Neuroimmune Interactions in Chronic Pain
Review

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Angiotensin receptors and neuropathic pain

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Abstract

Growing evidence implicates the renin–angiotensin system (RAS) in multiple facets of neuropathic pain (NP). This narrative review focuses primarily on the major bioactive RAS peptide, Angiotensin II (Ang II), and its receptors, namely type 1 (AT1R) and type 2 (AT2R). Both receptors are involved in the development of NP and represent potential therapeutic targets. We first discuss the potential role of Ang II receptors in modulation of NP in the central nervous system. Ang II receptor expression is widespread in circuits associated with the perception and modulation of pain, but more studies are required to fully characterize receptor distribution, downstream signaling, and therapeutic potential of targeting the central nervous system RAS in NP. We then describe the peripheral neuronal and nonneuronal distribution of the RAS, and its contribution to NP. Other RAS modulators (such as Ang (1-7)) are briefly reviewed as well. AT1R antagonists are analgesic across different pain models, including NP. Several studies show neuronal protection and outgrowth downstream of AT2R activation, which may lead to the use of AT2R agonists in NP. However, blockade of AT2R results in analgesia. Furthermore, expression of the RAS in the immune system and a growing appreciation of neuroimmune crosstalk in NP add another layer of complexity and therapeutic potential of targeting this pathway. A growing number of human studies also hint at the analgesic potential of targeting Ang II signaling. Altogether, Ang II receptor signaling represents a promising, far-reaching, and novel strategy to treat NP.

Keywords: Angiotensin, Neuropathy, ACE inhibitor, Neuroimmune interactions, Macrophages, Type 1 angiotensin receptor, Type 2 angiotensin receptor, Mas receptor

1. Introduction

Neuropathic pain (NP) is among the most challenging conditions to treat effectively, affecting 7% to 10% of those with chronic pain.[130] The most common NP conditions include diabetic, alcohol, and drug-induced neuropathy, and too commonly proper analgesia is not achieved by currently available medications.[144] As such, many alternative drug targets have been studied for their analgesic efficacy. Angiotensin receptor blockers are one such group of potential targets, and there are a growing number of promising preclinical and clinical studies showing the effectiveness of different Ang II receptor modulators in NP.[5,122] A large number of studies have explored the pathophysiological roles of the renin–angiotensin system (RAS) pertaining to the vasculature, heart, kidney, and brain, in the contexts of inflammation and damage repair, metabolic dysfunction, and aging. These topics have recently been reviewed comprehensively elsewhere.[40] The present review focuses on RAS signaling in NP and the novel therapeutic opportunities represented by recent discoveries.

The RAS was first described in the late 19th century as an endocrine modulator of systemic blood pressure.[8] Angiotensin II (Ang II) is the major bioactive octapeptide responsible for vasoconstriction and stimulating secretion of aldosterone and vasopressin, increasing sodium and fluid retention. Ang II is derived from the inactive precursor angiotensinogen, which was first shown to be released into the circulation by the liver (**Fig. 1**). Angiotensinogen is first cleaved by renin, yielding the inactive intermediate angiotensin I (Ang I). Angiotensin-converting enzyme (ACE) then catalyzes the conversion of Ang I into Ang II. Additional proteases that catalyze the production of Ang II were discovered more recently, such as cathepsins, carboxypeptidases, and aminopeptidases.[89] Ang II binds with similar affinity to the G-protein-coupled receptor subtypes AT1R and AT2R in multiple cell and tissue types (**Fig. 2**).[99] Unlike humans, mice and rats underwent a gene duplication event and express 2 pharmacologically identical isoforms of AT1R with different expression patterns, termed AT1a and AT1b.[61,112] Other fragments besides Ang II (Ang 1-8) are formed by alternative pathways, such as Ang III (Ang 2-8), Ang IV (Ang 3-8), and Ang (1-

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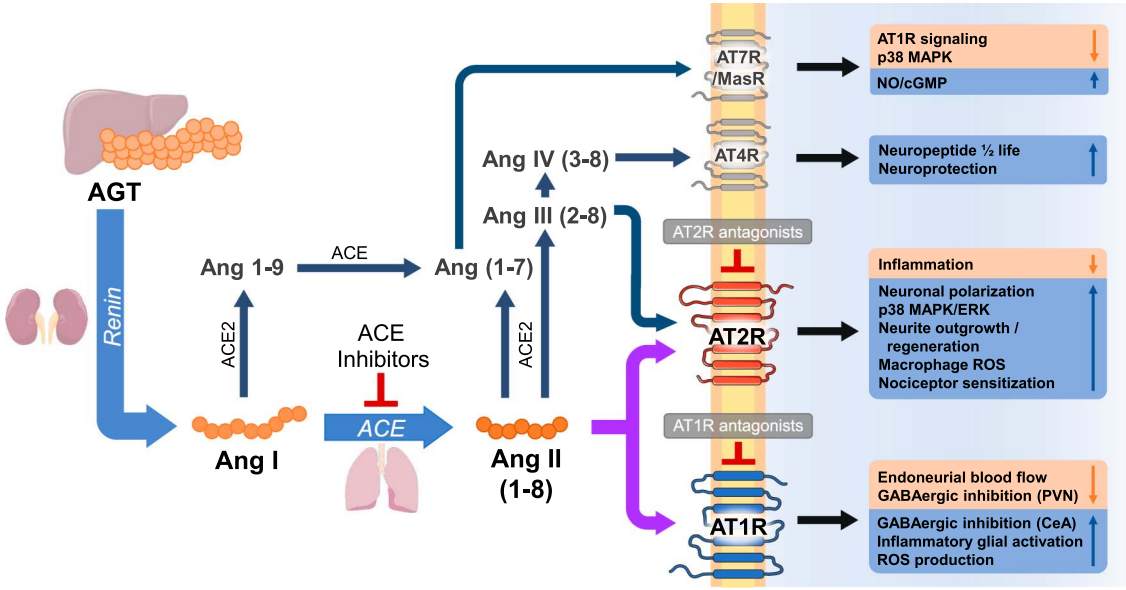

Figure 1. Schematic depicting the renin–angiotensin system with receptors, inhibitory drug molecules, and downstream processes relevant to NP. Historically, the sources of angiotensinogen, renin, and ACE were first identified as the liver, kidney, and lung epithelia, although more recent studies have identified expression of these components in many other cell types, including those in the CNS and peripheral nervous system. Physiological processes of relevance to NP described in this review are listed downstream of their respective receptors. AGT, angiotensinogen; ACE, angiotensin-converting enzyme; Ang, angiotensin; AT1R and AT2R, angiotensin II type 1 and type 2 receptors; AT4R, angiotensin IV-specific receptor; AT7R/Mas receptor, Ang (1-7) receptor; CeA, central nucleus of the amygdala; CNS, central nervous system; PVN, paraventricular nucleus of the hypothalamus.

7).[3,38] These endogenous products activate different receptors and play different physiological roles. Currently, there are 4 separate receptor types described: AT1R, AT2R, AT4R (receptor of Ang IV), and the AT7R/Mas receptor (receptor of Ang (1-7)) (**Fig. 1**). AT1R preferentially binds Ang II and Ang III. AT2R preferentially binds Ang II and Ang III with similarly high affinity and is a lower-affinity receptor for Ang IV and Ang (1-7).[18] Ang IV also binds to AT4R, also known as insulin-regulated membrane aminopeptidase/IRAP.[39] Ang IV inhibits IRAP, thereby extending the half-life of "analgesia-promoting" peptides that are otherwise cleaved by the receptor's enzymatic activity (eg, vasopressin, oxytocin, and somatostatin[48,57,84]). Ang (1-7) is formed from Ang I and Ang II by the activity of the ACE homolog angiotensin-converting enzyme-2 (ACE2), in an alternate RAS pathway to the ACE/Ang II pathway. Ang (1-7) binds to the Mas1 proto-oncogene GPCR, commonly referred to as the Mas receptor, MasR. This collection of ligands, receptors, and physiological functions has many important implications for NP because they operate in the central and peripheral nervous systems, as well as nonneuronal cells known to influence NP (eg, leukocytes). The modulators of these receptors reviewed in the present article are listed in **Table 1**.

2. Angiotensin receptors in the brain: relevance to neuropathic pain

2.1. Angiotensin signaling in the brain: angiotensin receptors and functions

AT1Rs at sites controlling autonomic and hormonal responses in the brain are the chief drivers of the classic Ang II-induced pressor and dipsogenic responses.[38,131,134] In addition to mediating the blood pressure and fluid balance functions of the RAS, Ang II plays a significant role in nervous system function and pain-related mechanisms.[4,116] The first study hinting at a central nervous system

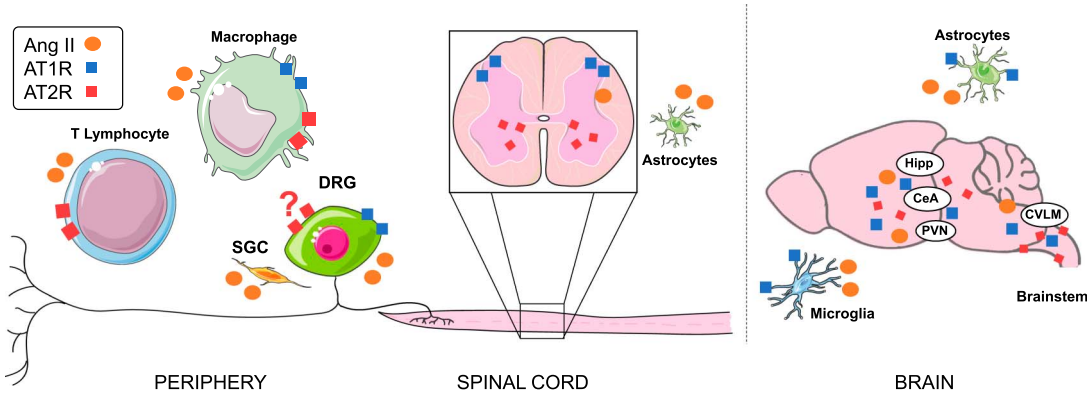

Figure 2. Overview of RAS expression in peripheral, central neuronal, and nonneuronal structures related to NP. Structures within the sagittal rodent brain of known relevance to RAS signaling are labeled accordingly. Expression patterns have been reported consistently in the literature, with the possible exception of AT2R in DRG neurons. CeA, central amygdala; CVLM, caudal ventrolateral medulla; DRG, dorsal root ganglion; Hipp, hippocampus; NP, neuropathic pain; RAS, renin–angiotensin system; PVN, paraventricular nucleus; SGC, satellite glial cells.

Table 1

List of drugs and compounds referenced in this review.

ACE inhibitors	AT1R antagonists	AT2R antagonists	Other compounds (*Function*)
Enalapril	Candesartan	PD123177	Compound 21 *(AT2R Agonist)*
Trandolapril	L-158809	PD123319/EMA200	A779 *(AT7R/MasR antagonist)*
Quinapril	Losartan	EMA300/400/401	Saralasin *(nonselective ATR antagonist)*
	Telmisartan		SB203580 *(p38 MAPK inhibitor)*
	ZD7155		L-NOarg *(nonselective NOS inhibitor)*
			L-NPA *(neuronal NOS inhibitor)*
			Glibenclamide *(K_{ATP} channel blocker)*

ACE, angiotensin-converting enzyme; MAPK, mitogen-activated protein kinase; NOS, NO synthase.

