NON-NEURONAL ATP: REGULATION OF RELEASE AND ACTION IN THE BLADDER

PhD Thesis

By

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SUMMARY OF THESIS

This thesis investigates the mechanisms involved in sensory signalling from the mouse urinary bladder. Sensory afferent firing is essential in the initiation of the micturition reflex and ultimately regulates the cycle of bladder filling and bladder emptying. Whilst bladder contraction and efferent function have been studied extensively, the processes that determine afferent signalling remain elusive and two distinct pathways are currently thought to underlie mechanotransduction: direct gating of mechanosensitive afferents in the bladder wall during bladder stretch, and an indirect mechanism via the release of mediators from the urothelium. Understanding more about bladder afferent transduction mechanisms may lead to the development of novel treatments for lower urinary tract disorders in which symptoms are associated with the filling phase of micturition such as overactive bladder and interstitial cystitis.

Mechanisms underlying bladder mechanosensation were investigated both directly, using an in vitro afferent nerve recording technique which allowed the concurrent recording of intravesical pressure and afferent nerve activity, and indirectly, examining urothelial mediator release and intracellular calcium signalling of urothelial cells and isolated DRG neurons. Using a combination of mechanical, pharmacological and genetic tools, a role for P2X, P2Y, TRPV1 and NK2 receptors in influencing mechanosensitivity was investigated.

Experiments employing pharmacological blockade or genetic deletion of the TRPV1 receptor implicated TRPV1 in bladder mechanosensitivity. Furthermore, experiments investigating the mechanisms of TRPV1 mechanosensitivity determined a role for TRPV1 in modulating purinergic responses on afferent nerves and ATP release from the urothelium. These mechanisms are thought to combine and underlie the decreased afferent nerve sensitivity to distension observed in TRPV1−/− knockout mice. This thesis also suggests that autocrine signalling of the urothelium through P2X and P2Y receptors may regulate intracellular calcium levels, an essential component of ATP release from the urothelium. Furthermore, a role for tachykinins in mediating mouse detrusor contraction acting through NK2 receptors was confirmed. A novel mechanism whereby stimulation of urothelial NK2 receptors was able to alter urothelial mediator release, and modify afferent nerve activity as a result of a change in detrusor function was also elucidated.

As modulation of bladder compliance and detrusor smooth muscle contraction during bladder distension was able to significantly alter afferent nerve discharge, these studies suggest that the major stimulus for afferent nerve output from the bladder is direct mechanical stretch of the bladder.
wall, and that in healthy mice, a lesser component is attributable to secondary indirect mechanisms mediated via non-neuronal ATP from the urothelium.

Further research is necessary to determine the relative contribution of the two mechanosensitivity pathways in disease states, as there is significant evidence that a phenotypic switch towards an indirect mechanism of mechanosensation could underlie increased sensation and reflex bladder symptoms.
DECLARATION AND ADDENDUM

This thesis is submitted to Bond University in fulfilment of the requirements of the degree of Doctor of Philosophy by Research

This research represents my own original work towards this research degree and contains no material which has been previously submitted for a degree or diploma at this university or any other institution, except where due acknowledgment has been made.

Luke Grundy

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