

## UROTHELIUM IS THE PRIMARY SOURCE OF ATP AND NO RELEASE IN THE RAT URINARY BLADDER: A NOVEL METHOD FOR AN UROTHELIUM FREE PREPARATION

### Hypothesis / aims of study

ATP and NO are released from both the urothelium and smooth muscle in the urinary bladder. The purpose of these experiments was to determine the relative contribution from each layer releasing ATP and NO.

### Study design, materials and methods

All animal experiments were carried out in accordance with the National Institute of Health Guidelines for the Care and Use of Laboratory Animals, and were approved by the Institutional Animal Care and Use Committee. 250-300g Sprague Dawley rats were used. The bladder was removed under isoflurane anesthesia, opened and halved. One half of the bladder was gently and evenly rubbed with a cotton swab while the other half was left intact. Two strips were prepared from each half and mounted in a heated organ bath in Krebs solution. Both neurally evoked contractions and NO and ATP release were measured.

### Results

The amplitude of the neurally evoked contractions (20Hz/100 shocks) and the carbachol (CCh) (50 uM) evoked contractions of the urothelium free strips were significantly smaller (by 43% and by 49% respectively) than that of the intact strips. Although the basal release of ATP was identical, both electrical stimulation and CCh (50 uM), evoked release were significantly reduced in the urothelium-free preparations (by 81% and by 85%, respectively). The microscopic picture of the swabbed preparation showed the complete lack of the urothelial layer while the smooth muscle layer was normal. There was no change in the basal and electrical stimulation evoked NO release in the urothelium-free preparations, however, the carbachol evoked NO release was reduced by 40 %.

### Interpretation of results

Swabbing the urothelial layer results in an urothelium-free preparation. These results clearly show that most of the ATP and a significant portion of NO derives from the urothelium. In addition, the contractile response of the strips was also diminished indicating that the urothelium positively interacts with the efferent system.

### Concluding message

This information is noteworthy because there are significant numbers of sensory and efferent terminals in the smooth muscle layer that also utilize ATP and NO as neurotransmitters.

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<b>What were the subjects in the study?</b>	<b>ANIMAL</b>
<b>Were guidelines for care and use of laboratory animals followed or ethical committee approval obtained?</b>	<b>Yes</b>
<b>Name of ethics committee</b>	<b>Baylor College of Medicine/Institutional Animal Care and Use Committee</b>