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Suprapubic Bladder Catheterization of Male Spinal-Cord–Injured Sprague–Dawley Rats

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The rat spinal-cord–injury (SCI) model is widely used to study the pathologic mechanisms that contribute to sensory and motor dysfunction in humans. This model is thought to mimic many of the negative outcomes experienced by humans after spinal contusion injury. We theorized that manual bladder expression contributed to the kidney and bladder lesions reported in previous studies using the rat SCI model. In the present study, rats were surgically implanted with bladder catheters after spinal contusion injury to provide continuous drainage of urine. After 72 h, the rats were euthanized and their kidneys and bladders examined histologically. BUN, serum creatinine, and urine protein were compared at 0 and 72 h after surgery. Kidney and bladder lesions were similar in SCI rats with and without implanted bladder catheters. BUN at 72 h was higher than baseline values in both groups, whereas serum creatinine was higher at 72 h compared with baseline values only in the catheterized rats. These findings indicate that suprapubic bladder catheterization does not reduce hydronephrosis in SCI rats and that the standard of care for bladder evacuation should continue to be manual expression of urine.

Abbreviations: SCI, spinal cord injury; TPER, tissue protein extraction reagent; VUR, vesicoureteral reflux.

The effects and outcomes of spinal cord injury (SCI) have been studied for decades in rats. This model reproduces many, if not all, of the aspects of the condition in humans. In addition to the primary spinal cord lesions, lesions of liver, intestine, lung, bladder, and kidney have been described in SCI rats.[10,11,15,17] These reports often focus on the resolution of lesions in organs other than the spinal cord that were associated with various treatment regimens.[21,29,30,33] In addition to an acute inflammatory response in rat kidneys after SCI, histologic findings included reduced space in the renal tubules and collapse of Bowman space in a previous study.[15] Other researchers have shown that renal tubular degeneration and glomerular dysfunction were present in rat kidneys at 14 d after SCI.[29]

In addition to morphologic evidence of kidney damage, hypertrophic changes in the bladder wall including transitional epithelial hyperplasia, degeneration of the lamina propria, and an increase in the tunica adventia layer, have been reported previously.[29] The hypertrophic bladder was 2 to 9 times heavier than were controls and had decreased ability to void urine.[20,29,31] SCI cranial to the lumbosacral level produced a loss of voluntary micturition that was attributable to detrusor–external sphincter dyssynergia, defined as a lack of coordination between the contractions of the detrusor muscle of the bladder wall and the external urethral sphincter.[19] Detrusor–external sphincter dyssynergia led to urinary retention after SCI. In previous studies, the urinary bladder was expressed manually 2 or 3 times daily according to a modified method.[13,30,35] Manual expression of the bladder was shown to cause vesicoureteral reflux by allowing retrograde flow of urine to the ureters and renal pelvis.[23] Vesicoureteral reflux was implicated in medullary-centered renal injury in previous studies.[7,23]

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Our aims were to evaluate an alternative method for evacuating the full bladder and to determine the effect of the new method on the kidneys and bladders of rats after SCI. We hypothesized that suprapubic bladder catheterization would reduce urine accumulation and lead to decreased bladder hypertrophy because of the constant passive drainage of urine. Consequently, vesicoureteral reflux would be eliminated and the kidney protected. To our knowledge, ours is the first study to evaluate the use of suprapubic bladder catheterization in SCI rats. We compared the histology of the kidneys and bladders of SCI rats with and without bladder catheters with those of controls. In addition, renal function was evaluated by measurement of BUN, creatinine, and urine protein levels at 0 and 72 h after SCI and compared with control values.

Materials and Methods

Animals. All experimental protocols were approved by the Animal Welfare Committee (IACUC) at the University of Texas Health Science Center at Houston, followed the recommendations set forth in the *Guide for the Care and Use of Laboratory Animals*,[18] and complied with the US Public Health Service Policy on the Humane Care and Use of Laboratory Animals.[27] Male Sprague–Dawley rats (age, 7 to 8 wk; weight, 225 to 250 g) were purchased from Harlan Laboratories (Houston, TX) and housed in the centralized animal facility at 22 ± 1 °C and 45% to 55% relative humidity with a 12:12-h light:dark cycle. Sentinels exposed to dirty bedding from all cages on the rack were negative for rat parvovirus, Toolan H1 virus, Kilham rat virus, rat minute virus, rat coronavirus, sialodacryoadenitis virus, Sendai virus, rat theilovirus, reovirus 3, *Mycoplasma pulmonis*, pinworms, and fur mites. Rats were fed a standard commercial rat chow (Rodent Diet 5001, PMI Labdiet, St Louis, MO) and given tap water ad libitum. Rats were divided randomly into 3 groups: unmanipulated controls ($n = 4$); SCI surgery without implantation of bladder catheter ($n = 4$), and SCI surgery with catheter implantation ($n = 6$).

