The endocannabinoid system in pain and inflammation: Its relevance to rheumatic disease
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Abstract

Pain is the most common manifestation of both acute and chronic inflammation that often challenges patients with rheumatic disease. Simply, we attribute this to local joint changes of pH in joints, the formation of radicals, enhanced joint pressure, or cytokine release acting on local nerves to produce pain. However, there is a more complex interplay of interactions between cytokines, mediators of inflammation, and ion channels that influence the final immune response and our perception of pain. Endocannabinoids, a group of less well-known endogenous bioactive lipids, have such manifold immunomodulatory effects able to influence both inflammation and pain. In this review, we overview the endocannabinoid system, its role in pain, inflammation, and immune regulation, and highlight the emerging challenges and therapeutic hopes.

Keywords: Endocannabinoids, pain, inflammation, arthritis

The Endocannabinoid System

Since the identification of endocannabinoid receptors, the potential of cannabinoid pharmacotherapy in clinical pain conditions has received much attention (1, 2). The three fundamental constituents of the endocannabinoid system include the endocannabinoid signaling molecules, G-protein-coupled cannabinoid receptors, and enzymes involved in ligand biosynthesis and inactivation. All these components have since been mapped throughout the peripheral nerve terminals and extend up to the supraspinal centers, which constitute the pain (nociceptive) pathway (3). Anti-nociception refers to the act or process of blocking the detection of painful or harmful stimuli by sensory neurons, thereby reducing one's sensitivity to pain. Preclinical studies of systemically administered cannabinoids have noted anti-nociception as a prominent feature in various models of pain (4). It is well accepted that endocannabinoids have anti-nociceptive effects (5-7). Under physiological conditions, potentially harmful stimuli are integrated by the nociceptors of primary afferent fibers and relayed for final processing in the supraspinal centers. In pathological states, the relaying of nocuous input by the nervous system is corrupted, resulting in abnormal nociceptive signaling and an aberration in pain responses (8). The analgesic effects of cannabinoids and their ligands are primarily mediated by the cannabinoid receptor 1 (CB1) via inhibition of presynaptic gamma-aminobutyric acid (GABA) and glutamatergic transmission. Within the nervous system, GABA transmission suppresses neuronal excitability (4, 9). Despite the endocannabinoids therapeutic appeal, its clinical application is marred by a plethora of psychoactive side effects, necessitating a cautious and critical evaluation as well as a search for an alternate cannabinoid-based approach or analogs to analgesia (10). In this review, the analgesic properties mediated by the endocannabinoid system is discussed and their potential use as novel pharmacotherapies in the treatment of arthritic pain is evaluated.

Endocannabinoid Ligands

The endocannabinoid system is regulated by a series of lipid signaling molecules known as “endocannabinoids,” belonging to the N-acylethanolamines. Of these, the two most widely investigated are anandamide (arachidonoyl ethanolamine [AEA]), initially isolated from porcine brain, and 2-arachidonoylglycerol (2-AG), initially isolated from canine intestines (11, 12). AEA is responsible for maintaining basal endocannabinoid signaling, binding to both the CB1 and CB2 receptors (13). At elevated concentrations, AEA also functions as a full agonist for the transient receptor potential vanilloid 1 (TRPV1), an ionotropic receptor responsible for the integration of noxious stimuli that cause pain (14, 15). In contrast to AEA, 2-AG functions as a full agonist for both CB1 and CB2 (16). In addition to these compounds, a series of other biochemically similar endocannabinoids such as 2-AG ether, vireodamine and N-arachidonoyl dopamine have been discovered. Knowledge of their function and regulatory role remains in its infancy.
Endocannabinoid Synthesis and Degradation

While the predominant endocannabinoids AEA and 2-AG are both lipid molecules generated from the breakdown of arachidonic acid, they share very few similarities in their biosynthetic pathways, as shown in Figure 1 (17). Endocannabinoid synthesis is a result of enzymatic cleavage of phospholipids within the cell membrane. Once released, the endocannabinoid ligands diffuse, acting locally as retrograde messengers to regulate the release of multiple presynaptic messengers. Following cellular uptake, the endocannabinoid ligands are quickly transported from the synaptic space and inactivated through subsequent catabolism via specific enzymes within the intracellular environment (18). This inactivation is catalyzed by intracellular enzymes which are unique to each endocannabinoid, and include fatty acid amide hydrolase (FAAH), the principal enzyme for AEA, and monoacyl glycerol lipase (MAGL) for the breakdown of 2-AG, as shown in Figure 2 (19). Enzymatic degradation of these endocannabinoids yields arachidonic acid and ethanolamine from AEA and glycerol from 2-AG, respectively (20).

