

Ipsilateral cerebellar lesions prevent learning of the classically conditioned nictitating membrane/eyelid response

JANN S. LINCOLN, DAVID A. McCORMICK and RICHARD F. THOMPSON

Stanford University, Department of Psychology, Stanford, CA 94305, (U.S.A.)

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Unilateral lesions of the cerebellum in the rabbit prior to any training completely prevented learning of the classically conditioned eyelid and nictitating membrane responses of the eye ipsilateral to the lesion. In marked contrast, subsequent training of the eye contralateral to the lesion yields normal learning.

A major goal in analysis of the neuronal substrates of learning and memory is to identify brain structures and systems that are essential for learned responses. We have recently found that the cerebellum is essential for retention and reacquisition of the classically conditioned eyelid and nictitating membrane (NM) responses^{10,12,14}. Thus, lateral lesions of the cerebellar hemisphere ipsilateral to the trained eye completely and permanently abolish the conditioned response and prevent relearning in well-trained animals but have no effect at all on the amplitude of the unconditioned reflex response. This lesion effect is selective in that the conditioned response is learned rapidly by the eye contralateral to the lesion. In the present study, unilateral cerebellar lesions were made prior to any training to evaluate two questions. (1) Will the eye ipsilateral to the lesion show any signs of learning if the lesion is made prior to training? (2) How does the rate of subsequent learning by the eye contralateral to the lesion compare if initial training to the ipsilateral eye is given before versus after the unilateral cerebellar lesion?

Lesions and training conditions are identical to those in a previous study¹². In brief, standard procedures for classical conditioning of the rabbit eyelid and NM responses were used^{12,13,14}: acoustic conditioned stimulus (CS) 36 dB spectral level white noise on for 350 ms, coterminating with a 100 ms

corneal airpuff unconditioned stimulus (UCS), intertrial interval approximately 60 s. Fifteen blocks of training were given per day, each block consisting of a restricted random sequence of 6 paired CS and UCS training trials and two CS-alone test trials, for a total of 120 trials per day. Extension of the NM was measured with a micropotentiometer and eyelid closure was also monitored; they behave essentially identically¹³ and all effects reported here occur equivalently for both responses.

In the present study the cerebellar lesions were made 7 days prior to any training. Animals were then given 4 full days of training to the eye ipsilateral to the lesion (left eye), subsequently given training to the right eye and finally given one additional day of training to the left eye. A total of 6 animals were operated and all 6 were run to completion in the study. Since all 6 animals learned to criterion (8 CRs in 9 successive trials) with the right eye, each animal served as its own control.

The mean amplitudes of conditioned responses (CR) and unconditioned responses (UCR) for the 6 animals are plotted over the course of training in Fig. 1. No conditioned responses at all are given over the 4 days of training to the left eye, the eye ipsilateral to the cerebellar lesion. When trained on the right eye, all animals learned to criterion. When shifted back to training of the left eye, they again showed no sign of conditioned responding.

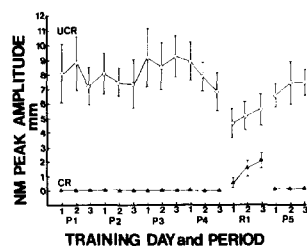


Fig. 1. Effects of ablation of left lateral cerebellum on learning of the nictitating membrane (and eyelid) responses (6 animals). Solid triangles, amplitude of conditioned response (CR); open diamonds, amplitude of unconditioned response (UCR). All training was to left eye (ipsilateral to lesion) except where labeled R1. The cerebellar lesion prevented conditioning of the ipsilateral eye but had no effect on the UCR. P1-P4 indicate the 4 days of post-lesion training to the left eye. The right eye was then trained and learned at a rate comparable to that of initial learning of non-lesioned animals. The left eye was again trained (P5) and showed no learning. Numbers on abscissa indicate 40-trial blocks.

The extent of the lesions is shown in Fig. 2. As in the previous study, the smallest lesion removed essentially all of the paramedian and ansiform lobes with damage to the pyramis, median lobe, anterior lobe and the dentate and interpositus nuclei. In no case did the lesion extend beyond the cerebellum.

