

EDITORIAL

Recent advances in targeting ion channels to treat chronic pain

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Introduction

'Divine is the task to relieve pain' (Hippocrates).

Pain represents a complex heterogeneous sensory experience, almost uniquely influenced by emotional condition and experience. This broad description alludes to the fact that pain is difficult to treat pharmacologically. Pain may be acute (a desirable physiological response warning of actual or impending tissue damage) or chronic (an undesirable state that must be treated). Chronic pain is estimated to affect ~1.5 billion people globally; in the UK alone, the Department of Health estimates at least 28 million adults (~40% of the population) suffer at some time from chronic pain, leading to poor quality of life, financial and societal consequences and increased risk of mortality. Equally, the pharmaceutical industry estimates that the global market in pain therapeutics currently exceeds US\$600 billion·year⁻¹.

The elucidation of nociceptive circuitry and the function of ion channels that control inherent excitability within such pathways (including the de novo synthesis of such channels during disease and their persistent expression, which can engender a profound hyperexcitability even after resolution of tissue damage) provides a rational basis for the development of novel analgesics. Current research activity focussed on targeting ion channels involved in pain transduction, axonal conduction and neurotransmitter release (in spinal cord

through to higher centres) provides important targets for discovery of new analgesics. For example, knowledge of voltage-gated ion channel function, such as **Na⁺ channels** (Cardoso and Lewis, 2018) and **K⁺ channels** (Du *et al.*, 2018) classically involved in neuronal excitability and **Ca²⁺ channels** (Patel *et al.*, 2018) responsible for transmitter release, has progressed *via* advances in, amongst others, measurement of protein up-regulation during pain states, knowledge of channel dysfunction in pain syndromes associated with human genetics and development of transgenic knock-down and knock-in technology to support *in vitro* and *in vivo* models. Alongside this, increased structural and functional knowledge of **transient receptor potential (TRP) channels** (Moran and Szallasi, 2018) and **acid-sensitive ion channels (ASICs)** (Lee *et al.*, 2018) has generated further potential targets. Equally, ionotropic receptors, in particular, excitatory NMDA **glutamate** (Haley and Dickenson, 2016) and inhibitory **GABA** and **glycine** (Zeilhofer *et al.*, 2012) receptors and additional targets such as **P2X ATP receptors** (Bernier *et al.*, 2018), offer further possibilities. In addition to pore-forming subunits as molecular targets, neuronal excitability may also be modulated *via* binding partners such as voltage-gated Ca²⁺ channel auxiliary $\alpha 2\delta$ subunits (the binding site for gabapentinoids; Gong *et al.*, 2018) and collapsin response mediator protein 2, which modulates voltage-gated Ca²⁺ (and potentially Na⁺) channel activity in pain states (Moutal *et al.*, 2018). It is also clear that

academic and pharmaceutical industry drug development programmes are starting to deliver lead low MW entities and biological agents to target ion channels or associated proteins. In this themed section, many of the above areas are the subject of contemporary review articles by leading experts in the pain field. Together with original research papers detailing recent pharmacological progress, this themed section provides a timely snapshot of the field and an evaluation of direction of travel in terms of potential for future clinical translation.

Measuring ion channel function in nociceptive pathways

Nociceptor activation generates afferent input, principally mediated by small diameter C and A δ fibres (typically mechanical and thermal nociceptors) of dorsal root ganglia (DRG). Free nerve endings of musculoskeletal/cutaneous afferents and hollow organs/smooth muscle afferents detect somatic and visceral pain respectively. Pain transduction at free nerve endings is mediated by channels such as non-selective TRP channel isoforms (including **TRPV1** and **TRPA1**) and voltage-gated Na⁺ channels (see Waxman and Zamponi, 2014) but also ATP-gated P2X ionotropic receptors (e.g. **P2X3**, Bernier *et al.*, 2018) and acid-sensing ion channels (in particular **ASIC3**; Deval and Lingueglia, 2015). Channels expressed in nociceptors with a role in controlling sensory neuron excitability include the voltage-gated Na⁺ channel subtypes **Nav1.1**, **Nav1.3**, **Nav1.6**, **Nav1.7**, **Nav1.8** and **Nav1.9** (Cardoso and Lewis, 2018); Cav3 voltage-gated Ca²⁺ channel isoforms (François *et al.*, 2015) and K⁺ channels such as **two-pore-domain (K2P)** TREK and TRESK (Mathie and Veale, 2015) and **KCNQ (Kv7)** M channels (Du *et al.*, 2018). Channels such as **Ca_v2.2** (Patel *et al.*, 2018) and associated $\alpha 2\delta$ auxiliary subunits (Gong *et al.*, 2018) have a key role in regulating neurotransmitter and neuropeptide release at central terminals in the dorsal horn of the spinal cord. Many ion channels in sensory neurons are regulated by changes in expression level/cellular location and modulated by inflammatory mediators in different neuropathic or inflammatory pain states. Recent work is also starting to reveal modulators such as miRNA that regulate gene expression of proteins involved in nociception. For example, miRNA regulates the expression of $\alpha 2\delta$ subunits in TrkB DRG neurons (Peng *et al.*, 2017).