(CNS)-related function of the RAS showed a centrally mediated pressor response initiated by intracerebroventricular injection of Ang II.[16] Because Ang II does not penetrate the blood–brain barrier, the study was the first indication of a new, nontraditional role of a discrete nervous system RAS. This discovery was followed some years later by the description of a separate, brain-based expression of renin by Jacques Genest's group.[44] Although products of the "circulating RAS" produced in the periphery can access the circumventricular organs,[46,81] the majority of brain structures are likely only able to respond to RAS components produced within the brain itself. Subsequent studies demonstrated widespread expression of all necessary precursors and enzymes required to produce angiotensin. The majority of CNS angiotensinogen is produced by astrocytes, although neurons can also contribute.[51] In addition, the receptors AT1R and AT2R are expressed by neurons, astrocytes, oligodendrocytes, and microglia throughout the brain.[3,67,128,131] This "cerebral RAS" has subsequently been implicated in learning and memory, regulation of affective responses, and processing of sensory input.[131] These findings formed the basis for the eventual exploration of the RAS—in the brain and elsewhere—in the context of pain.

AT1a, AT1b, and AT2 receptor subtypes are found in areas associated with pain relay, modulation and perception, such as thalamic and hypothalamic nuclei, locus coeruleus, central amygdala (CeA), and solitary tract nucleus[28,29,102] (**Fig. 2**). Ang II inhibits activity in the CeA, a highly important structure for NP, as it receives input from the dorsal horn through the parabrachial nucleus.[58] Ang II inhibits CeA activity by activating postsynaptic AT1Rs and facilitating GABAergic synaptic input.[56] Impairments in GABAergic inhibition occur in the CNS in NP,[70] but the extent to which this could be targeted by facilitating Ang II-AT1R signaling remains to be established. The activation of AT1Rs in the CNS has also been associated with neuroinflammatory responses, such as M1-like polarization of microglia.[67] This induction of a proinflammatory glial phenotype and ROS production are well-established features of many NP states.[50]

2.2. Processes induced by the dysregulation of the cerebral renin–angiotensin system

The diverse functions of the cerebral RAS are still not fully understood. A more complete understanding of the glial and neuronal subtypes involved, the circuitry, and circumstances triggering such signaling events will be required to delineate the functions of its different receptors and neuropeptides in different pathological processes, including NP. Nevertheless, the cerebral RAS seems to play an important role in neuroprotection. Its dysregulation seems to be an important factor in different neurodegenerative diseases, which hints at potential involvement in NP. The role of the cerebral RAS specifically in neurodegenerative diseases has recently been reviewed elsewhere.[38,135]

Kandalam and Clark postulated that Ang II might participate in inflammatory and proliferative pathways in the brain through JAK/STAT signaling and IL-6 production, which are in turn associated with astrocyte proliferation and maintenance of NP.[63,127] In addition, NP conditions are associated with dysregulation of dopaminergic signaling.[133] To the best of our knowledge, a direct connection between dopaminergic disruption and the cerebral RAS has not been established in the context of NP. However, dopaminergic deficiency observed in the early stages of Parkinson disease has been associated with induction of neuroinflammation by activation of glial RAS.[33] This raises the possibility that similar neuroinflammation in NP is driven at least in part by glial RAS. In accordance with these results, a study from the Garrido-Gil laboratory suggested that the cerebral RAS modulates dopamine release, possibly through mutual regulation of dopamine and angiotensin receptors, with dysregulation of these interactions leading to overactivation of local RAS, neuroinflammation, oxidative stress, and neuronal death.[45] It is now clear that the cerebral RAS influences the complex processes of neuroinflammation, neuroregeneration, and neuroprotection. However, how these changes might be linked to the pain modulatory action of different RAS components is not fully understood. Nevertheless, there are a growing number of studies aimed at improving our understanding of the role of cerebral RAS in different pain processes.

2.3. Possible future therapeutic implications of the brain renin–angiotensin system in neuropathic pain

There is evidence to suggest the cerebral RAS can promote or inhibit pain responses, depending on the cells/structures targeted and the receptor subtypes involved. For example, local administration of Ang II in the caudal ventrolateral medulla caused hyperalgesia in rodent tail-flick and formalin tests. This hyperalgesia was significantly attenuated by local administration of losartan (AT1R antagonist; **Tables 1 and 2**).[79] These results suggest that Ang II may exert pronociceptive effects in components of the supraspinal pain control system. However, Parlo et al.[101] showed that Ang I, II, or III injected into rat periaqueductal gray induced dose-dependent antinociception in the tail-flick test. This effect of Ang II on the ventrolateral PAG was inhibited by prior local administration of saralasin, a nonselective angiotensin receptor antagonist. Ang II also activates spinally projecting paraventricular nucleus (PVN) neurons by AT1R-mediated attenuation of GABAergic (but not glutamatergic) synaptic inputs into the PVN.[71] This observation is consistent with the finding that electrical stimulation of the PVN increases pain threshold.[139] Because the activity of the ACE2-Ang (1-7)-AT7R/Mas receptor axis tends to oppose the activity of AT1R signaling,[41] augmenting MasR signaling may offer an additional approach to achieve similar analgesia.

In addition to AT1R, cerebral AT2R might also be an important regulator of pain threshold, as indicated by Sakagawa et al.[111] They found increased pain sensitivity in tail-flick and tail-pinch tests in AT2R-deficient animals, a finding that was associated with reduced expression of β-endorphin in the arcuate nucleus of the medial basal hypothalamus in AT2R knockout mice.

In contrast to signaling through AT1R, the activation of AT2Rs exerts mainly neuroprotective effects. Recently, Bhat et al. showed that AT2R activation elicits neuroprotection by inhibiting proinflammatory processes in microglia; AT2R-mediated activation of protein phosphatase 2A prevented Ang II-induced activation of protein kinase C, phosphorylation of p47phox, and proinflammatory activation of microglia. ROS production, proinflammatory microglial activation, and sickness behaviors were also inhibited by AT2R activation in a mouse model of neuroinflammation.[15] Ang IV—AT4R/IRAP signaling is also thought to be broadly neuroprotective. Ang IV increases neuronal glucose uptake through inhibition of GLUT4 vesicular trafficking and inhibits proteolysis of neuropeptides associated with analgesia (eg, vasopressin, oxytocin, and somatostatin[38,48,57,84]). The activation of AT4R might offer novel therapeutic opportunities for NP, although more studies are needed to explore these complex effects.

The activation of ACE2 might offer beneficial therapeutic effects by enhancing Ang (1-7) availability. Ang (1-7) mainly exerts its actions by the activation of AT7R/Mas receptors (and to a lesser extent AT2R). In terms of neuronal processes (and feasibly pain transduction), the ACE2-Ang (1-7)-Mas receptor axis seems to counteract the ACE-Ang II-AT1R receptor axis, thereby counteracting Ang II-induced nociceptive behaviors.[91,92] Ang (1-7) and its postulated functions in the cerebral RAS were recently reviewed,[113] but the possible therapeutic role of ACE2 activators in NP is a question for the future.[103]

How the highly complex system of the brain RAS influences the development of different pain types, and how these processes might be used therapeutically is far from fully understood. As the neuroprotective effects and inhibitory facilitation of some angiotensin receptor agonists came to light, research began to focus on potential therapeutic implications of the peripheral RAS and angiotensin receptors. Indeed, EMA401 (the first AT2R antagonist analgesic reaching clinical trials) was not found to penetrate the CNS, yet proved to be a potent analgesic for the treatment of NP[5,121] (see section 4).

3. Angiotensin receptors in spinal and peripheral neurons and relations to neuropathic pain

3.1. Angiotensin signaling at the spinal cord and dorsal root ganglion

Although the majority of the research on RAS in NP focuses on peripheral injury, there are findings that are likely to be relevant to injury due to CNS trauma, eg, spinal cord injury. Such trauma is characterized by neuroinflammation, leukocyte infiltration, and neuronal apoptosis.[19,42,114] The ability of Ang II to induce chemokine expression in myocardium and vascular smooth muscle[69,132] suggests that Ang II production might also underlie leukocyte infiltration and subsequent pathology after CNS injury. Indeed, Füchtbauer et al. found that Ang II signaling to astrocytes through AT1R regulates leukocyte infiltration into the CNS in response to axonal lesioning.[43] Blocking AT1R with candesartan significantly increased numbers of infiltrating macrophages. The infiltration of macrophages at sites of neuronal injury is seen across multiple models of neuropathy and shows a strong connection with activation of the RAS, as described in section 4. Wu et al. hypothesized that cerebral AT2Rs are neuroprotective and support neuronal survival in response to ischemia-induced neuronal injury through inhibiting AT1Rs.[136] A recent study by Abdul-Muneer addressed neuroinflammation and apoptosis in a cell culture model of neuronal stretch injury.[1] They showed that stretch injury increases Ang II, causing the release of proinflammatory cytokines and oxidative stress. The AT1R antagonist losartan attenuated the oxidative stress, reducing neuroinflammation and cell death. AT1R expression is upregulated after injury in this model, which was also counteracted by losartan. These interesting findings help further our understanding of the mechanisms by which AT1R antagonists exert anti-inflammatory and neuroprotective effects, but it should be further explored in vivo across different pain models. Notably, some studies suggest that the beneficial (anti-inflammatory and neuroprotective) effects of competitive AT1R antagonists might be due to facilitation of AT2R signaling. This issue is discussed further in section 3.2.

Numerous studies indicate that the spinal cord expresses RAS peptides and receptors described in the cerebral RAS in the previous section.[95] Ang II acts as a neuromodulator in spinal pain transmission, which is supported by the finding that spinal administration of Ang II elicits nociceptive behaviors in different pain models.[90,95] Ang II and AT2R were shown in peripheral neuronal tissues that are crucial in pain transmission: rat and human dorsal root ganglion (DRG) and in trigeminal ganglia. Transcripts for renin, angiotensinogen, and ACE are all expressed in rat DRG neurons and other cell types (eg, blood vessels and satellite glia).[20] In addition, Ang II colocalized with substance P and calcitonin gene-related peptide (CGRP)-containing DRG neurons, which further supports its role in nociception.[4,59,98,99] Angiotensinogen and ACE mRNA were detected in rat DRG by Patil et al. Furthermore, a combination of in situ hybridization and immunohistochemistry revealed colocalization of angiotensinogen mRNA and Ang II immunoreactivity in most rat DRG neurons. This indicates angiotensinogen can be produced intraneuronally in the DRG and that uptake from circulation may not be necessary for the local formation of Ang II.[98] Neuronal expression in DRG is in contrast to the brain, where angiotensinogen is predominantly expressed by astrocytes, neurons, and cerebral endothelial cells to a lesser extent.[143]

Mitogen-activated protein kinase (MAPK) phosphorylation seems to be a key player in Ang II-mediated transduction of pain signals.[86,90,92] Mitogen-activated protein kinases are activated by phosphorylation and play crucial roles in cell signaling and gene expression. Evidence shows that spinal p38 MAPK is activated after neuronal injury; consequently, it is a major contributor to the development and maintenance of NP.[60] Several Ang II signaling modulators influence pain transmission, at least in part, by counteracting Ang II-induced MAPK signaling. Another key component of Ang II-mediated signaling is p42/p44 MAPK (ERK1/2), which is essential for induction of AT2R-mediated neurite outgrowth.[53] It is conceivable that such Ang II-mediated neurite outgrowth/regeneration is beneficial when target reinnervation is achievable after nerve injury. However, futile regeneration is maintained if the nerve injury is too severe to be repaired.[137] Taken together, AT2R activation induces pleiotropic effects, both of which have potential clinical significance. p38 MAPK activation contributes to NP symptoms, whereas the ERK1/2 pathway seems to moderate the neurotrophic effects. Although the exact role of different Ang II receptors (mainly AT1R vs AT2R) in the development and possible management of NP is not yet clear, the following

sections describe studies that have evaluated the relationships between different angiotensin receptor subtypes and pain.

