

microvilli are 2-3 times as long as those in PTS I and more densely packed. At the base of some microvilli there appear to be small pinocytotic vesicles. The lateral cell junction has a terminal bar approximately twice as long as that seen in PTS I. The adjacent lateral membranes interdigitate with long fingerlike projections arranged parallel to the long axis of the cell. In contrast to PTS I the lateral membranes are not always closely opposed. Occasional desmosomes are seen on the lateral cell surfaces. The basal membrane rests evenly on the basal lamina which is smooth and appears thinner in PTS I. The round and centrally located nucleus is of the same size as in PTS I. Again the most prominent cell organelles seen are mitochondria which are conspicuously absent just under the apical membrane but otherwise evenly and densely distributed. Rough endoplasmic reticulum and Golgi apparatus are not prominent.

In summary, the segment of the nephron which secretes the divalent ions (PTS II) differs in several respects from PTS I. The cells are much taller, the brush border more pronounced, and lateral cell borders interdigitate in different patterns.

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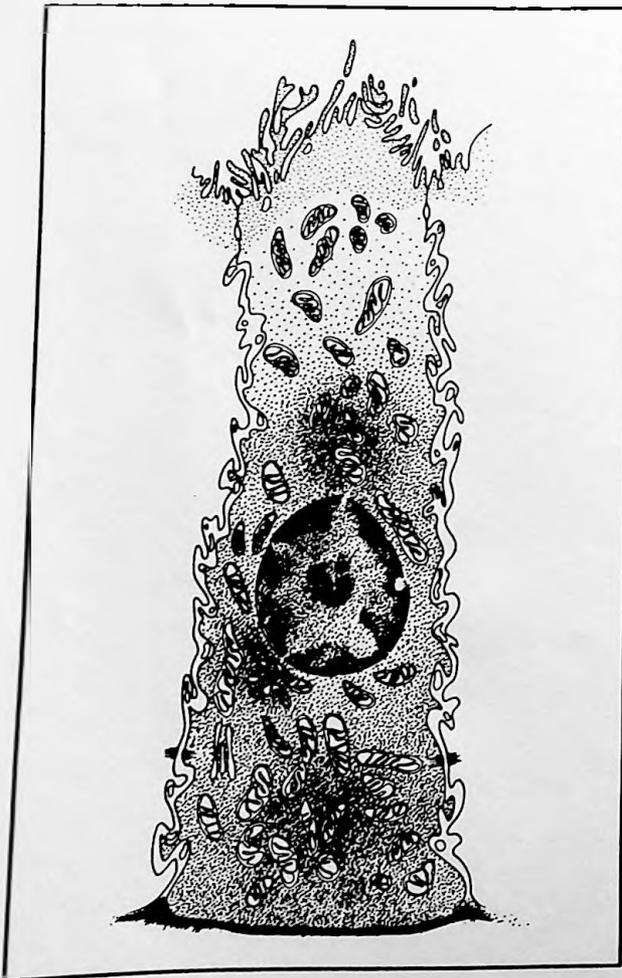


Figure 3

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The Urinary Bladder in the Little Skate, *Raja Erinacea*
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The gross anatomy of the lower urinary tract of the little skate, *Raja erinacea*, was investigated. By injecting phenol red into the tail vein 5-12 hours prior to sacrifice the urine became deeply red or yellow. Only the findings in the female skate will be reported here. The mature female has a distinct, rather large, bilobed urinary bladder (u. bl.) (Figure 1), attached caudally and dorsally to the urogenital sinus (u.s.). It is normally distended with urine (2 to 5 ml). It empties through a short urethra into the urogenital sinus via a small orifice. Eight to ten ureters (Figure 2) lead from each kidney to the dorsal surface of the bladder where they penetrate the epithelium individually. In immature female skates the bladder and urogenital sinus are much smaller. In very young specimens the urine is stored in the cloaca. Urine taken from the bladder of mature female skates is acid (pH 4.50 ± 0.08, n=6), while urine taken from the cloaca of immature specimens is less acid (pH 5.14 ± 0.05, n=4). The difference is significant, P < 0.05.

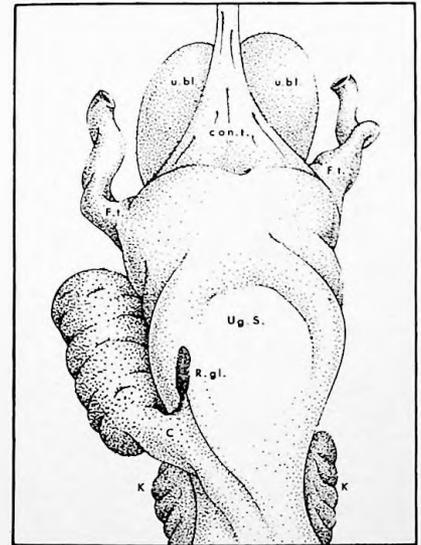


Figure 1

The mucosae of the bladders were dissected and analyzed for carbonic anhydrase content by the micro-method of Maren (*J. Pharm. Expt. Therap.* 130: 1960] using barbital buffer. The tissue contained small definite amounts of enzyme, about 3 units/gram mucosa (on this scale shark red cells have about 40 units/ml). The enzyme activity was entirely abolished by 4 M Metizolamide.

