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TIME-DEPENDENT CHANGES IN NITRIC OXIDE SYNTHASE ISOFORMS IN THE SPINAL CORD IN A RAT MODEL OF DETRUSOR OVERACTIVITY INDUCED BY BLADDER IRRITATION

Hypothesis / aims of study
Considerable evidence has demonstrated that peripheral inflammation can up-regulate the expression of neural nitric oxide synthase (nNOS) in the spinal cord immediately and then induce the expression of inducible NOS (iNOS) in the late phase (1). Spinal nNOS-dependent processes have also been implicated in detrusor overactivity associated with acute chemical irritation and inflammation of the lower urinary tract (2). However, little is known about the role of iNOS in spinal sensitization inducing detrusor overactivity. Therefore, we examined the role of nNOS and iNOS in an early phase (acute irritation by direct application of acrolein that is active metabolite of cyclophosphamide [CYP]) and a late phase (24h after intraperitoneal injection of CYP) of chemical cystitis.

Study design, materials and methods
Continuous cystometrograms (CMG, 0.1ml/min) was performed in female Sprague-Dawley rats under urethane anesthesia (1.2 g./kg., s.c.). Some rats received single intraperitoneal injection of CYP (200mg./kg.) 24hours before CMG (CYP group). In another group, following baseline CMG recordings with saline (0.1ml/min for 60min), 0.05 mg/ml acrolein was infused (acrolein group). Intrathecal (i.t.) injections of non-selective NOS [NG-nitro-L-arginine methyl ester (L-NAME)], inducible NOS [N-(3-(aminomethyl)benzyl)acetamidine (1400W) and 2-amino-5,6-dehydro-6-methyl-4H-1,3-thiazine (AMT)], neural NOS [N^{ω}-propyl-L-arginine (NPA)] and endothelial NOS [N^{5}-(1-iminoethyl)-L-ornithine (L-NIO)] inhibitors were tested in the CYP or acrolein group as well as the control group during saline infusion. Also, we measured the Ca^{2+}-dependent (neural and endothelial) and independent (inducible) NOS enzyme activities in the lumbar spinal cord (L6-S1) segments in CYP-treated and control rats.

Results
Bladder capacity (BC) in CYP (0.16±0.04 ml) and acrolein (0.29±0.05 ml) groups was significantly lower than that in the control group (0.68±0.09ml). I.t. injection of L-NAME (1 to 30μg) significantly increased BC in CYP and acrolein groups does-dependently without changes in the control group. I.t. injected 1400W and AMT as iNOS inhibitors significantly increased BC in the CYP group, but not in the acrolein group, while i.t. injected NPA as a nNOS inhibitor significantly increased BC in both groups. However, in the CYP group, increasing effect of NPA on the BC was significantly smaller than that of 1400W. L-NIO as an endothelial NOS inhibitor did not affect BC in either group. Both Ca^{2+}-dependent and independent NOS enzyme activities in the lumbar spinal cord segments in CYP-treated rats were significantly higher than those in control rats.

Interpretation of results
These findings indicate that spinal nNOS contributes to detrusor overactivity caused by acute chemical irritation (early phase) and CYP-induced tissue inflammation (late phase), while spinal iNOS plays an important role only in the late phase.

Concluding message
Our results suggest that endogenous NO produced by different NOS isoforms in the spinal cord can enhance afferent transmission resulting in detrusor overactivity and provide a basis for the potential use of different types of NOS antagonists for the treatment of detrusor overactivity/pain conditions.

References
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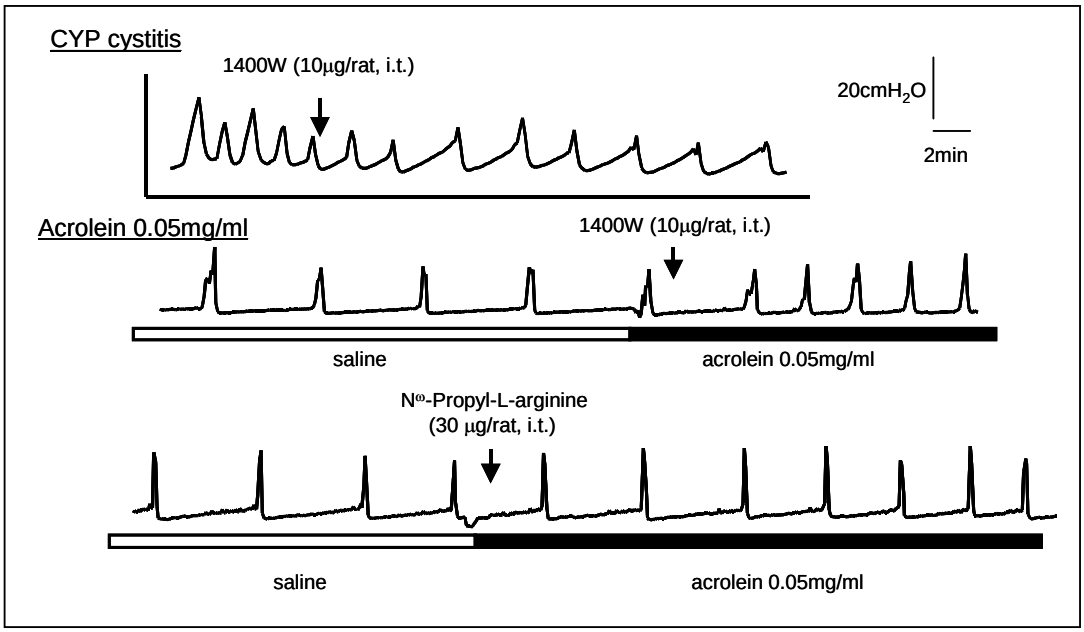
Fig. 1 Effects of intrathecal (i.t.) administration of 1400W, a iNOS inhibitor, or N^{ω}-propyl-L-arginine, a nNOS inhibitor, on the detrusor overactivity caused by CYP or acute chemical irritation with acrolein



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