Published in final edited form as:

Curr Opin Anaesthesiol. 2008 October ; 21(5): 570–579. doi:10.1097/ACO.0b013e32830eddbdf.

Neuron-glia crosstalk gets serious: Role in pain hypersensitivity

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Abstract

Purpose of review—Recent studies show that peripheral injury activates both neuronal and non-neuronal or glial components of the peripheral and central cellular circuitry. The subsequent neuron-glial interactions contribute to pain hypersensitivity. This review will briefly discuss novel findings that have shed light on the cellular mechanisms of neuron-glial interactions in persistent pain.

Recent findings—Two fundamental questions related to neuron-glial interactions in pain mechanisms have been addressed: 1) what are the signals that lead to central glial activation after injury and 2) how glial cells affect CNS neuronal activity and promote hyperalgesia.

Summary—Evidence indicates that central glial activation depends on nerve inputs from the site of injury and release of chemical mediators. Hematogenous immune cells may migrate/infiltrate to the brain and circulating inflammatory mediators may penetrate the blood brain barrier to participate in central glial responses to injury. Inflammatory cytokines such as IL-1 β released from glia may facilitate pain transmission through its coupling to neuronal glutamate receptors. This bidirectional neuron-glial signaling plays a key role in glial activation, cytokine production and the initiation and maintenance of hyperalgesia. Recognition of the contribution of the mutual neuron-glial interactions to central sensitization and hyperalgesia prompts new treatment for chronic pain.

Keywords

astroglia; microglia; cytokines; NMDA receptor; hyperalgesia

Introduction

Glia cells greatly outnumber neurons in the brain and have intimate relationships with neurons, yet brain functions have been attributed mainly to neurons and active involvement of glial cells in brain function has long been overlooked. However, cumulating evidence has been constantly challenging the limits of the neuron theory of Cajal. For example, gliapse, a concept that is based on an anatomical relationship between astrocyte and neurons and infers neuron-glial signaling, was proposed to argue that the glia and the neuron work together as the fundamental functional unit of the brain [1]. Astrocytes may modulate synaptic strength through a “tripartite” synapse that includes pre- and post-synaptic membranes and extrasynaptic astrocytic contacts [2,3**]. A functional synapse may even be tetrapartite that includes contributions from microglia [4] (Fig. 1). Ample work has demonstrated that glial cells participate in normal brain function and further contribute to neurological disorders including chronic pain [5–7*]

There has been a general consensus that neuron-glia interactions play critical roles in the development of central sensitization and hyperalgesia [7*,8*,9*]. Most recent studies have addressed two fundamental questions related to neuron-glia interactions in the mechanisms of persistent pain: 1) what are the signals that lead to glial activation after injury and 2) how do glial cells affect central neuronal activity and promote hyperalgesia. This essay will briefly summarize some key findings in the past year on the mechanisms of central neuron-glia interactions and persistent pain.

Glial activation after injury

Glial cells, primarily microglia and astroglia, exhibit dynamic plasticity by converting from an resting or quiescent state to a reactive state after injury and assume a more active role in modulating neuronal activity. Activation of glial cells is monitored by their expression of specific cellular markers and kinases, changes in morphology and a release of a variety of immune substances [8*]. Typically, activated astroglia and microglia exhibit hypertrophy and increased production of specific cellular proteins and/or cell-surface markers. Glial fibrillary acidic proteins (GFAP) are selectively increased in activated astrocytes and CD11b (cluster of differentiation 11b, integrin α M, Mac-1 α) is induced in activated microglia. Additionally, S100 β is a calcium-binding peptide produced mainly by astrocytes and can be used as a functional marker for astroglial activity [10]. Iba1 (ionized calcium-binding adapter molecule 1) is a calcium binding protein that is specifically expressed in microglia and can be used as a functional marker for microglia [11,12]. Glial activity can also be assessed by the phosphorylation state of molecules such as p38 mitogen-activated protein kinase (MAPK) [13**,14*] and extracellular signal-regulated kinases (ERK) [15], glial glutamate transporters [16] and gap junction proteins [17,18**,19], and toll-like receptors (TLRs) [20,21] and the complement components [22*] (Fig. 1).

