

Effect of Intraluminal Pressure on Proximal Tubular Sodium Reabsorption. A Shrinking Drop Micropuncture Study*

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Intrarenal oncotic and hydrostatic pressures have been shown to play an important role in the regulation of sodium transport across the proximal tubular epithelium. While peritubular changes in oncotic pressure in part control the rate of tubular sodium reabsorption(1), similar modifications of protein concentration at the intraluminal level are of much lesser significance(2). The decrease of net sodium reabsorption rate induced by a fall in peritubular protein concentration has been shown to be associated with an increase in ionic conductance of the intercellular shunt pathways, resulting in increased backflow of sodium to the lumen(3,4). Regarding hydrostatic pressure changes, peritubular hydrostatic pressure changes have been suggested to affect tubular sodium reabsorption(5,6). At present, evidence is lacking that modifications of the transepithelial hydrostatic gradient, when induced by intraluminal pressure changes, exert a reversed but symmetrical effect on proximal tubular sodium and fluid transfer.

In the present study a significant effect of intraluminal pressure on the rate of sodium reabsorption of *Necturus* proximal tubule has been clearly demonstrated. An intraluminal pressure clamp technique is described in this paper, and its application to the shrinking drop micropuncture technique is illustrated. Moreover, the pressure characteristics of the shrinking drops have been determined and are compared to the pressures occurring in various tubular and extratubular structures of the *Necturus* kidney.

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METHODS

Thirty adult specimens of either sex of *Necturus maculosus* ranging in weight from 75 to 160 g were used. The animals were obtained from the Mogul Corporation, (Oshkosh, WI) during the months from October to April, and kept in flowing, charcoal-filtered tap water at 15–20°. Animals kept over the summer were fed with living minnows. Anesthesia and surgical procedures were performed as described earlier(3) with the exception that the solution covering the gills was bubbled with air. A modified Ringer's solution (Na^+ 100.5 mM, K^+ 2.5 mM, Cl^- 98.1 mM, HCO_3^- 10 mM, H_2PO_4^- 0.5, Ca^{2+} 1.8 mM, and Mg^{2+} 1.0 mM) was infused via the cranial portion of the ventral abdominal vein for a period of 2 hr at a rate of $0.1 \mu\text{l min}^{-1}\text{g}^{-1}$. The exposed right kidney was superfused with a similar solution. The skin of the animal was maintained wet by means of water and moist cellulose tissue. All measurements were started 30–60 min after surgery.

The *shrinking drop micropuncture technique* was performed as previously described(3). Stained oil (1000 centistokes (cSt) silicone oil, General Electric SF-96, and 10% Automate Black No. 1, Patent Chemicals, Inc., Paterson, NJ) was injected into the proximal tubule through a single-barreled micropipet having a tip diameter of 10–15 μ (prepared from Friedrich-Dimmock, Millville, NJ, KG-33 borosilicate glass o.d. 0.98 mm, i.d. 0.68 mm). The oil droplet was split by means of a second, similar micropipet (tip diameter 8–12 μ) filled with Ringer's solution. The shrinking drops were placed in a straight segment of the non-pigmented late portion of the proximal tubule. Two techniques for preparing the split drop were used and are described in Fig. 1. In technique A (Fig. 1A), the Ringer's droplet was maintained in place by immobilization of the downstream oil droplet in the intermediate tubular segment, a tubule site offering a high resistance to the flow of oil. The upstream oil droplet (length: 5–10 tubular diameters) remained mobile and moved preferentially during the period of the fluid shrinkage. In this condition it is expected that the pressure in the Ringer's droplet be close to the "stop-flow" pressure in the corresponding early proximal convolutions. In technique B (Fig. 1B) the Ringer's droplet was maintained in place after injection of a long upstream oil block and a subsequent puncture of Bowman's capsule to eliminate the effects of the filtration pressure. In this situation, in contrast to the previous one, it is the downstream oil droplet (5–10 tubular diameters in length) which remains mobile during the shrinkage. In this condition it is to be expected that the intraluminal pressure is lower than that obtained using technique A. In both techniques, the impalement sites, separated more than three tubular diameters away from the Ringer's droplet, were carefully sealed by the stationary oil block. Observation of the decrease in length of the split-drop fluid sample determined by means of a filar micrometer, was started immediately after its deposition. Tubular diameters were estimated from the distance between the light reflex on either inner surface of the tubular wall. For a given intraluminal pressure condition, the tubular diameter remained constant throughout the time of reabsorption. As described previously(3), from

