WOUND HEALING IN THE NEURAL TUBE OF THE EARLY CHICK EMBRYO

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In a previous study by Birge and Hillemann (1953), an investigation was made of the regulatory potentiality (regenerative capacity) of the metencephalon of the early chick neural tube. In a subsequent study by Birge (1959), consideration was given to the regulatory potentiality of the mesencephalon of the early chick brain and the associated mechanisms involved in wound repair. In each of these studies regulatory potentiality was gauged as the response to induced unilateral deficiencies in which one alar (sensory) plate of the concerned brain division was ablated by micro-electrosurgery. While significant regenerative potentiality was demonstrated in the alar (sensory) plate system of the early mesencephalon of the chick, such capacity was found to be largely lacking in the metencephalon during early neural tube stages. Unilateral alar plate ablation in the mesencephalon was consistently followed by a pattern of regeneration in which cells migrated from the intact (hyperplastic) alar plate across the midline into the wound area, thus reconstituting the deficiency. Similar operations did not elicit the same response in the chick metencephalon. Though the wound was normally closed over by a simple choroidal-like epithelium which regenerated from the ependymal lining of the intact brain wall, there was no actual restitution of mantle (gray matter) or marginal (white matter) layer elements in the defective site (Fig. 1). However, on the 10th day of incubation, a compensatory hyperplasia consistently developed in the portions of the intact metencephalic alar plate destined to give rise to the anterior and middle cerebellar lobes. More than likely this represented a secondary regulatory response occurring as a result of the influx of fiber pathways into the developing cerebellum and, therefore, it was not considered to be a *direct* response to the imposed deficiency which was instituted at 28 to 38 hours of development.

In comparing the results of these two studies, attention is directed to one fundamental difference in the nature of the lesions produced in the metencephalic and mesencephalic brain divisions. In the experiments dealing with the mesencephalon, all coagulated tissue was removed from the wound by microsurgery. Concerning the metencephalic lesions on the other hand, much of the coagulated tissue was left intact in the wound area. Though such coagulated tissue may not persist in the lesion area for more than a few hours (Birge, 1959), the question must be entertained as to whether it might block the initial migration of new cells into the wound and thus inhibit the regenerative process. The study presented herein is intended to resolve this question.

Materials and Methods

A total of 10 chick embryos (White Leghorn strain) were utilized in this study. In each embryo the right alar plate was ablated in the mesencephalic and metencephalic brain divisions at 36 to 38 hours of development. The embryos were subsequently returned to incubation and later sacrificed and fixed in Bouin's solution at 4 to 5 days of development.

Alar plate ablation was carried out by micro-electrosurgery according to the method originally perfected by Hillemann (1943), and subsequently modified by Birge (1959). In each instance the coagulated tissue was carefully removed from the lesion area by conventional microsurgery.

Incubation was maintained at 100°F, with a relative humidity of 62 to 65%. Resultant alar plate regulation was gauged by the method previously described by Birge (1959).

Observations and Conclusions

In each of the 10 embryos examined, significant alar plate regeneration had occurred in the mesencephalic brain area by the time of sacrifice. The pattern of regulative development found was essentially identical to that previously described by Birge (1959). The lesion area had been entirely closed off by the extension of cells across the midline from the intact alar plate in 3 of the 5-day embryos. In the remaining 7 embryos, only small lesion openings persisted (Fig. 2). The frequency of mitotic figures in the regulating alar plates ranged 30 to 38% above the normal value previously established for this development period by Birge (1959).

Examination of the metencephalic alar plate system revealed no detectable regulative development in 4 of the embryos. In each case, head mesenchyme had condensed up to the outer margin of the open lesion. In 5 of the remaining embryos, the lesion opening had been closed over by 7 to 12 per cent. The cells responsible for this slight degree of wound repair had migrated into the lesion site from the intact metencephalic alar plate. In the single remaining embryo, the wound area had been closed by approximately 40% (Fig. 3). In this case, however, a small portion of the right alar plate had been left intact at the time of operation. Therefore, the degree of wound closure due to regulative development cannot be accurately fixed in this case. However, even if this should be entirely attributed to regeneration, it would not approach the degree of regulative development noted in the mesencephalic alar plate system.

On the basis of the foregoing observations it seems apparent that regenerative potentiality is distinctly limited in the metencephalic alar plate system of the chick during the developmental period considered in this study (36 hours to 5 days), at least as judged by the response given subsequent to unilateral alar plate ablation as effected by micro-electrocoagulation. On the other hand, the mesencephalic alar plates clearly possess a relatively high degree of regulatory potentiality. These conclusions are in agreement with the earlier findings of Birge and Hillemann (1953) and Birge (1959).