New evidence supports a role of glia in persistent pain. Moss et al. [23*] show that immaturity of the microglial response in newly born rats coincides with the absence of allodynia after nerve injury. Glial activation can spread into areas of the spinal cord innervated by uninjured nerve [24*], consistent with extraterritorial hyperalgesia [25]. Inhibition of lysosomal cysteine protease cathepsin S in spinal microglia suppressed microglial activation and reversed neuropathic pain [26**]. The activation of microglia in the spinal cord by sciatic nerve injury has been quantified by stereological techniques, which shows that the total number of spinal microglia after spared nerve injury was increased by almost 3-fold [24*]. The flow cytometry technique has been adapted to demonstrate and quantify microglial activation in the rat spinal cord after nerve injury [27]. It is appreciated that glial activation also occurs at multiple supraspinal levels [28–30].

Astrocytes interconnect by connexin 43, an astrocytic gap junction protein, and form a functional syncytium through which waves of calcium ions spread between astrocytes (Fig. 1). Astrocytes are uniquely situated to interact with neurons. A three-dimensional reconstruction analysis indicates that a single cortical astrocyte enwraps on an average four neuronal somata and contacts 300–600 neuronal dendrites [31*]. New studies also further demonstrate the contribution of astroglia to persistent pain [32]. Orofacial inflammation induces astroglial activation in the regions of the spinal trigeminal complex with a time course correlating with hyperalgesia [18**]. Hyperexcitability of trigeminal nociceptive neurons is attenuated by application of methionine sulfoximine, an inhibitor of astroglial glutamine synthetase that catalyzes conversion of glutamate to glutamine [33*]. This effect is likely a result of reduced supply of glutamate neurotransmitters due to inhibition of the astroglial glutamate-glutamine shuttle (Fig. 1). In mice over-expressing the chemokine CCL2 (MCP-1) in astrocytes under control of the GFAP promoter, the CFA-induced edema and thermal hyperalgesia are significantly enhanced [34*].

The functional consequence of astroglial activation is complex. Astrocytic glutamate transporters (GLT-1) uptake glutamate into astrocytes to help to maintain an appropriate level of extracellular glutamate concentration. Models of neuropathic pain have been associated with a decrease in GLT-1 activity [16,35,36] although astroglia are activated in response to injury [18**,37]. Thus, there appears to be a reciprocal relationship between the astrocytic activation and GLT-1 expression. A reduction in GLT-1 expression may lead to a build up of glutamate concentration in the synaptic cleft, leading to neuronal hyperexcitability and hyperalgesia. Interestingly, the often-used glial modulator/inhibitor propentofylline produces multiple effects on astrocytes [38]. In primary astrocytic cultures that exhibit an activated phenotype, propentofylline suppresses lipopolysaccharide (LPS)-induced chemokine release but induces GLT-1 expression and glutamate uptake [38]. These cellular actions of propentofylline are consistent with its anti-allodynic effect [39*].

There seems to be a coordinated activation of microglia and astroglia, the details of which are currently unclear. Studies have suggested that microglial activation precedes activation of astrocytes [15,29,40–42]. Activation of TLR4 on microglial cells may lead to astroglial activation [4,20,]. However, long-lasting microglial activation (28–42 d) has been observed post-L5 spinal nerve transection [39*]. Kawasaki et al. [43**] showed that early microglial and later astroglial activation were associated with neuropathic pain. Matrix metalloproteinase (MMP)-9 induces early microglial activation and MMP-2 is related to later astroglial activation after L5 spinal nerve ligation [43**] (Fig. 1). In MMP-9 knockout mice, neuropathic pain was attenuated early (1–3 d post-injury) but was fully expressed later at 10 d post-injury. These findings point to potential respective contributions of microglia and astroglia to initiation and maintenance, or earlier and later phases of central sensitization and persistent pain.

Signals leading to central glial activation after peripheral injury

The signals that carry peripheral injury signals and trigger glial activation are still elusive. New findings suggest that both neuronal and non-neuronal factors induce glial activation in the CNS. Neurotransmitters/modulators glutamate, brain-derived neurotrophic factor (BDNF), substance P (SP) and ATP released from presynaptic terminals act not only on postsynaptic neuronal receptors, but may also reach receptors on microglia or astrocytes to produce glial activation. (Fig. 1).

Role of neural input

An intriguing possibility for triggering central glial activation is through neuronal signals. An earlier study suggested that the increase in GFAP expression in the spinal cord after nerve injury depended on N-methyl-D-aspartate receptor (NMDAR) activity [44]. Zhuang et al. [15] show that ERK is sequentially activated in neurons, microglia, and astrocytes following spinal nerve ligation, suggesting an effect of neuronal activity on glial activation. In fact, increased neuronal activity will lead to a rise in extracellular K^+ , which may force an increased uptake of K^+ into the surrounding astrocytes and lead to a change in activity [45].

