Effect of Somatostatin on Ion Transport in the Rat Colon

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ABSTRACT The effect of somatostatin (SRIF) on ion transport was determined in the rat colon in vitro. SRIF produced a sustained decrease in the short circuit current (I_{sc}) (-0.8 ± 0.1 μeq/h·cm^2) and increased net Cl absorption (0.9 ± 0.3 μeq/h·cm^2). The threshold effect of SRIF on I_{sc} was observed at 6 nM. 10μM serotonin decreased net Na absorption (-2.6 ± 0.4 μeq/h·cm^2), net Cl absorption (-3.6 ± 0.5 μeq/h·cm^2) and increased I_{sc} (0.7 ± 0.1 μeq/h·cm^2); these changes were totally blocked by 0.1 μM SRIF. SRIF completely blocked net Cl secretion induced by 10 mM theophylline (-2.5 ± 0.7 to $+4.1\pm2.0$ μeq/h·cm^2) and partially blocked theophylline-induced inhibition of net Na absorption (0.7 ± 0.5 to 2.1 ± 0.4 μeq/h·cm^2). SRIF also blocked prostaglandin E_1 (PGE_1) induced increase in potential difference and I_{sc} ($P < 0.001$). Mucosal cyclic AMP levels were increased by theophylline and PGE_1 but not by serotonin. SRIF had no effect on basal or theophylline- and PGE_1-stimulated cyclic AMP levels. These results indicate that SRIF blocks both cyclic AMP and noncyclic AMP mediated changes in ion secretion and suggest that SRIF is acting at a step in the secretory process beyond the formation of cyclic AMP.

INTRODUCTION

Considerable evidence has accumulated in recent years suggesting that somatostatin (SRIF)[1] affects fluid and electrolyte transport in the intestine. Carter et al. have reported that SRIF blocked vasoactive-intestinal polypeptide-induced inhibition of water absorption in the rat colon in vitro (1). We have found that SRIF blocked water secretion induced by prostaglandin E_1 and theophylline in the rat jejunum in vivo without affecting cyclic AMP levels (2). SRIF infusion has been reported to inhibit watery diarrhea in two patients with the carcinoid syndrome (3, 4). We have also shown that SRIF stimulates Na and Cl absorption in the rabbit ileum in vitro (5). In this study, we determined the effect of SRIF on colonic ion transport in vitro and the ability of SRIF to block colonic secretion stimulated by cyclic AMP- and noncyclic AMP-mediated agents.

METHODS

Ion transport studies. Nonfasting male Sprague-Dawley rats weighing 250–300 g were killed with ether and the colon removed rapidly to cold Ringer's solution. After rinsing, serosal and muscle layers were removed as described (6). Segments of mucosa were mounted in lucite chambers with a surface area of 1.13 cm^2 and attached to mucosal and serosal reservoirs containing identical volumes of Ringer's solution, pH 7.4, at 37°C. The composition of the Ringer's solution used in these experiments was the following (mM): Na, 140; Cl, 119.8; HCO_3, 25; Ca, 1.2; Mg, 1.2; K, 5.2; HPO_4, 2.4; H_2PO_4, 0.4. 5 mM glutamine was added to all Ringer's solutions used in these experiments. All solutions were continuously oxygenated with 95% O_2 – 5% CO_2.

The potential difference (PD) across the mucosa was measured by calomel half cells in 3 M KCL and monitored with a potentiometer. The spontaneous tissue PD was nullified by an automatic voltage clamp with Ag:$AgCl_2$ electrodes throughout the experiments, except for 5–10 s every 5 min when the spontaneous PD was recorded. Tissue conductance was calculated from the PD and the short-circuit current (I_{sc}) according to Ohm's law. Na fluxes were determined with ^{22}Na and ^{24}Na oppositely directed across the same piece of tissue and Cl fluxes were determined with ^{36}Cl oppositely directed on adjacent tissue pairs. In some experiments Na and Cl fluxes were measured using ^{22}Na and ^{36}Cl oppositely directed on adjacent pieces of tissues. Tissue pairs were discarded if conductance differed by more than 30%. Isotopes were added immediately after the tissue was mounted. 15 min later the first 15-min flux period began after

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[1] *Abbreviations used in this paper*: I_{sc}, short circuit current; PD, electrical potential difference; PGE_1, prostaglandin E_1; SRIF, somatostatin.

which time SRIF, theophylline or serotonin was added to the seosal reservoir and 10 min later secretagogue or SRIF or no addition was made to the serosal reservoir. 10 min after this second addition, the second 15-min flux period was begun (Figs. 4 and 5). All agents were added to the serosal reservoir.

