Lovick, T. (2016). Central control of visceral pain and urinary tract function. *Autonomic Neuroscience: Basic and Clinical*, 200, 35-42. <https://doi.org/10.1016/j.autneu.2016.02.001>

Peer reviewed version

License (if available):
CC BY-NC-ND

Link to published version (if available):
[10.1016/j.autneu.2016.02.001](https://doi.org/10.1016/j.autneu.2016.02.001)

[Link to publication record in Explore Bristol Research](#)
PDF-document

This is the author accepted manuscript (AAM). The final published version (version of record) is available online via Elsevier at <http://www.sciencedirect.com/science/article/pii/S1566070216300066>. Please refer to any applicable terms of use of the publisher.

University of Bristol - Explore Bristol Research

General rights

This document is made available in accordance with publisher policies. Please cite only the published version using the reference above. Full terms of use are available: <http://www.bristol.ac.uk/red/research-policy/pure/user-guides/ebr-terms/>

Central control of visceral pain and urinary tract function

Thelma A Lovick

Physiology & Pharmacology, University of Bristol, Bristol BS8 1TD, UK

e-mail: Thelma.lovick@bristol.ac.uk

tel +44 07971361705

Abstract

Afferent input from A δ and C-fibres innervating the urinary bladder are processed differently by the brain, and have different roles in signaling bladder sensation. A δ fibres that signal bladder filling activate a spino-bulbo-spinal loop, which relays in the midbrain periaqueductal grey (PAG) and pontine micturition centre (PMC). The excitability of this circuitry is regulated by tonic GABAergic inhibitory processes. In humans and socialized animals micturition is normally under volitional control and influenced by a host of psychosocial factors. Higher nervous decision-making in a social context to 'go now' or 'do not go' probably resides in frontal cortical areas, which act as a central control switch for micturition. Exposure to psychosocial stress can have profoundly disruptive influence on the process and lead to maladaptive changes in the bladder. During sleeping the voiding reflex threshold appears to be reset to a higher level to promote urinary continence.

Under physiological conditions C-fibre bladder afferents are normally silent but are activated in inflammatory bladder states and by intense distending pressure.

Following prolonged stimulation visceral nociceptors sensitise, leading to a lowered threshold and heightened sensitivity. In addition, sensitization may occur within the central pain processing circuitry, which outlasts the original nociceptive insult.

Visceral nociception may also be influenced by genetic and environmental influences.

A period of chronic stress can produce increased sensitivity to visceral pain that lasts for months. Adverse early life events can produce even longer lasting epigenetic changes, which increase the individual's susceptibility to developing visceral pain states in adulthood.

Keywords: micturition, central nervous control, visceral pain, bladder

Despite its autonomic nature, the bladder is by no means an autonomous structure; normal bladder filling and emptying does not occur without the involvement of the central nervous system. Although there is some organisation at sacral spinal level (see ***** this issue), following spinal injury bladder dysynergia is a serious problem that impacts significantly on the individual's quality of life. Even following spinal injury at supra sacral levels voiding is not normal, implying a critical dependence on the brain.

The sophisticated nature of the control over micturition is perhaps under-appreciated.

In the newborn, urinary voiding is a reflex process driven by the fullness of the bladder, but as the individual matures voiding becomes a volitional process. In addition, the threshold for micturition is not fixed. In humans and many socialised animals, social mores dictate that voiding is appropriate only under certain circumstances. Humans may suppress voiding for relatively long periods of time despite fullness of the bladder, until the relevant social conditions are met. House training of domestic pets instills a similar degree of control. When a suitable opportunity presents itself, humans can empty a partly filled bladder, in order to maximize the interval before the next void. If needed, there is also the facility to halt the micturition process mid-stream i.e. switch on or off at will.

Volitional control is also exhibited in many undomesticated but socialised species.

For example, specific 'toilet' areas can be identified within the cages of laboratory rats and mice. Controlled partial voiding in order to release small quantities of urine for scent marking is another example of voiding behaviour that requires very precise

volitional control of the autonomic outflows. In addition, micturition does not normally occur during sleep. The high level of decision-making and precise motor control over the micturition reflex has lead to the concept of “cognitive voiding” (Fig 1; Gillespie, 2013), which implies significant forebrain involvement.

The act of voiding requires a co-ordinated sequence of events, which includes contraction of the detrusor and relaxation of the external urethral sphincter muscles, usually accompanied by various postural adjustments. In some species (e.g. rats and dogs), the sphincter muscle undergoes a period of rhythmic contraction and relaxation during voids, which is thought to increase the velocity of flow in the urethra and enable spurting of urine for scent marking (d’Amico et al, 2011; Conte et al., 1991; Maggi et al, 1986; Matsuura et al., 1998, 2000). These co-ordinated events require the functional integrity of the spino-midbrain-spinal loop and are absent after spinal transection (d’Amico et al, 2011).

Basic spinal-midbrain-spinal pathway for micturition

The spino-midbrain-spinal circuit, which relays in the midbrain periaqueductal grey matter (PAG) and pons (pontine micturition centre or Barrington’s nucleus) has been the subject of a number of comprehensive reviews in recent years (Fowler et al., 2008; Holstege 2010; Drake et al., 2010; de Groat and Wickens, 2013). It will therefore be considered only briefly here, for completeness.

