

The Co-localization of COX-1 and COX-2 in Aged Mouse Prostate and Human Prostate

Livianna Myklebust¹, Chandler Hudson², Teresa Liu¹, Laura Pascal³, Kai He², Donald B. DeFranco², William A. Ricke¹



University of Wisconsin Madison, Department of Urology, ²University of Pittsburgh, Department of Pharmacology and Chemical Biology



Introduction

Benign prostatic hyperplasia (BPH) is characterized by the enlargement of the prostate leading to lower urinary tract symptoms (LUTS) with the potential to have a severe negative impact on the quality of life in aging men. Prostatic inflammation may contribute to the pathogenesis of BPH, however the underlying mechanisms for this contribution have yet to be fully elucidated. Cyclooxygenase-2 (COX-2) is an inflammatory mediator that has been found to be selectively overexpressed in diverse cell types within benign and malignant disease, including the prostate. Cyclooxygenase-1 (COX-1) is constitutively expressed, and while primarily involved in maintaining homeostasis in various organs, it has been implicated in inflammation. Despite the association between prostatic inflammation and BPH/LUTS, treatment of symptomatic BPH with a combined therapy of a nonsteroidal anti-inflammatory agent (NSAID), which inhibits Cox-2, and a 5 alpha-reductase inhibitor (5ARI) only showed transient resolution of LUTS in clinical trials¹. In this study, we compare the changes in expression of both COX-1 and COX-2 in both normal and hyperplastic human prostate. Additionally, our lab has previously shown that lower urinary tract dysfunction spontaneously develops in aged mice². To also investigate a potential COX-1/COX-2 compensatory mechanism in an aging prostate, we also compared the protein expression of both enzymes in the lobes of young and old mice.

Hypothesis

We hypothesize that the expression of the inflammatory mediators COX-1 and COX-2 will be upregulated in old mouse and BPH tissues compared to young mouse and normal tissues as a component of inflammatory mediated prostatic hyperplasia.

Materials and Methods

Formalin fixed paraffin embedded human normal adjacent and BPH tissues from a tissue micro-array and formalin fixed paraffin embedded mouse prostate (AP, VP, and DLP) from 2 and 24 month old mice were stained for COX-1 and COX-2 using multispectral quantitative multiplex IHC. E-cadherin was also stained to aid in tissue segmentation. Fluorophores were spectrally unmixed and the optical densities for both proteins were quantified using InForm[®] software. Tissue and cell segmentation were performed for protein localization.

