

Figure 2.--Follicle diameter and serum 17- β estradiol levels in female Raja erinacea.

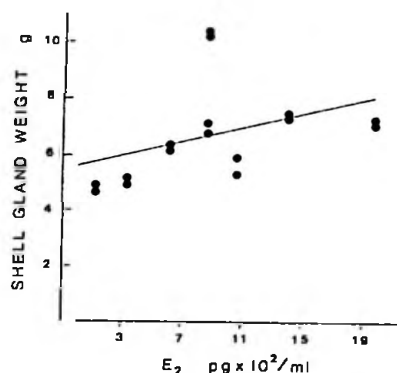


Figure 3.--Shell gland weight and serum 17- β estradiol levels in pre-spawning Raja erinacea.

gland growth is coincident with both follicle enlargement and rising estradiol levels, it is likely that follicle derived estradiol influences shell gland function. We, as yet, have no direct evidence that estradiol can regulate nidamental gland function but are currently investigating this possibility. This work was supported by NSF PCM 81-04144 to I.P.C.

THE EFFECT OF MAMMALIAN INSULIN ON PLASMA SUCROSE IN THE SKATE AND SPINY DOGFISH

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Although comparative biochemical studies of vertebrate insulins indicate structural conservation, significant differences exist between mammalian and elasmobranch molecules (Steiner et al., Amer. Zool. 13:591-604, 1973). Nevertheless, mammalian insulin induces hypoglycemia in some elasmobranchs (Patent, Gen. Comp. Endocrinol. 14: 215-242, 1970). In this study we sought to characterize the temporal changes in blood sugar level (BSL) following a single intraperitoneal injection of bovine insulin (100 iu/kg; Sigma). After injection of hormone or vehicle, blood samples were collected from the caudal aorta at 0.5, 3, 6, 12, 24 and, in the skate only, 48 hours post injection. Blood was centrifuged and plasma frozen until analysis by the glucose oxidase method (Worthington Diagnostics).

The results are shown in Table 1. Despite normal variability in starting values in a natural population each

TABLE I Percent change in blood sugar level (+ SD) in Squalus acanthias and Raja erinacea following a single i.p. injection of bovine insulin (100 iu/kg)

	Time (hours)					
	0.5	3	6	12	24	48
Shark						
Insulin	-6.7 \pm 17.3	-25.0 \pm 7.5	-32.4 \pm 11.5	-52.5 \pm 19.6	-62.2 \pm 33.4	-
Control	-4.9 \pm 16.3	- 1.5 \pm 17.1	13.2 \pm 30.6	13.1 \pm 35.2	15.3 \pm 27.9	-
p value	NS	<.025	<.025	<.01	<.005	
Skate						
Insulin	-8.62 \pm 23.7	-30.8 \pm 23.6	-45.0 \pm 22.4	-64.1 \pm 13.7	-76.9 \pm 5.0	-86.7 \pm 3.1
Control	-7.96 \pm 9.9	- 9.5 \pm 17.8	-21.2 \pm 18.5	1.6 \pm 56.0	-18.0 \pm 7.0	25.0 \pm 24.2
p value	NS	<.10	<.05	<.05	<.001	<.001

* p values are given for comparison between injected and control animals at each time interval tested.

n = 5 in all instances.

animal treated with hormone showed a decline in BSL following hormone injection, whereas control animals did not. The decline began within 30-60 minutes and BSL was significantly ($p < .05$) depressed by six hours post injections in both of these species. Mean absolute values of blood sugar ranged from 89.46 ± 11.40 (S.D.) mg% at 0 time to 31.24 ± 6.71 mg% at 24 hours for S. acanthias and 42.06 ± 4.95 mg% at 0 time to 13.42 ± 2.85 mg% at 48 hours for R. erinacea. Relaxin, a polypeptide hormone with similar tertiary structure to insulin (Schwabe et al., Ann. N.Y. Acad. Sci. 380:6-12, 1982), had no significant effect on BSL in these species in the same experimental design and conditions. Supported by NSF PCM 8104144 to I.P.C.

THE CANALICULAR PERMEABILITY BARRIER IN THE SMALL SKATE (RAJA ERINACEA)

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Among vertebrates bile formation appears at the onset to require movement of water and solute from sinusoidal perfusate across the hepatocellular plates to canalicular lumina. The dynamics of this process remains obscure although considerable evidence (Boyer, J., Review in Physiological Reviews 60:303-326, 1980) suggests that an osmotic pressure gradient produced by active secretion of bile salts into the canaliculi provides an energy source for directed fluid movement. Recent work in this laboratory (Reed, J.S., et al., Amer. J. Physiol. 242:G313, 1982) has added evidence that an hydraulic pressure gradient may also be operative in selachians. These gradients impell movement by routes across the hepatocytes and between them. Transcellular flows traverse a complicated succession of membrane and cytoplasmic barriers whereas paracellular flow is impeded only by the junctional complexes and thus presents a simpler system for study. In rats and dogs, simultaneous measurements of the biliary clearances of molecular weight matched molecular species, one anionic, one neutral (^3H -sucrose (S) and sodium ^{14}C -ferrocyanide (F) or ^3H -methoxy- and ^{14}C -carboxyl-inulin (H,C) yield data (Bradley, S.E. and Herz, R., Amer. J. Physiol., 235:E570, 1978) consistent with a relatively impermeable, negatively charged canalicular junctional barrier that selectively retards passive anionic movement. This approach has been employed in a study of bile formation in excised perfused liver of small skates (Raja erinacea) during changes in bile flow produced spontaneously or by changes in perfusion pressure in order to assess the properties and function of the canalicular membrane.

METHODS--Bile was collected from skate livers, isolated and perfused by 125 ml of Elasmobranch Ringers as previously described by Reed et al (ibid) with the following modifications. Gallbladder drainage was deferred until after isolation of the liver and establishment of adequate portal perfusion with the liver ventral sideup. The gallbladder was incised from the apex to the base. The flap over the common cystic duct on the dorsal side of the gallbladder was identified and a flared 3 cm length of PE205 tubing bent at a right angle was inserted under it. This cannula was secured by 5-0 silk sutured into the gallbladder wall close to the origin of the cystic duct. A preweighed 10 cm length of PE240 tubing was inserted over the PE205 tubing for bile collections. The liver was then inverted to prevent its weight from compressing portal vein, hepatic vein sinuses, and bile ducts.

Bile flow and labelled markers were measured during 3 experimental periods. During the initial period perfusion pressure was established at 10 cm Ringers. Approximately 25 μl of bile was collected in tared PE240 cannulas at intervals that depended on the rate of bile flow (5 to 60 min). After 2-5 collection periods, perfusion pressure was reduced to 5 cm Ringers for 2-5 additional collection periods and then again adjusted to 10 cm Ringers for the final period. In one of 11 experiments this sequence was reversed. The duration of experiments ranged from 3-7 hours.

The 125 ml recirculating fluid contained: sodium ^{14}C -ferrocyanide (25 μCi , NEN, 7135 mCi/m mol) and ^3H sucrose (5 μCi , NEN, 11.4 Ci/m mol) in six experiments; and ^{14}C carboxyl inulin (10 μCi , NEN, 2.1 mCi/g) and ^3H methoxy inulin (10 μCi 140.5 mCi/g) in five experiments. Perfusate samples (50 μl) were taken in duplicate, 1-4 times during the course of each experimental period and counted in 6 ml of scintillation fluid (Aquasol or