Filling of the bladder is detected by stretch-sensitive afferent nerves in the bladder wall. Among distension-sensitive bladder afferents, low threshold myelinated A-delta fibers acting as ‘in series’ tension receptors (Iggo, 1955) are considered to be mainly

involved in control of micturition and several functional classes of distension-sensitive afferents have been identified (Zagorodnyuk et al., 2007). In contrast, high threshold afferents (intravesical pressure >20 mmHg) have been associated with the generation of painful sensations (de Groat, 1997). Afferent information on filling status is relayed to the brain by a group of neurons in the lateral funiculus of the upper sacral cord, just lateral to the dorsal horn. Anatomical studies in cats indicate that cells in this region project rostrally to terminate bilaterally in the central part of the PAG (Vanderhorst et al., 1996) although in the rat, electrophysiological and anatomical evidence favours dorsal and ventrolateral targets for the projection (Ding et al., 1997; Duong et al., 1999; Marson, 1997; Mitsui et al., 2003). In rats there is also a direct projection from the sacral cord to the PMC (Ding et al., 2007), which if activated by bladder distension, could potentially short-circuit the PAG. However, this direct pathway does not appear to play a significant role in initiating voiding, at least under urethane anaesthesia, because lesions or pharmacological inhibition of synaptic transmission through the PAG prevent normal micturition from taking place (Matsumoto et al., 2004; Matsuura et al., 1998; 2000; Stone et al., 2011). Comparable information from humans is limited but even so, urinary retention was reported to accompany the development of a space occupying lesion in the PAG (Yaguchi et al., 2004) whilst in multiple sclerosis sufferers retention or incontinence have been associated with the presence of demyelinating lesions in the midbrain and pons close to the periaqueductal grey matter and pontine micturition centre (Charil et al., 2003).

Efferents from the PAG project to the pontine micturition centre (PMC, also known as Barrington's nucleus), which in turn sends projections directly to preganglionic parasympathetic neurones in the sacral cord that regulate the activity of motor

neurons innervating the detrusor and urethral sphincter muscles (Holstege, 2010; Fowler et al., 2008; Drake et al., 2010; deGroat and Wickens, 2013 for reviews). In cats, but not in rats, more laterally in the pons lies the pontine storage centre or ‘L-region’ where electrical stimulation induces contraction of the external urethral sphincter (EUS) and relaxation of the bladder (Holstege et al., 1986). Anatomical connections between this region, the PMC and the PAG have yet to be demonstrated however.

A central switch for micturition that requires higher nervous decision-making in a social context.

The spino-midbrain-spinal loop is undoubtedly the foundation of micturition control. However, this circuit does not function simply as a relay whereby a set level of afferent input arising from the bladder triggers a void. Integral to the concept of cognitive voiding (see above) is that cues from the external as well as internal environment control a neural “switch” for voiding. The nature of control of the switch is still a matter of debate. In one scenario the switch is “off” during the resting (filling) state but can be flipped to the “on” position to initiate a void. An alternative scenario would be a switch that is normally “on” and tonically inhibiting the micturition circuitry but is switched to the “off” position to permit voiding to occur. In practice, both mechanisms might be present.

The location of the “switch” is also a matter of debate. Beckel and Holstege (2014) have suggested that a neural switch that turns on voiding resides in a region of the dorsolateral pons they term the pelvic organ stimulating centre (POSC, corresponding to the pontine micturition centre), but the hand that “throws” the switch lies in the

PAG. This is an attractive proposition since the PAG cannot evoke voiding without engaging the PMC and the PMC and, with the exception of rats, does not receive direct information from the bladder (see de Groat and Wickens 2013 for a recent discussion of this point). Neurones in both the PMC and PAG display changes in firing rate that correlate with changes in vesicular pressure during isovolumetric contractions evoked by distending the bladder with the outlet ligated (e.g. Liu et al., 2004; Sakakibara et al., 2002; Sugaya et al., 2003; Tanaka et al., 2003). Based on the properties of such cells deGroat and Wickens (2013) have proposed a simple computer model of the micturition circuit. It will now be important to test their model with data obtained from cells recorded during physiological micturition i.e. when actual voiding takes place.

The location of the micturition on-off switch is of paramount importance as it could be key to understanding certain forms of urge incontinence. Studies in animal models are fleshing out the physiological and anatomical details of the basic voiding circuitry. However the question of switching may best be answered by recording activity during voiding in the conscious state. As the resolution of brain imaging technology improves, such studies are becoming feasible in animals. An fMRI study in sedated rabbits reported activation in both PAG and PMC after filling the bladder to 70% capacity, but without voiding (Xiang et al., 2010). In a study in rats under urethane anaesthesia the PAG but not the PMC, was activated during filling. However when bladder volume reached the voiding threshold, the intensity of the PAG signal increased further and activation was also detected in the PMC (Tai et al., 2009). The results of this elegant study are consistent with the PAG monitoring bladder volume during the filling phase and then ‘switching on’ the PMC to initiate a void.

Forebrain control of bladder function

Whilst the finer details of the micturition switch remain to be worked out, another important question to consider is what determines the status of the switch? For many years investigations into the control of voiding at forebrain level were restricted by the use of anaesthetized animal models in which synaptic transmission in forebrain structures is necessarily depressed. However, the field opened up with the advent of modern high resolution imaging techniques, which could be applied to conscious human subjects. In 2010 a meta-analysis of the data gathered from the first decade of brain imaging of micturition in humans enabled Fowler and Griffiths (2010) to propose a working model of lower urinary tract control by higher centres. In essence, they proposed that during storage, information on bladder status carried by spinal afferents is relayed directly to the midbrain periaqueductal gray (PAG) and also activates the hypothalamus, thalamus, dorsal anterior cingulate cortex, right insula and lateral prefrontal cortex. In the prefrontal cortex the decision to void or not may be made. If the decision is to “go” the medial prefrontal cortex, which during storage is tonically inhibiting transmission through the PAG and the downstream pontine micturition centre is disinhibited, thereby allowing a void to proceed. At the same time the hypothalamus may also provide a “safe” signal to the PAG. In their more recent comprehensive review of forebrain influences Griffiths and Fowler (2013) extended their original model and proposed in addition, the existence of a back-up continence mechanism which in cases of extreme urgency, can bypass the brainstem switch to promote continence by activating a pathway from the anterior cingulate region to the pontine storage centre (Fig 2).

