# Contents

# Chapter 1: Introduction.

**1.1** Anatomy of the urinary bladder……………………………………………………………………1

**1.2** The urothelium……………………………………………………………………………………..8

**1.3** Interstitial Cells……………………………………………………………………………………55

**1.4** Innervation of the bladder…………………………………………………………………………57

**1.5** Detrusor muscle contraction………………………………………………………………………66

**1.6** The micturition reflex……………………………………………………………………………..72

**1.7** Bladder pathology and clinical implications………………………………………………………75

**1.8** Aim of this thesis………………………………………………………………………………….83

# Chapter 2: Materials and Methods.

**2.1** Simultaneous recording of intraluminal pressure and afferent nerve activity...…………………..84

**2.2** Experimental Protocol……………………………………………………………………………..90

**2.3** Drugs, dilutions and their application……………………………………………………………..92

**2.4** Statistical analysis of data obtained from electrophysiology recordings……………………….....93

**2.5** Single unit analysis………………………………………………………………………………..95

**2.6** Bladder Histology…………………………………………………………………………………99

# Chapter 3: The response to bladder distension.

**3.1** Aim of this chapter……………………………………………………………………………….102

**3.2** The response to ramp distension of the bladder………………………………………………….103

**3.3** Stability of the preparation – Time………………………………………………………………115

**3.4** Discussion………………………………………………………………………………………..125

**3.5** Concluding comments……………………………………………………………………………135

# Chapter 4: Inhibition of mediator release from the urothelium.

*Investigating the effects of Ca2+ free Krebs exposure on compliance and*

*afferent nerve sensitivity of the mouse bladder*

**4.1** Introduction………………………………………………………………………………………137

**4.2** Aims and hypothesis……………………………………………………………………………..138

**4.3** Experimental method…………………………………………………………………………….139

**4.4** Results: Effects of Ca2+ free Krebs on afferent nerve firing and bladder compliance…………...142

*Investigating the effects of detrusor muscle paralysis on bladder compliance*

*and afferent nerve firing in the mouse*

**4.5** Aim and hypothesis………………………………………………………………………………152

**4.6** Experimental method…………………………………………………………………………….153

**4.7** Results: Effects of nifedipine on afferent nerve firing and bladder compliance………………...156

*Investigating the effects of Ca2+ free Krebs exposure, in the presence of PPADS,*

*on compliance and afferent nerve sensitivity of the mouse bladder.*

**4.8** Aim and hypothesis………………………………………………………………………………165

**4.9** Experimental method………………………………………………………………………...…..166

**4.10** Results…………………………………………………………………………………………..169

**4.11** Discussion………………………………………………………………………………………180

**4.12** Summary of findings.…………………………………………………………………………...188

# Chapter 5: Stimulation of mediator release from the urothelium.

**5.1** Introduction………………………………………………………………………………………189

*An investigation of the effects of induction of mediator release from the urothelium, by high K+ solution infusion, on afferent nerve sensitivity and compliance in the mouse bladder.*

**5.2** Aims and hypothesis…………………………………………………………………………......191

**5.3** Experimental Method……………………………………………………………………….........192

**5.4** Results……………………………………………………………………………………………195

*An investigation of the effects of urothelial damage by infusion of protamine sulphate on afferent nerve sensitivity and compliance in the mouse bladder.*

**5.5** Aims and hypothesis……………………………………………………………………………..222

**5.6** Experimental method…………………………………………………………………………….222

**5.7** Results……………………………………………………………………………………………226

*An investigation of the effects of high K+ perfusion on bladders with urothelial damage on afferent nerve sensitivity and compliance in the mouse bladder*

**5.8** Aims and hypothesis..……………………………………………………………………………243

**5.9** Experimental method………………………………………………………………………….....244

**5.10** Results…………………………………………………………………………………………..246

**5.11** Discussion………………………………………………………………………………………256

**5.12** Summary of findings, concluding remarks and future perspectives………………………...….269

# Chapter 6: Mechanism.

**6.1** Introduction………………………………………………………………………………………271

**6.2** Aims and hypothesis……………………………………………………………………………..273

**6.3** Experimental Method…………………………………………………………………………….275

**6.4** Vehicle control (full cocktail)……………………………………………………………………281

**6.5** Results – Full cocktail……………………………………………………………………………291

**6.6** Is the inhibitory response to high K+ stimulation mediated by the cannabinoid

signalling pathway?........................................................................................................................303

**6.7** Is the inhibitory response to high K+ stimulation mediated by ACh/ muscarinic

receptor signallinpathway?............................................................................................................315

**6.8** Is the inhibitory response to high K+ stimulation mediated by the purinergic

signalling pathway? …………………………………………………………………….………327

**6.9** Is the inhibitory response to high K+ stimulation blocked by prostaglandin

antagonist?..……………………………………………………………………………………...339

**6.10** The inhibitory response to high K+ stimulation is mediated by the nitric

oxide (NO) signalling pathway…………………………………………………………………351

**6.11** NO signalling mediates the inhibition of afferent nerve sensitivity in

response to high K+ stimulation. Is this effect attributable to eNOS?.......................................363

**6.12** Discussion………………………………………………………………………………………376

**6.13** Summary of findings presented in this chapter…………………………………………………383

# Chapter 7: General Discussion.

**7.1** Major findings……………………………………………………………………………………385

**7.2** Further study, clinical perspectives and pharmaceutical opportunities………………………….390

# References

………………………………………………………………………………………………………..392