Effects of Secretin on Peritubular Capillary Physical Factors and Proximal Fluid Reabsorption in the Rat

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ABSTRACT The effects of secretin vasodilation on peritubular capillary Starling forces and absolute proximal reabsorption were examined in the rat. Secretin was infused at 75 mU/kg per min into the aorta above the left renal artery. Efferent plasma flow increased from 125±28 to 230±40 nl/min with secretin infusion. Single nephron filtration rate (44±6 vs. 44±7 nl/min) and absolute proximal reabsorption (21±5 vs. 21±4 nl/min) were not significantly changed. Peritubular capillary and interstitial hydrostatic pressures increased with secretin infusions (from 9±0.4 to 15±0.7 mmHg and from 3±0.2 to 4±0.2 mmHg, respectively). Both peritubular capillary and interstitial oncotic pressures decreased (from 25±2 to 20±2 mmHg and from 10±1 to 4±1 mmHg, respectively) during secretin infusion. The net reabsorption pressure for peritubular capillary uptake significantly decreased from 9±2 to 5±2 mmHg and the coefficient of reabsorption increased from 3±1 to 6±2 nl/min per mmHg. We conclude that although secretin causes a vasodilation and decreases net reabsorption pressure, absolute proximal reabsorption is unchanged. Peritubular capillary uptake is maintained, and since net reabsorption pressure is decreased, the coefficient of reabsorption is increased.

INTRODUCTION

Most vasodilators (bradykinin, acetylcholine, and prostaglandin E_2) cause a diuresis and a natriuresis when infused into the kidney (1–3). Marchand et al. (4) have demonstrated that intrarenal infusions of pharmacologic doses of secretin causes a marked increase in renal blood flow without increases in renal interstitial pressure or sodium excretion in the dog. Absolute proximal reabsorption and peritubular capillary pressure were significantly increased with the renal artery infusion of secretin. The failure of increased peritubular capillary pressure to decrease proximal reabsorption was inconsistent with the generally accepted hypothesis linking peritubular capillary uptake with regulation of proximal sodium reabsorption (5). Since peritubular capillary and interstitial oncotic pressures were not measured, the relationship between proximal reabsorption and the net Starling forces for peritubular capillary uptake could not be determined.

To define the relationship between the peritubular capillary Starling forces and proximal tubule reabsorption for this unique vasodilator, all four Starling forces and absolute proximal reabsorption were determined before and during secretin infusion in the rat.

METHODS

Male Sprague-Dawley rats weighing 250–300 g were fasted for 12 h before the study. Inactin (100 mg/kg) was administered intraperitoneally to induce anesthesia. A tracheostomy was performed; both right and left jugular veins were cannulated, and the urinary bladder was catheterized. The left carotid artery was cannulated for monitoring blood pressure and for blood specimen collection. A cannula was positioned at the level of the left renal artery through the left iliac artery for infusion of secretin (Kabi Diagnostica, Studsvik, Sweden). Injection of 5% lissamine green dye was used to confirm this position. Animals were placed on a heated board and core body temperature maintained at 37°C. Surgical blood losses were replaced with isoncotic (5.0 g/100 ml) human serum albumin (1.2 cm^3/h for 2 h). An inulin solution was infused to maintain serum concentrations between 50 and 100 mg/100 ml. All solutions were diluted in 0.9% saline. The total rate of infusion was 3.5 ml/h.

After a 2-h recovery time from surgery, the kidney was prepared for micropuncture through a left flank incision. The kidney was placed in a lucite cup packed with cotton wool to reduce respiratory and vascular motion.

Recollection micropuncture was performed on two late proximal tubules in each rat before and during secretin in-

This work was presented in abstract form in May 1982 at the American Federation of Clinical Research, Washington, DC. Address reprint requests to Dr. Knox.