Endocannabinoid Receptors

Various inhibition studies of the cannabinoid receptors have shown that endocannabinoids attenuate and suppress the perception of pain (21). The anti-nociceptive potency of cannabinoid agonists is strongly correlated with their ability to displace binding ligands from the cannabinoid receptor, obstructing their signaling. Localization studies using receptor binding immunohistochemistry and in-situ hybridization have mapped the distribution of the cannabinoid receptors along all levels of the pain nexus, providing a neuroanatomical framework befitting to the function of the cannabinoid system in sensory processing. The widespread expression of the cannabinoid receptors along the principal pain processing sites offer boundless opportunities for the development of analgesics for various pain conditions.

Cannabinoid receptor 1

Cannabinoid receptor 1 is the principal receptor of the central nervous system (CNS) and is densely expressed in several areas of the brain and supraspinal regions involved with nociceptive transmission. These areas include the cortex, basal ganglia, hippocampus, dorsal root ganglion (DRG), spinal cord, thalamus, periaqueductal gray (PAG), and amygdala (22). More recently, CB1 was shown to be densely cumulated within the frontal-limbic brain circuits that are key in both the affective and emotion-
al manifestations of human pain. CB1 inhibition of ascending nociceptive transmission, mainly at the thalamus level, has been shown to modify the emotional pain component acting at the limbic system and cortical areas. At the supraspinal level, CB1 activates the descending inhibitory pathway by suppressing GABA release in the PAG and rostral ventral medulla, intercepting descending input to the spinal cord nociceptive system through CB1 function (22, 23). At the level of the spinal cord, CB1 is densely expressed at the presynaptic terminals of primary afferents and excitatory neurons and regulate the transmission of noxious stimuli to the brain by inhibiting neurotransmitter release (24). As well as these central effects, CB1 receptors localize on sensory terminals in the periphery, gating the propagation of pain signals, contributing to peripheral analgesia (25). Indeed, inhibition of peripheral CB1 receptor expression suppressed the analgesic effects of administered cannabinoids in a neuropathic pain model, demonstrating the importance of peripheral CB1 function in cannabinoid-mediated analgesia (26).

Cannabinoid Receptor 2

In the periphery, CB2 receptors are widely located on immune cells and therefore represent a target for influencing inflammatory pain processing. Direct evidence of CB2-mediated anti-nociceptive effects was first reported in 1999 using the selective CB2 agonist, HU-308, which markedly decreased nociceptive behavior in the hind paw model of formalin treated rats (27). CB2 receptor agonists contributed to anti-nociception in other models of both inflammatory and nociceptive pain by suppressing the local secretion of pro-inflammatory factors by non-neural cells, which sensitize neighboring nociceptive neuron terminals. Stimulation of peripheral CB2 receptors therefore mediate anti-nociceptive responses in settings of neuropathic pain or inflammatory hyperalgesia by acting locally on immune cells in the periphery and microglia in the CNS (28). Although originally described as being restricted to immune cells, evidence for CB2 receptor expression in neural cells involved in pain perception and modulation has emerged (29). In vitro studies show CB2 receptor expression in human DRG sensory neurons and nerve fibers within the synovium and digit skin (30). Recently, CB2 receptors were detected on β-endorphin-containing keratinocytes on the epidermis of rat hind paws. This expression supports the notion that CB2-mediated effects may be regulated by a functional interplay between the endocannabinoid and µ-opioid systems, resulting in an indirect activation of opioid receptors expressed in primary afferent pathways (31). Thus, cannabinoid compounds may modulate pain by a number of pathways and a number of different mechanisms.