Spinal cord injury surgery. All rats undergoing surgery were anesthetized by using a 'cocktail' of ketamine (80 mg/kg), xylazine (10 mg/kg), and acepromazine (0.75 mg/kg) at a dose of 0.1 mL/100 g body weight. SCI surgeries were performed by using a spinal injury device (Infinite Horizon Device, Precision Systems and Instrumentation, Lexington, KY).[28] A laminectomy was performed at the tenth thoracic vertebra and the vertebral column stabilized at thoracic vertebrae 9 through 11. A moderate contusion injury was delivered by using 150 kilodynes of force with a 1-s dwell time.[17] Overlying muscles then were sutured, and the skin was closed with stainless steel wound clips (EZ Clip, Stoelting, Wood Dale, IL). Control rats were not surgically manipulated.

Preparation of bladder catheter. The open flared end of a 3.5-French, 5.5-in. semirigid polypropylene catheter (Covidien, Mansfield, MA) with an inner diameter of 0.76 mm was trimmed to create an elliptical, pointed tip (Figure 1). The narrow open end was trimmed so that the catheter was no more than 3 in. in length. The catheters were sterilized by using ethylene oxide (Anprolene AN74i, Andersen Products, Haw River, NC). Prior to use, the catheters were rinsed with a heparin–saline solution (10 units of heparin in 1 mL 0.9% sterile saline).

Bladder catheterization surgery. The surgical technique for rats undergoing catheter implantation was modified from the previously described method.[36] The caudoventral abdomen was clipped with a no. 40 blade and the skin disinfected by wiping with 2% chlorhexidine digluconate scrub followed by 70% isopropyl alcohol; this process was repeated twice. The skin at the incision site was injected with 0.2 mL 0.25% bupivicaine. To expose the bladder, a 2-cm incision was made on the midline, extending cephalad from the pubic bone. The bladder was retracted gently cranially. A 16-gauge, 3-in. spinal needle with the stylet removed was used to puncture the bladder on the ventral side near the neck. The pointed, elliptical end of the catheter was introduced into the bladder through the puncture. A pursestring suture of 5-0 polypropylene was placed in the bladder wall around the catheter entry site (Figure 2). The site was checked for leakage of urine and the bladder replaced into its normal position. The abdomen was lavaged with warm 0.9% saline. The catheter exited the abdomen at an oblique angle at the caudal end of the skin incision. The muscular layer of the abdominal wall was sutured in a continuous pattern, and the skin incision was closed with wound clips. The external tip of the catheter exited caudally between the hindlimbs to allow for continuous passive emptying of the bladder. All rats recovered from anesthesia uneventfully. The bladder catheters were examined for urine flow before the rats were returned to their home cages.

Postoperative care. All rats that underwent surgery received buprenorphine (0.03 mg/kg SC) twice daily and enrofloxacin (10 mg/kg SC) once daily throughout the study. Each rat's hydration status was assessed twice daily by tenting the skin at the scruff of the neck and by observing the color of the urine produced. Subcutaneous fluids (0.9% sterile saline) were administered twice daily to all rats. All operated rats each were given 3 mL saline SC twice on the day after surgery. Thereafter, rats were given 2 mL saline twice daily, unless the catheter was plugged or the urine was dark in color compared with that of others in the group. Rats with these signs received 2.5 mL saline SC twice daily. Softened food pellets were placed in sterile plastic dishes on the floors of the cages within easy reach of the rats. Zinc oxide cream was applied daily to the medial thighs of the catheter-bearing rats to protect against urine scald. Manual bladder expression was performed twice daily in SCI-only rats by restraining the rat in one hand and gently expressing the bladder with the fingertips of the other hand. If urine did not flow from a bladder catheter, the bladder was examined for fullness in the same manner. Blood clots or debris that impeded urine flow were removed by flushing the catheter with a small amount of heparin–saline solution, alternating with gentle suction. Blockages that could not be flushed out of the catheter were removed by trimming 3 mm from the tip of the catheter once daily if the catheter was blocked.

Blood collection. Control rats were anesthetized briefly with 2% isoflurane for the collection of 0.25 mL whole blood from the ventral tail artery. For remaining rats, blood was collected from each rat while anesthetized for the surgical procedure. A second blood sample was collected from all rats at 72 h after surgery.

Urine collection. To collect the presurgical urine sample, each rat was placed in a clean, nonbedded plastic rat cage and observed for voluntary micturition. Urine was retrieved from the cage by using a new sterile syringe for each sample. A second sample was obtained from control rats in the same manner at the 72-h time point. SCI-only rats were restrained for manual bladder expression to collect the second urine sample. For catheter-bearing SCI rats, the second urine sample was collected directly from the catheter, in some cases with gentle abdominal massage.