Use of isolated DRG neurons to understand the role of ion channels in nociceptive transduction, electrogenesis and neurotransmitter release is a mainstay of *in vitro* pain research. Sensory neurones have traditionally been classified according to cell diameter or specific markers (e.g. **TrkB receptors** are associated with light-touch mechanoreceptors, **TrkA receptors** define peptidergic nociceptors, while isolectin B4 has been used to identify non-peptidergic nociceptors). However, there is considerable diversity of nociceptors as revealed by subgroups defined by differences in response (threshold and duration) to thermal or mechanical stimuli, which is supported by recent data from single cell RNA-Seq analysis of sensory neurons (Usoskin *et al.*, 2015); such knowledge may allow specific pain state-dependent targeting. Isolated DRGs are highly amenable to a number of recording techniques, principally electrophysiology, where whole-cell voltage and current clamp and multi-electrode array

(MEA) recordings are the routinely used techniques of choice to unravel the mechanistic role of individual ion channels in sensory neuron excitability. Similarly, ion channel-specific techniques such as imaging using selective fluorescence dyes to measure changes in intracellular ion concentrations or membrane potential are highly prevalent. In addition to measuring native, endogenous DRG ion channel activity using the patch clamp technique, heterologous expression of exogenous channels carrying mutations associated with human pain syndromes in DRGs has also provided an important understanding of the contribution of individual variants, for example, the hyperexcitability of Na_v1.7 gain-of-function mutants underlying erythromelalgia (Yang *et al.*, 2018). Higher throughput MEA recordings of cultured DRGs has been challenging due to limited spontaneous activity *in vitro*. However, there has been recent progress in MEA recordings from DRGs, including from those derived from human embryonic stem cells (Alshawaf *et al.*, 2018), which could provide important phenotypic screening platforms as a basis for identifying novel analgesics. It is often useful to support native neuronal studies with work in expression systems such as transiently transfected or stably expressed mammalian cells (Loucif *et al.*, 2018; McArthur *et al.*, 2018) or oocyte expression (Lee *et al.*, 2018; Mourot *et al.*, 2018). Such model systems, theoretically, allow ion channel pharmacology and/or biophysics to be examined in isolation. These efforts are supported by developments in automated patch clamp technology, predominantly using expression systems, to measure ion channel activity (Bell and Dallas, 2018; Loucif *et al.*, 2018).

Nociceptive afferent fibres predominantly innervate the dorsal horn of the spinal cord through interaction with projection neurons or interneurons in the superficial lamina I and II. Synaptic neurotransmission between nociceptors and dorsal horn is mediated by release of excitatory glutamate and neuropeptides such as **substance P** and inhibitory GABA and glycine; such transmitter release is controlled predominantly by **Ca_v2.2** voltage-gated Ca²⁺ channels (Patel *et al.*, 2018) but also potentially by **Ca_v3.2 channels** (Jacus *et al.*, 2012). The spinal cord dorsal horn slice preparation represents a useful *ex vivo* preparation to investigate pain pathways. Electrophysiology is again the primary technique, largely focussing on recordings of excitatory (glutamatergic) or inhibitory (GABAergic and/or glycinergic) postsynaptic currents from lamina I and/or II neurons in the superficial dorsal horn (Nerandzic *et al.*, 2018; Winters *et al.*, 2018). It is also possible to co-culture DRG and spinal cord neurons to investigate nociceptive function (Yu *et al.*, 2018). Postsynaptic expression of Ca_v1 family voltage-gated Ca²⁺ channels in the dorsal horn may also contribute to maintenance of pathological pain (Roca-Lapirot *et al.*, 2018). Thus, the **Ca_v1.2** channel is expressed somatically and at proximal dendritic shafts, and **Ca_v1.3 channels** at distal somato-dendritic compartments. It also appears that glial elements also regulate dorsal horn responses, with evidence for expression on **P2X4 receptors** on microglial cells within these regions (Bernier *et al.*, 2018).

