Kidney pressures after temporary renal artery occlusion in the rat

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Kidney pressures after temporary renal artery occlusion in the rat. Am. J. Physiol. 230(4): 1173-1181. 1976. — Acute kidney failure was produced in the anesthetized rat by 1 h of complete renal artery occlusion. Kidney function was studied either immediately after release of the occlusion or 1 day later using clearance, micropuncture, histological, and nephron dissection techniques. Polyfructosan clearance was decreased to 5% of normal after temporary occlusion. Proximal tubular pressure (PTP) averaged 13-14 mmHg in normal kidneys, 39 mmHg immediately after release of unilateral occlusion, 19 mmHg 1 day after unilateral occlusion, and 25 mmHg 1 day after bilateral occlusion. The increased PTP reduces the glomerular filtration rate (GFR). Glomerular capillary pressure, estimated from the sum of the stop-flow and arterial plasma colloid osmotic pressures, was not decreased after temporary ischemia. Single-nephron GFR, measured without intratubular pressure control, was only slightly below normal 1 day after bilateral occlusion. Most distal tubules from ischemia-damaged kidneys contained hyaline casts. Tubular obstruction is a major factor in this model of acute kidney failure.

METHODS

Experiments were done on Wistar rats of both sexes. Five experimental groups were studied. Group 1 consisted of 24 acute experiments on control (normal) rats. Group 2 consisted of 22 acute experiments in which kidney function was studied "immediately" (i.e., between 0.5 and 4 h) after release of a 1-h period of complete, left renal artery occlusion. Group 3 consisted of 14 experiments on rats subjected to a sham operation 1 day before; this group is a control for the 4th and 5th groups. Group 4 consisted of 18 experiments on rats 1 day after both renal arteries had been occluded for 1 h. Group 5 consisted of seven experiments on rats 1 day after unilateral (left) renal artery occlusion for 1 h.

The following procedure was used in groups 3-5 one day before the micropuncture experiment. Rats were allowed free access to food and water and were anesthetized with sodium pentobarbital, 40 mg/kg body wt ip. Rectal temperature was kept at 37°C with a heated animal board. Clean, but not sterile, techniques were used. The left and right (group 4) or left (group 5) kidneys were exposed by flank incisions, and a small bulldog clamp was placed on the artery. Prior to occlusion, 40 U heparin sodium in 1 ml 0.9% NaCl were injected intravenously. After 60 min, the clamps were removed, and the incisions were closed. The animal was allowed to recover from anesthesia, care being taken to maintain normal body temperature. The rat was placed overnight in a metabolism cage. Water was available but food was withheld. Group 3 animals were treated in the same way as group 4 animals (exposure of both kidneys), but the renal arteries were not occluded. The next day, function of the left kidney was studied using the same procedures for all five groups.

The micropuncture experiments were done as described previously (44). Briefly, rats were anesthetized with Inactin, 100 mg/kg body wt, ip. The animal was placed on a heated micropuncture table, and rectal temperature was kept at 37°C. Surgical procedures included a tracheotomy, cannulation of a femoral artery and vein, placement of the left kidney in a micropuncture cup, and cannulation of the ureter with polyethylene...
The kidney was covered with warm (37°C), light mineral oil. Blood pressure was recorded from the femoral artery with a Statham P23Db pressure transducer and a Sanborn 150 recorder. To estimate GFR, we gave a priming dose of 10% inulin (polyfructosan, PFS, Lavecusan Company, Linz, Austria) in 0.9% NaCl (0.2 ml/100 g body wt), followed by a maintenance infusion of 1–10% PFS in 0.9% NaCl at 0.02 ml/min. Timed urine samples were collected under oil, and urine volumes were estimated by weighing. Arterial blood samples (0.2 ml) were usually taken at the midpoint of urine collection periods and were replaced with an equal volume of 0.9% NaCl. A terminal blood sample (1 ml) was collected in groups 2–5 for measurement of plasma osmolality, urea, and potassium.

The following specific measurements were made.