Preparation of dispersed colonic mucosal cells. Isolated mucosal cells from rat colon were prepared using minor modifications of the procedure of Weiser et al. (7). Rats were killed by ether anesthesia and the entire colon was resected. The lumen of the resected colon was washed with an iced (4°C) solution containing 140 mM NaCl, 16 mM NaH_2PO_4 (pH 7.3), and 1 mM dithiothreitol until the effluent was clear. The distal end of the resected colon was clamped with a hemostat. The segment was filled with ~10 ml of a solution containing 96 mM NaCl, 1.5 mM KCl, 27 mM Na citrate, and 5.6 mM KH_2PO_4 (pH 7.3) and clamped with a hemostat. The intestine was incubated in 250 ml of solution, identical to that used to fill the lumen, at 37°C for 20 min and was gassed continuously with 100% O_2. At the end of the incubation the luminal contents of the segment were drained and discarded. The segment was filled with ~10 ml of a solution containing 140 mM NaCl, 16 mM NaH_2PO_4 (pH 7.3), 1.5 mM EDTA, and 0.5 mM dithiothreitol and then incubated in 250 ml of the same solution at 37°C for 20 min. The luminal contents of the intestine were drained, gassed with 100% O_2 and centrifuged at 500 *g* for 3 min. The supernate was discarded and the cells were washed twice in iced solution composed of 140 mM NaCl, 16 mM NaH_2PO_4 (pH 7.3), and 1.5 mM EDTA and once with the same solution without EDTA.

The mucosal cells were diluted to the desired concentration ($5-10 \times 10^{-6}$ cells/ml) in solution containing 110 mM NaCl, 4.74 mM KCl, 1.19 mM KH_2PO_4, 26 mM $NaHCO_3$, 1% bovine serum albumin, 0.5 mM $CaCl_2$, 1.2 mM $MgCl_2$, pH 7.4. The concentration of the cell suspension was determined by counting a properly diluted sample in a standard hemocytometer. The incubation solution was equilibrated with 100% O_2.

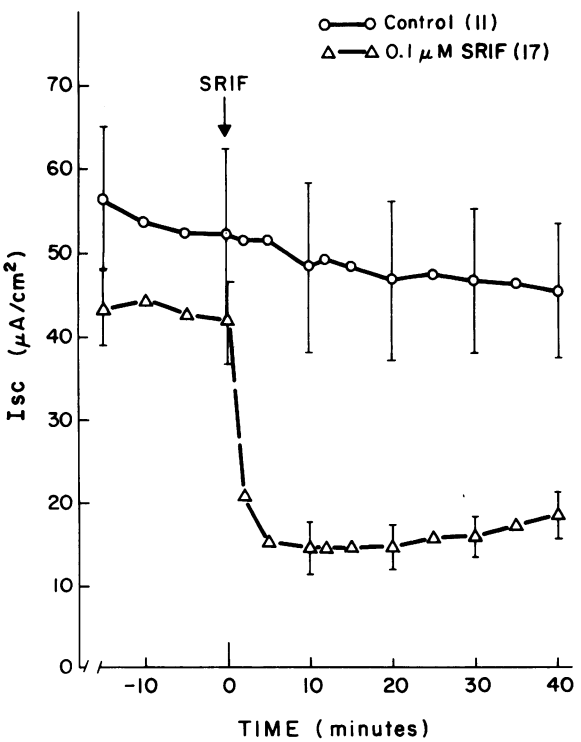

FIGURE 1 The effect of SRIF on the I_{sc}. 0.1 μM SRIF was added 30 min after mounting the tissue. All values are expressed as mean±SE. The number of tissues is indicated in parentheses. All values after addition of SRIF are significantly different from controls ($P < 0.05$).

Cyclic AMP measurement. Cyclic AMP was determined by radioimmunoassay using the technique of Harper and Brooker (8). After the isolated colonic cells were incubated at 37°C with the agents shown in Table II, 1 ml of absolute ethanol was added to 500 μl of cell suspension. The samples were kept at 4°C for 15 min and then centrifuged at 1,500 *g* for 20 min. Duplicate 25- or 50-μl samples of supernate were added to 100 μl of 50 mM sodium acetate (pH 6.2). The samples were acetylated at ambient temperature by adding 5 μl of a freshly prepared solution composed of two parts triethylamine and one part acetic anhydride. 100 μl of anticyclic AMP antiserum plus 100 μl of ^{125}I-cyclic AMP were added and the samples were incubated at 4°C for 18 h. Antibody-bound ^{125}I-cyclic AMP was measured by adding 2.0 ml of 50 mM sodium acetate (pH 6.2), centrifuging the samples at 1,500 *g* for 20 min, aspirating the supernate, and measuring the radioactivity in the precipitate. For each assay a standard curve was constructed by adding known amounts of cyclic AMP to standard incubation solution and processing these standards in the same way as the experimental samples.

