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BLADDER PAIN SYNDROME/INTERSTITIAL CYSTITIS: IS THIS CONDITION ASSOCIATED WITH A SYMPATHETIC DYSFUNCTION?

Hypothesis / aims of study

Bladder Pain Syndrome/Interstitial cystitis (BPS/IC) courses with suprapubic pain related to bladder filling, usually accompanied by frequency and nocturia, in the absence of urinary infection or other local pathology. BPS/IC aetiology is unknown. Several hypotheses have been proposed, such as a dysfunctional, thinner urothelium with a slow turnover, more permeable to toxic urine components, a defect of the glycosaminoglycan layer, infection by unknown bacteria, autoimmune mechanisms and neurogenic inflammation, among others.
The role of the autonomic nervous system in the development of BPS/IC has not been object of systematic investigation. Nevertheless, chronic bladder inflammation promotes nerve fibre sprouting (1). High levels of urinary catecholamines were reported in BPS/IC patients (2). Furthermore, OnabotA injection in the trigone of BPS/IC patients showed a decrease in pain and frequency, raising the possibility that impairment of sympathetic fibres, almost exclusively located in the trigone, could play a role in this improvement (3). All together, these data may suggest that BPS/IC courses with an abnormal sympathetic outflow to the bladder.
In this study we aimed to investigate the effect of a chronic adrenergic stimulation of naive rats in their bladder morphology and function, as well as the effect of chronic bladder inflammation of the activity of the bladder sympathetic system

Study design, materials and methods

Two groups of rats were injected with phenylephrine (PHE) 0.5mg/kg or 2.5mg/kg (s.c., daily for 14 days). At day 15, cystometries were performed for two hours, while saline was infused at a 6 ml/h rate. Afterwards, the bladder of animals was harvested and fixed by immersion in buffered formalin, and the whole animals were perfusion fixed and the L6 dorsal root ganglia (DRG) and L6 spinal cord were harvested. These same treatments were performed in rats treated with capsaicin 100 mg/kg, (s.c., 24h prior to PHE treatment).

The bladders were stained for haematoxylin-eosin, Periodic-Schiff acid and Toluidine Blue and immunoassayed for Bax, Caspase 3 and α1 adrenoreceptor+TRPV1. The L6 spinal cord segment was immunoreacted for Fos and L6 DRGs against the α1 adrenoreceptor.

In another group, animals were intravesically instilled with 2mg/kg lipopolysaccharide (LPS), for one hour. Twenty-four hours later, the urine of these animals was collected and the urinary bladder harvested and fixed by immersion in paraformaldehyde. The density of sympathetic fibres was investigated by performing an immunoassay for VMAT2 and the urinary levels of norepinephrine (NE) were determined by ELISA.

Results

PHE-treated animals presented an increase in micturition frequency (1.12±0.23 contractions/minute for PHE 0.5 mg/Kg, $p<0.01$; 1.47±0.24 contractions/minute for PHE 2.5 mg/kg, $p<0.001$) when compared with controls (0.43 ± 0.11 contractions/minute). The urinary bladder of PHE-treated animals was lined by a thin urothelium (25,99 µm height for PHE 0.5 mg/kg, $p<0.001$; 21,30 µm in height for PHE 2.5 mg/kg, $p<0.001$) when compared with controls (40.39 µm in height). Urothelial cells had a strong positive staining for the pro-apoptotic proteins Caspase 3 and Bax. The bladder wall was infiltrated by mast cells (21.32 mast cells/mm^2 for PHE 2.5 mg/kg and 8.25 mast cells/mm^2 for control). A double staining for α1 adrenergic receptor + TRPV1 was observed in nerve terminals in the bladder mucosa and in L6 DRG neurons of both control and PHE-treated animals. An intense α1 labelling was observed on urothelial cells. When compared with controls (9±6 Fos expressing cell/section), PHE-treated rats had an increase in spinal cord Fos expression (22±6 cFos expressing cell/section for PHE 0.5 mg/kg, $p<0.001$; 36±11 Fos expressing cell/section for PHE 2.5 mg/kg, $p<0.001$).

Systemic capsaicin treatment abolished the PHE-induced increase in micturition frequency and in spinal cord Fos expression.

The urine of LPS-inflamed animals had a massive increase in NE levels when compared to control group (2341,2 vs 3 µg/g creatinine, $p<0.0001$). LPS-inflamed bladders had more VMAT2 positive fibres, due to an increase of these fibres in the mucosa and in the detrusor of the bladder dome and body. No changes in VMAT2 fibre number could be seen in the trigone.

Interpretation of results

A strong prolonged adrenergic stimulation induces profound changes in the urothelium, including urothelium thinning and activation of the apoptotic cascade. This might increase urothelial permeability leading to inflammatory changes, including mast cell infiltration, a characteristic present in the bladder of BPS/IC patients. Interestingly, urothelial cells exhibited a strong immunoreaction for α1 receptors.
The PHE induced increased in micturition frequency and in L6 spinal Fos expression, a surrogate marker of bladder pain, was mediated by C-fibres, since systemic capsaicin pre-treatment, which knocks down the effect of PHE treatment. The observed effect occurs probably by intracellular cross-talk between α adrenoreceptors and TRPV1, since these two receptors are co-expressed in bladder sensory fibres.
Inflammation might, on the other hand, further enhance sympathetic drive. The observed sprouting of sympathetic fibres might be responsible for the increase in the urinary levels of catecholamines. Furthermore, the sprouting of sympathetic fibres throughout the bladder body and dome makes possible a new cross-talk between these fibres and sensory fibres. NE release from sympathetic fibres can sensitize bladder nociceptive fibres as TRPV1 immunoreactive fibres also express α1 adrenergic receptors.

Concluding message

Chronic adrenergic stimulation induced signs similar to the ones observed in BPS/IC patients. Therefore, future studies should investigate sympathetic dysfunction in patients with BPS/ IC. In addition, chronic adrenergic stimulation of naive rats may constitute a new, valuable experimental model to study BPS/IC in rodents.

References

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