THE RENIN-ANGIOTENSIN SYSTEM AND ITS INVOLVEMENT IN PAIN CONTROL. A NEW INSIGHT INTO AN OLD SYSTEM

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Abstract

Pain is a symptom usually associated with the development of diseases, but it is always a personal experience influenced by contextual, psychological, and social factors. The processing and interpretation of pain include a complicated brain network consisting variety of subordinated structures. At each level of this neural network, numerous cellular processes contribute to the modulation of pain sensitivity.

It has been found that the renin-angiotensin system (RAS) plays a significant role in controlling pain under normal and pathological conditions. Recently, new components of the RAS were discovered characterizing two interacting and balancing arms. The classical arm includes the angiotensin-converting enzyme (ACE), the octapeptide angiotensin II (Ang-II), its primary receptor AT1, and the less common AT2 receptor. The alternative arm includes ACE2, the heptapeptide angiotensin 1-7, and its primary receptor Mas-1. RAS has long been known as a homeostasis system primarily responsible for maintaining the physiological balance between the cardiovascular and renal systems. Accumulating data however enlarged scientific knowledge on the role of RAS in control of brain functions and modulation of pain susceptibility. Scientific evidence indicates a differential involvement of angiotensin receptors in the modulation of pain transmission and suppression. Some data seems contradictory, but a thorough analysis emphasized a specific role of receptor distribution and their selective activation/inhibition on the final effect of pain sensitivity. This review summarizes the available literature on the topic and characterizes some perspectives for further research.

Keywords: *Renin-angiotensin system, Nociception, ACE2, Ang 1-7*

Acknowledgment:

This work was financially supported by the Bulgarian National Scientific Fund project КП-06-Н71/9 of the Ministry of Education and Science, Bulgaria.

Introduction – pain definition and function

Pain is the most common clinical symptom associated with the development of many diseases, in which a part of the somatosensory system is activated. Still, it is always a personal experience influenced by contextual, psychological, and social factors [1,2]. According to the recently revised definition by the International Association for the Study of Pain, pain is accepted as: "An unpleasant sensory and emotional experience associated with, or resembling that associated with actual or potential tissue damage" [3]. In addition, pain can be described in terms of three components - sensory, emotional, and cognitive - reflected in painful stimuli's transmission and modulation mechanisms [4]. The sensory process that conveys noxious signals to the central nervous system (CNS) and triggers pain is known as nociception. Pain usually serves an adaptive role, but may adversely affect social, and psychological well-being. Depending on the duration of the stimulus and plastic changes in the information processing system, acute pain can differ from chronic pain. Acute pain has a protective function detecting harmful stimuli and preventing body damage. Chronic pain, however, persists long after the nociceptive stimulus and activates neuroplasticity processes, leading to the experience of exacerbated responses to both painful (hyperalgesia) and non-painful stimuli (allodynia) [5]. Chronic pain is considered a disease in

itself, which often is accompanied by emotional and cognitive impairments, leading to the development of anxiety and depression.

Mechanisms of nociception

Pain perception begins with the stimulation of specific receptors, nociceptors, the first-order sensory neurons classified as either unmyelinated C- or lightly myelinated A-delta fibers situated throughout the body: the skin, joints, viscera, and muscles [6,7,8,9]. High-threshold mechanical and heat stimuli, and a vast variety of chemical substances such as acetylcholine, adenosine triphosphate (ATP), substance P, potassium, serotonin, lactic acid, arachidonic acid, histamine, nerve growth factor, calcitonin gene-related peptide (CGRP), etc., can activate nociceptors [10,11]. Three transient receptor potential (TRP) cation channels, TRPV1, TRPM3, and TRPA1, were identified as chemical- and heat-induced pain sensors in nociceptor neurons. TRPV1 which can integrate diverse noxious stimuli, is expressed in both peptidergic and nonpeptidergic nociceptive fibers [12]. The complexity and multimodality of nociceptors require fine control to ensure the correct interpretation and prevent over-strengthening of the non-adaptable sensory reaction. The encoded nociceptive information is conveyed in the cell bodies in the dorsal root ganglion (DRG), and Rexed layer I and substantia gelatinosa of Rexed layer II of the dorsal horn of the spinal cord if the stimulus comes from the body, or the trigeminal ganglia (TG), if the stimulus is from the face. Nociceptive neurons make connections directly, or indirectly, through spinal interneurons, with second-order sensory neurons in the spinal cord which are activated through the released neurotransmitters, including glutamate, substance P (SP), CGRP, and others [6,7,8]. Interneurons, which can be referred to as "local circuit neurons", can be divided into two main classes: excitatory (glutamatergic) and inhibitory (GABA and/or glycineergic) [13].