SRIF was obtained from Bachem, Inc., Torrance, Calif. and Ciba-Geigy Corp., Summit, N. J.; ^{22}Na, ^{24}Na, ^{36}Cl, ^{125}I-cyclic AMP, and anticyclic AMP antisera from New England Nuclear, Boston, Mass.; prostaglandin E_1 from Upjohn Co., Kalamazoo, Mich.; theophylline and serotonin creatinine sulfate complex from Sigma Chemical Company, St. Louis, Mo.

The results are expressed as mean±S.E. Statistical analysis was performed using Student's *t* test (9).

RESULTS

Effect of SRIF

The addition of 0.1 μM SRIF to the serosal side of the rat colon, resulted in an almost immediate decrease in the PD and I_{sc} (Fig. 1). The maximal decrease occurred at ~5 min (-24 ± 3 μA/cm², $P < 0.001$). Thereafter, the PD and I_{sc} gradually increased but remained significantly lower than controls over the next 40 min. When Na in the bathing solution was replaced with equimolar amounts of choline, 0.1 μM SRIF ($n = 10$) and 1 μM SRIF ($n = 6$) had no effect on the I_{sc} (data not shown).

Fig. 2 illustrates the effect of increasing concentrations of SRIF in the serosal solution on the I_{sc}. Results are expressed as the ratio of the decrease in I_{sc} compared to the I_{sc} just before the addition of SRIF. The decrease in I_{sc} in response to SRIF varies directly with the initial I_{sc} before addition of SRIF. Thus, the lower the initial I_{sc} the smaller the response to SRIF. A significant decrease in the I_{sc} was first observed with 6 nM and the maximal decrease in the I_{sc} was observed with 60 nM. Therefore, 0.1 μM SRIF was used in all subsequent studies.

The effect of 0.1 μM SRIF on Na and Cl transport is

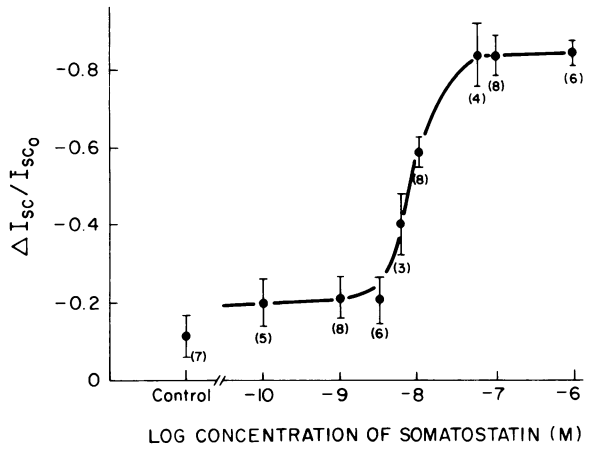

FIGURE 2 The effect of increasing concentrations of SRIF on the I_{sc} in the rat colon. SRIF was added to the serosal reservoir of the Ussing chamber 30 min after mounting the tissue. The maximal decrease in I_{sc} that occurred in the 10-min interval after the addition of SRIF was used for calculation. Results are expressed as the ratio of the decrease in I_{sc} (ΔI_{sc}) to the I_{sc} just before addition of SRIF (I_{sc_0}). The number of tissues is indicated in parentheses.

summarized in Table I. Although SRIF decreased the I_{sc} by 0.8 μeq/h·cm² ($P < 0.001$), there was no significant change in net or unidirectional Na and Cl flux or the residual flux when compared to pretreatment values (period I). However, in SRIF tissues net Cl absorption increased (0.9±0.6 μeq/h·cm²) and in control tissues (no addition between periods I and II) net Cl absorption decreased (−0.9±0.3 μeq/h·cm²); the difference in responses being significant ($P < 0.005$). Conductance was not affected by SRIF.

Effect of SRIF on secretagogue-induced changes in ion transport

Serotonin. 10 μM serotonin increased the PD and I_{sc}, which reached the peak within 2 min (89±7 μA/cm², $P < 0.001$). Thereafter, the PD and I_{sc} gradually declined (Fig. 3). Fig. 4 illustrates the effect of increasing concentrations of serotonin on the increase in the I_{sc}. An initial response was seen at 0.5 μM and the maximal effect was seen at 10 μM. Therefore 10 μM serotonin was used in all subsequent studies. Serotonin decreased net Na and Cl absorption (Table I) when compared to pretreatment values (period I) and when compared to control tissues.[2] Analysis of the unidirectional Na and Cl fluxes reveals that the decrease in net Na and Cl absorption was due primarily to a decrease in mucosal to serosal flux. Tissue conductance increased by 13%.

[2] The decrease in net Cl absorption in serotonin-treated tissues (−3.6±0.5 μeq/h·cm²) was greater than the decrease in control tissues (−0.9±0.3 μg/h·cm², $P < 0.001$).