the evolution in time of the length of the droplet between the two menisci and the tubular radius, the following calculations were made:

$$V/V_0 = (L + 2a) / (L_0 + 2a), \quad [1]$$

where V/V_0 is the ratio of the volume of the droplet at time t compared to its initial volume, L and L_0 are the corresponding lengths of the droplet, and a the tubular radius. Since the volume changes exponentially with time:

$$\ln V/V_0 = -k t \quad [2]$$

$$t_{1/2} = 0.693 / k, \quad [3]$$

where $t_{1/2}$ is the half-time of reabsorption (min) and k (min^{-1}) is the rate constant of the exponential function obtained from the slope of $\ln V/V_0$ against time. As shown by Gertz(7), J_v , the net volume flux ($\text{ml min}^{-1} \text{cm}^{-2}$), is given by:

$$J_v = k \left(\frac{a}{2} \right) \quad [4]$$

Note that the rate constant k has been referred to in the literature as the intrinsic reabsorptive capacity $\left(\frac{C}{\pi a^2} \right)$ (8).

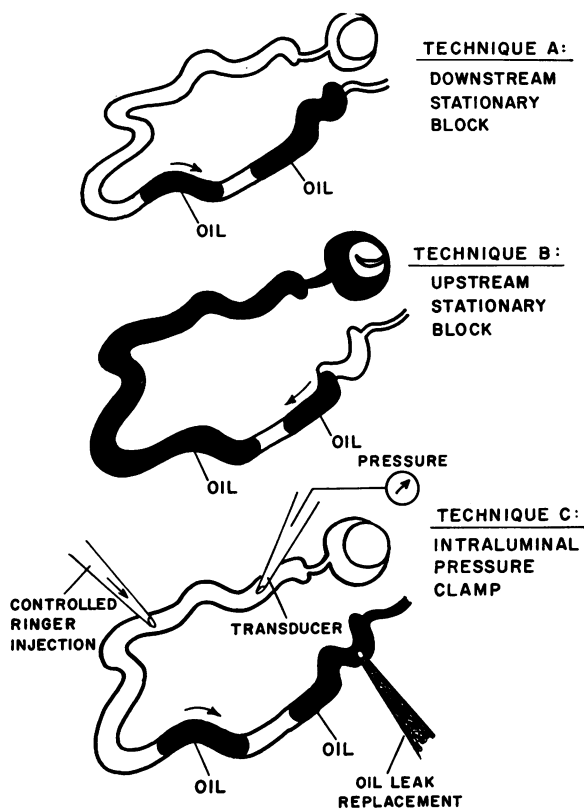


FIG. 1. Different methods for the preparation of split drops (techniques A and B). Method for intraluminal pressure clamping (technique C).

Hydrostatic pressures were measured in various vascular elements and in proximal tubules of the kidney. Stop-flow intratubular pressures proximal to the oil block were measured after a period sufficiently long to allow the oil drop to be blocked in the intermediate segment. Peritubular pressure was measured in venous peritubular capillaries of 30- to 100- μ diameter halfway between the medial and lateral borders of the kidney.

