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Review

Peripheral analgesia: Hitting pain where it hurts

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ABSTRACT

Pain is a complex biological phenomenon that encompasses intricate neurophysiological, behavioural, psychosocial and affective components. Protracted or chronic pain alerts an individual to a possible pathological abnormality and is the main reason why patients visit a primary care physician. Despite the pervasiveness of chronic pain in the population, the effectiveness of current pharmacological therapies remains woefully inadequate and prolonged treatment often leads to the development of undesirable side-effects. Since the vast majority of chronic pain originates in a specific tissue or group of tissues, it may be advantageous to target pain control in the periphery and thereby circumvent the known risks associated with non-specific systemic treatments. This review spotlights a number of promising targets for peripheral pain control including the transient receptor potential (TRP) family of neuronal ion channels, the family of proteinase activated receptors (PARs), cannabinoids, and opioids. A critical appraisal of these targets in preclinical models of disease is given and their suitability as future peripheral analgesics is discussed.

As most of us can attest, pain is an unpleasant sensory experience which instils negative feelings and as such is something we generally try to avoid. Pain can either be short-lasting (acute) or more protracted (chronic). Acute pain is typically beneficial to the individual as it warns of actual or impending tissue damage allowing rapid, reflex evasive action to be carried out. This nociceptive pain arises in the periphery via direct activation of pain-sensing nerve terminals by noxious mechanical, thermal or chemical stimuli. Acute pain can usually be relieved by local administration of pharmacological agents (*e.g.* lidocaine, non-steroidal anti-inflammatory drugs) or non-pharmacological approaches (*e.g.* rubbing the affected area or application of ice). Chronic pain, on the other hand, tends to be a maladaptive response to some underlying pathology. Pain is considered chronic if it continues unabated beyond about 3 months and typically outlasts the normal healing response or continues in the absence of any observable tissue damage [1]. Chronic pain is the primary reason why people seek medical attention, yet the arsenal of available effective remedies is limited. Furthermore, all of the currently available analgesics have some degree of negative side-effect associated with them. The time has come to develop more effective, safe and consistent analgesics.

Firstly an overview of the classic pain pathway is provided. Most types of pain begin in the periphery with a noxious stimulus activating unspecialised free nerve endings associated with small diameter primary afferent neurones (Fig. 1). This physical stimulus is subsequently transduced into an electrochemical signal which is transmitted from the periphery towards the central nervous system along slowly conducting primary afferent nerves. Upon entering the dorsal horn of the spinal cord, these impulses are transmitted via chemical synapses to second order neurones where they are subsequently conveyed along ascending tracts to higher centres in the brain. By relaying these impulses to specific areas of the brain such as the somatosensory cortex and the amygdala, the electrochemical signals are shaped into a psychophysical experience incorporating any protective motor reflexes and emotional responses. The critical details of this entire process are still inadequately understood and form the basis of a fascinating area of neuroscientific research.

Current pharmacological agents attempt to tackle pain in both the central as well as the peripheral divisions of the pain pathway. Since centrally acting analgesics tend to produce numerous secondary unwanted side-effects (*e.g.* dysphoria, motor deficits and addiction) it stands to reason that peripherally acting drugs have the advantage of circumventing these problems by targeting pain at the source. This review will address these issues and highlight the palpable benefits of peripherally-directed analgesia. While the peripheral mediators covered here are by no means exhaustive, the intent is to highlight emerging and possibly contentious targets worthy of further discussion.

1. Transient receptor potential ion channels

In an era of molecular cloning technology, a large number of cation channels have emerged belonging to the transient receptor potential (TRP) family. Identification of these ion channels on polymodal nociceptors triggered intense interest in the pain research community, many of whom consider TRP channels to be central integrators of