Intracellular Endocannabinoid Signaling

Cannabinoid receptors located on the presynaptic neurons regulate the synthesis and secretion of neurotransmitters to the synapse, as shown in Figure 3 (32). Action potentials in the presynaptic neuron cause the release of neurotransmitters within vesicles. Binding of the neurotransmitters to their postsynaptic receptors causes a depolarization of the postsynaptic membrane and the accumulation of Ca2+ in the cytoplasm, stimulating the activation of the calcium-dependent enzymes (phospholipase [PL] and diacylglycerol lipase [DAGL]) in charge of the biosynthesis of endocannabinoids. After synthesis, the endocannabinoid ligands are released and diffuse within the synapse, acting locally as retrograde messengers to regulate the release of multiple presynaptic messengers. Binding of the endocannabinoid ligands to the CB receptors induces a G-protein-dependent inhibition of presynaptic Ca2+ influx through voltage-gated Ca2+ channels. In addition to the pathway mentioned above, endocannabinoid binding may also cause the activation of enzymes PI3 kinase, sphingomyelinase, and phospholipase. As a result, hyperpolarization of the presynaptic membrane occurs, modulating the release of neurotransmitter and thus synaptic transmission. CB1 receptor stimulation regulates the intensity and duration of synaptic transmission (22). Similarly, in immune cells, CB2 activation has been shown to mediate an inhibitory effect on activation, cell motility and secretion of inflammatory mediators (28). Endocannabinoids are internalized by a selective transporter and degraded by specific enzymes (FAAH, MAGL) (32).

Endocannabinoids and Arthritic Pain

Arthritic pain is both nociceptive, resulting from the irritation of sensory nociceptors responsible for the detection of potentially noxious stimuli, and neuropathic, resulting from a malfunction in the somatosensory nervous system (33). Extensive innervation within the joint helps facilitate the sensation of pain in inflammatory joint conditions (34). Under normal physiological conditions, joint nociceptors are localized within the articular structure.
of the joint. Under inflammatory conditions, these silent nociceptors expand to adjacent tissues and function to propagate and amplify the sensation of pain (33, 34). The altered neuronal activity, also known as neuronal plasticity, is collectively believed to constitute the foundation of rheumatic pain and is characterized by hyperalgesia, an elevated noxious response to painful stimuli, and allodynia, a painful response to a normally mild and harmless stimulus. The primary components of the endocannabinoid signaling system (CB1, CB2, and FAAH) are characteristically expressed in the synovium of both osteoarthritis (OA) and rheumatoid arthritis (RA) patients, with compelling evidence to demonstrate an active participation in the pathophysiologic of joint pain (35). Preclinical and clinical studies support the therapeutic application of cannabinoids in the treatment of chronic pain, and to date, patients suffering from chronic arthritic and musculoskeletal pain represent the most prevalent users of medicinal cannabis (36). Despite this optimism, hesitation in clinical application is prevalent with extensive guidelines published in different countries (37-39).

**Changes in CB Receptor Expression**

In chronic pain states, central sensitization results in the reorganization of the spinal nociceptive circuitry through a localized up-regulation of CB1 and CB2 receptor expression along the pain nexus. As a result, hypersensitivity manifests as both hyperalgesia and allodynia occurs, for which standard analgesic treatments are unsuccessful. In neuropathic settings, both the up-regulation of spinal CB2 receptors (29, 40) and a greater effect of intrathecally administered cannabinoid CB2 agonists have been noted (40).

The functional importance of CB2 up-regulation in the integration and sensitization of OA pain has been demonstrated through the use of genetically modified mice (41). In pain experiments, mechanical induced allodynia was suppressed in transgenic mice overexpressing CB2 in the CNS54. By contrast, no significant changes in pain responses were observed in CB1 knockout mice, suggesting that the centrally controlled mechanisms of these nociceptive responses are primarily regulated via CB2, with minor input via CB1 (41). This is consistent with previous studies showing increased levels of AEA, 2-AG, and their synthesizing enzymes within the spinal cord of rat monoiodoacetic acid (MIA) models of OA (42). The tonic release of spinal endocannabinoid levels counteract peripheral sensitization through enhanced endocannabinoid signaling within the spinal cord (42). In a rat model of OA pain, systemic administration of CB2 agonist, JWH-133, suppressed pain behavior, while acute administration to the spine inhibited mechanically stimulated noxious neurotransmission (43).

