cal comparison of pawns of different genetic loci is necessary for further analysis.

The heat-sensitive pawns are interesting material for the studies of membrane excitation because the normal process can now be switched on and off and presumably only one molecular species was altered in the disruption and restoration of excitation.

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References and Notes

1. C. Kung, *Z. Vergl. Physiol.* **71**, 142 (1971); *Genetics* **69**, 29 (1971).
2. H. S. Jennings, *Behavior of the Lower Organisms* (Columbia Univ. Press, New York, 1906).
3. H Kinosita, S. Dryl, Y. Naitoh, *J. Fac. Sci. Univ. Tokyo Sect. 4* **10**, 291 (1964); R. Eckert and Y. Naitoh, *J. Gen. Physiol.* **55**, 467 (1970).
4. R. Eckert, *Science* **176**, 473 (1972).
5. C. Kung and R. Eckert, *Proc. Nat. Acad. Sci. U.S.A.* **69**, 93 (1972); C. Kung and Y. Naitoh, *Science* **179**, 195 (1973).
6. T. M. Sonneborn, in *Methods of Cell Physiology*, D. Prescott, Ed. (Academic Press, New York, 1970), p. 241.
7. The Na-rich solution contained 20 m*M* NaCl, 0.3 m*M* $CaCl_2$, 1 m*M* tris, *p*H 7.2. Cells in sucrose medium were injected through polyethylene tubing (inside diameter, 1.14 mm) with a syringe. The columns were 2.65 cm in inside diameter and 12 or 26.5 cm high, depending on the experiments.
8. We thank Dr. James Berger for the suggestion of using density difference to stabilize the column.
9. S. Dryl, *Bull. Acad. Pol. Sci. Ser. Sci. Biol.* **6**, 429 (1958).
10. ——— and A. Grebecki, *Protoplasma* **32**, 255 (1966).
11. H. Kinosita, A. Murakami, M. Yasuda, *J. Fac. Sci. Univ. Tokyo Sect. 4* **10**, 421 (1964); Y. Naitoh and R. Eckert, *Z. Vergl. Physiol.* **61**, 453 (1968); Y. Satow and C. Kung, in preparation.
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Retrograde Amnesia and the "Reminder Effect": An Alternative Interpretation

Abstract. *Recent findings suggest that amnesic agents block the retrieval of stored information. "Reminder" treatments, such as noncontingent punishments given after the production of amnesia for avoidance learning, improve the later retention performance of an animal. The data reported suggest that noncontingent treatments provide an additional learning experience which adds to the retention performance of partially amnesic or poorly trained animals.*

Electroconvulsive shock (ECS) and a variety of other treatments produce retrograde amnesia. Animals treated shortly after training perform poorly on subsequent retention tests. These facts are not in serious dispute. Controversy centers on interpretations of the basis of the retention deficit (*1*). One interpretation is that the treatments produce retrograde amnesia by interfering with time-dependent processes underlying memory storage (*2*). An alternative interpretation of experimentally produced retrograde amnesia is that amnesic treatments produce retrograde amnesia by interfering with the retrieval of stored information. The findings of several studies appear to support the "impaired retrieval" interpretation of retrograde amnesia (*3–6*). In many of these studies, animals are (i) trained on an inhibitory (passive) avoidance task, (ii) given a treatment (such as ECS), (iii) given an initial retention test, (iv) given a noncontingent aversive treatment [usually a noncontingent footshock (NCFS)] in a different apparatus, and (v) are then given a second retention test. The noncontingent aversive stimulation appears to attenuate the retention impairment produced by the amnesic treatment. According to the impaired retrieval interpretation, these findings indicate that the noncontingent aversive treatment serves as a "reminder" and thus enables the animal to retrieve the stored but previously unavailable information.