The results from basic studies in animal models lend support to the concept of top down gating of the ponto-medullary micturition circuit by the pre-frontal area (Griffiths and Fowler, 2013) via a neural micturition "switch" in the PAG, which permits the downstream PMC to initiate a void. In rats the critical synaptic relays in the micturition circuit lie within the ventrolateral PAG, the region that sends projections to the PMC (Kuipers et al., 2006). These regions are normally subject to a tonic GABAergic influence (Stone et al., 2011). When voiding takes place a decrease in concentration of extracellular GABA in the PAG occurs (Kitta et al., 2008), presumably reflecting a lift of the tonic inhibition. This would be expected to increase the excitability of the PAG, and thereby stimulate the efferent projection to the PMC to trigger voiding, in line with the results obtained during imaging by Tai et al. (2009) (see above).

The source of the inhibitory GABAergic tone in the PAG has not yet been established. However, in rats and in primates the prefrontal cortex sends dense projections to the ventrolateral PAG (An et al., 1998; Floyd et al., 2000), which might be a source of the tonic inhibition during filling. The inhibitory nature of the input to the PAG from the medial frontal lobe is suggested by the observation that stimulation in this area prolonged the interval between bladder contractions in conscious rats that were undergoing continuous cystometry (Nishijima et al., 2012). Whether the inhibitory projection is direct or mediated by activation of local GABAergic interneurons in the PAG is not known.

In humans the involvement of the medial prefrontal cortex (PFC) in decision-making, based on the social situation, is well established (Adolphs, 1999). More recently,

sophisticated multilevel path modelling approaches to imaging have shown that social evaluative threat (SET) can influence functional connections between the frontal cortical regions, the PAG and autonomic output (Wager et al., 2009). In the context of micturition, when mental appraisals are translated into an adaptive physiological response, SET might be experienced as the potential humiliating consequences of failing to maintain continence in a given social situation.

There are many other aspects of micturition control in which the forebrain is undoubtedly involved. For example, the sense of urgency is not always directly related to bladder volume and indeed, appears to be remarkably plastic. It is a common experience that the need to void imminently can be subsumed at least temporarily if the individual is distracted and attention is diverted elsewhere. On the other hand a sense of urgency can suddenly and dramatically increase on the final approach to home - the “latch key” phenomenon. Paruresis, as exemplified by the inability to urinate in public toilets or in other people’s homes is another example of transient dysfunction of the bladder control system. Little is known about the central nervous basis of these effects, yet they are an integral part of daily life in humans and may be relevant to understanding disorders of bladder control.

What is well established from work in animal models, is that exposure to psychosocial stress exerts a powerful influence on bladder function. In rats 30min of daily exposure to water avoidance stress for 7 days is sufficient to promote urinary retention and abnormal urodynamics, which resemble those produced by partial bladder outlet obstruction (pBOO) (Wood et al, 2013). Mice too showed a similar stress-induced urinary retention and increased bladder mass (Chang et al, 2009).

1 Activation of CRF-containing neurons within Barrington's nucleus (pontine
2 micturition centre) appears to be integral to inducing this effect. These neurones send
3
4 projections to the spinal cord (Valentino et al, 1996) where they may produce
5
6 inhibitory effects. Intrathecal application of CRF increased bladder capacity and
7
8 micturition volume in awake rats (Pavcovich and Valentino, 1995; Kiddo et al, 2006).
9
10 However, these findings are equivocal since others have reported that CRF facilitated
11
12 micturition (Klausner et al, 2005).
13
14
15
16
17
18

19 Interestingly pBOO induced experimentally in the absence of stress has been shown
20
21 to increase expression of CRF in Barrington's nucleus. Thus a positive feedback loop
22
23 could be activated whereby once exposure to psychosocial stress produces voiding
24
25 dysfunctions and bladder pathology via activation of the CRF pathway, signals from
26
27 the dysfunctional viscera activate components of the basic micturition circuit to
28
29 produce further neurobehavioral effects that are co-morbid with visceral pathology
30
31 (Valentino et al, 2011). This vicious circle of events could give rise to long-lasting
32
33 bladder dysfunction.
34
35
36
37
38
39
40

41 Central regulation of bladder function during sleep

42
43 Another challenge for the micturition control system is the sleep state. Humans
44
45 require several hours of uninterrupted sleep daily in order to thrive. Micturition
46
47 typically does not occur during the sleep period. This can be attributed in part to
48
49 reduced urine production as a consequence of the absence of fluid intake, combined
50
51 with the nocturnal increase in secretion of antidiuretic hormone. An endogenous
52
53 circadian variation of bladder compliance has also been demonstrated. Bladder
54
55 capacity increases during the sleep period due to functional changes in gap junctions
56
57
58
59
60
61
62
63
64
65

1 in bladder smooth muscle cells that are associated with oscillating expression of
2 connexin43 (Negoro et al., 2012; 2013).
3
4
5
6

7 The activity of brain circuitry controlling filling and voiding may also be reset during
8 sleep. In rats the micturition threshold increases during sleep (Kiddoo et al, 2006).
9
10 Whether this is due to a centrally-mediated increase in descending tonic inhibitory
11 control on sensory input from the bladder at spinal levels which raises the threshold
12 for micturition during sleep and/or whether the threshold of the central voiding
13 circuitry is re-set to a higher level during sleep is not clear. A dynamic centrally-
14 mediated feed forward afferent control system has been identified, which increases
15 bladder compliance during filling (Smith et al., 2012), but it is not known whether the
16 activity of this system changes during sleep. However, in favour of this idea, we
17 found that during continuous cystometry carried out in rats under urethane
18 anaesthesia, micturition threshold increased and compliance decreased as the EEG
19 cycled from an activated to a de-activated sleep-like state (Fig 3). Urethane
20 anaesthesia is characterized by cyclical changes in EEG waveform that have been
21 proposed to model changing sleep states (Clement et al., 2008). Thus there may be a
22 centrally-mediated modulation of responsiveness to bladder filling, which changes in
23 different sleep states.
24
25
26
27
28
29
30
31
32
33
34
35
36
37
38
39
40
41
42
43
44
45
46
47
48
49
50
51
52
53
54
55
56
57
58
59
60
61
62
63
64
65

Although decreased urine production and increased storage capacity of the bladder
reduce the likelihood of a need to void during the sleeping period, there are occasions
when micturition needs to take place. In conscious rats, EEG desynchronization
consistent with an arousal response (decrease in power in all frequencies) occurs just
prior to bladder pressure reaching the micturition threshold (Richenbacher et al, 2008;

Valentino et al, 2011). This effect is associated with increased firing of neurones in locus coeruleus. It has been suggested that the increased arousal level facilitates disengagement from ongoing behavior and promotes a shift to elimination-related behaviors (Richenbacher et al, 2008). It is possible that activation of this system during sleep could serve as an “alarm” to induce arousal and alert the individual of the need to void, thereby preventing nocturnal enuresis.

