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Transient urethral obstruction predisposes to ascending pyelonephritis and tubulo-interstitial disease: studies in rats

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Abstract Chronic tubulo-interstitial disease, an important cause of end-stage renal disease, often results from the combined effects of a disturbed urinary outflow tract and urinary tract infection. Acute unilateral ureteral obstruction in rats rapidly induces foci of medullary necrosis, confined to the region of the papilla and fornices. This injury may provide a nidus for bacterial invasion and may invoke reactive and regenerative changes, ultimately leading to chronic pyelonephritis and tubulo-interstitial nephropathy. To explore this possibility, adult rats underwent renal morphological evaluation 2–7 days following transient 24-h unilateral ureteral obstruction. In some experiments the bladder was inoculated with bacteria (10^8–10^9 cfu/ml *Escherichia coli* in 0.5 ml) after release of ureteral obstruction, with subsequent cultures obtained from the pelvis of both kidneys and from the urinary bladder. Morphologic evaluation of perfusion-fixed kidneys, 2–7 days after the release of 24-h ureteral obstruction disclosed papillary necrosis, urothelial proliferation, marked inner-stripe interstitial expansion, and fibrosis and proximal tubular (S_3) dilatation. The lateral (perihilar region) was predominantly affected, with lesions spreading from the fornices. There was some progression of interstitial fibrosis during the postobstructive time course or following more prolonged ureteral obstruction. By contrast, infection hardly contributed to the tubulointerstitial changes. In rats subjected to infection, cultures were positive in all 15 postobstructive kidneys, as opposed to five contralateral kidneys ($P < 0.0001$). Viable counts from the postobstructive kidney were also higher than those from the contralateral side (79,000 ± 12,000 vs 2900 ± 1600 cfu/ml, mean ± SEM, $P < 0.0001$), and were comparable to those obtained from the bladder (77,000 ± 13,000 cfu/ml). We conclude that transient ureteral obstruction predisposes to ascending pyelonephritis and to tubulointerstitial disease. This vulnerability may relate to altered urodynamics and medullary tissue destruction.

Key words Pyelonephritis · Renal medulla · Rat · Morphology · Urethral obstruction · Culture · *Escherichia coli*

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Introduction

Chronic tubulo-interstitial nephropathy is a common cause of end-stage renal disease. Anatomic abnormalities of the urinary tract, calculus disease, analgesic abuse, and diabetes are involved in the majority of cases [9], with urinary tract infection playing an important coexisting role in up to one-third of patients [38].

We have recently noted that acute transient ureteral obstruction results in early medullary damage confined to the papilla and fornices [19], identical to the rat injury pattern of reflux [11, 17, 39]. This lesion is rapidly followed by reactive and regenerative changes and may provide a nidus for bacterial invasion and pyelonephritis. The current studies were designed to evaluate the propensity of such kidneys to develop

ascending pyelonephritis and early tubulo-interstitial disease.

Materials and methods

Animals

A total of 167 male Sprague Dawley rats (343 ± 5 g weight, X ± SEM) were used for all experiments, fed on regular chow and drinking water ad libitum. Experiments were conducted following the principles of laboratory animal care (NIH publication No. 86–23, revised 1984). Under ether anesthesia the left ureter was exposed through a mid-abdominal incision and occluded by a silk suture thread or a small venous clip, placed 1–2 cm below the uretero-pelvic junction. The abdomen was closed in two layers. After 24 h, again under ether anesthesia, the suture or clip was removed and urinary flow was confirmed visually. The rats were allowed to recover for the subsequent 2–3 days ($n = 76$) or 5–7 days ($n = 79$) and were finally killed for morphologic evaluation. Rats were anesthetized with Inactin (100 mg/kg) and both kidneys were perfusion-fixed with glutaraldehyde as previously reported through a cannula inserted into the aorta [19]. This in vivo perfusion fixation technique facilitates the detection of the early outer medullary interstitial changes detailed below.

Overall, 8% of transiently obstructed kidneys were excluded, being severely hydronephrotic or pyonephrotic with renal parenchymal loss in control or infected rats, respectively. With the concern of persistent partial ureteral obstruction following the release of ureteral ligature, two control groups ($n = 6$ per group) were included: (1) rats in which the left ureters were kept continuously clamped for 3 successive days before removal for morphology, and (2) rats in which following the 24-h obstruction period the left ureter was totally severed above the ligature and allowed to drain into the peritoneum. These two control groups were designed to represent morphologically total continuous obstruction and confirmed ureteral patency, respectively.