Euthanasia and tissue processing. All rats were euthanized by pentobarbital–phenytoin overdose (1 mL IP per rat). Specific tissues of interest were collected, including both kidneys and the urinary bladder, and placed in 10% buffered formalin. Tissues were trimmed, placed in cassettes, and routinely processed to produce paraffin blocks. Sections (5 µm) were cut from the paraffin-embedded tissue, mounted on glass slides, and stained with hematoxylin and eosin.

Histology. Slides were examined by a veterinary pathologist. Images were acquired on an Olympus BX61 upright microscope at 10× and 20× magnification. Brightfield images were captured by using a Spot-Flex camera (Diagnostic Imaging, Sterling Heights, MI) and annotated within Adobe Photoshop CS4 (Adobe, San Jose, CA).

BUN and creatinine determination. Pre- and postsurgical blood samples were centrifuged at 1327 × *g* for 10 min and the serum collected. Samples were run on an inhouse chemistry analyzer (VetTest 8008, IDEXX, Westbrook, ME) to quantify BUN and creatinine levels.

Urine protein concentration assay. Urine samples were collected before SCI surgery and again 72 h after surgery. Samples were collected from control rats at 0 and 72 h. Frozen (−20 °C) samples were thawed, and urine protein concentrations were measured by using a bicinchoninic acid assay (Thermo Scientific, Rockford, IL) with BSA as the standard. Briefly, 10 mg BSA was added to 1 mL of tissue protein extraction reagent buffer and protease inhibitor and then serially diluted to obtain the set of BSA concentration standards. All urine samples were diluted 1:5, 1:10, and 1:20 with buffer. Aliquots (5 µL each) of BSA standards, blank (buffer only), and diluted urine samples were pipetted into a 96-well plate. Standards and blanks were tested in duplicate and urine samples in triplicate. The bicinchoninic acid assay protein standard reagent was mixed according to the kit directions, and 250 µL of this reagent was pipetted into each sample well. The plate was covered with foil and mixed on a rocker for 30 min. A microplate reader was used to measure absorbance of each BSA standard and urine sample at 490 nm. Protein levels in the collected urine samples were estimated based on the standard curve generated from the BSA standards.

Figure 1. A 16-gauge, 3-in. spinal needle (top) was used to puncture the bladder to create an opening for introduction of the polypropylene catheter (bottom).

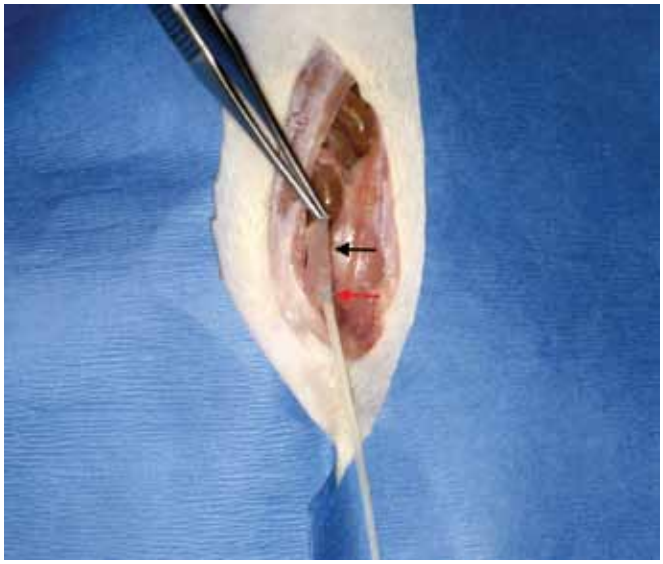

Figure 2. The bladder (black arrow) of a rat shown after placement of the catheter. A pursestring suture (red arrow) secured the catheter in place.

Statistical methods. Statistical analyses of BUN, creatinine, and urine protein values were performed as repeated-measures ANOVA by using SAS Proc GLM (version 9.2; SAS, Carey, NC). The models included group as a between-subjects variable and time as a within-subjects variable. Pairwise comparisons of the terms comprising significant interactions were evaluated for statistical significance ($P < 0.05$) by using the Tukey–Kramer test to control for type I error.

Results

Gross appearance of urinary bladders. Among the 6 rats that underwent SCI and catheter implantation, the urinary bladders of 3 were distended at necropsy and contained between 0.5 and 2 mL urine. The ends of the catheters were positioned against the bladder mucosa and appeared at least partially blocked. In the remaining 3 rats in this group, the ends of the catheters were not positioned against the bladder mucosa; these bladders were much less distended and contained less than 1 mL urine. These bladders more closely resembled the bladders of control rats, all of which were normal in appearance and contained less than 1 mL urine. The bladders of all 4 of the SCI-only rats were distended and contained between 1 and 3 mL urine.