The dependence of central glial activation on neuronal input has been directly examined by producing local anesthetic block of the primary afferent input [13**,18**]. A complete Freund's adjuvant (CFA)-induced increase in GFAP and hyperalgesia was abolished in lidocaine-treated rats [18**]. Electrical stimulation of nerve fibers increases intracellular calcium in glial cells [46]. Consistently, burst stimulation of the masseter nerve induced an increase in GFAP levels in the brain stem spinal trigeminal complex [47]. Pretreatment with a long-lasting local anesthetic, bupivacaine-loaded microspheres, above the nerve injury site prevented activation of p38 MAPK in spinal microglia [13**]. Further, brain stem descending input may induce spinal glial activation [48]. In a very interesting experiment by Kim et al. [49*], adding degenerating neurites of dorsal root ganglion neurons to the glial cell culture

induced proinflammatory gene expression in glia. This result implies that signals carried by damaged nerve may induce glial activation. Consistently, apoptotic neurons may release an active form of MMP-3 that activates microglia [50] (Fig 1). These findings indicate that primary afferent inputs associated with nociceptor activation after injury are necessary for central glial activation. However, glial activation may be *maintained* by signals that are independent of nerve input since post-treatment with bupivacaine after nerve injury did not reverse p38 activation in microglia [13**].

Chemical mediators

Arrival of neural input at primary afferent terminals is followed by release of neurotransmitters and other mediators. Apparently, these chemical mediators not only affect synaptic transmission, but may also induce glial activity. A number of mediators are potentially capable of mediating signals from neuron to glia, which include neurotransmitters such as SP and calcium gene-related peptide (CGRP) [18**], nitric oxide (NO) [18**,51], purinergic agents [52,53], glutamate [54] and opioid peptides [9*,55]; the chemokines fractalkine (CX3CL1) [56**], monocyte chemoattractant protein-1 (MCP-1) [41,57] and cysteine-cysteine chemokine ligand 21 (CCL21) [30]; and glucocorticoids [58]. Notably, increases in protease activity such as the serine protease tissue type plasminogen activator [59], lysosomal cysteine protease cathepsin S [26**] and MMPs [43**] in the spinal dorsal horn may be critical in cleavage and releasing signaling molecules for glial activation.

Substance P stimulates IL-1 production by astrocytes via intracellular calcium [60]. Glia may be activated by chemicals that are released from primary afferent terminals and involved in pain transmission [61]. Direct application of SP or CGRP to a medullary slice preparation induced a significant increase in GFAP and IL-1 β in the spinal trigeminal complex [18**]. Thus, activation of either CGRP or SP receptors can induce glial activation and cytokine induction. Interestingly, there is no known localization of neurokinin-1 tachykinin receptors, the primary binding site for SP, in activated astrocytes in adults *in vivo*. Other experiments show that pretreatment of a medullary slice preparation with an NO synthase inhibitor L-NAME blocked the SP-induced GFAP and IL-1 β [18**], suggesting that NO act as a messenger of SP between neurons and astroglia. These results are consistent with a role of NO in pain facilitation and the observation that NO may act upstream to glial activation [51,62,63].

Purinergic signals from the neuron may activate glia [3**]. Glial cells express multiple purinergic receptors [52] and ATP is released from synaptic terminals [64]. P2Y₁₂ receptors on microglia are upregulated after nerve injury [14*]. Microglial P2X₄ receptors are induced after spinal nerve injury with a time course that parallels allodynia [65]. The injury-induced upregulation of P2X₄ receptors requires signaling through autophosphorylation of Lyn tyrosine kinase in microglia [66]. Application of ATP directly to the spinal cord elevated CD11b and GFAP levels in the spinal dorsal horn and produced allodynia that was attenuated by glial inhibitors [67].

Excitatory amino acids can evoke membrane currents in glial cells in spinal slices [68]. It is notable that cortical astrocytes express functional NMDAR that are insensitive to magnesium block [69]. In the presence of magnesium, NMDA activated p38 MAPK in astrocytes that led to long-term potentiation of spinal neuronal activation visualized with optical imaging [70*]. Activation of microglial p38 MAPK by NMDA has also been reported [71]. These results suggest activation of astrocytes and microglia by excitatory amino acids and functional crosstalk between neurons and glia.