As is clear from Fig. 1, prior unilateral removal of the lateral cerebellum completely prevents learning of the classically conditioned eyelid and NM responses of the ipsilateral eye. Our previous study showed complete abolition of, and an inability to reacquire, the same responses when they were learned prior to lesion. Consequently, the ipsilateral cerebellum is essential for both learning and memory of the conditioned eyelid and NM responses.

In the previous study¹⁴, where animals were

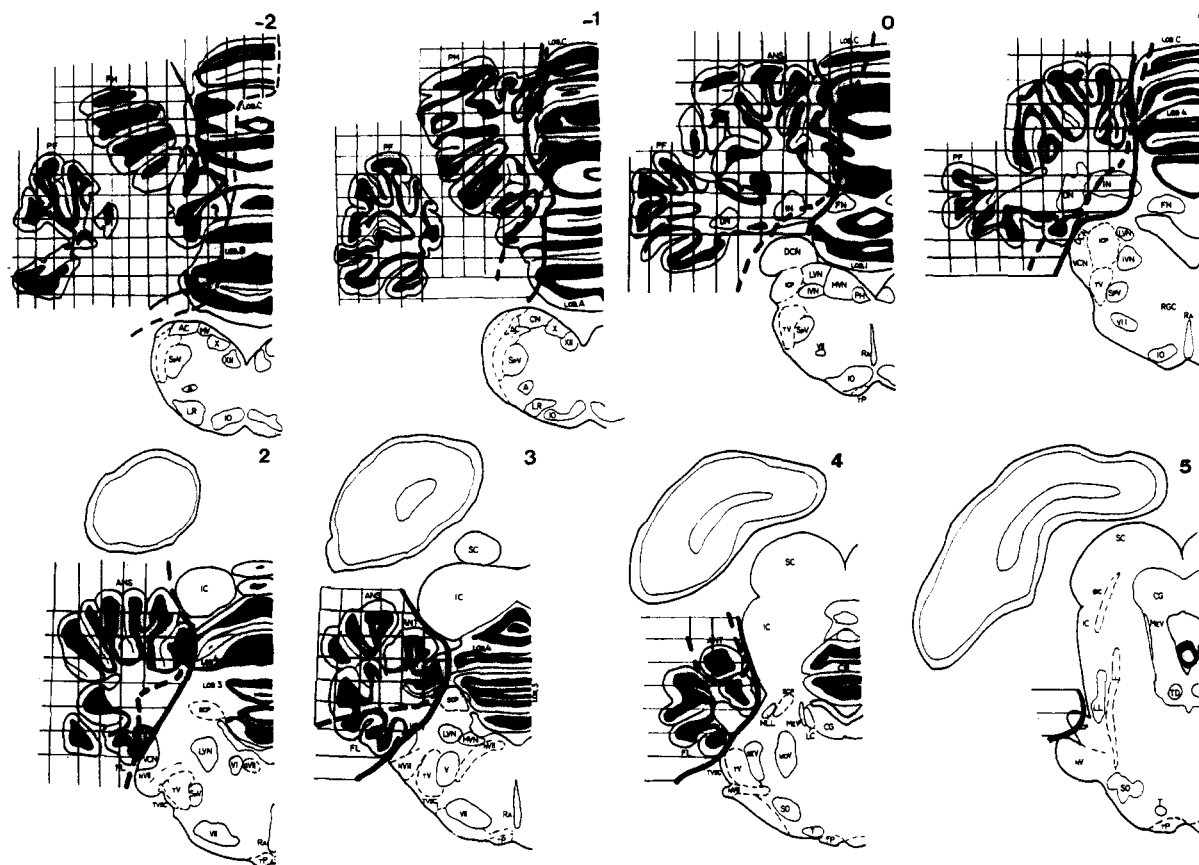


Fig. 2. Reconstructions of the smallest and largest aspirations. All tissue encompassed by the dashed border and vertical lines was removed in the animal with the smallest aspiration. All tissue encompassed by the solid border and the horizontal lines was removed in the animal with the largest aspiration. The number above each section represents mm anterior to lambda with top of the skull at lambda 1.5 mm lower than that at bregma. The abbreviations relating to areas within or near the lesions are: ANS, ansiform lobes; ANT, anterior lobe; DCN, dorsal cochlear nucleus; DN, dentate nucleus; FL, flocculus; FN, fastigial nucleus; IC, inferior colliculus; ICP, inferior cerebellar peduncle; IN, interpositus nucleus; MCP, middle cerebellar peduncle; PF, paraflocculus; PM, paramedian lobe; SCP, superior cerebellar peduncle; VCN, ventral cochlear nucleus.

trained before the cerebellar lesion, initial learning to criterion required a mean (\pm standard deviation) of 93.7 ± 44.0 trials. The animals were then subjected to the lesion, allowed to recover, given 4 days of training to the same (left-ipsilateral) eye and showed no conditioned responses. They were then trained on the right (opposite) eye and learned to criterion in a mean of 17.0 ± 14.8 trials, a substantial and statistically significant saving over original pre-operative learning of the left eye ($t = 3.3$, $df = 8$, $P < 0.01$). In the present study, with no training prior to lesion, animals subsequently learned to criterion with the right eye in a mean of 103.2 ± 76.3 trials (see Fig. 1). This number of trials does not differ significantly from the number of trials required in original learning of the left eye prior to lesion in the earlier study ($t = 0.37$, $df = 10$ n.s.). Consequently, initial training to one eye prior to ipsilateral cerebellar lesion results in significant savings in subsequent learning by the opposite eye but initial training after lesion does not. It is as though initial training to one eye in the normal animals establishes some degree of plasticity in the contralateral cerebellum.