Bacteriologic studies

The organism used was a strain of *E. coli*, recovered from the urine of a patient with acute pyelonephritis. Urinary tract inoculation with bacteria took place in 53 rats immediately after the release of ureteral obstruction: the bacterial inoculum (10^8–10^9 organisms, suspended in 0.5 ml saline) was injected directly into an empty bladder through a 25-gauge needle. At the conclusion of the experiment, 3–6 days after bacterial inoculation, the rats were killed and the kidney perfusion fixed for morphology as detailed above. In 15 rats urine was aspirated from the bladder and both renal pelvises before the rats were killed, and samples (10 μl) were seeded for bacterial culture using standard laboratory techniques.

Morphological evaluation

Perfusion-fixed kidneys were postfixed in buffered 2% OsO_4, dehydrated, and embedded in an araldite-EM bed 812 mixture. Large sections were cut, perpendicular to the renal capsule, containing cortex and medulla, including the papilla. One-micron sections, stained with 1% methylene blue, were analyzed in a blinded fashion for morphologic alterations, comparing the transiently occluded left kidney with the intact contralateral one.

Statistics

Values of bacteriologic studies are presented as the SEM. Chi square test and one-way analysis of variance with Neuman-Keuls post hoc test were applied for data evaluation, with statistical significance set at $P < 0.05$.

Results

Morphologic evaluation

Noninfected rats

The right contralateral kidneys were intact, both macroscopically and microscopically. By contrast, the left kidneys were consistently enlarged with mild to moderate hydronephrosis (enlarged urinary space without evident parenchymal loss). Macroscopically, 3 days after the release of ureteral obstruction, infarction, associated with hemorrhage and congestion (perhaps reflecting reduced blood flow and stasis [18] in the perfusion-fixed kidney) were often noted in the fornix region (Fig. 1a), extending into the outer medulla, medullary rays, and perihilar cortex. Hemorrhage was also frequently noted in the papilla, with occasional loss of the papillary tip. Microscopically, there was primarily medullary-centered injury and repair; uroepithelial proliferation was also noted (Fig. 1a). Papillary necrosis was often present, with variable stages of healing. A distinct interstitial fibrosis took place in the inner stripe of the outer medulla, predominantly centered in the interbundle zones (Fig. 1b). Tubular dilatation was most evident at the S_3 segments of the proximal tubules in the outer stripe of the outer medulla (Fig. 1c) and in medullary rays. Collecting ducts were dilated as well, though inconsistently, with occasional casts. These changes were somewhat more consistent in the control group with 3 days of continuous obstruction (Fig. 2). The second control group with the left ureter cut above the obstruction and drained into the peritoneum did not differ morphologically from the corresponding group of released obstruction (Fig. 1). At 5–7 days after the release of 24-h ureteral obstruction, frank macroscopic hemorrhage was no longer evident, but congestion was still occasionally noted. Microscopically, interstitial fibrosis at the inner stripe seemed more pronounced.

Infected animals

Again the right kidneys were grossly intact. Four out of 53 postobstructed left kidneys were transformed into pyonephrosis, with total loss of the renal parenchyma. The rest of the postobstructed kidneys were enlarged with what macroscopically appeared as pronounced medullary congestion, both at 3 days and 5–6 days after the release of ureteral obstruction and bacterial inoculation. Microscopically, acute inflammatory response was noted, particularly at the submucosa in the forniceal region, and uroepithelial proliferation was most pronounced (Fig. 3). Otherwise, in general the tubulointerstitial changes were comparable to the corresponding groups without infection.

Comparisons of morphological features in the various experimental groups are schematically presented in Table 1.