A few experiments were performed using the Landis technique(9) with buffered phosphate-filled micropipets of 8- to 12- μ diameter. The solution was stained with lissamine green (5%). A water manometer was used. The majority of the pressure measurements were obtained by means of the Wiederhielm technique(10,11). Although this method allows the use of pipets as small as 1- μ tip diameter, intrarenal measurements in *Necturus* required sharpened tips of 4-10 μ outer diameter. The pipets, filled with a solution of low resistivity (1.5 M NaCl) had a resistance of 0.3-1.0 10^6 ohms. Five percent lissamine green was added to the 1.5 NaCl solution in order to provide an optical control of the localization of the interface between the outer fluid of relatively high resistivity and the fluid filling the remainder of the micropipet.

The methods of Landis and Wiederhielm are based on a same basic principle. In both the pressure is measured which maintains zero net flux in the micropipet when the tip is immersed in a fluid at pressure P_x . Hence

$$P_x = P_c + P_h - P_{cap}, \quad [5]$$

where P_c is the counterpressure necessary for nulling any net flux in the micropipet, P_h is the hydrostatic pressure exerted by the fluid column in the micropipet for a given incline, and P_{cap} the capillary pressure. Since P_h and P_{cap} are constant, P_x is a linear function of P_c . As summarized in Table 1 the two methods differ solely in the procedure for nulling net flow in the pipet. Small changes in pressure dP_x produce microdisplacements dl of the interface within the tip of the pipet. Whereas, in the Landis technique this displacement is visible by means of the color transition of the interface, the Wiederhielm technique detects the shift dl as a change in resistance of the micropipet since a movement of the interface changes the amount of external fluid of high resistivity within the tip of the

TABLE 1
FLOW NULLING SYSTEMS

Steps of the feedback mechanism	Landis	Wiederhielm
dP_x induces interface displacement dl	Optical detection of dl	Electrical (resistive) detection of dl
Detected signal causes feedback response dP_c	Operator	Electronic servo-system
dP_c compensates dl	Operator	Linear motor and bellows

pipet. In Table 2 the resistance values of a typical micropipet are shown as a function of the localization of the interface. The resistance of the micropipet was linearly related to the position of the interface up to 100 μ (distance between interface and final tip), since no change of diameter occurred in that segment. Concurrent with the widening of the pipet diameter, relatively smaller resistance changes were produced by additional shifts of the interface. In the flow-nulling system only very small (optically undetectable) interface displacements dl occur since resistance changes of less than 1% of the overall resistance can be accurately detected. With respect to the position of the interface, both techniques require that the latter be accurately placed in the tip section of the micropipet where dl corresponds to minimal volume displacement. In addition, the small crosssectional surface of the interface reduces mixing of the two phases by diffusion. In the original technique of Wiederhielm the resistance is measured by means of a AC Wheatstone bridge(10,11). In the present study, a preamplifier phase detector served as the transducer headstage. An electronic servo loop maintained the appropriate feedback response from a linear motor and bellows. The counter-pressure P_c was applied through an air-filled system and read by means of a strain gauge(12).

A comparison of some performance features of the two methods is summarized in Table 3. Both techniques show a similar accuracy within 1 mm H₂O. The faster response time of the Wiederhielm technique is significant and allows re-

TABLE 2
CHARACTERISTICS OF A TYPICAL MICROPIPET IN WIEDERHIELM'S METHOD

Distance between interface and final tip of the micropipet (10 ⁻⁴ cm)	Micropipet outer diameter at the level of interface (10 ⁻⁴ cm)	Overall resistance of the micropipet (ohm 10 ⁶)
0	10	0.8
10	10	1.0
100	10	3.3
200	14	5.3
300	22	6.8
400	30	7.8

TABLE 3
COMPARISON OF THE METHODS OF LANDIS AND WIEDERHIELM

	Landis	Wiederhielm
Accuracy	Within 1 mm H ₂ O	Within 1 mm H ₂ O
Tip diameter	2-12 μ or more	0.5-10 μ or more
Response time	Several seconds	0.035 seconds
Operator involvement	Constant	Temporary
Pressure measurements in closed spaces (split drop)	Doubtful	Accurate

liable recordings of pulsatile pressures. Involvement of the operator is minimal in the Wiederhielm method. Reliable measurements of pressure in such closed spaces as split drops are possible as only minimal volume displacements occur with the servomechanism method of Wiederhielm. Accordingly, contamination of the split drop by the hypertonic pipet solution can be easily avoided.

