TIME-COURSE CHANGES IN THE MECHANICAL PROPERTIES OF THE RAT URINARY BLADDER FOLLOWING SPINAL CORD INJURY

[1, 3]Kevin K. Toosi, [1, 2]Jiro Nagatomi, [2, 3]Michael B. Chancellor, [1, 2]Michael S. Sacks
[1]Department of Bioengineering, University of Pittsburgh, Pittsburgh, PA
[2]McGowan Institute for Regenerative Medicine, University of Pittsburgh, Pittsburgh, PA
[3]Department of Urology, University of Pittsburgh, Pittsburgh, PA
email: msacks@pitt.edu , web: www.pitt.edu/~msacks/etml.html

INTRODUCTION

The urinary bladder is a smooth muscle organ whose main functions are to store urine and to void when necessary. Since the most important aspect of the storage function of the bladder is to maintain low intravesical pressure in order to protect the upper urinary tract from backflow of urine, the compliance of the bladder wall is one of the key functional parameters to assess the health of this organ. Previously, using planar biaxial testing, we demonstrated that the mechanical compliance of bladder wall tissue in spinal cord injury (SCI) rats at 10 days post-injury was significantly greater compared to that of normal bladders under biaxial stretch [1]. These results contrasted typical, non-compliant bladder conditions found in chronic SCI patients [2]. In order to determine long-term alterations in the urinary bladder mechanical behavior due to SCI, the present study examined the biaxial mechanical properties of bladder wall tissues at various time points post-SCI. In addition, biochemical assays were used to quantify collagen and elastin contents of SCI bladders to correlate with tissue-level findings of biomechanical properties.

METHODS

Female SD rats in the test group were subjected to complete transection of the spinal cord at the T9-T10 level, and the bladders were harvested at 3-week, 6-week and 10-week post-SCI. Normal rats were used as controls. Using our custom planar biaxial testing device, bladder specimens were subjected to equibiaxial stress (100kPa), and areal strain of the rat bladder wall, as an index of the mechanical compliance, was calculated for each group.

Following biaxial mechanical testing the bladder specimens were weighed, cut into smaller strips, and digested in 0.5N acetic acid supplemented with 1 mg/mL pepsin at 4 °C overnight. Acid-soluble collagen in the supernatant solution was quantified using a commercially available assay kit. The insoluble tissue materials (following acetic acid digestion) were further treated with 0.25M oxalic acid at 95 °C for 180 minutes. Elastin concentrations in these supernatants were also quantified using a commercially available assay kit. The data were normalized by wet tissue weight and expressed as average ± SEM, analyzed using a t-test when compared to that of normal and 3-week SCI bladders from our previous study and the difference was considered significant if $p<0.05$ [3].

RESULTS AND DISCUSSION

Bladder wall compliance was significantly greater at three and six weeks after spinal cord injury, when compared to normal. The increase in compliance, however, diminished by ten weeks post-injury, and was similar to that of normal bladder. Furthermore, when maximum axial stretches in two anatomical directions were compared, it was significantly ($p<0.05$) greater in the circumferential direction than in the longitudinal direction in 3-, 6- and 10-week SCI bladders. The different mechanical responses in two directions found in 3-, 6- and 10-week SCI bladder tissues indicate material anisotropy in these samples. This finding contrasted the isotropic behavior found in 10-day SCI bladders, suggesting that material class of bladder tissue continues to change from 10-day to 10-week after SCI.

Collagen concentrations (*i.e.* collagen mass normalized by wet tissue weight of bladder sample) of the 3-week and 10-week SCI rat bladders were similar to each other but significantly lower compared to the normal bladders, due to increased mass of hypertrophied bladder. However, the collagen content (*i.e.* total collagen mass per bladder sample) of 10-week SCI bladders was significantly greater than both normal and 3-week SCI samples, indicating increased collagen production by smooth muscle cells between three and ten weeks after injury. Elastin concentrations (and contents) of the 3-week and 10-week SCI bladders were similar to each other and significantly higher than that of normal bladders. These results suggest that elastin content primarily increases over the first three weeks following injury and remains constant after that.

CONCLUSIONS

The changes in the tissue mechanical compliance, composition and material class of bladder wall over a 10-week period of time post-SCI indicate that the bladder tissue continuously remodels after spinal cord injury. The results of the present study provide first evidence that early and late responses of bladder tissue following spinal cord injury were distinct from each other. Understanding that the bladder wall functional behavior continuously alters over the time, urologists may need to choose different clinical approaches regarding treatment of non-compliant bladder in SCI patients, based on injury's time-course.

REFERENCES

1. Gloeckner, D.C., et al., *Passive biaxial mechanical properties of the rat bladder wall after spinal cord injury.* J Urol, 2002. **167**(5): p. 2247-52.
2. Hackler, R.H., M.K. Hall, and T.A. Zampieri, *Bladder hypocompliance in the spinal cord injury population.* J Urol, 1989. **141**(6): p. 1390-3.
3. Nagatomi, J., et al., *Changes in the biaxial viscoelastic response of the urinary bladder following spinal cord injury.* Ann Biomed Eng, 2004. **32**(10): p. 1409-19.