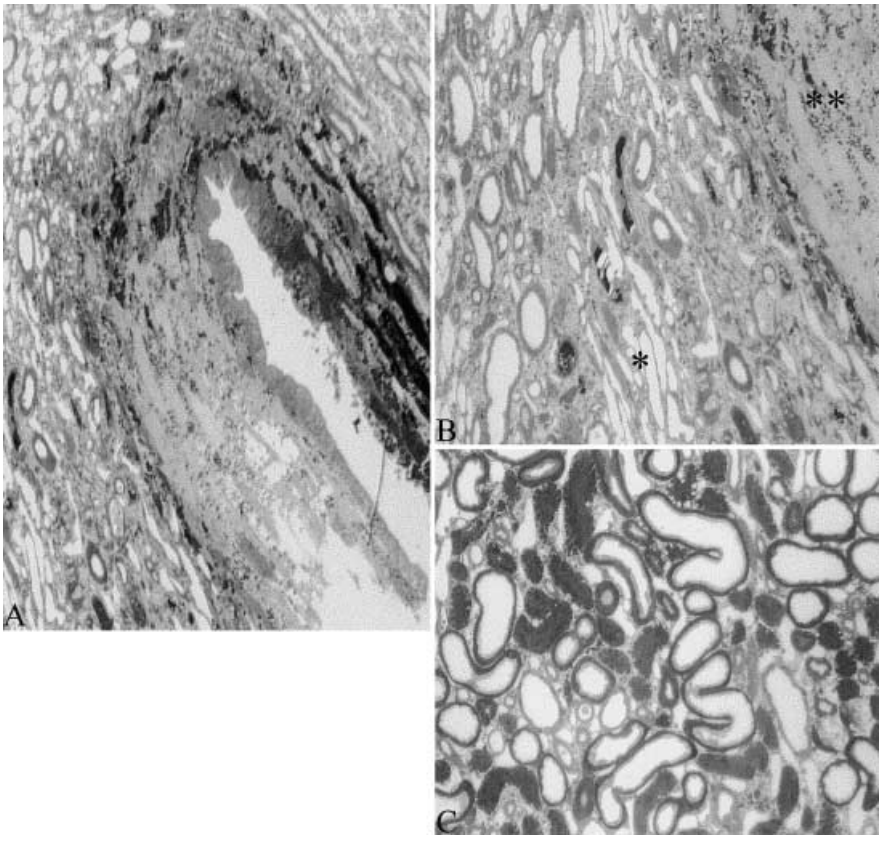

Fig. 1A, B Morphology of a rat kidney 3 days after a unilateral transient 24-h ureteral obstruction. Relief of obstruction was confirmed by ureteral transection above the ligature and drainage into the peritoneum. In **A**, the inner stripe bordering the fornix can be seen to be extensively infarcted. The urothelial lining is thickened. In **B**, adjacent to the infarcted area (**), the interbundle zones show fibrosis, but the vasa recta (*) are unchanged. The straight (S_3) segments of the proximal tubules in the outer stripe of the outer medulla are dilated (**C**). ×135, ×180, ×220

Microbiologic studies

Cultures were positive in all 15 postobstructive kidneys, as opposed to 5 out of 15 contralateral kidneys ($P < 0.0001$). As detailed in Fig. 4, viable counts from the postobstructive kidney were also higher than those from the contralateral side (79,000 ± 12,000 vs 2900 ± 1600 cfu/ml, mean ± SEM, $P < 0.0001$), and were comparable to those obtained from the bladder (77,000 ± 13,000 cfu/ml). The infection rate and its intensity were comparable at 3 and 6 days after inoculation. Cultures obtained from a control group of rats, administered intravesically with sterile saline following the release of ureteral obstruction, were all negative.

Discussion

The hallmark of chronic tubulointerstitial nephropathy is interstitial expansion with variable inflammatory reaction, progressive accumulation of fibroblasts and extracellular matrix, and concomitant tubular atrophy and nephron loss. Tubulointerstitial nephropathy, predominantly affecting the medulla, is mainly associated with reflux or renal outflow obstruction, a large host of drugs, toxins, and metabolic products, infection, or with their combination [9]. In most cases, cortical involvement occurs later [34], in part due to cortical tubular apoptosis, oxidative damage, and down-regulation of epidermal growth factor (EGF) [2, 28, 41]. More focal cortical scars may also develop from renal cortical vascular compromise due to dense surrounding scar tissue at the cortico-medullary junction [20, 39].

The current series describes a convenient rat model of early tubulointerstitial nephropathy that may evolve into the advanced diffuse or focal scarring, well described in the classical works by Cotran [20] in piglets with ureterovesical reflux and in other species with prolonged ureteral obstruction [34]. It indicates that *transient* ureteral obstruction with or without bacterial infection also predisposes to tubulointerstitial disease. Interstitial expansion with accumulation of intracellular matrix and fibroblasts was already noted 2–3 days following the release of 24-h ureteral obstruction, much earlier than previously reported [42], and has been predominantly noted in the interbundle zone of the inner stripe of the outer medulla, especially at its lateral perihilar aspects. Interstitial expansion increased somewhat at 5–7 days following obstruction and was more pronounced in kidneys subjected to more prolonged ureteral obstruction. At the papillary tip and in the inner medulla adjacent to the fornices, interstitial expansion was part of the reparative process, evolved from renal parenchymal destruction and hemorrhage, with subsequent inflammatory reaction. By contrast, in the mid-inner stripe, interstitial expansion occurred without evident local tissue injury. As opposed to the advanced injury pattern in control kidneys subjected to 3-day ureteral obstruction, renal morphology following transection of the ureter after 24-h obstruction was not