Pain from the urinary tract

In the healthy individual there is little sensation from the bladder at low volumes. However, sensations are evoked in humans once bladder volume exceeds around 400ml and increases until a volume is reached which represents the individual’s “absolute need” to void (De Wachter et al., 2013). At the high end there may be acute discomfort coupled with a certain amount of anxiety but on the whole, micturition is not an overtly painful experience. However, in certain pathophysiological states, frank pain can arise from the urinary tract.

The encoding of noxious events by the viscera is mediated by several types of visceral nociceptor. These include high threshold receptors, silent nociceptors and high intensity coding receptors (Cervero and Jänig, 1992). A δ bladder afferents signal bladder status related to filling under normal conditions. However, the urothelium and the detrusor muscle are innervated by C-fibres as well. Bladder activated C-fibre activity was first demonstrated in a classic study by Iggo (1955) but whether these fibres would be recruited under physiological conditions was not clear.

It now appears that the C afferents of the lower urinary tract are normally quiescent

during filling but may become activated by extreme intravesicular pressure or in inflammatory bladder states (see Gonzales et al., 2014 for a recent review). In the ureters, which are a source of intense pain under pathophysiological conditions, the majority of afferents function as true nociceptors, remaining silent unless exposed to intense distension, although there exists a minority of low threshold fibres, which are sensitive to contractions of the ureter (Pedersen et al., 2010).

The brain processes input from the A δ and C fibres separately. Imaging studies of responsiveness to activation of nociceptive input from the bladder in humans are scarce. However intravesical administration of cold (4-8°C) saline, which preferentially activates C-fibres (Mehnert et al., 2011), caused a different supraspinal activation pattern, albeit with some overlap, compared to that evoked by activation of A δ fibres by bladder filling (Table I). The cold stimulus did not however evoke overt pain in the subjects; thus brain activation reflecting the emotional component of bladder pain may not be evident from this study. It should be noted that there are areas of overlap between brain regions activated by A δ and C-fibre afferents, e.g. in the PAG. However, it is likely that different functional sets of neurons in this structure are engaged to process information from A δ and C-fibre afferents.

Plasticity of visceral pain circuits – peripheral sensitisation

An interesting feature of the processing of visceral afferent input via C-fibres is the plasticity within the nociception circuitry. Although most basic research into the central processing of visceral sensation has focused on nociceptor input from the gut, evidence is now starting to accumulate, which suggests that these findings will generalize to the urinary tract (e.g. Pedersen et al., 2010; Gonzales et al., 2014).

In the periphery persistent nociceptor stimulation due to intense pressure or the effects of inflammatory mediators released at sites of injury and tissue damage, can sensitise nociceptors by reducing thresholds for activation and enhancing responsiveness. The combination of prolonged noxious stimulation and peripheral sensitisation of nociceptors induces a long lasting increase in excitability of the spinal cord and higher centre neurons, a phenomenon known as central sensitization (Latremliere and Woolf, 2009). This effect may persist even when there is no longer any afferent nociceptor input. Visceral pain states can also induce a change in gap junction coupling between cells in dorsal root ganglia (Huang et al., 2010) and activation of spinal microglia (Bradesi, 2010; Liu et al., 2012) which both serve to amplify activity in the nociceptive circuitry and maintain the pain state.

Plasticity of visceral pain circuits – descending control systems

In addition to the ‘bottom up’ mechanism of visceral nociception a ‘top down’ modulation of afferent input from incoming signals occurs, which is mediated by activation of descending control systems from the brain. The descending modulation from cortical and limbic structures is a dynamic system. Emotional states such as stress, fear and anxiety exert potent, but complex, modulatory influences that can either suppress pain (stress-induced analgesia) or exacerbate painful states (stress-induced hyperalgesia; SIH) depending on the nature, duration and intensity of the stressor. This is exemplified in extreme conditions such as during a life threatening physical conflict, when pain may be completely suppressed. On the other hand, and perhaps more relevant to the clinical situation, milder stressors can exacerbate pain states. At the core of these effects is the modulation by cortical and subcortical

1 structures of activity in a pathway from the PAG, which via relays in the ventromedial
2 medulla, and modulates sensory transmission at the spinal level (Olango and Finn,
3 2014 for a recent review). Descending projections from the PAG also control bladder
4 voiding (see above). However, these neurons are almost certainly functionally distinct
5 from the pain modulatory circuit, although interactions may occur, particularly in
6 bladder pain states.

17 Epigenetic and environmental influences on visceral sensitivity

18 Genetic and environmental influences on visceral hypersensitivity in humans are now
19 well established. Traumatic early life events in particular, can produce long-term
20 effects on the brain circuitry involved in visceral pain processing. There is much
21 evidence for an association between abuse, particularly sexual abuse in childhood,
22 and strong and persistent effects on gastrointestinal health and visceral pain in
23 adulthood (Leserman and Drossman, 2008 for a review). The neuronal basis for
24 such effects is under investigation. Women suffering from irritable bowel syndrome,
25 who had a history of sexual or physical abuse, showed increased sensitivity to visceral
26 pain (rectal distension) compared to non-abused patients, which correlated with a
27 higher level of activation in the anterior and posterior cingulate cortices (Ringel et al.,
28 2008). At the same time the level of activation in the supragenual cingulate, a region
29 implicated in pain inhibition and arousal, was reduced (Ringel et al., 2008).