Received for publication 22 June 1982 and in revised form 2 March 1983.

fusion (75 mU/kg per min) and averaged for single nephron filtration rate and absolute proximal reabsorption. Late proximal tubules were identified with a 0.1 cm^3 venous injection of 5% lissamine green dye. Time-control experiments were performed in a similar manner except that 0.9% saline was infused throughout the experiment into the abdominal aorta. Three subcapsular pressures were averaged before and after secretin infusion to estimate renal interstitial pressure. Fluid from hilar lymphatics close to the kidney was used to measure interstitial proteins. One efferent arteriolar blood sample was obtained before and after secretin. One hilar lymph collection was also obtained before and after secretin infusion. Peritubular capillary hydrostatic pressures were measured and averaged from three medium-sized superficial capillaries before and after secretin infusion. Systemic and surface star vessel protein concentrations were used as estimates of afferent and efferent protein, respectively. All hydrostatic pressures were measured with the servo-nulling apparatus. Inulin was determined by the microfluorometric method (6) and protein by microadaptation of the Lowry method (7).

The following formulae were used in the calculations:

SNGFR (nanoliters per minute) = $V_c \times (TF/P)_{In}$, where *SNGFR* is single nephron glomerular filtration rate, V_c is tubule fluid flow in nanoliters per minute, and $(TF/P)_{In}$ is tubule fluid to plasma inulin ratio.

APR (nanoliters per minute) = *SNGFR* − V_c, where *APR* is absolute proximal reabsorption and *SNGFR* and V_c are as above.

SNFF = $1 - (C_A/C_E)$ where *SNFF* is single nephron filtration fraction, C_A is afferent protein concentration in grams per 100 ml, and C_E is efferent protein concentration in grams per 100 ml.

epf (nanoliters per minute) = *SNGFR/SNFF* − *SNGFR*, where *epf* is efferent plasma flow and *SNGFR* and *SNFF* are as above.

C_{MP} (grams per 100 ml) = $(epf \times C_E)/(epf + CU)$, where C_{MP} is end capillary protein concentration to the point of micropuncture, *CU* is capillary uptake assumed to equal absolute proximal reabsorption (*APR*), and *epf* and C_E are as above.

$\pi_{E,MP}$ (mmHg) = $1.74c + 0.28c^2$, where $\pi_{E,MP}$ is efferent or end capillary oncotic pressure and c is protein concentration in grams per 100 ml (8).

π_c (mmHg) = $\frac{(\pi_E + \pi_{MP})}{2}$, where π_c is peritubular capillary oncotic pressure, π_E is efferent capillary oncotic pressure, and π_{MP} is end capillary oncotic pressure at the point of micropuncture.

π_I (mmHg) = $2.4c + 0.17\ c^2 + 0.01c^3$, where π_I is interstitial oncotic pressure and c is interstitial protein concentration in g/100 ml (9).

NRP (mmHg) = $(\pi_c - \pi_I) - (P_c - P_I)$, where *NRP* is net reabsorption pressure, π_c is peritubular capillary oncotic pressure, π_I is interstitial oncotic pressure, P_c is capillary hydrostatic pressure, and P_I is interstitial hydrostatic pressure.

K_r (nanoliters per minute per mmHg) = *CU/NRP*, where K_r is coefficient of reabsorption and *CU* is as above.

All data were expressed as means±SE. *P*-values were calculated by the paired *t* test.

RESULTS

Table I summarizes the results in the rats before and during secretin infusion and in time-control experiments. Fig. 1 shows the results for all four Starling forces in each rat. The systemic protein concentration was 5.1±0.3 and 5.0±0.3 g/100 ml before and after secretin infusion, respectively. The efferent protein decreased from 7.3±0.4 to 6.1±0.4 g/100 ml ($P < 0.001$) with secretin vasodilation. Peritubular oncotic pressure decreased 5.0±1.1 mmHg ($P < 0.01$)

TABLE I

Peritubular Capillary Physical Factors and Proximal Reabsorption

	BP	*SNGFR*	*APR*	*SNFF*	*epf*	π_e	π_c	π_I	P_c	P_I	*NRP*	K_r
	mmHg	*nl/min*	*nl/min*		*nl/min*				*mmHg*			*ml/min/mmHg*
Effect of secretin (n = 9)												
Control												
(±1 SEM)	142 + 4	44±6	21±5	0.29±0.03	125±28	28 + 2	25±2	10±1	9±0.4	3±0.2	9±2	3±1
Secretin												
(±1 SEM)	138 + 4	44±7	21±4	0.17±0.02	230±40	21 + 1	20±2	4±1	15±0.7	4±0.2	5±2	6±2
$P \leq$	NS	NS	NS	0.001	0.001	0.001	0.01	0.001	0.001	0.001	0.02	0.05
Time controls (n = 5)												
Control												
(±1 SEM)	129 + 6	51±5	22±5	0.32±0.04	119±25	30 + 2	27±2	6±2	9±0.2	3±0.2	14±3	2±0.5
Saline Vehicle												
(±1 SEM)	125 + 4	52±6	21±5	0.41±0.04	78±15	34 + 4	29±3	6±2	8±0.3	3±0.2	17±4	2±0.9
$P \leq$	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS	NS