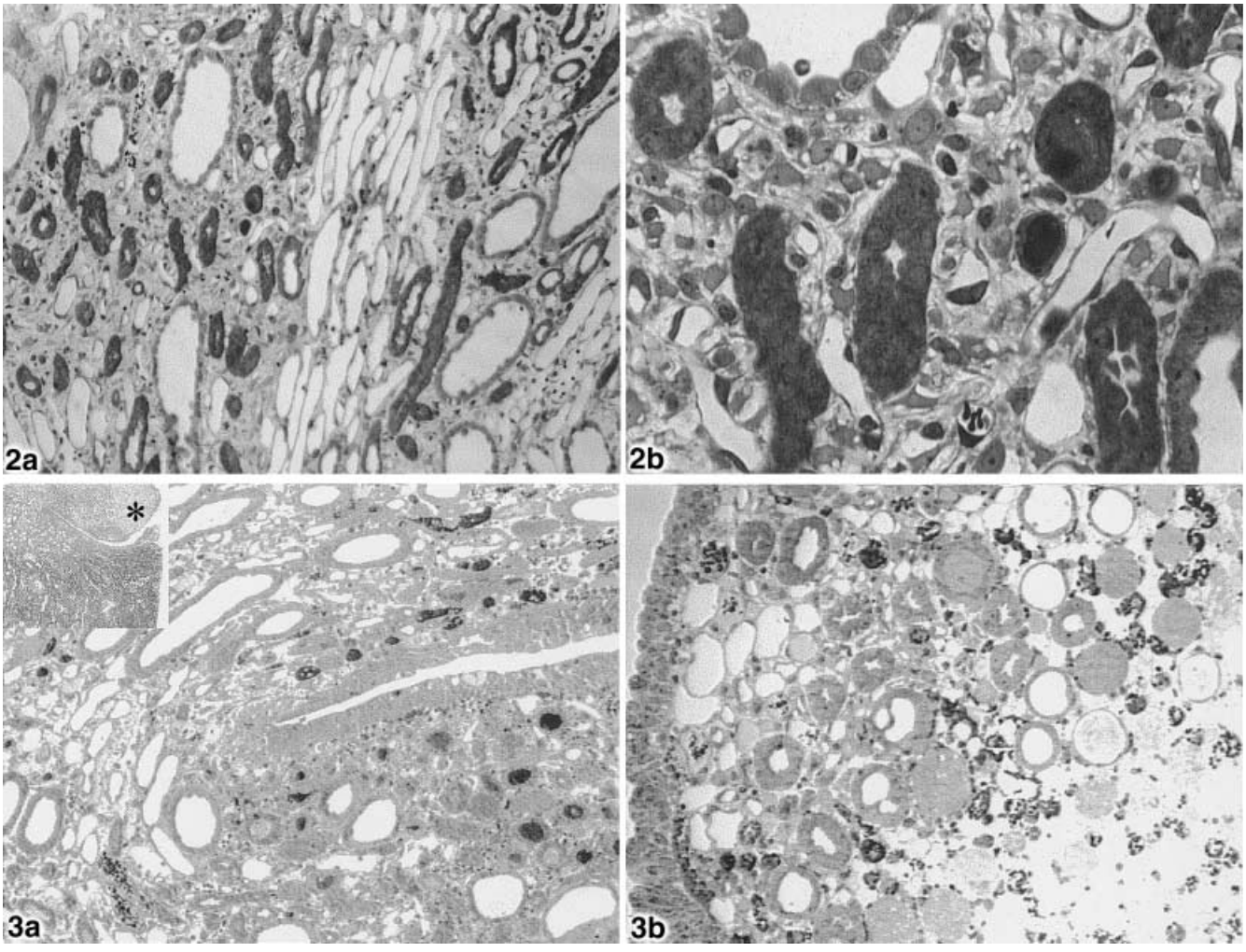

Fig. 2a, b Renal morphology following 3 days ongoing ureteral obstruction. In the inner stripe **a**, the interbundle areas are extensively fibrotic, but the vasa recta show no abnormalities. The fibroblastic proliferation can be seen at higher power, **b**, and is considerably more evident than after only 24 h of obstruction, shown in Fig. 1. ×180, ×600