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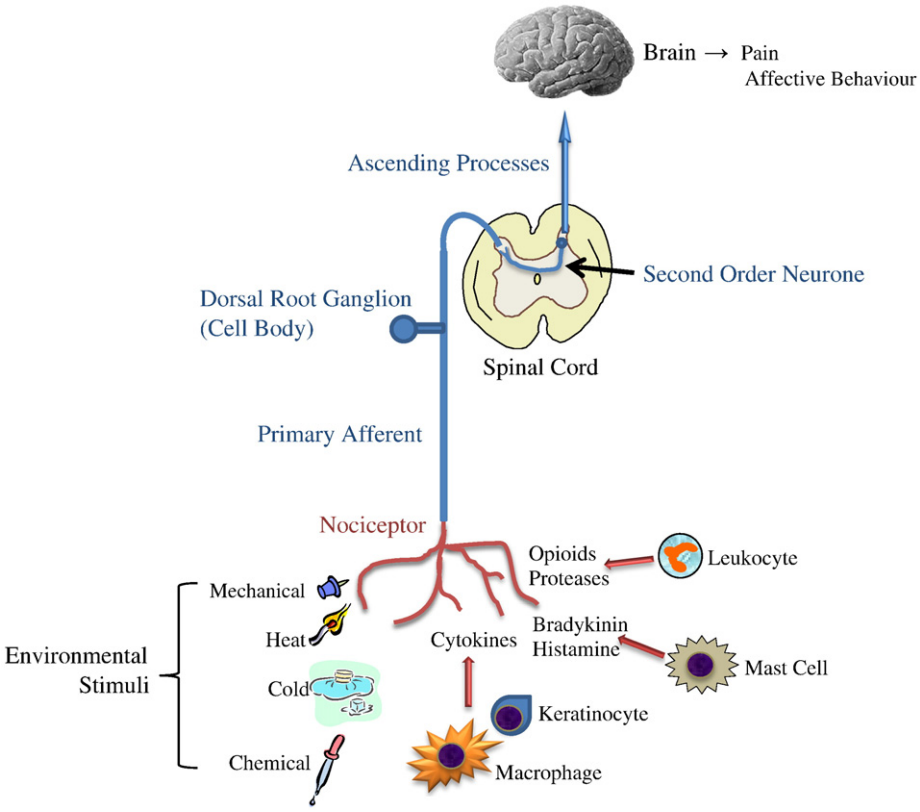

Fig. 1. The pain pathway from periphery to brain. Noxious environmental stimuli can activate peripheral nociceptors leading to the sensation of pain. Various cells residing in the vicinity of the nerve terminal can release mediators which either heighten or reduce the activation threshold of the nerve.

pain transduction and modulation. Indeed, TRP channels have been linked with all aspects of sensory physiology including mechanosensation, thermosensation, hearing, vision and taste. The latter sensory modality prompted a slew of molecular biology studies and at one point it felt like just about everything in the kitchen larder activated a TRP channel. It is plain to see why these prevalent and precocious ion channels have garnered favour as targets of interest by academia and the pharmaceutical industry alike. A summary of the most commonly studied TRP channels is shown in Table 1.

The channel which has received the greatest attention is the TRP-vanilloid 1 (TRPV1) channel. Cloned in 1997 [2], TRPV1 is known to be activated by protons, temperatures in excess of 42 °C, endovanilloids, cannabinoids, and a host of other chemical mediators known to be present in the "inflammatory brew". The promiscuous nature of the TRPV1 channel renders it a central modulator of pro-algesic stimuli from disparate sensory modalities. Consequently, several small molecule TRPV1 antagonists are currently being assessed for potential analgesic properties in various painful diseases. In joints, intra-articular injection of the TRPV1 agonist capsaicin was found to cause synovial vasodilatation [3] which can be blocked by the selective TRPV1 antagonist SB366791 [4]. Thus, a role for TRPV1 in arthritic processes was established. This was later confirmed by the observation that the severity of inflammatory joint disease was significantly less in TRPV1 knockout mice compared to wild-types [5,6]. Pain behaviour studies found that the tactile allodynia associated with Freund's complete adjuvant-induced arthritis was less in TRPV1 deficient mice compared to genetically normal controls [6,7]. Similarly, systemic treatment of osteoarthritic rats with the TRPV1 antagonist A-425619 reduced the weight bearing deficit associated with this disease [8].

In the gastrointestinal system, TRPV1 contributes to visceral hypersensitivity making it an intriguing target for intractable abdominal pain syndromes. Intestinal biopsies taken from subjects undergoing routine colonoscopy revealed that patients with inflammatory bowel syndrome had over three times as many TRPV1-positive nerve fibres compared to control subjects [9]. These additional TRPV1-positive afferents can then be sensitized by peripherally circulating inflammatory mediators released from enteric nerves, endothelial cells, and immunocytes. Data supporting a neuroimmune component of peripheral sensitization has recently been reported [10]. Supernatants of peripheral blood mononuclear

Table 1
Some of the commonly studied TRP channels in pain research and their activators/blockers.