**Pressure measurements.** PTP was estimated with a Kulite miniature pressure transducer (48) or a servonulling device (23). Pressures were recorded on a Sanborn 150 recorder. The pressure measuring pipettes had a tip diameter of about 8 μm, and were filled with 0.5 g/100 ml lissamine green dye in 0.9% NaCl or 0.5 M NaCl (with the servo system). The zero-pressure reference level was taken with the pipette tip immersed in a thin layer of fluid on the kidney surface. This fluid layer was maintained by periodically placing a few drops of 6 g/100 ml albumin solution on the kidney.

GCP was estimated from the sum of the stop-flow pressure (SFP) and arterial plasma colloid osmotic pressure (COP) (1, 7). To measure SFP, castor oil was injected into a tubule in a retrograde manner until the earliest accessible proximal loop was identified. The pressure in the tubule above the block was determined with a pressure measuring micropipette. COP was estimated by measuring the plasma protein concentration, and by using the Landis-Pappenheimer equation (28): COP = 2.1 C + 0.16 C' + 0.009 C^3, where C is the total protein concentration grams per 100 ml and COP is in millimeters Hg.

After ischemia, proximal tubule lumens were usually dilated, and a marked heterogeneity was apparent. An effort was made to puncture tubules of all sizes, not just the widest ones. Sometimes a few tubules were collapsed; we were not able to obtain reliable pressure measurements in these tubules. An average of eleven (range 4–38) measurements of PTP and three to four (range 1–8) estimates of GCP were done per animal.

**Tubular fluid protein concentration.** In three male rats, immediately after temporary ischemia (group 2), tubule-fluid samples were collected for analysis of total protein concentration. From 0.1 to 0.6 μl of tubule fluid was collected in 12–22 min. Protein was analyzed by the Lowry method (29), using a 0.05-ml cuvette. To test the analysis, 0.2 to 0.6 μl samples of rat plasma diluted 11 times with 0.9% NaCl were treated as unknowns. Recovery of protein averaged 99 ± 18% (SD), n = 20, of the expected recovery.

**Single-nephron glomerular filtration rate (SNGFR).** To estimate SNGFR and fluid reabsorption, we gave four rats of group 4 a priming dose of 0.4 ml 10% PFS-128 μCi [14C]inulin per milliliter 0.9% NaCl solution, followed by a sustaining intravenous infusion of a 10-fold dilution of this solution at 0.02 ml/min. The [14C]inulin (1.65 mCi/g) was obtained from New England Nuclear. Samples of tubule fluid were collected from superficial proximal tubules using sharpened, 8-μm tip diameter pipettes and a mercury leveling bulb. Intratubular pressure was not controlled during these collections. A column of Sudan black-stained mineral oil was maintained distal to the puncture site, and excessive suction was avoided. Collections usually lasted about 10 min. Puncture sites were localized from neoprene casts.

SNGFR was calculated as the product of the tubule fluid-to-plasma inulin concentration ratio (TF/P,,) times the rate of tubule fluid collection. Tubular water reabsorption was calculated as the difference between the rate of filtration of water (SNGFR x 0.93) and the collection rate. The factor 0.93 is a correction for the water content of plasma.

Since both PFS and [14C]inulin were infused in these experiments, a comparison of their renal clearances is possible. In 15 clearance periods, the PFS/[14C]inulin clearance ratio averaged 0.94 ± 0.07 (SD). Thus PFS had a slightly, but significantly (P < 0.01), lower clearance than inulin in these damaged kidneys.

**Histological and microdissection studies.** At the end of many experiments, the kidneys were fixed in vivo by perfusion with 1% glutaraldehyde-Tyrode (30). Histological sections for light microscopy were stained with hematoxylin and eosin or with the periodic acid-Schiff (PAS) technique. Tubules were microdissected as described by Oliver, MacDowell, and Tracy (35). The tubules were usually examined unstained with a Zeiss Universal microscope with Nomarski differential interference contrast. Photographs of representative tubules were taken. To estimate the percentage of the nephron population that contained casts, we examined, from different areas of each kidney, about eight collecting or connecting tubules, each with one to four attached distal tubules.

**Analyses.** Plasma and urine samples were analyzed for PFS by an anthrone method, for osmolality by freezing-point depression (Advanced Instruments osmometer), for urea by a urease-indophenol method, for potassium by atomic-absorption spectrophotometry, and for protein by the Lowry method. Before analysis for protein, urine samples were precipitated with phosphotungstic acid, and the protein precipitate was washed with 100% ethanol. [14C]Inulin in tubule fluid, plasma, and urine samples was assayed in a liquid scintillation counter, as described previously (44).