Fig. 3a, b Renal morphology 3 days after a transient 24-h unilateral ureteral obstruction. Intravesical inoculation by *E. coli* (10^8–10^9 organisms, suspended in 0.5 ml saline) took place immediately following the release of ureteral obstruction. The fornical area (**a**) is inflamed and the covering urothelium shows proliferative changes. Inner stripe infarction can be seen at low power (*inset**) and is more evident at high power (**b**; *lower right*). ×15 (*inset*), ×90, ×180

different from the experiments with simple removal of the ureteral clamp. This confirms that the described morphologic findings are related to transient, rather than prolonged partial ureteral obstruction.

Various mediators are involved in medullary tubulointerstitial fibrosis, including the induction and release of angiotensin II, transforming growth factor (TGF)-β, platelet-derived growth factor (PDGF), and osteopontin, as well as the down-regulation of epidermal growth factor (EGF) and catalase with enhanced oxidative stress, the recruitment of monocytes/macrophages, and myofibroblast transformation with collagen type IV accumulation [5–8, 10, 22, 25, 27–29, 42]. Interestingly, osteopontin, a major participant in this sequence of events, is principally produced by medullary thick ascending limbs [21], around which the early interstitial changes take place. In our short-term experiments, tubulointerstitial changes were restricted to the renal medulla. Prolonged total ureteral obstruction leads to a more delayed but extended interstitial fibrosis that may affect the cortex as well [34].

Experimental pyelonephritis has been reproduced by direct inoculation of the kidney [12, 33, 35], through hematogenous spread (where infection occurs predominantly [3, 14, 15] but not always [16] on the side with total or partial ureteral obstruction) or by retrograde inoculation. This latter method may better represent the progression of ascending pyelonephritis. In such experiments bacteria have been administered through the urethra or directly intravesically, with variable rates of ascending pyelonephritis, depending on bacterial species and virulence, and on the co-insertion of foreign bodies into the bladder [3]. Vesicoureteral reflux can be easily reproduced in rats simply by a relatively large volume (0.5–1.5 ml) inserted into the bladder [17, 30, 36] or by increasing hydrostatic pressure in the bladder. Accord-

Table 1 Schematic presentation of structural changes in obstructed kidneys with or without ascending urinary tract infection. Transient ureteral obstruction lasted for 24 h in all but one control group obstructed for 3 consecutive days. Morphologic evaluation took place 2–3 or 5–7 days following the release of obstruction. Findings in an additional group killed 3 days after transection and intraperitoneal drainage of 24-h obstructed ureter were similar to those noted in the corresponding group with 24-h obstruction and 2–3 days release. *OM* outer medulla

Morphologic features	24-h obstruction, 2–3 days release	Same + infection	24-h obstruction, 5–7 days release	Same + infection	72-h obstruction
Fornical necrosis/hemorrhage	+ + +	+ + +	+	+ +	+ + +
Papillary necrosis/hemorrhage	+ +	+ +	+ +	+ +	+ + +
Urothelial proliferation	+	+ +	+	+ + +	+ +
OM interstitial expansion	+	+	+ +	+ +	+ + +
Proximal tubular dilatation	+	+	±	±	+ +
Inflammatory response in fornix region	±	+	±	+ +	±

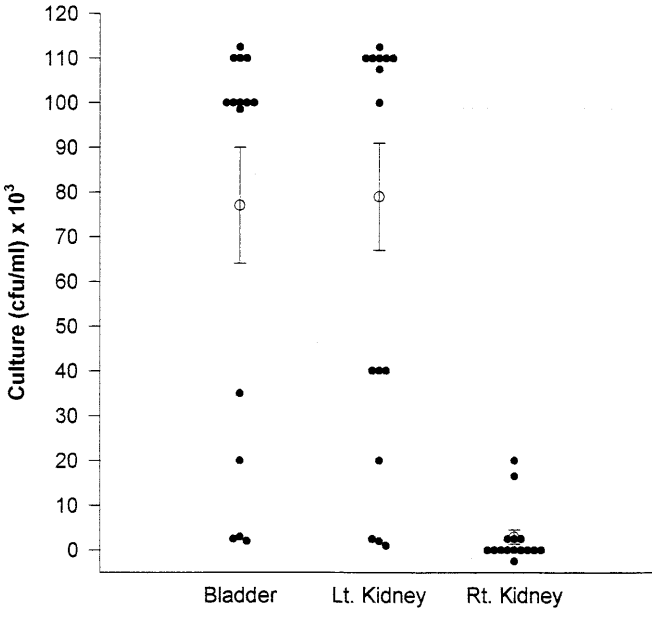

Fig. 4 Urine culture obtained from the bladder and renal pelvises 3–6 days after direct intravesical inoculation with *E. coli* (10^8–10^9 bacteria, suspended in 0.5 ml saline). Bacterial inoculation took place immediately following the release of 24-h left ureteral obstruction. The incidence (100%) and extent of left ascending infection is comparable to that in the bladder, as opposed to low-grade infection affecting only one-third of the contralateral kidneys ($P < 0.0001$, ANOVA)

