These data indicate that within minutes after adding ^{14}C-taurine to the bath, the label begins to appear in the secreted fluid exiting from the open end of the tubule. After 1 1/2 to 2 hours, taurine secretion rates and taurine lumen/bath ratios reached steady-state values for all tubules.

In all tubules, where these studies were attempted, taurine was secreted (Table 2). Only one experiment

Table 2. TAURINE SECRETION BY ISOLATED FLOUNDER RENAL TUBULES

tubules	$\dot{V}_s$ pl/min·mm	bath [TAU] mM	cell [TAU] mM	lumen [TAU] mM	L/B	C/B	C/L	TAU secretion rate fmole/min·mm
n=5	34.95 ±10.6	0.47 ±0.13	63.8 ±8.9	11.8 ±3.8	25	135	5.4	391±122

Values are means ± S.E. $\dot{V}_s$ represents fluid secretion rates. L/B = lumen/bath, C/B = cell/bath, and C/L = cell/lumen taurine concentration ratios.

was terminated early, due to epithelial cell lysis caused by dilution of the bath Ringer's with the temperature-control circulating water. Although the results of this experiment were in the same direction as the others, the magnitude of secretion was much reduced as expected. The results shown in Table 2 are steady-state data averaged from the last sample collections. Taurine secretion rates averaged 391 fmole/min·mm. The lumen/bath taurine concentration ratio for these tubules was 25 which is similar to the tissue/medium ratios for taurine uptake by teased flounder tubules (see King et al., this Bulletin). The taurine concentrations for bath, cell, and lumen were 0.47 mM, 63.8 mM, and 11.8 mM, respectively. Without knowledge of the taurine net electrical charge during secretion and the magnitude of the basolateral and apical membrane potentials, these data suggest an active step of taurine secretion across the basolateral membrane of the epithelium and a passive step from cell to lumen by passive or facilatated diffusion across the apical membrane.

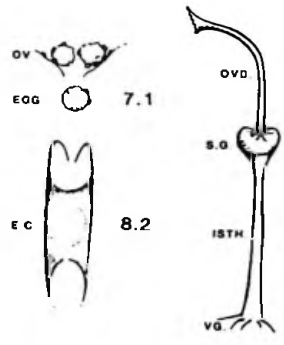
EGG-OVIDUCT SIZE RELATIONSHIPS IN RAJA ERINACEA

Thomas J. Koob, John Laffan and Ian P. Callard, Department of Biology, Boston University and Department of Orthopaedic Surgery, Children's Hospital Medical Center, Boston, Massachusetts

The little skate, Raja erinacea, spawns throughout the year in the North Atlantic and, like all skates, is oviparous (Richards et al., Bull. Bing. Oc. Coll. 18, 5-65, 1963). Characteristically shaped, leathery egg capsules are produced in pairs for periods up to six weeks by females of at least 200 grams. Egg development never progresses beyond the germinal ring stage in utero. Embryogenesis and fetal development are completed external to the maternal environment.

Egg case formation begins before ovulation and is nearly one third complete before the egg enters the shell gland. In passing through the shell gland, the egg is elongated and, with accompanying albumen, packaged in the egg case. The egg case enters the isthmus and is held there until oviposition. Egg case formation requires a minimum of 48 hours and in most cases is followed quickly by oviposition (Richards et al., op. cit.). The oviduct must facilitate passage of a rigid egg case unique in shape and larger in size than the egg (Figure 1). Eggs at ovulation measure 7/1 cm. in circumference while spawned egg cases average 8.2 cm. The reproductive tract could have either a circumference sufficient in size at all times to accommodate the egg case, or an ability to increase its natural circumference during follicle development or at spawning. This report investigates these possibilities.

The female reproductive tract of the little skate (Figure 1) consists of an anterior ostium, an albumen secreting oviduct, the shell gland, an isthmus or uterus and a vaginal constriction through which egg cases



	CIRCUMFERENCE (CM)				WET WEIGHT (MG)	
	NATURAL		MAXIMUM			
	RESTING	SPAWNING	RESTING (M/N)	SPAWNING (M/N)	RESTING	SPAWNING
	1.1±.1	1.3± .1	3.5± .1 (3.3)	4.1± .3 (3.3)	0.32± .04	.44± .02*
	1.6±.1	1.8± .2	7.0± .3 (4.2)	8.2± .9 (4.6)		
					2.19± .09	2.37± .15
	2.3± .3	3.8± .4*	6.1± .4 (2.6)	9.9± .7** (2.6)		
	0.8± .1	2.1± .8	5.1± .4 (6.3)	8.2± .6** (3.8)	1.05± .11	1.67± .24*

Figure 1.--Natural and maximum circumferences of the reproductive tract in resting and spawning female Raja erinacea. Natural circumference: circumference under minimal load. Maximum circumference: circumference at which tissue ruptures. M/N: ratio of maximum circumference to natural circumference. All values are means ± S.E.M. Abbreviations: ov.-ovary; e.c. - egg case; ovd. - oviduct; s.g.-shell gland; isth.- isthmus; va.-vagina. *$p < .05$, **$p < .01$.

gain entrance to the common urogenital sinus at spawning. Natural and maximum circumference of these regions in spawning and resting females were determined using an apparatus identical to that designed by Harkness and Harkness (J. Physiol. 148, 524-547, 1959). Rings of tissue were placed as a belt around two stainless steel rods arranged one above the other, the lower fixed in position and the upper parallel to it in a movable stirrup. Calibrated forces were applied directly to the upper rod to pull the rods apart and exert force on the ring of the tissue. The change in distance between the rods, and thus the circumference were recorded with time. Rings of tissue 1 cm in width were used throughout and all tests were performed in sea water with 1 g/L acetylcholine added. A small constant force exerted on the upper rod was necessary in order to measure natural circumference. Increasing force was then applied in small increments until the tissue ruptured. The circumference at which the tissue ruptured was taken as the maximum circumference. Measurements of 5 sexually mature non-pregnant and 4 pregnant skates are reported in Figure 1. All skates were between 950 and 1000 grams in body weight. It should be noted that two of the five non-pregnant skates contained large oocytes and were close to spawning. Values for non-pregnant skates are therefore artifically weighted towards the pregnant values.

