crovilli and an occasional long cilia. Nuclei stain evenly but lightly (Fig. 4).

The ultrastructure of these cells is characterized by very dense and richly-developed basal cell infoldings paralleling the well-developed, elongated mitochondria which occur in all the cell zones, basal, paranuclear and supranuclear. The endoplasmic reticulum is smooth and often whorling and dense supranuclearly. The cytoplasm exhibits large parallel vesicular spaces. An occasional Golgi apparatus lies near the nucleus and there is an infrequent lysosome.

Segment VI. (Segment VI is very variable in length, 0.05 to 0.15 mm and has an average diameter of 0.03 mm.) The terminal part of segment V courses across the whole width of the nephron toward its glomerulus, where it narrows rapidly and ends in the thin-walled, relatively wide and short segment VI. The transitional zone of segment V into segment VI approaches the glomerulus intimately on its lateral and hilar aspects. Located at this site in this segment are a group of cells resembling the cells of the mammalian macula densa. The segment elsewhere is lined by cuboidal translucent cells without a brush border. It has a variable course; it may empty into a collecting tubule at almost a right angle, or may run parallel to it for a short distance before joining.

The Collecting Duct System

The collecting tubule is lined by simple flat non-ciliated epithelium, which becomes cuboidal but remains non-ciliated as it is traced into the main collecting duct. Further along the main collecting duct the cells become columnar and ciliated, and the duct also acquires a sparse investment of smooth muscle. At this point the main collecting duct comes to lie on the dorsal surface of the kidney in the midline. It then turns caudally, coursing medial to the primary branches of the ureter for a short but variable distance before emptying into one of them. The primary branches of the ureter pass caudally, joining with each other along their course to form the definitive ureter. These primary ureteric structures as well as the ureter have a prominent smooth muscle coat and a wide lumen, and they are lined by low columnar non-ciliated epithelium.

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UREA METABOLISM AND OSMOREGULATION IN THE SKATE, *Raja erinacea*

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Skates were transferred gradually during one week from 100% to approximately 50% seawater. Steady-state conditions with respect to solute and water balance were established by the end of the dilution program. Hematocrits of the fish in 50% seawater were somewhat lower than those in 100% seawater (12.5 ± 0.1 vs 15.5 ± 0.7) indicating that the skates did not osmoregulate completely in the dilute environment. Plasma concentrations of urea, trimethylamine oxide (TMAO) and chloride were 45, 23, and 30% lower respectively in the skates in 50% seawater than in the same fish in 100% seawater (Table 1). The rate of urea excretion (which reflects urea production in the steady-state) in skates in 100% seawater was 239 ± 42 μmoles/kg x hr (mean ± S.E. of six fish), 126 ± 16 in 50% seawater ($P < .05$) and 214 ± 29 when returned to 100% seawater. Urine flow was increased nearly sixfold and glomerular filtration rate doubled (although not significant statis-

Table 1

EFFECT OF ENVIRONMENTAL DILUTION ON RENAL FUNCTION IN THE SKATE, Raja erinacea

Seawater	Plasma concentration (mM)			$\dot{V}$ (ml/kg x hr)	GFR (ml/kg x hr)	$\frac{\text{Reabsorbed}}{\text{Filtered}}$ (%)		
	Urea	Chloride	TMAO			Urea	Chloride	TMAO
100%	396 ± 11 (7)	287 ± 4 (7)	48 ± 3 (7)	0.18 ± .02 (4)	0.71 ± .23 (4)	94 ± 3 (4)	73 ± 6 (3)	94 ± 2 (3)
50%	220 ± 9 (9)	202 ± 9 (9)	35 ± 5 (9)	1.04 ± .27 (3)	1.46 ± .42 (3)	62 ± 7 (3)	50 ± 5 (3)	91 ± 3 (3)
"P" value	< .01	< .01	< .05	< .02	< .2	< .01	< .05	<.5

cally, $P < .2$) in skates maintained in 50% seawater as compared to the values obtained in 100% seawater (Table 1). The fractions of filtered urea and chloride reabsorbed in the renal tubules of skates were reduced 44 and 42% respectively by environmental dilution (Table 1). The fact that TMAO reabsorption was not similarly altered in skates kept in diluted seawater argues against the possibility that the decrease in reabsorption of urea and chloride was due to generalized renal tubular failure or to some non-specific "flushing-out" of urinary solute during diuresis resulting from a greater flux of water through the fish maintained in the dilute medium. Although the renal clearance of urea was elevated in skates maintained in dilute seawater, the total clearances of urea from the body fluids of skates (calculated from total urea appearing in seawater bath) kept in diluted and undiluted seawater were similar (0.72 ± 0.20 and 0.83 ± 0.15 ml/Kg/hr respectively).

This study indicates that as in other elasmobranchs urea plays an important role in the adaptation of the skate, Raja erinacea, to alterations of the salinity of the medium. The adjustment of urea concentration in the body fluids is achieved mainly by altering rates of biosynthesis rather than total excretion of the end-product. However, the kidneys do play an important role in this adaptation by regulating the fraction of filtered urea that is reabsorbed by the renal tubules.

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IMMUNOLOGIC INVESTIGATION OF ELASMOBRANCH PITUITARY HORMONES

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As an approach toward elucidation of phylogenetic variation of pituitary hormone principles immunologic techniques have recently been applied to advantage. On the basis of bioassays, endocrine principles corresponding to mammalian prolactin appear throughout the vertebrate series. The present investigation consisted of an attempt to demonstrate and characterize the prolactin form in the anterior pituitaries of the dogfish (Squalus acanthias) and skate (Raja erinacea) by immunologic means.

Potent rabbit antisera were developed individually with regularly scheduled injections (w/ adjuvant, Freund's) of purified ovine prolactin (NIH-P-S8), shark anterior pituitary brei, or skate pituitary brei. In-gel immunoelectrophoretic analysis (IEA) presented consistent, multiple-antigen patterns for both elasmobranch pituitaries. A combination organ culture/IEA system for the shark pituitary indicated that the IEA pattern represented soluble products of the intact, isolated anterior lobe.

Numerous double diffusion (Ouchterlony) and IEA systems were designed to show cross-reactivity among the several antigens and antisera available. No indications of any common immunologic affinity were obtained. More sensitive analyses were then employed, based on neutralization of antigens, or on absorption of antibodies, before their entry into IEA or diffusion. In addition, to enhance antibody potency, globulin fractions were isolated (salt precipitation) and used as antibody agent. Absorbants included mammalian (ovine) prolactin, (bovine) STH, and synthetic ACTH, as well as elasmobranch pituitary brei. No participant antigen of either the shark