3.2. Angiotensin II type 1 receptor modulators and neuropathic pain

Considering that AT1R signaling is proinflammatory in other tissues and organ systems[82] and is expressed along the pain neuraxis, it was hypothesized that AT1R also contributes to pathological pain states (the reviewed AT1R modulators are listed in **Table 2**). For example, Nemoto et al. found that intrathecal losartan (an AT1R antagonist) produces an antinociceptive effect through the inhibition of p38 MAPK phosphorylation in the mouse formalin test.[93,94] Subsequent studies in NP models showed that systemic administration of losartan was analgesic in paclitaxel-induced NP, an effect achieved by inhibiting the inflammatory mediators p-NFκB, IL-1β, TNF-α, and CCL2/MCP-1 in rat DRG.[66] AT1Rs were found to be expressed in both neurons and satellite cells in the DRG. Affirming these results, Yuksel et al.[141] found that systemic delivery of telmisartan (also an AT1R antagonist) significantly improved nerve healing by inhibiting production of IL-1β and caspase-3 in nerve crush and transection NP models in rats. Based on these findings, the authors concluded that AT1R inhibitors could be used to improve peripheral nerve regeneration after injury.

Diabetic neuropathy represents a unique intersection of the "classic" hemodynamic effects of Ang II signaling and its more recently described role as an inflammatory mediator. Increased circulating Ang II in diabetes is known to contribute to renal injury, which can progress to chronic kidney disease and uremic neuropathy.[120] It is also well established that vascular dysfunction in diabetic neuropathy drives nerve injury by impairing endoneurial blood flow.[17,22,126] This leads to energetic dysfunction, inflammation, and cell death. In one study, streptozotocin-induced diabetic rats were treated for 12 weeks with enalapril, an ACE inhibitor, or L-158809, an AT1R antagonist.[22] Interestingly, treatment of diabetic rats with either compound reduced superoxide levels in the aorta, prevented the diabetes-induced impairment of vascular relaxation, and improved acetylcholine-mediated vasodilation. Similarly, Ogata et al. showed that neuropathic symptoms of streptozotocin-induced diabetic mice were alleviated by intrathecal administration of losartan, an AT1R antagonist, but not by PD123319, an AT2R antagonist.[95] It seems that expression of spinal ACE is increased under diabetic conditions. This leads to an increased level of Ang II, accompanied by the AT1R-dependent phosphorylation of p38 MAPK. Collectively, this supports the observations that ACE inhibitors and/or Ang II receptor blockers may be effective treatments for diabetes and the attendant vascular and neural dysfunction[75,107] (**Tables 2 and 3**). However, mechanistically disentangling direct effects of Ang II on neuronal excitability from effects secondary to cardiovascular/renal dysfunction may prove challenging.

Studies using inflammatory pain models suggest that AT1R antagonists may not be universally analgesic or may differ in their effectiveness between inflammatory and NP models. Costa et al.[24] showed that carrageenan-evoked inflammatory pain was augmented by the AT1R antagonist losartan, as well as the AT7R/Mas receptor antagonist A779. These antagonists showed no effect in chronic constriction injury (CCI). This serves as a clear indication that suitability of analgesics must be tested across different pain models and animal strains. Clearly, the type of pain-inciting insult highly influences, or even determines, the effectiveness of different therapeutics.

A recent study by the Snyder laboratory showed a reciprocal interaction of AT1R with the ion channel transient receptor potential vanilloid 4 (TRPV4) in choroid plexus epithelia.[142] The results indicated that TRPV4 activation and Ca^{2+} influx inhibits AT1R signaling. These results also hint at the possibility of AT1R-mediated inhibition of TRPV4. Such reciprocal regulation is likely conserved across other GPCR–TRP channel pairs, building upon an earlier hypothesis that Ang II receptors expand their functional diversity through dimerization with other GPCRs.[83] Because there is an apparent role of TRPV4 in NP,[49,104] the implications for NP should be further investigated.

3.3. Angiotensin II type 2 receptor modulators and neuropathic pain

Anand et al. showed that expression of AT2R in small-/medium-diameter human and rat DRG is partly colocalized with TRPV1 expression. A study by the Benitez laboratory also showed that AT2R is preferentially expressed in nonpeptidergic (IB4-binding) likely C-nociceptors and it is also present in peptidergic ($trkA^+$/SP^+) neurons that encompass both small C-nociceptors and medium-size A-neurons, which are crucial in pain transmission. In cultured DRG neurons, Ang II potentiated Ca^{2+} flux evoked by the

Table 2

Effects of AT1R antagonists.

Compound	Model	Outcome	Citation
Losartan	CVLM injection of Ang II (rat)	Reduced hyperalgesia (tail-flick, formalin tests)	79
	Formalin injection (mouse)	Antinociceptive (through p38 MAPK inhibition)	93
	In vitro neuronal stretch injury (rat)	Attenuated oxidative stress, neuroinflammation, cell death	1
	Ang II (i.t.; mouse)		90
	Streptozotocin-induced diabetic neuropathy (mouse)	Antinociceptive (delivered i.t.)	95
	Cultured human, rat DRG neurons	No effect on capsaicin responses	4
	Paclitaxel-induced neuropathy (rat)	Alleviated mechanical hyperalgesia	66
	Chronic constriction injury (rat)	No effect in Randall–Selitto test	24
	SNI (mouse)	No effect on von Frey sensitivity	117
	Sciatic nerve crush (rat)	No effect on nerve regeneration	106
	Astrocyte culture (rat)	Reduced Ang II-induced IL-6 secretion	63
Candesartan	Vincristine-induced neuropathy (mouse)	Restored normal tactile sensitivity	12
	LPS-induced neuroinflammation (rat)	Alleviated neuroinflammation	14
Telmisartan	SNI (rat; axonotmesis vs anastomosis)	Improved nerve regeneration	141
L-158809	Streptozotocin-induced diabetic neuropathy (rat)	Prevented vascular abnormalities	22

CVLM, caudal ventrolateral medulla; dorsal root ganglion; MAPK, mitogen-activated protein kinase; SNI, spared nerve injury.

Table 3

Effects of ACE inhibitors.

Compound	Model	Outcome	Citation
Perindopril	LPS-induced neuroinflammation (rat)	Alleviation of neuroinflammation	14
Trandolapril	Normotensive patients with type I or II diabetes and mild neuropathy	Improved conduction velocity, AP amplitude	75
Lisinopril	Diabetic patients with hypertension	Improved temperature and vibration perception thresholds	107
Enalapril	Streptozotocin-induced diabetic neuropathy (rat)	Prevented vascular abnormalities	22

ACE, angiotensin-converting enzyme.

TRPV1 agonist capsaicin, along with increasing cAMP levels and promotion of neurite outgrowth.[4] Treatment with EMA401, a selective AT2R antagonist, inhibited capsaicin-evoked responses (compounds acting on AT2R are listed in **Table 4**). Although human limb peripheral nerve segments proximal to injury showed reduced AT2R, expression levels were preserved in painful neuromas. This suggests maintained expression in regenerating nerve fibers. By contrast, AT1R immunoreactivity was absent from neurons, but strongly positive in vascular structures, human DRG, and in other tissues. This is in agreement with other studies, which have indicated that the expression pattern of AT2R may contribute to its viability as a target for the treatment of NP.[7] Accordingly, the AT1R antagonist losartan had no effect on DRG neuron responses to capsaicin. The possible therapeutic role of AT2R is also underlined by the finding that an inflammatory soup (histamine, ATP, PGE_2, bradykinin, serotonin, IL-6, TNFα, and substance P) was found to upregulate AT2R but not AT1R mRNA levels in cultured rat DRG neurons.[9] Indeed, Smith et al. showed that augmented Ang II/AT2R signaling in the DRG of CCI rats is attenuated by the AT2R antagonist EMA300,[123,124] with similar attenuation in a bone cancer pain model.[86] In CCI mice, the analgesic effect was abolished in AT2R knockouts and had an intermediate effect in the hemizygotes, affirming a role for AT2R. AT2R blockade by EMA300 resulted in the inhibition of p38 MAPK and p44/p42 MAPK activation in the rat DRG. These results also corroborated the colocalization of Ang II with the small-/medium-diameter nociceptive neuronal markers, substance P, and CGRP in rat and human tissues. Further supporting the possible future therapeutic use of AT2R antagonists, different compounds produced analgesic effects in several different animal models of NP, including the following: spared nerve injury-induced mechanical hypersensitivity in mice (PD123319/EMA200;[115]), a rat model of antiretroviral toxic polyneuropathy (EMA200, EMA300;[122]), and the CCI model in rats (EMA200, EMA300, and EMA400;[124]). Our recent findings also underline the promising effects of AT2R antagonists in NP.[116–118] Mechanical and cold pain hypersensitivity (assessed using reflexive and nonreflexive measures of pain sensitivity) were measured in spared nerve injury mice after systemic administration of PD123319/EMA200. Despite observing analgesic efficacy, AT2R expression was not detected in DRG neurons by RNA sequencing. AT2R-GFP reporter mice were also negative in DRG histology. These apparently conflicting data could indicate that the injury model, duration, and species used can influence AT2R expression, and that cell types other than DRG neurons may also be targets of AT2R antagonists. This is discussed further in section 4. Nonetheless, these results independently verified that the analgesic action of AT2R antagonism is conserved across the different subtypes of EMA compounds used.

Although blockade of AT2R signaling seems to be analgesic, it seems that AT2R agonists are neuroprotective and/or neurotrophic. More than 2 decades ago, Lucius et al.[73] found that AT2R promotes the axonal elongation of postnatal rat retinal explants and DRG neurons in vitro, and axonal regeneration of retinal ganglion cells after optic nerve crush. Cotreatment with the AT2R antagonist PD123177 (but not the AT1R antagonist losartan) abolished the Ang II-induced axonal regeneration. In a more recent study, Ang II was administered through osmotic minipump to injured sciatic nerve in rats. The authors found that Ang II treatment promoted functional recovery, which was fully inhibited by AT2R antagonist PD123319. Once again, the AT1R antagonist losartan had no effect.[106] This was the first study to present direct evidence for an involvement of the AT2R in peripheral nerve regeneration. There are now several publications

Table 4

Agents acting on AT2R.

