

later investigators found sodium concentrations of brain at 120 mEq/L  $H_2O$  and red cell at 33 mEq/L  $H_2O$ . The Column-values for  $Na^{22}$  are different for various tissues, and one may assume that these counts are more or less equilibrium values for the whole tissues. Column 2 of Table 3 presents counts corrected to gm tissue water.

It turns out that of the counts injected, 12 percent wind up in the muscle which is 43 percent of the total body weight, while 8 percent are in the skin which is only 4 percent of total body weight. Apparently the largest sodium pool is the extracellular fluid, including blood, and then, strangely, followed by the skeleton.

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#### CONTINUED STUDIES ON SODIUM FLUXES AND RATES OF TISSUE UPTAKE OF SODIUM IN THE DOGFISH, Squalus acanthias

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Whether unidirectional or not, the exchange of water and electrolyte through the external surfaces of aquatic organisms is a basic problem. Data of Burger and Tosteson, Comp. Biochem. Physiol. 19:649-53, 1966, and Burger and Horowicz, Bull. M.D.I.B.L. 1966: 8-9, indicate a net sodium influx through the head end of the spiny dogfish. One of us (PH) developed a unique mathematical treatment which slightly revised the values published here in 1966. Mean efflux was 0.16 mEq Na/kg hr; mean influx was 0.68. Using the same methodology as previously, but calculating by conventional methods, the following new data were secured.

1. Sodium influx is not due to or augmented by the drinking of sea water during the experimental periods. All fish that were in an external bath containing  $Na^{22}$  had their stomach contents analyzed. There was no evidence of even minute drinking.

2. Sodium efflux into a choline chloride branchial perfusate averaged 0.064 mEq/kg hr (Farber, Gerstein, and Boylan, Bull. M.D.I.B.L. 1965:14). Immersing a dogfish head in a pure sucrose bath, isotonic to sea water gave values like those of Farber *et al.* The average efflux from four fish was 0.069 mEq/kg hr (0.053 - 0.086). It is noted that the efflux is reduced over what it was in sea water, possibly because of the absence of external ions for exchange. Since the dogfish has a positive arterial pressure, it seems reasonable to regard the efflux as hydrostatically induced leakage. As seen below, the skin is rich in sodium.

3. Influx from a sucrose bath isotonic to sea water was studied. In order to have some basis for calculation,  $Na^{22}$  was mixed in 5 ml sea water which was added to a 2 liter sucrose bath. For ten periods in three fish, influx averaged 0.0093 mEq Na/kg hr. If the bath were sea water (440 mEq/1), this rate would give an influx of 0.52 mEq/kg hr, which is in the range of previously determined normal influx. It appears that whatever the mechanism effecting influx, it operates at the same rate whether the external medium is low or rich in sodium.

4. Dogfish without a rectal gland maintain normal plasma electrolyte and osmolarity for a 21 day test period (Burger, Physiol. Zool. 38:191-96, 1965). It was reasoned that these glandless fish must have a decreased sodium uptake. To test this directly, four dogfish had their rectal glands removed and four controls had a dummy operation. After fourteen days, only two

glandless fish had their sutures intact, so two glandless fish were compared to two control fish for sodium uptake from an external bath. No differences in sodium uptake were found, nor did these unfed laboratory-penned fish become increasingly leaky inwardly to external sodium. The mechanism whereby rectal glandless fish maintain normal plasma is still not resolved.

5. Attention was given to the rate of sodium exchange within the dogfish.

a. Arterial-venous differences. In four fish, the dorsal aorta was cannulated by caudal puncture with the catheter pushed well forward. The lateral abdominal vein was catheterized with the catheter well forward. This vein drains the lateral body wall and is free from the action of the kidneys or gut. It is emphasized, that the blood is not mixed venous blood as can be got only from the heart or ventral aorta, but a specialized sample draining the striated muscle and skin. Isotope was provided by natural influx through the head from an external bath. The A-V difference in the first half hour when the tissues had presumably little isotope to return to the veins, averaged 25 percent (range: 17-41 percent). It seems reasonable to suggest that as the blood circulates through muscle 25 percent of the capillary blood sodium is filtered and reabsorbed. If one further postulates a rough correspondence between the capillary filtration of water and sodium, and calculates from the average data given in the preceding report, the rate of filtration in the muscle is in the range of 0.4 ml/g hr. For the kidney where the rate of one capillary bed, the glomerulus is known, the filtration rate is in the range of 0.7 ml/g kidney hr, but here of course the glomeruli are only a fraction of the total kidney mass. It should be added that the above calculations do not imply that this is rate of penetration into the muscle cells themselves.