Natural and maximum circumferences of the regions tested in the skate reproductive tract are shown in Figure 1. Regression in size of the reproductive tract is indicated by the smaller natural, as well as maximum, circumferences in resting females. The lower isthmus and vagina undergo marked cyclic variations with maxima occurring at spawning, while minimal chages occur in the oviduct and upper isthmus. The maximum circumferences of all regions tested in resting females are well below that necessary for passage of the rigid egg case. These measurements demonstrate that oviposition could not occur without an alteration in the size or mechanical properties of the reproductive tract. Wet weight determinations (Figure 1) parallel changes in circumference and again indicate that regression in size occurs between spawning periods. These data suggest that it is growth of the reproductive tract which facilitates passage of the egg case at spawning.

Additional evidence supporting this suggestion is derived from measurements of extensibility (change in length/time) and tensile strength (force required to rupture tissue). Neither parameter of any region was significantly different in spawning versus resting skates. Furthermore, the ratio m/n (Figure 1) is virtually identical in all tissues with the exception of the vagina, the tissue which shows the greatest increase in wet weight.

It would appear from the above data that oviposition requires growth of the reproductive tract in order to accommodate the passage of the skate egg case. It is likely that this requisite growth occurs during follicle development and thus might be under hormonal stimulation. The possible hormonal regulation of the events associated with spawning is under investigation. This work was supported by NSF PCM 78-08201 to I.P.C.

28 FINE STRUCTURAL CHANGES ASSOCIATED WITH CELL VOLUME REGULATION IN SLICES OF DOGFISH (SQUALUS ACANTHIUS) RECTAL GLAND

S.K. Masur, J. Goldstein and A. Kleinzeller, Departments of Physiology and Biophysics, Mount Sinai School of Medicine and Department of Biological Sciences, Columbia University, New York, N.Y., Brown University, Providence, R.I. and Department of Physiology, University of Pennsylvania, Philadelphia, Pa.

Evidence for cell swelling induced by high external K^+ medium in incubated shark rectal gland slices has been derived from electrolyte and water content analysis (Kleinzeller et al., Bulletin, MDIBL 20:75, 1980). In other tissues, reversal of such swelling is accompanied by the appearance of cytoplasmic vesicles and vacuoles (Van Rossum and Russo, J. Membrane Biol. 59:191, 1981). We are reporting preliminary observations of similar changes in fine structure of rectal gland slices when swelling is reversed. We have begun morphological and cytochemical studies of these slices in normal medium, in swelling conditions and during swelling reversal.

Slices of rectal gland prepared as described by Goldstein et al. (Bulletin, MDIBL 19:3, 1979) were incubated for 1 to 2 hrs in either A) control medium (6 mM K^+) or B) high K^+ medium (280 mM K^+ replacing equivalently the Na^+ in the standard medium) or C) 1 hr in high K^+ followed by 5, 15 or 60 min in control medium.

For electron microscopy (EM), one or two slices were removed from an experimental sample and fixed in a modified Karnovsky's fixative (buffered, double aldehyde) (Doyle, Bulletin, MDIBL 17:34, 1977) for 1 1/2-3hrs, briefly rinsed in 0.2 M cacodylate, pH 7.4 and postfixed in "aged" 0.05% ruthenium red (RR) in 1% OsO_4 in 0.1M cacodylate for 1 - 3 hrs. Handley and Chan (Histochemistry 71:249, 1981) reported that ageing this mixture for 3 hrs prior to use allows the OsO_4 to oxidize the RR. RR is a morphologically detectable marker for regions which are surface connected at the time of fixation (Chambers, J. Cell Biol 57:874, 1973). Oxidized RR according to Handley and Chan penetrates tight junctions and therefore the aged RR could provide a marker for extracellulary connected compartments on luminal as well as basal-lateral borders. We found no evidence of RR penetration between the cells of the rectal gland slice, but the tissue was well preserved by this mixture. Following fixation the tissue was dehydrated in a graded series of alcohols, propylene oxide, embedded in Epon and 1-2 μm sections were cut on glass knives, stained with toluidine blue and examined by light microscopy (Figures 1,2,3). Thin sections were stained with uranyl and lead and examined in a Philips 201 EM (Figures 4 and 5).

Observations of Incubated rectal gland slices

A. Control medium, 1 - 2 hrs

Parenchymal cells of the incubated rectal gland slices maintain their polarity and their junctional complexes (Eveloff et al, J. Cell Biol. 83:16, 1979) for prolonged periods (Doyle, personal communication) (Fig. 1). Blunt microvilli project into the lumen which is bounded by the junctional complexes between adjacent cells. Immediately below the membrane, the apical region is filled with microfilaments and some microtubules; within this cytoskeletal material are membrane-bound vesicles. Going in a basal direction, we find bounded irregular tubules some of which have ribosomes and are therefore connected with the endoplasmic reticulum. Mitochondria are also found in this region along with the Golgi apparatus. The nucleus sits in the center of the cell. The basal-lateral membranes are plicated with little extracellular space between them and occasional coated vesicles near them. At the basal end of the cell, narrow bands of cytoplasm contain mitochondria and some rough ER vesicles.