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Yamada S[1], Yoshida A[1], Kageyama A[1], Mori F[1], Ito Y[1]
***1.*** *University of Shizuoka*

# MUSCARINIC AND PURINERGIC RECEPTORS IN THE RAT BLADDER ARE ALTERED BY CHEMICALLY INDUCED CYSTITIS.

Hypothesis / aims of study

Interstitial cystitis (IC) is a chronic, abacterial inflammatory disease of the urinary bladder characterized by urinary frequency, urgency and suprapubic pain associated with bladder filling and relieved by voiding [1], but its exact etiology and pathogenesis remain unclear. Currently, there are increasing evidences to suggest the idea that the abnormality of muscarinic and purinergic signaling transduction in the bladder is implicated in the development of interstitial cystitis [2, 3]. In this regard, it is of interest to note that expressions of $P2X_2$ and $P2X_3$ receptors were significantly altered in the bladder urothelium of patients with interstitial cystitis [3]. This study was conducted to clarify the mechanisms involved in the pathophysiology of IC by measuring muscarinic and purinergic receptors and urodynamic parameters in the bladder of rats with cystitis induced by HCl, cyclophosphamide (CYP) and protamine (Pro)

Study design, materials and methods

The mechanical responses of bladder in cystitic rats were monitored by the cystometric method under an anaesthesia. Then, rats were sacrificed by the exsanguination from descending aorta, and the bladder was excised. Muscarinic and ATP receptors in the rat bladder homogenates were measured by measuring specific binding of [$^3$H]NMS and [$^3$H]□ □-MeATP, and binding parameters of apparent dissociation constant ($K_d$) and maximal number of binding sites ($B_{max}$) for [$^3$H]NMS were estimated by nonlinear regression analysis using Graph Pad Prism.

Results

Compared with that of sham rats, the body weight of HCl- and Pro-treated rats was similar and that of CYP-treated rats was significantly decreased. The bladder weights of HCl- and CYP-treated rats were significantly greater than those of sham rats, while they were not significantly altered by Pro treatment. In the cystometry of these cystitic rats compared with sham rats, micturition interval and micturition volume were significantly decreased and the frequency of micturition was significantly increased (Fig. 1). The $B_{max}$ values for specific [$^3$H]NMS binding were significantly (55%, 40% and 16%, respectively) decreased in the bladder of HCl-, CYP- and Pro-treated rats compared with sham rats (Table 1). Similarly, the $B_{max}$ values for specific [$^3$H]□ □-MeATP binding were significantly (72% and 30%, respectively) decreased in the bladder of HCl- and CYP-treated rats compared with sham rats, but specific [$^3$H]□ □-MeATP binding was not altered by Pro treatment. On the other hand, the $K_d$ value for specific binding of [$^3$H]NMS and [$^3$H]□ □-MeATP in the bladder was not significantly alterd in cystitic rats except for that for specific [$^3$H]□ □-MeATP binding in the bladder of HCl-treated rats (Table 1).

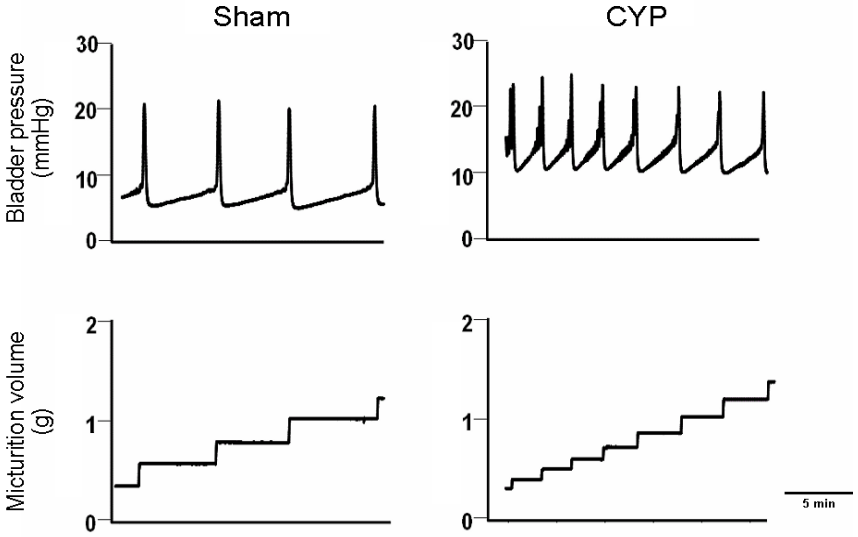


Fig. 1. Representative cystometry traces in rats with vehicle (sham) and CYP-induced cystitis

Table 1. $K_d$ and $B_{max}$ for specific binding of [$^3$H]NMS and [$^3$H]□ □-MeATP in the bladder of rats with HCl, cyclophosphamide (CYP) and protamine (Pro)-induced cystitis

| | [$^3$H]NMS | | [$^3$H]□ □-MeATP | |
|---|---|---|---|---|
| | $K_d$ (pM) | $B_{max}$ (fmol/mg protein) | $K_d$ (pM) | $B_{max}$ (pmol/mg protein) |
| HCl-treated rat | | | | |
| Sham | 264 ± 20 | 98.7 ± 7.5 | 532 ± 33 | 9.55 ± 0.93 |
| HCl | 227 ± 13 | 44.1 ± 3.1*** | 715 ± 57* | 2.66 ± 0.21*** |
| CYP-treated rat | | | | |
| Sham | 255 ± 20 | 84.8 ± 4.6 | 853 ± 126 | 9.91 ± 0.87 |
| CYP | 278 ± 23 | 51.0 ± 5.0*** | 940 ± 85 | 6.89 ± 0.95* |
| Pro-treated rat | | | | |
| Sham | 329 ± 23 | 141 ± 10 | 1735 ± 274 | 9.23 ± 1.34 |
| Pro | 290 ± 18 | 119 ± 6* | 1511 ± 234 | 8.67 ± 1.14 |

Values are expressed as mean±S.E. (n=6-9). Asterisks show a significant difference from the values in control, *P<0.05, ***P<0.001.

Interpretation of results

Our data revealed down-regulation of both muscarinic and purinergic receptors with the induction of overactive bladder in the bladder of rats with chemically induced cystitis. Furthermore, the alteration of pharmacologically relevant receptors in Pro-treated rats compared with HCl- and CYP-treated rats seemed to be mild. Thus, muscarinic and ATP receptors may be partly involved in the pathophysiology of cystitis.

Concluding message

Pharmacologically relevant (muscarinic and ATP) receptors may be down-regulated in the bladder of rats with chemically induced cystitis.

References

1. J Am Acad Nurse Pract 15: 64-71, 2003
2. Auton Neurosci 122: 9-20
3. BJU International 93: 1344-1348, 2004



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|---|---|
| ***Specify source of funding or grant*** | **None** |
| ***Is this a clinical trial?*** | **No** |
| ***What were the subjects in the study?*** | **ANIMAL** |
| ***Were guidelines for care and use of laboratory animals followed or ethical committee approval obtained?*** | **Yes** |
| ***Name of ethics committee*** | **This study was done in accordance with recommendation of the Experimental Animal Ethical Committe of the University of Shizuoka.** |