Pain transmission

The spinal cord's nociceptive neurons form projections comprising the three major ascending pain pathways: the neospinothalamic, paleospinothalamic, and archispinothalamic tracts.

The neospinothalamic tract begins with second-order nociceptive neurons located in Rexed layer I. They decussate and ascend via the lateral spinothalamic tract to third-order neurons in the thalamus but with collaterals to autonomic homeostatic brainstem noradrenergic cell groups A1–A2 and A5–A7, the parabrachial nucleus (PB), and the periaqueductal gray (PAG). From the ventral posterolateral nucleus (VPL) and the ventral posterior inferior nucleus (VPI) of the thalamus, nociceptive information projects to the somatotopically organized primary somatosensory cortex (S1) responsible for the immediate awareness and the exact location of the painful stimulus. Nociceptive information from the TG projects to the ventral posteromedial nucleus (VPM), parafascicular nucleus (PF), and the centromedian nucleus (CM) then the information further projects to SI [4].

The paleospinothalamic tract starts from Rexed layer II, in which fibres have diffuse projections to Rexed layers IV through VIII. They ascent anteriorly and project bilaterally into the midbrain reticular formation (RF), PAG, tectum, PF, and CM. The neurons in the two latter thalamic structures send projections to the somatosensory cortex, brainstem nuclei, and limbic areas (cingulate gyrus, insulate cortex). The interplay between the limbic structures, hypothalamus, and brainstem nuclei mediates emotional and visceral responses to pain.

The archispinothalamic tract starts with second-order neurons found in Rexed layer II (substantia gelatinosa), which project to neurons in Rexed layers IV and VII. Diffuse projections from the latter 2 layers are sent to the RF and the PAG, then to the hypothalamus, limbic system nuclei, PF, and CM nucleus. This tract also mediates visceral and emotional reactions to pain [4].

Pain modulations, processing, and interpretations

There are many different classifications of pain based on various criteria such as etiology, intensity, duration, and pathophysiological mechanisms. Based on the latter, there are three types of pain: nociceptive, neuropathic, and nociplastic [14,15,16]. Pain accumulates the subjective expression of sensory/discriminative and motivational/affective experiences. Its interpretation may result from the interplay between aversive and rewarding processes that elicit specific motivated behavioral responses. For example, acute pain leads to an adaptive escape/avoidance reflex, and relief of acute pain is rewarding in facilitating learning to anticipate danger.

Under normal pain transmission, GABAergic neurons within the PAG have tonic activity, however, upon opioidergic activation, GABA release is inhibited which results in the disinhibition of rostral ventromedial medulla (RVM) off-cells and on-cells [17]. The endogenous opioidergic system is one of the well-known mechanisms supporting this fine-tuning. Still, the role of other modulatory systems including some neurohypophyseal hormones has recently been revealed [18]. Endogenous opioids bind to four major types of G protein-coupled receptors widely distributed in the pain modulatory network, which includes the ventrolateral PAG, RVM, and dorsal horn of the spinal cord [18].

Serotoninergic neurons in the nucleus raphe magnus (NRM) have descending pathways in the dorsal horn of the spinal cord exerting direct or indirect inhibitory control on pain transmission and participating in peripheral and central sensitization processes. [19].