TRP channel	Activator	Temperature sensitivity	Selective channel blocker
TRPV1	Protons Vanilloids Ethanol	43–52 °C	SB366791 Capsazepine
TRPV2	Δ9-Tetrahydrocannabinol 2-aminoethoxydiphenyl borate Mechanical swelling	≥52 °C	None
TRPV3	Eugenol 2-aminoethoxydiphenyl borate Vanillin Camphor	32–39 °C	None
TRPV4	4α-Phorbol 12,13-didecanoate Epoxyeicosatrienoic acids Mechanical	25–34 °C	HC-067047 RN-1734
TRPA1	Cinnamaldehyde Allicin Acrolein Formalin Mustard oil	17 °C	AP18 HC-030031
TRPM8	Menthol Eucalyptol Spearmint Icilin	18–24 °C	5-Benzyloxytryptamine

cells taken from patients with post-infectious inflammatory bowel syndrome caused increased firing of mouse visceral primary afferents. This observation suggests that during inflammatory bowel syndrome, immunocytes can locally release mediators that sensitize colonic nociceptors leading to intestinal pain.

Having established TRPV1 as an integrator of peripheral pain processes, a number of small molecule TRPV1 antagonists rapidly emerged and touted as potential novel analgesics. Subsequent tests with small molecule TRPV1 antagonists, however, revealed that some of these pharmacological agents produced profound hyperthermia [11,12] and as such could not be used therapeutically. Later studies found that the primary site of TRPV1 antagonist-induced hyperthermia was the periphery and that targeting TRPV1 channels in the central nervous system may be more beneficial for pain control while avoiding thermoregulatory complications [13,14]. Some researchers claim to have discovered TRPV1 antagonists with differential pharmacology i.e. compounds which inhibit TRPV1-induced pain but have no effect on core body temperature control [15]. Since TRPV1 is activated by a large array of sensory stimuli and chemical agents, it remains to be seen whether it is possible to isolate the pain regulatory aspect of TRPV1 without evoking undesirable side-effects.

Another vanilloid channel receiving increasing attention is the TRPV4 channel. It is expressed on sensory free nerve endings [16] and is known to be activated by moderate temperatures (25–34 °C), mechanical stimuli [17], and chemical ligands such as 4α-phorbol 12,13 didecanoate (4α-PDD) and epoxyeicosatrienoic acids [18,19]. The identification of TRPV4 channels on cochlear hair cells and vibrissal Merkel cells gave the first indication that this particular TRP channel may be involved in mechanosensation [20]. Subsequent studies showing TRPV4 expression on Aδ and C fibres in mouse skin highlighted the possibility that TRPV4 could be involved in peripheral mechanonociception [16]. In fact, TRPV4 knockout mice have a higher threshold to noxious mechanical stimuli compared to wildtype animals [21]. In the gut, TRPV4 is co-localised with inflammatory neuropeptides in small diameter sensory neurones and administration of a TRPV4 agonist greatly enhances visceral mechanosensation [22]. Furthermore, behavioural responses to noxious colonic distension were inhibited in animals lacking TRPV4 cation channels [22].

Moving from hot to cold, the first channel found to be responsive to lower temperatures was found to be expressed by a subset of TRPV1 expressing polymodal nociceptors and due to its large number of ankyrin repeating motifs was named TRPA1 [23,24]. TRPA1 channels are believed to be activated by moderate temperatures (approximately 17 °C) and their expression has been shown to be increased in dorsal root ganglia (DRG) ipsilateral to Freund's complete adjuvant-induced inflammation [25] or nerve injury [26]. Mice missing functional TRPA1 channels show reduced thermal and mechanical pain responses to intraplantar injection of bradykinin or allylisothiocyanate [27,28]. Peripheral injection of the TRPA1 antagonist AP18 [(Z)-4-(4-chlorophynyl)-3-methylbutyl-3-en-2-oxime] significantly reduced mechanical hyperalgesia and cold allodynia in a model of chronic inflammation [29]. Another TRPA1 antagonist HC-030031 [(2-[1,3-dimethyl-2,6-dioxo-1,2,3,6-tetrahydro-7H-purin-7yl)-N-(4-isopropylphenyl)acetamide] dose-dependently reduced flinching in the formalin model of chemonociception [30] as well as mechanonociception in Freund's complete adjuvant model of inflammatory pain and in the spinal nerve ligation model of neuropathic pain [31]. More recently, mechanical hyperalgesia produced by either intra-plantar administration of tumour necrosis factor-α or intra-articular injection of Freund's complete adjuvant was attenuated by AP18 suggesting that TRPA1 channels are involved in inflammatory and arthritic pain [32].

