

## MODELING THE EFFECT OF NITRIC OXIDE UPON INFLAMMATION IN THE RAT URINARY BLADDER

### Hypothesis / aims of study

Painful bladder syndrome is somewhat of an umbrella term for chronic inflammatory diseases characterized by urgency, urinary frequency and pain. One of these diseases is interstitial cystitis. Many of the symptoms of interstitial cystitis are directly associated to changes occurring in the bladder contractile response. A common way to induce a syndrome similar to interstitial cystitis is by injection with the alkylating agent cyclophosphamide, a drug often used in cancer treatment. In the current study, the aim was to study the effect of nitric oxide upon cholinergic and purinergic contraction during cyclophosphamide-induced inflammation in the rat urinary bladder. Further, the data were modelled in order to detect minute changes which would otherwise be indiscernible.

### Study design, materials and methods

A total of 48 male rats were used in the study, of which 24 were injected intraperitoneally with cyclophosphamide (100 mg/kg). The healthy and cyclophosphamide-injected rats were further divided into two treatment groups of 12 rats per group. For a statistical power of 80% ( $\alpha$ -error 0.05) a sample size of at least 8 rats per groups was predicted. The contractile response of the urinary bladder to methacholine (MeCh), a muscarinic receptor agonist, and adenosine-5'-triphosphate (ATP), in the presence and absence of L-NG-monomethyl arginine citrate (L-NMMA), a non-selective nitric oxide synthase inhibitor, was studied in an organ bath setup. The concentration-response data was modeled by using WinNonlin Phoenix, Pharsight Professional Software version 6.2, to find a model that describes the data adequately and which could detect minute changes upon inflammation.

### Results

In cyclophosphamide pre-treated rats, both the cholinergic and purinergic maximal contraction was significantly lower than in normal rats in the absence of L-NMMA. In cyclophosphamide pre-treated rats, the methacholine-evoked contractile response curve was shifted to the right in the presence of a low dose L-NMMA (1  $\mu$ M), but in the presence of a high dose of L-NMMA (10  $\mu$ M) a slight left-shift of the response curve occurred. In the control group, a right-shift was seen in the presence of both doses L-NMMA. In both control and inflamed rats, the maximal contraction evoked by ATP was lower in the presence of L-NMMA (1  $\mu$ M), but in the presence of a high dose of L-NMMA (10  $\mu$ M) an increase of the maximum contraction was seen.

### Interpretation of results

As shown previously, both the cholinergic and purinergic contractile function of the urinary bladder is altered upon inflammation, leading to a decrease in maximal contraction of the bladder. This is likely due both to changes in receptor expression, structural detrusor damage caused by the inflammation and influence by factors arising during the inflammation, such as nitric oxide. The current study shows that nitric oxide affects both cholinergic and purinergic contraction in the rat bladder, more during inflammation than in the normal state. Further, our data indicate dual roles of nitric oxide, as it has varying effects upon contraction and when given in a high or low dose. Possibly this duality is due to separate pools of nitric oxide synthase, generating opposing effects in different parts of the urinary bladder.

### Concluding message

The current study gives rise to the possibility that nitric oxide can act in both a facilitatory and inhibitory fashion in the rat urinary bladder during inflammation, affecting both cholinergic and purinergic contraction.

### Disclosures

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