

Experiments were conducted as previously described (Hogben, Proc. Soc. Exp. Biol. Med. 124:890-893, 1967). For the serosal (or interstitial) bathing solutions, the HCO_3^- concentrations were 30, 10, 3 or 1 mEq to yield a pH at room temperature of 7.4, 7.0, 6.5, and 6.0 respectively. All serosal solutions were gassed by 95 percent O_2 and 5 percent CO_2 . A constant $[\text{Na}^+]$ and osmolality were maintained by increasing the $[\text{Cl}^-]$ when $[\text{HCO}_3^-]$ was decreased. As previously reported the mucosal (or luminal) solution was an unbuffered saline gassed by 100 percent O_2 . Mucosal samples were withdrawn and replaced hourly for three periods to determine the H^+ secretion rate. Carbachol (a parasympathomimetic stimulus) was added to all the serosal solutions to a final concentration of 10 $\mu\text{M/L}$.

The result of comparison of paired portions of gastric mucosa from the same fish are given in Table 1. Though not established at the 5 percent level of confidence, H^+ secretion was greater at pH 6.5 relative to pH 7.4 and relative to pH 6.0. The H^+ secretory rates were indistinguishable at pH 6.5 and 7.0

This work was supported by grants NIH AM 05848 and NSF GB 28139.

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THE EFFECT OF DIPHENYLHYDANTOIN AND VASOPRESSIN ON IONS TRANSPORT ACROSS THE FLOUNDER INTESTINE

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Recent reports on the effect of diphenylhydantoin (DPH) on the electrical properties of frog skin and ion fluxes in rat diaphragm have demonstrated that this compound stimulates Na^+ transport across these tissues by increasing the membrane permeability to sodium ion (Watson and Woodbury, J. Pharm. Exp. Therap. 180: 767, 1972; Bihler and Sawh, BBA 249:240, 1971). It has not been determined whether DPH also affects the membrane transport of other electrolytes such as chloride. In the present investigation the intestine of winter flounder, *P. americanus*, was used as an experimental model for the study of Na^+ and Cl^- ion transport and the effects of DPH on these processes.

The small intestine was mounted in a lucite Ussing chamber and bathed with seven ml. of Forster's teleost Ringer solution containing 5.5 mM dextrose as substrate. The details of the experimental procedures have been reported previously (Huang and Chen. Am. J. Physiol. 220:1734, 1971 and J. Pharm. Exp. Therap. 180:777, 1972). A potential difference (PD) ranging from one-four mV was measured across the membrane with the serosa negative to the mucosa. A short circuit current (I_{sc}) was applied to zero the PD. An average I_{sc} of -29 μAmp . was recorded. When DPH was added to the mucosal bathing solution at a concentration of 140 $\mu\text{g/ml}$ or 350 $\mu\text{g/ml}$, both PD and I_{sc} increased slightly (moved toward zero), suggesting either an increase in net Na^+ flux or a decrease in net Cl^- flux or both. Radioactive ^{22}Na and ^{36}Cl were then used to measure the ionic fluxes in neighboring intestinal sections of the same fish.

TABLE 1

The effect of DPH and vasopressin on ion flux

Exp.	No. of Animal	^{22}Na Flux			^{36}Cl Flux		
		J_{ms}	J_{sm}	J_{net}	J_{ms}	J_{sm}	J_{net}
		$\mu\text{Eq.cm}^{-2}\text{hr}^{-1}$			$\mu\text{Eq.cm}^{-2}\text{hr}^{-1}$		
I:	8						
Control		4.20+ 0.27 ⁻	3.19+ 0.35 ⁻	1.01+ 0.32 ⁻	2.98+ 0.21 ⁻	1.98+ 0.19 ⁻	1.14+ 0.16 ⁻
DPH 140 $\mu\text{g}/\text{ml}$, mucosal		5.89+ 0.32 ⁻	4.77+ 0.50 ⁻	1.11+ 0.42 ⁻	3.63+ 0.29 ⁻	2.11+ 0.36 ⁻	1.38+ 0.27 ⁻
II:	4						
Control		4.05+ 0.78 ⁻	2.85+ 0.63 ⁻	1.19+ 0.33 ⁻	3.54+ 0.32 ⁻	1.16+ 0.21 ⁻	2.58+ 0.11 ⁻
DPH 350 $\mu\text{g}/\text{ml}$, mucosal		7.06+ 0.35 ⁻	5.54+ 0.28 ⁻	1.52+ 0.08 ⁻	5.27+ 0.76 ⁻	5.04+ 0.60 ⁻	0.23
--		8.15+ 0.66 ⁻	6.22+ 0.44 ⁻	1.93+ 0.54 ⁻	7.30+ 0.63 ⁻	5.00+ 0.46 ⁻	2.30+ 1.08 ⁻
III:	5						
Control		4.53+ 0.76 ⁻	3.73+ 0.57 ⁻	0.80+ 0.23 ⁻	3.73+ 0.67 ⁻	2.03+ 0.62 ⁻	1.70+ 0.24 ⁻
DPH 350 $\mu\text{g}/\text{ml}$, mucosal		8.06+ 1.17 ⁻	6.20+ 0.84 ⁻	1.85+ 0.54 ⁻	5.03+ 0.56 ⁻	4.00+ 0.41 ⁻	1.03+ 0.40 ⁻
Vasopressin 0.14 I.U./ml, m.+s.		8.01+ 0.88 ⁻	7.21+ 0.70 ⁻	0.80+ 0.61 ⁻	5.45+ 0.50 ⁻	4.74+ 0.88 ⁻	0.71+ 0.59 ⁻

As shown in Table 1 three sets of experiments were performed. In the first set DPH at a concentration of 140 $\mu\text{g}/\text{ml}$ increased Na^+ and Cl^- fluxes in both directions across the intestine with slight but not significant increase in the net absorption (J_{ms}) of both Na^+ and Cl^- . In the second set of experiments DPH at 350 $\mu\text{g}/\text{ml}$ caused a greater increase in Na and Cl fluxes. The increase in I_{sc} produced by DPH can be explained by the simultaneous effects of increasing net Na^+ flux (absorption) and a greater stimulation of Cl^- secretion (J_{sm}) than Cl^- absorption (J_{ms}). In the third set of experiments the addition of vasopressin to the bathing solution after DPH did not additionally increase either the Na or the Cl flux, indicating a lack of synergistic effect between these two compounds.

Based on these preliminary observations it appears that a major effect of DPH on flounder intestine is similar to that demonstrated in other tissues, namely an increase in membrane permeability to Na^+ resulting in a stimulation of Na transport. In addition stimulation of Cl^- secretion is responsible in part for the observed effect on the electrical properties of flounder intestine.

This work was supported by grants from NSF GB-27495 and NIH AM-2217-14.