ingly, increasing reflux by a large volume of bacterial suspension inserted into the bladder or by mechanical manipulation of the bladder with the urethra clamped [17, 30, 35] markedly increased the rate of ascending infection. Reflux induced by such maneuvers causes mechanical damage to the mucosa at the fornix region that may serve as the site of ascending bacterial invasion into the renal parenchyma and the blood stream [31] or may even serve as a nidus for blood-borne pyelonephritis [11]. It is worth noting that 24-h ureteral obstruction produces fornical lesions comparable to those induced by acute reflux [19]. The predominant exudative reaction in this region in infected kidneys, previously described by others [11, 17, 39] may indicate the site of bacterial invasion, underscoring the importance of such predisposing parenchymal damage. The fornix region is also the site of physiologic pyelorenal urinary backflow [37, 40] that may serve as an additional mechanism for spread of infection from the renal pelvis into the renal parenchyma and blood stream.

Urethral manipulation in experimental ascending pyelonephritis has been tried before by Prat et al. [30]: 1 hr after the release of 4–6-h unilateral ureteral obstruction, a large-volume inoculum (0.8–1.0 ml) was inserted into the bladder. The urethra was clamped and the bladder massaged repeatedly. Reflux was demonstrated radiographically and after the manipulation a delayed antegrade emptying of the renal pelvis was noted in the affected kidney. Urethral manipulation resulted in increased incidence of ascending infection on the involved side. In addition, these authors have also demonstrated that ureteral ligature and transection prevented the ascending infection, confirming the luminal or periureteral ascending route of infection. Since in humans vesicoureteral reflux is absent without evident anatomic changes or infection, we avoided induced reflux, using a smaller volume of inoculum (0.5 ml), without massaging the bladder. The incidence of bacterial infection in the right unaffected kidney (33%) is well within the expected reported 20% chance of reflux at the volume of inoculum used [36] and the possibility of ascending infection with a virulent *E. coli* strain [31]. It was a low-grade infection, with only 2900 cfu/ml on the average, and was not accompanied by morphologic alterations suggesting fornical reflux-induced damage or infection. By contrast, all manipulated left kidneys (100%) were heavily infected, 79,000 cfu/ml on the average. This high rate extensive ascending infection probably reflect delayed antegrade emptying of the affected upper urinary collecting system, combined with parenchymal invasion at injured renal sites.

Pyelonephritis alters renal parenchymal structure, ultimately leading to tubular atrophy, interstitial fibrosis and renal scarring [9, 20, 23]. Urinary infection inhibits ureteral peristalsis and induces intrarenal reflux, predisposing to medullary damage from altered urinary flow [24]. Moreover, the inflammatory process directly leads to tissue damage [11, 17, 24, 39] and tubular atrophy [4, 23, 33], even at the absence of reflux. This is mediated by cytotoxic and oxidative injury as well as by hypoxic damage secondary to interstitial edema and granulocyte aggregation within capillaries [1, 13, 32].

Transient or partial ureteral obstruction and infection could perpetuate each other, with the former providing nidus for bacterial invasion in damaged tissue and the latter exacerbating urine stasis. Intensification of morphologic tubulointerstitial changes has therefore been anticipated in transiently obstructed infected kidneys. Our major morphologic findings associated with infection have been focal acute inflammatory reaction and marked pelvic urothelial proliferation. By contrast, the extent of early outer medullary interstitial expansion was comparable in both infected and noninfected transiently obstructed kidneys (Table 1). Outer medullary interstitial expansion was not reported in a 72-h mouse model of ascending unobstructed pyelonephritis [24]. However, interstitial edema, compromising outer medullary microcirculation, has indeed been demonstrated in piglets subjected to sustained reflux pyelonephritis [1]. Therefore, more prolonged morphologic studies may be needed to explore possible intensification of obstruction-induced outer medullary interstitial changes by concomitant infection.