Compound	Model	Outcome	Citation
Agonists			
Diminazene aceturate (DIZE)	Formalin test (mouse)	Attenuates the second phase of formalin-induced nociception	94
Compound 21	Vincristine-induced neuropathy (mouse)	Restored normal tactile sensitivity	12
CGP42112A	Optic nerve crush (rat)	Axonal regeneration	73
Antagonists			
PD123319	Astrocyte culture (rat)	Did not prevent Ang II-induced IL-6 secretion	63
	SNI (mouse)	Reduced pain behaviors after systemic delivery	117, 118
	Sciatic nerve crush (rat)	Prevented Ang II-induced recovery of sensorimotor function	106
EMA200, EMA300, EMA400	CCI (rat)	Dose-dependent pain relief	65, 123, 124
EMA200 & EMA300	Antiretroviral toxic polyneuropathy (rat)	Significant analgesia	122
EMA401	Human and rat DRG	Inhibited capsaicin responses, reduced neurite length/density	4
	Postherpetic neuralgia patients	Significantly reduced pain scores	109

CCI, chronic constriction injury; DRG, dorsal root ganglion; SNI, spared nerve injury.

highlighting the role of the RAS in neuronal development and protection.[26,31,68] For example, Chakrabarty et al.[20] showed that locally synthesized Ang II in rat DRG plays a role in estrogen-mediated sprouting. Estrogen-induced neurite outgrowth was inhibited by the ACE inhibitor enalapril. 17β-Estradiol was also found to upregulate AT2R expression, and the neurotrophic action of estradiol was counteracted by the AT2R antagonist PD123319. Based on the neurotrophic role of AT2Rs, it is hypothesized that AT2R activation might be able to improve NP by facilitating neuronal protection or regeneration. In a recent study, Bessaguet et al.[13] analyzed the effects of PD123319/EMA200 (AT2R antagonist) in resiniferatoxin-induced sensory small-fiber neuropathy in mice. They showed that AT1R blockade by the competitive antagonist candesartan prevented functional sensory neuropathy. This effect was not ascribed to AT1R inhibition per se, but to the resultant increase in Ang II binding to AT2R. This conclusion is based on the finding that AT2R blockade does not prevent resiniferatoxin-induced neuropathy, and the effect of candesartan could be blocked by PD123319/EMA200 in wild type but not AT2R-deficient mice. Therefore, the beneficial effects of candesartan could result from blunting of AT1R-mediated negative feedback on the release of renin, thereby increasing Ang II-AT2R signaling and enhancing neuroprotection. In a murine model of vincristine-induced mechanical allodynia, both candesartan and the AT2R-selective agonist Compound 21 completely restored normal tactile sensitivity. However, only Compound 21 displayed neuroprotective effects against vincristine-induced neuronal damage. This study provides further evidence of AT2R agonist-induced neuroprotection.[12] A study by Bhat et al.[14] similarly connected the beneficial effects of candesartan to facilitation of AT2R signaling. Candesartan treatment alleviated neuroinflammation in rats and in astroglial and microglial cells. Such effects were associated with elevation of AT2R expression and blocked by AT2R antagonists.

Marion et al.[78] described a novel mechanism involving AT2Rs, in which a mycolactone toxin, *Mycobacterium ulcerans,* exerts analgesia. Mycolactone-induced AT2R signaling elicited $G\alpha_i$ signaling, triggering a cascade culminating in PGE_2 synthesis and leading to the opening of TRAAK channels, hyperpolarization, and analgesia. These results should be explored further using other well-established AT2R agonists (eg, Compound 21) under NP or other pain conditions to better understand mechanism(s) underlying AT2R-induced analgesia and the ability of PGE_2 to participate in analgesia and hyperalgesia in different pathologies.[72] These findings are intriguing because several clinical and preclinical studies suggest the utility of AT2R antagonism for NP. It is possible that the analgesic effects of AT2R depend on the nature of the pain-inducing insult or the cell type(s) expressing AT2R. Perhaps AT2R antagonists inhibit a $G\alpha_s$ pathway in nociceptors, which converges with the nerve growth factor-TRPV1 pathway. By contrast, pathways activated by compounds such as mycolactone perhaps predominate in leukocytes and/or the CNS.[27] Of note, our study indicates the involvement of macrophage AT2Rs and neuronal TRPA1 channels in the analgesic actions of AT2R antagonists, as described in section 4.

In summary, AT2R agonists have been found to be neuroprotective because neuronal regenerative processes are commonly hampered under neuropathic conditions. However, there are numerous studies showing the effectiveness of AT2R antagonists, as they seem to alleviate NP symptoms. Further studies are needed to investigate these somewhat competing therapeutic possibilities, and the extent to which these neurotrophic and analgesic properties of AT2R (or AT1R) might be used clinically in NP. For example, careful optimization of dosing, delivery, and NP stratification may be required to offset nerve regeneration against analgesia. Based on our current knowledge, AT2R antagonists are among the most promising novel compounds for the future management of NP.[108]

3.4. Other angiotensin receptors, modulators, and neuropathic pain

Ang (1-7) is mainly produced by ACE2 from Ang II and binds to the G-protein-coupled AT7R/Mas receptor, reducing inflammation, fibrosis, and insulin resistance.[119] Ang (1-7) and AT7R/Mas activation seems to counteract the harmful effects of Ang II-mediated signaling (pronociception and proinflammation) by inhibition of MAPK activation[55] (Mas receptor modulators are listed in **Table 5**). In a recent study, Yamagata et al.[138] postulated that Ang (1-7) production is downregulated in leptin-deficient *ob/ob* diabetic mice, which is accompanied by a loss of ACE2-positive neurons. Intrathecal administration of Ang (1-7) in *ob/ob* mice attenuated hyperalgesia. This effect was counteracted by A779, a Mas receptor antagonist. SB203580, a p38 MAPK inhibitor, also attenuated hyperalgesia, and Ang (1-7) inhibited the phosphorylation of spinal p38 MAPK. This study again underlines the important role of RAS-mediated p38 MAPK phosphorylation in pain transmission and the beneficial counteracting role of the ACE2/Ang (1-7)/Mas receptor pathway. Ang (1-7) signaling has also been linked to the NO/cyclic GMP pathway. NO is capable of activating ATP-sensitive K^+ channels (K_{ATP}) through increased intracellular cGMP, which has been implicated as an antinociceptive mechanism for several different drugs.[36] Costa et al.[23] investigated this pathway in relation to Ang (1-7). They showed that Ang (1-7) induced NO release and demonstrated an antinociceptive effect in rats after hind paw injection of PGE_2. This antinociception was antagonized by the nonselective NO synthase inhibitor L-NOarg, the selective neuronal NO synthase inhibitor L-NPA, and the K_{ATP} channel blocker glibenclamide. Although these findings indicate that Ang (1-7) activates the NO/cyclic GMP/K_{ATP} channel pathway, a role for Ang (1-7) in the treatment of NP should be further evaluated.

Table 5

Agents acting on MasR.

Compound	Model	Outcome	Citation
Agonists			
Ang (1-7) (i.p.)	Metastatic bone pain (mouse)	Attenuated spontaneous, evoked pain	41
Ang (1-7) (i.t.)	Ang II (i.t.)-induced nociception (mouse)	Attenuated nociceptive behavior	91
	Obese diabetic (*ob/ob*) mouse		138
Antagonists			
A779	CCI (rat)	No effect	24

CCI, chronic constriction injury.

ACE2 is of tremendously growing interest after the discovery that it is the main cell entry receptor for the novel coronavirus SARS-CoV-2.[80] The spike protein of SARS-CoV-2 binds ACE2. Given the expression of ACE2 in the peripheral and CNS and several reports of neurological complications of COVID-19,[76,105,145] pain is a raising concern as a potential complication of this novel highly contagious infection.[2,6] The dysregulation of ACE2/Ang (1-7)/Mas receptor pathway by the virus-induced ACE2 depletion may lead to the development of lasting pain symptoms. This is a rapidly evolving area of research, which underscores the numerous gaps in our knowledge of neuronal RAS in health and disease.

4. Effect of the renin–angiotensin system on immune system function and implications for neuropathic pain

Ang II and AT2R have been shown to be expressed by sensory neurons and by nonneuronal cells. The density of angiotensin receptors is developmentally regulated. Although AT1R is expressed in most adult tissues including the heart, blood vessels, brain, and kidney, AT2R expression was (until recently) thought to be largely restricted to embryonic, fetal, and neonatal tissues.[100] Immune cells are now considered to be crucial in the development of pain, and the infiltration of various immune cells into peripheral nervous structures and their profound influence on pain development is a well-known phenomenon.[77] Circulating mononuclear cells, macrophages, and T-lymphocytes are known to express ACE.[25,37,89] Elevated ACE expression is associated with enhanced myeloid cell activity,[10,11] although the implications of this for NP have not been explored. Nevertheless, these findings shed light on the possible intriguing neuroimmune interaction in the complex mechanism of pain transmission and the development of different pain states. However, the circumstances that trigger immune system RAS contribution to NP, and/or the changes in leukocyte trafficking and inflammatory phenotype that may result from RAS signaling require further study.

A study by Nataraj et al.[88] was one of the first to describe the capability of Ang II to influence immune responses; the immunosuppressive effects of calcineurin inhibitors were potentiated by RAS antagonism. Migrating leukocytes are crucial in lesion development, in which Ang II is an essential mediator. In addition, macrophages express all components of the RAS. These cells infiltrate local tissues after neuronal injury, and further promote the development of NP symptoms by fostering neuronal damage. A study by Okamura et al.[96] showed that monocytes/macrophages upregulate RAS expression during differentiation, a phenotypic change that underlies the contribution of macrophages to development of atherosclerosis. Moreover, an in vitro study by Guo et al.[54] showed that Ang II acts directly on macrophages; Ang II contributes to the production and release of proinflammatory cytokines TNF-α and IL-1β in macrophages through AT1R activation.

Several studies show that the RAS is highly involved in immunological responses under pathological conditions (eg, during inflammation). Nabah et al.[87] showed how Ang II induces neutrophil accumulation. In this study, Ang II provoked rapid neutrophil recruitment, mediated through the release of CXC chemokines such as CINC/KC and MIP-2 in rats and IL-8 in humans. This may contribute to neutrophil infiltration in acute myocardial infarction. In a study by Moreno et al., Ang II infusion resulted in the infiltration of inflammatory cells and stimulated cell proliferation. Interestingly, these pathogenic events were attenuated by atorvastatin, which can inhibit Ang II-induced activation of NF-κB and chemokine expression.[85] Although Guo et al. did not investigate NP as a readout, their results highlight the pathophysiological role of macrophage AT1Rs in inflammatory processes. Treating macrophages with Ang II upregulated AT1R expression and increased the production of inflammatory mediators (TNF-α, IL-1β, IL-6, and IL-10) and reactive oxygen species. These effects were blocked by ZD7155, an AT1R antagonist. These results indicate a possible mechanism for the analgesic actions of different AT1R antagonists (see section 3.3) and should be further investigated in relation to NP and other different pain diseases. AT1R antagonists are associated with reduced perineural invasion in head and neck cancer,[35] although it is not yet clear if this is due to an AT1R-mediated effect on chemokine expression. Collectively, these findings are likely to be relevant for NP because leukocyte infiltration and the consequent proinflammatory actions are known to be significant contributors to development and maintenance of NP.[97,129,140]

Numerous studies show that the RAS is involved in (auto) immune responses and modulation of T-cell and macrophage function, with Ang II typically thought to drive proinflammatory macrophage responses.[55] Khan et al.[65] investigated the effects of the AT2R antagonist EMA300 on CCI rats. Consistent with their earlier studies,[123] they observed a CCI-induced increase in Ang II expression in DRG. This increase was underpinned at least in part by infiltration of Ang II/AT2R-expressing $CD3^+$ T cells. This increase in Ang II levels was reversed at the time of peak analgesia by administration of a single intraperitoneal dose of EMA300. At the point of peak analgesia, there was also a significant reduction in Ang II/AT2R signaling and its downstream mediators, p38 MAPK and p44/p42 MAPK. These observations are consistent with the hypothesis that cells infiltrating sites of inflammation/tissue injury establish a local RAS. Subsequent AT2R activation then elicits nociceptor hypersensitivity and sprouting. This results in heightened thermal and mechanical sensitivity, as shown in a recent study of CFA-induced inflammation in rats.[21] This study showed that the Ang II-synthesizing proteins renin and angiotensinogen were largely absent from the hind paw after saline injection, but abundant in T cells and macrophages in CFA-injected paws. These findings correlate with an earlier study by Thomas and Hoffman, who discovered decades ago that macrophages express specific receptors for Ang II and related peptides that were thought to be responsible for uptake of Ang II by macrophages.[125]