Data are presented as means ± SD (number of observations in parentheses). Statistical comparisons were made by t tests after a preliminary test for homogeneity of variances.

**RESULTS**

Table 1 summarizes overall functions as measured on the day of micropuncture experiments. Body weights...
TABLE 1. Functions in normal rats and rats subjected to 1 h of renal artery occlusion

<table>
<thead>
<tr>
<th>Group</th>
<th>Body Wt, g</th>
<th>MABP, mmHg</th>
<th>V, µl/min/100 g body wt</th>
<th>C, µl/min/100 g body wt</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>178</td>
<td>105</td>
<td>2.28</td>
<td>462</td>
</tr>
<tr>
<td>± 26</td>
<td>± 12</td>
<td>± 0.93</td>
<td>± 104</td>
<td></td>
</tr>
<tr>
<td>(24)</td>
<td>(21)</td>
<td>(24)</td>
<td>(24)</td>
<td></td>
</tr>
<tr>
<td>2</td>
<td>191</td>
<td>101</td>
<td>5.42</td>
<td>26</td>
</tr>
<tr>
<td>± 39</td>
<td>± 12</td>
<td>± 5.72</td>
<td>± 23</td>
<td></td>
</tr>
<tr>
<td>(22)</td>
<td>(21)</td>
<td>(22)</td>
<td>(22)</td>
<td></td>
</tr>
<tr>
<td>3</td>
<td>196</td>
<td>107</td>
<td>2.61</td>
<td>538</td>
</tr>
<tr>
<td>± 30</td>
<td>± 15</td>
<td>± 1.02</td>
<td>± 90</td>
<td></td>
</tr>
<tr>
<td>(14)</td>
<td>(12)</td>
<td>(11)</td>
<td>(12)</td>
<td></td>
</tr>
<tr>
<td>4</td>
<td>218</td>
<td>103</td>
<td>3.49</td>
<td>35</td>
</tr>
<tr>
<td>± 48</td>
<td>± 8</td>
<td>± 2.29</td>
<td>± 62</td>
<td></td>
</tr>
<tr>
<td>(18)</td>
<td>(16)</td>
<td>(15)</td>
<td>(15)</td>
<td></td>
</tr>
<tr>
<td>5</td>
<td>210</td>
<td>108</td>
<td>0.15</td>
<td>17</td>
</tr>
<tr>
<td>± 26</td>
<td>± 12</td>
<td>± 0.40</td>
<td>± 45</td>
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<tr>
<td>(7)</td>
<td>(7)</td>
<td>(7)</td>
<td>(7)</td>
<td></td>
</tr>
</tbody>
</table>

Values are means ± SD (no. of experiments). Data are for the left kidney only. Abbreviations: MABP, mean arterial blood pressure; V, urine flow rate; C, clearance; PFS, polyfructosan; group 1, control; group 2, shortly after release of 1 h left renal artery occlusion; group 3, sham-operated (control); group 4, 1 day after 1 h bilateral renal artery occlusion; and group 5, 1 day after 1 h unilateral (left) renal artery occlusion.

PTP (Fig. 1 and Table 2) was markedly elevated shortly after release of temporary unilateral renal artery occlusion (group 2). It averaged 39 mmHg, and was much more variable from one tubule to another than in the normal kidney. One day after unilateral occlusion (group 5) PTP averaged 19 mmHg, considerably less than the previous day. One day after bilateral occlusion (group 4), PTP averaged 25 mmHg, nearly twice the normal value.

GCP (Table 2) averaged 56 mmHg in group 1 and 53 mmHg in group 3, the two control groups. Immediately after temporary unilateral artery occlusion (group 2), GCP was significantly increased to 65 mmHg. One day after temporary occlusion, GCP was not significantly different from control (group 3) values. These data suggest that a decrease in GCP is not the explanation for the marked decrease in whole kidney PFS (or inulin) clearance observed after temporary ischemia.

Tubular fluid protein concentration. Proximal tubular fluid protein concentration in three rats immediately after temporary occlusion averaged 82 ± 83 mg/100 ml (n = 14), range, 16-340 mg/100 ml. The final urine protein concentration averaged 328 ± 119 mg/100 ml (n = 7).