In conclusion, our series provides morphologic evaluation of early renal medullary changes that take place following transient ureteral obstruction, underscoring the rapid interstitial expansion particularly in the fornix region and in the interbundle zone of the outer medulla. These early changes may relate to altered urodynamics and medullary tissue destruction. Transient ureteral obstruction also predisposes to ascending pyelonephritis that does not seem to enhance early medullary interstitial expansion.

References

1. Androulakakis PA, Ransley PG, Risdon RA, Sorger K, Hohenfellner R (1987) Microvascular changes in the early stage of reflux pyelonephritis. An experimental study in the pig kidney. Eur Urol 13: 219
2. Chevalier RL (1996) Growth factors and apoptosis in neonatal ureteral obstruction. J Am Soc Nephrol 7: 1098
3. Cotran RS, Thrupp LD, Hajj SN, Zangwill DP, Vivaldi E, Kass EH (1963) Retrograde E. coli pyelonephritis in the rat: a bacteriologic, pathologic, and fluorescent antibody study. J Lab Clin Med 61: 987
4. Damadian RV, Shwayri E, Bricker NS (1965) On the existence of non-urine forming nephrons in the diseased kidney of the dog. J Lab Clin Med 65: 26
5. Diamond JR (1995) Macrophages and progressive renal disease in experimental hydronephrosis. Am J Kidney Dis 26: 133
6. Diamond JR, Kees-Folts D, Ding G, Frye JE, Restrepo NC (1994) Macrophages, monocyte chemoattractant peptide-1, and TGF-beta 1 in experimental hydronephrosis. Am J Physiol 266: F926
7. Diamond JR, Ricardo SD, Klahr S (1998) Mechanisms of interstitial fibrosis in obstructive nephropathy. Semin Nephrol 18: 594
8. Diamond JR, Kreisberg R, Evans R, Nguyen TA, Ricardo SD (1998) Regulation of proximal tubular osteopontin in experimental hydronephrosis in the rat. Kidney Int 54: 1501
9. Ekonoyan G (1997) Chronic tubulointerstitial nephropathies. In: Schrier RW, Gottschalk CW (eds) Diseases of the kidney. Little Brown, Boston, p 1983
10. Fern RJ, Yesko CM, Thornhill BA, Kim HS, Smithies O, Chevalier RL (1999) Reduced angiotensinogen expression attenuates renal interstitial fibrosis in obstructive nephropathy in mice. J Clin Invest 103: 39
11. Fierer J, Talner L, Braude AI (1971) Bacteremia in the pathogenesis of retrograde E coli pyelonephritis in the rat. Am J Pathol 64: 443
12. Gilbert RM, Weber H, Turchin L, Fine LG, Bourgoignie JJ, Bricker NS (1976) A study of the intrarenal recycling of urea in the rat with chronic experimental pyelonephritis. J Cin Invest 58: 1348
13. Glauser MP, Lyons JM, Braude AI (1978) Prevention of chronic experimental pyelonephritis by suppression of acute suppuration. J Clin Invest 61: 403
14. Gorrill RH (1956) The effect of obstruction of the bladder on the renal localization of bacteria. J Path Bact 72: 57
15. Guze LB, Beeson PB (1956) Experimental pyelonephritis, I. Effect of ureteral ligation on the course of bacterial infection in the kidney of the rat. J Exper Med 104: 803
16. Guze LB, Hubert E, Kalmanson GM (1965) Pyelonephritis VI. Observations of the effects of congenital, partial ureteral obstruction on susceptibility of the rat kidney to infection. J Infect Dis 115: 500
17. Heptinstall RH (1964) Experimental pyelonephritis: bacteriological and morphological studies on the ascending route of infection in the rat. Nephron 1: 73
18. Heyman SN, Brezis M, Epstein FH, Spokes K, Silva P, Rosen S (1991) Early renal medullary hypoxic injury from radiocontrast and indomethacin. Kidney Int 40: 632
19. Heyman SN, Fuchs S, Yafe R, Beeri R, Elazarian L, Brezis M, Rosen S (1997) Renal microcirculation and tissue damage during acute ureteral obstruction: the effect of saline infusion, indomethacin and radiocontrast. Kidney Int 51: 653
20. Hodson CJ, Cotran RS (1983) Vesicoureteral reflux, reflux nephropathy and chronic pyelonephritis. In: Cotran RS, Brenner BM, Stein JH (eds) Tubulo-interstitial nephropathies. Churchill Livingstone, New York, p 83 (Contemporary issues in nephrology, vol 10)
21. Hudkins KL, Giachelli CM, Cui Y, Couser WG, Johnson RJ, Alpers CE (1999) Osteopontin expression in fetal and mature human kidney. J Am Soc Nephrol 10: 444
22. Ishidoya S, Morrissey J, McCracken R, Klahr S (1996) Delayed treatment with enalapril halts tubulointerstitial fibrosis in rats with obstructive uropathy. Kidney Int 49: 1110
23. Johnson JR, Manivel JC (1991) Vesicoureteral reflux induces renal trauma in a mouse model of ascending, unobstructed pyelonephritis. J Urol 145: 1303
24. Johnson JR, Berggren T, Manivel JC (1992) Histopathologic-microbiologic correlates of invasiveness in a mouse model of ascending unobstructed urinary tract infection. J Infect Dis 165: 299
25. Kaneto H, Morrissey J, McCracken R, Reyes A, Klahr S (1998) Osteopontin expression in the kidney during unilateral ureteral obstruction. Miner Electrolyte Metab 24: 227
26. Klahr S, Morrissey JJ (1998) The role of growth factors, cytokines, and vasoactive compounds in obstructive uropathy. Semin Nephrol 18: 622
27. Klahr S, Ishidoya S, Morrissey J (1995) Role of angiotensin II in the tubulointerstitial fibrosis of obstructive nephropathy. Am J Kidney Dis 26: 141
28. Kuemmerle NB, Brandt RB, Chan W, Krieg RJ Jr, Chan JC (1997) Inhibition of transforming growth factor beta 1 induction by vitamin E in unilateral ureteral obstruction in rats. Biochem Mol Med 61: 82
29. Pimental JL, Sundell CL, Wang S, Copp JB, Montero A, Martinez-Maldonado M (1995) Role of angiotensin II in the expression of transforming growth factor-beta in obstructive uropathy. Kidney Int 48: 1233
30. Prat V, Losse H, Konickova L, Ritzerfeldd W (1970) Experimental ascending escherichia coli pyelonephritis in the rat. Invest Urol 8: 311