b. Rate of penetration of sodium into tissue masses. Table 3 of the preceding report gives isotope counts for various organs. There is wide variation, undoubtedly reflecting differences in sodium content of various tissues, e.g. it is known that muscle sodium is very low. Table 1 is an attempt to relate the distribution of  $\text{Na}^{22}$  from the blood into the various tissues as a function of time. Again, it is noted that the whole sodium pool of a tissue is involved, not

Table 1  
SODIUM UPTAKE BY DOGFISH TISSUES WITH TIME

	$\text{Na}^{22}$ counts per min per gram wet tissue/counts per min per ml terminal plasma				
	0.33 hr	1.5 hr	3 hr	3.5 hr	11 hr
Skin	0.40	0.54	0.54	0.54	0.55
Cartilage	0.026	0.15	0.38	0.36	0.56
Brain	0.071	0.089	0.11	0.19	0.31
Muscle	0.044	0.083	0.055	0.067	0.086
Rectal gland	0.37	0.33	0.36	0.34	0.33
Pancreas	0.20	0.21	0.22	0.19	0.24
Liver	0.011	0.077	0.062	0.058	0.056
Stomach	0.28	0.19	0.37	0.47	0.39
Uterus	-	0.51	0.59	0.57	0.67
Kidney	-	0.47	0.32	0.38	-

the intracellular sodium alone. After preparing Table 1 from several different bases, it was decided to present the data in a simple ratio: counts per gram of wet tissue/counts per milliliter of terminal plasma. Another method was counts per milliliter of tissue water/half-time counts per milliliter of plasma. One can calculate the ratios tissue water/plasma using Table 1, and Table 3 in the preceding report.

In a general way, these ratios can be taken as a measure of the rate at which the sodium spaces of the various tissues are filled. For brain and cartilage the ratios increase with time. For the rectal gland, the ratio remains constant with time. Except for a few out-of-line values the ratios form an orderly picture.

These ratios seem to reflect the intracellular sodium content. If the ratios merely measured extracellular space/plasma, one would expect that in eleven hours the ratios would approach one, which they do not do. Indeed if one takes the few reported intracellular concentrations for sodium in brain and muscle then the ratio mEq Na per ml tissue water/mEq sodium per ml plasma is nearly equal to the ratio of counts per ml of tissue water/counts per ml plasma. Of course certain tissues such as cartilage and skin have large non-cellular spaces. It does seem possible from the above data and that in the preceding report, to approximate the sodium content of various organs.

The ratios indicate that since the various tissues do not have a uniform rate of sodium exchange, it is not possible to define when the animal is in equilibrium with an injected dose. However, for practical purposes, two hours seems a generalized time for good internal mixing. Graphically, falling plasma curves begin to flatten out at this time, and the counts per ml plasma correspond to what one would expect if the total counts were mixed in a volume equal to 20% of the body weight.

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#### SODIUM EXCHANGE THROUGH THE EXTERNAL SURFACES OF Myxine

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Unlike marine elasmobranchs which produce a hypertonic plasma by adding to blood electrolyte osmotically active organic moieties, the marine cyclostomes effect a slight hypertonicity largely through electrolytes. It is of interest to know whether electrolyte exchange takes place through the skin and gills in all marine forms, and if so to estimate the rate of exchange. This statement carries no implication that there must be a directed flux.

Morris (J. Exp. Biol. 42:359-71, 1965) gives numerous data on the salt and water content of the hagfish, Myxine, together with data on urine flows, drinking sea water, etc. The study here is concerned solely with sodium exchange between the external surfaces of hagfish and its environment as measured with  $\text{Na}^{22}$ .

The handling of the fish (M. glutinosa) was essentially that described by Rall and Burger (Am. J. Physiol. 212:354-56, 1967). In addition, a cord was tied around the animal anterior to the cloaca to prevent fluid loss while the fish was mounted for sampling. In efflux studies this same technique was used prior to injection of the isotope. Some fish (A, D series) were used