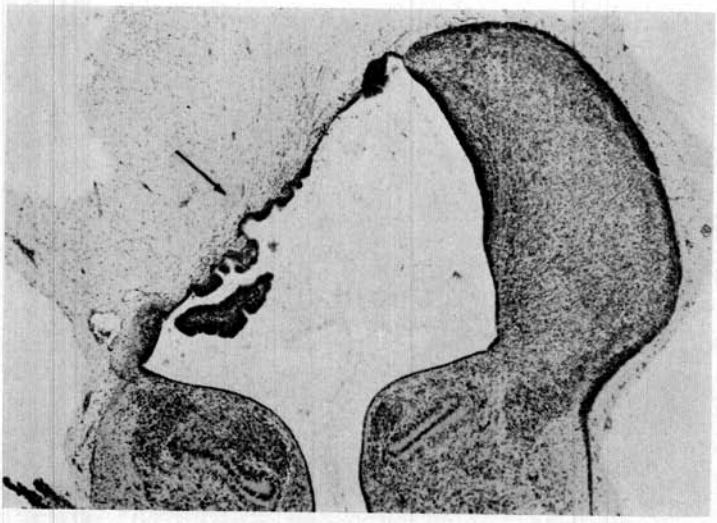

Fig. 1.—Section through metencephalon of a 10-day chick embryo. Right alar plate was ablated at 36 hours of development; left alar plate was left intact. Coagulated tissue was not removed from the lesion site (arrow). A regenerated epithelium, which remains continuous with the intact ependyma, has closed the lesion opening. However, no mantle or marginal layer elements have migrated into the defective area (from Birge and Hillemann, 1953). X32.

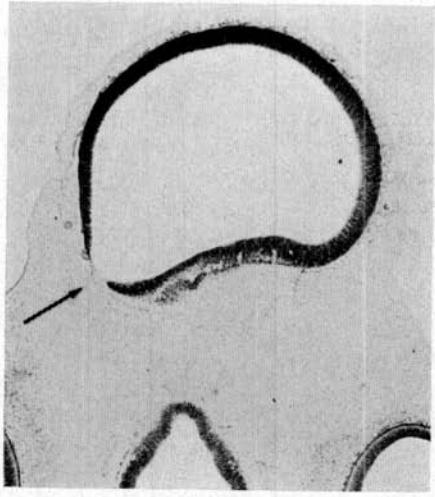

Fig. 2.—Section through mesencephalon of 4-day embryo showing effective wound closure. Arrow demarks small remnant of lesion opening. X32.

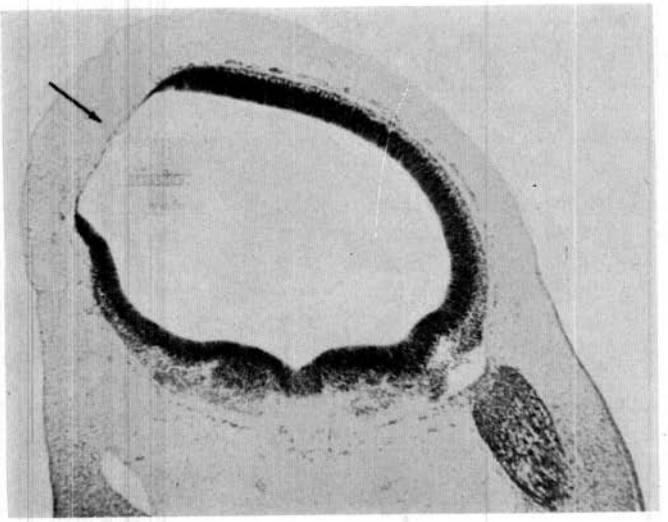

Fig. 3.—Section through metencephalon of 4-day embryo. Lesion opening has been closed by approximately 40%. Wound was cleared of coagulated tissue. X42.

It may also be concluded that the regulatory response to such unilateral metencephalic deficiencies *may* in some cases be impeded to a limited extent when a significant amount of coagulated tissue is left intact in the lesion site. Presumably this partially blocks the initial migration of new cells into the lesion area. This should be regarded as a temporary or transient effect, as the coagulated tissue is soon lost from the lesion site (Birge, 1959). The inhibitory effect produced by such remnants of coagulated tissue is probably most pronounced in systems with low regulatory potentiality. It should be noted, however, that removal of coagulated debris from metencephalic alar plate lesions does not necessarily insure a detectable degree of wound closure, at least during the developmental span considered.

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Summary

In the study presented herein, unilateral alar plate ablations were effected in the mesencephalic and metencephalic brain areas of chick embryos at 36 to 38 hours of development by micro-electrosurgery. The embryos were later sacrificed at 4 to 5 days of development and the mesencephalic and metencephalic brain areas were analyzed to determine the nature and extent of alar plate regulation.

Literature Cited

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