Single-nephron glomerular filtration rate. Figure 2 illustrates data from four rats 1 day after bilateral artery occlusion. SNGFR averaged 27 ± 11 nl/min (n = 22), not significantly lower than 30 nl/min we previously found in normal rats (44). Tubule fluid inulin concentration and the volume of fluid reabsorbed increased pro-

and arterial blood pressures in all five groups were similar. Immediately after temporary artery occlusion, an increase in urine flow was sometimes observed. One day after bilateral occlusion, urine flow averaged close to normal, but was more variable. One day after unilateral occlusion, there was practically no urine flow from the damaged kidney in six out of seven experiments. In these six experiments, flow was so slow that urine did not reach the end of a 3-cm-long PE-50 ureteral cannula in a 3 to 4-h period. Polyfructosan clearance (C_{PFS}), normally equal to GFR, was decreased to about 5% of normal after temporary artery occlusion (groups 2, 4, and 5).

One day after bilateral artery occlusion, plasma osmolality was 337 ± 25 mosmol/kg H_2O (n = 15); plasma urea, 43.1 ± 16.0 mM (n = 17); and plasma potassium, 8.36 ± 2.39 meq/liter (n = 9). After unilateral artery occlusion (group 5), the corresponding values were 305 ± 6 (n = 7), 8.0 ± 5.1 (n = 7), and 4.86 ± 0.68 (n = 7); and in the sham-operated group (group 3), 289 ± 4 (n = 12); 4.8 ± 0.9 (n = 14); and 4.88 ± 0.54 (n = 6). Large, significant (P < .01), increases in plasma osmolality, urea, and potassium were induced by bilateral occlusion. Modest, but significant (P < .01), increases in plasma osmolality and urea were produced by unilateral occlusion, but plasma potassium was normal.

Pressure measurements. Since two different pressure measuring techniques were used, we compared results obtained with the Kulite transducer with those obtained with the servo-null device. In normal kidneys (group 1), with the Kulite transducer PTP averaged 13.3 ± 2.0 mmHg (n = 181); SFP, 40.5 ± 5.7 mmHg (n = 24); and GCP, 55.5 ± 5.2 mmHg (n = 24). With the servo-null system, PTP averaged 13.8 ± 1.8 mmHg (n = 29); SFP, 40.6 ± 6.6 mmHg (n = 13); and GCP, 56.6 ± 7.8 mmHg (n = 13). These results indicate that both techniques give identical values.

FIG. 1. Histograms of proximal tubular hydrostatic pressure.
Temporary complete renal ischemia has often been used to induce acute renal failure in experimental animals (4, 11, 15-17, 21, 26, 27, 35, 38, 40, 42-44, 46, 47). Interruption of the renal circulation is an obvious ischemic insult, and if prolonged, severely damages the renal parenchyma. In the rat, 1 h of temporary renal artery occlusion results in extensive tubular cell necrosis (27, 37, 38) and a mortality of 13-25% (27, 40). Since an ischemic insult is generally considered to be the leading cause of acute renal failure in man (32, 35, 46), the present study may be relevant to understanding the human disease.

**Significant differences between unilateral and bilateral cases.**

**Histological and microdissection studies.** Figure 3 shows histological sections from kidneys fixed in vivo at the end of some experiments. Sections from kidneys obtained 3-4 h after release of the occlusion typically showed normal looking glomeruli and degenerative tubular cell changes. Proximal tubules were dilated, and distal tubules frequently contained an eosinophilic material. One day after temporary artery occlusion, extensive cell necrosis was present in the cortex and outer medulla, and many medullary tubules were filled with eosinophilic casts.

Microdissected tubules from normal and ischemia-damaged kidneys are illustrated in Fig. 4. In the normal kidney, the tubule lumens were open, probably owing to rapid in vivo fixation (30), and the lumens contained no debris. In damaged kidneys, the distal nephron was often completely filled with a hyaline cast of orange or golden colored material. This material corresponds to the eosinophilic casts seen in the histological sections.