Addition of serotonin to SRIF-pretreated tissue produced an increase in the I_{sc} (26±10 μA/cm²), which was significantly smaller than tissues treated with serotonin alone (89±7 μA/cm² $P < 0.001$, Fig. 3A). 0.1 μM SRIF added to serotonin-treated tissues decreased the I_{sc} (−66±8 μA/cm²) significantly more than SRIF alone (−34±6 μA/cm², $P < 0.05$, Fig. 3B). Addition of 0.1 μM SRIF to serotonin-treated tissues completely blocked the effect of serotonin on net and unidirectional Na and Cl fluxes (Table I).

Theophylline. Addition of 10 mM theophylline resulted in an immediate and sustained increase in the I_{sc} (130±11 μA/cm², $P < 0.001$, Fig. 5). 10 mM theophylline abolished net Na absorption and net Cl absorption was reversed to net secretion, primarily because of a decrease in mucosal to serosal flux of Na and Cl (Table I). Tissue conductance increased 22%.

Addition of 10 mM theophylline to SRIF-treated tissues resulted in only a 55% increase in the I_{sc} compared to the addition of theophylline alone (72±13 vs. 130±11 μA/cm², $P < 0.02$, Fig. 5A). Addition of 0.1 μM SRIF to theophylline-treated tissue resulted in a much larger decrease in the I_{sc} (−128±11 μA/cm², Fig. 5B) than seen with SRIF alone (−20±4 μA/cm², $P < 0.001$). 0.1 μM SRIF added to theophylline-treated tissues partially blocked the decrease in net Na absorption and reversed theophylline induced Cl secretion to net absorption (Table I). There was no effect of SRIF on theophylline-induced increase in tissue conductance.

Prostaglandin E_1. 0.1 mM prostaglandin E_1 (PGE_1) increased the I_{sc} 137±23 μA/cm² (Fig. 6). This concentration of PGE_1 produced the maximal increase in I_{sc}. Addition of 0.1 mM PGE_1 to SRIF-treated tissues increased the I_{sc} only 42% of that seen with PGE_1 alone (57±10, $P < 0.025$, Fig 6A). Addition of 0.1 μM SRIF to PGE_1-treated tissues resulted in a decrease in the I_{sc} (−61±12 μA/cm²) that was larger than the effect of SRIF alone (−29±4 μA/cm², $P < 0.05$).

Effect of SRIF on cyclic AMP levels

The effect of SRIF and secretagogues on cyclic AMP content in isolated colonic epithelial cells is shown in Table II. Theophylline and PGE_1 increased cyclic AMP levels but serotonin had no effect. SRIF did not affect basal cyclic levels, nor inhibit the increase in cyclic AMP induced by theophylline and PGE_1.

DISCUSSION

The purpose of this study was to determine the effect of SRIF on colonic ion transport and to determine the ability of SRIF to block the effect of secretagogues. SRIF decreased the I_{sc} in the rat colon, indicat-

TABLE I
Effect of SRIF on Na And Cl Transport

	Na fluxes			Cl fluxes				
	$J_{m\to s}$	$J_{s\to m}$	J_{net}	$J_{m\to s}$	$J_{s\to m}$	J_{net}	I_{sc}	G
				$\mu eq/h \cdot cm^2$				$mmhos/cm^2$
SRIF								
Period I	13.5±1.0	8.1±0.5	5.4±0.7	18.3±0.8	14.2±1.0	4.1±1.3	1.5±0.1	12.0±0.5
Period II	12.5±0.7	7.3±0.4 (n = 21)	5.1±0.6	17.2±0.7	12.2±0.7 (n = 10)	5.0±1.1	0.7±0.1*	11.3±0.7
Serotonin								
Period I	13.0±0.6	7.5±0.6	5.5±0.4	19.0±0.5	13.0±0.5	6.0±0.7	1.6±0.1	12.5±0.6
Period II	11.3±0.6*	8.5±0.4* (n = 21)	2.8±0.5*	16.6±0.5*	13.9±0.5* (n = 12)	2.7±0.6*	2.4±0.1*	14.1±0.7*
Serotonin/SRIF								
Period I	13.9±1.2	8.9±1.1	5.0±0.7	18.1±0.5	13.1±0.6	5.0±0.7	1.3±0.1	11.7±0.8
Period II	13.8±1.3‡	9.7±1.3 (n = 16)	4.1±0.5‡	18.4±1.1	13.5±1.0 (n = 7)	4.9±1.6‡	0.6±0.1‡*	13.0±1.0
Theophylline								
Period I	14.1±1.1	7.2±0.6	6.9±0.7	19.5±0.7	14.1±0.7	5.4±0.6	1.6±0.1	12.5±0.7
Period II	8.7±0.6*	7.9±0.6 (n = 13)	0.7±0.5*	12.6±0.9*	15.1±0.4 (n = 6)	−2.5±0.7*	5.0±0.2*	15.2±0.8*
Theophylline/SRIF								
Period I	14.0±0.7	7.7±0.5	6.3±0.6	19.1±0.9	13.6±0.8	5.5±1.4	1.4±0.1	12.5±0.5
Period II	10.0±0.5*	8.0±0.4 (n = 25)	2.1±0.4‡*	17.0±1.8	12.9±1.2 (n = 7)	4.1±2.0‡	2.3±0.2‡*	15.2±0.9*
Control								
Period I	14.2±1.1	8.8±0.6	5.4±0.9	16.5±0.6	12.1±0.4	4.4±0.7	1.7±0.2	14.5±0.9
Period II	13.9±1.1	9.1±0.8 (n = 12)	4.9±0.8	15.6±0.5	12.2±0.5 (n = 14)	3.5±0.6	1.5±0.1	14.9±1.0

