

either urine flow was greater than the GFR, indicating renal tubular fluid secretion, or inulin and PEG were reabsorbed. In none of the sea water-acclimated eels did the urine flow exceed the GFR. In both groups of eels the urine was osmotically dilute. The average free water clearance (C_{H_2O}) was 1.57 ml/kg hr. in fresh water and 0.41 ml/kg hr. in sea water-acclimated eels. The difference in C_{H_2O} (1.15 ml/kg hr), corresponds closely to the difference in urine flow between the two groups, 1.04 ml/kg hr.

Aglomerular urine secretion is typical of the aglomerular marine teleosts. It has, however, been observed sporadically in glomerular teleosts, e.g., Grafflin (J. Morphol. 61:165-173, 1937) and Hickman (Canadian J. Zool. 46:427-437, 1968). It is unusual that it should be found in the fresh water- rather than in the sea water-acclimated eels.

Supported by research grant NIH 09975-06 and training grant GMO 1699-04.

1971 #42

RENAL COLLECTING DUCT FUNCTION IN THE LITTLE SKATE, *Raja erinacea*.

H. Stolte, G.M. Eisenbach, D. Antkowiak and J.W. Boylan; State University of New York at Buffalo, Buffalo, New York and Medizinische Hochschule, Hannover, W. Germany

The renal collecting ducts of the skate originate segmentally on the dorso-lateral surface of the kidney and receive the terminal portions of nephrons from each segment. The ducts turn, coursing medially between lobules, and enter the kidney substance at its dorso-medial aspect. Emerging at the ventral surface the ducts enter the urinary sinus, either directly or after joining the other tributary ducts. The collecting ducts are visible and accessible to micropuncture throughout their dorsal surface portions.

We have examined fluid taken from early and successively more distal portions of the collecting ducts for osmolality and electrolyte composition ($Na^+ + K^+$) and compared these to more proximal tubular fluid and to final urine. In some experiments collecting ducts were filled retrograde with colored microfil and the more distal surface tubules visualized. The collected fluid was analyzed for osmolality by the freezing point method of Ramsay-Brown and some samples for sodium and potassium concentration by the microflame photometer of Hampel. (We are indebted to Dr. Bodil Schmidt-Nielsen for the use of this equipment and to Mr. Yogendra who did the flame photometry).

Studies were completed on 20 male and female skates weighing from 0.88 to 1.59 kg and anesthetized by injection of 0.45 mgs/kg sodium pentobarbital and 0.45 kgs/kg curare into a lateral tail vein. Fish were placed on a board dorsal or ventral side up, the spiracles perfused with aerated sea water (100 or 75% at 1 - 1.5 L/min. thermostated at 10°C.) A paramedian incision exposed the dorsal or ventral surface of the kidney for direct visualization. To minimize interference with the renal blood supply, the tail was not fixed. The exposed surface of the kidney and surrounding tissue were covered with a layer of 2% liquid agar which hardened in a few minutes. The site of micropuncture was then exposed by a small window cut in the agar. Samples were aspirated through sharpened micro glass capillaries ground to a tip diameter of 8-15 μ . The appearance of the area of the puncture site is shown in Figure 1, after retrograde injection of microfil. The collecting ducts,