More recently, an increased expression of CB1 accompanied by OA development has been demonstrated (36). CB1 receptors are located on peripheral sympathetic nerve terminals as well as on nociceptive nerve fibers, where they modulate adrenergic signaling to influence cytokine production (44). Stimulation of CB1 receptors desensitize neurons by modulating ion channels of the Transient Receptor potentials (TRP), suppressing action potentials and reducing pain. Externally administered AEA and CB1 agonist, arachidonyl-2-chloroethylamide (ACEA), have been shown to significantly reduce the firing rate of afferent nerve fibers in OA joints, but not in the control joints, suggesting a tonic release of endocannabinoids at the joint level in OA. Considering the close positioning of transient receptor potentials type 1 (TRPV1) and CB1 receptors in relation to nociceptive transmission both have drawn interest in the development of novel therapies of OA pain and is discussed in more detail below (3, 9).

**Changes in TRP Stimulation**

TRP channels, also known as capsaicin receptors, are a group of ligand-gated ion channels responsible for the detection and integration of noxious stimuli (14, 44). Primarily found in the nociceptive neurons of the peripheral nervous system, stimulation of the transient receptor potential vanilloid receptor type 1 (TRPV1) results in a cation influx and the production of an action potential that consequently results in the sensation of pain. As such, the TRPV1 channels represent a prime focus for the development of novel analgesics. Modulators of TRPV1 function include agonists such as capsaicin, used clinically as creams or patches, to desensitize TRPV1 activity and reduce pain. Indeed, it has been well-documented that heavy capsaicin dosing depletes neuropeptides levels, with a resultant suppression in the severity of adjuvant-induced joint disease (45). Other antagonists that block TRPV1 activity (capsazepine, ruthenium red) as well as clinical trial compounds (AMG517, GRC6211, NGD 8243) have also been shown to be effective in reducing pain but have not progressed in development due to undesirable side effects such as hyperthermia.

In arthritis, TRPV1 receptors are located on peripheral cells and sensory neurons abundant ly expressed in arthritic synovial tissue. The importance of these receptors in arthritis is highlighted in TRPV1/- knockout animals that demonstrate elevations in pain threshold and an associated reduction in joint inflammation (46). In addition to enhanced neurotransmitter release, stimulation of TRPV1 is associated with increases in inflammatory mediators contributing to joint inflammation (47). A recent study found that tumor necrosis factor-alpha (TNF-α)-induced mechanical hyperalgesia in an arthritic model could be blocked by a central, but not peripheral, injection of a TRPV1 antagonist (48). Similarly, interleukin-1 beta (IL-1β) can upregulate TRPV1 expression in arthritic rat DRG neurons, as well as sensitize articular C-fibers (48). Together, these findings indicate a novel link between TRP channels and cytokines in the generation of joint pain through central sensitization (44). Similar to cytokines, endocannabinoid’s AEA, OEA, and palmitoylethanolamide (PEA) can activate the TRPV1 receptors (49, 50). AEA is believed to modulate synaptic plasticity, a key component in arthritic pain, through actions at both the pre and postsynaptic TRPV1 channels (15). On exposure, the TRPV1 channels are rapidly desensitized, resulting in reduced calcium influx and increased pain thresholds. In a recent study, TRPV1 activation via AEA produced nociceptive behavior through the excitation of C-fibers (51). Additionally, cross-talk between CB1 and TRPV1 co-expressed on sensory nerves, modulate pain and inflammation in arthritis (52). When co-expressed, CB1 agonists suppressed TRPV1 activation through dephosphorylation, increasing the threshold level for agonists (47). Pharmacological elevations of AEA in an arthritic rat were shown to suppress hypersensitivity of afferent nociceptors and elevated pain thresholds by a process containing CB1 and TRPV1 channels (15). This mechanism was confirmed through the use of joint blood flow experiments, which demonstrated that the vasomotor effects of a CB1 agonist in rat knees could be inhibited by TRPV1 antagonism (53). Similarly, in a model of OA pain, local administration of CB1 agonist ACEA reduced mechanosensitivity of afferent nerve fibers, suppressing nociceptive transmission in OA and healthy rat knee joints. Inhibition of both the TRPV1 and CB1 receptors suppressed ACEA responses, indicating the dual involvement of both channels in ACEA-mediated nociception. At present, TRPV1 has been shown to co-localize with CB1 in sensory DRG and the spinal cord, and brain neurons and with CB2 receptors in sensory neurons and osteoclasts (30).

