PROCEEDINGS OF THE SOCIETY FOR EXPERIMENTAL BIOLOGY AND MEDICINE 156, 321-325 (1977)

# Glucose Reabsorption in Experimental Glomerulonephritis (39928)

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*Introduction.* Since the classical studies of Shannon *et al.* (1, 2), glucose reabsorption by the mammalian kidney has been used to assess tubular functional capacity (1-3). The characteristic glucose titration curve shows a direct relationship between filtered glucose load and glucose reabsorptive rate until a maximal glucose reabsorptive rate appears ($Tm_G$). A small degree of splay is present under normal conditions. It is now known that the glucose titration curve may be altered by a number of factors. Prominent among these is the volume of the extracellular fluid compartment and its associated sodium excretion rate. Acute expansion of extracellular fluid volume in dogs and rats has been shown to increase the degree of splay in the glucose titration curve and to lower $Tm_G$ (4-7). Glomerulotubular balance for glucose appears to be set at a new level under these conditions. The characteristics of the glucose titration curve may also be modified appreciably in chronic renal disease. While the relationship between $Tm_G$ and glomerular filtration rate has been shown to persist in the chronically diseased kidney, the degree of splay increases markedly at low levels of renal function (8). The question of whether this reflects an increase in the heterogeneity of the nephron population in uremia or whether it reflects the presence of circulating inhibitors of this transport system is still unsettled (6, 9). The present studies were designed to examine the relationship between glucose reabsorption and glomerular filtration rate in rats with experimental glomerulonephritis. The model employed exhibits a wide range of filtration rates in superficial nephrons and thus lends itself to an examination of glucose reabsorption in individual nephrons exposed to varying glucose loads in the same animal (10, 11). Since serum albumin concentration is reduced in such animals, the model also permits an examination of the effects of changes in peritubular colloid osmotic pressure on single nephron glucose reabsorption *in vivo*.

*Materials and methods.* Experimental anti-glomerular basement membrane nephritis was induced in female Sprague-Dawley rats as previously described (12). Experiments were conducted from 3 to 6 weeks after the last antibody injection, at which time the animals weighed between 200 and 280 g. They were allowed an *ad lib.* intake of food and water, but food was withdrawn 14 hr preceding the experiments. Inactin, 100 mg/kg of body weight, was administered, and PE-50 catheters were inserted for infusion and sample collections. A PE-10 catheter was inserted in the left carotid artery and advanced into the abdominal aorta to a site approximately 0.5 cm above the level of the right renal artery. The position of this catheter was verified at the completion of all experiments. The left kidney was prepared for micropuncture as previously described (10). Fluid replacement for surgical losses was given as isotonic saline in a volume equivalent to 2% of body weight. All animals received a priming dose of 150 $\mu$Ci of [$^3$H]inulin in 1 ml of normal saline, and the modified Ringer's sustaining solution contained sufficient para-aminohippurate to provide plasma PAH levels of approximately 1 mg%. This was infused at a rate of 0.013 ml/min and, concurrently, an infusion of 35% glucose containing 20 $\mu$Ci/ml of uniformly labeled [$^{14}$C]glucose was infused at a rate of 0.047 ml/min. After a 90- to 120-min equilibration period, two or three control clearance periods, each 20-30 min in duration, were obtained. Immediately thereafter, the modified Ringer's solution was replaced by a solution of 30% fraction V bovine serum albumin in Ringer's solution of the same composition which was infused at 0.03 ml/





ISSN 0037-9727

min. In some rats, 2 m*M* calcium chloride and 1 m*M* magnesium chloride were added to the albumin solution. Except for a diminished natriuresis during volume expansion, the latter animals exhibited the same findings as the non-calcium-infused group. Therefore, all animals were grouped together for purposes of analysis.

Two to six tubular fluid samples were obtained from late proximal tubules during the control clearance periods. The sites of these collections were carefully mapped, and a re-collection from the same site was made 10–20 min after beginning the albumin infusion. Second re-collections at the same sites were obtained between 50 and 70 min after initiating the albumin infusion. Tubular fluid samples contained at least twice as many counts (measured as counts per minute) as background and were counted for a minimum of 5000 counts.