However, another possible explanation of the "reminder effect" is that the NCFS is itself a weak training experience which an animal with some retention of inhibitory avoidance training will generalize to the task on which it is tested. This generalization hypothesis predicts that the retention performance of animals which would otherwise show poor retention will be increased by the NCFS treatment. Studies on the "reminder effect" have two results that support the hypothesis presented here: (i) the retention latencies of the retrograde amnesia control groups are generally higher than the latencies of the naive level, indicating incomplete retrograde amnesia (*3, 4*), and (ii) retention latencies of the control group that received footshocks (FS) are also increased by the NCFS (*3, 5*). These findings suggest that the NCFS provides an additional training experience which summates with retention of an avoidance task.

We tested the generalization hypothesis directly by (i) comparing the effectiveness of "reminder" treatments on animals with different degrees of retrograde amnesia and (ii) examining the effectiveness of "reminder" treatments on animals that are poorly trained initially but never given an amnesic treatment.

Sixty-day-old male Sprague-Dawley rats (250 to 300 g) (Simonsen) were used. Under Nembutal anesthesia, we implanted stainless steel skull screw electrodes bilaterally over frontal and posterior cortex (2 mm anterior to bregma, 2 mm lateral; 7 mm posterior to bregma, 2 mm lateral) in 21 of the rats. These rats were then allowed a 1-week recovery period. Other animals were unoperated.

All animals were water deprived to 80 percent of their initial weight over a period of 6 to 8 days. The rats were then given 4 days of preliminary training during which they were allowed to lick for a 30-second period from a water tube at one end of a straight alley. The alley was divided into two compartments: a white safe compartment (24 by 14 by 12 cm) separated by a sliding door from a black shock compartment (37 by 14 by 12 cm). Each day the animals were placed in the white compartment and the door was opened, allowing the animals to run to the water tube that protruded from the far end of the black compartment.

On day 5, animals were divided into seven groups (Table 1). (i) The strong-FS control group ($N = 8$) received a 2-ma, 1-second FS administered from a constant current source during the 10th second of licking on day 5. These animals, as well as animals in all other groups were given retention tests 24 hours (T_1) and 48 hours (T_2) later. (ii) The retrograde amnesia control group ($N = 6$) received the strong FS followed 0.1 second later by 6-ma (0.5-second, 60-hertz, sine wave, constant current) stimulation of posterior cortex. This amnesia treatment was chosen on the basis of prior studies that used the same task (*7*). One hour after T_1 the animals were placed in the NCFS apparatus but received no FS. (iii) The retrograde amnesia + NCFS group ($N = 15$) received a 2-ma, 1-second (strong) FS followed 0.1 second later by 6-ma stimulation of posterior cortex. One hour after T_1 each animal received

Table 1. Retention performance as tested 24 hours (T_1) and 48 hours (T_2) after training. Median latencies to lick and interquartile ranges are shown. Abbreviations used are: FS, footshock; NCFS, noncontingent footshock; RA, retrograde amnesia.

Group	N	Median latency to lick (seconds)		
		T_1	Treatment after T_1	T_2
Strong-FS controls	8	300 (300–300)	None	300 (300–300)
RA controls	6	6 (3–16)	None	8 (6–15)
RA + NCFS	15	6 (3–85)	NCFS	168 (28–300)
Complete RA + NCFS	6	5 (3–6)	NCFS	7 (6–28)
Incomplete RA + NCFS	9	30 (10–210)	NCFS	300 (300–300)
Weak-FS controls	8	32 (5–122)	None	18 (7–38)
Weak-FS + NCFS	9	7 (6–39)	NCFS	300 (84–300)
NCFS prior to weak-FS training	8	8 (7–9)	Weak-FS training	300 (300–300)
Repeated NCFS only	8	4 (3–4)	NCFS	5 (3–7)

a NCFS (2 ma, 1 second) in a novel compartment that differed considerably from the training apparatus. This was a trough-shaped compartment that had four metal plates forming the sides and floor, through which scrambled FS could be administered. (iv) The weak-FS control group ($N = 8$) received a weak training FS (2 ma, 0.4 second) during the 10th second of drinking on day 5. (v) The weak-FS + NCFS group ($N = 9$) was trained and tested as in the previous group but received a NCFS 1 hour after T_1. (vi) For the NCFS prior to weak-FS training group ($N = 8$), the NCFS preceded T_1 by 1 hour. On T_1 the animals received the weak training FS during the 10th second of licking. (vii) The repeated NCFS-only group ($N = 8$) was not trained but received only the NCFS 1 hour after the test trial on each of 4 days.

