as operating in a different domain. Kidder determined for the dogfish gastric mucosa that there is a hypopolarization inflection point at +24 mV and a possible hyperpolarization inflection point about −50 mV (Bull. Mt. Desert Island Biol. Lab. 18: 6, 1978).

Though the present study was not designed to advance understanding of anomalous "base" secretion, a few comments are in order. Conditions for evoking "base" secretion by the isolated fundic mucosa have been established by Flemström (Am. J. Physiol. 233: E1, 1977) and by Rehm (Am. J. Physiol., in press, 1981). However the rates reported to date for amphibia have been modest, about 0.4 μEq.cm^{-2}hr^{-1}, and an order of magnitude less than that of the dogfish fundic mucosa. It is not known whether the exceptional rate would be maintained. Clearly study of "base" secretion by the isolated fundic mucosa is intrinsically important, but to use a pejorative adjective it is by no means certain it is "physiological" and can be extrapolated to the mammal in vivo. To date "base" secretion has been accompanied by a deteriorated conductance. Whether sporadically or consistently, it has only been elicited by adverse circumstances, e.g., excessive voltage-clamping, anoxia or inhibition (Sanders SS personal communication). "Stat" titration at pH 4.5 deemed meritorious by some should be regarded as an adverse circumstance for the isolated gastric mucosa. The implicit and at times explicit inference that "base" secretion represents bicarbonate secretion is so far without foundation. It is noteworthy as noted here as well as by others that the open-circuit PD does not change importantly with the onset of base-secretion. Thus the net change of transmucosal ionic currents is at best small. The overall process is one of ion exchange. That the exchange is passive is remote. Though the nature of the exchange is unknown a forced H^+:Na^+ exchange (Transport Mechanisms in Epithelia. Alfred Benzon Symposium V. Editors, H.H. Ussing and N.A. Thorn, Academic Press, New York and London, pp. 236-253, 1973) should not be considered remote

Though it may be regrettable to close on an editorial note, it should be acknowledged that we may be at the end of an era. Much if not all of the world literature on the isolated gastric mucosa of the last 45 years must now be re-evaluated.

ACTIVE POTASSIUM SECRETION BY FLOUNDER URINARY BLADDER: ROLE OF A BASOLATERAL NA-K PUMP AND APICAL POTASSIUM CHANNEL

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We reported previously (Dawson and Andrew, Bull. MDIBL, 19: 46, 1979) that the urinary bladder of the winter flounder exhibited a short circuit current (I_{sc}) which was consistent with the flow of positive charge from serosa to mucosa. Although I_{sc} was abolished by ouabain the current was not directly related to NaCl absorption. Bladders with no detectable I_{sc} nevertheless absorbed NaCl. Furthermore, I_{sc} could be abolished with either lidocaine or the calcium ionophore, A23187, while NaCl absorption was unaffected.

In this paper we present evidence that I_{sc} in the flounder bladder is attributable to active potassium secretion. The results are consistent with a simple model (Fig. 3) in which potassium is actively accumulated by the epithelial cell via a basolateral Na-K ATPase and then leaves the cell, down a favorable electrochemical potential gradient, through a barium-sensitive potassium channel in the apical membrane.

METHODS

Urinary bladders were removed from flounder which had been maintained for several days to a week in flowing sea water and mounted in Ussing chambers (Area = 1.25 cm^2) as previously described (Dawson and Andrew, loc cit).

The bathing solutions, except where otherwise indicated, contained (mM/L) Na: 147.5, Cl: 147.5, K: 2.5, Ca: 1.5, Mg: 1.0, Hepes: 15.0, Glucose: 5.0. Both sides of the isolated bladder were vigorously stirred with air, the pH being approximately 7.5. All experiments were conducted at room temperature which averaged about 20°C. Verapamil (10^{-5}M) was added to the serosal bathing solutions to reduce contraction of bladder smooth muscle (Dawson and Andrew, loc cit).

RESULTS & DISCUSSION

Active Potassium Transport and the Na-K Pump

Several observations suggested that I_{sc} in the flounder urinary bladder could be the result of active potassium secretion. The addition of $BaCl_2$ (1-5 mM) to the mucosal bathing solution abolished I_{sc} in seconds, but the inhibition was readily reversed by two or three replacements of the mucosal bathing solution (*vide infra*). I_{sc} was also reduced to near zero by repeated washing of the serosal side of the tissue with potassium-free Ringers, but was rapidly restored to control values by the addition of 2 to 5 mM potassium to the serosal bathing solution. Several washes of the mucosal surface with Na-free, choline-Ringers also abolished I_{sc}, and the inhibition was rapidly reversed by adding 20 mM NaCl to the mucosal bathing solution.