More recently, the transient receptor potential canonical 5 (TRPC5) expressed on fibroblast like synoviocytes cells within the joint has been shown to protect against pain and inflammation in arthritic mice (54).
using knockouts or pharmacological agents is correlated well with the propagation of joint inflammation and hyperalgesia, providing evidence that TRCP5 is a negative regulator of inflammation. The proposed mechanism of action suggested by Alawi et al. (54) is that TRPC5 acts to suppress inflammation by influencing the early cytokine-immunity axis discussed below (54). These exciting results highlight how TRP receptors protect against pain and vascular joint inflammation in arthritis.

Endocannabinoids and Inflammation

There is increasing and exciting evidence showing that endocannabinoids regulate the immune response at both the innate (monocytes, macrophages, neutrophils, NK cells, eosinophils, basophils, mast cells) and adaptive immune level (55). Immune cells are not only able to be influenced, but are also able to generate and secrete endocannabinoids that lead to changes in immune-cell behavior as well as the production of other inflammatory factors that subsequently influence tissue inflammation (56, 57).

The Role of Endocannabinoids in Joint Inflammation

Chinese healers, who have known about the healing properties of endocannabinoids since 2000 BC, have claimed that cannabis "undoes rheumatism" (58). Evidence supporting the anti-inflammatory effects of endocannabinoids come from preclinical studies that have shown that all classes of cannabinoids including phytocannabinoids (tetrahydrocannabinol, cannabidiol) and synthetic analogs such as Ajulemic acid, "Nabibone," and elmeric acid possess anti-inflammatory effects (18). These anti-inflammatory effects may be due to direct action on participating immune cells, or by changes in the local endocannabinoid concentrations that then carry out anti-inflammatory actions. In arthritis, persistent inflammation results in the infiltration of immune cells and the subsequent development of hypersensitivity. Synovial serum samples from patients with RA consistently express elevated cytokine levels such as TNF-α, interleukin-6 (IL-6), and IL-1β, which act directly to sensitize joint nociceptors and stimulate the release of prostaglandins (59, 60). In an elegant study by Sancho et al. it was shown that AEA can inhibit TNF-α-induced NF-κB activation by direct inhibition of the IκB kinase (59, 60). With both cannabinoid receptors and endogenous ligands present in inflamed human joints, targeting this system may hold therapeutic promise for both inflammatory, as well as degenerative arthritis (60). Administration of cannabinoid agonists, WIN55212 and CP55940, have shown the ability to reduce inflammatory IL-6 and interleukin-8 (IL-8) cytokine production by fibroblast-like synoviocytes cells, ameliorating acute inflammation and associated pain in arthritic joints (2, 63). Similarly, systemic administration of the CB2 agonist, JWH133, suppressed pain and corrected deviation in circling pro- and anti-inflammatory cytokines in the rat MIA model (43). These anti-inflammatory effects are limited by the rapid cellular uptake and degradation of endocannabinoid metabolites but can overcome through the inhibition of the catalytic enzyme FAAH allowing longer physiological effects (35). In vivo studies by Krstev et al. (64) reported that FAAH inhibition with compound URBS97 can elevate tissue concentrations of AEA by inhibiting local endocannabinoid degradation and dampen inflammatory pain in rodent models of OA. In a similar study, URBS97 suppressed inflammatory hyperemia in a mouse model of acute arthritis (64). In the periphery, FAAH inhibition mediates anti-inflammatory effects by down regulating cytokine production and the desensitization of TRPV1, resulting in analgesia (15). Inhibition of AEA catalobism is said to have promising effects in the management of OA pain mediated by both anti-inflammatory (65) and anti-hyperalgesia actions (62). Therapeutic intervention in peripherally restricted CB1 antagonist and FAAH inhibition are promising strategies to ameliorate chronic inflammation and pain in RA.