Abbreviations used in this table: $J_{m\to s}$, mucosal to serosal flux; $J_{s\to m}$, serosal to mucosal flux; J_{net}, net flux; G, conductance. Period I is the 15-min flux period immediately before addition of the agents in the first column. Period II is a 15-min flux period beginning 20 min after the addition of 0.1 μM SRIF, 10 μM serotonin, or 10 mM theophylline alone. In the serotonin/SRIF experiment, 10 μM serotonin was added at the end of period I, 0.1 μM SRIF was added 10 min later and after a 10-min equilibration, period II began as shown in Fig. 3B. In the theophylline/SRIF experiment, either 10 mM theophylline or 0.1 μM SRIF was added after period I and 10 min later the other substance added as shown in Fig. 5. Since the results were similar, they were combined for presentation. n = number of tissues used for the Na fluxes and the number of tissue pairs for the Cl fluxes. Results are expressed as mean±SE.

* $P < 0.05$, period I compared to period II.

‡ $P < 0.05$, period II theophylline or serotonin alone compared to Period II, theophylline or serotonin with SRIF.

ing an effect on ion transport. The decrease in I_{sc} induced by SRIF was seen at concentrations as low as 6 nM. Although SRIF did not significantly change ion flux from pretreatment values in the same tissues, it increased net Cl absorption compared to control tissues (Table I). Thus the decrease in I_{sc} induced by SRIF is probably due to increased Cl absorption; this probability being strengthened by the fact that the decrease in the I_{sc} (-0.8 $\mu eq/h \cdot cm^2$) was virtually identical to the increase in net Cl absorption (0.9 $\mu eq/h \cdot cm^2$). The relatively small effect of SRIF on ion transport in the rat colon is in agreement with the studies of Carter et al., who found that 10 μM SRIF had no effect on water movement in the rat colon, using the everted gut sac preparation (1). SRIF had no effect on I_{sc} in Na-free media, suggesting that Na is required for the SRIF effect on Cl absorption.

We have shown that SRIF significantly alters the effect of serotonin, PGE_1, and theophylline on ion transport. SRIF inhibited the increase in I_{sc} induced by these agents and when SRIF was added after the secretagogue, the decrease in I_{sc} was much greater than with SRIF alone, indicating that SRIF can reverse the effect of these agents on I_{sc}, rather than just having an additive effect. SRIF completely or partially blocked the effect of serotonin and theophylline on Na and Cl transport. The partial blockade of theophylline effect on Na transport is compatable with the results of Carter et al. (1) who found that SRIF only partially blocked the effect of theophylline on water move-