The use of the Landis method involves larger volume displacements accompanying the nulling procedure. This aspect of the technique could affect the unknown P_x of the closed space of the split drop. In addition, contamination of the Ringer droplet by the stained pipet solution breaks down the color demarcation of the interface. These difficulties are avoided using the Wiederhielm technique. Choosing the appropriate starting point of resistance, the interface is placed at a distance of 100–300 μ from the final tip and thus assures that a sufficient amount of uncontaminated external fluid remains always in the tip of the pipet. Moreover, the addition of the input resistance of the tubular wall in series with a high pipet resistance does not affect appreciably the localization of the interface after impalement. It proved of advantage to temporarily open the servo feedback loop during the insertion of the pipet. With a closed servo loop the obstruction of the tip by membrane material during tubular impalement produces a rise in tip resistance, a buildup of P_c (see the transient hyperpressure artifact during the impalement in the example of Fig. 3), and a transient outflow of hypertonic NaCl at the moment when the tip penetrates the tubule wall. Such outflow was made visible by staining the 1.5 M NaCl solution within the pipet tip with lissamine green. The hyperpressure artifact is avoided in the open-loop condition. In the latter situation the interface rises in the pipet at the end of the impalement, up to the time when closing the loop restores the initial interface level.

The use of an *intraluminal pressure-clamp technique* shown as technique C in Fig. 1, made it possible to study the effect of different steady intraluminal pressure levels on the rate of volume reabsorption in individual split drop experiments. As illustrated in Fig. 1C, hydrostatic pressure was measured in the tubular convolutions proximal to the split-drop assembly similar to technique A. Such pressure measurements correspond closely to the directly measured pressure within the Ringer's droplet. Indeed, virtually complete transmission of hydrostatic pressure across the proximal upstream mobile oil drop (similar to that of technique A) has been demonstrated (see Results and Table 4). Except in the experiments summarized in Table 4, we have chosen not to undertake direct pressure determinations in the split drop to avoid leaks and damage of the tubular wall. Different absolute levels of intraluminal pressure (continuously monitored by the Wiederhielm technique) were achieved in stop-flow conditions or during controlled injection of Ringer's into the tubule proximal to the split-drop assembly. Appropriate proximal infusion rates were used to raise the pressure to levels compatible with the maintenance of the stationary downstream block. The stability of the latter was assured by means of an oil-replacement pipet (see Fig. 1C). Ringer's injection was controlled by constant adjustment of an air-pressure pump.

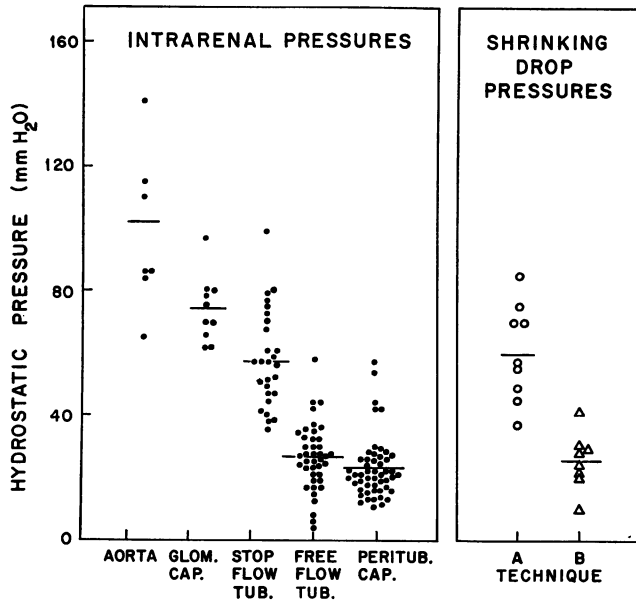


FIG. 2. Comparison of intrarenal hydrostatic pressures with pressures occurring within shrinking split drops.