BP, mean arterial blood pressure; *SNGFR*, single nephron glomerular filtration rate; *APR*, absolute proximal reabsorption; *SNFF*, single nephron filtration fraction; *epf*, efferent plasma flow; π_e, efferent arteriolar oncotic pressure; π_c, peritubular capillary oncotic pressure; π_I, interstitial oncotic pressure; P_c, peritubular capillary hydrostatic pressure; P_I, interstitial hydrostatic pressure; *NRP*, net reabsorption pressure; K_r, coefficient of reabsorption.

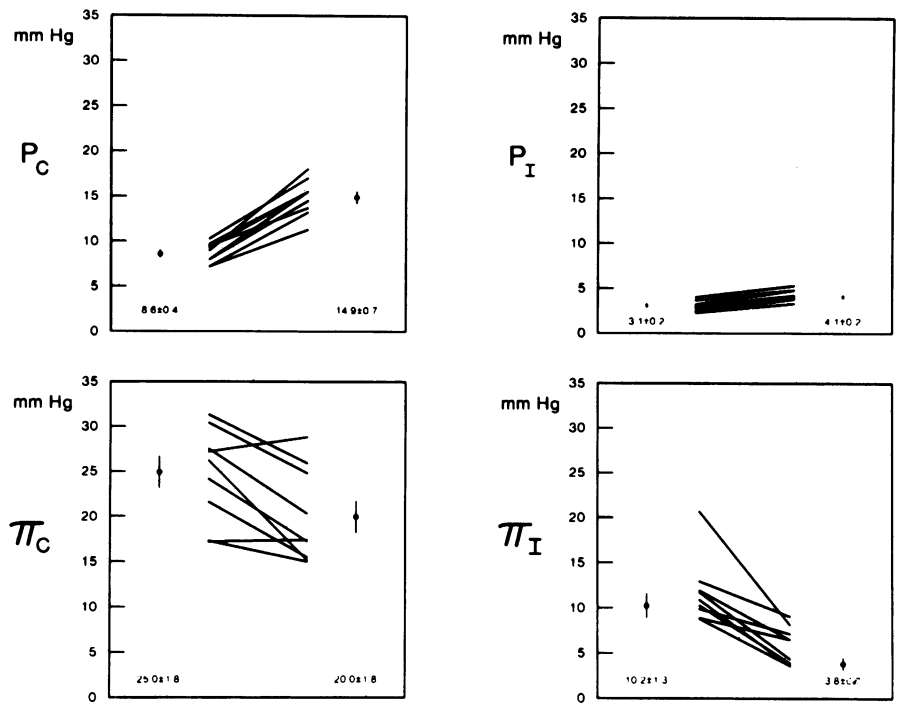

FIGURE 1 Peritubular capillary physical factors before and during secretin infusion, left- and right-hand numbers in boxes, respectively (n = 9). P_c, peritubular capillary hydrostatic pressure (mean change = 6.31; $P < 0.001$); P_I, interstitial hydrostatic pressure (mean change = 0.97; $P < 0.001$); π_c, peritubular capillary oncotic pressure (mean change = −5.0; $P < 0.01$); π_I, interstitial oncotic pressure (mean change = −6.3; $P < 0.001$). Mean values shown±SE.

and interstitial oncotic pressure decreased 6.3±1.1 mmHg ($P < 0.001$). Peritubular capillary hydrostatic pressure increased 6.3±0.5 mmHg ($P < 0.001$) and interstitial hydrostatic pressure slightly increased, 1.0±0.1 mmHg ($P < 0.001$). Secretin infusion did not significantly change glomerular filtration rate or absolute proximal tubule reabsorption. The net reabsorption pressure significantly decreased from 9±2 to 5±2 mmHg ($P < 0.02$). Thus, the coefficient of reabsorption increased from 3.4±0.8 to 6.4±1.9 nl/min per mmHg ($P < 0.05$). Time-control experiments showed no statistically significant changes.