30 The development of animal models has started to shed further light of the underlying
31 neuronal processes. To date most studies have focused on gastrointenstinal disorders
32 (Maloney et al, 2015) but as data becomes available on bladder sensitivity, it seems
33 likely that similar findings will be made. As in humans, early life stress in rats has

1 been shown to produce a significant impact on visceral sensitivity in adulthood.
2 Maternal separation in early life is an especially powerful force, which increases
3 susceptibility to stress-induced visceral hypersensitivity, as shown by responsiveness
4 to colonic distension (Coutinho et al, 2002). Moreover, this effect could be transferred
5 to the next generation, even without exposure of second generation individuals to
6 maternal separation (van dem Wijngaard et al., 2013). Interestingly, fostering the
7 offspring of maternally deprived dams with dams that had not been exposed to
8 maternal separation prevented the development of hypersensitivity (van dem
9 Wijngaard et al., 2013).

10 Stress in adulthood has also been shown to lead to visceral hypersensitivity, including
11 the bladder. In a recent study (Lee et al, 2015) 10 days of 1h water avoidance stress in
12 anxiety-prone rats induced bladder hyperalgesia (increased visceromotor response to
13 instilling cold saline or rapid inflation of the bladder) and suprapubic hyperalgesia (an
14 index of referred bladder pain in rat). These effects were still evident months later,
15 after cessation of the stress.

16 Changes in the characteristics of the prefrontal cortex-PAG descending control
17 system may contribute to the long-term effect of early life stress in animals. Increased
18 responsiveness of the PAG to electrical stimulation has been reported in adult rats that
19 had been subjected to early life separation stress, suggesting that the circuitry in this
20 region had become hyper-excitable (Quintino-dos-Santos et al., 2014). Imaging using
21 PET in maternally separated rats, which developed stress-induced hyperalgesia in
22 adulthood revealed activation in the PAG in response to colorectal distension
23 (Wouters et al., 2012). After exposure to water avoidance stress, in addition to the
24 PAG, colorectal distension evoked significant deactivation of the frontal cortex

(Wouters et al., 2012), perhaps reflecting a stress-induced disinhibition of tonic forebrain influence on the descending pain control system in the PAG.

Exactly how exposure to chronic stress predisposes to development of visceral pathology and pain states is not clear. However, a number of studies have now shown that epigenetic mechanisms can contribute to the symptomatology of stress-evoked visceral hyperalgesia. Epigenetics refers to processes that lead to stable and/or heritable changes in gene function without alterations of the primary DNA sequence. Chronic social stress in rats increases DNA methylation and histone acetylation of genes that regulate visceral pain sensation in the peripheral nervous system. In rats in which hypersensitivity to colonic distension had been induced by exposure to repeated water avoidance stress, expression of the gene encoding the glucocorticoid receptor and cannabinoid CB1 receptors was reduced but expression of TRPV1 receptor increased in lumbo-sacral dorsal root ganglia (Hong et al, 2015). Within the brain reduced expression of the glucocorticoid receptor gene, with a concomitant increase in CRF expression, was also reported in amygdala (Tran et al., 2013).

CRF was first characterized as the hypothalamic neurohormone released in response to stressors in order to initiate the endocrine limb of the stress response. However, it also acts as a brain neurotransmitter that is released from neurons in limbic brain regions to mediate behavioral responses to stress. Interestingly, CRF is expressed in neurons of Barrington's nucleus (pontine micturition centre), which project to the spinal cord. These cells also collateralise to locus coeruleus, which is involved in arousal mechanisms (Valentino et al, 1996). Activation of spinally projecting CRF-containing neurons inhibits micturition and promotes urinary retention (Valentino et al, 2013). It has been suggested that prolonged retention may lead to the development

of bladder pathology and that afferent signals from the damaged bladder feed forward via locus coeruleus and Barrington's nucleus to further contribute to bladder dysfunction and a stressed phenotype. It will be of interest to discover whether epigenetic mechanisms in Barrington's nucleus contribute to these long-term changes.

Conclusion

The urinary bladder is a remarkable organ that is subject to a very sophisticated level of central nervous control that is influenced by emotional and psychosocial factors, which can produce surprisingly long-lasting effects. Recent findings provide a glimpse of the powerful influence not only of long-term stress but also the effect of early environment on visceral pain processing circuitry in adulthood. It is becoming clear that stress-induced visceral pain and its psychiatric comorbidities have a multifaceted etiology in which dysregulation of the hypothalamic-adrenal axis via inadequate or excessive activation, may contribute to the development of wide array of pathologies. An in depth understanding of neurophysiological mechanism which mediates voiding reflexes, including their modulation by emotional states and the factors which can give rise to pain from the urinary tract is essential to fully understand bladder function in health and will inform the development of new therapeutic strategies to treat conditions of dysfunctional bladder.

Acknowledgements

Financial support from the Medical Research Council Project Grant G1002251

- Adolphs R (1999). Social cognition and the human brain. *Trends Cogn Sci* **3**, 469-479.
- An X, Bandler R, Ongür D, Price JL (1998). Prefrontal cortical projections to longitudinal columns in the midbrain periaqueductal gray in macaque monkeys. *J Comp Neurol* **401**, 455-479.
- Beckel JM, Holstege G. (2014) The lower urinary tract. In: *The rat nervous system*. 4th Ed. Ch 12. Paxinos G, Sydney - Tokyo: Acad. Press.
- Bradesi S (2010). Role of spinal cord glia in the central processing of peripheral pain perception. *Neurogastroenterol Motil.* **22**:499-511.
- Cervero F, Jänig W (1992). Visceral nociceptors: a new world order? *TINS* **15** 374-378.
- Charil A, Zijdenbos AP, Taylor J, Boelman C, Worsley KJ, Evans AC, Dagher A. (2003). Statistical mapping analysis of lesion location and neurological disability in multiple sclerosis: application to 452 patient data sets. *Neuroimage.* **19**:532-544.
- Chang A, Butler S, Sliwoski J, Valentino R, Canning D, Zderic S (2009). Social stress in mice induces voiding dysfunction and bladder wall remodeling. *Am J Physiol Renal Physiol.* **29**: F1101-F1108.
- Clement EA, Richard A, Thwaites M, Ailon J, Peters S, Dickson CT (2008). Cyclic and sleep-like spontaneous alternations of brain state under urethane anaesthesia. *PLoS One* **3** e2004.
- Conte B, Maggi CA, Parlani M, Lopez G, Manzini S, Giachetti A. Simultaneous recording of vesical and urethral pressure in urethane-anesthetized rats: effect of neuromuscular blocking agents on the activity of the external urethral sphincter. *J Pharmacol Methods.* **1991**;26:161–171.