Glucose concentrations in plasma and urine were measured using a Beckman glucose analyzer, and glucose concentrations in tubular fluid samples were calculated from [$^{14}$C]glucose counts (counts per minute) present. Glucose concentration was also measured microenzymatically in six tubular fluid samples containing [$^{14}$C]-labeled glucose (13). The specific activity of [$^{14}$C]-glucose in these samples was 139 cpm/$\mu$g of glucose. The concurrent specific activity of [$^{14}$C]glucose in plasma was 141.6 cpm/$\mu$g of glucose. The ratio of [$^{14}$C]glucose specific activities in plasma samples to those in urine samples for all experiments was 1.03 ± 0.01.

PAH concentrations in plasma and urine were measured using the method of Smith *et al.* (14), and serum albumin concentration was determined as described by Ness *et al.* (15). Standard statistical procedures were used in analyzing the data (16).

*Results.* Table I summarizes the clearance data obtained before and during the infusion of hyperoncotic albumin. Serum albumin concentration increased significantly soon after beginning the hyperoncotic albumin infusion, but was still lower than values observed in normal rats. Hematocrit values and filtration fraction decreased during this interval but absolute and fractional sodium excretion rates were unchanged. Continuation of the intra-arterial infusion of albumin resulted in a further rise in serum albumin concentration and a rise in fractional sodium excretion. The change in fractional sodium excretion was blunted in those rats which were infused with calcium-containing albumin. Glucose reabsorptive rates increased slightly, but not significantly, and then fell to control levels.

Data from individual nephrons are summarized in Table 2. Superficial nephron filtration rates (SNGFR), which varied from 4.4 nl/min to as high as 38.2 nl/min during control periods, rose by 20% after beginning the albumin infusion and remained elevated throughout the experiment. Proximal tubular glucose reabsorption also rose

TABLE I. CLEARANCE VALUES BEFORE AND DURING ALBUMIN INFUSION ($n$ = 14).[a]

| | Plasma albumin (g%) | Hematocrit (%) | $C_{IN}$ (ml/min) | F.F. | F.E.$_{Na}$ (%) | Plasma glucose (mmole/liter) | $T_G$ ($\mu$mole/min) | $T_G/C_{IN}$ |
|---|---|---|---|---|---|---|---|---|
| Control | 1.77 ± 0.15 | 37.6 ± 2.5 | 0.43 ± 0.09 | 0.26 ± 0.02 | 0.46 ± 0.06 | 55.7 ± 4.17 | 8.1 ± 1.61 | 20.5 ± 2.61 |
| 10–20 min | 2.54 ± 0.25[b] | 30.2 ± 3.0[b] | 0.45 ± 0.10 | 0.23 ± 0.02[b] | 0.52 ± 0.07 | 57.7 ± 5.22 | 11.6 ± 2.56 | 24.1 ± 2.84 |
| 50–70 min | 3.57 ± 0.34[b] | 23.4 ± 2.9[b] | 0.38 ± 0.02[b] | 0.21 ± 0.02[b] | 0.78 ± 0.12[c] | 45.8 ± 4.22 | 6.3 ± 1.0 | 18.1 ± 2.15 |

[a] Values are means ± SE for the left kidney. $n$ = number of animals.
[b] Different from control at $p < 0.01$.
[c] Different from control at $p < 0.05$.

TABLE II. INDIVIDUAL NEPHRON VALUES BEFORE AND DURING ALBUMIN INFUSION ($n$ = 57).[a]

| | SNGFR (nl/min) | TF/$P_{IN}$ | $t_G$ (nmole/min) | $t_G$/SNGFR |
|---|---|---|---|---|
| Control | 17.7 ± 2.85 | 1.45 ± 0.06 | 0.35 ± 0.07 | 0.019 ± 0.001 |
| 10–20 min | 21.3 ± 2.83[b] | 1.70 ± 0.10[c] | 0.61 ± 0.10[c] | 0.028 ± 0.002[c] |
| 50–70 min | 21.0 ± 2.31[b] | 1.42 ± 0.07 | 0.42 ± 0.04 | 0.021 ± 0.001 |

[a] Values are means ± SE. $n$ = number of nephrons.
[b] Different from control at $p < 0.05$.
[c] Different from control at $p < 0.01$.

after beginning the albumin infusion and then fell to control levels. The ratio of glucose load to glucose reabsorptive rate was in excess of 2 in all nephrons sampled, as well as in the kidney as a whole during these collections.