Another ion channel known to be involved in cold sensation belongs to the melastatin family of TRP channels and was termed TRPM8 [33,34]. Triggered by temperatures in the range 18–24 °C, as well as other cooling compounds such as menthol, eucalyptol and icilin, TRPM8 is expressed by about 15% of small diameter DRG and trigeminal neurones [33–35]. Being a non-selective cation channel, stimulation of TRPM8 channels leads to membrane depolarization and afferent nerve firing. Involvement of TRPM8 channels in cold hypersensitivity is not yet certain although the emerging evidence is highly suggestive of the involvement of TRPM8 in cold pain processing. In several neuropathic and inflammatory pain models, TRPM8 expression is increased and the channel offsets thermal as well as mechanical hypersensitivity although it is unclear whether this is achieved by TRPM8 activation or blockade [26,36,37]. Experiments examining acute inflammatory pain induced by intraplantar injection of Freund's complete adjuvant found that pain was inhibited by peripheral TRPM8 agonism [36]. Similarly, neuropathic pain elicited by chronic constriction injury to the sciatic nerve or peripheral nerve demyelination using lysolecithin could be ameliorated by topical cooling, menthol and icilin [36]. Katsura et al., however, found that antisense inactivation of central TRPM8 channels had no effect on L5 spinal nerve ligation-induced neuropathic pain [25]. The role of TRPM8 in pain modulation still requires further analysis; however, some of the controversy could be related to whether the agonists/antagonists are administered centrally versus peripherally leading to differential responses.

2. Proteinase activated receptors

Proteinases hydrolyse the peptide bonds in long chain proteins and are involved in multiple physiological processes such as tissue degradation, apoptosis, the blood clotting cascade and tissue remodelling. Further to this classic enzymatic activity, proteinases can also act as signalling molecules by triggering a small group of G-protein coupled receptors. There are currently four members of these proteinase activated receptors (PAR_{1-4}) and they are expressed in a plethora of organs such as joints, skin, gut and lungs. Unlike other G-protein coupled receptors which rely on the binding of a specific chemical ligand to cause receptor stimulation, PARs are activated by a proteinase causing a change in receptor conformation. Tethered to the extracellular N-terminal loop of the receptor there is a short amino acid sequence which has the potential to stimulate the same receptor but is impeded from doing so by an amino acid cap. A locally released proteinase cleaves the receptor at a specific point thereby removing the cap and exposing the functional amino acid sequence which can now bind to its complementary domain on the extracellular N-terminal loop of the receptor rather like the hammer of a gun engaging with a cartridge. The stimulated receptor then initiates a series of second messenger cascades leading to intracellular signalling. Experimentally, it is possible to activate the PARs with a small peptide agonist which matches the tethered ligand sequence (see Table 2 for a list of the various PAR activating peptides). This approach has the benefit of selectively stimulating the PAR of interest without possible off target effects that would undoubtedly be initiated by addition of the proteinase.

With the exception of PAR_3, all of the PARs have been implicated in the control of peripheral nociception. PAR_1 is primarily activated by thrombin and plays an important role in cardiovascular disease and inflammation [38]. Recent evidence has shown that PAR_1 is expressed by mouse DRG and treating these sensory neuronal cell bodies with a PAR_1 agonist leads to calcium mobilization [39]. Local injection of the PAR_1 agonist TFLLR-NH_2 dose-dependently blocks hyperalgesia in both acute and chronic models of inflammation [39–41]. It is believed that PAR_1 achieves anti-nociception by causing the secondary release of endogenous opioids since PAR_1 responses are attenuated by the non-specific opioid antagonist naloxone [39]. Interestingly, PAR_1 knockout mice respond normally to noxious thermal and mechanical stimuli suggesting that PAR_1 is only active during inflammation and does not affect physiological pain [39].

PAR_2 has been localised on sensory nerves and is primarily activated by trypsin and mast cell tryptase resulting in the release of

Table 2
Summary of PAR modulators and the role of PARs in pain.

