Trypan Blue and Cell Migration in the Adrenal Cortex of Rats

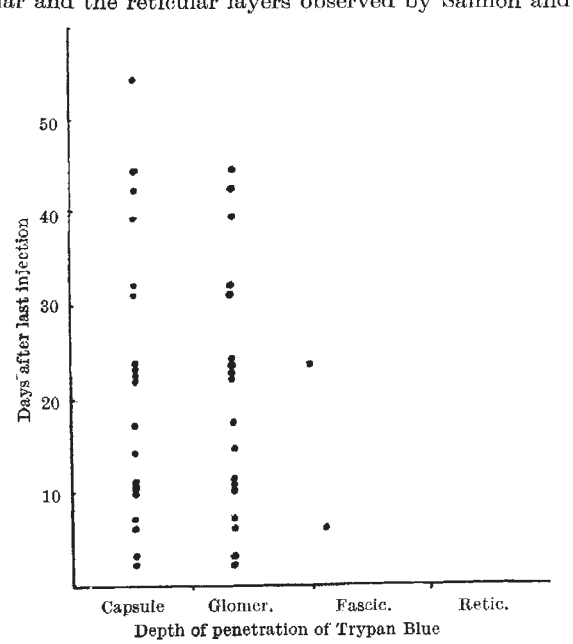
THE hypothesis has recently been put forward that there is an inward cell migration in the adrenal cortex. According to Zwemer *et al.*[1,2], in several species capsular cells multiply and move inwards, becoming in turn cells of the glomerular, fascicular and reticular layers. In guinea pigs Blumenthal[3] found that there is a zone of mitotic activity comprising the inner glomerular and outer fascicular layers, and Hoerr[4] found after chloroform narcosis increased mitotic activity in the outer fascicular layer.

Recently, Salmon and Zwemer[5] claimed that in rats Trypan Blue injected subcutaneously was first taken up by fibroblast-like cells in the capsule, and that afterwards dry droplets were to be found at varying intervals of time in the glomerular, fascicular and reticular zones successively, while the outer layers ultimately became free from the dye. On the basis of these findings they concluded that there was a progressive inward cell migration.

In the hope that this method might provide a reliable technique for the standardization of substances acting upon the cortex, we carried out similar experiments on seven batches, each of six male rats, some of the batches being composed of immature animals. The Trypan Blue was injected in doses of 1–3 c.c. of a 1 per cent solution in saline. The rats were killed 2–54 days after the last injection. Both glands were removed, and stained and unstained sections were examined.

In all sections examined, Trypan Blue droplets were present in the capsule, independent of the lapse of time after injection and the dosage given. Trypan Blue was also found frequently in the glomerular zone and very occasionally in the outer fascicular layer; but the depth of penetration of the dye bore no relation whatever, either to the time interval after the last injection, or to the dose given. The results obtained are plotted on the accompanying chart.

This method thus yields no evidence for centripetal cell migration in the adrenal cortex. The Trypan Blue uptake of the cells of the deeper fascicular and the reticular layers observed by Salmon and Zwemer may perhaps be due to the massive doses used (21–30 c.c. of a 1 per cent solution), which resulted in a generalized absorption of the dye by the whole cortex. It seems possible that the uptake of Trypan Blue and its subsequent appearance in droplet form in the fibroblast-like and glomerular cells is due to the macrophagic activity of the former and to some metabolic peculiarity of the latter, not normally possessed by the cells of the fascicular and reticular zones.



We would like to thank Prof. Samson Wright and Dr. J. H. Woodger for their interest and criticism.

ITALO CALMA.
CHARLES L. FOSTER.

Departments of Physiology and Biology,
Middlesex Hospital Medical School,
London, W.1. Oct. 5.

[1] Zwemer, R. L., Wotton, R. M., and Norkus, M. G., *Anat. Rec.*, **72**, 249 (1938).
[2] Wotton, R. M., and Zwemer, R. L., *Anat. Rec.*, **86**, 409 (1943).
[3] Blumenthal, H. T., *Endocrinology*, **27**, 477 (1940).
[4] Hoerr, N., *Amer. J. Anat.*, **48**, 139 (1931).
[5] Salmon, T. N., and Zwemer, R. L., *Anat. Rec.*, **80**, 421 (1941).

Amount of Heterochromatin as a Specific Character

ONE general fact which emerges from recent work in animal cytology is that the extent of the heterochromatic ('inert') segments of the chromosomes often varies very greatly from one species to another, even within the same genus. Thus, among the grasshoppers, *Stauroderus scalaris* possesses very large proximal heterochromatic regions in all the chromosomes, while in other species of the genus these regions are very much shorter and less conspicuous[1]. The same difference has long been known to exist between species of Drosophila, where *D. virilis* has the proximal half of each autosome heterochromatic, while in some other species such as *busckii* there is far less heterochromatin. These species belong to different subgenera; but it has recently been shown by Wharton[2] that *D. melanopalpa* has considerably more heterochromatin than the very closely related *repleta*. In the Pentatomids, Schrader[3] has shown that *Edessa irrorata* has prominent heterochromatic blocks at the ends of most or all of its chromosomes, which are not obvious in several other members of the same genus. An identical situation occurs in the Corixidæ, where Slack[4] has shown that *Corixa punctata* is a species with unusually extensive heterochromatin.

Since this phenomenon is clearly one of very widespread occurrence in animals (and possibly in plants as well) it seems desirable to have words to designate species with more or less than the average amount of heterochromatin for the group to which they belong. I should like to suggest that species like *Stauroderus scalaris*, *Edessa irrorata*, *Drosophila virilis* and *D. melanopalpa* be called 'megaheterochromatic', the species with little heterochromatin being referred to as 'microheterochromatic'. It is unfortunately not always easy to distinguish between species in which the heterochromatic regions are long and those in which they are of average length but unusually strongly heteropycnotic at certain stages of the nuclear cycle. It is believed, however, that all the examples cited do actually represent cases where the heterochromatic segments are unusually long, although they may also be more strongly heteropycnotic than usual.

Heterochromatin, in all those forms where its