The mesolimbic dopamine system in the ventral tegmental area (VTA) and its projections to the medial prefrontal cortex (mPFC) and nucleus accumbens (NAc) have recently focused attention as a hub for both the control of motivation, reward, and aversion and the development of comorbid chronic pain depression [20]. The VTA receives input from several brain regions, including those involved in nociceptive information processing (e.g., the parabrachial nucleus, PAG, central amygdala, and lateral habenula) [21]. At each level of this neural network, numerous cellular processes contribute to the modulation of pain sensitivity.

Renin-angiotensin system

The Renin-Angiotensin-Aldosterone System (RAAS) is a cascade including three significant components: renin, angiotensin II (Ang II), and aldosterone which plays a major role in the regulation of blood pressure, fluid balance, and electrolyte homeostasis [22,23,24]. The discovery of new peptides in the last few decades has increased our understanding and the complexity of the RAS [25]. These peptides form a counter-regulatory pathway of the RAS, which opposes the actions of the classic arm [26]. RAS is now well recognized as a dual vasoactive system, acting as a circulating endocrine system and a local tissue paracrine system. Contemporary studies continue to expand our knowledge of newer peptides in the RAS and the cross-talk between the two major pathways and their receptors [27].

Classic RAS Pathway

Prorenin is an inactive precursor of renin and it is produced in multiple tissues (a part of local RAS), including juxtaglomerular cells in the kidney, adrenal gland, placenta, uterus, retina, testes, and submandibular glands [28]. While prorenin is constitutively secreted in its inactive form, juxtaglomerular cells can cause prorenin's proteolytic (by enzymes proconvertase 1 and cathepsin B) or non-proteolytic activation to renin. [29,30,31]. Mature renin is then stored and released into the circulation for several physiological reasons: changes in renal perfusion, low plasma sodium concentration, increased activity of beta1 adrenergic receptors, or negative feedback from Ang I, potassium (increased by hypokalemia

and decreased by hyperkalemia). Plasma renin is the rate-limiting enzyme in classical RAS with an activity half-life of 10-15 minutes [32]. Angiotensinogen is a precursor molecule (85 amino acid alpha 2-globulin) primarily synthesized and constitutively secreted by the liver [33]. Renin cleaves the N-terminal of this polypeptide forming a biologically inert decapeptide Ang I [34]. Angiotensin-converting enzyme 1 (ACE1) is an exopeptidase on the plasma membranes of vascular endothelial cells, mainly in the pulmonary circulation, as well as in the tissues of the intestinal and urogenital tracts, the heart, adipose tissue, and the nervous system [35]. It cleaves the two amino acids from the carboxy-terminal of Ang I to make the octapeptide Ang II.

Ang II is the main mediator of the physiological and pathophysiological effects of the RAS, including volume regulation, blood pressure, and aldosterone secretion [36]. It mediates these effects by binding with close affinity to type 1 (AT1) and type 2 (AT2) receptors [37]. These receptors have different and often opposite responses attributed to cellular signalling pathways: the AT1 receptor stimulates protein phosphorylation and the AT2 receptor stimulates dephosphorylation [38]. Physiological activation of AT1 receptors results in vasoconstriction, sodium and water reabsorption, and aldosterone secretion. Pathological overactivation of AT1-R leads to inflammation, fibrosis, oxidative stress, tissue remodelling, and increased blood pressure [39]. Despite the low expression of AT2-R (compared to AT1-R), it is essential for mediating protective and opposing effects of AT1-R: inhibition of inflammation, fibrosis, central sympathetic outflow, vasodilation, natriuresis, and neurodegeneration [38].