DISCUSSION

These experiments confirm and extend the studies of Marchand et al. (4) of secretin infusion in the dog. Both their studies and our studies show marked increases in blood flow and peritubular capillary hydrostatic pressures during secretin infusion without marked increases in interstitial pressure or decreases in absolute reabsorption by the proximal tubule. Marchand et al. (4) did not measure the peritubular capillary or interstitial oncotic pressures. Nevertheless, they postulated an increase in capillary oncotic pressure, or a decrease in interstitial oncotic pressure, or both, to account for the lack of decrease in absolute proximal reabsorption. Our studies demonstrate not only a decrease in interstitial oncotic pressures (10–4 mmHg) with secretin infusion, but also a decrease in peritubular capillary oncotic pressure (25–20 mmHg).

In 1967, Martino and Earley (10) concluded from clearance studies in the dog that net fluid reabsorption is determined by the rate of removal of reabsorbate by the capillary circulation. Quinn and Marsh (9), using a rat model, examined the relationship between absolute proximal reabsorption and net interstitial pressure ($\pi_I - P_I$). Using Ringer's loaded, plasma loaded, and hydropenic rats, they found a positive correlation between absolute proximal reabsorption and net interstitial pressure. They hypothesized that net interstitial pressure regulated proximal reabsorption and, therefore, was the link between changes in pressures in the peritubular capillaries and reabsorption by the proximal tubule. In our study, net renal interstitial pressure decreased from 7 to 0 mmHg without a change in proximal reabsorption. The net interstitial pressure fell because of a significant decrease in

interstitial oncotic pressure (10–4 mmHg) with only a slight increase in the opposing interstitial hydrostatic pressure (3–4 mmHg). Consequently, using a vasodilator that increases interstitial hydrostatic pressure minimally, no correlation between net interstitial pressure and absolute proximal reabsorption was found. Stimulation of proximal reabsorption by secretin may have prevented a fall in absolute proximal reabsorption when net interstitial pressure decreased, or, alternatively, this pharmacologic tool demonstrated that there is not a causal relationship between changes in net interstitial pressure and proximal reabsorption. It is clear that during secretin vasodilation the renal interstitial Starling forces do not regulate absolute proximal reabsorption.

In the steady state, assuming negligible rate of lymph production, the rate of uptake of peritubular capillaries must equal the tubular reabsorption (5). Quinn and Marsh (9) estimated not only absolute proximal reabsorption, but also absolute distal reabsorption in Ringer's expanded, normal hydropenic, and plasma-expanded rats. The inclusion of distal reabsorption increased total reabsorption (proximal plus distal) by ~20% in each group. Inclusion of distal reabsorption would increase calculated capillary uptake and the coefficient of reabsorption both before and after secretin infusion. Since secretin did not significantly alter absolute proximal reabsorption, it is unlikely that superficial distal reabsorption would selectively change after secretin and alter capillary uptake.

Peritubular capillary and interstitial oncotic pressures decreased by 5.0 and 6.3 mmHg, respectively. The decrease in peritubular capillary oncotic pressure can be attributed to the fall in filtration fraction. Although there is some variation in the absolute values of hilar lymph protein, all values decreased with secretin vasodilation. Thus, the changes in capillary and interstitial oncotic pressures were largely in parallel. Since the hydrostatic pressures were not offset, the net reabsorption pressure for uptake in the peritubular capillaries decreased from 9±2 to 5±2 mmHg. Since uptake was unchanged, the coefficient of reabsorption increased from 3±1 to 6±2 nl/min per mmHg with the infusion of secretin.

The coefficient of reabsorption is the product of the hydraulic conductivity and surface area of the peritubular capillaries. Therefore, secretin infusion could either increase the hydraulic conductivity, the surface area available for uptake, or both. These in turn provide possible mechanisms for sustained capillary uptake in the presence of renal vasodilation with secretin.

ACKNOWLEDGMENTS

We appreciate the fine secretarial assistance of June Hanke and the technical assistance of Marcy Onsgard.

This work was supported by National Institutes of Health grants HL 14133 and AM 07013 and by Mayo Foundation.

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