The amplitudes of UCRs in the eye ipsilateral to the prior cerebellar lesion in the present study (mean of 8.2 ± 3.6 mm) do not differ from the amplitudes of the UCRs in the earlier study (mean of 8.5 ± 3.3 mm; $t = 0.15$, $df = 10$ n.s.). In the earlier study there was no significant difference in UCR amplitudes between the left and right eyes. However, in the present study the right eye UCRs are somewhat lower than the left eye UCRs (see Fig. 1), a decrease that just reaches statistical significance ($F = 1.89$, $df = 14,70$, $P < 0.05$). It is not clear whether this decrease is due to the sequence of training and lesion in the present study or simply to such factors as exact location of airpuff nozzle when moved to the right eye. In any event, learning in the right eye developed as though the animals were normal and new to the situation.

The present study and our earlier studies demonstrate that the lateral cerebellum ipsilateral to the trained eye is essential for original learning, retention (memory), and relearning of the conditioned eyelid and NM responses. We have found the same effect on retention and reacquisition with electrolytic lesions of the ipsilateral dentate-interpositus nuclei, and of the ipsilateral superior cerebellar peduncle in the pontine brainstem and midbrain^{10,12}. One other laboratory has independently found that ipsilateral lesions in the pontine brainstem have similar behavioral effects³. These lesion reconstructions have not yet been published but the authors kindly made them available to us. It appears that all their effective lesions included damage to the superior cerebellar peduncle.

In summary, the ipsilateral cerebellum and its major efferent pathway are essential for learning and memory of the eyelid and NM responses. Although alternative interpretations are possible, we would like to suggest that the primary memory trace for this response may develop within the cerebellum, consistent with several theories of cerebellar function^{1,5,7,11}. In support of this view, it has been reported that lesions of the cerebellar flocculus prevent plasticity of the vestibulo-ocular reflex⁸. Furthermore, cooling of the cerebellar dentate nucleus in monkeys was found to reverse the performance of a learned response to pretraining levels². In addition, earlier literature indicated that ablation of the entire cerebellum impairs performance of a variety of conditioned responses⁹. Finally, neuronal recordings from the Purkinje cells of the cerebellar cortex have implicated these cells in the plasticity of various responses^{4,6}.

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