Table 1

INCREASED FRACTIONAL UREA EXCRETION WITH ECF EXPANSION AND ENVIRONMENTAL DILUTION

Period	$\dot{V}$	GFR	Clearance (ml/kg x hr)		Percent excreted (of filtered load)	
	(ml/kg x hr)		Urea	Cl^-	Urea	Cl^-
1.85 kg Squalus						
Control 1	1.48	3.86	0.15	0.97	3.9	24.9
Control 2	1.41	3.30	0.30	1.07	9.1	32.3
4% of body wt balanced isotonic solution i.v.						
Experiment 1	1.75	4.61	2.01	1.64	43.5	35.3
Experiment 2	1.89	3.76	2.21	1.39	59.0	36.9
1.97 kg Squalus in 70% seawater						
1	2.80	5.30	1.97	1.33	37.4	25.1
2	3.30	5.49	2.34	1.62	42.7	29.6

Also included in Table 1 are data from a single dogfish after its "accommodation" to diluted (70%) seawater. The spiny dogfish is not truly a euryhaline fish and clearance values were found to vary widely in these animals which have a limited capacity to adapt to hypotonic conditions. However, the most consistent feature again was the striking rise in percentage of urea excreted, 37.4 and 42.7% of the filtered load in this instance. Corresponding values in 3 other dogfish under similar conditions averaged 42, 29 and 37%, respectively. Reductions in hemoglobin content of 35, 33, 25 and 34%, indicate appreciable expansion of fluid volume in the 4 fish maintained in this hypotonic environment.

Extraglomerular control mechanisms are indicated in these experiments involving exposure to a dilute environment, administration of epinephrine, and the expansion of extracellular fluid volume. Further direct micropuncture measurements are needed to confirm the indication that the tubular reabsorption of urea, water and electrolytes in these instances is controlled by variations in perfusion pressures of the peritubular vascular bed.

Supported by PHS Grant HE-04457 and NSF Grant GB 8200.

1970 #13

UREA BIOSYNTHESIS AND EXCRETION IN FRESHWATER AND MARINE ELASMOBRANCH

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Urea concentration is markedly lower in the body fluids of freshwater elasmobranchs than in marine forms. Since the concentration of urea in the body fluids is the difference between rates of biosynthesis and excretion, we compared the urea biosynthetic capacity and rate of excretion of urea in the freshwater stingray Potamotrygon with these same parameters in the marine elasmobranch Squalus acanthias. The levels of ornithine-urea cycle enzymes assayed in liver homogenates from Potamotrygon were significantly lower than the levels of these enzymes . assayed in liver homogenates from Squalus. The activity of carbamoylphosphate synthetase, the rate limiting enzyme of the cycle, was 1/10 the activity in Potamotrygon liver compared to Squalus (0.4 vs 4.0 μmoles/g liver x hr, means of four fish and two fish, respectively). The rate of incorporation of ^{14}C-bicarbonate into urea by liver slices from Potamotrygon was 1/100 that of Squalus (3 x 10^{-2} vs 3.0 μmoles/g liver x hr, means of three fish each).

Using ^{14}C-urea we found that its rate of excretion in Potamotrygon was approximately 20-50 times greater than in Squalus (Figure 1). Thiourea, the sulfur analogue of urea which is not actively reabsorbed by the renal tubules of elasmobranchs (B. Schmidt-Nielsen and L. Rabinowitz, Science 146:1587, 1964) was used as a standard of reference to determine whether or not urea was actively reabsorbed by the renal tubules of Potamotrygon. Approximately 10 μC ^{14}C-urea (S.A. 4.86 μC/μM) and 10 μC ^{14}C-thiourea (S.A. 2.69 μC/μM) per kg were injected into the caudal vessels of four Potamotrygon (P$_1$ - P$_4$ and four Squalus (S$_1$ - S$_4$). Blood samples were drawn 6 hours (equilibration period) and 22 or 30 hours later. Plasma samples (0.1 - 0.5 ml) were either (1) directly mixed with 15 ml scintillation solution and counted in a liquid scintillation counter or (2) treated first with urease to convert ^{14}C-urea (but not ^{14}C-thiourea) to $^{14}CO_2$, gassed with $^{12}CO_2$ for 2 hours and then mixed with scintillation solution and counted. The percents of injected ^{14}C-urea and ^{14}C-thiourea that were excreted over a 24 hour period were calculated from these results. As shown in Figure 1 the rate of excretion of ^{14}C-thiourea in the freshwater stingray was similar to that of ^{14}C-urea indicating that the two compounds