Results

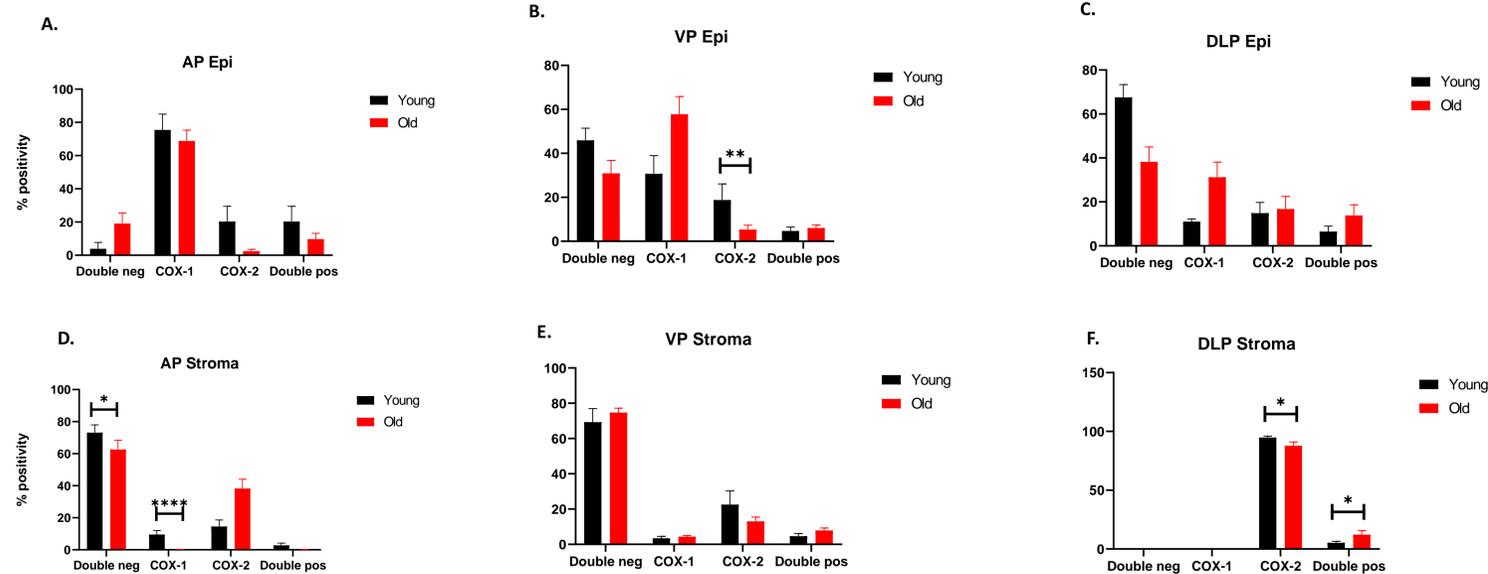
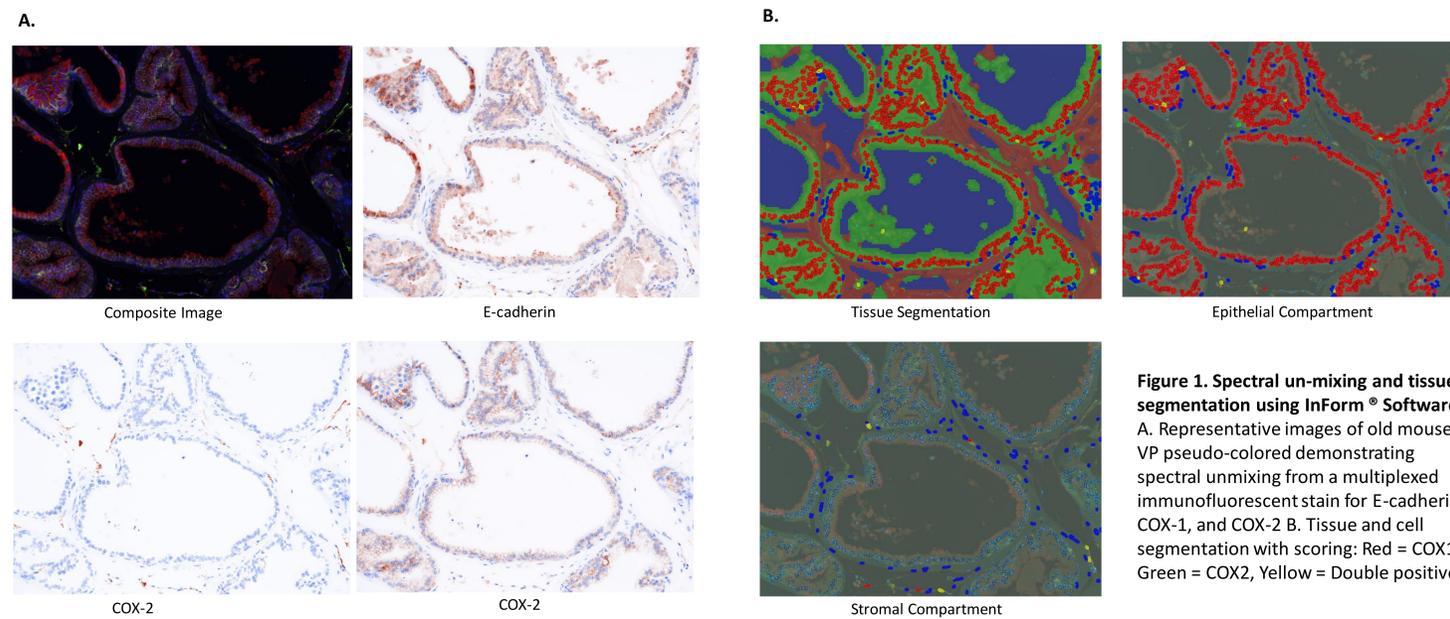


