Table 1 is a compilation of some of the enzymic activities measured in the nucleated erythrocytes of several marine species in comparison with activities of related enzymes in human and harbor seal *(Phoca vitullina)* erythrocytes. Particularly noteworthy is the high activity of nucleoside diphosphokinase (NDP kinase) found in the erythrocytes of all of the species examined. Strict comparison of these relative enzymic activities is not possible because assays with human erythrocytes were measured at 30° whereas some of the determinations in other species were made at room temperature. This laboratory has recently reported the occurrence of marked heterogeneity of NDP kinase in the human erythrocyte and has identified at least six isozymes with isoelectric points varying from 5.4 to 8.3 (Biochemistry 10, 2139, 1971). It was of considerable interest to find that similar marked electrophoretic heterogeneity occurs in the erythrocytes of all of the species examined.

Figure 2 presents the results of an isoelectric focusing experiment with the centrifuged, dialyzed supernatant fluid of hemolyzed hagfish erythrocytes. Here it was found that four distinct isozymes of NDP kinase occur with isoelectric points of 5.2, 6.0, 6.3 and 7.3. The true function in erythrocytic metabolism of these isozymes of NDP kinase is not apparent, but it seems possible that more detailed comparative studies with a number of species may provide useful insights.

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UREOSMOTIC REGULATION BY THE KIDNEYS AND GILLS OF THE LITTLE SKATE, *Raja erinacea*

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The renal and branchial excretion of urea were studied under various experimental conditions: in skates adapted to sea water or 50 percent sea water and in skates acutely transferred to 75 percent sea water. Urine collections were made by means of a cloacal catheter attached to a toy balloon. The catheter was generally sewn in place the day before the experiment. Branchial efflux of urea was studied with ^{14}C-urea. The skates (with cloacal catheters) were placed in aerated aquaria containing a known volume of external medium (about 4 liters). The branchial flux was calculated by dividing the cpm appearing in the external medium by the specific radioactivity in plasma of fish. The results are shown in Table 1. Two groups of fish were used. One group was studied in sea water and again after acute transfer to 75 percent sea water. The second group was gradually (5 days) adapted to and studied in 50 percent sea water. Osmotic pressures of the external media were

TABLE 1

EFFECT OF ACUTE AND PROLONGED ENVIRONMENTAL DILUTION ON RENAL AND BRANCHIAL UREA EXCRETION IN *Raja erinacea*.

SEAWATER	PLASMA		UREA EXCRETION	
	UREA (mmoles/l)	OSMOTIC PRESSURE (mosmoles/kg)	KIDNEYS (μmoles/100gxhr)	GILLS (μmoles/100gxhr)
100%	389±2.4 (4)	932±11.0 (4)	2.2±0.54 (4)	57.0±9.64 (6)
75%	371±4.3 (4)	883±13.4 (4)	11.3±1.17 (4)	71.1±12.41 (6)
P value *	<0.01	<0.05	<0.001	
50%	227±26.3 (5)	632±12.6 (5)	23.4±5.81 (5)	19.8±5.99 (5)
P value **	<0.001	<0.001	<0.02	<0.02

Values are means ± S.E. Numbers of fish per group shown in parentheses. * P Values refer to group in 75% seawater compared to group in 100% seawater. ** P Values refer to group in 50% seawater compared to group in 100% seawater. Fish in 100% and 50% seawater were fully adapted. Fish in 75% seawater were studied during the first 4 hours after transfer.

927 ± 3.4 (mean ± S.E. of four determinations), 706 ± 2.0 and 498 ± 11.9 mosmoles/kg for 100, 75 and 50 percent sea water. As shown previously by Goldstein and Forster (Am. J. Physiol. 220:742, 1971), the renal excretion of urea is significantly augmented in skates adapted to 50 percent sea water compared to skates maintained in 100 percent sea water. The urine flows were 29.1 ± 2.45 (mean ± S.E. of four fish) and 274.2 ± 41.06 (mean ± S.E. five fish) μl/100g x hr respectively in the two groups. The differences in urine flows between the two groups are in accord with the differences in osmotic gradient across the gills: 135 ± 2.65 (mean ± S.E. five fish) and 7 ± 4.8 (four fish) mosmoles/kg in 50 percent and 100 percent sea water, respectively. The osmotic permeability of the gills of this species of skate does not seem to vary with changes in environmental salinity. We have shown in a separate study that the diffusional water permeability of the gills is the same in *R. erinacea* adapted to 50 and 100 percent sea water (Payan and Goldstein, this issue).

After acute transfer of skates from 100 to 75 percent sea water there was an immediate renal response. Urine flow increased from 29.1 ± 2.45 (n=4) to 168.5 ± 16.3 (n=4) μl/100g x hr. This was accompanied by a 5-fold increase in renal urea excretion (Table 1). The increase in urea excretion following transfer to the dilute medium was due mainly to an increase in the fractional excretion of urea (3-4 fold) with only a modest increase in GFR (30 percent). GFR was measured as the inulin clearance, with inulin concentrations determined simultaneously in plasma and cloacal "urine". Branchial efflux of urea did not change during the first few hours after transfer of skates from 100 to 75 percent sea water. However in skates adapted to 50 percent sea water over a period of 5 days, by gradually lowering the salinity of sea water with freshwater, branchial excretion of urea was reduced to one-third. This result was predicted by Goldstein and Forster in a previous study (Am. J. Physiol. 220:742, 1971).

It is interesting to note the respective roles that the kidneys and gills play in the adaption of urea excretion to changes in environmental salinity. In undiluted sea water the renal excretion is

about 3 percent of the total urea excretion, the remainder being excreted by the gills. During acute transfer to 75 percent seawater there is an increase in total urea excretion of about 20 percent due to an elevation in renal urea excretion. In skates adapted to 50 percent sea water the total urea excretion is similar to that in sea water but then about 60 percent is excreted by the kidneys.

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DIFFUSIONAL WATER FLUXES IN SKATES, *Raja erinacea* and *Raja radiata*

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Diffusional water permeability ($^3H_20$ efflux) was measured in the skates, *Raja erinacea* and *radiata*, which are able to adapt to a dilute environment. All experiments were carried out at a temperature of 13° ± 1°C. The values obtained for the two species of skates were similar and were grouped together. No significant difference was observed in the turnover of internal water (λ) between the two groups of fish, those in sea water and those adapted to 50% sea water (Table 1). The cor-

TABLE I

TURNOVER OF INTERNAL WATER IN INTACT AND HYPOPHYSECTOMIZED SKATES ADAPTED TO SEAWATER AND 50% SEAWATER

	SW		50% SW	
	INTACT	HYPOX	INTACT	HYPOX
λ(%.h^{-1})	64.2±1.91 (n=7)	64.1±2.78 (n=7)	57.5±1.48 (n=5)	66.6±5.57 (n=6)

Values are means ± S.E. with numbers per group in parentheses.