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The Role of the Renal Lymphatics in Fluoride Handling[1]

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Summary

The role of the renal lymphatics in the handling of fluoride ion by the canine kidney was studied during IV sodium fluoride infusion. It was found that renal lymph and renal venous plasma have similar fluoride concentrations and that there was less fluoride in lymph than in arterial plasma. Fluoride did not alter renal lymphatic flow rate over a wide range of plasma fluoride concentrations.

Introduction

The renal handling of inorganic fluoride ion (F^-) has received much attention recently due to the potential for kidney toxicity when the ion is present in high concentration. Fluoride ion appears in the plasma as a result of the metabolism of certain halogenated anesthetics (1), inclusion in dental preparations (2) and increasing therapeutic use in osteoporosis (3). The present study was designed to show what role the renal lymphatic system plays in renal F^- handling and if the presence of F^- alters renal lymphatic flow.

Methods

Six mongrel dogs of either sex (19–28 kg) were anesthetized with IV sodium pentobarbital (30 mg/kg), intubated and allowed to breathe room air spontaneously. The right femoral artery and vein were exposed and catheterized with polyethylene tubing for collection of arterial samples and infusion of solutions, respectively. The left kidney was exposed via a flank incision and the left ureter catheterized with polyethylene tubing to facilitate urine collection. The kidney was retracted dorsally and the left gonadal vein was exposed and catheterized with polyethylene tubing (Clay Adams PE-90). The catheter was advanced into the left renal vein for collection of renal venous blood. In five dogs, a renal hilar lymphatic was carefully isolated and catheterized with a 30 cm polyethylene tube (Clay Adams PE-10) which had about a 15 μl volume. When free lymphatic flow was assured, the animals were systemically heparinized to prevent clotting of the lymph (300 U/kg). Aortic blood pressure was monitored by connecting a NARCO LDI-5 pressure transducer to the arterial catheter. Heart rate and electrocardiogram were recorded using needle electrodes and NARCO 7172 High Gain Coupler. Respiratory rate was monitored by a NARCO 7212 Impedence Pneumograph Coupler. Blood pressure, heart rate/ECG and respiratory rate were continuously monitored on a NARCO Bio-Systems PMP-4A recorder.

Following stabilization of the preparation, an inulin priming dose (50 mg/kg) was given, followed by a sustaining infusion of 40 mg/kg/hr at 0.1 ml/kg/min. One hour of infusion prefaced the beginning of the first control period.

Control and experimental periods were set at 20 minutes since the required 0.2 ml lymph for F^- analysis could be collected in that time. Lymph was collected in 0.2 ml micropipettes (Yankee disposable micropet) and immediately analyzed for F^-. Three control periods were followed by six experimental periods during which an isotonic solution of sodium fluoride (0.1 mg/kg/min) was infused at 0.1 ml/kg/min. This infusion rate of sodium

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fluoride raised plasma F^- concentrations to values associated with nephrotoxicity in surgical patients (1) and experimental animals (4, 5). Arterial and renal venous blood samples were collected at the mid-point of each period. Urine, collected in graduated tubes, was sampled at the end of each period. Lymph and arterial plasma F^- concentrations varied with time in a linear manner (correlation coefficient: arterial = 0.9795, lymph = 0.9956) so that the lymph F^- concentration at the mid-point of each lymph collection period could be estimated with certainty.

Inorganic F^- in urine, lymph and plasma was analyzed using an Orion 96-09 specific ion electrode and Orion model 801A pH meter by a modification of the method of *Fry and Taves* (6). Inulin in urine and plasma was determined by the method of *Heyrovsky* (7). Urine osmolality was determined by freezing point depression (Advanced Instruments Model 3W Osmometer). Statistical analysis of data was accomplished by a multi-group, repeated measures ANOVA (8). Differences within groups were partitioned into types of trends by use of orthoganal, polynomial coefficients while differences between groups were determined by a Tukey Post Hoc comparison (9).