Our laboratory has previously investigated a possible role of the RAS in neuroimmune interactions in NP conditions. We found that local AT2R activation by Ang II in peripheral macrophages leads to chronic pain hypersensitivity associated with nerve injury/neuropathy.[116–118] AT2R antagonism provided effective analgesia in neuropathic but not inflammatory pain, although any qualitative difference between acute and more chronic dosing regimens was not explored. In addition, Ang II was elevated in the injured sciatic nerves of mice, and an AT2R antagonist dose-dependently attenuated mechanical hypersensitivity. Our findings suggest that macrophages infiltrating sites of injury induce persistent neuropathic mechanical and cold pain hypersensitivity through the activation of AT2Rs. It is worth noting that macrophages at the site of injury seem to be major contributors to pain hypersensitivity in this model, but macrophage infiltration/proliferation elsewhere along the pain neuraxis (eg, in the DRG) is an area of active study and likely also contributes.[140] Activation of macrophage AT2R induces intercellular crosstalk between peripheral macrophages and sensory neurons, mediated by AT2R-to-TRPA1 redox signaling.[116] Ang II was again shown to induce macrophage infiltration, and AT2R activation macrophages triggered ROS/RNS production, which can

transactivate TRPA1 channels on sensory neurons to induce NP, as has been previously described.[97] The lack of detectable expression of AT2R in mouse and human DRG tissue in these studies indicates that the relative contribution of macrophage-centric mechanisms vs those mechanisms involving other leukocytes and/or direct modulation of DRG neurons (as described in section 3.3) requires further study. Nevertheless, selective AT2R antagonists such as EMA401 and PD123319 have now attenuated NP in humans and in multiple pain model studies conducted by independent research groups. Our study directly identifies immune cell-to-sensory neuron signaling crosstalk, which underlies peripheral nociceptor sensitization. These findings collectively describe a prominent neuroimmune interaction in the development of pain and represent a newly discovered mechanism underlying the analgesic action of AT1R and AT2R antagonists. Ultimately, the emerging ability of RAS modulators to simultaneously target neuronal physiology and pathophysiology, leukocyte infiltration, and inflammation make them a uniquely promising class of analgesic drug candidates.

5. Human studies showing the involvement of the renin–angiotensin system in the development and possible management of pain

Although the majority of data on the role of RAS in pain development originate from preclinical rodent studies, there are a few human studies supporting those findings. As described previously, Ang II and AT2R were shown both in rat and human DRG and in trigeminal ganglia, key points in pain transmission (see section 3.1.).[20] Colocalization of Ang II with the small-/medium-diameter nociceptive neuronal markers, substance P, and CGRP was also demonstrated in human tissues. There are numerous studies involving human tissues suggesting the mechanistic role of Ang II receptors in pain development, which strongly validates the results of the preclinical studies discussed above. One of the early examples of human clinical studies is a study by Drummond, which showed that Ang II administered to healthy volunteers by iontophoresis increased thermal sensitivity, presumably through vasoconstriction and ischemia.[34] In the late 1990s, Malik et al. investigated the beneficial effects of the ACE inhibitor trandolapril in patients with diabetic NP.[74,75] In this double-blind, placebo-controlled trial, trandolapril improved neuronal function, assessed by peroneal nerve conduction velocity. These beneficial effects were observed even in normotensive diabetic patients. However, neuropathy symptom and deficit scores showed no improvement. In addition, the sample size was relatively small (41 patients with diabetic NP), and the Ang II receptor signaling underlying this effect was not addressed. In a recent study by Didangelos et al.,[32] another ACE inhibitor (quinapril) showed beneficial effects in patients with diabetic cardiovascular autonomic neuropathy by improving parasympathetic dysfunction. Similarly, symptoms of NP were not affected in patients with peripheral diabetic NP in this study. Nevertheless, these studies underline the involvement of RAS in NP and hint at possible future applications.

Juhi Kalra et al. studied the modulation of pain perception by ramipril and losartan, an ACE inhibitor and AT1R blocker, respectively.[62] The pain perception threshold of 30 healthy individuals was assessed after a single dose of placebo or test compound using a sphygmomanometer-based pressure pain test. Ramipril lowered the measured pain thresholds, and both compounds lowered the maximal tolerated pain 4 hours after administration. The mechanism by which these drugs might influence pain perception would benefit from further study in a larger number of individuals. Notably, a study by Costa et al. resonates with these observations because they showed the ability of losartan to augment acute pain (section 3.2). There is additional evidence that hemodynamics can influence pain sensitivity, which could partially explain the observed effects of different RAS inhibitors in humans. Some studies have shown the association of hypertension with hypoalgesia in dental pain.[47,52] The authors attributed the observed reduction in pain threshold of hypertensives receiving ARBs to AT1R signaling. It is possible that the inhibited degradation of substance P and/or bradykinin underlie these contradictory effects, which have been hypothesized to contribute to complex regional pain syndrome.[30] Ultimately, the systemic/hemodynamic effects and the local/proinflammatory effects may interact and underlie the inconsistencies, but further work is needed to fully exploit targeting of Ang II signaling for pain relief. Nevertheless, acute pain perception in healthy or not neuropathic individuals highly differs from the complex changes observed in NP conditions.

Probably the most investigated and recognized novel RAS modulator is the AT2R antagonist EMA401. This high-affinity receptor antagonist proved to be effective in a phase 2 clinical trial for the treatment of postherpetic neuralgia.[109] Unfortunately, EMA401 was withdrawn from further studies due to adverse events, although its analgesic efficacy was clear.[64] A retrospective study by Roldan et al. conducted on patients with chemotherapy-induced peripheral neuropathy also supports the hypothesis that blockade of Ang II receptor signaling would be beneficial in NP.[110] They suggest that ACE inhibitors and Ang II receptor blockers may have a neuroprotective effect on the myelinated Aβ and Aδ fibers in cancer patients receiving neurotoxic chemotherapy. Although promising, the limitations of such retrospective studies (along with limited sample size) should be acknowledged.

There are a few findings in humans that also support the existence of neuroimmune interactions during pain development, in which the RAS seems to be a major contributor (as described in section 4.). Ang II infusion was found to increase expression of the adhesion molecule ICAM-1 (CD54; intercellular adhesion molecule-1) in human endothelial cells and soluble ICAM-1 release in human volunteers, an effect that was inhibited by losartan. These findings are intriguing in terms of neuroimmune interactions because ICAM-1 is a crucial mediator of leukocyte adhesion to the vascular endothelium and subsequent tissue infiltration. There is also evidence that human monocytes and macrophages express Ang II, AT1R, and AT2R. A study by Nabah showed the role of Ang II in neutrophil recruitment during myocardial infarction, hence the neuroimmunological concept of pain development should be further investigated in humans.[87] Taken together, there are very few clinical studies investigating the possible therapeutic role of the RAS and Ang II receptors in pain management, but they support their possible future therapeutic role. More studies with AT2R blockers likely will be conducted because they seem to be the most promising group for NP alleviation.

6. Summary and conclusions

Renin–angiotensin system signaling has expanded far beyond its initial description as an endocrine controller of hemodynamics. Additional enzymes, receptors, peptide fragments, and sites of expression discovered in the past several decades led to the discovery of RAS signaling within the nervous system. It is quickly becoming clear that there are pathophysiological ramifications of angiotensin signaling at all levels of the pain neuraxis, whether in the brain, spinal cord, DRG, or leukocytes infiltrating sites of neurological damage. There remains a lack of consensus

regarding the circumstances under which RAS inhibition would be most beneficial for NP; this may depend to a large extent on the desired outcome (neurotrophic vs analgesic). Clearly, Ang II signaling is a powerful modulator of inflammatory-immunological responses involved in different painful pathological conditions that influence central and peripheral neuronal functions, including NP. A more detailed understanding of the role of RAS in neuroprotection (eg, AT2R initiated neuronal protection) or subsequent activation of different pain pathways (eg, the pain-sensitizing effect of AT2R activation through TRPA1) is likely to offer more opportunities for therapeutic modulation. The current literature supports targeting the RAS to treat NP as a worthwhile endeavor because a preexisting suite of medications indicated for hypertension can be used to guide future therapeutic development. To fully understand the potential role of AT1Rs, AT2Rs, and other components of the RAS for the future management of different pain states, more studies evaluating their impact on multiple clinically relevant pain modalities should be explored.

Disclosures

The authors have no conflicts of interest to declare.

