female fish and there also tended to be an inverse relationship between fish weight and hepatic AHH activity. However, these differences were relatively small and the statistical significance was in part a reflection of the large sample sizes employed in the analysis.

There was also a positive correlation between hepatic AHH activity and 7-ERF activity in the individual flounder studied. This is not surprising since cytochrome P-448 from rat hepatic microsomes supports the rapid oxidative metabolism of both benzo(a)pyrene and 7-ethoxyresorufin. This is also the case in hepatic microsomes from flounder, little skates (*Raja erinacea*), and sheepshead (*Archosargus probatocephalus*) that were treated with polycyclic hydrocarbons.

We were also interested in determining whether or not the date of sacrifice (and assay) was related to hepatic AHH activity of flounder. However, a plot of AHH activity *versus* date of assay did not illustrate any clear-cut relationship between these two parameters although it did reemphasize the variability of enzyme activity in the fish studied.

The causative factor for the induction of AHH activity in about half of the flounder examined has still not been clearly elucidated. However, since individual induced fish were found in all groups tested over the course of the summer, it does not appear to be related to spawning. It is also interesting that both male and female fish were affected. These observations are consistent with the enzyme response being caused by an exogenous factor, such as the presence of a polycyclic hydrocarbon-like inducer in the environment. This possibility is further enhanced by the fact that the enzyme responses monitored in the fish with high hepatic AHH activity are identical to those found in flounder that have been pretreated with polycyclic hydrocarbons. However, additional studies are still required before pollutants are definitely associated with the elevated hepatic AHH activities in many (but not all) Maine flounder, and more importantly, before enzyme induction in fish is used as a biochemical monitor for toxic environmental contaminants.

THE EGG CASE OF *Raja erinacea*: MECHANICAL PROTECTION OR OSMOREGULATORY DEVICE?

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The little skate, *Raja erinacea*, is oviparous and lays egg cases throughout the year (Bigelow and Schroeder, Fishes of the Gulf of Maine, p. 69, 1953). Smith (Biol. Rev. 11:49-82, 1936) suggested that the cases (as well as the female uteri in ovoviviparous and viviparous species of elasmobranchs) provide for an osmotically isolated environment until the embryo is capable of urea retention and osmoregulation. However, Reed (Comp. Biochem. Physiol. 24:668-675, 1968) found that encapsulated embryos of the skate *Raja binoculata* do possess the enzymes of the Krebs ornithine-urea cycle and are able to retain near adult levels of both urea and trimethylamineoxide (Reed, Biol. Bull. 135:537-547, 1968). The retention of urea seems to be secondary to an urea-impermeable egg membrane since Needham and Needham (J. Exp. Biol. 7:7-18, 1930) had shown that the egg cases are permeable to urea.

Despite the ability to maintain high urea levels, it appears that osmoregulation in seawater is limited since Libby (Nat. Geogr. Mag. 116:412-420, 1959) found that if embryos of *R. eglanteria* are removed before day 20 of the 64-day developmental period (after day 20 the mucous plug of the egg case is dissolved and seawater enters the egg case), they are unable to survive in seawater.

It, therefore, appears that the full complement of osmoregulatory mechanisms is not developed before approximately 1/3 of the developmental period is over. It is obvious that the data on the ontogeny of elasmobranch (and teleost, for that matter) osmoregulation is sketchy at best. To initiate our investigations of this system we measured the Na^+, K^+ and Cl^- concentration of the intracapsular fluids and the Na^+ efflux with newly-shed egg cases of the little skate, *Raja erinacea*.

TABLE 1

Intracapsular ionic concentrations and Na^+ efflux from *R. erinacea* egg case

[Na]*	[K]	[Cl]	Na efflux**
261 ± 21	7.4 ± 0.5	557 ± 61	24 ± 3%.hr^{-1}
(9)	(9)	(9)	(10)

*Ionic concentrations in mM.l^{-1} ± S.E. (N)

**Na efflux in % intracapsular Na.hr^{-1} (KX 100)

Ten egg cases were collected within 24 hours of extrusion from the female and maintained at 15°-17°C in running seawater for 5 days or less. They were injected with 10 µl (2 µCi) of ^{22}Na solution and placed into 100 ml of seawater (in an 800 ml plastic beaker) maintained at 15°-17°C by placing the experimental containers in a running seawater system. The site of injection was plugged with silicone grease. (This may have been unnecessary since subsequent loss of the silicone plug did not alter the efflux.) At various times post-injection (for up to 24 hours), 5 ml samples of the solution were removed and analyzed on the Packard Autogamma system for ^{22}Na activity. An aliquot of the diluted injection solution was also counted to determine the amount of radioactivity injected into the egg case. Effluxes were calculated from the formula $K = 1/t$ in C_o/C_t where K is the rate constant (fraction of isotope lost perhour), t is the time of the experimental period in hours, and C_o and C_t are the radioactivity in the egg case at the start and end of an experimental period, respectively. Since preliminary experiments indicated that it took approximately 2 hours for equilibrium of the isotope within the fluids of the egg ccase, the rate constant was calculated from time periods subsequent to 3 hours. After the final sample of the efflux bath, the egg cases were placed into another, nonradioactive bath for at least 48 hours (to allow total washout of the radioactivity) and then opened and samples of the intracapsular fluids taken. The samples were frozen, shipped to Miami, and analyzed for Na^+ and K^+ (via flame photometry), and Cl^- (via amperometric titration).

Table 1 shows that the capsular fluids are not ionically equivalent to either the surrounding seawater (Na^+ = 380 mM, Cl^- = 442 mM, K^+ = 9.3 mM) or the blood of the adult (Na^+ = 299 mM and K^+ = 4.96 mM, Forster et al., Bull. MDIBL 14:20-25, 1974). Unfortunately, we have no data on the total osmolarity or the urea content of the capsular fluids. The extremely high concentrations of Cl^- are especially interesting; it is not known whether these represent experimental error or a substantial ionic imbalance in the capsular fluids.

Table 1 also indicates, for the first time, that the egg case is extremely permeable to atleast Na^+. Similar efflux rates were found in the egg case of the Mediterranean shark, *Scyliorhinus canicula* (Evans, unpublished). This extremely high rate of efflux of injected ^{22}Na is of the same order of that described for intact teleost fish but some 50-100 times that normally found in elasmobranchs. It is, therefore, obvious that the skate egg case represents a rather leaky barrier between the external seawater and the ionically dissimilar capsular fluids. One might assume that this indicates some ionic regulatory role played by some capsular membrane between the two solutions, but further experiments are necessary before this conclusion can be made. Research supported by NSF PCM 77-09915.