Nociceptive information from dorsal horn to the thalamus and cerebral cortex is carried by five major second-order ascending pathways: the chief spinothalamic tract and the spinoreticular, spinomesencephalic, cervicothalamic and spinohypothalamic tracts. Thalamic nuclei such as the lateral

and medial nuclei receive inputs from these tracts and pass information to areas of the cerebral cortex pain-processing centres, including the cingulate gyrus and the insular cortex. Electrophysiological recordings of thalamocortical neurons either acutely isolated or in thalamic brain slice preparations are well supported by electroencephalography (and also by functional neuroimaging techniques). Thalamic nuclei express high levels of Ca_v3 T-type voltage-gated Ca^{2+} channel isoforms (Talley *et al.*, 1999), which control neuronal excitability around the resting membrane potential. In particular, **$\text{Ca}_v3.1$ channel** activity in the thalamus and somatosensory cortex has been implicated in signalling trigeminal neuralgia pain (Choi *et al.*, 2016), commonly held as the most excruciating manifestation of neuropathic pain, and $\text{Ca}_v3.2$ represents a major molecular target in pain (Snutch and Zamponi, 2018).

Descending inhibitory pathways, such as those from periaqueductal grey (PAG) region, locus ceruleus and rostral ventromedial medulla, also feedback directly or indirectly onto dorsal horn neurons. These pathways are likely to be inhibited during neuropathy, while descending excitatory pathways are promoted (Boadas-Vaello *et al.*, 2016). Principal neurotransmitters in the descending inhibitory pathways include biogenic amines such as **5-HT** and **noradrenaline**. Within the nociceptive pathways described above, the concerted activation of a number of proteins, including key ion channels, can lead to peripheral sensitization (an increased sensitivity to pain stimuli) and a resultant hyperalgesia (an increased pain response to noxious stimuli) causing central sensitization. For example, glutamate acts on AMPA receptors to mediate fast baseline responses and on NMDA receptors to mediate delayed responses to pain; neuropeptides, such as substance P and **CGRP**, and also gasotransmitters, such as **NO**, enhance glutamatergic effects (hypersensitivity), which enhances further transmission of pain signals (hyperalgesia). Sensitization can also lead to allodynia (where non-noxious stimuli are perceived as noxious pain). Of relevance here is that a range of animal models of pain are used to determine the effects of potential analgesics on parameters such as hypersensitivity and allodynia and on parameters related to central sensitization such as wind-up (an activity-dependent persistent increase in neuronal firing). These models of chronic pain include not only the widely used spinal nerve ligation, partial sciatic nerve ligation and chronic constriction injury models but also more specific models such as Freund's complete adjuvant-induced inflammatory pain (Lee *et al.*, 2018), the carrageenan model of peripheral inflammation (Nerandzic *et al.*, 2018), post-surgical pain and HIV-induced sensory neuropathy models (Moutal *et al.*, 2018) and the chronic visceral hypersensitivity model (Castro *et al.*, 2018).

Current pharmacology and future directions

As described above, nociceptive pathways are well defined physiologically but also can possess an emotional component that adds to the difficulty in providing effective treatment. Pharmacologically, the mainstay to treat acute and palliative pain are opioids, which mimic the body's