On the retention tests the animals were placed in the apparatus as on previous days. Latency to lick and the amount of time spent licking during the 30-second period after the first lick were taken as the retention measures.

Drinking behavior was stable by the end of day 4 of the preliminary training. All animals licked within 10 seconds (median, 5 seconds), and all animals licked for at least 20 seconds during the 30-second period that followed the first lick (median, 30 seconds). After training and treatment, animals for which the latency to lick was less than 10 seconds and the amount of time spent licking was more than 20 seconds were classified as having complete retrograde amnesia. Animals not meeting these rigorous criteria were classified as having incomplete retrograde amnesia.

The results are presented in Table 1. (i) Median latencies for animals in the strong-FS control group were 300 seconds on both T_1 and T_2. (ii) The retrograde amnesia control group latencies were significantly lower than those of the strong-FS control group on both test trials (*8*). (iii) Latencies for animals in the cortical stimulation group that received the NCFS were significantly lower than those of the FS control group on T_1. However on T_2, after the NCFS, the latencies no longer differed significantly ($P > 0.1$) from those of the FS control group. The increase in latencies from T_1 to T_2 was significant ($P < .05$). (iv) The cortical stimulation group that received the NCFS was divided into groups which had complete or incomplete retrograde amnesia on T_1. For both subgroups, the latencies on T_1 were significantly lower than those of the strong-FS control group. Latencies of those animals that performed at a naive level on T_1 (complete retrograde amnesia) were not significantly altered by the NCFS. However, the animals judged to have some retention of prior training (incomplete retrograde amnesia) had significantly higher T_2 latencies to lick than those of either the retrograde amnesia control group, which did not receive the NCFS, or the complete–retrograde amnesia subgroup. Thus, the results of the incomplete–retrograde amnesia subgroup provide a replication of the basic "reminder effect." Our results are consistent with the findings of Cherkin (*9*) that noncontingent punishment is effective in increasing avoidance behavior in chicks only if amnesia is incomplete.

(v) The weak-FS groups performed in a manner comparable to that of the retrograde amnesia groups. Animals that were trained with weak FS and later received NCFS displayed significantly longer latencies to lick on T_2 than did animals in the weak-FS group that did not receive the NCFS. These animals received no amnesic treatment yet their avoidance behavior was significantly improved by the NCFS. (vi) Also, the T_2 latencies of animals that received NCFS prior to training with weak FS were significantly higher than those of the animals in the weak-FS control group. (vii) Repeated NCFS did not increase latencies of the animals that received no training in the avoidance task, even after four tests, each followed by NCFS.

Our results indicate that avoidance performance of animals with poor retention of an inhibitory avoidance task, produced either by weak training or by strong training followed by an amnesic treatment, is improved by a NCFS. Presumably then, animals with weak retention of inhibitory avoidance training generalize the effects of NCFS to the avoidance task. Animals with no retention of inhibitory avoidance training, either untrained or completely amnesic animals, do not generalize from the NCFS experience. The conclusion that weak retention of the training experience is necessary in order for NCFS to influence avoidance performance is supported by the results of the retrograde amnesia + NCFS subgroups. This view is consistent with interpretations of other investigators (*4, 9*). For example, after examining carefully the conditions under which noncontingent punishment can add to a memory trace that is weakened by an amnesic treatment, Cherkin (*9*, p. 954) suggested that "noncontingent learning may result in a subthreshold engram [for behavioral expression] which co-operates with the subthreshold engram that survived the RA [retrograde amnesia] treatment, to such a cumulated level that can be recalled." Our results provide further support for this view.