Bladders from mature female skates were removed from the animal and fixed with a phosphate buffered glutaraldehyde-papaformaldehyde fixative, embedded in Epon plastic and sectioned for light and electron

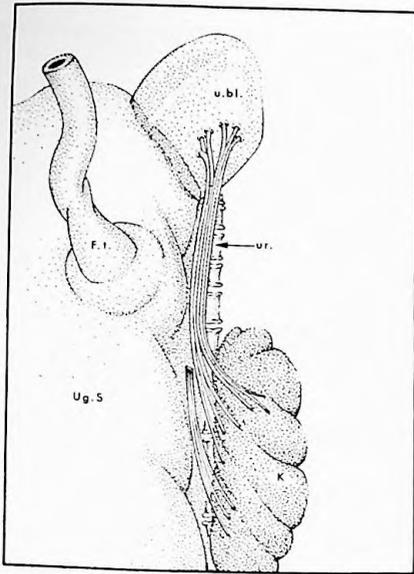


Figure 2

Electron microscopy. One micron thick sections were stained with toluidine blue-borax solution. Thin sections for electron microscopy were stained with uranyl acetate and lead citrate.

Light microscopy shows that irregularly shaped cuboidal epithelia cells line the female bladder lumen (Figure 3). Cuboidal epithelia consist of three to five cells from the lumen to a strongly basophilic staining basement membrane. Large, irregularly shaped nuclei with prominent and peripherally located nucleoli occupy the central portion of each cell. Scattered throughout the epithelia are cells intensely stained with the basic toluidine blue stain. Large, rounded, clear vesicles are apparent throughout the cytoplasm in all cells but are most numerous and positioned apically in the luminal cells. Large, open, intercellular canals (lateral spaces) are seen. Cytoplasmic extensions form a mesh-work between cells.

Electron microscopy of the female bladder (Figure 4) reveals a slightly undulating apical (luminal) membrane. A prominent junctional complex is present between adjacent luminal cells. Desmosomes with long tonofilaments are present at the contacts between thin cellular extensions of adjacent cells. Intercellular canals between cellular extensions of adjacent cells appear extremely large. Their appearance may be exaggerated as the bladder tissue was not in a distended state when fixed. Deep basal infoldings of the plasma membrane are prominent adjacent to the basal lamina where hemi-desmosomes are present. Nuclei in each cell appear centrally located, elongated and slightly irregular. The distribution, type and density of organelles are variable in each cell from the lumen to basal lamina. Small round vesicles border the perinuclear Golgi apparatus of luminal cells and are filled with a lightly staining homogenous material. Vesicles become progressively larger near the apical cell border where they fuse with the plasma membrane emptying their contents into the lumen. Densely packed mitochondria occupy the space between apical vesicles. Subluminal cells are rich in

tonofilaments with a paucity of cell organelles. Mitochondria as well as other cell organelles are not abundant in cells that do not border either the lumen or basal lamina. Between the basal infoldings mitochondria are positioned with their long axis perpendicular to the basal lamina. There is a paucity of cell organelles in the remaining regions of the basal cells. Loose connective tissue, fibroblast appearing cells and single collagen fibrils in an extensive ground matrix constitute the lamina propria. Large bundles of smooth muscle cells between numerous blood vessels are present in the deeper regions of the lamina propria.

The apically positioned vesicles with their homogeneously staining contents suggest a secretory function for these cells. Periodic Acid Schiffs Reaction applied to 8 μ m thick section of paraffin-embedded female bladder is positive in the apical region of luminal cells identifying the contents of the vesicles as mucous.

A urinary bladder is also found in teleost fish and it has been shown that Na and Cl ions are actively transported across the epithelium (Renfro, *Am. J. Physiol.* 228: 52-61, 1975). In the skate the transport characteristics of the bladder epithelium with the wide open intracellular spaces, the infoldings of the basal membrane, the apically positioned vacuoles, etc. would suggest that the epithelium is involved in transepithelial transport.

The combination of the facts that there is carbonic anhydrase in the tissue with the pH of bladder fluid (4.9) much more acidic than that of urine from sharks lacking this bladder (5.8) leads us to suspect that the bladder of female sharks is an acidifying organ.

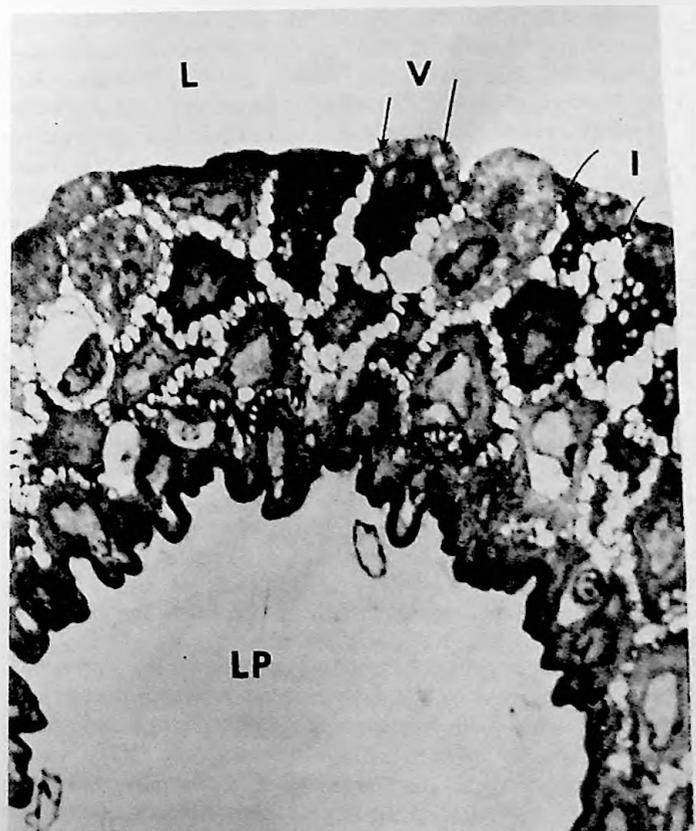


Figure 3