Opioid receptor mRNA and receptors have been found in primary astroglial cultures [55,72]. Repeated dosing of morphine activates glia in the brain and spinal cord and this opioid-induced glial activation is blocked by AV411, a glial activation inhibitor [73]. Kappa opioid receptors

may also contribute to glial activation after nerve injury (Fig. 1). In dynorphin knock-out mice or mice lacking κ opioid receptors, upregulation of GFAP in the spinal dorsal horn after nerve ligation was abolished [55]. The consequences of opioid-induced glial activation may include opioid tolerance, pain hypersensitivity, and withdrawal syndrome [9*].

Thus, multiple lines of evidence favor neurotransmitter/modulator signaling from neurons to glia. These chemical mediators may act together to affect glial activity. It should be noted, however, that most evidence regarding neuron-to-glia signaling is indirect and a direct relationship between a neuronal derived substance and its action on glial cells is difficult to prove *in vivo*. The overlapping distribution of neurotransmitter receptors in neurons and glial cells further complicates the issue [74–76] since receptor antagonists are not differentially selective for either neurons or glia cells. Recognition of the different properties of neurotransmitter receptors in glia vs. neurons may help to address this problem. For example, NMDARs in astrocytes do not appear subject to magnesium block [69] and glial and neuronal opioid receptors may exhibit different stereoselectivity [9*]. The p38 MAPK isoforms are differentially distributed in spinal neurons (p38alpha) and microglia (p38 beta) [77]. Furthermore, the localization of many neurotransmitter receptors in glial cells has yet to be confirmed *in vivo*. For example, glial neurokinin-1 tachykinin receptor (NK-1R) immunoreactivity has been localized only in cultures including spinal astroglial culture [78, 79]. When saporin-conjugated SP was used to selectively destroy NK-1R-containing neurons in the spinal cord, this procedure did not affect GFAP immunoreactivity in the spinal cord [80]. This result indicates that the saporin-SP conjugate does not affect adult spinal astrocytes, possibly due to a lack of NK-1Rs.

Besides neurotransmitters, chemokine fractalkine (CX3CL1, neurotactin) may convey signals to microglia. Fractalkine mRNA and immunoreactivity are localized to rat spinal cord and dorsal root ganglion neurons [81]. In the spinal cord, the fractalkine receptor CX3CR1 is expressed only by microglia [81,82] (Fig. 1). Spinal nerve ligation in rats induced reduction of the membrane-bound fractalkine in the dorsal root ganglion [56**], which is consistent with an increased cleavage and release of fractalkine. While the spinal fractalkine level was not altered by chronic constriction injury and sciatic inflammatory neuropathy, its exclusive receptor CX3CR1 expression in the spinal cord was upregulated after nerve injury and intra-articular injection of CFA [56**,81–83]. Lindia et al. [82] report that fractalkine was also observed in astrocytes in the spinal cord after nerve injury. Application of fractalkine to the rat spinal cord enhanced responses of dorsal horn neurons, an effect that was blocked by minocycline and likely mediated by an action on microglia [84]. Intrathecal fractalkine produced allodynia/hyperalgesia that was attenuated by antibodies to CX3CR1 [85,86]. Anti-CX3CR1 antibody also blocked activation of p38 MAPK after nerve injury and attenuated allodynia/hyperalgesia [56**,83]. Fractalkine antagonizes opioid analgesia at both spinal and supraspinal levels [87,88], an effect supporting a role of fractalkine-mediated glial activation in opioid tolerance. Clark et al. [26**] show that after nerve injury, the lysosomal cysteine protease cathepsin S in microglia is responsible for the liberation of neuronal fractalkine, which in turn stimulates p38 MAPK phosphorylation in microglia. Injection of rat recombinant cathepsin S induced allodynia in wild-type but not CX3CR1-knockout mice [26**]. MCP-1 [57,89] and CCL21 [30] are other chemokines that are expressed in neurons and may trigger glial activation. Knockout of the MCP-1 receptor CCR2 on microglia prevents microglial activation and mechanical allodynia [90**]. Glucocorticoids may induce microglia activation through NMDARs [58], although there is no direct evidence regarding a role of glucocorticoids in injury-induced glial activation [see 91].