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References

[1] Abdul-Muneer PM, Bhowmick S, Briski N. Angiotensin II causes neuronal damage in stretch-injured neurons: protective effects of losartan, an angiotensin T1 receptor blocker. Mol Neurobiol 2018;55:5901–12.
[2] Aksan F, Nelson EA, Swedish KA. A COVID-19 patient with intense burning pain. J Neurovirol 2020;26:1–2.
[3] Allen AM, O'Callaghan EL, Mendelsohn FAO, Chai SY. Neuronal angiotensin. In: LR Squire, editors. Encyclopedia of neuroscience. Oxford: Academic Press, 2009. pp. 697–702.
[4] Anand U, Facer P, Yiangou Y, Sinisi M, Fox M, McCarthy T, Bountra C, Korchev YE, Anand P. Angiotensin II type 2 receptor (AT2 R) localization and antagonist-mediated inhibition of capsaicin responses and neurite outgrowth in human and rat sensory neurons. Eur J Pain 2013;17:1012–26.
[5] Anand U, Yiangou Y, Sinisi M, Fox M, MacQuillan A, Quick T, Korchev YE, Bountra C, McCarthy T, Anand P. Mechanisms underlying clinical efficacy of Angiotensin II type 2 receptor (AT2R) antagonist EMA401 in neuropathic pain: clinical tissue and in vitro studies. Mol Pain 2015;11:38.
[6] Assersen KB, Sumners C, Steckelings UM. The renin-angiotensin system in hypertension, a constantly renewing classic: focus on the angiotensin AT2-receptor. Can J Cardiol 2020;36:683–93.
[7] Bali A, Singh N, Jaggi AS. Renin-angiotensin system in pain: existing in a double life? J Renin Angiotensin Aldosterone Syst 2014;15:329–40.
[8] Basso N, Terragno Norberto A. History about the discovery of the renin-angiotensin system. Hypertension 2001;38:1246–9.
[9] Benitez S, Seltzer A, Messina D, Foscolo M, Patterson S, Acosta C. Cutaneous inflammation differently regulates the expression and function of Angiotensin II types 1 and 2 receptors in rat primary sensory neurons. J Neurochem 2020;152:675–96.
[10] Bernstein KE, Gonzalez-Villalobos RA, Giani JF, Shah K, Bernstein E, Janjulia T, Koronyo Y, Shi PD, Koronyo-Hamaoui M, Fuchs S, Shen XZ. Angiotensin-converting enzyme overexpression in myelocytes enhances the immune response. Biol Chem 2014;395:1173–8.
[11] Bernstein KE, Khan Z, Giani JF, Cao DY, Bernstein EA, Shen XZ. Angiotensin-converting enzyme in innate and adaptive immunity. Nat Rev Nephrol 2018;14:325–36.
[12] Bessaguet F, Danigo A, Bouchenaki H, Duchesne M, Magy L, Richard L, Sturtz F, Desmouliere A, Demiot C. Neuroprotective effect of angiotensin II type 2 receptor stimulation in vincristine-induced mechanical allodynia. PAIN 2018;159:2538–46.
[13] Bessaguet F, Danigo A, Magy L, Sturtz F, Desmoulière A, Demiot C. Candesartan prevents resiniferatoxin-induced sensory small-fiber neuropathy in mice by promoting angiotensin II-mediated AT2 receptor stimulation. Neuropharmacology 2017;126:142–50.
[14] Bhat SA, Goel R, Shukla R, Hanif K. Angiotensin receptor blockade modulates NFκB and STAT3 signaling and inhibits glial activation and neuroinflammation better than angiotensin-converting enzyme inhibition. Mol Neurobiol 2016;53:6950–67.
[15] Bhat SA, Sood A, Shukla R, Hanif K. AT2R activation prevents microglia pro-inflammatory activation in a NOX-dependent manner: inhibition of PKC activation and p47(phox) phosphorylation by PP2A. Mol Neurobiol 2019;56:3005–23.
[16] Bickerton RK, Buckley JP. Evidence for a central mechanism in angiotensin induced hypertension. Proc Soc Exp Biol Med 1961;106:834–6.
[17] Biessels GJ, Bril V, Calcutt NA, Cameron NE, Cotter MA, Dobrowsky R, Feldman EL, Fernyhough P, Jakobsen J, Malik RA, Mizisin AP, Oates PJ, Obrosova IG, Pop-Busui R, Russell JW, Sima AA, Stevens MJ, Schmidt RE, Tesfaye S, Veves A, Vinik AI, Wright DE, Yagihashi S, Yorek MA, Ziegler D, Zochodne DW. Phenotyping animal models of diabetic neuropathy: a consensus statement of the diabetic neuropathy study group of the EASD (Neurodiab). J Peripher Nerv Syst 2014;19:77–87.
[18] Bosnyak S, Jones ES, Christopoulos A, Aguilar MI, Thomas WG, Widdop RE. Relative affinity of angiotensin peptides and novel ligands at AT1 and AT2 receptors. Clin Sci 2011;121:297–303.
[19] Carpenter RS, Jiang RR, Brennan FH, Hall JCE, Gottipati MK, Niewiesk S, Popovich PG. Human immune cells infiltrate the spinal cord and impair recovery after spinal cord injury in humanized mice. Sci Rep 2019;9:19105.
[20] Chakrabarty A, Blacklock A, Svojanovsky S, Smith PG. Estrogen elicits dorsal root ganglion axon sprouting via a renin-angiotensin system. Endocrinology 2008;149:3452–60.
[21] Chakrabarty A, Liao Z, Smith PG. Angiotensin II receptor type 2 activation is required for cutaneous sensory hyperinnervation and hypersensitivity in a rat hind paw model of inflammatory pain. J Pain 2013;14:1053–65.
[22] Coppey LJ, Davidson EP, Rinehart TW, Gellett JS, Oltman CL, Lund DD, Yorek MA. ACE inhibitor or angiotensin II receptor antagonist attenuates diabetic neuropathy in streptozotocin-induced diabetic rats. Diabetes 2006;55:341–8.
[23] Costa A, Galdino G, Romero T, Silva G, Cortes S, Santos R, Duarte I. Ang-(1–7) activates the NO/cGMP and ATP-sensitive K+ channels pathway to induce peripheral antinociception in rats. Nitric Oxide 2014;37:11–16.
[24] Costa ACO, Romero TRL, Pacheco DF, Perez AC, Savernini A, Santos RRA, Duarte IDG. Participation of AT1 and Mas receptors in the modulation of inflammatory pain. Peptides 2014;61:17–22.
[25] Costerousse O, Allegrini J, Lopez M, Alhenc-Gelas F. Angiotensin I-converting enzyme in human circulating mononuclear cells: genetic polymorphism of expression in T-lymphocytes. Biochem J 1993;290(pt 1):33–40.
[26] Cote F, Do TH, Laflamme L, Gallo JM, Gallo-Payet N. Activation of the AT(2) receptor of angiotensin II induces neurite outgrowth and cell migration in microexplant cultures of the cerebellum. J Biol Chem 1999;274:31686–92.
[27] Danser AHJ, Anand P. The angiotensin II type 2 receptor for pain control. Cell 2014;157:1504–6.
[28] de Kloet AD, Steckelings UM, Sumners C. Protective angiotensin type 2 receptors in the brain and hypertension. Curr Hypertens Rep 2017;19:46.
[29] de Kloet AD, Wang L, Ludin JA, Smith JA, Pioquinto DJ, Hiller H, Steckelings UM, Scheuer DA, Sumners C, Krause EG. Reporter mouse strain provides a novel look at angiotensin type-2 receptor distribution in the central nervous system. Brain Struct Funct 2016;221:891–912.
[30] de Mos M, Huygen FJ, Stricker BH, Dieleman JP, Sturkenboom MC. The association between ACE inhibitors and the complex regional pain syndrome: suggestions for a neuro-inflammatory pathogenesis of CRPS. PAIN 2009;142:218–24.

[31] Diaz-Ruiz C, Villar-Cheda B, Dominguez-Meijide A, Garrido-Gil P, Guerra MJ, Labandeira-Garcia JL. Aging-related overactivity of the angiotensin/AT1 Axis decreases sirtuin 3 levels in the substantia nigra, which induces vulnerability to oxidative stress and neurodegeneration. J Gerontol Ser A Biol Sci Med Sci 2020;75:416–24.
[32] Didangelos T, Tziomalos K, Margaritidis C, Kontoninas Z, Stergiou I, Tsotoulidis S, Karlafti E, Mourouglakis A, Hatzitolios AI. Efficacy of administration of an angiotensin converting enzyme inhibitor for two years on autonomic and peripheral neuropathy in patients with diabetes mellitus. J Diabetes Res 2017;2017:6719239.
[33] Dominguez-Meijide A, Rodriguez-Perez AI, Diaz-Ruiz C, Guerra MJ, Labandeira-Garcia JL. Dopamine modulates astroglial and microglial activity via glial renin-angiotensin system in cultures. Brain Behav Immun 2017;62:277–90.
[34] Drummond PD. The effect of noradrenaline, angiotensin II and vasopressin on blood flow and sensitivity to heat in capsaicin-treated skin. Clin Auton Res 1998;8:87–93.
[35] Du KN, Shepherd AJ, Ma IV, Roldan CJ, Amit M, Feng L, Desai S, Cata JP. Lack of association between angiotensin-converting enzyme inhibitors and angiotensin receptor blockers and pain improvement in patients with oral cancer. ecancer 2020;14:1121.
[36] Duarte ID, dos Santos IR, Lorenzetti BB, Ferreira SH. Analgesia by direct antagonism of nociceptor sensitization involves the arginine-nitric oxide-cGMP pathway. Eur J Pharmacol 1992;217:225–7.
[37] Eklund A, Blaschke E, Danielsson B. Subcellular localization of angiotensin-converting enzyme in the human alveolar macrophage. Scand J Clin Lab Invest 1987;47:47–54.
[38] Farag E, Sessler DI, Ebrahim Z, Kurz A, Morgan J, Ahuja S, Maheshwari K, John Doyle D. The renin angiotensin system and the brain: new developments. J Clin Neurosci 2017;46:1–8.
[39] Fernando RN, Larm J, Albiston AL, Chai SY. Distribution and cellular localization of insulin-regulated aminopeptidase in the rat central nervous system. J Comp Neurol 2005;487:372–90.
[40] Forrester SJ, Booz GW, Sigmund CD, Coffman TM, Kawai T, Rizzo V, Scalia R, Eguchi S. Angiotensin II signal transduction: an update on mechanisms of physiology and pathophysiology. Physiol Rev 2018;98: 1627–738.
[41] Forte BL, Slosky LM, Zhang H, Arnold MR, Staatz WD, Hay M, Largent-Milnes TM, Vanderah TW. Angiotensin-(1-7)/Mas receptor as an antinociceptive agent in cancer-induced bone pain. PAIN 2016;157: 2709–21.
[42] Fu H, Li F, Thomas S, Yang Z. Hyperbaric oxygenation alleviates chronic constriction injury (CCI)-induced neuropathic pain and inhibits GABAergic neuron apoptosis in the spinal cord. Scand J Pain 2017; 17:330–8.
[43] Füchtbauer L, Groth-Rasmussen M, Holm TH, Løbner M, Toft-Hansen H, Khorooshi R, Owens T. Angiotensin II Type 1 receptor (AT1) signaling in astrocytes regulates synaptic degeneration-induced leukocyte entry to the central nervous system. Brain Behav Immun 2011;25:897–904.
[44] Ganten D, Marquez-Julio A, Granger P, Hayduk K, Karsunky KP, Boucher R, Genest J. Renin in dog brain. Am J Physiol 1971;221: 1733–7.
[45] Garrido-Gil P, Rodriguez-Perez AI, Fernandez-Rodriguez P, Lanciego JL, Labandeira-Garcia JL. Expression of angiotensinogen and receptors for angiotensin and prorenin in the rat and monkey striatal neurons and glial cells. Brain Struct Funct 2017;222:2559–71.
[46] Gebke E, Müller AR, Jurzak M, Gerstberger R. Angiotensin II-induced calcium signalling in neurons and astrocytes of rat circumventricular organs. Neuroscience 1998;85:509–20.
[47] Ghione S, Rosa C, Mezzasalma L, Panattoni E. Arterial hypertension is associated with hypalgesia in humans. Hypertension 1988;12:491–7.
[48] Goodin BR, Ness TJ, Robbins MT. Oxytocin—a multifunctional analgesic for chronic deep tissue pain. Curr Pharm Des 2015;21: 906–13.
[49] Grace MS, Bonvini SJ, Belvisi MG, McIntyre P. Modulation of the TRPV4 ion channel as a therapeutic target for disease. Pharmacol Ther 2017; 177:9–22.
[50] Grace PM, Gaudet AD, Staikopoulos V, Maier SF, Hutchinson MR, Salvemini D, Watkins LR. Nitroxidative signaling mechanisms in pathological pain. Trends Neurosci 2016;39:862–79.
[51] Grobe JL, Xu D, Sigmund CD. An intracellular renin-angiotensin system in neurons: fact, hypothesis, or fantasy. Physiology 2008;23:187–93.
[52] Guasti L, Zanotta D, Diolisi A, Garganico D, Simoni C, Gaudio G, Grandi AM, Venco A. Changes in pain perception during treatment with angiotensin converting enzyme-inhibitors and angiotensin II type 1 receptor blockade. J Hypertens 2002;20:485–91.
[53] Guimond MO, Roberge C, Gallo-Payet N. Fyn is involved in angiotensin II type 2 receptor-induced neurite outgrowth, but not in p42/p44mapk in NG108-15 cells. Mol Cell Neurosci 2010;45:201–12.
[54] Guo F, Chen XL, Wang F, Liang X, Sun YX, Wang YJ. Role of angiotensin II type 1 receptor in angiotensin II-induced cytokine production in macrophages. J interferon Cytokine Res 2011;31:351–61.
[55] Hammer A, Stegbauer J, Linker RA. Macrophages in neuroinflammation: role of the renin-angiotensin-system. Pflugers Archiv 2017;469:431–44.
[56] Hu B, Qiao H, Cao T, Sun B, Luo X, Jia R, Fan Y, Wang N, Lu Y, Yan J. Angiotensin II facilitates GABAergic neurotransmission at postsynaptic sites in rat amygdala neurons. Neuropharmacology 2018;133:334–44.
[57] Huang J, Polgár E, Solinski HJ, Mishra SK, Tseng P-Y, Iwagaki N, Boyle KA, Dickie AC, Kriegbaum MC, Wildner H, Zeilhofer HU, Watanabe M, Riddell JS, Todd AJ, Hoon MA. Circuit dissection of the role of somatostatin in itch and pain. Nat Neurosci 2018;21:707–16.
[58] Ikeda R, Takahashi Y, Inoue K, Kato F. NMDA receptor-independent synaptic plasticity in the central amygdala in the rat model of neuropathic pain. PAIN 2007;127:161–72.
[59] Imboden H, Patil J, Nussberger J, Nicoud F, Hess B, Ahmed N, Schaffner T, Wellner M, Müller D, Inagami T, Senbonmatsu T, Pavel J, Saavedra JM. Endogenous angiotensinergic system in neurons of rat and human trigeminal ganglia. Regul Pept 2009;154:23–31.
[60] Ji RR, Suter MR. p38 MAPK, microglial signaling, and neuropathic pain. Mol Pain 2007;3:33.
[61] Kakar SS, Sellers JC, Devor DC, Musgrove LC, Neill JD. Angiotensin II type-1 receptor subtype cDNAs: differential tissue expression and hormonal regulation. Biochem Biophys Res Commun 1992;183: 1090–6.
[62] Kalra J, Chaturvedi A, Kalra S, Chaturvedi H, Dhasmana DC. Modulation of pain perception by Ramipril and Losartan in human volunteers. Indian J Physiol Pharmacol 2008;52:91–6.
[63] Kandalam U, Clark MA. Angiotensin II activates JAK2/STAT3 pathway and induces interleukin-6 production in cultured rat brainstem astrocytes. Regul peptides 2010;159:110–16.
[64] Keppel Hesselink JM, Schatman ME. EMA401: an old antagonist of the AT2R for a new indication in neuropathic pain. J Pain Res 2017;10: 439–43.
[65] Khan N, Muralidharan A, Smith MT. Attenuation of the infiltration of angiotensin II expressing CD3(+) T-cells and the modulation of nerve growth factor in lumbar dorsal root ganglia - a possible mechanism underpinning analgesia produced by EMA300, an angiotensin II type 2 (AT2) receptor antagonist. Front Mol Neurosci 2017;10:389.
[66] Kim E, Hwang SH, Kim HK, Abdi S, Kim HK. Losartan, an angiotensin II type 1 receptor antagonist, alleviates mechanical hyperalgesia in a rat model of chemotherapy-induced neuropathic pain by inhibiting inflammatory cytokines in the dorsal root ganglia. Mol Neurobiol 2019; 56:7408–19.
[67] Labandeira-Garcia JL, Rodríguez-Perez AI, Garrido-Gil P, Rodriguez-Pallares J, Lanciego JL, Guerra MJ. Brain renin-angiotensin system and microglial polarization: implications for aging and neurodegeneration. Front Aging Neurosci 2017;9:129.
[68] Laflamme L, Gasparo M, Gallo JM, Payet MD, Gallo-Payet N. Angiotensin II induction of neurite outgrowth by AT2 receptors in NG108-15 cells. Effect counteracted by the AT1 receptors. J Biol Chem 1996;271:22729–35.
[69] Li C, He J, Zhong X, Gan H, Xia Y. CX3CL1/CX3CR1 Axis contributes to angiotensin II-induced vascular smooth muscle cell proliferation and inflammatory cytokine production. Inflammation 2018;41:824–34.
[70] Li C, Lei Y, Tian Y, Xu S, Shen X, Wu H, Bao S, Wang F. The etiological contribution of GABAergic plasticity to the pathogenesis of neuropathic pain. Mol Pain 2019;15:1744806919847366.
[71] Li DP, Chen SR, Pan HL. Angiotensin II stimulates spinally projecting paraventricular neurons through presynaptic disinhibition. J Neurosci 2003;23:5041.
[72] Lin C-R, Amaya F, Barrett L, Wang H, Takada J, Samad TA, Woolf CJ. Prostaglandin E2 receptor EP4 contributes to inflammatory pain hypersensitivity. J Pharmacol Exp Ther 2006;319:1096–103.
[73] Lucius R, Gallinat S, Rosenstiel P, Herdegen T, Sievers J, Unger T. The angiotensin II type 2 (AT2) receptor promotes axonal regeneration in the optic nerve of adult rats. J Exp Med 1998;188:661–70.
[74] Malik RA. Can diabetic neuropathy be prevented by angiotensin-converting enzyme inhibitors? Ann Med 2000;32:1–5.
[75] Malik RA, Williamson S, Abbott C, Carrington AL, Iqbal J, Schady W, Boulton AJ. Effect of angiotensin-converting-enzyme (ACE) inhibitor trandolapril on human diabetic neuropathy: randomised double-blind controlled trial. Lancet 1998;352:1978–81.