Figure 2. COX-1 and COX-2 expression changes between young and old tissues. A – E. COX-1 and COX-2 scoring between young and old mouse prostate lobes separated into the stromal and epithelial compartments. COX-2 decreases in the old mouse VP epithelium and DLP stroma whereas COX-1 decreases in the old mouse AP stroma. Double negativity decreases in the old AP stroma and the double positivity of both COX-1 and COX-2 increases in the DLP stroma. *, p-value <0.05, **, p-value <.01, ****, p-value <.0001

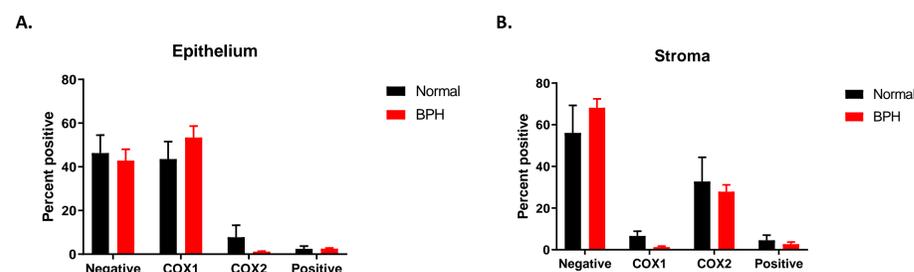


Figure 3. COX-1 and COX-2 expression is not altered in BPH tissues compared to normal prostate. A. COX-1/COX-2 expression does not change between BPH and normal prostate epithelial tissues B. COX-1/COX-2 expression also does not change between BPH and normal prostate stromal tissues.

Discussion

- Multiplex IHC revealed the presence of both the constitutive and inducible forms of cyclooxygenase, COX-1 and COX-2, respectively, within individual cells of prostate tissue.
- Clinical use of COX-2 specific NSAIDs may have unpredictable effects on the production of arachidonic acid metabolites, products of cyclooxygenase activity, that play an important role in the promotion and/or resolution of inflammation.
- Distinct alterations in the balance between COX-1 and COX-2 in aged mice or in human BPH provide the impetus for future studies to investigate the role of specific arachidonic acid metabolites in BPH/LUTS progression, particularly in the context or prostatic inflammation.
- Furthermore, age as a factor in the human tissue was not examined in this study.
- The potential impacts of the changes in COX-1/COX-2 colocalization and alterations to the expression of COX-1 in the aged stroma of the human prostate will require further exploration.

References

- Di Silverio F, Bosman C, Salvatori M, Albanesi L, Proietti Pannunzi L, Ciccariello M, Cardi A, Salvatori G, and Sciarra A. (2005) Combination therapy with rofecoxib and finasteride in the treatment of men with lower urinary tract symptoms (LUTS) and benign prostatic hyperplasia (BPH). *Eur. Urol.* 47, 72–78; discussion 78–79
- Liu TT, Thomas S, Mclean DT, Roldan-Alzate A, Hernando D, Ricke EA, Ricke WA. Prostate enlargement and altered urinary function are part of the aging process. *Aging (Albany NY)*. 2019 May 13;11(9):2653-2669. doi: 10.18632/aging.101938. PMID: 31085797; PMCID: PMC6535061.

Acknowledgements

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