Transmural fluxes of ^{42}K were measured under short circuit conditions in the presence and in the absence of mucosal barium. The results of a representative experiment are shown in Figure 1 where the serosa to mucosa potassium flux (J^K_{sm}), I_{sc} and tissue conductance (G_T) are plotted for successive half-hour flux periods. Under these circumstances the mucosa to serosa flux of potassium averaged (0.01 ± .005 μEq/cm^{2}hr) so that J^K_{sm} was nearly equal to *net* potassium secretion. Figure 1 shows that net potassium secretion was identical to I_{sc} and that *both* potassium secretion and I_{sc} were abolished by mucosal barium.

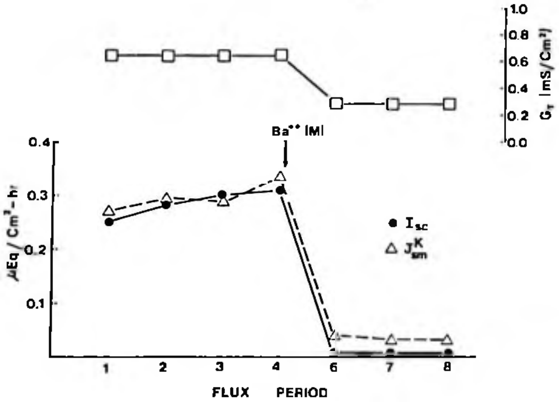

Fig. 1.--Values of short circuit current (I_{sc}), serosa to mucosa potassium flux (J^K_{sm}) and tissue conductance (G_T) plotted as a function of flux period. Note that values for period 5 have been omitted due to non-steady tracer fluxes immediately following inhibition of I_{sc} by barium.

If the basolateral membrane of the epithelial cells contains an electrogenic sodium-potassium pump, then under appropriate conditions it should be possible to detect a *positive* current from *mucosa* to *serosa*. To test for the presence of such a cation exchange pump in the flounder bladder we conducted experiments in which the cation-selectivity of the apical membrane was altered by treatment with the polyene antibiotic, amphotericin B. Polyene antibiotics can form pores in natural and artifical membranes which are moderately cation selective but which do not discriminate appreciably between sodium and potassium. For these experiments both sides of the bladder were bathed by a Ringer's solution which was identical to that described above except that all of the NaCl was replaced by sodium benzene sulfonate, in order to prevent swelling due to net salt entry into the epithelial cells. Figure 2 shows the results of a representative experiment. In the presence of symmetric, Na-benzene sulfonate Ringers I_{sc} was near zero. The addition of amphotericin-B (10 μM) to the mucosal bathing solution resulted in the rapid development of an I_{sc} consistent with net positive charge movement from mucosa to serosa, i.e., opposite to that observed in the normal state. This current was promptly increased by the addition of 3 mM potassium to the serosal bath. The direction of this change was opposite to that expected for a potassium diffusion current but was consistent with stimulation of a coupled, electrogenic Na-K exchange. The polyene-induced I_{sc} was abolished by serosal ouabain (10^{-4}M). It is also of interest that in the presence of ouabain the introduction of substantial K gradients, by the addition of 20-30 mM potassium methyl sulfate to the mucosal bath, did not generate significant diffusional currents across the tissue, suggesting that the basolateral membrane has a relatively low potassium conductance.

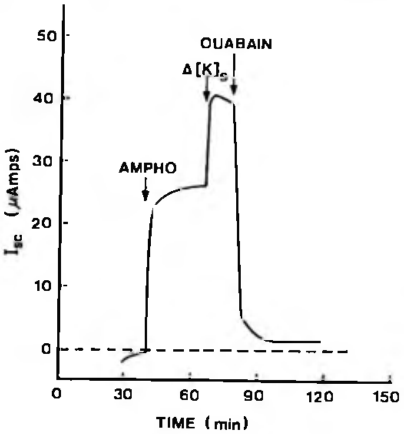

Fig. 2.--Effect of amphotericin-B on I_{sc} in the presence of symmetric Na-benzene sulfonate Ringers. Note that I_{sc} has the orientation of positive change flow from mucosa to serosa and that I_{sc} is increased by serosal potassium and abolished by serosal ouabain.

