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PELVIC ORGAN CROSS-SENSITIZATION MODULATES EXPRESSION AND RELEASE OF NEUROPEPTIDES AND NEUROTROPHIC FACTORS IN THE URINARY BLADDER OF FEMALE RATS: ROLE OF THE ESTROUS CYCLE

Hypothesis / aims of study

Painful bladder syndrome and interstitial cystitis affect women more often than men suggesting the role of ovarian hormones in pain perception and transmission. The underlying mechanisms regulating pain fluctuations within a menstrual cycle in women are not fully understood [1]. Recent studies suggested that pelvic organ cross-sensitization triggered by pathology in one of the pelvic organs can lead to functional changes in adjacent organs. It was demonstrated that inflammation of the distal colon causes the development of a neurogenic bladder [2]. We hypothesize that changes in ovarian hormones associated with the estrous cycle will result in changes in the expression of bladder neuropeptides in female rats after colonic inflammation. The objective of this study was to identify major neurotransmitters/ neuropeptides released in the bladder during rising and falling estrogen phases of the estrous cycle using an animal model of a neurogenic bladder. Our secondary aim was to determine if similar differences in urinary neuropeptides and neurotransmitters were noted among premenopausal women with and without bladder pain.

Study design, materials and methods

The stage of the estrous cycle in female Sprague–Dawley rats (N= 12) was determined by histological examination of the cells in daily vaginal lavage. Gene and protein expression of calcitonin gene-related peptide (CGRP), brain-derived neurotrophic factor (BDNF), Substance P (SP), and transient receptor potential vanilloid 1 (TRPV1) was measured using qRT-PCR and ELISA. Tissue samples from the urinary bladder, lumbosacral (L6-S2) spinal cord and dorsal root ganglia were collected from female rats during either diestrus (low estrogen) or proestrus (high estrogen) phases of the estrous cycle in the control group and from animals that underwent acute colonic inflammation. Neuropeptides (CGRP, BDNF, and Substance P) and TRPV1 were also measured using ELISA in urine samples from 5 premenopausal women (mean $\pm$ SD age, 47 $\pm$ 10 years) with painful bladder syndrome and scored > 1 on the Neuropathic Pain Questionnaire (NPQ) which is a self-administered validated tool to measure neuropathic pain [3]. Neuropeptides were also measured in urine samples collected from 6 similarly aged women (mean $\pm$ SD age, 48 $\pm$ 5 years) that did not have complaints of bladder pain and scored $\leq$ 1 on the NPQ. Neuropeptide levels are expressed as mean $\pm$ SEM and were compared using ANOVA.

Results

Comparison of CGRP and SP protein expression in the control group of animals during diestrus and proestrus phases did not reveal significant changes within the estrous cycle. However, acute inflammation in the distal colon caused 2-fold increase in CGRP in the urinary bladder and 30% increase in the spinal cord during proestrus when compared to the diestrus phase ($p < 0.05$). Substance P was similarly increased in the bladder of proestrus animals compared to the diestrus group (1.34 $\pm$ 0.85 pg/ml vs. 0.03 $\pm$ 0.016 pg/ml, respectively, $p < 0.05$). Analysis of the expression of the neurotrophic factor BDNF showed more than a 5-fold increase in the bladder during proestrus in female rats with colonic inflammation as compared to the control group and during the diestrus phase ($p < 0.05$). Gene expression of TRPV1 in the female rat bladder was not affected by colonic inflammation during diestrus phase. In proestrus, TRPV1 mRNA was up-regulated in the rat bladder compared to the diestrus group (3.3 $\pm$ 1.0 vs. 1.1 $\pm$ 0.3 arbitrary units, respectively, $p < 0.05$). Urine from women with painful bladder syndrome and neuropathic pain trended towards a higher expression of Substance P compared to urine from women without bladder pain but this difference did not reach significance (0.0030 $\pm$ 0.0005 vs. 0.0017$\pm$ 0.0002 pg/ml, $P = 0.07$). There were no differences noted in urinary CGRP, BDNF, or TRPVI peptide levels between groups of women ($p \geq 0.50$).

Interpretation of results

Active colonic inflammation trigger an increased release of neurotrophic factors and neuropeptides during proestrus phase in the female rat bladder. The absence of similar differences noted in urinary neuropeptide expression between women with and without bladder pain may, in part, be explained by the fact that they were not examined with respect to their menstrual cycle phase (luteal vs. follicular). The menstrual cycle in women with bladder pain may be an important factor in urinary neuropeptide and neurotransmitter expression.

Concluding message

Taken together, these data suggest that ovarian hormones may modulate painful sensations in women with bladder pain of neurogenic origin and further research is required to fully elucidate these mechanisms.

References

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Specify source of funding or grant	**NIH/NIDDK DK077699, DK077699-S2, and NIH/NICHD Women's Reproductive Health Research Award K12- HD00165-11**
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	**University of Pennsylvania Institutional Review Board (for humans) #809736, and** **University of Pennsylvania Institutional Animal Care and Use Committee apprival # 802389**