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Glutamate Transporter Activators as Anti-Nociceptive Agents

Anti-Nosiseptif Ajanlar Olarak Glutamat Taşıyıcı Aktivatörleri

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Abstract

The effective management of chronic pain remains enigmatic. There is a paucity of effective mechanistically-based approaches employed. Chronic visceral pain is a particularly difficult subcategory to manage. Glutamate is the most predominant excitatory neurotransmitter and mediates many aspects of sensory function including acute and chronic pain. There is a growing literature describing the efficacy of physiologically dominant glutamate transporter GLT-1 up-regulation in attenuating chronic visceral and somatic nociception. Since glutamate is the major excitatory neurotransmitter released in the first central synapse of the pain-transmitting afferent neurons, augmentation of GLT-1 activity, which reduces extracellular levels of glutamate, may be an important target for pain management strategies. This review summarizes studies in our laboratory and others which highlight findings that GLT-1 up-regulation by transgenic, pharmacologic and viral transfection approaches attenuate a host of nociceptive responses emanating from visceral or somatic sources in animal models. The study also outlines the future work that will be required to ascertain the translational potential of this approach.

Key Words: Glutamate, Pain, Transporters

Özet

Kronik ağrının etkili tedavisi halen çözümlenmemiştir. Etkin çalışan az sayıda mekanistik tabanlı yaklaşım bulunmaktadır. Özellikle kronik visseral ağrı yönetilmesi zor bir alt kategoridir. Glutamat, en baskın eksitatör nörotransmiterdir ve akut ve kronik ağrı da dahil olmak üzere, duyusal fonksiyonun pek çok yönüne aracılık etmektedir. Kronik visseral ve somatik nosisepsiyonu azaltmak için, fizyolojik olarak baskın glutamat taşıyıcı GLT-1'in yukarı regülasyonunun etkinliğini açıklayan, giderek artan bir literatür bulunmaktadır. Glutamat, ağrı ileten afferent nöronların ilk merkezi sinapsından salınan başlıca eksitatör nörotransmitter olduğu için, GLT-1 aktivitesinin artırılması, böylelikle ekstrasellüler glutamatın azaltılması, ağrı yönetimi stratejilerinin önemli bir hedefi olabilir. Bu derleme, hayvan modellerinde transgenik, farmakolojik ve viral transfeksiyon yaklaşımlarıyla GLT-1 yukarı regülasyonunun visseral ve somatik kaynaklardan köken alan nosiseptif yanıtların çoğunu yavaşlattığının altını çizen bizim ve diğer laboratuvarların çalışmalarını özetlemekte ve bu yaklaşımın translasyonel potansiyelini ortaya koymak için gelecekte yapılması gereken çalışmaların taslağını çizmektedir.

Anahtar Kelimeler: Ağrı, Glutamat, Taşıyıcılar

Introduction

There is a paucity of effective therapeutic options for the millions of patients suffering from chronic and persistent pain disorders. Chronic pain, defined as the persistence of pain in the absence of injury or long after an inciting injury has resolved, represents a significant clinical challenge [1]. Progressive increasing drug treatment "ladders" are often utilized in the management of persistent pain, starting with non-steroidal anti-inflammatory agents and progressing to the use of mild (codeine) and then strong (morphine) opioid drugs. Anticonvulsants and antidepressants have also been employed to reduce neuronal excitability characteristic of many pain disorders; however these drugs act through pathways not specific to pain-related pathways, and thus off-target effects of these drugs limit their utility. For example, opiate agonists have major side effects including somnolence, constipation and urinary retention; continuous use of opiates results in the development of tolerance requiring the escalation of dosage to achieve analgesia, which in turn increases the risk of drug abuse.

Chronic visceral pain, or pain associated with the internal organs, is a particularly difficult pain subcategory to manage. These disorders consume the practice of gastroenterologists, cardiologists, urologists, gynecologists and internists. These disorders may be exacerbated by inflammatory processes [2], but a significant proportion are of unknown etiology, termed "functional bowel disorders" [3]. It is clinical lore that visceral pain evokes particularly strong emotional responses, given that dysregulation of the internal organs often signals a medical emergency. At present, the best treatment modalities only ameliorate symptoms [4], and are not specific to the underlying cause of these disorders.