Neurogenic Inflammation

In neurogenic inflammation, local afferent neurons secrete inflammatory mediators such as neurokinin A, substance P (SP), and calcitonin gene-related peptide which perpetuate inflammation in rodent models of arthritis. The anti-inflammatory potential of CB2 has been confirmed in mice models of arthritis (61, 62). The protective CB2 effects include the suppression of pro-inflammatory cytokine and damaging proteinases secretion; as well as regulating immune cell adhesion and migration to the inflamed joint. Together, this helps slow the perpetuation of disease and alleviate associated arthritic pain primarily derived from localized inflammation (36). Further to this, elevated levels AEA and 2-AG are detected in the synovial fluid of RA and OA patients, but absent in healthy controls, suggest that local endocannabinoid secretion may assist in minimizing inflammation in the arthritic joints (4, 60). With both cannabinoid receptors and endogenous ligands present in inflamed human joints, targeting this system may hold therapeutic promise for both inflammatory, as well as degenerative arthritis (60). Administration of cannabinoid agonists, WIN55212 and CP55940, have shown the ability to reduce inflammatory IL-6 and interleukin-8 (IL-8) cytokine production by fibroblast like synoviocytes cells, ameliorating acute inflammation and associated pain in arthritic joints (2, 63). Similarly, systemic administration of the CB2 agonist, JWH133, suppressed pain and corrected deviation in circling pro- and anti-inflammatory cytokines in the rat MIA model (43). These anti-inflammatory effects are limited by the rapid cellular uptake and degradation of endocannabinoid metabolites but can overcome through the inhibition of the catalytic enzyme FAAH allowing longer physiological effects (35). In vivo studies by Krstev et al. (64) reported that FAAH inhibition with compound URBS97 can elevate tissue concentrations of AEA by inhibiting local endocannabinoid degradation and dampen inflammatory pain in rodent models of OA. In a similar study, URBS97 suppressed inflammatory hyperemia in a mouse model of acute arthritis (64). In the periphery, FAAH inhibition mediates anti-inflammatory effects by down regulating cytokine production and the desensitization of TRPV1, resulting in analgesia (15). Inhibition of AEA catalobism is said to have promising effects in the management of OA pain mediated by both anti-inflammatory (65) and anti-hyperalgesia actions (62). Therapeutic intervention in peripherally restricted CB1 antagonist and FAAH inhibition are promising strategies to ameliorate chronic inflammation and pain in RA.

Neural-immune Circuits in Inflammation

In RA, sustained stimulation of the sympathetic nervous system propagate inflammation through norepinephrine signaling. The influence of adrenergic signaling and the loss of sympathetic nerve fibers in the inflamed tissue in RA has been demonstrated in several animal models of arthritis (68). Sympathetic injury in the early phase of the disease has been shown to ameliorate experimental arthritis, indicating the pro-inflammatory influence of adrenergic signaling (69). During arthritic inflammation, nerve repulsion factors released by macrophages result in the withdrawal of sympathetic neurons from the synovial tissue and the subsequent depletion of synovial noradrenergic concentrations (68). As a result, inadequate norepinephrine levels causes a shift from previously stimulated anti-inflammatory α-adrenergic to β-adrenergic signaling, favoring pro-inflammatory cascades (70). Peripheral norepinephrine release from the sympathetic terminals is regulated through the CB1 receptors and can be stimulated by respective CB1 endocannabinoid agonists such as AEA and 2-AG. In adjuvant arthritis, β2 receptor stimulation suppresses levels of TNF-α, increases anti-inflammatory interleukin-10 (IL-10), shifts the T-cell profile, and enhances T-regulatory immune responses potentiating a widespread reduction in joint destruction, inflammation, and pain (71). Similarly, it has been demonstrated that antagonism of CB1 at splenic sympathetic terminals mediates strong anti-inflammatory effects and improves collagen-induced arthritis in an in vivo mouse model, which was reversed by β2 adrenergic antagonism (72). Recent research using CB1 knockout mice showed impaired neurogenesis when compared to wild type, suggesting endogenous CB1 signaling may promote basal levels of neurogenesis (73, 74). While further studies are required, this pos-
sible application to sympathetic nerves may promote the extension of nerves previously lost from the synovial space during arthritic inflammation (68, 70).

