

PARTIAL BLADDER OUTLET OBSTRUCTION INDUCES URETHRAL SMOOTH MUSCLE HYPERTROPHY AND DECREASED FORCE GENERATION

Hypothesis / aims of study

Partial bladder outlet obstruction (PBOO) leads to increased urinary frequency, decreased void volume, urinary retention, and hypertrophy of the detrusor smooth muscle. Obstructed animals reveal detrusor smooth muscle hypertrophy and alteration in contractile and regulatory proteins. This study was conducted to determine whether PBOO-induced increase in urinary frequency is associated with urethral smooth muscle hypertrophy and alteration in the contractility and the expression of myosin isoforms.

Study design, materials and methods

PBOO was surgically induced in male New Zealand White rabbits, and sham-operated rabbits served as a control. Twelve days after obstruction, urine output and voiding frequency were monitored by keeping the animals in metabolic cages prior to euthanasia. Animals with increased urinary frequency (43 ± 12 voids/ 24 hrs) and sham-operated rabbits with (6 ± 3 voids/24 hrs) were euthanized two weeks after surgery. Morphology of the urethra was studied using light and immunofluorescence microscopy. RT-PCR and Western Blotting were used to study the expression of myosin isoforms. Urethras were immersed and stored in Tyrodes buffer at 37°C and equilibrated with 95% oxygen 5% CO₂. Longitudinal strips of urethral wall free of the mucosa and serosa (~3 mm X 10 mm) were prepared and used for force measurements using a Grass Model 7D Polygraph. Muscle strips were contracted either by adding KCl (125 mM), phenylephrine (250 µM) or the cholinergic agonist, carbachol (100 µM). The smooth muscle content of the muscle strips used for force measurements was confirmed by histology and the force was expressed per gram of smooth muscle tissue.

The morphology of the urethral smooth muscle was studied using light and immunofluorescence microscopy. RT-PCR and Western Blotting were used to study the expression of myosin isoforms.

Results

The urethral smooth muscle of PBOO rabbits with increased urinary frequency showed hypertrophy. The force produced by the longitudinal muscle strips from the urethra of PBOO animals in response to phenylephrine, KCl, or electrical field stimulation was diminished 50%, 37% and 40%, respectively, compared to that of sham. Immunofluorescence microscopy using neurofilaments antibody revealed a reduction in nerve density. RT-PCR and Western blotting showed a significant decrease in the expression of myosin isoform SM-B with a concomitant increase in SM-A, both at the mRNA and protein levels.

Interpretation of results

These results indicate that there is an alteration in the contractility in the hypertrophied urethral smooth muscle in PBOO. A decrease in the neurofilament protein and innervation of the urethra suggests that the alteration in the myosin isoform is related to decreased innervation. Both hypertrophy and a switch to an isoform that is compatible with tonic-type smooth muscle in the proximal urethra might affect the urethral tone.

Concluding message

Our data show hypertrophy of the urethral smooth muscle distal to the ligation and alteration in the contractile characteristics, innervation, and myosin isoform composition are associated with PBOO-induced bladder dysfunction. Supported by George O'Brien Urology Research Center grant P50 DK52620.

