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Purinergic Signaling in Spinal Cord Injury Induced-neurogenic Bladder: A Mini Review Study

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Abstract

Normal micturition (urination) including storage and periodic elimination of urine requires proper function of both the bladder and the urethra and a healthy central nervous system (CNS), which coordinates sympathetic, parasympathetic, and somatic nervous system activity. Any disruption in this pathway leads to the neurogenic bladder (NGB). Adenosine triphosphate (ATP) besides acetylcholine leads to bladder contraction and voiding reflex in many species. The main neurotransmitter of healthy human for initiating muscle contraction is acetylcholine. However, in pathological conditions such as NGB, purinergic components increase and its signaling is introduced as a new pathway. This review discusses the purinergic signaling pathway and its role in NGB following spinal cord injury (SCI)

Keywords: *Purinergic Signaling, Spinal Cord Injury,neurogenic bladder*

Introduction

Scientists discovered that the exocytotic vesicular release of Adenosine triphosphate (ATP: a purine) plays a role as a co-transmitter with acetylcholine from parasympathetic neurons. this process leads to bladder contraction and voiding reflex in many species [1]. Purinergic signaling in living bladder and urethra tissues have various functions such as controlling of contraction/relaxation of mammalian bladder and relaxation of the urethra [2]. Besides, ATP (a fast excitatory neurotransmitter or neuromodulator) plays vital roles in cell proliferation, differentiation, development and regeneration [3], as well as in pathological conditions [4,5]. There are four subtypes of P1 (adenosine) receptor, seven subtypes of P2X ion channel receptors, and eight subtypes of the P2Y G protein-coupled receptor [6]. Pathophysiology and therapeutic potential of purinergic signaling are considered as novel tool for managing many pathologies. For example, P1 receptor agonists are used for the treatment of supraventricular tachycardia. Clopidogrel is a P2Y12 antagonist. It blocks P2Y12 receptor-mediated platelet aggregation, and hence is the best choice for treatment of thrombosis and stroke. P2X3 receptor antagonists are used for urinary incontinence, or P2X7 receptors antagonists are being investigated for the treatment of neurodegenerative diseases [7-10].

The purinergic pathway has been proposed in many bladder dysfunctions due to interstitial cystitis/painful bladder syndrome, lower urinary tract symptoms, diabetes, aging, or secondary to spinal cord injury (SCI). This pathway is active in bladder [detrusor muscle or urothelium], central or peripheral nervous system, as well as motor and sensory nerves [11-14]. Herein, we discuss the purinergic signaling pathway and its role in the neurogenic bladder (NGB) following spinal cord injury (SCI).

Neuropathway of micturition

The normal function of the bladder consists of storage and emptying of the urine in a coordinated and controlled manner [15]. This coordination is regulated by the central and peripheral nervous systems [16] by integrated neural circuits at the level of the forebrain, brain and spinal cord. The storage of urine depends on the lumbosacral spinal cord reflexes, while voiding involves a spino-bulbo-spinal reflex. The areas which are controlled by the cortex, (e.g. the frontal, cingulate gyri regions,

and subcortical regions) have an inhibitory effect on the urine at the pons level and an excitatory effect on the external urinary sphincter (EUS). This allows for urinary voluntary control, and delay the drainage of the bladder until achieving an appropriate time and location. Pontine micturition center (PMC) which often known as the Barington's nucleus or M-region, is essential for urine coordination. At the emptying phase, PMC sends a stimulatory effect to the spinal cord that causes contraction of the detrusor, while simultaneously has an inhibitory effect on the thoracolumbar spinal cord, which causes relaxation of intrinsic urinary sphincter (IUS). The overall effect is to allow bladder contents to be drained. In contrast, in the bladder storage phase, PMC inhibition leads to spinal cord suppression which relaxes detrusor muscle, while simultaneously stimulate the thoracolumbar spinal cord, and leads to sphincter contraction. The overall effect is to permit the filling or storage of urine in the bladder [17-20].

The lower urinary tract is innervated by both autonomic and somatic innervations. Peripheral innervation comes from the pelvic (sacral parasympathetic), hypogastric (thoracolumbar sympathetic) and pudendal (somatic) nerves and any injury to these nerves can result in partial or complete denervation of the bladder [21]. The parasympathetic system is derived from S2–4 roots and innervates the bladder and smooth muscle of the IUS. Its efferent nerves control the bladder emptying using cholinergic neurotransmitters with relaxation of the sphincter valve smooth muscle and excitation bladder muscle wall [22].