Protective Pathway

The RAS has a second, protective arm as important as the well-known classical pathway that balances the physiological effects of the classical arm. The initiating enzyme of this arm is the monocarboxypeptidase angiotensin-converting enzyme type 2 (ACE2) with close structural homology to ACE1 [40,41]. Substrates for the enzyme are Ang I and Ang II, but its catalytic efficiency is 300 times higher for Ang II than for Ang I [41]. ACE2 is widely expressed in the lungs, cardiovascular system, kidneys, adipose tissue, and brain [42]. Membrane-bound levels of ACE2 are regulated by the metalloproteinase ADAM17, which cleaves it to create soluble ACE2 [43]. ACE2 converts Ang I to angiotensin 1-9 (Ang1-9), which is thought to affect the cardiovascular system through its interaction with AT-R, resulting in antihypertrophic, [44], antihypertensive and anti-inflammatory effects [45].

The most important active product of ACE2 is perhaps angiotensin 1-7 (Ang 1-7), formed by the hydrolysis of the C-terminal phenylalanine of Ang II [46]. Ang 1-7 can also be produced by NEP (neutral endopeptidase) activity on Ang I or propyl carboxypeptidase on Ang II [47]. Although the half-life of circulating Ang (1-7) is approximately 10 seconds due to rapid metabolism by peptidases such as ACE 1 and dipeptidyl peptidase 3 [48], it has multiple biological activities opposing Ang II [49]. Binding to its G-protein-coupled receptor, Mas-R It induces anti-inflammatory, vasodilatory, antiangiogenic, antihypertensive, antifibrotic [50], lipid, and glucose homeostasis-improving effects [51]. Mas-R is widely expressed in the brain, testis, heart, kidney, and blood vessels [26]. The Mas G-protein coupled receptor member D (MrgD) is the second receptor for Ang (1-7) with an affinity for Ala1-(Ang-(1-7), also called alamandin, another peptide member of the protective arm of RAS [52]. MrgD is expressed in the central nervous system, heart, kidney, dorsal root and trigeminal ganglia sensory neurons, gastrointestinal tract, respiratory tract, and intracellular mechanism of MasR and MegD activation increases levels of cyclic adenosine monophosphate (cAMP), phosphokinase A (PKA) and cAMP response element phosphorylation (CREB) [53]. Ang (1-7) was found to activate the phosphatidylinositol-3-kinase-Akt pathway, and nitric oxide synthase (NOS) in the heart [54].

The existence of an independent brain RAS in which Ang II is produced locally by angiotensinogen independently of the systemic RAS was established decades ago. Although the renin was found in neurons and astrocytes, local production of Ang II is also regulated by other brain-specific enzymes such as chymase and cathepsin G. The brain RAS is represented by the ACE1, Ang II, AT1-R, AT2-R, and components for the counterregulatory RAS including ACE2, Ang 1-7, MasR and MgrD [55]. It contributes to the brain's control of various physiological processes in the central nervous system (CNS), including blood pressure control, water balance, neuroendocrine regulation, stress responses, emotional and memory modulation, and pain susceptibility. Dysregulation of brain tissue RAS has been implicated in the development of various neurological disorders, including stroke, Alzheimer's disease, and Parkinson's disease [56]. Interest in functions of the protective axis RAS increased after the invasion of the SARS-CoV2 virus, which was found to bind to the peptidase domain of ACE2 through its spike protein receptor-binding domain as part of the viral infection process [57].

Pain control and RAS

Recent research data indicates that pathological changes in the expression of RAS components are involved in processes of inflammation or neuropathy, conditions associated with changed pain interpretation. The RAS has a complex role in tissue-specific nociception, signal transduction, pain processing, and interpretation. Ang II may play a dual role depending on its site of action in the existing pain processing network. Initial data on the effects of Ang II administered into the brain ventricles indicate brief inhibition of the phasic pain response [58]. In addition, the established involvement of brain angiotensin receptors in stress-induced analgesia and morphine-induced analgesia suggests an interaction with the opioidergic system [58]. Further study revealed that intracerebroventricular infusion of Ang II had a longer-lasting antinociceptive effect in a visceral model of nociception and that this effect was attenuated by blocking brain AT2 receptors [59]. These findings were later supported by a study on AT2-deficient mice, which showed a lower pain threshold accompanied by decreased levels of beta-endorphin in the brain [60]. Single doses of a selective AT2R agonist provoked short-term antinociception, and chronic activation of brain AT2R increased nociception and attenuated the physiological diurnal rhythm of phasic nociception [61].