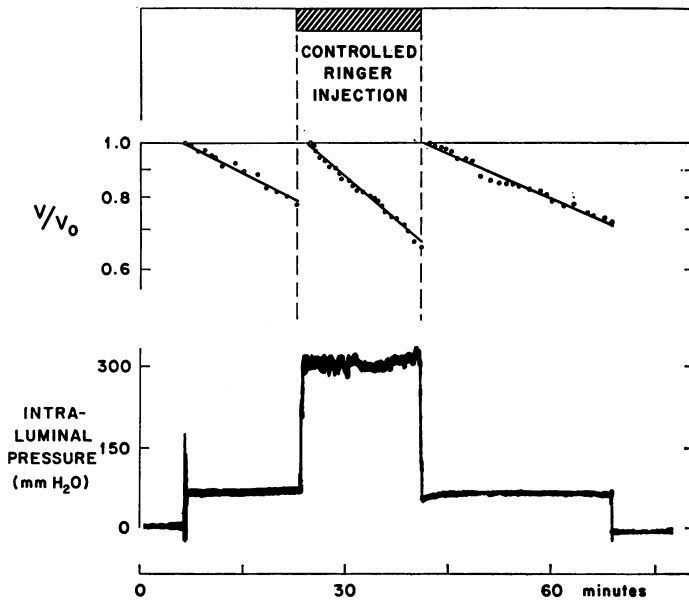


FIG. 3. Top: Relative volume of a single Ringer's droplet plotted against time in three successive periods at different intraluminal pressures. Bottom: Continuous recording of the intraluminal pressure during the same periods.

The decrease in volume of a single split drop was observed at one to three pressure levels, each being maintained for a period of 10 min, or for periods corresponding to a volume decrease of 30%. During each period of clamp pressure the tubular diameter was constant. However, since tubular diameters changed as the hydrostatic pressure level was varied, all volume changes were expressed as the fraction of the initial volume. Care was taken to randomize the sequence of pressure levels.

RESULTS

Intravrenal pressures. As shown on the left of Fig. 2, the mean pressure in the glomerular capillaries was 74.0 ± 3.3 (SEM) mm H₂O ($n=10$), corresponding to a pressure 28% below that of the aortic pressure. Proximal tubular pressures were 57.4 ± 3.0 ($n=27$) and 26.6 ± 1.7 ($n=41$) mm H₂O in stop-flow and free-flow conditions, respectively. The mean pressure in portal peritubular capillaries was 23.0 ± 1.4 ($n=51$) mm H₂O corresponding to a small positive transepithelial

TABLE 4
TRANSMISSION OF INTRALUMINAL PRESSURE ACROSS THE MOBILE OIL DROP (TECHNIQUE C)

Tubule number	Upstream clamp pressure*	Shrinking drop pressure*	D*
1	50	50	0
	185	190	5
	102	105	3
	67	67	0
	45	48	3
	180	185	5
	130	135	5
	110	115	5
	50	55	5
	2	45	50
140		132	- 8
132		120	-12
150		135	-15
135		132	- 3
195		200	5
3	45	45	0
	105	102	- 3
	35	42	7
	132	137	5
	45	48	3
	82	88	6
	62	65	3
	42	45	3
	110	112	2
165	170	5	
Mean difference			1.4 ± 1.2

* mm H₂O.

pressure gradient of 3.6 mm H₂O, statistically not significant ($0.05 < P < 0.1$). Preliminary studies with the Landis technique had shown similar values of stop-flow (56.6 ± 2.8 ($n=21$)), free-flow (22.2 ± 1.5 ($n=25$)), and peritubular capillary pressure levels (21.3 ± 0.8 ($n=36$)) mm H₂O.