Coutinho SV, Plotsky PM, Sablad M, Miller JC, Zhou H, Bayati AI, McRoberts JA, Mayer EA. 2002; Neonatal maternal separation alters stress-induced responses to viscerosomatic nociceptive stimuli in rat. *Am J Physiol* 282: G307-G316

D'Amico SC, Schuster IP, Collins WF 3rd (2011). Quantification of external urethral sphincter and bladder activity during micturition in the intact and spinally transected adult rat. *Exp Neurol*. 228: 59-68.

de Groat WC (1997). A neurologic basis for the overactive bladder. *Urology* 50: 36–52.

de Groat WC, Wickens C (2013) Organization of the neural switching circuitry underlying reflex micturition. *Acta Physiol (Oxf)* 207: 66–84.

De Wachter SG, Heeringa R, Van Koevinge GA, Winkens B, Van Kerrebroeck PE, Gillespie JI (2014). "Focused introspection" during naturally increased diuresis: description and repeatability of a method to study bladder sensation non-invasively. *Neurourol Urodyn*.33: 502-506.

Ding YQ, Zheng HX, Gong LW, Lu Y, Zhao H, Qin BZ (1997). Direct projections from the lumbosacral spinal cord to Barrington's nucleus in the rat: a special reference to micturition reflex. *J Comp Neurol* 389:149–160.

Duong M, Downie JW, Du HJ. (1999). Transmission of afferent information from urinary bladder, urethra and perineum to periaqueductal gray of cat. *Brain Res*. 819: 108-119.

Drake MJ, Fowler CJ, Griffiths D, Mayer E, Paton JF, Bird L. (2010). Neural control of the lower urinary and gastrointestinal tracts: supraspinal CNS mechanisms. *Neurourol Urodyn*. 29:119–127.

Floyd NS, Price JL, Ferry AT, Keay KA, Bandler R (2000). Orbitomedial prefrontal cortical projections to distinct longitudinal columns of the periaqueductal gray in the

1 rat. J Comp Neurol 422, 556-578.

2 Fowler CJ, Griffiths DJ (2010) .A decade of functional brain imaging applied to
3 bladder control. Neurourol Urodyn 29:49–55.

4
5
6
7 Fowler CJ, Griffiths D, de Groat WC. (2008). The neural control of micturition. Nat
8
9
10 Rev Neurosci. 9: 453–466.

11
12 Gillespie J. (2013) What determines when you go to the toilet? The concept of
13
14 cognitive voiding. BJOG 120:133–136.

15
16
17 Gonzalez EJ, Merrill L, Vizzard MA (2014). Bladder sensory physiology: neuroactive
18
19 compounds and receptors, sensory transducers, and target-derived growth factors as
20
21 targets to improve function. Am J Physiol Regul Integr Comp Physiol 306: R869–
22
23 R878.

24
25
26 Griffiths DJ, Fowler CJ (2013). The micturition switch and its forebrain influences.
27
28 Acta Physiol 207, 93–109.

29
30
31 Holstege G. The emotional motor system and micturition control. (2010). Neurourol
32
33 Urodyn. 29: 42–48.Holstege G, Griffiths D, de Wall H, Dalm E (1986). Anatomical
34
35 and physiological observations on supraspinal control of bladder and urethral
36
37 sphincter muscles in the cat. J Comp Neurol 250: 449-461.Hong S, Zheng G, Wiley
38
39 JW (2015). Epigenetic regulation of genes that modulate chronic stress-induced
40
41 visceral pain in the peripheral nervous system. Gastroenterology 148:148–157.

42
43
44 Huang TY, Belzer V, Hanani M (2010). Gap junctions in dorsal root ganglia: possible
45
46 contribution to visceral pain. Eur J Pain 14:49.e1-11.

47
48
49
50
51 Iggo A (1955). Tension receptors in the stomach and the urinary bladder. J Physiol
52
53 128, 593–607.

Kiddoo DA, Valentino RJ, Zderic S, Ganesh A, Leiser SC, Hale L, Grigoriadis DE (2006). Impact of state of arousal and stress neuropeptides on urodynamic function in freely moving rats. *Am J Physiol* 290: R1697-R1706.

Kitta T, Matsumoto M, Tanaka H, Mitsui T, Yoshioka M, Nonomura K (2008). GABAergic mechanism mediated via D receptors in the rat periaqueductal gray participates in the micturition reflex: an in vivo microdialysis study. *Eur J Neurosci* 27, 3216-3225.

Klausner, AP, Streng T, Na Y-G, Raju J, Batts TW, Tuttle JB, Andersson K-E, Steers WD (2005). The role of corticotropin releasing factor and its antagonist, astressin, on micturition in the rat. *Auton Neurosci* 123 26-35.

Kuipers R, Mouton LJ, Holstege G (2006) Afferent projections to the pontine micturition center in the cat. *J Comp Neurol*. 494:36-53.

Latremoliere A, Woolf CJ (2009). Central sensitization: a generator of pain hypersensitivity by central neural plasticity. *J Pain* 10: 895–926.

