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BAY 41-2272, A SOLUBLE GUANYLATE CYCLASE STIMULATOR, IMPROVES BLADDER FUNCTION IN PARTIAL BLADDER OUTLET OBSTRUCTED MICE

Hypothesis / aims of study

Bladder outlet obstruction (BOO) results in an impaired ability of the urinary bladder to store and empty urine. In animal models, BOO results in bladder dysfunction and alterations of the contractile machinery in detrusor smooth muscle (DSM), similar to those found in men with Benign Prostatic Hyperplasia [1], and can be associated to increased oxidative stress which can inactivate nitric oxide (NO) and inhibit activity of soluble guanylate cyclase (sGC). A class of agents called sGC stimulators and activators has been studied for improving enzymatic conversion of GTP to cyclic GMP, and promoting smooth muscle relaxation [2]. The sGC stimulator BAY 41-2272 (5-Cyclopropyl-2-[1-(2-fluoro-benzyl)-1*H*pyrazolo[3,4-*b*]pyridine-3-yl]pyrimidin-4-ylamine) is to effective in producing cavernosal relaxations in rabbits and human [3], and could be used to ameliorate bladder function when urethral obstructive process is present. This study aimed to evaluate bladder function in mice with BOO treated chronically with BAY 41-2272.

Study design, materials and methods

All animal procedures were approved by the Ethical Principles in Animal Research. C57BL/6 male mice (28-32 g) were anaesthetized with xilazine (30 mg/kg i.p.) and ketamine (2 mg/kg i.p.) for surgical procedure. BOO was performed using nylon ligature in the bladder neck. Four groups with 6 to 9 animals per group were obtained: Sham + vehicle, Sham + BAY, BOO + vehicle and BOO + BAY. Vehicle (transcutol, chremofor and water at 1:2:7 parts, respectively) and BAY 41-2272 (1.3 mg/ml, approximately 10 mg/Kg) were given by gavage (0.2 ml/animal) from 3 to 5 weeks after surgery. At 5 weeks, *in vivo* and *in vitro* studies were performed. Continuous cystometry was carried out by infusing saline into the bladder at a rate of 0.6 mL/h for 30 min after the first micturition cycle in anaesthetized mice. In separate experimental groups, bladder and body weights were obtained and bladder weight to body weight ratio was calculated. Bladder contractility was also evaluated *in vitro* by constructing concentration-response curves to carbachol (CCh; 1 nM – 30 µM), KCl (1 mM – 1 M) and electrical-field stimulation (EFS; 2-16 Hz, 50 V, 0.2 ms, 10 s interval). Relaxant responses were obtained using the β_3-agonist mirabegron (1 nM – 30 µM).

Results

BOO + vehicle animals had increased bladder mass when compared to Sham + vehicle. BOO + BAY treated mice had less non-voiding contractions and fewer micturition frequency when compared to BOO + vehicle. Compliance was also augmented in BOO + BAY mice in relation to the other groups (Figure 1). Tissue bath protocols revealed that BOO + BAY mice had lower contractile response to CCh when compared to BOO + vehicle (E_{max}: 0.82 ± 0.06 and 1.78 ± 0.18 mN, respectively), but no differences were seen for EFS. BOO+BAY mice had lower contractile responses to KCl when compared to Sham+BAY (E_{max}: 0.9 ± 0.07 and 1.451 ± 0.2 mN, respectively). Mirabegron produced lower bladder relaxations in BOO + vehicle had less compared to Sham+vehicle and BOO+BAY (E_{max}: 49.0 ± 5.1, 85.5 ± 8.8 and 88.3 ± 4.2%, respectively).

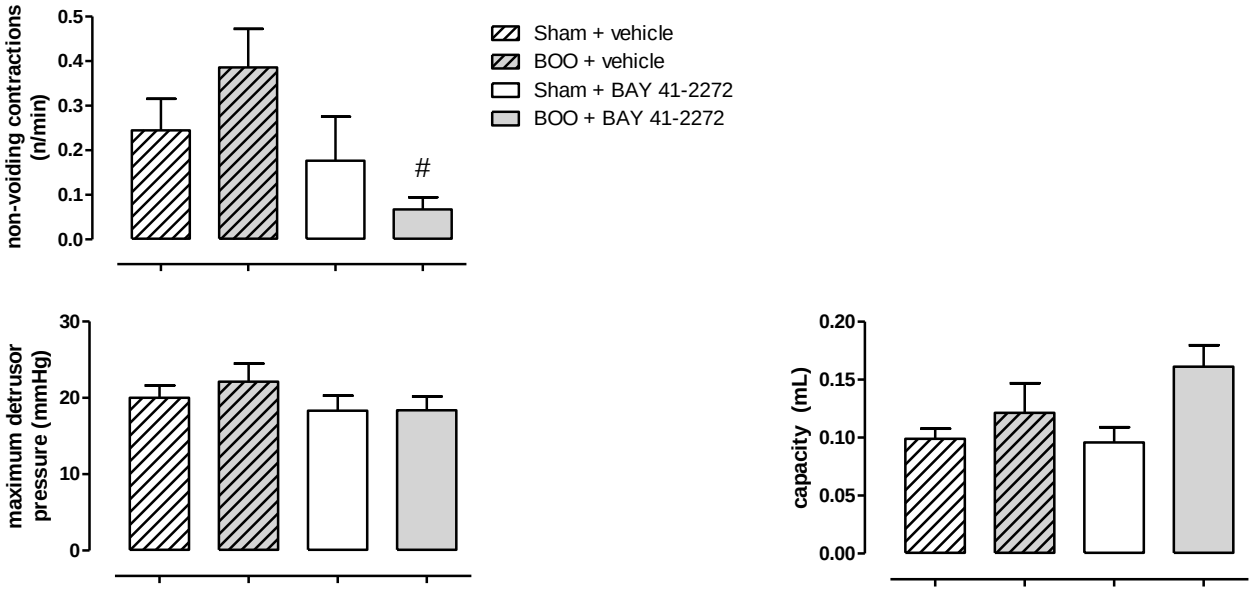

Figure 1. Cystometric parameters in SHAM and BOO treated or not with BAY 41-2272: non-voiding contractions (A), maximum detrusor pressure (B), bladder capacity (C), compliance (D), and micturition frequency (E). Data are shown as mean ± SEM of 3 to 5 experiments per group. *P<0.05 versus Sham. #P<0.05 versus BOO. # #P<0.001 versus BOO.

Interpretation of results

BAY 41-2272 had a protective effect in BOO mice, since it prevented development of hyperactive bladder, as shown in CCh and KCl protocols. Besides, cystometric parameters showed that BAY normalized spontaneous bladder contractions and reduced micturition frequency, suggesting that direct stimulation of sGC in a NO-independent manner by BAY 41-2272 counteracts the bladder dysfunction in BOO mice.

Concluding message

Our results indicate that BAY 41-2272 ameliorates bladder function in obstructed mice, and the decrease in NO bioavailability in bladder could be responsible for detrusor hyperactivity.

References

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Disclosures

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