Gross appearance of the kidneys. The right kidney from each rat was examined. Kidneys from rats in both groups that underwent SCI were grossly larger than those from control rats. Each kidney was cut longitudinally and examined. The renal pelvis in kidneys from each of the SCI rats was dilated compared with those of control rats.

Histologic appearance of urinary bladder. Histologically, the urinary bladders of control rats were normal with no edema or inflammation (Figure 3 A). In rats that underwent SCI only, the bladders had moderate edema and a multifocal infiltrate consisting primarily of neutrophils within the submucosa and muscularis layers (Figure 3 B). The bladders of rats that received SCI and catheters exhibited focal moderate to marked edema in the submucosa of the bladder wall (Figure 3 C), with marked, diffuse, chronic, active inflammation that extended from the submucosa into the muscularis. In addition, mild hemorrhage was present in the submucosa and mucosa of the bladders in both groups that underwent SCI.

Histologic appearance of kidneys. In control rats, the kidney sections appeared normal (Figure 4 A). The kidneys of SCI-only rats had moderate to marked dilatation of the collecting tubules, with degeneration, necrosis, and loss of tubular epithelium (Figure 4 B). The kidneys of rats that underwent SCI and catheter implantation displayed numerous areas in the renal medulla where collecting tubules were dilated with loss, flattening, or degeneration of tubular epithelium (Figure 4 C). No inflammatory cell populations were identified in the kidneys of either of the groups of operated rats, and the cortexes of these kidneys appeared normal.

Comparison of BUN, creatinine, and urine protein levels. BUN displayed a significant group × time interaction ($F[5, 22] = 21.38$, $P < 0.0001$). Post hoc contrasts, corrected by using the Tukey Studentized range test, indicated that both groups of rats that underwent SCI developed significant increases in BUN after surgery (Figure 5 A); In control rats, BUN was not different between samples collected at 0 and 72 h. In contrast, BUN (mean ± 1 SD) was 15.75 ± 2.06 mg/dL at 0 h and 22.25 ± 2.87 mg/dL at 72 h in SCI-only rats and 16.17 ± 1.94 mg/dL at 0 h and 29.50 ± 3.21 mg/dL at 72 h in those that received SCI and catheters. Creatinine showed a significant group × time interaction ($F[5, 22] = 3.50$, $P < 0.02$). Post hoc contrasts, corrected by using the Tukey Studentized range test, showed that control rats had a significant increase between the 0- and 72-h time points, as did rats that underwent both SCI and catheter implantation (Figure 5 B). Creatinine values were 0.325 ± 0.096 mg/dL at 0 h and 0.475 ± 0.096 mg/dL at 72 h for control rats. Rats that received SCI and catheters had a creatinine level of 0.433 ± 0.082 mg/dL at 0 h and 0.583 ± 0.117 mg/dL at 72 h. The creatinine values of SCI-only rats did not differ significantly at 0 and 72 h. Urine protein did not differ between the 0- and 72-h time points in any group (Figure 5 C).

Discussion

Complications arising from bladder management and care are a crucial concern in humans with SCI.[9] Increasing severity of kidney disease in persons with SCI was found to be a positive predictor of likelihood of death.[14] The present study compared the degrees of renal injury in SCI rats with and without bladder catheters. All rats that underwent SCI without implantation of bladder catheters developed hydronephrosis and various amounts of tubular nephrosis in the renal medulla. The kidneys of rats with SCI and bladder catheters had similar pathologic changes. Compromised renal function was reflected in the rise in BUN in blood samples collected from both groups of rats with SCI and in the increase in creatinine in rats with catheters at 72 h after SCI. Changes in creatinine for control rats were significantly different at the 0- and 72-h time points, but both values were within the normal range for rats. Given that neither BUN nor creatinine values were abnormal at either time point in control rats, kidney damage is an unlikely cause of