We attempted to estimate what fraction of the nephron population contained casts. Table 3 summarizes our findings. Only distal tubules were systematically examined for several reasons: a) it was not possible to dissect out entire nephrons, b) the proximal tubule epithelium was too thick and pigmented to get a clear view of its lumen, and c) histological studies suggested that casts were present mainly in the distal nephron. In group 2, 92 ± 7% (n = 7 kidneys) of the distal tubules contained casts, and in group 4, 84 ± 14% (n = 6). In normal kidneys (often the right, untouched kidney from a group 2 or group 5 experiment), 6 ± 8% (n = 7) of the distal tubules contained casts.

**DISCUSSION**

Temporary complete renal ischemia has often been used to induce acute renal failure in experimental animals (4, 11, 15-17, 21, 26, 27, 35, 38, 40, 42-44, 46, 47). Interruption of the renal circulation is an obvious ischemic insult, and if prolonged, severely damages the renal parenchyma. In the rat, 1 h of temporary renal artery occlusion results in extensive tubular cell necrosis (27, 37, 38) and a mortality of 13-25% (27, 40). Since an ischemic insult is generally considered to be the leading cause of acute renal failure in man (32, 35, 46), the present study may be relevant to understanding the human disease.
renal artery occlusion exist. After bilateral occlusion, a severe uremia develops (e.g., plasma urea and potassium levels are elevated), but this does not occur after unilateral occlusion. For this reason, bilateral artery occlusion is probably the better model for clinical acute kidney failure.

Other differences between these two models are observed. PTP in damaged kidneys was higher one day after bilateral occlusion than after unilateral occlusion (Table 2). After bilateral occlusion, urine flow was always measurable and averaged close to normal; by contrast, 1 day after unilateral occlusion there was no measurable urine flow from the damaged kidney in six of seven animals (Table 1). Similar differences in PTP and in urine flow have been found after chronic bilateral and unilateral ureteral obstruction (25). Thus, the function of a damaged kidney depends on whether the opposite kidney has also been insulted.

The central problem in acute kidney failure is the apparent decrease in whole-kidney GFR. One hour of renal artery occlusion reduced the whole-kidney PFS or inulin clearance to 5% of normal (Table 1). This drastic reduction may be due to a change in the physical conditions which govern filtration across the glomerular capillary membrane and/or abnormal tubular leakiness to the substances used to estimate GFR (PFS, inulin).

**Proximal tubular pressure**: PTP was higher than normal after temporary artery occlusion (Table 2 and Fig. 1). This increase would act to decrease GFR.

Several factors might produce an increase in PTP: a) an increase in SNGFR, b) a decrease in tubular water reabsorption, and c) an increase in downstream flow resistance. An increase in SNGFR does not contribute to the rise in PTP, for SNGFR appeared reduced immediately after temporary artery occlusion (44) or 1 day after bilateral artery occlusion. A decrease in tubular fluid reabsorption may contribute somewhat to the rise in PTP, since fluid reabsorption was modestly decreased after temporary ischemia (Fig. 2) (44). If this were the sole explanation for the increase in PTP, one would predict, as a consequence of decreased tubular fluid reabsorption, a striking diuresis. This was not observed. The major factor causing the rise in PTP is an increase in downstream flow resistance, i.e. tubular obstruction.

It is difficult to state precisely how much of the decrease in GFR is due to the rise in PTP. Davis, Schne-
mann, and Horster (14) reported that in normal kidneys an increase in PTP of 4.5 mmHg would decrease SNGFR by 30%. Andreucci et al. (2) found that an increase in PTP of 8 mmHg, produced by elevated ureteral pressure, would decrease whole-kidney or single-nephron GFR by about 50%. Thus small increases in PTP can lead to substantial decreases in GFR. In animals 1 day after bilateral artery occlusion, in which PTP was 25 mmHg compared to a normal 13 or 14 mmHg (Table 21, it is likely that the elevated tubule pressure contributed in a major way to the fall in GFR.