Intraluminal pressure in shrinking drops. Pressure values of shrinking drops of technique A and B are also given in Fig. 2. In technique A with a downstream stationary block the mean pressure was 59.9 ± 5.2 ($n=9$) mm H₂O. In technique B using an upstream stationary block, a significantly lower mean pressure of 25.8 ± 3.2 ($n=8$) mm H₂O was found. The difference between both groups of split drops is significant below the 0.001 level. Actually, the pressure level in split drops using technique A is identical to the pressure in control stop-flow tubules ($0.6 < P < 0.7$). In contrast, the pressure in split drops using technique B is similar to the pressure value in free-flow tubules and peritubular capillaries. These results confirm the thesis that the method for preparing a split-drop can influence its intraluminal pressure. The data also demonstrate that the presence of what has been described in our technique as a "mobile oil block" assures complete transmission of the hydrostatic pressure head into the split-drop fluid column (technique A). Further evidence supporting this notion was obtained using technique C. In three tubules pressures were monitored simultaneously with two Wiederhielm pressure devices, one located in the shrinking droplet and the other in the adjacent upstream proximal tubular convolution. This arrangement allowed stepwise changes of the intratubular pressure above the split drop to be monitored during a controlled injection of Ringer's through a micropipet inserted at that level. Two minutes were allowed for equilibration after each pressure change. As shown in Table 4, the transmission of pressure through the mobile oil drop was complete as indicated by the insignificant pressure differences between paired measurements ($0.20 < P < 0.30$). Rapid equilibration of pressure across the mobile oil block is also found when the constancy of pressure in a split drop is tested during the time of reabsorption. Table 5 shows no significant change in intraluminal pressure between the beginning of a reabsorptive period and after 10 min of reabsorption. Accordingly, the data on the right of Fig. 2

TABLE 5
EFFECT OF SHRINKAGE ON THE INTRALUMINAL PRESSURE OF SPLIT DROP
WITH ONE MOBILE AND ONE STATIONARY OIL DROP

Tubule number	Technique	Initial pressure*	Pressure after 10 min*	D*
1	A	55	50	- 5
2	A	85	95	10
3	A	75	80	5
4	B	25	17	- 8
5	B	22	20	- 2
6	B	30	37	7
Mean difference				1.2 ± 2.9

* mm H₂O.

are pooled values of pressure measurements obtained at different times during the shrinkage.

Effect of intraluminal pressure on proximal tubular fluid and sodium reabsorption. Since striking pressure differences exist in split drops depending on the use of either technique A or B, the reabsorption rate was compared in both techniques. The scatter of the values of rate constants does not allow us to conclude that there is a significant effect of intratubular pressure on the rate constant of fluid reabsorption (= intrinsic reabsorptive capacity). This explains the absence of marked differences between the half-times of reabsorption reported by different laboratories using a technique similar to A(3), or, possibly closely resembling our technique B(13).

A much wider pressure range was obtained and could be explored by means of technique C. Single split drops were exposed to one to three levels of intraluminal pressure and their reabsorption was simultaneously monitored. A typical experiment is shown in Fig. 3. In this particular example, it was possible to determine the rate of fluid and sodium reabsorption successively at 60, 300, and 60 mm H₂O. The rate constant, or intrinsic reabsorption capacity, k , calculated according to Eq. [2], was 15, 24, and 12 10^{-3} min^{-1} , respectively.¹ A summary of the results of measurements obtained in 15 tubules kept at one to three levels of intraluminal clamp pressures are shown in Fig. 4. It is apparent that intraluminal pressure changes significantly affect the sodium reabsorptive capacity. The effect was obvious in paired values obtained in single tubules (dots joined by lines) as well as between different tubules. The mean of the slopes of single tubules is $0.110 \pm 0.031 \text{ } 10^{-3} (n=15) \text{ min}^{-1} (\text{mm H}_2\text{O})^{-1}$ and the regression line for all values of Fig. 4 (including those obtained with split drops of technique A and B) has a slope of $0.075 \pm 0.022 \text{ } 10^{-3} \text{ min}^{-1} (\text{mm H}_2\text{O})^{-1} (n=40)$. The Students t test for null hypothesis of the values of both the mean of individual slopes and the regression slope gives a $P < 0.01$. The nonlinearity test for the regression line is not statistically significant(14).