We suggest that "reminder" treatments do not release a memory from an ECS-produced retrieval block. Instead, noncontingent "reminder" treatments provide a learning experience that adds to the performance of animals which would otherwise have poor retention of avoidance training. Such treatments are effective (i) whether animals have partial retrograde amnesia for the training, or are simply poorly trained and (ii) whether the treatment precedes or follows the weak training experience.

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References and Notes

1. J. L. McGaugh and M. J. Herz, *Memory Consolidation* (Albion Publishing Co., San Francisco, 1972).
2. J. L. McGaugh and R. G. Dawson, *Behav. Sci.* **16**, 45 (1971); J. L. McGaugh, *Science* **153**, 1351 (1966).
3. A. D. Springer and R. R. Miller, *Science* **177**, 628 (197[illegible] D. J. Lewis, J. R. Misanin, R. R. Miller, *Nature* **220**, 704 (1968).
4. D. Quartermain, B. S. McEwen, E. C. Azmitia, *Science* **169**, 683 (197[illegible] *J. Comp. Physiol. Psychol.* **79**, 360 (1972); R. J. Koppenaal, E. Jagoda, J. A. F. Cruce, *Psychonom. Sci.* **9**, 293 (1967).
5. E. H. Galluscio, *J. Comp. Physiol. Psychol.* **75**, 136 (1971).
6. R. R. Miller and A. D. Springer, *Physiol. Behav.* **8**, 645 (1972).
7. P. E. Gold, O. F. Bueno, J. L. McGaugh, *Physiol. Behav.*, in press.
8. All significantly different results are at the $P < .02$ level, two-tailed, unless otherwise indicated. Mann-Whitney U test or sign test, as appropriate [from S. Siegel, *Nonparametric Statistics for the Behavioral Sciences* (McGraw-Hill, New York, 1956)].
9. A. Cherkin, *Physiol. Behav.* **8**, 949 (1972).

10. Supported by postdoctoral fellowship 1 FO2 MH-51265-01 PS to P.E.G. and research grant MH-12526 to J.L.M. from NIMH and PHS and predoctoral fellowship to J.W.H. and undergraduate research participation program fellowship GY-10031 from NSF.

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Pain Perception: Modification of Threshold of Intolerance and Cortical Potentials by Cutaneous Stimulation

Abstract. *Cutaneous electrical stimulation temporarily raises the threshold of intolerance for pain elicited by electric shock. Similar stimulation suppresses the somatosensory cortical evoked potential.*

Threshold of intolerance for painful electrical stimuli was determined for three adult male subjects. The subjects in all cases faced away from the experimenter and the stimulation apparatus. In addition, their eyes were closed. Each subject was instructed to indicate when the limits of his pain tolerance had been exceeded by saying "stop." The pain stimuli were applied by silver electrodes 1 cm in diameter separated by a fixed distance of 3 cm. These electrodes were applied with pressure to the skin surface after cleansing and the application of standard electroencephalogram (EEG) electrode paste. Square-wave stimuli of constant duration were delivered at 1 sec^{-1} frequency to the skin of the dorsal surfaces of both great toes and thumbs, over the lateral malleolus of both ankles, and over the ulnar and radial borders of both wrists. Voltage was increased from 100 volts in 25-volt steps every 2 seconds to the point that the subject described it as intolerable. Subjects were given no cues other than the cutaneous shocks. Each subject was studied many times over several days so that the threshold for each area was determined a minimum of five times. The maximum tolerable voltage for a square-wave pulse of 0.2-msec duration was remarkably constant for all areas studied and among the three subjects (300 ± 25 volts).