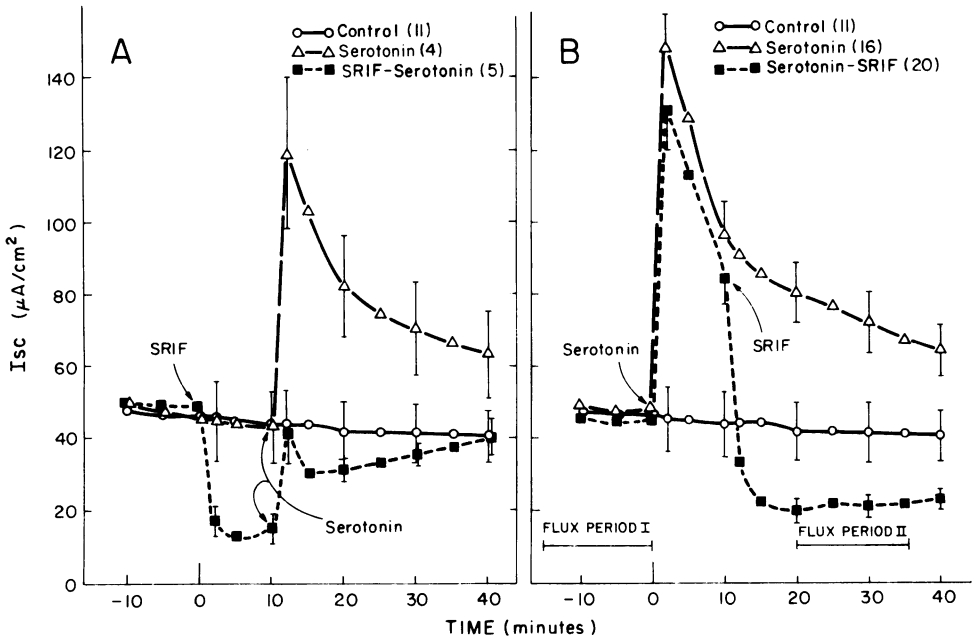

FIGURE 3 The interaction of SRIF and serotonin on the I_{sc}. Circles represent control tissues; tissues treated with 10 μM serotonin are shown as triangles and tissues treated with both 0.1 μM SRIF and 10 μM serotonin are shown as squares. (A) SRIF was added 10 min before serotonin. (B) Serotonin was added 10 min before SRIF. The number of tissues is indicated in parentheses.

ment in the rat colon. Ion flux studies were not performed with PGE_1, however, this agent has been previously shown to stimulate fluid and electrolyte secretion in jejunum (2), ileum (10), and colon.[3] SRIF had no effect on basal cyclic AMP levels nor the increase in cyclic AMP induced by theophylline or PGE_1. Thus SRIF can block the effect of cyclic AMP-mediated (PGE_1 and theophylline) and noncyclic AMP-mediated (serotonin) agents on ion transport without affecting cyclic AMP levels. This suggests that SRIF is working at some distal step in the secretory process beyond cyclic AMP formation.

It is possible that intracellular calcium concentration is the final common mediator for both cyclic AMP and noncyclic AMP-mediated processes. Frizzell (11) found that cyclic AMP increased calcium efflux from the rabbit colon, suggesting that cyclic AMP is mobilizing intracellular stores of calcium. Donowitz has found that the effect of serotonin on ion transport can be blocked by verapamil, a calcium channel blocker or by removing calcium from the bathing solution (12). Ilundain and Naftalin (13) have demonstrated that stelazine will block both cyclic- and noncyclic-mediated secretion in the rabbit ileum and that stelazine binds to calcium dependent regulatory protein (calmodulin). Thus, one could speculate that cyclic AMP, by mobilizing intracellular stores of calcium, and serotonin, by increasing calcium influx, increase cytosolic calcium, which results in activation of calmodulin. Calmodulin then activates some process resulting in net electrolyte secretion or inhibition of

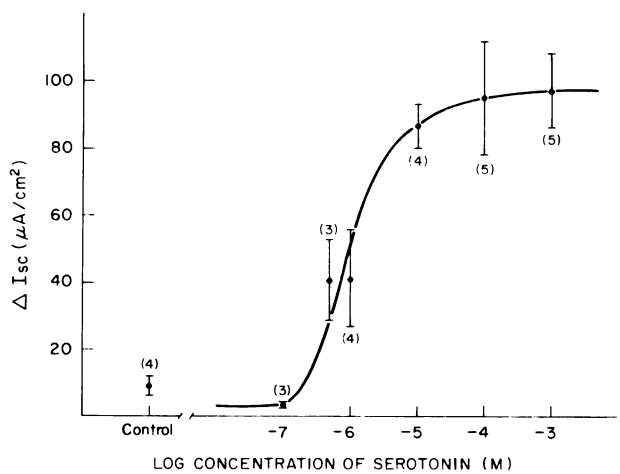

FIGURE 4 The effect of increasing concentration of serotonin on the I_{sc}. Serotonin was added to the serosal reservoir of the Ussing chamber in the concentration shown 30 min after mounting the tissue. The maximal increase in I_{sc} that occurred in the first 10-min interval after addition of serotonin was used for calculation. The number of tissues is indicated in the parentheses.

[3] Racusen, L. C., and H. J. Binder. Effect of prostaglandin on ion transport across isolated colonic mucosa of the rat. Submitted for publication.

