GUANYLIN/UROGUANYLIN REGULATION OF DOGFISH SHARK (*SQUALUS ACANTHIAS*) RECTAL GLAND CHLORIDE SECRETION

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The guanylin/uroguanylin family of peptides regulate ion and fluid transport in a wide variety of epithelia, acting through the second messenger, cyclic guanosine monophosphate (cGMP) [Forte and Hamra, *News Physiol. Sci.* 11,17-24, 1996]. Uroguanylin and prouroguanylin were purified from opossum intestine and are found in plasma, indicating that uroguanylin is a circulating peptide. It has been proposed that uroguanylin is an endocrine link between intestine and kidney (Forte et al., *Amer. J. Kid. Dis.* 28,296-304, 1996). Uroguanylin has natriuretic, kaliuretic, and diuretic activities in both opossum in vivo and perfused rat kidney (Freeman et al., *FASEB J.* 8,A552, 1994; Fonteles et al., *Am. J. Physiol.* 275,F191-F197, 1998). Uroguanylin stimulates short-circuit current when applied to the basal side of the mouse colon (L. Forte, unpublished observations).

In the dogfish shark (*Squalus acanthias*) rectal gland, the effects of guanylin or the homologous peptide, heat-stable enterotoxin (STa), have been studied in perfused glands, perfused secretory tubules, monolayer cultures of the secretory epithelial cells, and fresh tissue slices. Both ligands stimulate a robust chloride secretion in the perfused gland (Silva et al., *Bull. MDIBL* 36,53-54, 1997). The nerve blocker, procaine, does not inhibit this stimulatory effect of STa (Silva et al., *Bull. MDIBL* 37,73, 1998), suggesting that guanylin and STa are not acting through nerves.

Interestingly, guanylin does not stimulate chloride secretion in perfused tubules (personal communication, R. Greger). Likewise, STa barely increases (84%) cGMP levels in monolayer cultures whereas the same concentration of atrial natriuretic peptide stimulates cGMP levels 40 fold (Kennedy et al., *Bull. MDIBL* 30,102-103, 1991). In the present study we have confirmed earlier observations that STa does not stimulate chloride secretion in monolayer cultures (Kennedy et al., op. cit.). Also, we report that rat and human guanylin are without effect on monolayer cultures and that STa does not stimulate cGMP generation in fresh rectal gland tissue slices incubated for 40 min at 16 °C with 10^{-6} M STa. It was previously shown that guanylin decreases Na reabsorption in perfused rat kidney (Fonteles et al., op. cit.), but fresh kidney slices do not respond to the ligand with increased cGMP production.

In both shark rectal gland and rat kidney, guanylin-like molecules may act on only a few cells, which in turn produce a second, potent ligand which regulates transport. In the case of the rectal gland, two potential candidates for this second molecule are ATP and a natriuretic peptide-like molecule. First, Silva et al. (*Bull. MDIBL* 37,63-64, 1998) reported that stimulation of

chloride secretion in perfused shark rectal gland by vasoactive intestinal peptide is accompanied by a corresponding, proportional, secretion of ATP into the fluid secretion. We report here that 10^{-4}M MgATP, added to the apical side of monolayer cultures of rectal gland epithelial cells, will stimulate chloride secretion five fold within 10 min. Second, we have partially characterized a natriuretic peptide-like secretagogue from the shark rectal gland (Karnaky et al., *Bull. MDIBL* 31,126-128, 1992). The nature and source of the guanylin-like molecule that acts on the shark rectal gland and the detailed nature of this regulation await further studies.

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