Results

Average control values for arterial F^-, renal venous F^- and renal lymph F^- were 1.92 ± .32 μM/l, 1.61 ± .29 μM/l and 1.52 ± .27 μM/l, respectively. After 120 minutes of NaF infusion these values had reached 517 ± 7/ μM/l (arterial), 433 ± 62 μM/l (renal venous) and 475 ± 82 μM/l (lymph). Table 1 has complete mean values for all F^- determinations.

Table 2 shows the F^- concentration ratios calculated for renal lymph to arterial plasma (L/P_A), renal lymph to renal venous plasma (L/P_V) and renal venous to arterial plasma (P_V/P_A). There was no significant difference among the average control values. P_V/P_A was less than unity during the infusion and an analysis of variance indicated that the ratio itself did not change as the experiments progressed. L/P_A and L/P_V increased from

Table 1 Fluoride in renal hilar lymph, arterial plasma, renal venous plasma and urine before and during sodium fluoride infusion

	Average Control	10	20	30	40	50	60	70	80	90	100	110	120 min
Lymph F^- μM/l (n = 5)	1.52 ± .27	27 ± 8		111 ± 26		208 ± 40		291 ± 52		368 ± 64		437 ± 80	
Arterial Plasma F^- μM/l (n = 6)	1.92 ± .32	137 ± 22		271 ± 49		347 ± 46		405 ± 53		443 ± 58		516 ± 71	
Renal Venous F^- μM/l (n = 6)	1.61 ± .29	123 ± 18		244 ± 41		298 ± 44		339 ± 39		398 ± 60		433 ± 62	
Urine F^- mM/l (n = 6)	0.7 ± .20		4.3 ± 1.0		13.6 ± 4.0		21.0 ± 4.8		25.9 ± 5.6		28.2 ± 5.1		31.5 ± 6.6

Table 2 Renal lymph to arterial plasma (L/P_A, n = 5), renal venous plasma (L/P_V, n = 5) and arterial to renal venous plasma (P_A/P_V, n = 6) ratios for fluoride ion before and during sodium fluoride infusion. (*indicates a significant, linear trend, $P < .01$)

	Average Control	20	40	60	80	100	120 min
L/P_A*	.77 ± .09	.17 ± .02	.38 ± .03	.57 ± .04	.68 ± .03	.80 ± .01	.81 ± .01
L/P_V*	.90 ± .11	.20 ± .03	.43 ± .04	.66 ± .05	.83 ± .03	.89 ± .02	.96 ± .02
P_V/P_A	.94 ± .11	.91 ± .04	.90 ± .01	.85 ± .01	.85 ± .02	.89 ± .02	.84 ± .02

Table 3 Renal fluoride clearance (C_F), renal inulin clearance (C_{IN}) and the fractional fluoride clearance (C_F/C_{IN}) before and during NaF infusion (n = 6). (*indicates a significant, linear trend, $P < .01$)

	Average Control	20	40	60	80	100	120 min
C_F (ml/min)	16.9 ± 3.5	10.3 ± 1.8	19.0 ± 4.7	22.2 ± 5.4	24.9 ± 6.1	22.2 ± 6.4	20.2 ± 6.7
C_{IN} (ml/min)	27.1 ± 4.5	27.8 ± 5.6	30.1 ± 6.9	32.1 ± 7.9	30.7 ± 6.1	25.1 ± 4.5	25.2 ± 6.8
C_F/C_{IN}*	.55 ± .07	.41 ± .05	.65 ± .09	.72 ± .08	.84 ± .10	.81 ± .12	.72 ± .11

Table 4 Mean arterial pressure, respiratory rate, urine flow, urine osmolality and lymph flow rate before and during NaF infusion (n = 6)

	Average Control	20	40	60	80	100	120 min
Arterial Pressure (mmHg)	141.3 ± 6.4	143.3 ± 5.6	145.3 ± 5.4	143.3 ± 6.8	149.3 ± 4.0	145.2 ± 6.1	144.0 ± 4.0
Respiratory Rate (Resp/min)	4.2 ± 0.2	4.3 ± 0.6	4.5 ± 0.7	4.5 ± 0.6	4.5 ± 0.3	4.8 ± 0.5	4.3 ± 0.3
Urine Flow Rate (ml/min)	0.48 ± .10	0.34 ± .06	0.39 ± .06	0.38 ± .07	0.36 ± .06	0.31 ± .05	0.26 ± .05
Urine Osmolality (mOsm/kg)	658 ± 131	755 ± 155	717 ± 147	696 ± 133	726 ± 122	717 ± 101	720 ± 101
Lymph Flow (µl/min, n = 5)	13.5 ± 2.1	13.3 ± 2.0	11.9 ± 2.0	12.7 ± 2.2	14.5 ± 2.1	15.1 ± 2.2	14.4 ± 2.7