[76] Mao L, Jin H, Wang M, Hu Y, Chen S, He Q, Chang J, Hong C, Zhou Y, Wang D, Miao X, Li Y, Hu B. Neurologic manifestations of hospitalized patients with coronavirus disease 2019 in Wuhan, China. JAMA Neurol 2020;77:683–90.
[77] Marchand F, Perretti M, McMahon SB. Role of the immune system in chronic pain. Nat Rev Neurosci 2005;6:521–32.
[78] Marion E, Song OR, Christophe T, Babonneau J, Fenistein D, Eyer J, Letournel F, Henrion D, Clere N, Paille V, Guérineau Nathalie C, Saint André J-P, Gersbach P, Altmann KH, Stinear Timothy P, Comoglio Y, Sandoz G, Preisser L, Delneste Y, Yeramian E, Marsollier L, Brodin P. Mycobacterial toxin induces analgesia in buruli ulcer by targeting the angiotensin pathways. Cell 2014;157:1565–76.
[79] Marques-Lopes J, Pinto M, Pinho D, Morato M, Patinha D, Albino-Teixeira A, Tavares I. Microinjection of angiotensin II in the caudal ventrolateral medulla induces hyperalgesia. Neuroscience 2009;158: 1301–10.
[80] Marquez A, Wysocki J, Pandit J, Batlle D. An update on ACE2 amplification and its therapeutic potential. Acta Physiol 2020:e13513.
[81] McKinley MJ, Allen AM, Burns P, Colvill LM, Oldfield BJ. Interaction of circulating hormones with the brain: the roles of the subfornical organ and the organum vasculosum of the lamina terminalis. Clin Exp Pharmacol Physiol Suppl 1998;25:S61–67.
[82] Mehta PK, Griendling KK. Angiotensin II cell signaling: physiological and pathological effects in the cardiovascular system. Am J Physiol Cell Physiol 2007;292:C82–97.
[83] Mogi M, Iwai M, Horiuchi M. Emerging concepts of regulation of angiotensin II receptors. Arterioscl Thromb Vasc Biol 2007;27:2532–9.
[84] Mogil JS, Sorge RE, LaCroix-Fralish ML, Smith SB, Fortin A, Sotocinal SG, Ritchie J, Austin J-S, Schorscher-Petcu A, Melmed K, Czerminski J, Bittong RA, Mokris JB, Neubert JK, Campbell CM, Edwards RR, Campbell JN, Crawley JN, Lariviere WR, Wallace MR, Sternberg WF, Balaban CD, Belfer I, Fillingim RB. Pain sensitivity and vasopressin analgesia are mediated by a gene-sex-environment interaction. Nat Neurosci 2011;14:1569–73.
[85] Moreno M, Ramalho LN, Sancho-Bru P, Ruiz-Ortega M, Ramalho F, Abraldes JG, Colmenero J, Dominguez M, Egido J, Arroyo V, Gines P, Bataller R. Atorvastatin attenuates angiotensin II-induced inflammatory actions in the liver. Am J Physiol Gastrointest Liver Physiol 2009;296: G147–156.
[86] Muralidharan A, Wyse BD, Smith MT. Analgesic efficacy and mode of action of a selective small molecule angiotensin II type 2 receptor antagonist in a rat model of prostate cancer-induced bone pain. Pain Med 2014;15:93–110.
[87] Nabah YN, Mateo T, Estelles R, Mata M, Zagorski J, Sarau H, Cortijo J, Morcillo EJ, Jose PJ, Sanz MJ. Angiotensin II induces neutrophil accumulation in vivo through generation and release of CXC chemokines. Circulation 2004;110:3581–6.
[88] Nataraj C, Oliverio MI, Mannon RB, Mannon PJ, Audoly LP, Amuchastegui CS, Ruiz P, Smithies O, Coffman TM. Angiotensin II regulates cellular immune responses through a calcineurin-dependent pathway. J Clin Invest 1999;104:1693–701.
[89] Nehme A, Cerutti C, Dhaouadi N, Gustin MP, Courand P-Y, Zibara K, Bricca G. Atlas of tissue renin-angiotensin-aldosterone system in human: a transcriptomic meta-analysis. Sci Rep 2015;5:10035.
[90] Nemoto W, Nakagawasai O, Yaoita F, Kanno S, Yomogida S, Ishikawa M, Tadano T, Tan-No K. Angiotensin II produces nociceptive behavior through spinal AT1 receptor-mediated p38 mitogen-activated protein kinase activation in mice. Mol Pain 2013;9:38.
[91] Nemoto W, Ogata Y, Nakagawasai O, Yaoita F, Tadano T, Tan-No K. Angiotensin (1-7) prevents angiotensin II-induced nociceptive behaviour via inhibition of p38 MAPK phosphorylation mediated through spinal Mas receptors in mice. Eur J Pain 2014;18:1471–9.
[92] Nemoto W, Ogata Y, Nakagawasai O, Yaoita F, Tadano T, Tan-No K. Involvement of p38 MAPK activation mediated through AT1 receptors on spinal astrocytes and neurons in angiotensin II- and III-induced nociceptive behavior in mice. Neuropharmacology 2015;99:221–31.
[93] Nemoto W, Ogata Y, Nakagawasai O, Yaoita F, Tanado T, Tan-No K. The intrathecal administration of losartan, an AT1 receptor antagonist, produces an antinociceptive effect through the inhibiton of p38 MAPK phosphorylation in the mouse formalin test. Neurosci Lett 2015;585: 17–22.
[94] Nemoto W, Yamagata R, Nakagawasai O, Nakagawa K, Hung WY, Fujita M, Tadano T, Tan-No K. Effect of spinal angiotensin-converting enzyme 2 activation on the formalin-induced nociceptive response in mice. Eur J Pharmacol 2020;872:172950.
[95] Ogata Y, Nemoto W, Nakagawasai O, Yamagata R, Tadano T, Tan-No K. Involvement of spinal angiotensin II system in streptozotocin-induced diabetic neuropathic pain in mice. Mol Pharmacol 2016;90:205.
[96] Okamura A, Rakugi H, Ohishi M, Yanagitani Y, Takiuchi S, Moriguchi K, Fennessy PA, Higaki J, Ogihara T. Upregulation of renin-angiotensin system during differentiation of monocytes to macrophages. J Hypertens 1999;17:537–45.
[97] Old EA, Nadkarni S, Grist J, Gentry C, Bevan S, Kim KW, Mogg AJ, Perretti M, Malcangio M. Monocytes expressing CX3CR1 orchestrate the development of vincristine-induced pain. J Clin Invest 2014;124: 2023–36.
[98] Patil J, Schwab A, Nussberger J, Schaffner T, Saavedra JM, Imboden H. Intraneuronal angiotensinergic system in rat and human dorsal root ganglia. Regul Pept 2010;162:90–8.
[99] Paul M, Poyan Mehr A, Kreutz R. Physiology of local renin-angiotensin systems. Physiol Rev 2006;86:747–803.
[100] Porrello ER, Delbridge LM, Thomas WG. The angiotensin II type 2 (AT2) receptor: an enigmatic seven transmembrane receptor. Front Biosci 2009;14:958–72.
[101] Prado WA, Pelegrini-da-Silva A, Martins AR. Microinjection of renin-angiotensin system peptides in discrete sites within the rat periaqueductal gray matter elicits antinociception. Brain Res 2003; 972:207–15.
[102] Premer C, Lamondin C, Mitzey A, Speth RC, Brownfield MS. Immunohistochemical localization of AT1a, AT1b, and AT2 angiotensin II receptor subtypes in the rat adrenal, pituitary, and brain with a perspective commentary. Int J Hypertens 2013;2013:175428.
[103] Qaradakhi T, Gadanec LK, McSweeney KR, Tacey A, Apostolopoulos V, Levinger I, Rimarova K, Egom EE, Rodrigo L, Kruzliak P, Kubatka P, Zulli A. The potential actions of angiotensin-converting enzyme II (ACE2) activator diminazene aceturate (DIZE) in various diseases. Clin Exp Pharmacol Physiol 2020;47:751–8.
[104] Qu YJ, Zhang X, Fan ZZ, Huai J, Teng YB, Zhang Y, Yue SW. Effect of TRPV4-p38 MAPK pathway on neuropathic pain in rats with chronic compression of the dorsal root ganglion. BioMed Res Int 2016;2016: 6978923.
[105] Ray PR, Wangzhou A, Ghneim N, Yousuf MS, Paige C, Tavares-Ferreira D, Mwirigi JM, Shiers S, Sankaranarayanan I, McFarland AJ, Neerukonda SV, Davidson S, Dussor G, Burton MD, Price TJ. A pharmacological interactome between COVID-19 patient samples and human sensory neurons reveals potential drivers of neurogenic pulmonary dysfunction. Brain Behav Immun 2020;89:559–68.
[106] Reinecke K, Lucius R, Reinecke A, Rickert U, Herdegen T, Unger T. Angiotensin II accelerates functional recovery in the rat sciatic nerve in vivo: role of the AT2 receptor and the transcription factor NF-kappaB. FASEB J 2003;17:2094–6.
[107] Reja A, Tesfaye S, Harris ND, Ward JD. Is ACE inhibition with lisinopril helpful in diabetic neuropathy? Diabetic Med 1995;12:307–9.
[108] Rice AS, Smith MT. Angiotensin II type 2-receptor: new clinically validated target in the treatment of neuropathic pain. Clin Pharmacol Ther 2015;97:128–30.
[109] Rice ASC, Dworkin RH, McCarthy TD, Anand P, Bountra C, McCloud PI, Hill J, Cutter G, Kitson G, Desem N, Raff M. EMA401, an orally administered highly selective angiotensin II type 2 receptor antagonist, as a novel treatment for postherpetic neuralgia: a randomised, double-blind, placebo-controlled phase 2 clinical trial. Lancet 2014;383: 1637–47.
[110] Roldan CJ, Song J, Engle MP, Dougherty PM. Angiotensin-converting enzyme inhibitors and angiotensin receptor blockers modulate the function of myelinated fibers after chemotherapy: a quantitative sensory testing study. Pain Phys 2017;20:281–92.
[111] Sakagawa T, Okuyama S, Kawashima N, Hozumi S, Nakagawasai O, Tadano T, Kisara K, Ichiki T, Inagami T. Pain threshold, learning and formation of brain edema in mice lacking the angiotensin II type 2 receptor. Life Sci 2000;67:2577–85.
[112] Sandberg K, Ji H, Clark AJ, Shapira H, Catt KJ. Cloning and expression of a novel angiotensin II receptor subtype. J Biol Chem 1992;267: 9455–8.
[113] Santos RAS, Sampaio WO, Alzamora AC, Motta-Santos D, Alenina N, Bader M, Campagnole-Santos MJ. The ACE2/angiotensin-(1-7)/MAS Axis of the renin-angiotensin system: focus on angiotensin-(1-7). Physiol Rev 2018;98:505–53.
[114] Sekiguchi M, Sekiguchi Y, Konno S, Kobayashi H, Homma Y, Kikuchi S. Comparison of neuropathic pain and neuronal apoptosis following nerve root or spinal nerve compression. Eur Spine J 2009;18:1978–85.
[115] Sheahan TD, Siuda ER, Bruchas MR, Shepherd AJ, Mohapatra DP, Gereau RW, Golden JP. Inflammation and nerve injury minimally affect mouse voluntary behaviors proposed as indicators of pain. Neurobiol Pain 2017;2:1–12.
[116] Shepherd AJ, Copits BA, Mickle AD, Karlsson P, Kadunganattil S, Haroutounian S, Tadinada SM, de Kloet AD, Valtcheva MV, McIlvried

