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EFFECTS OF ESTROGEN RECEPTOR ACTIVATION ON PROSTATIC INFLAMMATION AND BLADDER OVERACTIVITY IN A RAT MODEL OF CHRONIC NONBACTERIAL PROSTATITIS

Hypothesis / aims of study
Although it is known that chronic nonbacterial prostatitis (CNBP) is one of the common urological diseases among men, the treatment of CNBP is often unsuccessful. CNBP often causes irritative lower urinary tract symptom such as frequent urination or urgency. It has recently been reported that estrogen receptors (ERs), ERα and ERβ, modulate tissue inflammatory conditions and that ERβ stimulation can improve inflammation whereas ERα stimulation could be an accelerator of local inflammation. In this study, we investigated the role of ERβ in prostatic inflammation and bladder overactive condition using a rat model of CNBP since ERβ has been recognized as a therapeutic target for inflammation diseases in the skin or the central nerve system [1, 2].

Study design, materials and methods
Male Sprague–Dawley rats at age of 8 weeks old were used. In the first set of experiments, rats were divided into formalin injection prostatitis (FG, n=5) and saline injection control groups (SG, n=5). Prostatic inflammation was induced by 5% formalin injection into bilateral ventral lobes of the prostate. In another set of experiments, rats with formalin-induced prostatitis were divided into the ERβ agonist therapy group (TG, n=4) and the placebo group (PG, N=4). TG rats was treated with 3αdiol, which is a selective agonist for ERβ, dissolved in olive oil at a dose of 3mg/kg daily from 2 days before induction of prostatitis for 30days whereas PG rats received olive oil only. In each group, continual filling cystometry was performed in a conscious condition on day 28 after induction of prostatitis (FG, TG, PG groups) or sham treatment (SG group). Urodynamic parameters including non-voiding contractions (NVCs) during the storage phase, voiding interval (VI) and postvoid residual volume (RV) were investigated. After cystometry, the prostate and bladder were excised, and the bladder was separated into mucosa and detrusor muscle layers under a microscope. Expression levels of P2X2 and TRPA1 mRNA in bladder mucosa and detrusor layers as well as mRNA expression levels of ERα, ERβ, TNF-α, iNOS, and COX2 in the prostate were investigated by real-time PCR in each group. Statistical analysis was performed using Mann-Whitney U test. P value less than 0.05 was considered statistically significant.

Results
In cystometric investigation, the mean number of NVCs was significantly greater in FG rats than in SG rats ($P<0.05$), and VI were significantly decreased in FG rats compared with SG rats ($P<0.05$) (Figure 1). There was no significant difference in RV between FG and SG groups. In RT-qPCR analyses, mRNA expression of P2X2 and TRPA1 receptors was significantly increased in the bladder mucosa but not in the detrusor in FG rats compared with SG rats ($P<0.05$). In the prostate, expression levels of ERα, TNF-α, iNOS, and COX2 mRNA were significantly increased in FG rats compared with SG rats ($P<0.05$). However, expression of ERβ in the prostate was significantly decreased in FG rats compared with SG rats ($P<0.05$).
In comparison between TG and PG prostatitis groups, TG rats showed decreased NVCs and increased VI ($P<0.05$) (Figure1), and mRNA expressions of P2X2 and TRPA1 in the bladder mucosa as well as those of ERβ, TNF-α, iNOS, and COX2 in the prostate were significantly decreased in TG rats compared with PG rats (Figure2). Furthermore, ERβ mRNA expression in the prostate increased in TG rats compared with PG rats ($P<0.05$) (Figure2). The relative expression ratio of ERβ against ERα in FG rats was significantly decreased compared with SG rats ($P<0.05$); however, activation of ERβ by 3αdiol reversed the decreased ERβ/ERα ratio in the TG group compared with the PG group ($p<0.05$).

Interpretation of results
Patients with CNBP often exhibit irritative bladder symptoms such as urinary frequency or urgency. In this study, prostatitis rats showed bladder overactive conditions evidenced by increased NVCs and decreased VI and also increased expression of P2X2 and TRPA1 in the bladder mucosa after prostatic inflammation, suggesting that the prostate-to-bladder cross-organ sensitization might be involved in the induction of bladder overactivity associated with upregulation of ATP (P2X2) and TRPA1 receptors in the bladder following CNBP. This study also showed that ERβ expression in the prostate is reduced after prostatitis and that ERβ stimulation by 3αdiol in prostatitis rats (TG group) improved not only prostatic inflammation evidenced by decreased expression TNF-α, iNOS and COX2, which is associated with increased ERβ/ERα ratio in the prostate, but also bladder overactivity as shown by decreased NVCs and increased VI in association with normalization of P2X2 and TRPA1 mRNA expression in the bladder mucosa when compared with placebo-treated prostatitis group (PG). These results indicate that ERβ activation that can normalize ERβ expression in the prostate has anti-inflammatory effects to reduce bladder overactivity and expression of the related molecules in the bladder as well as to improve prostatic inflammation.