PAR	Activating peptide	Inactive peptide	Antagonist	Role in pain
PAR1	TFLLR-NH_2	FTLLR-NH_2	RWJ-58259 SCH-205831 SCH-5303048	↓ Pain [36–38]
PAR2	SLIGRL-NH_2 2-Furoyl-LIGRLO-NH_2	LSIGRL-NH_2 2-Furoyl-OLRGIL-NH_2	ENMD-1068	↑ Pain [40–42]
PAR3	None	None	None	Unknown
PAR4	AYPGKF-NH_2	YAPGKF-NH_2	Pepducin p4Pal-10	↑ Pain joints [45,46] ↓ Pain skin, gut [49]

algogenic neuropeptides such as substance P and calcitonin gene-related peptide [42]. As such, PAR_2 is considered to be a pro-nociceptive receptor whose activation causes increased pain in joints [43], gut [44], and hindpaws [45]. The mechanism by which PAR_2 promotes pain appears to involve capsaicin-sensitive TRPV1 ion channels. Evidence supporting this concept comes from the fact that the PAR_2 agonist SLIGRL-NH_2 does not cause secondary hyperalgesia in TRPV1 knockout animals and PAR_2-mediated nociception can be blocked by the TRPV1 receptor antagonist SB366791 [43]. Furthermore, Grant et al. found that TRPV4 ion channels are sensitised by PAR_2 agonists and PAR_2-induced mechanical hyperalgesia is absent in TRPV4 knockout mice and attenuated by the TRPV4 antagonist 4αPDD [46]. Mast cells have also been implicated in PAR_2 responses as the receptor has been localised on the surface of these cells and PAR_2 activation leads to mast cell degranulation [45,47]. The endogenous proteinases released during inflammation and which activate PAR_2 *in vivo* leading to pain have yet to be fully ascertained. Serine protease levels are known to be elevated in the synovial fluid of arthritis patients [48,49] and in the faeces from inflammatory bowel syndrome patients [50]. In the latter study, faecal supernatants taken from inflammatory bowel syndrome patients caused colono-rectal mechanical allodynia in wildtype mice which was less pronounced in PAR_2 knockout animals. This finding suggests that enzymatic mediators released during inflammatory bowel syndrome have the potential to sensitize visceral afferents leading to pain.

Recent studies have demonstrated PAR_4 expression in rat DRG [51] as well as in neural elements associated with joints [51,52] and the bladder [53]. Immunolocalisation and reverse transcription-polymerase chain reaction studies on rat cultured DRG found that PAR_4 is co-localised with the inflammatory neuropeptides substance P and calcitonin gene-related peptide [54]. The role of PAR_4 in nociceptive processing is still unclear with conflicting reports of hyperalgesia and analgesia. Peripheral administration of the PAR_4 agonist AYPGKF-NH_2 to knee joints, for example, results in increased mechanosensitivity of joint afferents and a reduction in pain threshold [51,52]. This pro-nociceptive effect of PAR_4 activation can be blocked by the bradykinin B_2 antagonist HOE140 but not by TRPV1 antagonism. In contrast, intraplantar and intracolonic injection of AYPGKF-NH_2 caused a reduction in nociception by deactivating the pain promoting effects of the TRPV4 channel [55]. Furthermore, feacal supernatants taken from patients suffering from ulcerative colitis produced visceral hyposensitivity in mice which was found to be due to the actions of the serine proteinase cathepsin G on PAR_4 [56]. Thus, the effect of PAR_4 activation on peripheral pain modulation appears to be organ specific and suggests the possible existence of PAR_4 subtypes (*e.g.* PAR_{4a}, PAR_{4b}, etc.) This hypothesis requires further investigation and characterisation.

3. Cannabinoids

Cannabinoids are highly lipophillic alkaloids derived from the hemp plant *Cannabis sativa*. At least 66 distinct chemicals have been identified in *C. sativa* and these plant-borne agents are collectively called phytocannabinoids. There is a growing collection of man-made cannabinoids (synthetocannabinoids) which are based on the structure of their plant counterparts. Finally, it has been found that the body produces its own natural cannabinoids called endocannabinoids of which anandamide and 2-arachidonylglycerol are the most prominent.

Cannabinoids bind to two G protein-coupled receptor subtypes (CB_1 and CB_2) with a third putative cannabinoid receptor (GPR55) having recently been described [57]. The CB_1 receptor was originally identified in the central nervous system [58] where it is believed to be the most abundant G protein-coupled receptor [58–60]. The CB_2 receptor was later identified on peripherally circulating macrophages and in the spleen [61]. Originally thought to be restricted to the periphery, CB_2 receptors have also been localised in the brainstem where they regulate emesis [62]. CB_1 receptors are present throughout the pain pathway including on peripheral neurones, spinal neurones and in pain processing areas of the brain [63–66]. Stimulation of neuronal CB_1 receptors by a cannabinoid ligand leads to hyperpolarisation and a decrease in calcium dependent neurotransmitter release. Since CB_2 receptors are relatively scarce in the central nervous system, they were originally thought to be an ideal target for pain control as selective CB_2 receptor agonists would not be able to produce the psychotropic effects commonly associated with CB_1 receptor activation. The reality has been less impressive with a number of CB_2 receptor agonists having low selectivity and producing multiple off-target responses. Some of the pharmacological tools used in cannabinoid research are listed in Table 3.

