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Targeting TRP Channels For Novel Migraine Therapeutics

Gregory Dussor,[†] J. Yan,[‡] Jennifer Y. Xie,[§] Michael H. Ossipov,[§] David W. Dodick,[∥] and Frank Porreca*[,§]

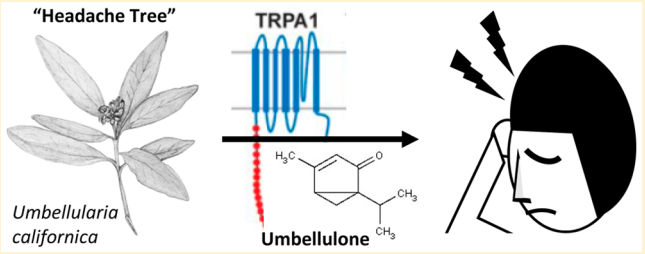
[†]School of Behavioral and Brain Sciences, University of Texas at Dallas, Dallas, Texas 75080, United States

[‡]Department of Pharmacology, University of Washington, Seattle, Washington 98195, United States

[§]Department of Pharmacology, University of Arizona College of Medicine, Tucson, Arizona 85724, United States

[∥]Department of Neurology, Mayo Clinic Arizona, Phoenix, Arizona 85054, United States

ABSTRACT: Migraine is increasingly understood to be a disorder of the brain. In susceptible individuals, a variety of "triggers" may influence altered central excitability, resulting in the activation and sensitization of trigeminal nociceptive afferents surrounding blood vessels (i.e., the trigeminovascular system), leading to migraine pain. Transient receptor potential (TRP) channels are expressed in a subset of dural afferents, including those containing calcitonin gene related peptide (CGRP). Activation of TRP channels promotes excitation of nociceptive afferent fibers and potentially lead to pain. In addition to pain, allodynia to mechanical and cold stimuli can result from sensitization of both peripheral afferents and of central pain pathways. TRP channels respond to a variety of endogenous conditions including chemical mediators and low pH. These channels can be activated by exogenous stimuli including a wide range of chemical and environmental irritants, some of which have been demonstrated to trigger migraine in humans. Activation of TRP channels can elicit CGRP release, and blocking the effects of CGRP through receptor antagonism or antibody strategies has been demonstrated to be effective in the treatment of migraine. Identification of approaches that can prevent activation of TRP channels provides an additional novel strategy for discovery of migraine therapeutics.



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Migraine is a common neurological disorder that ranks among the leading causes of disability in the world. According to the recent Global Burden of Disease Survey 2010 conducted by the World Health Organization,[1] migraine, with a global prevalence of 14.7%, ranked as the third most common disease in the world.[2] When considered on the basis of years lost to disability (YLDs), migraine accounts for 2.9% of all YLDs, placing it seventh among specific causes of disability.[2] Among neurologic disorders, migraine is responsible for more than one-half of YLDs, thus making it the leading neurological disorder causing disability.[2] Notably, these rankings of disease burden also take into consideration the treatments that are available, and in use, underscoring the inadequate treatment of headache disorders.[2,3] The introduction of triptans represented a remarkable advance in medical management of migraine. Triptans, however, are effective in only a subset of migraineurs, and few appreciable advances in available therapeutics for migraine have been introduced in the last 20 years.[4] Migraine remains a critical unmet medical need. Recent advances in our understanding of migraine pathophysiology point to several avenues that could lead to the discovery of novel therapies.

■ ACUTE MIGRAINE

A migraine episode generally consists of four phases, beginning with the premonitory phase.[5] This initial phase of migraine, reported by a majority of migraine sufferers, can appear hours to days prior to the headache itself and may include symptoms such as sore neck, tiredness, and difficulty in concentrating.[5] A sizable minority of migraineurs (15 to 30%) also experience aura up to 1 h prior to the migraine headache.[5−8] The aura can include visual disturbances with expanding regions of scintillations or light accompanied by some vision loss termed scotomas. The scotomas normally begin in the center of the visual field and appear to progress toward the periphery.[6] Aura may also present with somatosensory, motor, or language disturbances. Aura typically develops slowly over 5−20 min and lasts under 1 h.[5] The majority of migraineurs experience migraine without aura (previously "common migraine"). The migraine headache is usually characterized by a moderate to severe throbbing headache with unilateral localization.[9] Migraine headache may be exacerbated by movement of the head or by physical activity. It is often accompanied by sensory excitability, expressed as sensitivity to light (photophobia), noise (phonophobia), or smells (osmophobia) as well as nausea and vomiting.[6,7] It can last from 4 to 72 h if left untreated.[5−7] Migraine may be followed by a postdrome phase lasting hours or even days and often consists of fatigue, irritability, sensory excitability, and impaired concen-