0.17 ± .02 and 0.20 ± .03, respectively, at ten minutes of infusion, to 0.81 ± .01 and 0.96 ± .02, respectively, at the end of the experiments.

Fluoride clearance (C_F) and fluoride fractional clearance (C_F/C_{IN}) increased slightly at the beginning of the infusion and remained constant, as did inulin clearance (C_{IN}), for the remainder of the experiments (Table 3). Similarly, arterial pressure, respiratory rate, urine flow, urine osmolality and lymph flow rate were unaltered (Table 4).

Discussion

The presence of elevated F^- in the blood and urine is associated with a syndrome of polyuric failure in surgical patients (1) and similar dysfunction in experimental animals (4, 5). The kidney responds to F^- in the filtrate by tubular reabsorption of 20–95 % of the F^- tubular load and urinary excretion of the remainder. The portion of the filtered F^- reabsorbed is closely related to urinary pH in that reabsorption increases in the presence of an acid urine (10). The mechanism by which F^-

alters renal function is not understood but may be related to an alteration of the action of antidiuretic hormone on the collecting tubule portion of the nephron (11) when F^- accumulates to toxic concentrations. The present investigation demonstrated that the renal lymphatics return a small, but relatively constant percentage of the reabsorbed F^- to the circulation and that, even at high blood and urine concentrations, F^- did not affect renal lymphatic flow rate.

In these experiments it was difficult to characterized lymph to plasma F^- ratios prior to NaF infusion. A standard F^- curve was not consistently linear in the range of 1–2 μM/l with the specific ion electrode (6), but control lymph F^- concentrations were consistently less than in arterial or renal venous plasma (Table 1). This is probably due to mixing with tubular reabsorbate from which F^- has been removed by excretion. Further experiments will be conducted to characterize lymph F^- concentrations resulting from dietary and environmental sources. Fluoride entered the renal lymph from capillary filtrate with a contribution from tubular reabsorbate which gave it a F^- concentration not unlike renal venous plasma. Under pre-infusion conditions, the ratio of lymph F^- to arterial F^- was less than unity, suggesting that the capillary filtrate is "diluted" by relatively F^- free reabsorbate. About 55% of the fluoride filtered was excreted, under control conditions (Table 3). As the plasma F^- concentration was increased by infusion, the percent excreted increased significantly and leveled off at about 80%. It may be that, in these experiments, F^- excreting ability was limited by urine flow rate or urinary pH. It was clear that plasma F^- concentration, and therefore tubular load, was continuously increasing. The ratio of F^- in renal venous and arterial blood was unchanging, so that the excess reabsorbed F^- was not entering the peritubular capillaries but was sequestered in the renal medulla. *Whitford and Taves* (13) have demonstrated that, in F^- infused rats, there was from three to four more times F^- in medulla than cortex. While the absolute amount of F^- removed by the lymphatics seemed small if based on accepted criteria of ten lymphatics per kidney (12) and a flow rate of about 13–15 μl/min, it may be that the continual removal of even small amounts of F^- from the cortical interstitium aided in preventing alteration of cortical function. There is no evidence of alteration of cortical function attributed to F^- and no data as to the cortical concentration which may be toxic.

The unchanging lymph flow rate (Table 3) was surprising since F^- has been associated with peripheral vasodilatory effects (14) and alterations in glomerular filtration rate (3) at similar plasma concentrations. We suspected that changes in total renal blood flow or intrarenal blood flow distribution would occur and, most likely, increase lymph flow. This indirect evidence leads us to believe that F^- does not alter renal hemodynamics and is a worthy topic of further investigation.

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