Infiltration of hematogenous immune cells and chemical mediators (Fig. 1)

It has been observed that local anesthetic block of the sciatic nerve does not eliminate cyclooxygenase-2 (COX-2) mRNA induction in the spinal cord or increased prostaglandin E₂ levels in the cerebrospinal fluid [92], or NMDAR NR1 serine 897 phosphorylation in the spinal cord [93] after hindpaw inflammation. Some mediators of glial activation may reach their CNS target via the circulation. Inflammatory cells are capable of migrating into the CNS through the choroid plexus and activated blood brain barrier with disrupted tight junctions [94,95]. Gordh et al. [96] report that nerve injury induced a localized increase in blood-spinal cord barrier permeability that correlated with activation of astrocytes. Further evidence indicates that hematogenous macrophages are capable of invading the nervous system after nerve injury [90**]. In mice subject to transplantation of green fluorescent protein (GFP)-expressing bone marrow stem cells, sciatic nerve ligation led to infiltration of these cells into the spinal cord. The GFP donor cells that penetrated the blood-spinal cord barrier proliferate, differentiate, and exhibit the phenotype of microglia. The time course of the increased GFP cells in the spinal cord parallels that of microglial activation after nerve injury. Thus, central microglial activation may constitute both resident microglia and blood-borne macrophage components. CD4(+) T lymphocytes infiltrate into the lumbar spinal cord after L5 spinal nerve transection and may interact with resident glial cells to produce neuropathic pain [*97]. In addition, inflammatory mediators such as kinins are produced at the site of injury and may enter the brain and act on glia cells [98,99]. Interleukin-6 released into the peripheral circulation may also reach the brain [100*] (Fig. 1).

Glial modulation of neuronal activity and hyperalgesia

Activation of glial cells initiates cellular signal transduction pathways [26**,56**,99,101,102,103**] that lead to a release of a variety of substances including inflammatory cytokines [101,104,105], prostaglandins (PG) [103**], BDNF [106], ATP [53,107], NO [49*], D-serine [108] and glutamate [54,109, also see 8*,9*]. These chemical mediators in turn modulate neuronal activity and facilitate pain transmission, although the mechanisms by which such modulation occurs is only beginning to be understood.

One interesting observation is that a prototypic proinflammatory cytokine interleukine-1 β (IL-1 β) is selectively induced in astrocytes in animal models of bone cancer pain [110], CFA-induced inflammation [18**] and intracerebral hemorrhage [111]. MMP-2 is related to later astroglial activation and cleavage/release of IL-1 β after L5 spinal nerve ligation [43**]. These results suggest that astrocytes are a source of IL-1 β , although previous studies have indicated that pro-IL-1 β is produced primarily in microglia in the CNS and cleaved into bioactive IL-1 β by cysteine protease caspase 1 after secretion [104,112]. D-serine, a co-agonist of NMDAR, may also be released from astrocytes [4,108]. The inflammatory cytokines released by activated glia play important roles in persistent pain. Administration of IL-1 receptor antagonists (IL-1ra) attenuates hyperalgesia [18**,113]. Allodynia induced by spinal nerve ligation is reduced by intrathecal IL-1 β -neutralizing antibody [43**]. Deletion of IL-1 receptors and over-expression of IL-1ra significantly attenuate neuropathic pain behavior [114].

IL-1 β released from glia may modulate neuronal activity. It is known that NMDARs are involved in central sensitization and hyperalgesia. The IL-1 β signaling facilitates NMDAR activation in neurons [18**,115,116]. NMDAR antagonists blocked IL-1 β -produced hyperalgesia [117] and IL-1 receptor colocalizes with NMDAR in neurons [18**]. Incubation of medullary slices with IL-1 β induced a significant increase in P-ser896-NR1 levels in a subregion of the spinal trigeminal complex involved in pain processing. In primary mouse neuronal cultures, IL-1 β activates Src kinase that further triggers the phosphorylation of the NMDAR NR2B subunit [118**]. The effect of IL-1 β on NMDAR appears selective since

another prototype inflammatory cytokine TNF- α did not affect P-ser896-NR1 levels [18**]. The IL-1 β -induced NR1 phosphorylation was blocked by IL-1ra, but not by fluorocitrate, a glial inhibitor, suggesting that the effect of IL-1 β on NMDAR is downstream to glial activation. The signal pathways leading to IL-1 β -induced NMDAR phosphorylation involve protein kinase C, phospholipases and intracellular calcium release [18**]. Thus, IL-1 β signaling is coupled to an increased activity of neuronal NMDARs that leads to pain hypersensitivity (Fig. 1).