A plethora of cultures have used phytocannabinoids for millennia to treat a whole host of illnesses. Public acceptance of cannabinoids as a viable means of treating disease has been slow to catch on despite a significant body of evidence supporting a role for cannabinoids in the safe treatment of various chronic pain disorders. In joints, for example, the CB_1 receptor has been identified on nerve terminals innervating the synovium [67] and peripheral administration of the selective CB_1 receptor agonist arachidonyl-2-chloroethylamide (ACEA) dramatically reduced nociception in a rat model of osteoarthritis [68]. Local injection of the CB_1 receptor antagonist AM251 into arthritic joints led to an increase in nociceptor activity suggesting that endocannabinoids are released into the joint to help offset articular pain [68]. Other studies looking at models of inflammatory joint disease have found that both CB_1 and CB_2 receptors are involved in mediating articular analgesia [69–71]. Thus, there is a strong rationale for putting cannabis in our joints. The mechanism by which cannabinoids achieve anti-nociception in joints appears to involve TRPV1 ion channels [68,72,73]. During inflammation, TRPV1 channels

Table 3
Overview of some of the commonly used cannabinoid reagents and their target receptor.

	Cannabinoid receptor		
	CB_1	CB_2	GPR55
Non-selective agonists	WIN55,212-2 CP55,940 HU210		N/A
Full agonists	Arachidonyl-2-chlroethylamide (ACEA) O1812 Methanandamide Arachidonyl-cyclopropylamide	AM1241 JWH015 JWH133 GW405833 HU308 L-759,633 L-759,656	O1602
Selective antagonists	AM251 AM281 SR141716A LY320135	AM630 SR144528	O1908

are upregulated [74,75] and TRPV1/ CB_1 receptor co-expression is enhanced [76]. In inflamed tissue, the TRPV1 channel exists in a phosphorylated state [77] rendering it more sensitive to endocannabinoids such as anandamide [78–80]. Endocannabinoids then bind directly to either TRPV1 channels or TRPV1/ CB_1 receptor heterodimers leading to channel deactivation and pain relief.

Cannabis has been used as an adjunct to chemotherapy in cancer patients due to its anti-emetic effects and palliation properties. A seminal study by Noyes et al. found that low dose Δ-9 tetrahydrocannabinol was equipotent to codeine in alleviating pain in cancer patients, although higher doses produced psychotropic side-effects such as ataxia, dizziness and blurred vision [81]. In pancreatic cancer patients, it was noted that peripheral CB_1 receptor density was inversely proportional to subjective pain scores suggesting that cannabinoid receptors may be involved in controlling cancer pain [82]. Animal studies also support a role for cannabinoids in reducing cancer pain. The non-selective synthetocannabinoids WIN55,212-2 and CP55,940 were found to reduce bone cancer pain and this effect could be blocked by a CB_1 receptor antagonist but not a CB_2 receptor blocker [83,84]. The cancer pain produced by intraplantar injection of human oral carcinoma cells could also be attenuated by peripheral injection of WIN55,212-2 or the CB_2 receptor agonist AM1241 [85].

Cannabinoids have also been shown to be analgesic in painful disorders of the gastrointestinal system. The writhing response elicited by intraluminal deposition of formic acid in mice could be alleviated by oral administration of either Δ-9 tetrahydrocannabinol or cannabinol [86]. Pain induced by colorectal distension could also be reduced by treating animals with a local injection of either WIN55,212-2 or the CB_2 receptor agonist JWH015 [87]. By administering the same cannabinoid agonists, these investigators were also able to reduce mechanical hyperalgesia in a model of colitis [87] confirming that peripherally administered cannabinoids may be viable analgesics for the treatment of visceral pain.