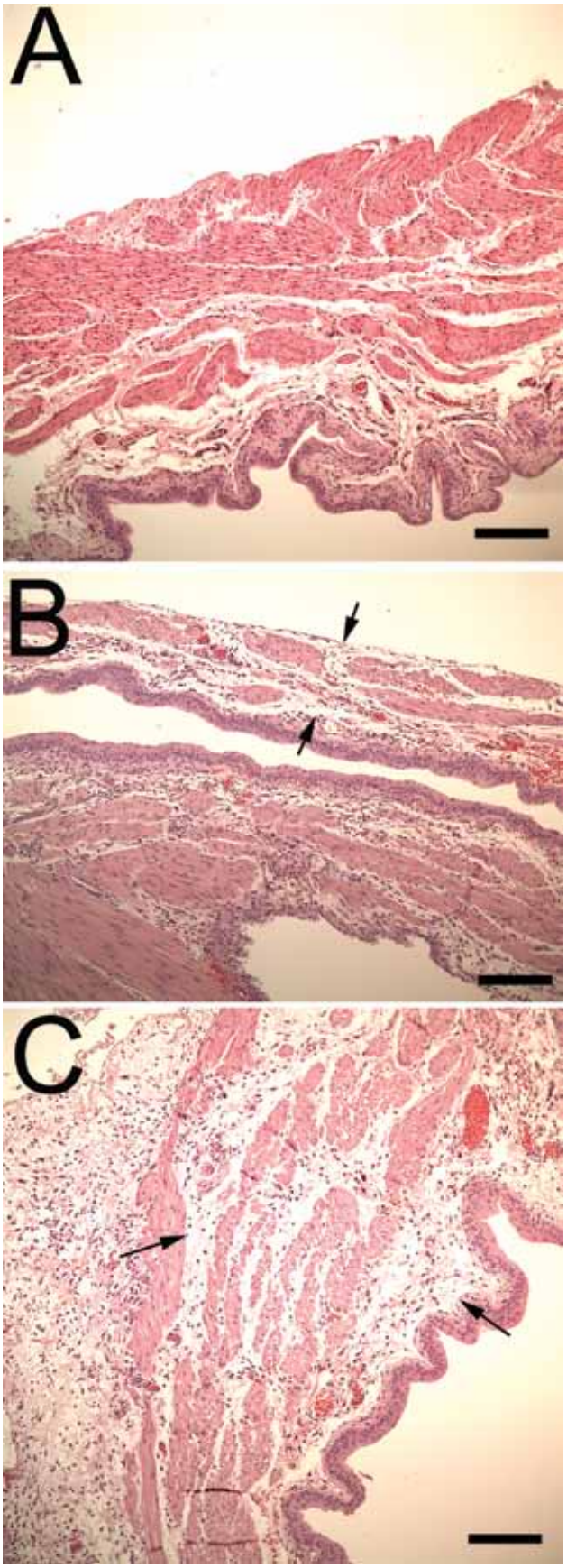

Figure 3. Photomicrographs of the bladder wall. (A) Control rat with normal submucosa and muscularis. (B) SCI-only rat and (C) catheterized SCI rat with edema in submucosa and muscularis (arrows). Hematoxylin and eosin stain; magnification, ×10; bar, 200 µm.

the increased creatinine values at 72 h in this group. Collection of 6-h urine samples for a 24-h period in a previous study of creatinine excretion revealed variation in the levels excreted throughout the day in normal rats.[8] Creatinine values at 72 h were significantly higher than baseline values in SCI rats with catheters and were outside the normal range. The significant change in creatinine in SCI rats with catheters compared with SCI-only rats was an unexpected finding that led us to theorize that more renal damage occurred in rats that were catheterized. In addition, the mean creatinine value at the 72-h time point was higher in these rats than in the other 2 groups.

The pathogenesis of hydronephrosis in kidneys of SCI rats can be explained by examining the effects of vesicoureteral reflux (VUR), the retrograde flow of urine from the bladder to the upper urinary tract.[16] As the bladder fills, the pressure in the renal pelvis increases.[2] Manual expression of a full urinary bladder might induce VUR in rats, particularly when the urinary output from the ureters is low. Normal intravesical pressure during micturition in rats is 15 to 20 mm Hg (20.4 to 27.2 cm H_2O).[23] In one study, intravesical pressures during manual expression of an obstructed bladder were as high as 250 mm Hg (340 cm H_2O), whereas the corresponding intrapelvic pressures increased to 125 mm Hg (170 cm H_2O).[23] In another study, VUR occurred in rats at bladder pressures as low as 45 cm H_2O, and the pressure at which VUR was induced fell as the number of manual bladder expressions increased.[34] During studies of SCI rats, manual bladder expression could occur as often as 2 or 3 times daily, thereby exposing the renal pelvis to cycles of low to high pressure, similar to those found during the cited VUR experiments.[23,34] The medullas of both groups of our SCI rats displayed tubular dilatation with loss, flattening, or degeneration of the tubular epithelium. This observation was consistent with histopathologic findings in rats with transient ureteral obstruction[7] and rabbits with induced VUR.[5] In those studies[5,7] as in the present one, tubular dilatation occurred as a consequence of increased renal pelvic pressure. In contrast to another study,[15] kidneys of rats in the current study did not stain positive for myeloperoxidase at the 72-h time point. This difference may reflect varying amounts of systemic inflammation between 2 different surgically induced models of SCI.