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tration, although some individuals may feel euphoric after migraine.[5]

In spite of the significant socioeconomic impact of migraine, the effective management of acute migraine remains an important unmet medical need. The most commonly employed acute therapies are the triptans and nonsteroidal anti-inflammatory drugs (NSAIDs),[4,10] with triptans accounting for up to 80% of medications prescribed for migraine.[11] This class of drugs represents the therapy of choice for acute treatment of moderate or severe migraine headache.[12] However, triptans are not always effective, as fewer than one-half of patients taking oral triptans report being pain-free at 2 h, and up to one-third of these individuals report headache recurrence within 24 h.[13,14] Moreover, triptans are contraindicated in patients with risk factors for cardiovascular disease or with hypertension,[15] which account for one-fifth of the migraine population. An important clinical concern is that triptan overuse can lead to medication-overuse headache (MOH).[16,17] Consequently, it is recommended that acute treatments not be used more than twice per week, thus rendering these drugs inappropriate as preventive treatments.[18] Opioids in combination with NSAIDs or acetaminophen, and barbiturates are also used, principally in the United States. However, as few as 5−8 doses per month of either opioid or barbiturate containing medications may lead to medication overuse headache.[19] A recent review of randomized controlled trials (RCT) suggests that high-dose aspirin may be effective against acute migraine attacks,[20] and NSAIDs are effective against mild migraine episodes.[10] There are also few options available for the acute treatment of severe migraine in the emergency department setting.[21] Only parenteral prochlorperazine has a high level of evidence for acute treatment of migraine, although the side effects including akathesia, dyskinesia, and sedation and risk of QT interval prolongation indicate that dopamine antagonists are not ideal for intractable migraine pain. Lysine acetylsalicylic acid (not available in the United States) and metoclopramide have a moderate level of evidence, while only a low level of evidence exists to support the use of intravenous ketorolac.[22]

CHRONIC MIGRAINE AND MEDICATION OVERUSE HEADACHE

The rate of occurrences of migraine episodes form a continuum ranging from low-frequency episodic migraine to high-frequency episodic migraine and culminating in chronic migraine.[23,24] Chronic migraine is defined by the International Headache Society (IHS) as headaches that occur 15 or more days per month over a period of 3 or more months.[17] In addition, headaches occurring on at least 8 of those days must meet the criteria for migraine with aura, migraine without aura, and/or be perceived by the patient to be migraine at onset and be relieved by a triptan or ergot derivative.[17] It has been estimated that approximately 14% of patients with episodic migraine will develop chronic migraine, representing 1.3−5.1% of the global population.[23−25] As would be expected, chronic migraine is more disabling than episodic migraine, owing to the high frequency of attacks and accompanying comorbidities associated with this condition.[23−25] Recent epidemiologic studies identified possible risk factors for the progression to chronic migraine that include female sex, age, low education, low socioeconomic status, and head injury.[26−28] Risk factors considered to be modifiable include stressful life events, sleep disturbances, obesity, depression, increased caffeine consumption, and elevated baseline headache frequency (10 or more per month).[23,26−28]

The excessive use of acute migraine medications represents an especially important risk factor, leading to chronic migraine and medication overuse.[23] Approximately 50−75% of patients diagnosed with chronic migraine have a history of overuse of medications, in particular, triptans, analgesics, and barbiturates.[23,29−32] Medication overuse headache is diagnosed in those with 15 or more headache days per month and the consumption of simple analgesics (acetaminophen, NSAIDs) for more than 15 days per month or the intake of combination analgesics, opioids, ergots, or triptans on more than 10 days per month.[23] Clearly, the acute treatment of frequent migraine episodes is not appropriate for chronic migraine, and prophylactic therapies to manage migraine are needed.