As discussed above, neural circuits can modulate immune responses by secreting compounds that influence cell function or through the detection of inflammatory mediators, which in turn causes the nerves to relay signals or release immunomodulatory peptides back to the immune system. The release of IL-1 following inflammation, for instance, act on the hypothalamic-pituitary adrenal axis via neuronal input to release glucocorticoids that then influence the peripheral immune response outcome. Such neural-immune reflex circuits that regulate innate immunity are well understood. Other neural-immune reflexes such as dopamine and electro-acupuncture based circuits also exist but are less-well defined, as shown in Figure 4 (75). The effects of acupuncture stimulation on neural reflex immunity circuits provides important and exciting insights into the regulation of innate immunity and the discovery of new targets as anti-inflammatory therapeutics (76).

Current Work and Future Direction of Research

In this review, we have focused on the basic sciences of endocannabinoids and overviewed their role in inflammation and pain. While compelling evidence suggests the therapeutic potential of endocannabinoids therapy in arthritis and chronic musculoskeletal pain syndromes, barriers to research include insufficient legally registered marijuana manufacturers and a limited number of clinical trials. In the study by Blake et al. (77), the effect of cannabis spray, “Nabiximols,” improved pain scores and disease activity when compared with placebo in 28 joints observed. Treatment took place over a 5-week period in a randomized, double-blinded trial group of 58 RA patients. Although adverse events in the active treatment group were not serious, they were common (77). In studies examining the endocannabinoid-based drug “Nabilone” on pain outcomes in fibromyalgia, the statistically significant effects were outweighed when side effects were taken into consideration (78-80). A separate review of cannabinoid-based therapies in chronic pain conditions, three of which were rheumatic pain (2 fibromyalgia, 1 RA), concluded that while the majority demonstrated improvements in pain, no direct evidence on effects of herbal marijuana in rheumatic pain (78).

The FAAH inhibitor, PF-04457845, showed both analgesic and anti-inflammatory effects in animal studies comparable to naproxen (79). However, when compared to naproxen, PF-04457845 was ineffective for OA pain when compared to placebo-control in a randomized phase II clinical trial (79). As endocannabinoids do not solely mediate their effects via the CB1/CB2 receptors, it is thought activity mediated via the TRPV channels hampered analgesic potential (80). Also interesting and worth mentioning is the relationship between cyclooxygenase enzyme (COX) inhibitors and endocannabinoids. COX enzyme is involved in the generation of prostaglandins from arachidonic acid that mediates inflammation. What is less well appreciated is that COX enzyme also metabolizes endocannabinoids to prostaglandin-glycerol esters for 2-AG and prostaglandin ethanolamines for AEA, as shown in Figure 2. These bioactive lipids may have a role in inflammation (81). What

Figure 4. Neural reflex immunity circuits. Various stimuli such as (a) inflammatory (e.g., IL1), (b) bacterial, (c) acupuncture, or (d) pressure/acupuncture are detected by sensory afferent nerves and relayed to interneurons located in the spinal cord/brainstem. The generated efferent signals work to dampen the innate immune responses signaling through (a) efferent neurons of the hypothalamic-pituitary adrenal (HPA) axis, (c) via the release of dopamine via the adrenal medulla, or (b) via the local axon-axon reflex which suppress the innate immune responses by cytokine release and immune cell activation. Efferent signals sent via the sympathetic system (d) or vagus nerve (e), release neurotransmitters that influence the immune response (75).
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is also not known is the functional consequences between cyclooxygenase-2 (COX-2) inhibition and endocannabinoid effects. More recently, focus has been drawn to dual-acting compounds such as OMDM-198. OMDM-198 works to increase FAAH substrate concentrations while simultaneously inhibiting TRPV1 receptors. In an MIA rat model of OA, OMDM-198 exhibited a meaningful reversal of hypersensitivity in joint pain, representing a promising avenue in endocannabinoid pain management.

While the benefits of pharmacologically prepared cannabinoid treatments have been inconsistent with large variations between species and population groups, they appear to have clinical benefits warranting the need for further investigation. Comprehensive evaluations through well-controlled randomized trials are also required to clarify the true clinical efficacy and long-term risks associated with cannabinoid therapy. Advancements in our understanding of the endocannabinoid system and cannabinoid pharmacology, has raised the hope of exciting new pharmacological entities. Cannabis-based medications which enhance endocannabinoid function may represent a novel therapeutic solution to disorders associated with chronic pain and remains a promising avenue of contemporary importance.

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