31. Roberts JA, Suarez GM, Kaack B, Kallenius G, Sevenson SB (1985) Experimental pyelonephritis in the monkey. VII. Ascending pyelonephritis in the absence of vesicoureteral reflux. J Urol 133: 1068
32. Roberts JA (1990) Pathogenesis of nonobstructive urinary tract infections in children. J Urol 144: 475
33. Serlachius E, Sundelin B, Eklof AC, Jahnke M, Laestadius A, Aperia A (1997) Pyelonephritis provokes growth retardation and apoptosis in infant rat renal cortex. Kidney Int 51: 1855
34. Sharma A, Mauer SM, Kim Y, Michael AF (1993) Interstitial fibrosis in obstructive nephropathy. Kidney Int 44: 774
35. Slotki IN, Asscher AW (1982) Prevention of scarring in experimental pyelonephritis in the rat by early antibiotic therapy. Nephron 30: 262
36. Sommer JL (1961) Experimental pyelonephritis in the rat with observations on ureteral reflux. J Urol 86: 375
37. Stenberg A, Bohman SO, Morsing P, Muller Suur C, Olsen L, Persson AEG (1988) Back-leak of pelvic urine to the blood stream. Acta Physiol Scand 134: 223
38. Tolkoff-Rubin NE, Rubin RH (1983) Urinary tract infection. In: Cotran RS, Brenner BM, Stein JH (eds.) Contemporary issues in nephrology (10): tubulo-interstitial nephropathies. Churchill Livingstone, New York, p 49
39. Tomas G, Tange JD (1985) Experimental pyelonephritis and papillary necrosis in the Gunn rat. Pathology 17: 420
40. Thomsen HS, Dorph S, Olsen S (1981) Pyelorenal backflow in normal and ischemic rabbit kidneys. Invest radiol 16: 206
41. Truong LD, Seheikh-Hamad D, Chakraborty S, Suki WN (1998) Cell apoptosis and proliferation in obsructive uropathy. Semin Nephrol 18: 641
42. Yamate J, Okado A, Kuwamura M, Tsukamoto Y, Ohashi F, Kiso Y, Nakatsuji S, Kotani T, Sakuma S, Lamarre J (1998) Immunohistochemical analysis of macrophages, myofibroblasts, and transforming growth factor-beta localization during rat renal interstitial fibrosis following long-term unilateral ureteral obstruction. Toxicol Pathol 26: 79