**Glomerular capillary pressure.** GCP is the driving pressure responsible for glomerular filtration. Its magnitude is of obvious importance in determining GFR. We estimated GCP indirectly, from the sum of SFP plus COP. Blantz et al. (7) first showed that this method of estimation of GCP gave values identical to direct measurements in the same nephron obtained by glomerular puncture. In hydropenic rats, however, the indirectly determined values are 4–8 mmHg higher than directly measured values when these are determined in separate nephrons (6, 7). The specific reason for this disparity is not known. Our indirect estimates of GCP in normal Wistar rats (56 mmHg in group 1 and 53 mmHg in group 3) are higher than values measured directly in Munich-Wistar rats by Brenner et al. (8)–mean, 44 mmHg; and by Blantz et al. (7)–mean, 47 mmHg. Our higher values may reflect differences in methodology, rat strain, and condition of the animals.

The assumptions and limitations of the indirect estimation of GCP have been discussed before (1, 7). In the ischemia-damaged kidney, a potential pitfall is the assumption that the protein concentration (and hence colloid osmotic pressure) in Bowman’s capsule is negligible. Cain and Fazekas (11), on the basis of histological studies, suggested that there was a transient increase in glomerular permeability to plasma proteins, most significant in the first 3 h after release of temporary renal ischemia. We measured the protein concentration of proximal tubular fluid at this time, and found it averaged 82 mg/100 ml. This value is higher than the 20 mg/100 ml protein concentration measured by Van Liew et al. (45) with the Lowry method in normal rats. If we assume that the measured tubular fluid proteins are plasma proteins and that they are in the same proportions as in plasma, then the calculated colloid osmotic pressure after temporary ischemia, 0.17 ± 0.18 mmHg (n = 14), is negligible.
A) Normal Kidneys  
<table>
<thead>
<tr>
<th>No. Distal Tubules Examined</th>
<th>No. Distal Tubules with Casts</th>
<th>% Distal Tubules with Casts</th>
<th>Cinf, 100 g body wt</th>
</tr>
</thead>
<tbody>
<tr>
<td>3/6/73</td>
<td>13</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>1/7/74*</td>
<td>13</td>
<td>1</td>
<td>8</td>
</tr>
<tr>
<td>2/18/74*</td>
<td>15</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>2/28/74*</td>
<td>16</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>3/5/74*</td>
<td>10</td>
<td>1</td>
<td>10</td>
</tr>
<tr>
<td>4/29/74*</td>
<td>9</td>
<td>2</td>
<td>22</td>
</tr>
<tr>
<td>4/30/74*</td>
<td>14</td>
<td>0</td>
<td>0</td>
</tr>
</tbody>
</table>

Mean ± 1 SD 6 ± 8

B) Shortly after 1 h renal artery occlusion (group 2)  
| 1/11/73                     | 7                             | 6                           | 86                  |
| 1/16/73*                    | 13                            | 13                          | 100                 |
| 1/25/73                     | 12                            | 12                          | 100                 |
| 2/1/73                      | 11                            | 10                          | 91                  |
| 2/12/73                     | 27                            | 26                          | 96                  |
| 4/29/74*                    | 16                            | 13                          | 81                  |
| 4/30/74*                    | 10                            | 9                           | 90                  |

Mean ± 1 SD 92 ± 7 32 ± 30

C) One day after 1 h bilateral renal artery occlusion (group 4)  
| 3/1/73                      | 16                            | 15                          | 94                  |
| 3/20/73                     | 14                            | 8                           | 57                  |
| 3/24/73                     | 14                            | 13                          | 93                  |
| 3/30/73                     | 21                            | 17                          | 81                  |
| 4/5/73                      | 17                            | 15                          | 88                  |
| 7/20/73*                    | 18                            | 16                          | 89                  |

Mean ± 1 SD 84 ± 14 15 ± 11

The normal kidneys were usually right kidneys from a unilateral (left) renal artery occlusion experiment, and no measurements of polyfructosan clearance (Cinf) were made. *Tubules were microdissected by K.L.S. G.A.T. evaluated the tubules and did not know whether they were from normal or damaged kidneys.

Our estimates of GCP suggest that decreases in this parameter are not responsible for the marked decrease in GFR after temporary ischemia. Thus, in groups 4 and 5, GCP was not significantly different from normal (Table 2). GCP was actually higher than normal immediately after temporary ischemia (Table 2). The reason for this increase is not known.

Single-nephron glomerular filtration rate. SNGFR after bilateral artery occlusion was only slightly below normal. This is consistent with the finding that GCP was close to normal (Table 2). Furthermore, if SNGFR were drastically reduced after temporary ischemia, then high PTP values would not have been observed.