Bladder histology in both groups of SCI rats showed moderate to marked edema in the submucosa of the bladder wall. The cause of this edema is unknown. Early changes to the wall of the bladder that occur during the acute phase of SCI in rats have been identified and described.[3,4,6,13,17,20,29,31] The cells lining the interior of the bladder wall, the uroepithelium, were noted to extend from the urethra to the renal pelvis, forming a barrier between urine and the underlying tissues.[6] The uroepithelium comprised a basal cell layer attached to a basement membrane, an intermediate cell layer, and a superficial layer of hexagonal 'umbrella' cells.[3] The umbrella cells in particular were responsible for maintaining high transepithelial resistance and low permeability to urea and water.[4] Tight junction complexes between umbrella cells prevented the paracellular movement of small molecules, such as water and ions.[4,6] After SCI, there was disruption in the barrier function of the uroepithelium. By 24 h after SCI, a decrease in transepithelial resistance and an increase in permeability to water occurred concurrently with a significant reduction in umbrella cells in the uroepithelium.[4] In addition, there was a loss of tight junction proteins by 48 h after SCI that contributed to the dysfunction of the uroepithelium.[17] The extensive breakdown of the uroepithelial barrier allowed the influx of water and solutes and caused inflammation in the

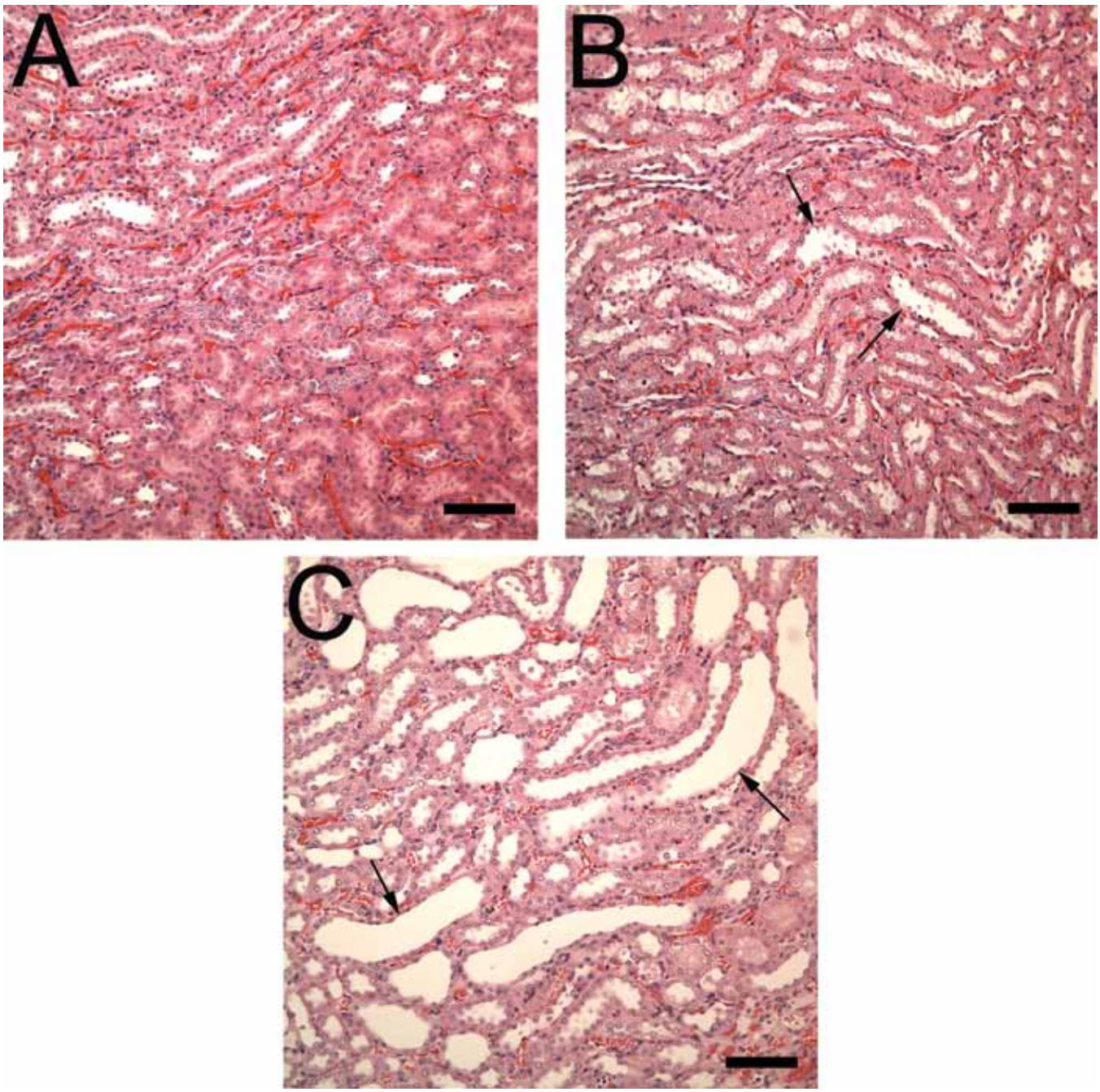

Figure 4. Photomicrographs of the kidney. (A) Control rat with normal renal tubular structure. (B) SCI-only rat and (C) catheterized SCI rat with tubular dilatation and degeneration of tubular epithelium (arrows). Hematoxylin and eosin stain; magnification, ×20; bar, 100 µm.

bladder wall,[4] although the underlying mechanisms of SCI-induced uroepithelial breakdown and inflammation were unclear.