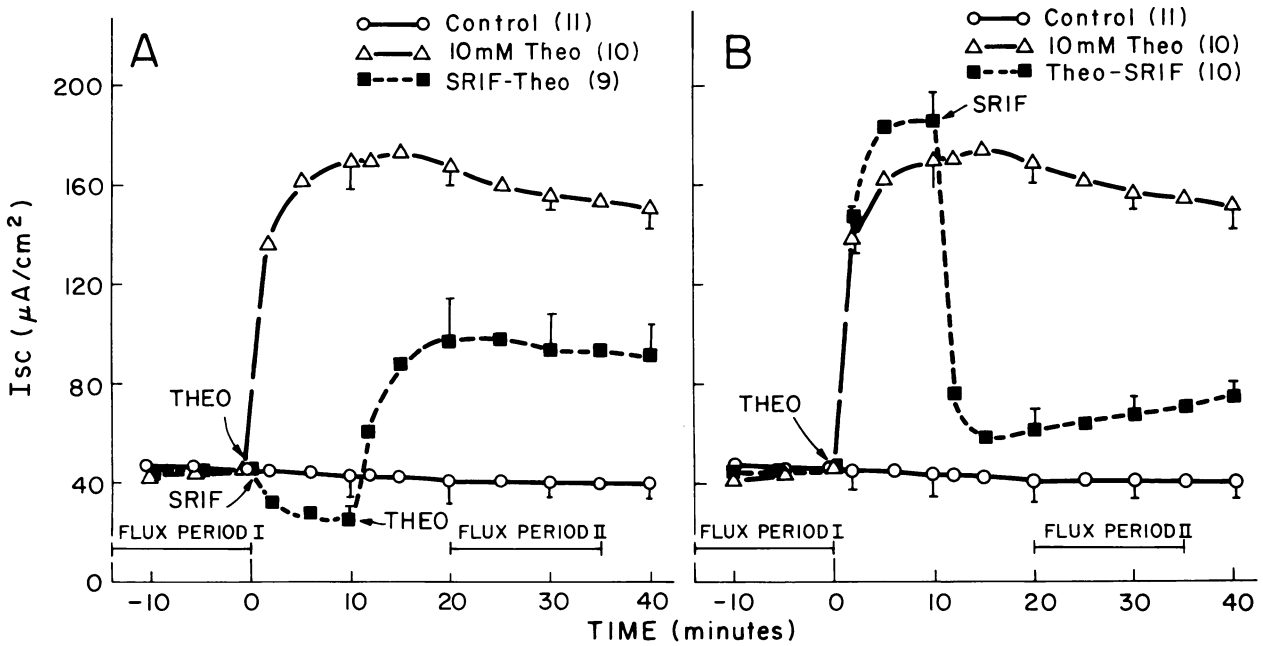

FIGURE 5 The interaction of SRIF and theophylline on the I_{sc}. Circles represent control tissues; tissues treated with 10 mM theophylline are shown as triangles and tissues treated with 10 mM theophylline and 0.1 μM SRIF are shown as squares. (A) SRIF was added 10 min before theophylline. (B) Theophylline was added 10 min before addition of SRIF. The number of tissues is shown in parentheses.

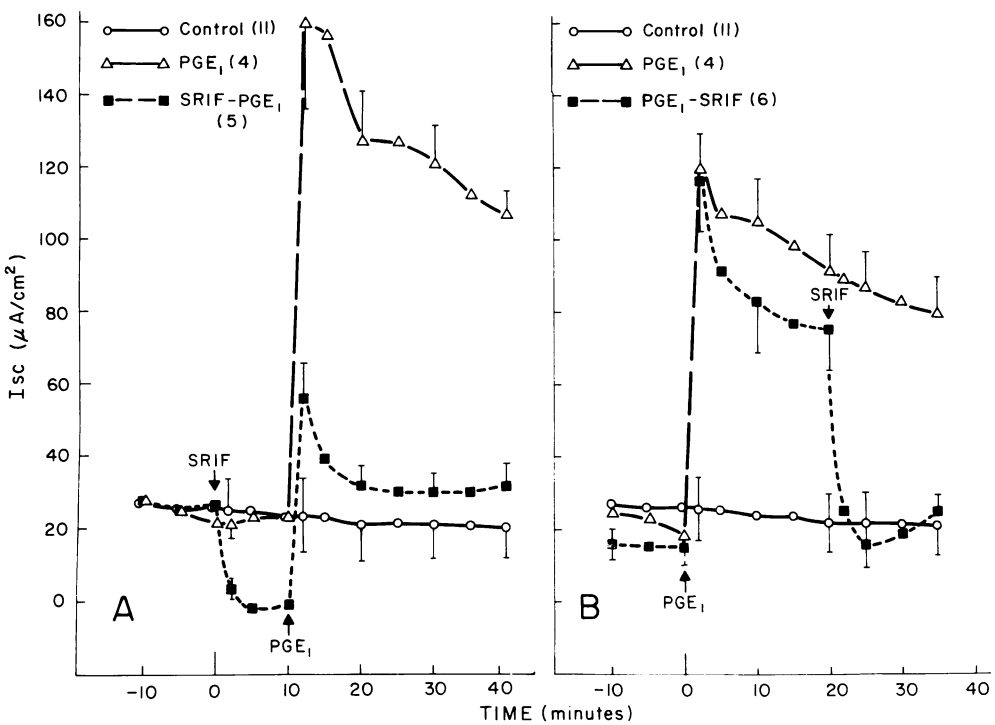

FIGURE 6 The interaction of PGE_1 and SRIF on the I_{sc}. Circles represent control tissues, tissues treated with 0.1 mM PGE_1, are shown as triangles and tissues treated with both 0.1 mM PGE_1 and 0.1 μM SRIF are shown as squares. (A) SRIF was added before PGE_1. (B) PGE_1 was added before SRIF. The number of tissues is shown in parentheses.