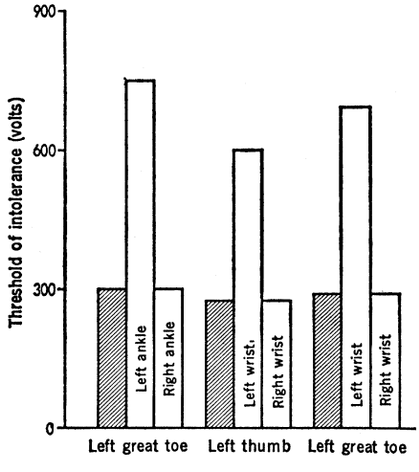

Fig. 1. Change in voltage tolerated after vibratory stimuli, subject J.B. Pain stimuli to areas labeled at bottom were given without previous vibratory stimuli (shaded bars) or immediately after vibratory stimuli to areas indicated in open bars. Four other series of determinations gave values within 25 volts of those diagrammed. Each cutaneous area was also studied five times in two other subjects, with similar results.

Vibratory electrical stimuli (50 sec^{-1}, 25 volts, 0.1-msec duration) were then delivered for 60 seconds to various portions of the extremities by electrodes constructed and applied in the same way as those used for the painful stimulation. After approximately 15 seconds, tingling sensations were reported by each subject in the extremity being stimulated. This tingling outlasted the stimulation by variable amounts of time. Pain thresholds were again determined immediately after the vibratory stimuli. In all observations, the vibratory points were at least 5 cm from the pain stimulation points, and usually the two points chosen were in cutaneous areas supplied by different peripheral nerves. A marked reproducible increase in maximum tolerance to the electrical impulse was noted after the vibration. This effect lasted for 20 minutes in our subjects, after which the original intolerance threshold values returned. Following the vibratory stimulus, the threshold for pain intolerance was often elevated in extremities other than the one receiving vibration. While this effect was primarily limited to ipsilateral limbs (Fig. 1), increased tolerance to the painful stimulus in contralateral limbs was observed several times. There was no evidence of tissue damage from the brief electrical stimulation used, despite high voltages.

Cortical evoked potentials (CEP) were obtained by averaging bipolar EEG recordings made with contralateral central region versus vertex electrodes while 100 nonpainful stimuli were delivered to the dorsal thumb (75 volts, 0.1-msec duration, 0.5 sec^{-1}) (Fig. 2). Following application of the vibratory stimulus described above, marked suppression of the CEP was observed. After the vibration, CEP's were recorded every 5 minutes until the original evoked potential configuration returned. Waves V and VI gradually reappeared, followed by a gradual increase in their voltage and then by return of the initial wave after 15 minutes. Configuration and latency of the CEP waves were identical to the initial recordings 20 minutes after the vibratory stimulus.

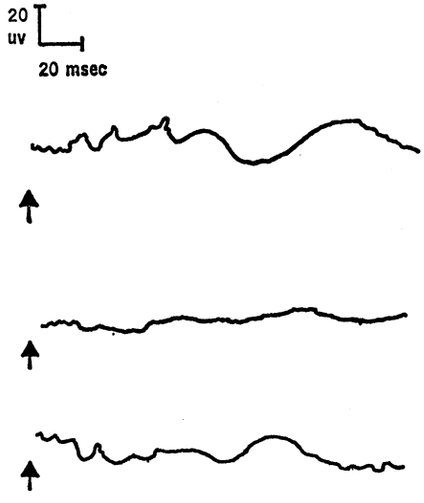

Fig. 2. Averaged cortical potentials evoked by cutaneous stimuli, subject J.B. Upper record was obtained prior to vibration (the irregular protuberance in the third major negative deflection represents artifactual reduplication of a point "dropped" by the averager). The middle record was obtained immediately after vibration; and the lower record, 15 minutes later. By 20 minutes after vibration, the CEP was indistinguishable from that in the upper trace. Arrows indicate stimulus marker. (Redrawn from photographic recordings.)