The present study was designed to allow continuous drainage of the bladder of SCI rats to examine the effect on the development of hydronephrosis in the kidneys. Kidney histopathology did not support the hypothesis that passive continuous drainage of the bladder in male SCI rats would prevent or attenuate hydronephrosis in these rats. In addition, serum BUN increased significantly in both groups of SCI rats, and creatinine significantly increased after surgery in SCI rats with catheters compared with control rats. This result indicated that mild renal dysfunction developed despite fluid therapy in SCI-only rats and despite fluid therapy plus bladder catheterization in catheterized SCI rats. Consistent with previous findings,[17] urine protein levels were not significantly different between the 2 time points in any group of our rats.

To understand the failure of catheterization to address the development of hydronephrosis and tubular degeneration, SCI-dependent changes in bladder function must be considered. Normal micturition requires coordination between contraction of the smooth muscle of the bladder wall and relaxation of the striated muscle of the external urethral sphincter. Spinal and supraspinal pathways are involved in this coordination.[26] These pathways are disrupted after SCI, causing bladder areflexia during the acute period after injury, as demonstrated in female rats that underwent SCI at the level of T_9 to T_{11}, with rats recovering the spontaneous bladder-voiding reflex by 4 to 7 wk after SCI.[19] In addition, the bladder wall of SCI rats shows increased com-

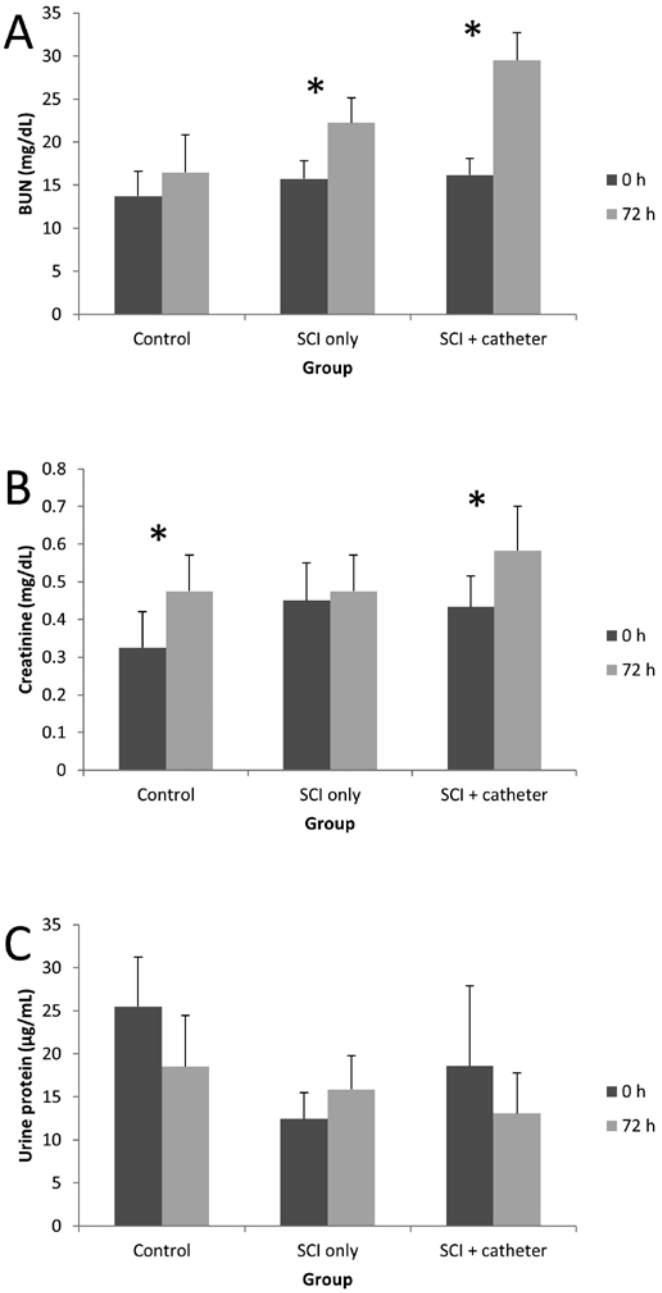

Figure 5. (A) BUN, (B) creatinine, and (C) urine protein results from all groups. Results are expressed as mean ± 1 SD. *, Significant difference between values at 0- and 72-h time points.

