# 48A

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**Title:** MESSENGER RNA EXPRESSION OF VANILLOID RECEPTOR SUBTYPE 1 IN UROTHELIUM AND SMOOTH MUSCLE AFTER BLADDER OUTLET OBSTRUCTION IN RAT

**Aims of the Study**

Bladder outlet obstruction (BOO) results in significant changes in bladder structure and function [1] including detrusor hypertrophy/hyperplasia, elevated voiding pressures and detrusor instability and irritability. Sensory symptoms of BOO such as bladder irritability are attributable in part to changes in the properties of afferent neurons [2]. In rats, the bladder afferent neurons affected by BOO appear to be the small size, capsaicin-sensitive neurons that have unmyelinated (C-fiber) axons. Although the urothelium is generally thought to function primarily as a barrier to block the penetration of urinary constituents into the bladder, recent studies have suggested that the urothelium also has “neuronal-like” properties. Like afferent nerves, the urothelial cells also express vanilloid receptors and are able to respond to capsaicin and protons [3]. This raises the possibility that VR1 containing urothelial cells may participate in nociceptive as well as non-nociceptive sensory mechanisms in the lower urinary tract.The present study was undertaken to investigate whether bladder outlet obstruction (BOO) in rats alters the expression of the capsaicin receptor in urothelium following bladder hypertrophy, and whether similar changes in the expression of capsaicin receptor occur in the urothelium and smooth muscle.

**Methods**

Female rats were divided into non-operated, sham control, and 1, 4, 7, and 14 days BOO groups. The bladder body of each rat was excised at the level of the ureteral orifices, weighed, and the urothelium was dissected away from muscle layer. Messenger RNA (mRNA) expression of vanilloid receptor subtype 1 (VR1) in the urothelium and smooth muscle tissue was analyzed by semi-quantitative reverse transcriptase polymerase chain reaction.

**Results**

The bladder weight 1 day after BOO was similar to that of control animals. Thereafter, the bladder weight gradually increased (approximately 400 % increase). The levels of VR1 mRNA were not altered at 1 day or 4 days after BOO. However, in smooth muscle, the level of VR1 mRNA increased significantly at 7 days after BOO as compared with control group. This increase was not detected at 14 days after BOO. In urothelium, the level of VR1 mRNA increased significantly 7 days after BOO and remained elevated at 14 days. The VR1 expression in the urothelium at 7 and 14 days were not significantly different.

**Conclusions**

These data suggest that VR1 might be involved in the sensory functions of the urothelium and that these functions might be altered by BOO. In addition, the VR1 may contribute to the pathophysiology of urothelium and smooth muscle associated with bladder outlet obstruction.

**References**

1. Effects of acute in vitro overdistension of the rabbit urinary bladder on DNA synthesis. J Urol 148:1347-1350,

1992.

2. Alterations in afferent pathways from the urinary bladder of the rat in response to partial urethral obstruction. J Comp Neurol 310:1-10, 1991.

3. Evidence for functional VR1 receptors in bladder epithelial cells. Soc Neurosci Abstr 26:1688, 2000.

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# 48B

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**Title:** CHANGES OF NERVE GROWTH FACTOR EXPRESSION IN UROTHELIUM AND MUSCULAR TISSUE OF THE URINARY BLADDER AFTER PARTIAL BLADDER OUTLET OBSTRUCTION IN RAT

## Aims of the Study

Nerve growth factor (NGF) is a secretory protein which plays a critical role in the development of the peripheral nervous system including the maturation of sensory pathways. It is also necessary throughout adulthood for maintenance of the normal properties of small size afferent neurons with unmyelinated axons (i.e., C-fiber afferents). Recent studies in adult rat with chronic partial urethral obstruction have demonstrated a role of NGF in target organ-neuronal interactions leading to neural plasticity [1]. Unmyelinated, VR1-containing afferent nerves are located near the luminal surface of the bladder in close proximity to and extending into the urothelium [2]. In addition to its barrier function the urothelium appears to have “neuronal-like” properties which allow it to respond to and release neurotransmitters which could in turn influence the activity of adjacent afferent nerves. Thus it is possible that bladder outlet obstruction (BOO) also alters the chemical signaling/sensory functions of the urothelium. This study was performed to investigate the changes in NGF mRNA and NGF protein levels in the urothelium and the muscular tissue of the rat bladder 1 to 14 days after partial BOO.

## Methods

Bladders were removed from non-operated, sham operated control and outlet obstructed female Wistar rats 1, 4, 7, and 14 days after partially ligating the urethra. After weighing the bladder, the urothelium was separated from the smooth muscle tissue and NGF mRNA was analyzed in both tissues by semi-quantitative reverse transcriptase polymerase chain reaction. NGF protein levels were measured by an enzyme linked immunosorbent assay in both tissues.

## Results

One day after BOO bladder weights (80 mg) were similar in control and BOO animals. Thereafter, the bladder weight gradually increased in BOO animals reaching 3-4 times control values at 14 days after obstruction. The levels of NGF mRNA in urothelium and muscle tissue increased significantly at 4 and 14 days, but not at 1 and 7 day after BOO. The concentration of NGF protein in muscle tissue increased significantly only at 7 days after BOO but did not change in the urothelium at any time point.

## Conclusions

Parallel increases in the NGF mRNA expression in urothelium and bladder muscle tissue after BOO indicate that NGF arises from multiple sources in the hypertrophied bladder. This change in NGF may lead to increased afferent nerve activity and contribute to irritative symptoms accompanying outlet obstruction. However, mRNA expression was not reflected in the measurements of NGF protein levels which increased only in muscle tissue at one time point. Thus, NGF protein levels may be stabilized by neuronal binding/uptake of free NGF and may

not be useful in predicting the pathophysiological consequences of upregulated NGF gene expression.

**References**

1. Immunity to nerve growth factor prevents afferent plasticity following urinary bladder hypertrophy. J Urol, 155: 379-385, 1996.

2. Evidence for functional VR1 receptors in bladder epithelial cells. Soc Neurosci, 26:1688, abstract 632.4, 2000.

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