LA, Sheahan TD, Jain S, Ray PR, Usachev YM, Dussor G, Krause EG, Price TJ, Gereau RW, Mohapatra DP. Angiotensin II triggers peripheral macrophage-to-sensory neuron redox crosstalk to elicit pain. J Neurosci 2018;38:7032–57.
[117] Shepherd AJ, Mickle AD, Golden JP, Mack MR, Halabi CM, de Kloet AD, Samineni VK, Kim BS, Krause EG, Gereau RW, Mohapatra DP. Macrophage angiotensin II type 2 receptor triggers neuropathic pain. Proc Natl Acad Sci U S A 2018;115:E8057–66.
[118] Shepherd AJ, Mohapatra DP. Attenuation of unevoked mechanical and cold pain hypersensitivities associated with experimental neuropathy in mice by angiotensin II type-2 receptor antagonism. Anesth Analg 2019; 128:e84–7.
[119] Simões e Silva AC, Silveira KD, Ferreira AJ, Teixeira MM. ACE2, angiotensin-(1-7) and Mas receptor axis in inflammation and fibrosis. Br J Pharmacol 2013;169:477–92.
[120] Siragy HM, Carey RM. Role of the intrarenal renin-angiotensin-aldosterone system in chronic kidney disease. Am J Nephrol 2010;31: 541–50.
[121] Smith MT, Anand P, Rice AS. Selective small molecule angiotensin II type 2 receptor antagonists for neuropathic pain: preclinical and clinical studies. PAIN 2016;157(suppl 1):S33–41.
[122] Smith MT, Lau T, Wallace VC, Wyse BD, Rice AS. Analgesic efficacy of small-molecule angiotensin II type 2 receptor antagonists in a rat model of antiretroviral toxic polyneuropathy. Behav Pharmacol 2014;25:137–46.
[123] Smith MT, Woodruff TM, Wyse BD, Muralidharan A, Walther T. A small molecule angiotensin II type 2 receptor (AT(2)R) antagonist produces analgesia in a rat model of neuropathic pain by inhibition of p38 mitogen-activated protein kinase (MAPK) and p44/p42 MAPK activation in the dorsal root ganglia. Pain Med 2013;14:1557–68.
[124] Smith MT, Wyse BD, Edwards SR. Small molecule angiotensin II type 2 receptor (AT(2)R) antagonists as novel analgesics for neuropathic pain: comparative pharmacokinetics, radioligand binding, and efficacy in rats. Pain Med 2013;14:692–705.
[125] Thomas DW, Hoffman MD. Identification of macrophage receptors for angiotensin: a potential role in antigen uptake for T lymphocyte responses? J Immunol 1984;132:2807–12.
[126] Thrainsdottir S, Malik RA, Dahlin LB, Wiksell P, Eriksson KF, Rosén I, Petersson J, Greene DA, Sundkvist G. Endoneurial capillary abnormalities presage deterioration of glucose tolerance and accompany peripheral neuropathy in man. Diabetes 2003;52:2615.
[127] Tsuda M, Kohro Y, Yano T, Tsujikawa T, Kitano J, Tozaki-Saitoh H, Koyanagi S, Ohdo S, Ji RR, Salter MW, Inoue K. JAK-STAT3 pathway regulates spinal astrocyte proliferation and neuropathic pain maintenance in rats. Brain 2011;134(pt 4):1127–39.
[128] Tsutsumi K, Viswanathan M, Strömberg C, Saavedra JM. Type-1 and type-2 angiotensin II receptors in fetal rat brain. Eur J Pharmacol 1991; 198:89–92.
[129] Uceyler N, Braunsdorf S, Kunze E, Riediger N, Scheytt S, Divisova S, Bekircan-Kurt CE, Toyka KV, Sommer C. Cellular infiltrates in skin and sural nerve of patients with polyneuropathies. Muscle Nerve 2017;55: 884–93.
[130] van Hecke O, Austin SK, Khan RA, Smith BH, Torrance N. Neuropathic pain in the general population: a systematic review of epidemiological studies. PAIN 2014;155:654–62.
[131] von Bohlen und, Halbach O, Albrecht D. The CNS renin-angiotensin system. Cell Tissue Res 2006;326:599–616.
[132] Wang L, Zhang YL, Lin QY, Liu Y, Guan XM, Ma XL, Cao HJ, Liu Y, Bai J, Xia YL, Du J, Li HH. CXCL1-CXCR2 axis mediates angiotensin II-induced cardiac hypertrophy and remodelling through regulation of monocyte infiltration. Eur Heart J 2018;39:1818–31.
[133] Wawrzczak-Bargieła A, Ziółkowska B, Piotrowska A, Starnowska-Sokół J, Rojewska E, Mika J, Przewłocka B, Przewłocki R. Neuropathic pain dysregulates gene expression of the forebrain opioid and dopamine systems. Neurotoxicity Res 2020;37:800–14.
[134] Wright JW, Harding JW. Brain angiotensin receptor subtypes in the control of physiological and behavioral responses. Neurosci Biobehav Rev 1994;18:21–53.
[135] Wright JW, Kawas LH, Harding JW. A role for the brain RAS in alzheimer's and Parkinson's diseases. Front Endocrinol 2013;4:158.
[136] Wu CY, Zha H, Xia QQ, Yuan Y, Liang XY, Li JH, Guo ZY, Li JJ. Expression of angiotensin II and its receptors in activated microglia in experimentally induced cerebral ischemia in the adult rats. Mol Cell Biochem 2013;382:47–58.
[137] Xie W, Strong JA, Zhang J-M. Active nerve regeneration with failed target reinnervation drives persistent neuropathic pain. eNeuro 2017;4: ENEURO.0008-0017.2017.
[138] Yamagata R, Nemoto W, Nakagawasai O, Takahashi K, Tan-No K. Downregulation of spinal angiotensin converting enzyme 2 is involved in neuropathic pain associated with type 2 diabetes mellitus in mice. Biochem Pharmacol 2020;174:113825.
[139] Yang J, Chen JM, Yang Y, Liu WY, Song CY, Lin BC. Investigating the role of hypothalamic paraventricular nucleus in nociception of the rat. Int J Neurosci 2008;118:473–85.
[140] Yu X, Liu H, Hamel KA, Morvan MG, Yu S, Leff J, Guan Z, Braz JM, Basbaum AI. Dorsal root ganglion macrophages contribute to both the initiation and persistence of neuropathic pain. Nat Commun 2020;11: 264.
[141] Yuksel TN, Halici Z, Demir R, Cakir M, Calikoglu C, Ozdemir G, Unal D. Investigation of the effect of telmisartan on experimentally induced peripheral nerve injury in rats. Int J Neurosci 2015;125:464–73.
[142] Zaccor NW, Sumner CJ, Snyder SH. The nonselective cation channel TRPV4 inhibits angiotensin II receptors. J Biol Chem 2020;295: 9986–97.
[143] Zeisel A, Hochgerner H, Lonnerberg P, Johnsson A, Memic F, van der Zwan J, Haring M, Braun E, Borm LE, La Manno G, Codeluppi S, Furlan A, Lee K, Skene N, Harris KD, Hjerling-Leffler J, Arenas E, Ernfors P, Marklund U, Linnarsson S. Molecular architecture of the mouse nervous system. Cell 2018;174:999–1014.e1022.
[144] Zeng L, Alongkronrusmee D, van Rijn RM. An integrated perspective on diabetic, alcoholic, and drug-induced neuropathy, etiology, and treatment in the US. J Pain Res 2017;10:219–28.
[145] Zhou Z, Kang H, Li S, Zhao X. Understanding the neurotropic characteristics of SARS-CoV-2: from neurological manifestations of COVID-19 to potential neurotropic mechanisms. J Neurol 2020;267: 1–6.