Concluding message
We demonstrated that: (1) CNBP in rats induces prostatic inflammation that involves cytokine production (TNF-α) and upregulation of nitric oxide (iNOS) and prostaglandin (COX2) production systems and also bladder overactivity, possibly due to prostate-to-bladder cross-organ sensitization, and (2) ERβ activation effectively improves not only prostatic inflammation but also overactive bladder conditions, and normalizes ERβ expression in the prostate. Therefore, ERβ could be a therapeutic target for the treatment of prostatic inflammation and irritative bladder symptoms in patients with CNBP.

Figure1

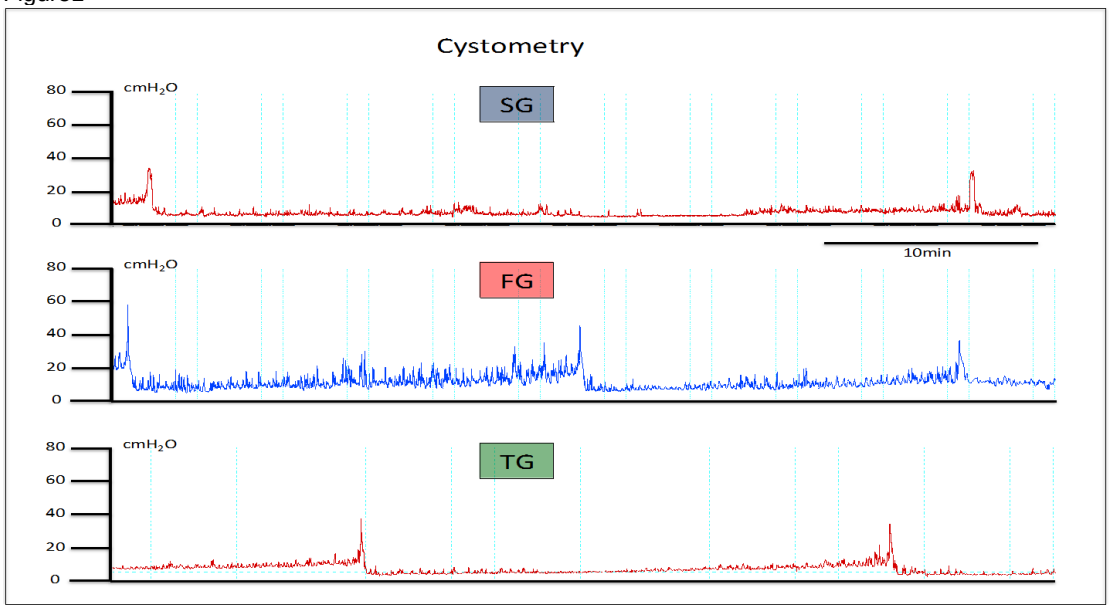
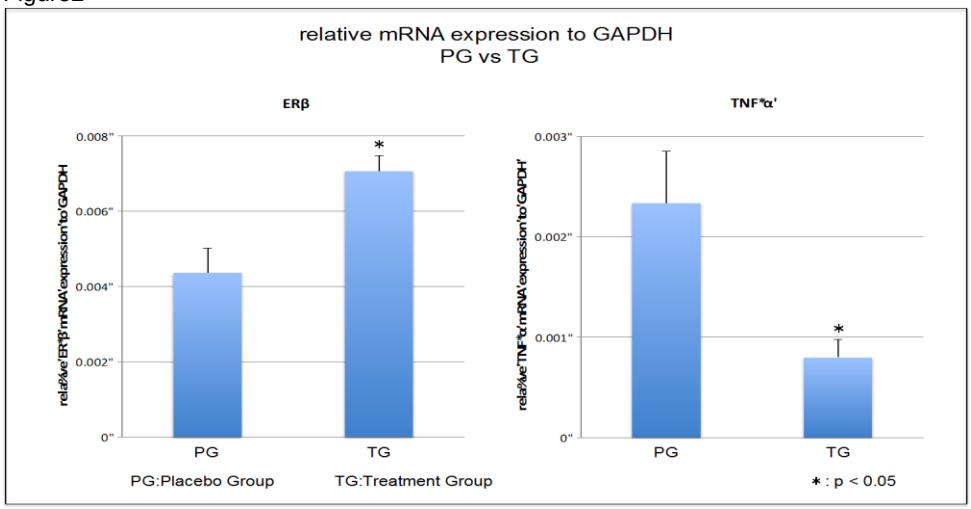


Figure2



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Disclosures

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