Damage to peripheral sensory nerves by either injury, altered metabolism or viral infection often results in neuropathic pain which may be episodic or constant. Neuropathic pain is often difficult to diagnose and hence treat since there is often no overt sign of tissue injury. Animal studies indicate that cannabinoids can alleviate neuropathic pain, offering a much needed alternative to the current first order treatment options of anti-convulsants and tricyclic antidepressants. In the rat spinal ligation model of neuropathic pain [88], CP55,940 was able to reverse tactile allodynia while WIN55,212-2 has been shown to reduce pain in the sciatic nerve chronic constriction model [89]. Unfortunately, these non-selective cannabinoid agonists were administered systemically in these studies rendering it impossible to know which cannabinoid receptor subtype was involved and where the site of analgesic action was taking place. Peripheral injection of WIN55,212-2 into the hindpaw was found to be anti-nociceptive in neuropathic pain rats with the effect being blocked by the CB_1 receptor antagonist SR141716A [90]. In contrast, spinal *cfos* expression is elevated in CB_1 receptor knockout mice compared to wild-type littermates [91] while CB_1 receptor deficiency has no effect on neuropathic pain responses [92]. In an ingenious model in which peripheral CB_1 receptors were deleted and central CB_1 receptors were maintained, the analgesia produced by systemic injection of cannabinoids was reduced [93]. These data indicate that peripheral CB_1 receptors are critical for neuropathic pain control and suggest that peripherally-restricted CB_1 receptor agonists could be efficacious in the treatment of neuropathic pain.

CB_2 receptors have also been implicated in neuropathic pain control. The CB_2 receptor agonists AM1241 and JWH133 have been found to attenuate neuropathic pain in the spinal nerve ligation model [94,95]. Furthermore, neuropathic pain generated by sciatic nerve constriction was ameliorated by GW405833 [96,97], although it should be noted that extremely high doses of the compound were required to achieve anti-nociception and compound selectivity is equivocal.

4. Opioids

Lauded for their therapeutic benefits, infamous for their euphoric effects, opioids are the archetypical pain killer. Opioids are a group of alkaloids derived from the poppy plant *Papaver somniferum*. The earliest description of poppies being actively cultivated as a crop was in c. 3400 BC by the Sumerians in Mesopotamia who referred to the plant as Hul Gil or "Joy Plant". For millennia opioids have been used medically to treat health problems as diverse as dysmenorrhoea, cough, teething, asthma and arthritis. The most familiar opioid is morphine which was first isolated by the pharmacologist Friedrich Sertüner in 1806. Named after Morpheus the Greek god of dreams, morphine is one of the most widely used analgesics and is still a first line treatment for most chronic pain sufferers.

The opioids act on four distinct G-protein-coupled receptors *viz*. the δ-opioid receptor (DOR), κ-opioid receptor (KOR), μ-opioid receptor (MOR), and the nociceptin/orphanin FQ receptor (NOP). Stereospecific binding assays originally discovered that opioid receptors were located in the central nervous system [98–100] although the identification of the individual opioid receptor subtypes would not be determined for another 3 years. Martin et al. [101] found that morphine and ketacyclozine acted via the MOR and KOR respectively while Hughes et al. determined that the endogenous opioids Met- and Leu-enkephalin bound to a receptor in the vas *de*ferens and called it the DOR [102]. In 1995, a novel opioid peptide with significant homology to dynorphin A was discovered by two independent laboratories [103,104]. One group called the peptide orphanin FQ since it did not bind to any known receptor subtype, while the other group referred to the opioid as nociceptin based on its ability to modulate pain processing in mammals. Based on these two independent findings, the peptide was referred to the rather clumsy concatenation of nociceptin/orphanin FQ.

Following on from their identification in the central nervous system, opioid receptors were found to be expressed in the periphery [105]. Functional studies showed that prostaglandin-induced hyperalgesia in the rat hindpaw could be ameliorated by locally applied low dose morphine thereby establishing a role for opioids in peripheral pain control [106]. This effect was confined to the treated hindpaw since contralateral morphine injection was unable to offset ipsilateral

Table 4
Overview of some of the commonly used opioid agonists/antagonists and their target receptor.

	Opioid receptor			
	DOR	KOR	MOR	NOP
Selective agonists	DPDPE [DAla^2]deltorphin-I or II SNC80	U69593 CI977 Salvinorin A	DAMGO Endomorphin-1 or -2 Sufentanil	N/OFQ, Ro646198 UFP112
Selective antagonists	Naltrindole	Nor-binaltorphimine GNTI	CTOP	J113397 SB612111 UFP101