APICAL POTASSIUM CHANNEL

The reversible inhibition of I_{sc} by mucosal barium strongly suggests that the apical membrane of the flounder bladder contains a barium-sensitive potassium channel similar to those demonstrated in the basolateral membrane of frog skin (Nagel, Biochim. Biophys. Acta 552: 356, 1979) and turtle colon (Kirk, et al., Nature 287: 237, 1980). We therefore examined the effect of mucosal potassium concentration on I_{sc}. Figure 3 shows a representative experiment in which the isolated bladder was initially bathed on both sides by identical Ringer's solutions containing 2.5 mM potassium. As shown in the figure the addition of 1 mM barium to the mucosal bathing solution reduced I_{sc} to near zero and resulted in a significant decrease in G_T. Subsequent additions of 1 mM barium produced a small additional inhibition. The decreases in I_{sc} and G_T were readily reversed by washing the mucosal surface of the bladder. After I_{sc} had returned to its steady-state, pre-barium value the potassium concentration in the mucosal bathing solution was progressively raised by the addition of small volumes of concentrated KCl. Note that each addition resulted in a decrease in I_{sc} and an increase in G_T. Although G_T was increased by more than 2.5 fold at 32.5 mM mucosal potassium, the subsequent addition of barium to the mucosal bath reduced G_T to less than its control value. We analyzed the data according to a simple equivalent circuit by computing the cellular resistance, R_{cell}, and the apparent emf, E_T, at each mucosal potassium concentration. Plots of the data from individual ex-

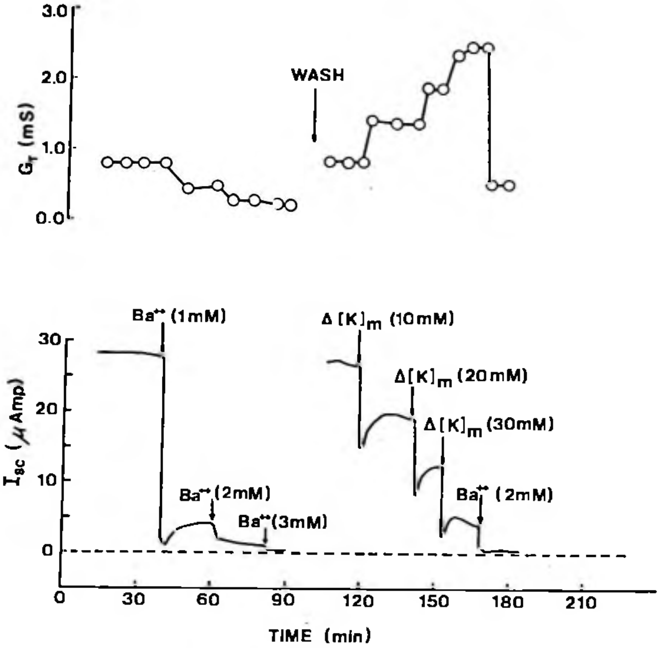

Fig. 3.--Reversible inhibition of I_{sc} by mucosal barium and effect on I_{sc} and G_T of increasing mucosal potassium concentration.

periments indicate that R_{cell} is linearly related to the reciprocal of $[K]_m$ whereas E_T decreases linearly with the log of $[K]_m$. The electrical behavior of the flounder bladder is thus at least qualitatively consistent with the presence in the apical membrane of a cation channel which is highly selective for potassium and is blocked by barium. Furthermore, this behavior is consistent with the notion that the sole driving force for potassium exit from the cell is the electrochemical potential gradient of this ion across the apical membrane. This research was supported by a grant from the NIH-NIAMDD (AM18776) and D.C.D. was the recipient of a Research Career Development Award from NIH-NIAMDD (AM00702).

POTASSIUM TRANSPORT BY THE INTESTINE OF THE WINTER FLOUNDER, SPEUDOPLEURONECTES AMERICANUS: EVIDENCE FOR KCl CO-TRANSPORT

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Prior studies of electrolyte transport across flounder intestine under short-circuit conditions revealed active transport of Na and Cl from mucosa to serosa. The short-circuit current could be attributed to Na and Cl absorption, according to the relation, $I_{sc} = J^{Na}_{net} - J^{Cl}_{net}$. However, the high Na permeability of this tissue produces uncertainty with regard to the absolute magnitude of the net Na flux and evaluation of the relation between I_{sc} and net ion movements. In the present study, the possibility that flounder intestine carries out net K transport was examined. The finding of net K secretion led to an examination of the permeability properties of flounder intestinal cells to K and Cl.

Methods

Transepithelial ion fluxes were determined under short-circuit conditions as previously described (Field, et al., J. Memb. Biol. 41: 265, 1978). Care was taken to assure that the unidirectional fluxes of K and Rb were measured