pliance as compared with that of normal specimens.[13] Urinary retention was exacerbated acutely after SCI due to a period of increased bladder compliance[31] that allowed large amounts of urine to remain in the bladder until manual expression was performed. In the present study, the loss of neural control coupled with the change in bladder compliance likely led to a complete inability of the bladder to generate the necessary force to void urine through the small diameter of the implanted catheter. However, some urine flowed through unblocked catheters during the twice-daily examination of the rats.

Three of the rats with SCI and catheters had bladders that were grossly smaller than those of the other 3 rats and all of the SCI-only rats. Some passive drainage of urine was occurring in at least these 3 catheter-implanted animals, most likely because of pressure on the external abdominal wall while the rats moved around the cages. Although the hindlimbs of these rats were completely paralyzed, the forelimbs exhibited no deficits, resulting in the rats' ability to locomote and rear up slightly. A complication in this model was the inability to prevent catheter blockage in 3 of the rats. Despite maintaining the rats' hydration status with subcutaneous injections of saline twice daily, crystalluria developed to a greater extent in some rats. Hematuria, which also contributed to catheter blockages, is a known complication of SCI.[4,17] The catheters in 4 of the 6 rats in the group were obstructed to various degrees by crystals, blood clots, or a combination of both. Clots were removed through a system of minute injections of heparin–saline mixture followed by gentle suction with a 22-gauge needle attached to a 3-mL syringe. In general, crystals could not be removed in the same way, and the catheter had to be trimmed to the level of the obstruction. How long the catheters in these rats remained obstructed was unknown. Although rats with unobstructed catheters had grossly smaller bladders, their bladder and kidney histology was not appreciably different from that of rats with obstructed catheters. Therefore, we conclude that in male SCI rats, passive continuous drainage of urine cannot easily be accomplished by suprapubic bladder catheterization. An alternative surgical approach using cutaneous vesicostomy may avoid the complications associated with the use of the catheters but could present other difficulties. Cutaneous vesicostomy, in which the bladder wall is incised, everted, and sutured to the skin, has been used in humans with SCI,[24] canines in a study of bladder reinnervation techniques,[1] cattle with urethral obstruction,[12] and goats with induced urethral obstruction[22] but has not been used in SCI rats. Further studies are needed to evaluate whether vesicostomy eliminates the histopathologic changes associated with bladder hypertrophy, VUR, and hydronephrosis in SCI rats.

In one study of VUR in rats, 25% of female Sprague–Dawley rats were found to have congenital VUR, and the bladder pressure necessary to cause VUR differed significantly between rats with and without congenital VUR.[2] A review of the literature revealed that SCI researchers overwhelmingly study female rats,[13,19,20,26,29-33] perhaps because of the greater ease of handling female rats while manually expressing the urinary bladder. During future studies on the effects of therapies on kidney and bladder histology and function in female SCI rats, particularly Sprague–Dawley rats, researchers should keep in mind that any findings should be interpreted in light of the potential for some of the rats to have changes associated with VUR. Transurethral bladder catheterization is possible in the female rat and may present a way of ameliorating the effect of VUR on kidney and bladder histology in future studies.

In the current study, we used male rather than female rats because the initial lesion of hydronephrosis was identified in male SCI rats, and most human patients with SCI are male. Transurethral bladder catheterization cannot easily be accomplished in male rats because of the flexure of the urethra as it travels over the pubic bone. Although PE10 tubing has successfully been used as a transurethral catheter to deliver solution to the level of the prostate,[25] the inner diameter of PE10 tubing is only 0.28 mm. Because we wanted urine to flow as freely as possible, the catheter used in the current study had a diameter of 0.76 mm, almost 3 times larger than that of PE10 tubing. Urine flow was established in all 6 rats with SCI and catheters immediately after the surgical procedure. Catheter blockage subsequently occurred 12 to 24 h after catheterization in 4 of these 6 rats and was managed as stated previously.

In conclusion, suprapubic bladder catheterization of male SCI rats did not prevent the development of kidney and bladder le-

sions. Evidence of mild renal dysfunction was present despite careful postoperative management of the hydration status of the rats. As the preferred method for emptying the bladder, manual expression 2 or 3 times daily should continue to be included in the postoperative care procedures for SCI rats.

Acknowledgments

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