The mechanisms that lead to development of chronic migraine are still not well understood. Consequently, the development of prophylactic agents against this condition is based on treatments that were discovered serendipitously and/or because efficacy was presumed based on pharmacologic class.[33−36] Notably, even within class, some drugs are effective (e.g., antiepileptics such as valproate and topiramate)[13,37,38] while others (e.g., oxcarbazepine) are not.[39] Moreover, many of these treatments are associated with troublesome side effects, including sedation, difficulty in thinking, and weight gain, and are not well tolerated in many patients. Because many patients do not respond to current prophylactic medications, a strong medical need for discovery of new therapeutics exists.[13,25,38,40] In addition to antiepileptic drugs such as valproate and topiramate, onabotulinumtoxinA has been shown to be effective in migraine prophylaxis[23,41] Double-blind, placebo-controlled trials showed reduced frequency of migraine headaches and improvement in "functioning, vitality, psychological distress, and overall quality of life".[41]

PATHOPHYSIOLOGY OF MIGRAINE

A critical impediment to the development of effective and well-tolerated therapeutics for migraine management has been a relative lack of understanding of the molecular, biochemical, and physiological mechanisms of migraine.[34,35] Migraine was initially thought to be primarily of vascular origin, since antimigraine therapies had potent vasoconstrictor properties.[42] However, it is becoming increasingly evident that vasodilation of dural vessels during migraine is an epiphenomenon unrelated to the genesis of migraine.[43,44] Functional imaging studies have shown that migraine headache can occur in the absence of vasodilation, and appears driven from sites within the brain.[45,46] Migraine is now considered to be a neurovascular disorder, where activation and sensitization of primary afferent neurons that innervate the dural vasculature can promote the headache phase.[6,7,47,48] Although the events that actually initiate a migraine attack remain unknown, the ultimate activation of the trigeminovascular system is considered to be essential.[49,50] Trigeminovascular activation may provoke release of multiple excitatory neurotransmitters including substance P, neurokinin A, and CGRP from dural afferent terminals, resulting in neurogenic vasodilation of dural blood vessels, release of proinflammatory mediators, degranulation of mast cells and plasma protein extravasation.[7,51,52] CGRP has been shown to be released in humans during migraine headache and in animal models following dural stimulation (see below[53,54]). Activation and sensitization of thinly myelinated and unmyelinated nociceptive afferent fibers that innervate the dura can elicit pain.[49,50] Additionally, sensitization of the second-order neurons of the trigeminal n. caudalis[49,50] can occur resulting in enhanced

nociceptive inputs to higher brain centers including the thalamus, hypothalamus and cortical sites, collectively manifesting as migraine pain.[49,50,55,56] Peripheral sensitization and central sensitization[57,58] amplify signaling along the entire pain transmission pathways. Evidence supports the clinical relevance of centrally sensitized pathways in migraine[59,60] and drugs clinically effective in migraine, such as NSAIDs, may elicit their actions primarily through central sites.[61] As perivascular stimulation of the dura results in pain referred to the head,[7,62] many animal models have applied artificial inflammatory stimuli to the dura to activate and sensitize afferent fibers.[63−65] Electrical stimulation of the trigeminal ganglion causes plasma extravasation and mast cell degranulation,[66,67] which is blocked by sumatriptan. Unilateral extravasation of proteins has been demonstrated during migraine attack,[68] but experimental antimigraine drugs designed to block extravasation, such as an NK1 antagonist,[69] an endothelin antagonist,[70] and a selective blocker of plasma extravasation,[71] all failed against migraine headache in clinical trials.[7]