The sympathetic nerve (from T10-L2 via the hypogastric nerve) innervates the bladder base, internal sphincter and proximal urethra. It relaxes the detrusor by releasing noradrenergic transmissions and excites the smooth muscle of sphincter [21,23]. Somatic innervation comes from the S2–4 roots via the pudendal and pelvic nerves, terminating in the striated sphincter, and by contracting the EUS cause storage of urine [24]. Any disruption in this pathway leads to NGB which is urinary bladder dysfunction caused by neurological diseases like as SCI, Parkinson disease (PD), multiple sclerosis (MS), cerebral vascular accident (CVA)/stroke, as well as spina bifida [25-27]. Symptoms of NGB are often among the early manifestations of neurological disease with a range from detrusor underactivity to overactivity, depending on the site of neurologic injury. For example, suprapontine causes such as stroke or traumatic brain injury; degenerative diseases (PD, Alzheimer disease, dementia with Lewy bodies); hydrocephalus, normal pressure hydrocephalus, cerebral palsy, and neoplasm are predominantly accompanied with storage phase symptoms, in which the bladder is overactive and sphincter of the urethra is normo-active. Infrapontine–suprasacral causes such as demyelinative (multiple sclerosis, transverse myelitis, trauma, SCI); vascular disease (arteriovenous malformations, spinal cord infarction); neoplasm [metastasis, primary]; hereditary (hereditary spastic paraparesis); infections [tropical spastic paraparesis (HTLV- I) lead to overactivity of urethra as well as bladder. In these lesions, normal bladder function as well as coordination between bladder and urinary sphincter is impaired and neurogenic detrusor overactivity (NDO) manifests which is recognized by uninhibited bladder contraction and detrusor-sphincter- dyssynergia (DSD) [28,29]. For example, after upper motor neuron lesions above T11 level in mice, disruption of descending neural control between higher centers and bladder / urethral lead to bladder areflexia and DSD that leads to urinary retention. Following remodeling the connection between neurons, NDO exhibits [30,31].

Infrasacral causes due to spina bifida, intervertebral disk prolapses, arachnoiditis, diabetes mellitus, pelvic surgery and nerve injury lead to bladder underactivity. In these conditions, the sphincter of urethra's function is normo-active or underactive.

Role of Purinergic Signaling in rodents

Scientists discovered the exocytotic vesicular release of ATP as a co-transmitter with acetylcholine from parasympathetic neurons leads to bladder contraction and voiding reflex in many species [1]. Physiological roles of purinergic signaling in living bladder and urethra tissues consist of control of contraction/relaxation of the mammalian bladder and relaxation of mammalian urethra [2]. Besides, ATP (a fast excitatory neurotransmitter or neuromodulator) plays a key role in cell proliferation, differentiation, and death in development and regeneration [4,5].

ATP is a signaling molecule that can act as a neurotransmitter and bind with 2 groups of receptors: ionotropic (P2X) and metabotropic (P2Y) [1,32]. There are seven P2X receptors (P2X1-7) and eight for P2Y (P2Y1, 2, 4, 6, 11, 12, 13 and14). P2X and P2Y receptors are present in urothelial cells in both sensory transduction [by releasing ATP from the umbrella cells], and the function of the bladder [33]. The relationship between purinergic signaling and bladder function is demonstrated by the knockout of P2X2/P2X3 in experimental models [34]. After ATP release, P2X3 receptors on suburothelial sensory nerves initiate the voiding reflex and mediate the sensation of bladder filling and urgency. The other mechanism of ATP is its effect on suburothelial interstitial cells/myofibroblasts generating via Ca (2+) transient through gap junctions with sending signals from urothelium to detrusor muscle [35].

Prolonged purinergic receptors activation leads to excitotoxicity and neurodegeneration [36,37]. The results of immunohistochemistry staining of these receptors showed that P2X2 staining is stronger. An experimental study showed a therapeutic effect in SCI developed overactive bladder by using A-317491 (as a selective P2X2/P2X3 purinergic receptor antagonist) via increment of contractions intervals [34]. moreover Brilliant blue G (P2X7 receptor antagonist) decreases astrocytes and microglia activation and lead to neuron protection from excitotoxicity and inflammatory responses [38]. In SCI, ATP release is seen in response to mechanosensory cholinergic receptor activation followed by P2X7 receptor activation. Hence, inhibition of P2X7 receptors can improve SCI recovery by oxidizing ATP and decrease cell death [39]. Also, purinergic signaling affects bladder function in both central and peripheral (afferent and efferent components) nervous system. Besides, this signaling can alter smooth muscle of bladder and also urethra [40].