Lee UJ, Ackerman AL, Wu A, Zhang R, Leung J, Bradesi S, Mayer EA, Rodríguez LV (2015). Chronic psychological stress in high-anxiety rats induces sustained bladder hyperalgesia. *Physiol Behav*. 139:541-548.

Leserman J, Drossman DA (2008). Relationship of abuse history to functional gastrointestinal disorders and symptoms: some possible mediating mechanisms. *Trauma Violence Abuse*. 8:331-343.

Liu PY, Lu CL, Wang CC, Lee IH, Hsieh JC, Chen CC, Lee HF, Lin HC, Chang FY, Lee SD. (2012). Spinal microglia initiate and maintain hyperalgesia in a rat model of chronic pancreatitis. *Gastroenterology*. 142:165-173.

Liu Z, Sakakibara R, Nakazawa K, Uchiyama T, Yamamoto T, Itoa T, Hattoria T (2004). Micturition-related neuronal firing in the periaqueductal gray area in cats.

Neuroscience 126: 1075–1082.

Maggi CA, Giuliani S, Santicioli P, Meli A (1986). Analysis of factors involved in determining urinary bladder voiding cycle in urethan-anesthetised rats. *Am J Physiol Regul Integr Comp Physiol* 251: R250–R257.

Moloney RD, O'Mahony SM, Dinan TG, Cryan JF (2015). Stress-induced visceral pain: toward animal models of irritable-bowel syndrome and associated comorbidities. *Front Psychiat* 6: 15.

Marson L (1997) Identification of central nervous system neurons that innervate the bladder body, bladder base, or external urethral sphincter of female rats: a transneuronal tracing study using pseudorabies virus. *J Comp Neurol* 389: 584–602.

Matsumoto S, Levendusky MC, Longhurst PA, Levin RM, Millington WR (2004). Activation of mu opioid receptors in the ventrolateral periaqueductal gray inhibits reflex micturition in anesthetized rats. *Neurosci Lett* 363, 116-119.

Matsuura S, Allen GV, Downie JW (1998). Volume-evoked micturition reflex is mediated by the ventrolateral periaqueductal gray in the anesthetized rats. *Am J Physiol* 275, R2049–2055.

Matsuura S, Downie JW, Allen GV (2000). Micturition evoked by glutamate microinjection in the ventrolateral periaqueductal gray is mediated through Barrington's nucleus in the rat. *Neuroscience* 101, 1053-1061.

Mehnert U, Michels L, Zempleni M-Z, Schurch B, Kollias S (2011). The supraspinal neural correlate of bladder cold sensation—An fMRI Study. *Human Brain Map* 32:835–845.

Mitsui T, Kakizaki H, Matsuura S, Tanaka H, Yoshioka M, Koyanagi T (2003). Chemical bladder irritation provokes c-fos expression in the midbrain periaqueductal gray matter of the rat. *Brain Res* 967: 81–88. Negoro H, Kanematsu A, Doi M,

1 Suadicani SO, Matusuo M, Imamura M, Okinami T, Nishikawa N, Oura T, Matsui S,
2 Seo K, Tainaka M, Urabe S, Kiyokage E, Todo T, Okamura H, Tabata, Y Ogawa O
3
4 (2012) Involvement of urinary bladder Connexin43 and the circadian clock in
5
6 coordination of diurnal micturition rhythm. *Nat Commun.* 3: 809.
7
8 doi: 10.1038/ncomms1812. Negoro H, Kanematsu A, Yoshimura K, Ogawa O (2013)
9
10 Chronobiology of micturition: putative role of the circadian clock. *J Urol* 190: 843-
11
12 849.
13
14 Nishijima S, Sugaya K, Kadekawa K, Ashitomi K, Yamamoto H (2012).
15
16 Effect of chemical stimulation of the medial frontal lobe on the micturition reflex in
17
18 rats. *J Urol* 187, 1116-1120.
19
20 Olango WM, Finn DP (2014). Neurobiology of stress-induced hyperalgesia. *Curr Top*
21
22 *Behav Neurosci* 20:251-280.
23
24 Pavcovich LA, Valentino RJ (1995). Central regulation of micturition in the rat the
25
26 corticotropin-releasing hormone from Barrington's nucleus. *Neurosci Lett.* 196: 185-
27
28 188.
29
30 Pedersen KV, Asbjørn Mohr Drewes AM, Frimodt-Møller PC, Osther PJS (2010).
31
32 Visceral pain originating from the upper urinary tract. *Urol Res* 38:345–355.
33
34 Quintino-dos-Santos JW, Müller CJ, Bernabé CS, Rosa CA, Tufik S, Schenberg LC
35
36 (2014). Evidence that the periaqueductal gray matter mediates the facilitation of panic-
37
38 like reactions in neonatally-isolated adult rats. *PLoS One* 9: e90726.
39
40 Rickenbacher E, Baez MA, Hale L, Leiser SC, Zderic SA, Valentino RJ (2008)..
41
42 Impact of overactive bladder on the brain: central sequelae of a visceral pathology.
43
44 *Proc Natl Acad Sci U S A.* 105:10589-10594.
45
46 Ringel Y, Drossman DA, Leserman JL, Suyenobu BY, Wilber K, Lin W, Whitehead
47
48 WE, Naliboff BD, Berman S, Mayer EA (2008). Effect of abuse history on pain
49
50
51
52
53
54
55
56
57
58
59
60
61
62
63
64
65

reports and brain responses to aversive visceral stimulation: an FMRI study.

Gastroenterology. 134:396-404.

Sakakibara R, Nakazawa K, Shiba K, Nakajima Y, Uchiyama T, Yoshiyama M, Yamanishi T, Hattori T (2002). Firing patterns of micturition-related neurons in the pontine storage centre in cats. *Auton Neurosci: Basic and Clinical* 99 24-30.

Smith PP, DeAngelis AM, Kuchel GA (2012). Evidence of central modulation of bladder compliance during filling phase *Neurourol Urodyn*. 31: 30–35.

