IN THE LABORATORY

Intravenous Methylene Blue for Studying Fiber Degeneration in the Central Nervous System

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A technique based upon intravenous perfusion of methylene blue into an anaesthetized animal (*1*, *2*) permits a much wider application of the unique property of this dye in selectively staining nervous tissue *intra vitam*, and thus makes possible studies of both the general pattern and the cytological details of neural structure throughout the nervous system. It has been noted (*2*) that the value of this method is enhanced by its demonstration of degenerating fibers, both myelinated and nonmyelinated, in the peripheral nervous system, a feature of intravital methylene blue previously described in detail by Weddell and Glees (*3*), who used the technique of local injection of the dye.

The intravenous method has now been found to give a remarkably clear demonstration of degenerating fibers in

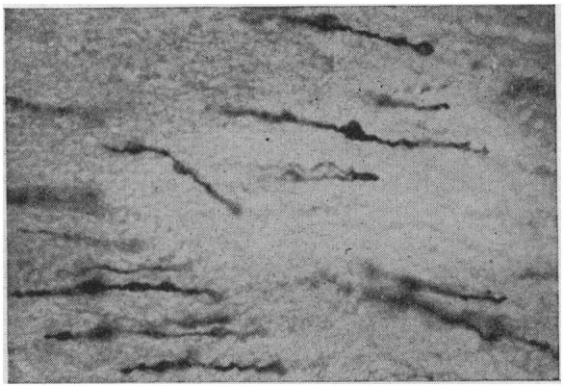

FIG. 1. Degenerating fibers in optic tract (×270).

the central nervous system. In rabbits perfused with methylene blue 5 days after section of one optic nerve, degenerating fibers in the optic pathways stand out distinctly as compared with normal fibers by reason of their more intense and metachromic staining. The contrast, even in macroscopic appearance, between the normal and degenerating optic nerves, is striking. In paraffin sections the degenerating fibers are rendered conspicuous both by their intense and characteristic reddish-purple shade of staining and by the morphological changes indicative of early degeneration, such as fibrillation and vesiculation of the axis cylinder, which they clearly exhibit (Fig. 1).

It is evident from these results that the method has a particular application to the problem of mapping out the fiber degeneration resulting from localized lesions in the central nervous system, and that it is therefore a technique likely to prove of great value in the field of experimental neurology.

The technical details of the method (*2*) have been modified to provide for more critical handling of experimental material from the central nervous system. The use of a more concentrated solution of dye has obviated the necessity of injecting a large volume of fluid which may result in cerebral edema. In addition, more satisfactory fixation has been obtained by taking the tissues through formol-saline after the ammonium molybdate stage. Neither of these modifications interferes with successful staining. Further studies are now being made on a series of rabbits in which section of an optic nerve has been selected as a control lesion, and details of these studies will be published elsewhere.

References

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A New Series of Reagents for the Colorimetric Determination of Steroids[1]

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In an investigation of the factors involved in the antimony trichloride reaction for certain ketosteroids described by Pincus (*1*), a new series of colorimetric reactions involving the use of antimony trichloride, bismuth trichloride, and aluminum chloride has been discovered. Using the Friedel-Crafts ketone synthesis as an analogy for combining catalysts and acyl chlorides, as well as other reactants, a remarkably specific and waried series of reagents has been prepared. Although only a few of the reagents have been developed far enough to give quantitative color responses following the Beer-Lambert law, it seems desirable to report the results obtained thus far.

[1] The steroids used in this study were generously provided by Ciba Pharmaceutical Products, Inc., through the courtesy of E. Oppenheimer.