Role of Purinergic Signaling in human

In healthy human the principal neurotransmitter that initiates muscle contraction is acetylcholine that acts on detrusor muscle cells by transmembrane muscarinic receptors and the role of ATP is minor [32]. All five muscarinic receptor subtypes exist in human detrusor; however, M2 and M3 receptor subtypes are predominantly expressed [41]. In pathological conditions such as NGB, purinergic components increased to about 40% and its signaling [the binding of ATP to its receptors] is introduced as a new pathway in the pathogenesis of many types of NGB [1].

Purinergic Signaling in rodents after SCI-induced NGB

The mechanism of acting the selective P2X2/P2X3 purinergic receptor antagonist (A-317491) in the treatment of SCI-induced NDO in an animal model [42] is increasing voiding interval, reducing non-voiding contractions, and increasing pressure threshold for voiding. A study on the SCI model on rabbits showed that from the efferent motor side, neural mediated bladder contractility (with electrical field stimulation (EFS)) shifted from purinergic predominance to cholinergic predominance [43]. Urothelial mechanosensory cholinergic receptor activation in SCI rats leads to ATP release from the spinal cord [44]. The mechanism of excessive release of ATP by the traumatized spinal tissue is the activation of P2X7 receptors [45] and inhibition of P2X7 receptor can improve recovery after SCI by oxidizing ATP and diminishing the cell death in the peritraumatic zone.

The other mechanism of voiding dysfunction following SCI is prolongation of purinergic receptor activation, which results in excitotoxicity-based neuronal degeneration. After SCI, P2X7 receptor antagonist acts by direct reduction of local activation of astrocytes and microglia, as well as neutrophil infiltration [38] and thus protect spinal cord neurons from purinergic excitotoxicity and reduce local inflammatory responses [46]. Purinergic signaling is important in the regulation of neural myelination and plays a role in plasticity and neural development in general [47]. The role of purinergic signaling related to SCI is complex. Purinergic signaling affects bladder function at both the central and peripheral nervous system levels (by affecting both the afferent and efferent components regulating bladder function), neural development, plasticity, and repair, alteration at the detrusor smooth muscle and bladder urothelial level [48].

Treatment methods influencing on purinergic pathway

Anticholinergic drugs are the most common form of treatment used for NDO [20,49]. The mechanisms of action are inhibition of acetylcholine release in post-junctional muscarinic receptors and consequently increase the bladder capacity and improve the storage phase of micturition. These drugs also target muscarinic receptors on the bladder smooth muscle [50,51].

Botulinum neurotoxins (BoNTs) are bacterial proteases produced by Clostridium botulinum and related species [52]. There are seven serotypes of BoNTs that termed A–G. They bind to and enter synaptic terminals and cleave one of the soluble N-ethylmaleimide-sensitive factor attachment protein receptor (SNARE) proteins to mediate the fusion of synaptic vesicle and lead to inhibition the exocytosis of neurotransmitters, vesicle-associated membrane protein (VAMP), synaptosomal-associated protein 25 (SNAP25), or syntaxin [53].

A subtype of Botulinium toxin A (BTX-A) is one of the most treatment strategies of NGB and act by decreasing ATP release [54] and modulating the release of substance P, calcitonin gene-related peptide and glutamate [55]. It binds to an extracellular receptor (ganglioside and presumably synaptic vesicle protein 2C) and disrupts the fusion of calcium- mediated release of acetylcholine vesicle [56] within the neuronal wall by cleaving the synaptosomal-associated protein-25 in the synaptic fusion complex in the neuronal cytosol [57]. Consequently, the paralysis of the low-grade contractions of the unstable detrusor while still allowing high-grade contraction that initiates micturition is happening. Additionally, it acts on the afferent nerve activity by modulating the release of ATP in the urothelium; blocking the release of substance P, calcitonin gene-related peptide, and glutamate from afferent nerves. It also reduces the levels of nerve growth factor [58].

Conclusions

Alterations in purinergic signaling have been described in bladder dysfunction secondary to SCI-induced NDO. In SCI, ATP release is seen in response to mechanosensory cholinergic receptor activation. After ATP release, P2X3 receptors on suburothelial sensory nerves initiate the voiding reflex and mediate the sensation of bladder filling and urgency. Activation of P2X7 receptor following SCI is reported the other injury mechanism. The prolonged purinergic receptors activation leads to excitotoxicity and neurodegeneration. Hence inhibition of P2X7 receptors can improve SCI recovery by oxidizing ATP and decrease the cell death. Pharmacotherapies such as muscarinic receptor antagonists and botulinum toxin type-A may act through suppression of afferent activity or inhibition of remodeling.

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Conflict of Interest

None

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