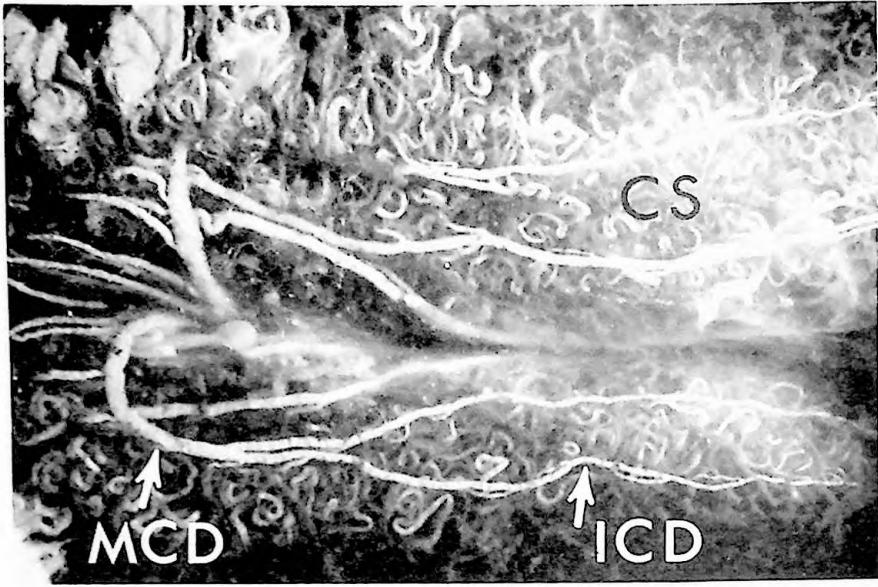


Figure 1

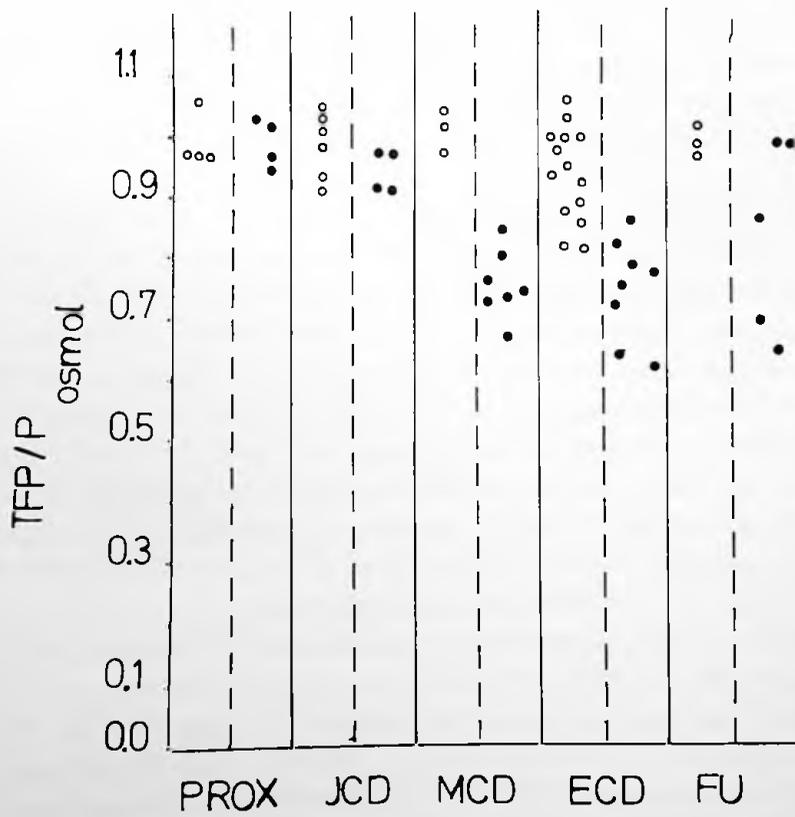


Figure 2

distal coiling segments and immediately proximal segments of the nephron are visualized.

In Figure 2 the calculated osmolar tubular fluid to plasma ratios (TF/P_{osm}) for fish in 100% sea water are plotted against the micropuncture sites (open circles). Proximal tubular fluid, probably segments III and V of Deetjen and Antkowiak (Bull. MDIBL 10:5, 1970) was always isosmotic to plasma. Fluid from initial collecting duct (ICD) or midcollecting duct (MCD) also was not significantly different from plasma. End collecting duct (ECD) fluid was slightly but significantly hypo-osmotic ($TF/P_{osm} = 0.93 \pm 0.02$ SE.) The data indicate the collecting duct as the most important site of urinary dilution under these experimental conditions and in a normal sea water environment.

Dilutional activity is exaggerated when the skate has been kept in 75% aerated sea water for more than 24 hours (closed circles, Figure 2). Under these conditions dilution probably is demonstrated in the ICD and is quite marked in MCD and ECD, $TF/P_{osm} 0.76 \pm 0.02 - 0.75 \pm 0.03$ SE. Samples taken from fish exposed to diluted sea water for 14 hours were indistinguishable from controls (TF/P_{osm} of MCD 0.94 - 1.01). Adaptation of the final nephron segments to the stress of a dilute environment, therefore, exhibits a time course.

The effect of 24 hours exposure to 75% sea water on plasma osmolality, hematocrit and plasma Na and K concentration are shown in Table 1. Dilution is accomplished by the reabsorption of sodium. The calculated sodium and potassium ratios of ECD fluid to plasma in control and 75% sea water are also shown in the Table. As the concentration ratio for sodium between collecting duct fluid and plasma decreased, the ratio for potassium increased remarkably (Table). The osmolality difference in CD fluid between control and dilute environments can be accounted for completely by sodium and potassium differences:

Table 1 Hematocrits, plasma and end collecting duct solutes after 24 hrs. in 75% sea water (values are means and S.E.).

SEA WATER	HEMATOCRIT		PLASMA		END COLLECTING DUCT	
	%	Osm (mosmol/l)	Na ⁺ (mEq/l)	K ⁺ (mEq/l)	Na ⁺ (TF/P)	K ⁺ (TF/P)
100%	20.32 ± 2.08 (6)	982.7 ± 22.2 (6)	269.5 ± 17.3 (3)	4.1 ± 1.8 (3)	0.91 ± 0.03 (13)	2.57 ± 0.63 (12)
75%	16.32 ± 1.13 (6)	806.1 ± 36.7 (8)	181.8 ± 10.7 (7)	4.3 ± 1.5 (3)	0.58 ± 0.13 (10)	14.47 ± 5.71 (6)

In summary: (1) The collecting duct is the principle site of urinary dilution in the skate kidney. (2) Dilution of the environment is followed after about 24 hours delay, by an increase in the trans-epithelial concentration gradient for sodium in the terminal collecting duct sufficient to account for the dilution.

Supported by National Institutes of Health Grant No. AM - 14424-02.