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An emerging aspect of visceral pain disorders is the spread of nociception to organs or somatic sites that were not originally the foci of the nociceptive response [5]. For example, patients diagnosed with interstitial cystitis present with symptoms of irritable bowel syndrome [6]. Interactions between afferent inputs at the spinal or supraspinal level are thought to be responsible for these findings (termed "viscera-viscero" or "somato-viscero" convergence) [6].

Glutamate is the most predominant excitatory neurotransmitter and mediates many aspects of sensory function including acute and chronic pain [2]. Strategies to reduce glutamatergic neurotransmission can mitigate nociception. Extracellular glutamate is regulated by the actions of five excitatory amino acid transporters located in neurons and glia. The physiologically dominant subtype, GLT-1 (rodent homologue) or human homologue (EAAT2) is expressed predominantly in glia throughout the CNS. Studies suggest that this transporter is responsible for the removal of over 90% of extracellular glutamate by rapidly removing it from the synaptic cleft after release from pre-synaptic terminals [7]. Since glutamate is the major excitatory neurotransmitter released in the first central synapse of the pain-transmitting afferent neurons, and perhaps from non-neuronal sources, augmentation of GLT-1 activity may be an important target of pain management strategies [8]. Indeed, substances that augment glutamate transporter function alleviate nociception and glutamate transporter blockers worsen pain [9-14].

Described below are studies in our laboratory and others which highlight the finding that GLT-1 up-regulation by transgenic, pharmacologic and viral transfection approaches attenuate a host of nociceptive responses in animal models.

GLT-1 activators relieve visceral nociception

The most common technique used in pre-clinical study of visceral nociception is the visceromotor response to hollow organ distension, described in detail elsewhere [15]. Transgenic and pharmacologic approaches demonstrate that GLT-1 up-regulation attenuates the visceromotor response to colo-rectal distension [16]. The pharmacological anti-nociceptive effect, produced by 1 week treatment with daily ceftriaxone (200mg/kg), was attenuated by pretreatment with the selective GLT-1 antagonist dihydrokainate, which implies that the GLT-1 transporter has a role in mediating anti-nociception [16].

Recently, the anti-nociceptive effect of enhanced GLT-1 expression was demonstrated in the bladder [17]. A one-week ceftriaxone treatment was effective to attenuate the visceromotor response to bladder distension, irritant-induced bladder hyperalgesia, colon-to-bladder cross organ sensitization, and chronic visceral nociception after colonic or bladder inflammation [18, 19]. Importantly, inflammogen-mediated hyperalgesia was attenuated by both pre-emptive and therapeutic administration of 1-week ceftriaxone, providing evidence of translational relevance of glutamate transporter activators [18].

Previous studies have characterized the anti-nociceptive effect of GLT-1 up-regulation have been conducted. A spinal site of action has been suggested, based on the effectiveness of intrathecal, but not intracisternal dihydrokainate to reverse the anti-nociceptive effect of 1-week ceftriaxone [18]. Studies in other laboratories have corroborated a spinal site of GLT-1 up-regulation to attenuate somatic nociception [20, 21]. An important area in the study of functional bowel disorders is role of early stressors mediating adult hyperalgesia [22]. A recent report shows effectiveness of glutamate transporter activator riluzole to mitigate the visceral hypersensitivity caused by premature maternal separation in the rat [23]. Thus, in the most important pre-clinical models of visceral pain, GLT-1 up-regulation showed significant anti-nociceptive effects.

Pharmacologic GLT-1 up-regulation is effective in selective categories of somatic pain. Ceftriaxone was effective in reversing thermal and to a lesser extent mechanical hyperalgesia after chronic constriction nerve injury in the rat [20]. Both pre-emptive and therapeutic administration of ceftriaxone was effective. Effectiveness of GLT-1 up-regulation was also shown recently in the hyperalgesia and allodynia associated with streptozocin-induced diabetic neuropathy [24]. In contrast, GLT-1 up-regulation was not effective in the pre-clinical measures of acute thermal or tactile somatic nociception [18, 21].