The SNGFR measured should be viewed as a "potential" rate of filtration (44). During the collection of fluid for this measurement, we could not avoid a fall in PTP. In effect, we cancelled out distal nephron obstruction by venting the nephrin upstream. The measured SNGFR probably approximates the rate of filtration that would occur if PTP were not elevated due to tubular obstruction.

Tubular casts. After temporary ischemia we observed casts in histological slides and microdissected tubules (Figs. 3 and 4). It seems likely that the casts obstruct the flow of urine in vivo and thereby increase proximal tubular pressure. On the basis of observations on fixed tissue, it is, however, not possible to prove this point. In agreement with others (13, 34, 35), the casts were found especially in the distal, connecting, and collecting tubules. Hence they are downstream to the site of our pressure measurements. The casts probably consist mainly of protein and debris from tubular epithelial cells damaged by 1 h of complete ischemia (26, 37, 38). The casts stained strongly with the PAS technique, suggesting a glycopolypeptide component. Agglutinated masses of Tamm-Horsfall protein may be present (36).

It is difficult to appreciate the full significance of intratubular casts from an ordinary histological slide (34). With the microdissection technique, we saw that many distal tubule lumens were completely filled with a solid-looking, hyaline material. We estimated that roughly 90% of nephrons from damaged kidneys contained casts (Table 3).

This estimate of nephron-cast frequency is, of necessity, crude. First, it is based only on distal tubules. We did not examine proximal or collecting tubules for casts. Potentially, a cast that obstructs a single large collecting duct could block the outflow from several thousand nephrons. Second, we observed casts in at most 90 tubules from normal kidneys (Table 3). This is surprising. It may reflect fixation artifacts or errors in judgment. It was often difficult to evaluate the condition of the tubule lumen, because we had to look through the tubule epithelium. Despite shortcomings, our results suggest that after temporary ischemia the majority of nephrons are blocked by casts.

Comparison to other studies. The mechanisms responsible for acute kidney failure are still controversial. Three mechanisms are most often discussed: a) a reduction in GFR due to a decrease in renal blood flow, b) back leakage of filtrate through an abnormally permeable tubule wall, and c) tubular obstruction (18, 32). Most studies suggest that alterations in renal hemodynamics are responsible for the near cessation of GFR (18). However, acute kidney failure may be caused by many different clinical and experimental circumstances, and it seems unlikely that it can be explained by a single pathophysiologic abnormality. In different models and in different patients, indeed at different stages of the disease in a single individual, all three mechanisms may contribute to renal insufficiency in varying degrees. We will highlight the role of tubular obstruction, and its consequence, increased PTP, since this contributes in a major way to renal failure in our study.

The kidneys of patients dying from acute kidney failure are typically swollen and firm. This has led to the hypothesis that intrarenal pressure is increased (31). Intrarenal pressure measured in patients with the wedged venous catheter technique was, however, the same as in normal subjects (9). De Wardener (15) first attempted to measure intrarenal pressure in experimental ischemic renal failure. He used a relatively crude technique (needle pressure) in the dog, and found no increase. Our finding that PTP is elevated after temporary ischemia in the rat is in good agreement with studies by Walther and Schocpepe (47). They reported that proximal tubule lumens were dilated and pressure
was increased to 21 mmHg 3-4 days after 1 h of bilateral ischemia. Arendshorst, Finn, and Gottschalk (4) have also found a marked increase in PTP shortly after ischemia. Arendshorst, Finn, and Gottschalk (4) have

We have emphasized the role of obstruction, but other factors contribute to renal insufficiency in ischemic failure. Renal blood flow was found to be about 10% below normal 3 days after 1 h of unilateral artery occlusion (16, 42). We observed in this and in a previous study (44) that SNGFR was below normal after temporary ischemia. The kidney tubules appear to be abnormally leaky after 1 h of temporary ischemia, since even a molecule as large as inulin can pass through the tubule wall (17, 44). To the extent that inulin or PFS is reabsorbed, the whole-kidney clearance of these substances will underestimate the actual GFR. Obstruction and leakiness often occur together, since both reflect damage to the tubular epithelium (5, 44). Several, often interrelated, mechanisms contribute to insufficiency in ischemia-induced renal failure.

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