The relationship between renal glucose reabsorptive rates and inulin clearance in all animals before and during hyperoncotic albumin infusion is shown in Fig. 1. Rats with higher glomerular filtration rates had proportionately higher rates of glucose reabsorption under conditions of these experiments. This relationship covered a range of glomerular filtration rates (single kidney) from 0.09 to 0.95 ml/min.

Figure 2 illustrates the relationship between glucose reabsorptive rates and SNGFR in superficial nephrons. There was a direct linear correlation between proximal tubular glucose reabsorptive rates and nephron filtration rates over a wide range of SNGFRs. This relationship did not appear to be influenced by the infusion of hyperoncotic albumin or by the presence of calcium and magnesium in the infusate.

*Discussion.* The application of saturation techniques to the study of renal physiology has provided valuable insight into tubular transport mechanisms (17). In the case of glucose, sequential increments in the amount of glucose filtered are associated

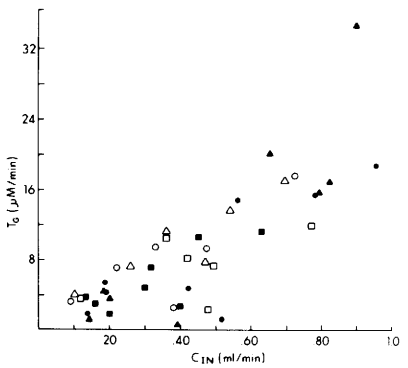


FIG. 1. Relationship between left kidney glucose reabsorption rates and glomerular filtration rates. The open symbols represent experiments in which a calcium-free infusate was used, and the closed symbols represent values from experiments employing calcium-containing infusate. (○, ●) Control; (△, ▲) first re-collection period; (□, ■) second re-collection period. $y = 23.2x - 1.1; r = 0.79$.

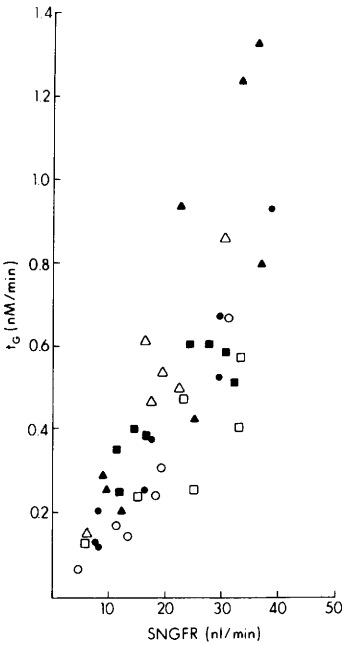


FIG. 2. Relationship between glucose reabsorptive rates and nephron filtration rates in superficial proximal tubules. The symbols refer to the same conditions as in Fig. 1. $y = 0.024x - 0.033; r = 0.84$.

with a progressive rise in renal glucose reabsorption until a constant reabsorptive rate ($Tm_G$) is achieved (1, 2). Additional increases in filtered glucose load are accompanied by proportional increases in urinary glucose excretion. The transition from first-order to zero-order kinetics is not abrupt but, rather, involves a small degree of splay in the glucose titration curve. Acute extracellular fluid volume expansion lowers $Tm_G$ and increases the degree of splay (4–7).