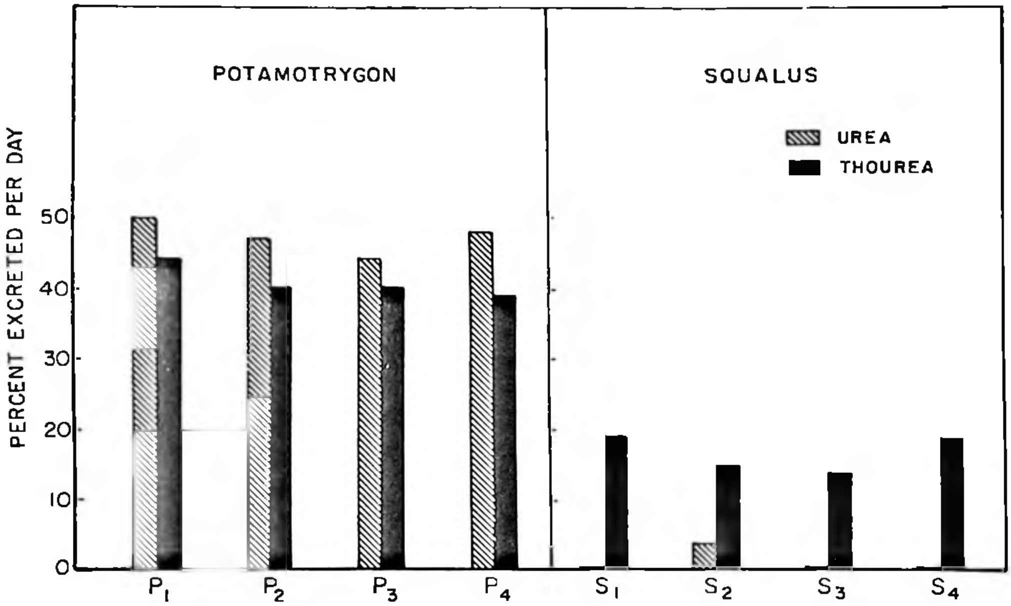

Figure 1. Comparative excretion rates of ^{14}C-urea and ^{14}c-thiourea in Potamotrygon and Squalus acanthias.

are handled similarly by the renal tubules of this elasmobranch. It was difficult to determine the manner in which urea is handled by the kidneys of Potamotrygon because of the inability to collect urine samples uncontaminated with cloacal contents in these small fishes (ca 100g) with no urinary bladder. Glomerular filtration rates estimated from inulin clearances average 8.3 ml/kg x hr as determined in four clearance periods in two freshwater rays. These values are 10-15 times those found in small marine skates Raja erinacea and significantly higher than glomerular filtration rates observed in Squalus. In contrast to marine elasmobranchs in which 85-90% of the urea filtered at the glomerulus is reabsorbed by the renal tubules, a renal clearance study done on a single Potamotrygon indicated that only about 50% of the filtered urea is reabsorbed by the renal tubules of this fish. Thus, acclimatization of this elasmobranch to freshwater environment is accompanied both by increased glomerular filtration rate and decreased tubular reabsorption of urea, both factors leading to elevated rates of renal excretion of the compound. These studies indicate that the low level of urea in Potamotrygon is due to both increased rate of excretion and decreased rate of biosynthesis of the compound.

Supported by NSF Grant GB 8200 and PHS Grant HE-04457.

1970 #14

INVESTIGATIONS OF LIVER RESPIRATION IN DOGFISH (Squalus acanthias) AND LITTLE SKATE (Raja erinacea)

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Previous studies (Physiol. Zool. 42:231-47, 1969) have shown that in the skate, Raja erinacea norepinephrine is twice as effective as epinephrine in elevating blood glucose levels but has little effect in the dogfish, Squalus acanthias. The present investigation was initiated as a pilot study to investigate the effect of norepinephrine and other substances on respiration in whole homogenates of elasmobranch liver. Routinely, 700 or 350 mg of tissue were homogenized in a 3 ml chilled medium of sterilized seawater or modified Elasmobranch Ringers (no glucose component). Both media proved equally successful. Respiration was determined using a YSI model oxygen electrode in test runs of 15 minute duration at 15, 17, 19 and 25° C, and recorded in μl O_2 consumed ml homogenate/minute. In one series of experiments respiration was studied on 100 mg slices of skate liver.

Rise in temperature from 15° C to 25° C in 700 mg of dogfish homogenate produced a slight rise in respiration from an average of 0.062 to 0.083 μl O_2/ml/min, but a threefold rise in the skate from an average 0.094 to 0.312 μl O_2/ml/min. This species differential is probably due in part to the higher fat content of the dogfish liver preparations. However, as nitrogen determinations were not made, species comparisons remain obscure. Determinations were eventually mad on 350 mg of skate tissue at 25° C so as to achieve oxygen consumption ranges closer to those fo 700 mg of Squalus liver.

The results of metabolic substances and mammalian hormone preparations on homogenate respiration are described below. During each trial an experimental sample was run simultaneously with that of a control. Recordings from the experimental and control electrode were alternated at approximately two minute intervals.