TABLE II

Effect of SRIF and Secretagogues on Cyclic AMP Content in Isolated Colonic Epithelial Cells

Experiment	min . . .	Cyclic AMP 0	2	5	10	20
				pmol/10⁶ cells		
Control		0.40±0.08	0.32±0.08	0.28±0.08*	0.28±0.08	0.25±0.06*
Theophylline		0.41±0.07	0.63±0.08‡	0.84±0.12‡	0.96±0.10§	0.95±0.06§
PGE_1		0.41±0.06	0.58±0.08‖	0.62±0.06§	0.58±0.07*	0.50±0.08
Serotonin		0.47±0.05	0.49±0.05	0.48±0.07	0.43±0.06	0.38±0.05¶
SRIF		0.36±0.05	0.33±0.08	0.35±0.09	0.25±0.07*	0.23±0.05¶
Theophylline plus SRIF		0.27±0.06	0.52±0.13*	0.62±0.17*	0.75±0.18¶	0.67±0.10§
PGE_1 plus SRIF		0.31±0.06	0.43±0.07	0.50±0.09¶	0.54±0.12*	0.49±0.12
Serotonin plus SRIF		0.43±0.07	0.36±0.08	0.34±0.05	0.36±0.07	0.30±0.06¶

0.5-ml samples of isolated rat colonic cells (pooled from three rat colons) were incubated with one or two agents as shown in the first column. The experiments were terminated by adding 1 ml of absolute ethanol to the sample at the designated time. The results shown represent the mean±SE of at least six experiments. Concentrations used in these experiments were the following: theophylline, 10 mM; PGE_1, 0.1 mM; serotonin, 10 μM; and SRIF, 0.1 μM. No significant effect of SRIF was seen either alone or when added to theophylline, PGE_1, or serotonin.

* $P < 0.05$ compared to 0′ control by paired t test.

‡ $P < 0.005$ compared to 0′ control by paired t test.

§ $P < 0.001$ compared to 0′ control by paired t test.

‖ $P < 0.01$ compared to 0′ control by paired t test.

¶ $P < 0.02$ compared to 0′ control by paired t test.

absorption. SRIF could thus work by (*a*) blocking calcium entry or mobilization, (*b*) blocking the activation of calmodulin, or (*c*) blocking the effect of calmodulin. Of course, at this time these hypotheses are purely speculative as there is no evidence that SRIF affects calcium-mediated processes in the intestine. In other tissues, however, there is evidence that SRIF may inhibit calcium-mediated secetion (14–17). The role of calcium in SRIF-mediated changes in intestinal ion transport is currently being investigated.

Chronic serotonin administration to rabbits results in net fluid and electrolyte secretion in the ileum in vivo (18). Serotonin produces a transient rise in the I_{sc}, abolishes Na absorption, and produces net Cl secretion in the rabbit ileum in vitro (19). No effect of serotonin on colonic ion transport has been previously demonstrated. Serotonin produced a sustained increase in the I_{sc} and decreased net Na and Cl absorption. Thus, serotonin can produce net fluid and electrolyte secretion in the ileum and inhibit electrolyte absorption in the colon. The minimal effective concentration is 2.6 nM in the ileum and 0.5 μM in the colon. These effects make serotonin a prime candidate for the agent causing diarrhea in the carcinoid syndrome, although other agents have been reported to be elevated in this syndrome (calcitonin, PGE_1, and $PGF_{2\alpha}$, histamine, etc.) (20–26). Three of four patients with the carcinoid syndrome were found to have net jejunal fluid secretion (4, 27).

SRIF has been shown to inhibit diarrhea in two patients with the carcinoid syndrome (3, 4). In one patient, the serotonin blood level was elevated 100-fold and unchanged by the SRIF infusion (3). In both patients however, urinary 5-hydroxyindole acetic acid excretion was reduced, but not to normal levels. Thus SRIF may work by inhibiting the release of serotonin, blocking its effect, or both, in the carcinoid syndrome. Alternatively, SRIF may work by inhibiting the release or blocking the effect of some other secretagogue produced by the tumor (20–26).

The observations made in this study, that SRIF will block secretagogue induced secretion or decreased absorption in the rat colon, plus our previous observations that SRIF will block secretion in the rat jejunum (2), stimulate Na, and Cl absorption in the rabbit ileum (5), and its inhibition of diarrhea in patients with the carcinoid syndrome (3, 4), strongly suggest that SRIF may be a useful therapeutic agent in the treatment of secretory diarrheas.

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