and reversal of a visual discrimination in which the stimuli were restricted to the goal-arm walls. The acquisition deficit is not consistent with the results of the brightness and roughness discriminations since acquisition fell into the low task-difficulty category. We consider it unlikely that the acquisition deficit is due to a sensory impairment since black and white are highly discriminable stimuli. An alternate hypothesis is that restricting the stimuli to the goal-arm walls reduced the noticeability of the stimuli and that the irradiated rats are impaired in attending to the relevant cue when stimulus noticeability is low. Support for this hypothesis is provided by a study reported by Raphelson, Isaacson, and Douglas (1965). They trained control rats and rats with hippocampal damage to run down a black straight alley for food reward. After pretraining, a novel stimulus consisting of white posterboard was added to the sides of the straight alley near the start box. On the following trial, the response latencies of the control rats increased, which indicated that they had noticed the novel white stimulus. The response latencies of the hippocampal rats did not change, which indicated that they had not noticed the novel stimulus. These results suggest that hippocampal rats are less likely to notice black and white stimuli located on the walls. Conversely, other studies have suggested that learning deficits in hippocampal rats are eliminated when highly noticeable stimuli are present (e.g., Leaton, 1969; Pellegrino & Clapp, 1971; Plunkett & Faulds, 1979; Winocur & Breckenridge, 1973).

We cannot at present adequately define "noticeability." By using a procedure similar to that described in the Raphelson et al. (1965) study, it may be possible to determine which stimuli rats with hippocampal damage are likely or not likely to notice. If these stimuli are then used in a discrimination

task, the relation between stimulus noticeability and discrimination learning in hippocampal rats can be determined. The causes of the learning handicaps of hippocampal rats with discriminanda of low noticeability must also remain uncertain. Altman, Brunner, and Bayer (1973) postulated that hippocampal rats are "inattentive" due to hyperactivity. They suggested that hyperactivity, which occurs when the hippocampal rats are placed in situations associated with arousal, interferes with the motor aspects of attention. The results of our study are consistent with this hypothesis, since the irradiated rats tested in an open field were found to be hyperactive.

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