The degree of splay in the glucose titration curve also increases in far-advanced renal disease (8). However, severely damaged kidneys in animals with unilateral renal disease elicit a normal glucose titration curve (9), and patients with glomerular filtration rates as low as 15 ml/min do not exhibit an exaggerated degree of splay (8). The latter two observations are at variance with the view that the increased splay seen in chronic renal disease reflects an increase in the degree of heterogeneity in the functional nephron population (17). An alternative explanation for the changes seen in chronic renal disease stems from studies showing that extracellular fluid volume ex-

pansion produces changes in the glucose titration curve similar to those seen in uremia (6, 9). In both cases, proximal tubular sodium reabsorption is inhibited and a natriuresis accompanies the increased splay. Since a close association between proximal sodium reabsorptive rates and proximal glucose reabsorptive rates is present in normal rats (18), it seems possible that renal glucose transport is linked in some manner to renal sodium transport (4, 5). This thesis is supported by studies in isolated nephron segments and intestinal transport systems which also suggest that transepithelial sodium and glucose transport are interconnected (19, 20).

A significant correlation between inulin clearance and glucose reabsorptive rates is present in normal humans, and animal studies have also shown that renal glucose transport is characterized by glomerulo-tubular balance for this solute (17-23). Glucose reabsorptive rates have also been shown to vary directly with perfusion rates in superficial nephrons and in isolated nephron segments (24, 25). However, the relationship between glucose reabsorption rates and filtration rates in individual nephrons has not been examined over a wide range *in vivo*. The experimental model employed in the present studies exhibits a wide dispersion of nephron filtration rates, and nephrons with varying filtration rates are present in the same kidney, where they are simultaneously exposed to the same plasma composition and external influences. The data depicted in Fig. 2 show that glomerulo-tubular balance for glucose persisted over the entire spectrum of single nephron filtration rates which was present. The level at which glomerulo-tubular balance was set was not fixed, but rather varied under different conditions. Thus, $t_G$/SNGFR ratios averaged 0.019 ± 0.001 under control conditions, increased to 0.028 ± 0.002 shortly after beginning the hyperoncotic infusion period, and then fell to 0.020 ± 0.001 as fractional sodium excretion increased. However, the direct linear relationship shown in Fig. 2 was maintained under all three experimental conditions. The linear regression equations and correlation coefficients were: $y = 0.023x - 0.06$, $r = 0.95$; $y = 0.030x - 0.06$, $r = 0.87$; and $y = 0.014x + 0.12$, $r = 0.79$ for values obtained during control and first, and second re-collection periods, respectively. Juxtamedullary nephrons were not examined in this study, but there is evidence that glomerulo-tubular balance for glucose is also present in this nephron population (22).

The rise in both fractional filtrate reabsorption and glucose reabsorption in superficial nephrons during the first 20 min of hyperoncotic albumin infusion is explicable in terms of an increase in peritubular oncotic pressure. Since the mean glucose load/$t_G$ ratio was greater than 2.5 in the nephrons sampled, it seems likely that glucose reabsorptive sites were saturated at this time. If this inference is correct, the increase in tubular glucose reabsorptive rate would appear to relate to the increase in bulk flow across the tubular epithelium which occurred during this interval. The subsequent fall in glucose reabsorptive rates and fractional filtrate reabsorption in the proximal tubule is attributable to the development of extracellular fluid volume expansion. Back-leak of glucose across the tubular epithelium has been shown in the isolated rabbit tubule preparation (25), but evidence for glucose back-leak was not found during *in vivo* studies in the dog when hyperoncotic albumin was given (4).

*Summary*. The relationship between glomerular filtration rate and tubular glucose reabsorption was examined in rats with experimental glomerulonephritis. Glomerular filtration rates (single kidney) varied from 0.09 to 0.95 ml/min and superficial nephron filtration rates ranged from 4.4 to 38.2 nl/min. A direct linear relationship between filtration rate and maximal glucose reabsorption rates was observed in individual superficial nephrons as well as in the whole kidney. Changes in peritubular oncotic pressure and extracellular fluid volume modified glucose reabsorptive rates, but the linear relationship between filtration rate and glucose reabsorption persisted. These data support the thesis that glomerulo-tubular balance for glucose is a characteristic feature of renal glucose transport.

The studies referred to in this manuscript were supported by Public Health Service Grant No. T501 HL06008. We are also grateful to Dr. Oliver H.

Lowry and Ms. Margaret Philips for performing the microenzymatic glucose determinations.

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Received June 6, 1977. P.S.E.B.M. 1977, Vol. 156.