GLT-1 up-regulation and anti-nociception; gene therapy approaches

Gene therapy is the introduction of genetic material to a patient's cells for therapeutic benefit [25-28]. Available strategies to evoke specific cellular/tissue tropism of virus penetration and thus anatomically selective gene expression make this approach attractive for putative treatment of numerous acquired and inherited disorders. Over the past several years the field has witnessed significant advances both pre-clinically and in clinical trials. The first example of gene therapy for pain therapeutics was introduced to clinical trials [29]. Utilizing this approach with respect to GLT-1 up-regulation, our laboratory showed that injection of neonatal mice with adeno-associated serotype 9 (AAV9)-GLT at 19-26 d produced a 62% reduction in the visceromotor response to colonic distension compared to animals injected with vehicle [18]. This was correlated with a 111% enhanced glutamate uptake seen in AAV9-GLT injected animals compared to controls. Thus, proof of concept was achieved regarding the potential of viral-vector mediated gene therapy approaches for GLT-1 transfection to mitigate visceral pain.

Recently, transfection of adenoviral linked GLT-1 gene was effective to attenuate nociception in animal models of neuropathic and inflammatory pain [21]. Intra-spinal infusion of adenoviral vectors expressing the GLT-1 gene increased GLT-1 expression in the spinal cord 2-21 days after the infusion. Spinal GLT-1 gene transfer had no effect on acute mechanical and thermal nociceptive responses in naive rats, whereas it significantly reduced the inflammatory mechanical hyperalgesia induced by hindlimb intraplantar injection of carrageenan/kaolin. Spinal GLT-1 gene transfer 7 days before partial sciatic nerve ligation recovered the extent of the spinal GLT-1 expression in the membrane fraction that was decreased following the nerve ligation, and prevented the induction of tactile allodynia. However, the partial sciatic nerve ligation-induced allodynia was not reversed when the adenoviruses was infused therapeutically, 7 or 14 days after the nerve ligation.

Translational potential of GLT-1 up-regulation in the treatment of chronic pain

Given the marked therapeutic potential of enhancement of glutamate transporters, there are ongoing efforts of drug screening for potent and selective regulators of glutamate levels [30-32].

Potential clinical utility of the approach to augment glutamate transporter activity depends on the lack of potential untoward effects. Pre-clinical studies performed thus far utilizing the rotarod, plus maze and open field locomotion tests demonstrated that GLT-1 up-regulation does not alter motor or cognitive function [18, 33]. More sophisticated tests of motor, cognitive and neuronal function will be required.

The interest in anti-nociceptive effects of GLT-1 up-regulation adds to the widespread interest in utilizing spinal GLT-1 up-regulation in a wide range of CNS disorders mediated by enhanced glutamatergic neurotransmission [8]. Down regulation of GLT-1 occurs in activated astrocytes; seen often following chronic opioid therapy and in chronic pain models. Efficacy has been demonstrated after intraspinal ceftriaxone in 1) hyperalgesia and allodynia following chronic morphine [8], 2) pain following peripheral neuropathy, inflammation or spinal cord injury [20, 21, 34] 3) tactile hyperalgesia and progression of motor weakness and paralysis in animal models of multiple sclerosis [8]. There are also ongoing studies regarding the efficacy of GLT-1 up-regulation in other conditions mediated by enhanced extracellular glutamate such as Alzheimer's disease, Huntington's disease, epilepsy, amyotrophic lateral sclerosis and the regulation of addictive behaviors [35-39]. Indeed, in the United States, GLT-1 enhancer ceftriaxone has reached Phase III clinical trials for the treatment of amyotrophic lateral sclerosis (ClinicalTrials.gov Identifier: NCT00349622).

Conclusion

There is a growing literature describing the efficacy of GLT-1 up-regulation to attenuate chronic visceral and somatic nociception. With respect to potential analgesic utility, there are several important questions to answer: 1) duration of anti-nociceptive effect of GLT-1 up-regulation, 2) optimal dosing regimen of administration to provide beneficial effect 3) assessment of blunted supraspinal neuronal response related to pain transmission [40] and 4) demonstration of anti-nociceptive effect of GLT-1 up-regulation in non-rodent vertebrates.

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