From the corresponding tubular radius for each pressure level and k , J_v was calculated from Eq. [4]. Using paired values of J_v obtained in single tubules, a change in volume flux, ΔJ_v was calculated and divided by the corresponding change in pressure ΔP . The mean ratio corresponding to 15 pressure shifts was $0.537 \pm 0.143 (n=15) \text{ } 10^{-7} \text{ cm}\cdot\text{sec}^{-1} (\text{cm H}_2\text{O})^{-1}$. This value has the dimension of an apparent hydraulic conductivity L_{app} for the effective barrier between lumen and blood vessel. The pressure-induced change in tubular diameter was on an average $0.27 \text{ } 10^{-4} \text{ cm}\cdot(\text{cm H}_2\text{O})^{-1}$.

DISCUSSION

This paper describes the hydrostatic pressure levels in tubular and vascular

¹ Since the rate constant k is linearly related to the effective net sodium transport, it was used instead of $T_{1/2}$. $T_{1/2}$ indeed is a hyperbolic function of both volume flux and intrinsic reabsorptive capacity, as mentioned in Eqs. [3] and [4].

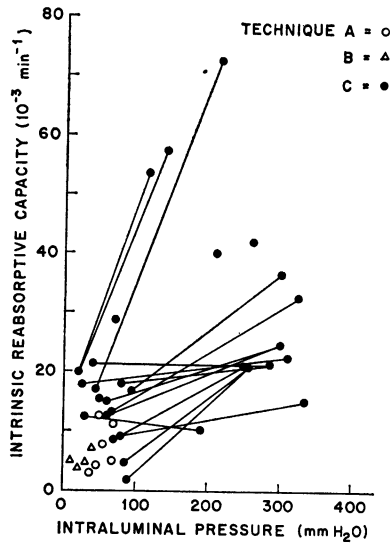


Fig. 4. Effect of intraluminal pressure on the rate constant of volume reabsorption in split drops.

structures of the *Necturus* kidney and some of their functional implications. In addition, two methods of measuring pressures, that of Wiederhielm(10,11) and that of Landis(9) were compared. Both techniques have the same accuracy. The advantages of the Wiederhielm method arise from its fast response time, the minimal involvement of the operator, and its applicability to pressure measurements in very small closed spaces such as shrinking drops.

Aortic pressures are slightly lower than systemic arterial pressures in another amphibian species, the frog(10). The difference between glomerular capillary pressure and tubular stop flow pressure of 16.6 mm H₂O should be equal to the mean effective oncotic pressure within the glomerular tuft. Considering a mean plasma protein concentration measured in the aorta of 2.6 g/100 ml,² an oncotic pressure of 91 mm H₂O would be calculated according to the Pappenheimer formula. The discrepancy of this value and the observed pressure difference between glomerular capillaries and early proximal tubular pressure under stopped-flow conditions can be interpreted to be due to an incorrect estimate of the effective oncotic pressure in the glomerular capillaries. Alternatively, it could be evidence for a functional role of the nephrostome in generating a significant pressure between the surface and the early proximal tubule in *Necturus* kidney. Free-flow proximal tubule and peritubular capillary pressures in the present study are similar to corresponding values reported by Bentzel *et al.*(13). All intrarenal pressures of *Necturus* are at least one order of magnitude lower than the corresponding pressures in the mammalian kidney(15,16). The transepithelial hydrostatic pressure gradient is also smaller than in the rat(15), possibly because

² Unpublished observation.

of the presence in Necturus of occasional negative pressure gradients due to a fall in glomerular filtration in the presence of continued portal perfusion.