endogenous painkillers, and short-term use of agents such as **paracetamol** and non-steroidal anti-inflammatory drugs which typically act to limit endogenous inflammation due to release of chemical mediators that can cause peripheral sensitization. Opioids act predominantly *via* μ opioid receptors, which are highly expressed in areas such as the PAG and superficial dorsal horn. Mechanistically, opioids act to inhibit presynaptic Ca_v2 voltage-gated Ca^{2+} channels *via* generation of inhibition $\text{G}\beta\gamma$ subunits; opioids can also activate postsynaptic K^+ channels to reduce neuronal excitability. Overall, opioids act to promote descending inhibitory pathways to suppress activity of ascending tracts such as the major spinothalamic projection neurons in the dorsal horn. Although opioids are widely used to treat acute pain, chronic pain is often neuropathic in classification, and traditionally, opioids are regarded to lack therapeutic efficacy in neuropathies. Alongside these are issues of tolerance and addiction and the abuse potential associated with opioids, as well as the widespread media concern over opioid abuse in countries such as the USA, where abuse of **fentanyl** and **oxycodone** is now rivalling or even surpassing the illicit use of **heroin**. Such concerns have propelled pharmacological imperatives for safer analgesic alternatives for chronic conditions to the fore. In terms of neuropathic pain, the current therapeutic approach is exemplified by the UK NICE Guidelines, which recommend a range of drugs from tricyclic antidepressants, such as **amitriptyline**, **serotonin-noradrenaline reuptake** inhibitors such as **duloxetine**, **gabapentinoids** or topical **capsaicin**. Despite these interventions, the unmet clinical need associated with chronic pain means that academia and industry are continuing their search for low MW compounds and biological agents that target ion channels or associated proteins. In particular, advances in high-throughput electrophysiology screening, transcriptomic and proteomic studies have generated small molecular entities to target Na^+ channels. Here, $\text{Na}_v1.7$ channels have emerged as a rational target due to human genetic associations of pain syndromes with both loss- and gain-of-function mutations; in particular, the identification of loss-of-function recessive mutations that lead to congenital insensitivity to pain (Cox *et al.*, 2006). Human trials of the leading aryl sulfonamide $\text{Na}_v1.7$ blocker PF-05089771 showed only modest efficacy in a phase II trial for painful diabetic peripheral neuropathy (Donnell *et al.*, 2018); however, there is initial evidence of efficacy of PF-05089771 at mutant $\text{Na}_v1.7$ channels associated with erythromelalgia (Cao *et al.*, 2016). In general, there continues to be great interest in $\text{Na}_v1.7$ channels across the pharmaceutical industry; for example, Genentech/Xenon are developing the oral small molecule $\text{Na}_v1.7$ inhibitor GDC-0310 (completed phase I). In addition, there are two positive phase II results with the Vertex $\text{Na}_v1.8$ channel inhibitor (VX-150) in acute bunionectomy pain and osteoarthritic pain. Other companies have embraced non-selective inhibition of Na^+ channels to target pain (by focusing on frequency-dependent inhibition); for example, Biogen's pyrrolidine-based compound vixotrigine (formerly raxatrigine, CNV-1014802) is currently undergoing phase III clinical trials for trigeminal neuralgia. There are also a range of naturally occurring toxins which act as voltage-gated Na^+ channel pore blockers and/or gating modifier which may provide lead compounds; such agents include **tetrodotoxin**, which

continues to move through clinical trials for treatment of chemotherapy-induced pain (Cardoso and Lewis, 2018). It may also transpire that current therapeutic agents have utility here; for example, **carbamazepine** has reported positive effects at mutant $\text{Na}_v1.7$ channels associated with erythromelalgia (Yang *et al.*, 2018). Development of alternative Na^+ channel blockers such as local anaesthetics containing photo-switchable moieties has also been proposed to be of clinical interest by allowing potential 'opto-pharmacology' approaches to target pain (Mouro *et al.*, 2018).

An important proof-of-concept for ion channel therapeutic agents has come from the introduction of the direct $\text{Ca}_v2.2$ channel blocker **ziconotide** and the indirectly acting gabapentinoid drugs, which bind $\alpha2\delta$ and probably function by modulating $\text{Ca}_v2.2$ trafficking within nociceptive pathways (Bauer *et al.*, 2010), but may also involve prevention of synaptogenesis associated with development of pain states (Yu *et al.*, 2018). For gabapentinoids, numbers needed to treat are ~ 7 , and side effects can include dizziness and nausea (Patel *et al.*, 2018) but pregabalin has been approved for painful diabetic neuropathy and fibromyalgia. Ziconotide, a synthetic version of the marine cone snail toxin ω -conotoxin MVIIA, is approved for treatment of chronic pain but is limited by a relatively narrow therapeutic index and associated side effects and is a large molecule with poor blood-brain barrier permeability that must be delivered intrathecally. The search for alternative low MW, CNS-penetrant, activity state-dependent blockers that can be delivered orally has generated a number of potential lead compounds, including ZC88, A-1264087, N-triazole oxindole (**TROX-1**) and CNV-2197944 (Patel *et al.*, 2018). Clearly it will be important to translate promising preclinical data into human trials, and a note of caution also exists in that the $\text{Ca}_v2.2$ channel blockers Z160 and **ABT-639** failed to show efficacy in phase II trials. Alongside $\text{Ca}_v2.2$ channels, development of small molecules to target Ca_v3 subtypes, and in particular $\text{Ca}_v3.2$ channels, is a major area of drug development. Here, clinical development of two candidates ABT-639 and MK-8998 have stalled, whereas development of another compound, **Z944**, remains on-going (Snutch and Zamponi, 2018). An area of optimism is the pipeline of alternative small molecules that target Ca_v3 which are showing clear preclinical promise. Expression system work has also identified the ortho-phenoxyanilide derivative, MONIRO-1, which showed preferential activation state-dependent and use-dependent block of $\text{Ca}_v2.2$ and $\text{Ca}_v3.2$ channels when tested amongst different Ca_v family subunits (McArthur *et al.*, 2018).