Prostaglandin E2 may activate signals between microglia and neurons after injury [103**] (Fig. 1). Rats receiving spinal contusion injury develop dorsal horn hyperexcitability and hyperalgesia. A microglia- and ERK1/2 phosphorylation-dependent PGE2 release was identified in these rats. Immunostaining shows location of PGE2 receptor (EP2) in neurons. Inhibition of microglia with Mac-1-SAP immunotoxin and EP2 receptor blockage suppressed microglia and reduced PGE2 levels, and reversed pain hypersensitivity [103**].

It has been suggested that astrocytes release gliotransmitters including calcium-dependent release of glutamate to affect neuronal activity [119,120,121]. Glial-derived glutamate may have an impact on neuronal hyperexcitability through an effect on extrasynaptic NMDARs [3**]. Recent work has raised questions about the ability of astrocytes to directly affect neuronal activity *in situ* by calcium-dependent glutamate release [122**]. When recording from hippocampal slices from transgenic mice that express a Gq-coupled receptor only in astrocytes, selective stimulation with an agonist that does not bind endogenous receptors induces widespread calcium elevation in astrocytes but without an effect on synaptic activity. The NMDAR channel blocker MK-801 does not affect IL-1 β -induced NMDAR phosphorylation [18**], suggesting that this effect is not mediated by extracellular glutamate including that from glial cells. Further studies should determine whether there is a direct contribution of glia-derived excitatory amino acids to central sensitization and hyperalgesia.

Recognition of the contribution of the mutual neuron-glia interactions to central sensitization and hyperalgesia prompts new treatment for chronic pain conditions. In addition to direct inhibition of neuronal activity, a variety of non-neuronal components of the brain also may be targeted for pain relief. Several glial inhibitors, antiinflammatory cytokines and other genetic approaches targeting at glial and cytokine signaling have been used in preclinical studies to produce anti-hyperalgesia [8*,18**,26**,39*,66,67,73,113,123**–128]. It is hoped that more selective agents that block glial activation of pain processing pathways will soon be developed and proven effective in fighting chronic pain.

Abbreviations

BDNF, brain-derived neurotrophic factor
 CCL21, cysteine-cysteine chemokine ligand 21
 CD11b, cluster of differentiation 11b
 CFA, complete Freund's adjuvant
 CGRP, calcium gene-related peptide
 COX-2, cyclooxygenase-2
 ERK, extracellular signal-regulated kinases
 GFAP, Glial fibrillary acidic protein
 GFP, green fluorescent protein
 GLT, glutamate transporter
 Iba1, ionized calcium-binding adapter molecule 1
 IL-1 β , interleukine-1beta
 IL-1ra, IL-1 receptor antagonist
 LPS, lipopolysaccharide

MAPK, mitogen-activated protein kinase
 MCP-1, monocyte chemoattractant protein-1
 MMP, matrix metalloproteinase
 NK-1R, neurokinin-1 tachykinin receptor
 NMDAR, N-methyl-D-aspartate receptor
 NO, nitric oxide
 PG, prostaglandins
 SP, substance P
 TLRs, toll-like receptors
 TNF, tumor necrosis factor

Acknowledgements

The authors' work is supported by NIH grants DE11964, DE15374, NS060735, NS059028.

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Fig. 1.

Schematic summary of recent findings on neuron-glia interactions in central sensitization and pain hypersensitivity. Note a synapse between an axon terminal and sensory neuron and close apposition of astrocytes and microglia. **A. Signals leading to central glial activation:** 1. Injury-generated input (small block arrows). 2. Chemical mediators released from nerve terminals (chemokines and neurotransmitters), damaged axons (MMP-3, MCP-1) and postsynaptic neurons (CCL21, DYN). 3. Hematogenous immune cells and inflammatory mediators. **B. Activation of microglia.** Note that activated microglia release a variety of mediators that in turn affect neuronal activity. **C. Role of astroglia.** Note the involvement of glutamate-glutamine shuttle and glutamate transport GLT-1, MMP-2 mediated cleavage/release of IL-1β, induction coupling of IL-1R signaling with NMDAR. Interactions between neurons and glia during nociceptive processing lead to amplified neuronal output and pain hypersensitivity. See text for further details. BK, bradykinin; CX43, connexin 43; D-ser, D-serine; DYN, dynorphin; EP2, prostaglandin E receptor subtype; gln, glutamine, glu, glutamate; GS, glutamine synthetase; KOR, kappa opioid receptor; P, phosphorylation.