Ang II and its specific receptors were shown to participate in the descending pain inhibitory network controlled by PAG. The research showed that administering Ang II the PAG can reduce pain caused by conditions such as plantar incision-induced allodynia [62]. Microinjection of Ang II into the caudal ventrolateral medulla (CVLM) induces hyperalgesia [63], which is not mediated by a direct neuronal connection between the CVLM and the spinal cord, but AT1 receptor-positive neurons in CVLMs project to the A5 noradrenergic nucleus of the pons, which further activates the neural circuit, amplifying the pain signal [64]. Ang II provoked a delayed short-lasting decrease in the mechanic pain threshold after intracerebroventricular injection and this effect was not abolished by nonselective angiotensin receptor antagonists [65].

The pronociceptive effect (scratching, licking, biting) of Ang II has been provoked at the spinal level by intrathecal administration of the peptide. These octapeptide-induced responses were attenuated by local injection of a selective AT1 receptor blocker rather than an AT2 receptor antagonist. Pain-related behavior was associated with p38 MAPK activation of spinal AT1 receptor-positive neurons and astrocytes. Furthermore, administration of the heptapeptide Ang (1-7) attenuated Ang II-induced nociceptive behaviour and inhibited p38 MAPK phosphorylation mediated through Mas1 receptors [66]. AT1 receptors have been shown to take a part in the regulation of circadian rhythms of nociception in

normotensive and spontaneously hypertensive rats. Chronic AT1 receptor blocking increased the pain threshold in normotensive rats only during the resting phase [67].

Based on the latest knowledge about the composition and functions of RAS, we note that Ang II does not remain in its original form for long but is metabolized to Ang-(1-7), suggesting that Ang-(1-7) is the main molecule responsible at least in part for the observed effects. It has been confirmed that the dorsal spinal cord expresses all the components required for the construction of both the ACE/Ang II/AT1 receptor axis and the ACE2/Ang (1–7)/MAS1 receptor axis [68].

A recent study provided data on the co-localization of MAS1, NK1, and NMDA receptors in the lumbar superficial dorsal horn. Ang (1–7) administered intrathecally inhibited the nociceptive response induced by spinal SP and NMDA. This study evidenced the significant modulation of the ascending nociceptive information by spinal MAS1 receptor activation [69]. All the components of the protective RAS arm were found in the DRG of the spinal cord. Moreover, the expression of MAS1 receptors in the DRG marked a daily increase in rats with a model of neuropathy pain. The neuropathic pain was markedly alleviated by the administration of Ang 1-7 [70]. RAS components have been found in the joint and bone tissues as well. Ang (1–7) was shown to inhibit cancer-induced bone pain (CIBP) via the MAS1 receptor without affecting tumor volume or bone metabolism [71]. The peripheral anti-nociceptive effects of Ang (1–7) were first demonstrated in rats experiencing PGE2-induced pain [72]. PGE2-induced hyperalgesia was subsequently suppressed by intraplanar administration of Ang (1–7) demonstrating its analgesic effect directly on nerve endings. This effect was further blocked by the MAS1- antagonist A779 but was not affected by the opioid receptor antagonist naloxone indicating an opioid-independent mechanism. A subsequent study revealed that the anti-nociceptive effect of Ang (1–7) was due to the activation of the neuronal nitric oxide synthase/cyclic GMP pathway and ATP-sensitive K+ channels locally at the site of administration [73].

Conclusions

Accumulating evidence describes the RAS as a homeostasis maintenance system and a complex balanced system controlling structural changes in response to environmental and behavioral challenges. Changes in the structures and dominance of one of the balancing arms of the RAS in various acute and chronic diseases emphasize the importance of this system for the correct understanding of pathological processes. The role of the RAS in the brain's pain control networks suggests further studies to uncover the importance of angiotensins in developing and treating various types of pain.

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