In comparison to voltage-gated Na^+ and Ca^{2+} subtypes, K^+ channels have received less attention; however, openers of M channels have been proposed as analgesics (Du *et al.*, 2018). Here, **retigabine** lacked efficacy in trials for postherpetic neuralgia; however, the less potent analogue **flupirtine** has been used to treat cancer pain, menstrual pain and post-surgical pain since the 1980s in several European countries. In addition, K2P channels represent potential targets, with the rationale that K2P openers may stabilize aberrant depolarization during chronic pain. Here, high-throughput screening has identified GI-530159 as one of the first TREK subtype activators (Loucif *et al.*, 2018). Another current area of focus is the targeting of TRP channels, which represent key players in transduction of sensory signals. Initial studies on TRPV1

antagonists were hampered by thermoregulatory side effects; however, capsaicin, delivered *via* patches, has shown therapeutic value *via* agonist (and subsequent local sensory nerve terminal retraction) actions. Modulation of TRPV1 channels by endogenous agents such as the lipid precursor N-arachidonoyl phosphatidylethanolamine (Nerandzic *et al.*, 2018) may open further avenues for drug development. Small molecule development has led to trials of antagonists of TRPA1 and **TRPM8** channels, while preclinical promise has also been shown for agents targeting **TRPV3**, **TRPV4**, **TRPM2** and **TRPM3** (Moran and Szallasi, 2018). Progress is also being made in the pharmacological targeting of ASIC subtypes, an area where initial ligands were largely non-selective and lacked potency. Characterization of agents such as diminazene and **APETx2** and demonstration of their efficacy in preclinical models of pain augur well for future development (Lee *et al.*, 2018). Finally here, a P2X3 receptor antagonist (**AF-219**) is currently in clinical trials to alleviate chronic cough and possibly chronic visceral pain (Abdulqawi *et al.*, 2015).

Overall, the search for new analgesics is very much an ongoing priority for drug development in the pharmaceutical industry and academia. Notably, targets arising from this themed section include $\text{Na}_v1.7$, $\text{Ca}_v2.2$ and $\text{Ca}_v3.2$, TREK and K_v7 channels, as well as specific TRP and ASIC isoforms. The key challenge is the translation of impressive results in animal models of chronic pain to the human condition. Work in this themed section provides hope for patients with chronic pain that their clinical needs will be better met in the next few years.

Conflict of interest

E.B.S. wishes to acknowledge a conflict of interest and highlight that he is a co-author on the paper by Loucif *et al.* (2018) and has co-authored papers with Stephen Waxman. G.J.S. has co-authored papers with Anthony Dickenson, Damian Bell and Mark Dallas.

References

- Abdulqawi R, Dockry R, Holt K, Layton G, McCarthy BG, Ford AP *et al.* (2015). P2X3 receptor antagonist (AF-219) in refractory chronic cough: a randomised, double-blind, placebo-controlled phase 2 study. *Lancet* 385: 1198–1205.
- Alshawaf AJ, Viventi S, Qiu W, D'Abaco G, Nayagam B, Erlichster M *et al.* (2018). Phenotypic and functional characterization of peripheral sensory neurons derived from human embryonic stem cells. *Sci Rep* 8: 603.
- Bauer CS, Rahman W, Tran-van-Minh A, Lujan R, Dickenson AH, Dolphin AC (2010). The anti-allodynic $\alpha2\delta$ ligand pregabalin inhibits the trafficking of the calcium channel $\alpha2\delta$ -1 subunit to presynaptic terminals in vivo. *Biochem Soc Trans* 38: 525–528.
- Bell DC, Dallas ML (2018). Using automated patch clamp electrophysiology platforms in pain-related ion channel research: insights from industry and academia. *Br J Pharmacol* 175: 2312–2321.

