STEROID HORMONE IMBALANCE STIMULATES OSTEOPONTIN EXPRESSION AND INFLAMMATION IN THE MOUSE PROSTATE

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Background

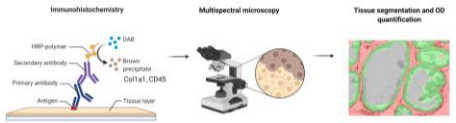
Inflammatory processes in the prostate are linked to the development of lower urinary tract symptoms (LUTS) in men. Steroid hormone imbalance is a major age-associated factor driving LUTS, but its relationship to inflammation is not fully understood. Our previous studies identified that osteopontin (OPN), a pro-inflammatory cytokine, is abundant in the prostate of men with LUTS and its secretion is stimulated by inflammation.

This study investigates whether an increase in estradiol: testosterone ratio drives inflammation in the prostate and whether OPN is involved in this process.

Hypothesis

Methods and Materials

Male wildtype C57BL/6J (WT) or Spp1tm1Blh/J (OPN-KO) mice were surgically implanted with slow-releasing subcutaneous pellets containing 25 mg testosterone (T) and 2.5 mg estradiol (E2) (T+E2). Mice were euthanized two or six weeks later.



The protein expression of OPN, and inflammatory markers, cyclooxygenase-2 (COX2) and nuclear factor kappa B (NFκB), was detected by immunohistochemistry. Nuclear and cytoplasmic optical density was detected and quantified by the inForm software package.

Results

OSTEOPONTIN EXPRESSION IS ELEVATED IN RESPONSE TO STEROID HORMONE TREATMENT

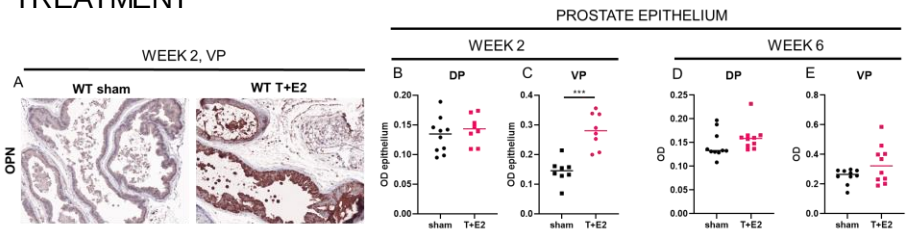
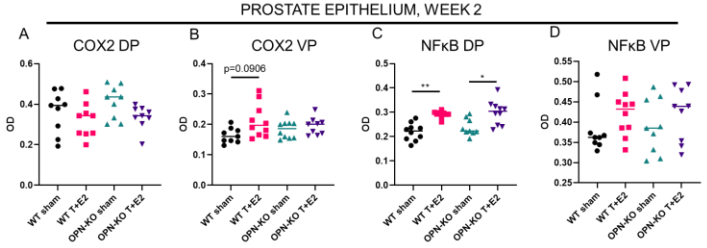


Figure 1: OPN expression is elevated in the ventral prostate two weeks after T+E2 pellet implantation. Optical density of OPN staining shows an increase in the epithelium of the ventral (VP, **A** and **C**) but not that of the dorsal prostate (DP, **B**). OPN expression was not significantly increased six weeks after pellet implantation. Significance was assessed with the Mann-Whitney test. ***: p<0.001.

NUCLEAR NFKB IN THE PROSTATE EPITHELIUM IS ELEVATED BY STEROID HORMONES



We tested two inflammatory markers, COX2 and NFκB, to determine whether their expressional changes correspond to the increased inflammatory cell infiltration previously found in T+E2-treated tissues. We found that nuclear NFκB serves as a better indicator of prostatic inflammation.

Figure 2: NFκB optical density, but not COX2, is upregulated in response to T+E2 treatment in prostate epithelial cells. Cytoplasmic COX2 in the DP (**A**) or in the VP (**B**) was not significantly affected by steroid hormone treatment. Nuclear NFκB expression was significantly upregulated by T+E2 treatment in the DP (**C**) but not in the VP (**D**). Significance was assessed with the Kruskal-Wallis test. *: p<0.05, **: p<0.01.

OPN LOSS DIMINISHES NFKB INCREASE IN THE STROMA

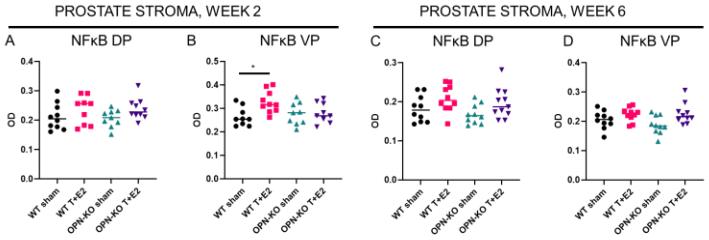
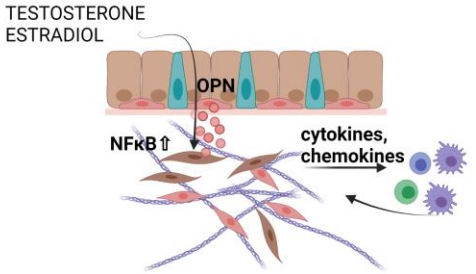


Figure 3: Stromal NFκB is increased in response to steroid hormone treatment in the VP which effect is abolished in OPN-KO mice. Stromal nuclear NFκB expression was unaffected by T+E2 treatment in the DP (**A**) but was significantly increased in the VP in OPN-KO mice (**B**) 2 weeks after pellet implantation. No significant change in nuclear NFκB was observed after 6 weeks in the DP (**C**) or the VP (**D**). Significance was assessed with the Kruskal-Wallis test. *: p<0.05.

Conclusions



We identified that testosterone and estradiol treatment specifically stimulates osteopontin expression in epithelial cells in the ventral prostate. Elevated OPN secretion will potentially trigger NFκB expression and its translocation to the nucleus. NFκB is known to promote the expression of various cytokines and chemokines which will attract immune cells to the prostate causing inflammation.

Future Goals:

- Investigate the expression of cytokines and chemokines in WT and OPN-KO mouse prostates
- Determine whether OPN and NFκB expression are increased in the prostatic urethra where pathological changes can be associated with urinary function

Acknowledgements

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