hyperalgesia. Similarly, the MOR agonist fentanyl reduced the pain produced by bilateral intraplantar carrageenan injection, but only in the paw in which the opioid was administered [107]. Systemic treatment with low dose fentanyl had no effect on paw withdrawal threshold confirming a peripheral site of action of the opioid. In addition to MOR-mediated analgesia, peripheral administration of DOR, KOR, and NOP agonists can also modulate inflammatory pain. Table 4 highlights some of the commonly used opioids. The selective KOR agonist U50488 reduced nociceptor spontaneous activity in acutely inflamed cat knee joints [108] as well as reducing pain behaviour in Freund's complete adjuvant inflamed paws [109]. Confirmation that U50488 was producing antihyperalgesia via KORs was indicated by the blocking effects of the KOR antagonist nor-BNI [110] but not the MOR antagonist CTAP [111] nor the DOR antagonist ICI174864 [112]. The synthetic DOR agonist DPDPE has been found to be antihyperalgesic when injected locally into the inflamed rat hindpaw [109] and when administered around the primary tumour site in an animal model of bone cancer pain [113]. Some of the original studies looking at a peripheral site of action for nociceptin/orphanin FQ found that the opioid acted on mast cells and caused secondary release of substance P from nerve terminals leading to peripheral sensitization and pain [114–117]. Curiously, during inflammation nociceptin/orphanin FQ desensitized nerve fibres suggesting that the peptide has a dual role in pain modulation depending upon the inflammatory status of the tissue [118]. This anti-nociceptive effect of nociceptin/orphanin FQ has been demonstrated in models of arthritis [118], colitis [119], capsaicin-induced neurogenic inflammation [120], as well as in response to noxious heat [121] and formalin-induced hyperalgesia [122]. Thus, it appears that under normal conditions peripherally administered nociceptin/orphanin FQ is pro-nociceptive but switches to having anti-nociceptive properties in tissues where there is pre-existing pain.

A number of reports describe an augmentation in the analgesic capacity of opioids in various inflamed tissues [123–126]. The main explanation proffered to account for this phenomenon is that during inflammation there is believed to be an increased synthesis and peripheral axonal transport of opioid receptors resulting in an upregulation of receptors in the inflamed tissue [127–129]. Since endogenous opioid expression is also increased following inflammation [130,131], it begs the question as to why these tissues are still painful. Closer inspection of the preceding studies reveals that opioid receptor expression was only assessed in the acute phase of inflammation (*i.e.* up to four days after induction) and overlooks the more clinically relevant chronic phase of the inflammatory process. In a chronic model of arthritis, it was found that peripheral MORs are actually downregulated leading to a loss in opioid anti-nociception [132]. The decrease in receptor number is thought to be due to an overaccumulation of endogenous ligand in the chronically inflamed tissue leading to internalisation of membrane-bound MOR [130]. This hypothesis would explain the inability of endogenous opioids to offset chronic pain as well as accounting for the poor analgesia reported by patients receiving intra-articular μ-opioid therapy [133–135]. Recovery strategies aimed at preserving opioid receptor number during chronic inflammation could improve endogenous opioid analgesia and improve the efficacy of peripherally restricted opioid agonists.

5. Summary

At first glance, the pain pathway appears to be a straightforward line: pain signals originate in the periphery, travel centrally to the spinal cord and thence to the brain. This overly simplistic notion is, however, deceptive and belies complex neuroanatomical circuitry and neurophysiological processing of which we still only have a rudimentary understanding. Spinal and supraspinal mechanisms augment, diminish and refine the nociceptive signal shaping it into the emotional and psychophysical experience we call pain. Central plasticity changes are thought to be responsible for the chronicity of long-term pain, but are these higher centres truly necessary for the ultimate treatment of nociceptive pain? Management of neuropathic pain is of course entirely different since the source of pain appears to be in the various divisions of the nervous system itself. However, for the majority of chronic pain, targeting nociceptive activity in the periphery should enable us to directly control pain generation at the source and thereby limit input to the central nervous system. It could be argued, however, that by the time a patient seeks medical assistance and ultimately receives appropriate pain management, the central nervous system has undergone plasticity changes and is now driving the pain. But surely the opposite must also hold true. A prolonged and targeted reduction in nociceptor activity by pharmacological means and/or physical therapy would allow the central nervous system to undergo structural reorganisation back to normal thereby returning the patient to a pain free state. As we have seen, controlling nociceptor activity is a difficult task with a multitude of chemical mediators and neurophysiological processes being involved. Adding to the complexity is that peripheral targets can change their sensitivity and even phenotype depending upon the severity of tissue pathology, concentration of agonist and crossreactivity of other inflammatory mediators. Nevertheless, by tackling pain in the periphery it should be possible to provide symptomatic relief to millions of chronic pain sufferers while avoiding the hazardous side-effects of pervasive systemic treatments.

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