- Bernier L-P, Ase AR, Séguéla P (2018). P2X receptor channels in chronic pain pathways. *Br J Pharmacol* 175: 2219–2230.
- Boadas-Vaello P, Castany S, Homs J, Álvarez-Pérez B, Deulofeu M, Verdú E (2016). Neuroplasticity of ascending and descending pathways after somatosensory system injury: reviewing knowledge to identify neuropathic pain therapeutic targets. *Spinal Cord* 54: 330–340.
- Cao L, McDonnell A, Nitzsche A, Alexandrou A, Saintot PP, Loucif AJ *et al.* (2016). Pharmacological reversal of a pain phenotype in iPSC-derived sensory neurons and patients with inherited erythromelalgia. *Sci Transl Med* 8: 335ra56.
- Cardoso FC, Lewis RJ (2018). Sodium channels and pain: from toxins to therapies. *Br J Pharmacol* 175: 2138–2157.
- Castro J, Grundy L, Deiteren A, Harrington AM, O'Donnell T, Maddern J *et al.* (2018). Cyclic analogues of α -conotoxin Vc1.1 inhibit colonic nociceptors and provide analgesia in a mouse model of chronic abdominal pain. *Br J Pharmacol* 175: 2384–2398.
- Choi S, Yu E, Hwang E, Llinás RR (2016). Pathophysiological implication of $\text{Ca}_v3.1$ T-type Ca^{2+} channels in trigeminal neuropathic pain. *Proc Natl Acad Sci USA* 113: 2270–2275.
- Cox JJ, Reimann F, Nicholas AK, Thornton G, Roberts E, Springell K *et al.* (2006). An SCN9A channelopathy causes congenital inability to experience pain. *Nature* 444: 894–898.
- Deval E, Lingueglia E (2015). Acid-sensing ion channels and nociception in the peripheral and central nervous systems. *Neuropharmacology* 94: 49–57.
- Donnell AM, Collins S, Ali Z, Iavarone L, Surujbally R, Kirby S *et al.* (2018). Efficacy of the Nav1.7 Blocker Pf-05089771 in a randomised, placebo-controlled, double-blind clinical study in subjects with painful diabetic peripheral neuropathy. *Pain*. <https://doi.org/10.1097/j.pain.0000000000001227>
- Du X, Gao H, Jaffe D, Zhang H, Gamper N (2018). M-type K^+ channels in peripheral nociceptive pathways. *Br J Pharmacol* 175: 2158–2172.
- François A, Schüetter N, Laffray S, Sanguesa J, Pizzoccaro A, Dubel S *et al.* (2015). The low-threshold calcium channel $\text{Ca}_v3.2$ determines low-threshold mechanoreceptor function. *Cell Rep* S2211–S1247.
- Gong N, Park J, Luo ZD (2018). Injury-induced maladaptation and dysregulation of calcium channel $\alpha_2\delta$ subunit proteins and its contribution to neuropathic pain development. *Br J Pharmacol* 175: 2231–2243.
- Haley JE, Dickenson AH (2016). Evidence for spinal N-methyl-D-aspartate receptor involvement in prolonged chemical nociception in the rat. *Brain Res* 1645: 58–60.
- Jacus MO, Uebele VN, Renger JJ, Todorovic SM (2012). Presynaptic $\text{Ca}_v3.2$ channels regulate excitatory neurotransmission in nociceptive dorsal horn neurons. *J Neurosci* 32: 9374–9382.
- Lee JYP, Saez NJ, Cristofori-Armstrong B, Anangi R, King GF, Smith MT *et al.* (2018). Inhibition of acid-sensing ion channels by diminazene and APETx2 evoke partial and highly variable antihyperalgesia in a rat model of inflammatory pain. *Br J Pharmacol* 175: 2204–2218.
- Loucif AJC, Saintot P-P, Liu J, Antonio BM, Zellmer SG, Yoger K *et al.* (2018). GI-530159, a novel, selective, mechanosensitive two-pore-domain potassium (K_{2P}) channel opener, reduces rat dorsal root ganglion neuron excitability. *Br J Pharmacol* 175: 2272–2283.
- Mathie A, Veale EL (2015). Two-pore domain potassium channels: potential therapeutic targets for the treatment of pain. *Pflugers Arch* 467: 931–943.