An important finding in the present study was the observation that the measurement of pressure within the split drop shows marked differences depending on the mobility of the oil blocks used. Using one mobile oil block the pressure within the fluid column under observation remains constant during the shrinkage time. Depending on the localization of the mobile oil block it can be either close to stop-flow or to free-flow pressures. The data also show that the luminal hydrostatic pressure is perfectly transmitted across a mobile oil block about 5 tubular diameters in length.

It was not possible to demonstrate an effect of intratubular pressure on the reabsorptive rate of fluid and sodium in split drops of technique A and B because of the large scatter of the rate constant in each group compared to their pressure differences. Therefore, the effect was studied over a wider range of pressure changes induced artificially by technique C. Under such conditions it can be shown that net volume flux is significantly altered in that range of intratubular pressures such that an elevation accelerates net reabsorption. A similar effect was reported in the mammalian kidney by Schnermann(17).

The apparent hydraulic conductivity calculated from these data is $0.537 \cdot 10^{-7}$ cm sec⁻¹ (cm H₂O)⁻¹. Several assumptions are required in order to relate this value to the true L_p , i.e., the filtration permeability coefficient. Thus, the relationship between pressure and flow is supposed to be linear, and alterations in active transport(18) or any interference with intraepithelial exchange compartments(3) by the induced pressure changes should be negligible. At present there is no information bearing on this point. The calculated apparent hydraulic conductivity is one order of magnitude higher than the L_{app} obtained from variations of osmotic gradients across the Necturus epithelium(2,19) ($0.33 \cdot 10^{-11}$ cm³ dyne⁻¹ sec⁻¹(19) is equal to $0.32 \cdot 10^{-8}$ cm sec⁻¹ (cm H₂O)⁻¹). It should be noted that, for an L_{app} derived from osmotic pressure differences, the above assumptions concerning no changes in active sodium reabsorption or in epithelial structure might also not be fulfilled. Thus, osmotic gradients might affect the structure and function of the epithelium in a different manner from hydraulic gradients. Accordingly, the existence of a true discrepancy in L_{app} values may be explained in this manner. Despite the fact that the L_{app} may not be a correct estimate of L_p , it represents the global effect of changes of intraluminal pressure on net transepithelial volume flow. As such it can be used to compute the role of any intraluminal pressure change on proximal tubule reabsorption. Provided the effect is linear, it can be estimated that the pressure differences arising from variations in split-drop techniques over the range observed in techniques A and B would result in a change in sodium reabsorption of $0.18 \cdot 10^{-9}$ liter cm⁻² sec⁻¹. The magnitude of this effect corresponds to 11% of the actually observed net reabsorptive volume flow of $1.64 \cdot 10^{-9}$ liter cm⁻² sec⁻¹(3). This fraction could be even more important in the mammalian kidney where the osmotic L_{app} is higher(20).

In conclusion, the presently measured apparent hydraulic conductivity is higher than previous estimates in Necturus proximal tubule, which were based

on osmotic pressure differences. The values reported in this study may be more relevant to predict the effects of intratubular pressure changes on the rate of proximal fluid and sodium reabsorption.

SUMMARY

1. Intrarenal pressures were determined by means of the Wiederhielm technique on *Necturus* kidney and compared to measurements obtained with the Landis technique. Some advantages of the Wiederhielm technique are described.

2. The technique used for the preparation of a split drop affects the intraluminal pressure measured within the droplet.

3. Changes of intraluminal pressure significantly affect the reabsorptive rate in proximal tubules. This effect yields an apparent L_{app} of $0.537 \cdot 10^{-7} \text{ cm sec}^{-1} (\text{cm H}_2\text{O})^{-1}$.

4. It is concluded that intraluminal pressure is a determinant of sodium reabsorption and should be controlled in the split-drop technique.

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