Dr. W. Pitney, of the Royal Perth Hospital, Perth, who performed the alkali denaturation tests and examined the peripheral blood and marrow smears, and Prof. J. B. Chatterjea of the School of Tropical Medicine, Calcutta, who confirmed the presence of hæmoglobin F in two of the cases, being the first to demonstrate it in one case. Mr. Ian Parsons, of the Peter MacCallum Institute, Melbourne, carried out the estimations of glucose-6-phosphate dehydrogenase.

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## HISTOLOGY

## Antidiuretic Hormone and Renal **Collecting Tubules**

GINETZINSKY<sup>1</sup> has reported that, in the kidneys of rats treated with posterior pituitary extract, the epithelium of the collecting tubules becomes flattened and its basement membrane less distinct. We have attempted to repeat Ginetzinsky's observations.

Fifty-two young male white rats, 150-200 gm. in weight, were fasted overnight with free access to water and then treated in one of four ways:

(1) Eighteen rats were given water by stomach tube (5 per cent of body-weight followed by 3 per cent after 1 hr., or 7 per cent of body-weight with replacement of lost water every  $\frac{1}{2}$  hr.) and were killed during a diuresis of 2.0-3.5 ml. (11 rats) or 1.0-2.0 ml. (7 rats) per ¼ hr.

(2) Sixteen rats were treated similarly, but the water diuresis was inhibited by a subcutaneous injection of 'Pitressin' (Parke Davis), 25-30 m.units per 100 gm. body-weight. These rats were killed when urine flow, which was rapidly inhibited, had fallen to 0.3 ml./15 min. or less; in 4 rats the flow had ceased completely for a half-hour or more.

(3) Nine rats were given the hormone but no water, and (4) nine were given neither water nor hormone but killed after being handled similarly to the other groups. In groups (3) and (4) the output of urine did not exceed 0.3 ml. in any single 15-min. period.

The following methods were used for staining sections of kidneys of these rats: (a) Bharadwaj and Love's method for mitochondria<sup>2</sup>; (b) LaCour, Chayen and Gahan's method for lipids<sup>3</sup>; fixation in Zenker's fluid, followed by (c) hæmatoxylin and eosin, (d) Heidenhain's azan, (e) toluidine blue or (f) the periodic acid-Schiff reaction (PAS); (g) fixation in formol saline, followed by Heidenhain's iron hæma-In each group, Bharadwaj and Love's toxylin. method, the PAS reaction and either azan or hæmatoxylin and eosin were each used for at least half the rats and the other methods for two animals of the group.

The staining of mitochondria and lipids showed differences between rats and between tubules in individual kidneys. These variations are reported elsewhere<sup>9</sup>.

From the examination of sections stained by the PAS reaction, by toluidine blue, by azan, and by Heidenhain's iron hæmatoxylin, the following generalizations can be made:

(1) In all rats the collecting ducts were lined by cuboidal epithelium, which became low columnar in form near the tip of the pyramid. The height of the epithelium of these ducts did not differ perceptibly from one rat to another or between groups of rats, if similar distances from the tips of the pyramids were taken in choosing fields for comparison.

(2) The collecting ducts converge early in their course, so that sections parallel with the plane of the pelvis and ureter must be central if the ducts are to be seen in the pelvic half of the pyramid. Besides these ducts, the only tubular organs in the innermost medulla are blood vessels and thin segments of Henle's loops. Thus it is possible to obtain longitudinal sections of the pelvic half of the medulla which show only tubules lined by low cells, such as those in Ginetzinsky's Fig. 4, but it does not follow that some of these must be collecting ducts, which in this region are to be found only in the centre of the pyramid.

(3) The basement membranes varied in appearance between sections of the same block when toluidine blue was used, but, after the PAS reaction, these membranes were clearly seen, constant in appearance for each part of the nephron, and did not vary appreciably between the experimental groups.

(4) The PAS reaction showed the collecting ducts to differ between individual kidneys in the proportion of intercellular boundaries which showed stained cement. In the ducts of some kidneys PAS-positive material was present in the lumina in masses up to  $50\mu$  in These two features were not obviously diameter. correlated with each other, nor did either provide a point of distinction between experimental groups.

These findings do not support the suggestion that hyaluronidase, which Ginetzinsky<sup>1,6</sup> reported to be secreted in increased concentration in the urine under the influence of antidiuretic hormone, acts by altering the visible form of basement membranes and intercellular cement in the collecting ducts. Since such variability in tubular mucopolysaccharides as was found in our experiments could not be correlated with the treatment of the rats, our results may be compared with those of Berlyne<sup>4,5</sup>, who, estimating hyaluronidase in the presence of a constant concentration of electrolytes', reported that the rate of excretion of hyaluronidase in the urine was unrelated to the rate of urine flow. We did not obtain any evidence for the apocrine activity which, according to Ginetzinsky<sup>1</sup>, is indicated by a loss in height of the epithelium of the collecting ducts.

The findings reported here agree with those of Heller and Lojda<sup>8</sup>, who examined renal histology under the influence of water and antidiuretic hormone but did not record rates of urine flow.

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