- McArthur JR, Motin L, Gleeson EC, Spiller S, Lewis RJ, Duggan PJ *et al.* (2018). Inhibition of human N- and T-type calcium channels by an *ortho*-phenoxyanilide derivative, MONIRO-1. *Br J Pharmacol* 175: 2284–2295.
- Moran MM, Szallasi A (2018). Targeting nociceptive transient receptor potential channels to treat chronic pain: current state of the field. *Br J Pharmacol* 175: 2185–2203.
- Mourou A, Herold C, Kienzler MA, Kramer RH (2018). Understanding and improving photo-control of ion channels in nociceptors with azobenzene photo-switches. *Br J Pharmacol* 175: 2296–2311.
- Moutal A, Li W, Wang Y, Ju W, Luo S, Cai S *et al.* (2018). Homology-guided mutational analysis reveals the functional requirements for antinociceptive specificity of collapsin response mediator protein 2-derived peptides. *Br J Pharmacol* 175: 2244–2260.
- Nerandzic V, Mrozkova P, Adamek P, Spicarova D, Nagy I, Palecek J (2018). Peripheral inflammation affects modulation of nociceptive synaptic transmission in the spinal cord induced by N-arachidonoylphosphatidylethanolamine. *Br J Pharmacol* 175: 2322–2336.
- Patel R, Montagut-Bordas C, Dickenson AH (2018). Calcium channel modulation as a target in chronic pain control. *Br J Pharmacol* 175: 2173–2184.
- Peng C, Li L, Zhang MD, Bengtsson Gonzales C, Parisien M, Belfer I *et al.* (2017). miR-183 cluster scales mechanical pain sensitivity by regulating basal and neuropathic pain genes. *Science* 356: 1168–1171.
- Roca-Lapirot O, Radwani H, Aby F, Nagy F, Landry M, Fossat P (2018). Calcium signalling through L-type calcium channels: role in pathophysiology of spinal nociceptive transmission. *Br J Pharmacol* 175: 2362–2374.
- Snutch TP, Zamponi GW (2018). Recent advances in the development of T-type calcium channel blockers for pain intervention. *Br J Pharmacol* 175: 2375–2383.
- Talley E, Cribbs LL, Lee JH, Daud A, Perez-Reyes E, Bayliss DA (1999). Differential distribution of three members of a gene family encoding low voltage-activated (T-type) calcium channels. *J Neurosci* 19: 1895–1911.
- Usoskin D, Furlan A, Islam S, Abdo H, Lönnnerberg P, Lou D *et al.* (2015). Unbiased classification of sensory neuron types by large-scale single-cell RNA sequencing. *Nat Neurosci* 18: 145–153.
- Waxman SG, Zamponi GW (2014). Regulating excitability of peripheral afferents: emerging ion channel targets. *Nat Neurosci* 17: 153–163.
- Winters BL, Rawling T, Vandenberg RJ, Christie MJ, Bhola RF, Imlach WL (2018). Activity of novel lipid glycine transporter inhibitors on synaptic signalling in the dorsal horn of the spinal cord. *Br J Pharmacol* 175: 2337–2347.
- Yang Y, Adi T, Efrain PR, Chen L, Dib-Hajj SD, Waxman SG (2018). Reverse pharmacogenomics: carbamazepine normalizes activation and attenuates thermal hyperexcitability of sensory neurons due to $\text{Na}_v1.7$ mutation I234T. *Br J Pharmacol* 175: 2261–2271.
- Yu YP, Gong N, Kweon TD, Vo B, Luo ZD (2018). Gabapentin prevents synaptogenesis between sensory and spinal cord neurons induced by thrombospondin-4 acting on pre-synaptic $\text{Ca}_v\alpha_2\delta_1$ subunits and involving T-type Ca^{2+} channels. *Br J Pharmacol* 175: 2348–2361.
- Zeilhofer HU, Wildner H, Yévenes GE (2012). Fast synaptic inhibition in spinal sensory processing and pain control. *Physiol Rev* 92: 193–235.