Stone E, Coote, JH Allard, J Lovick TA (2011) GABAergic control of micturition within the periaqueductal grey matter of the rat. *J Physiol*. 589 2065-2078.

Sugaya K, Ogawa Y, Hatano T, Nishijima S, Matsuyama K, Mori S (2003). Ascending and descending brainstem neuronal activity during cystometry in decerebrate cats. *Neurourol Urodyn*. 22:343-350.

Tai C, Wang J, Jin T, Wang P, Kim S-G, Roppolo JR, de Groat WC (2009). Brain switch for reflex micturition control detected by fmri in rats. *J Neurophysiol* 102: 2719–2730.

Tanaka Y, Koyama Y, Kayama Y, Kawauchi A, Ukimura O, Miki T (2003). Firing of micturition center neurons in the rat mesopontine tegmentum during urinary bladder contraction. *Brain Res*. 965:146-154.

Tran L, Chaloner A, Sawalha AH, Greenwood Van-Meerveld B (2013).

Importance of epigenetic mechanisms in visceral pain induced by chronic water avoidance stress. *Psychoneuroendocrinology* 38: 898-906.

Valentino RJ, Chen S, Zhu Y, Aston-Jones G (1996). Evidence for divergent projections of corticotropin-releasing hormone neurons of Barrington's nucleus to the locus coeruleus and spinal cord. *Brain Res* 732:1–15.

Valentino RJ, Wood SK, Wein AJ, Zderic SA (2011). The bladder-brain connection:

Putative role of corticotropin releasing factor. *Nat Rev Urol* 8: 19–28.

van den Wijngaard RM, Stanisor OI, van Diest SA, Selting O, Wouters MM, Cailotto C, de Jonge WJ, Boeckxstaens GE (2013) Susceptibility to stress induced visceral hypersensitivity in maternally separated rats is transferred across generations. *Neurogastroenterol Motil* 25, e780–e790.

Vanderhorst VG, Mouton LJ, Blok BF, Holstege G (1996). Distinct cell groups in the lumbosacral cord of the cat project to different areas in the periaqueductal gray. *J Comp Neurol* 376:361-385.

Wager TD, van Ast VA, Hughes BL, Davidson ML, Lindquist MA, Ochsner KN (2009). Brain mediators of cardiovascular responses to social threat, part II: Prefrontal-subcortical pathways and relationship with anxiety. *Neuroimage* 47, 836-851.

Wood SK, McFadden K, Griffin T, Wolfe JH, Zderic S, Valentino RJ (2013). A corticotropin-releasing factor receptor antagonist improves urodynamic dysfunction produced by social stress or partial bladder outlet obstruction in male rats. *Am J Physiol* 304: R940-R950.

Wouters MM, Van Wanrooy S, Casteels C, Nemethova A, de Fries A, van Oudenhove L, van den Wijngaard RM, van Laere K, Boeckxstaens G (2012). Altered brain activation to colorectal distention in visceral hypersensitive maternal separated rats. *Neurogastroenterol Motil* 24: 6786–85.

Xiang B, Biji S, Liu JX, Chu WC, Yeung DK, Yeung CK (2010). Functional brainstem changes in response to bladder function alteration elicited by surgical reduction in bladder capacity: a functional magnetic resonance imaging study. *J Urol* 184, 2186-2191.

Yaguchi H, Soma H, Miyazaki Y, Tashiro J, Yabe I, Kikuchi S, Sasaki H, Kakizaki

1 H, Moriwaka F, Tashiro K. (2004). A case of acute urinary retention caused by
2 periaqueductal grey lesion. *J Neurol Neurosurg Psychiatry*. 75:1202-1203.
3

4 Zagorodnyuk VP, Gibbins IL, Costa M, Brookes SJH, Gregory SJ (2007). Properties
5 of the major classes of mechanoreceptors in the guinea pig bladder *J Physiol* 585
6
7
8
9 147–163.
10
11
12
13
14
15
16
17
18
19
20
21
22
23
24
25
26
27
28
29
30
31
32
33
34
35
36
37
38
39
40
41
42
43
44
45
46
47
48
49
50
51
52
53
54
55
56
57
58
59
60
61
62
63
64
65

Fig 1. Concept of 'cognitive voiding'. Afferent input from the bladder is subject to a high level of analysis based on context and social situation before the command to void is issued. Reproduced from Gillespie (2013) with permission.

Fig 2 Circuitry for micturition. The basic spino-midbrain-spinal circuit (purple arrows) is normally inhibited by tonic activity in a midbrain-thalamo-cortical loop (red arrows), which must be disinhibited to allow a void to occur. Under conditions of extreme urgency the anterior cingulate area may be stimulated to activate an emergency pathway, which bypasses the midbrain (yellow arrows). Depending on emotional state, activity in hypothalamic circuits (blue arrows) may further modulate the excitability of the midbrain circuit.

Abbreviations: dACC: dorsal anterior cingulate; Hip: hippocampus; Hyp: hypothalamus; INS: insula; PAG: periaqueductal gray matter; med PFC, lat PFC: medial and lateral prefrontal cortex; PMC: pontine micturition centre; SMA: supplementary motor area. Reproduced from the Griffiths and Fowler (2013) with permission.

Fig 3. Continuous cystometry in response to infusion of saline into the bladder (6ml h⁻¹) in a male adult urethane-anaesthetised rat. As the EEG cycles from an activated (low amplitude high frequency activity, low 0.5-1.5Hz band power) to a de-activated state (high amplitude, low frequency EEG waveform, high 0.5-1.5Hz band power) voiding threshold increases (red broken lines) and end-filling compliance decreases (increase in slope of red oblique lines). Crook and Lovick unpublished work.

Figure

[Click here to download high resolution image](#)

Figure
[Click here to download high resolution image](#)

Figure

[Click here to download high resolution image](#)

Supplementary Material

[Click here to download Supplementary Material: Lovick Auton Neurosci rev annotated.docx..docx](#)