Considerable evidence suggests that calcitonin gene-related peptide (CGRP) plays a cardinal role in migraine headache. Blood levels of CGRP, but not of substance P, have been reported to be elevated during migraine attacks,[53] though this finding was not confirmed by all studies.[72] Additionally, the intravenous infusion of CGRP[73] produces migraine headache in migraineurs, but not in normal volunteers.[74] Precipitating migraine in susceptible individuals by administration of nitroglycerin causes elevations in levels of CGRP in the jugular venous blood,[75−77] although it should be noted that inhibiting CGRP does not block nitroglycerin-induced migraine.[78] It is likely that CGRP does not directly activate trigeminal dural afferents but potentiates the release of agents into the perivascular space, possibly because CGRP can act as a mast-cell degranulator and mast cell degranulation has been proposed to contribute to migraine.[79−82] Although the development of first-generation triptans was based on their vasoconstrictor properties, triptans likely act by inhibiting the release of pronociceptive transmitters, including CGRP, by activation of $5HT1_{B/D}$ presynaptic receptors on either the peripheral or central terminals of trigeminal afferents.[60,83−86] Clinical investigations have now demonstrated that CGRP receptor antagonists are as efficacious as triptans against migraine.[87,88] Their site of action however remains to be determined. CGRP receptor antagonists did not have activity in preclinical models when given into the trigeminal ganglia or onto the dura,[89,90] but can inhibit activity of second-order neurons when given into central sites in the nucleus caudalis.[91] However, this is in contrast to recent reports of efficacy for CGRP antibodies for migraine,[92] which likely do not gain access to the CNS and thus work in the periphery. These observations nonetheless indicate that modulation of trigeminal dural afferents, or of their postsynaptic pathways, can provide effective therapy for migraine, raising possibilities for the discovery of new medications that may provide efficacy in specific populations of migraineurs.

TRANSIENT RECEPTOR POTENTIAL (TRP) CHANNELS AND HEADACHE

TRP channels are a large family of membrane ion channels that have been implicated in a variety of pain states due to their response to stimuli such as temperature, changes in extracellular osmolarity, pH, and an extensive list of natural products.[93−95] There are six subfamilies of TRP channels, designated with different lettering systems and grouped according to their primary amino acid sequences including TRPC, TRPM, TRPV, TRPA, TRPP, and TRPML.[96,97] TRP channels are nonselective ion channels and contribute to membrane depolarization and activation of second messenger signaling cascades due to the influx of Na^+ and Ca^{2+}.[93] They are proposed to contribute to an extensive list of sensory encoding including processes in the visual, gustatory, auditory, and somatosensory systems.[98] A growing list of studies implicates TRP channels in the pathophysiology of headache and suggests that this family of channels might represent novel targets for headache therapeutics.

TRANSIENT RECEPTOR POTENTIAL V1 (TRPV1)

TRPV1 is highly expressed on peripheral nociceptors;[99,100] its expression in the central nervous system, however, is controversial. Neuronal TRPV1 expression has been reported[101] to be largely restricted to nociceptors in primary sensory ganglia, with CNS expression limited to only discrete brain regions including the caudal hypothalamus, consistent with a role of this channel in thermoregulation (see below). The restricted peripheral distribution was conserved across species in rat, monkey, and human brain.[101] The TRPV1 channel is activated by noxious heat and by chemicals such as capsaicin, an extract of chili peppers that produces burning sensations in humans.[102] Its activity is potentiated by low extracellular pH, implicating this channel in pain due to ischemic and inflammatory events.[103] TRPV1 channels are therefore transducers that can result in activation of nociceptors that can produce pain.[104,105] Importantly, the TRPV1 channel is also a molecular integrator of nociceptive signaling. Activation of multiple transducers found on nociceptors including bradykinin, serotonin, prostaglandin, and prokineticin receptors can result in activation of intracellular signaling pathways to sensitize the channel so that it is responsive at lower stimulus intensities.[106] It should also be noted that TRPV1 is distributed on the central terminals of nociceptive fibers where its activity may promote activation and sensitization of postsynaptic pain transmission pathways.[107−109]

Relevant to migraine, single nucleotide polymorphisms have been found in the TRPV1 gene in Spanish migraine patients but how these channel mutations may contribute to migraine is not clear.[110] Preclinically, TRPV1 has been shown on dural nerve fibers[111] as well as on trigeminal ganglion cell bodies retrogradely labeled from the dura.[112] Capsaicin can dilate dural vessels in a TRPV1-dependent mechanism,[113] and application of capsaicin to the rat dura produces ERK activation in trigeminal ganglion neurons[114] and behavioral responses consistent with headache.[115,116] Sumatriptan can also inhibit the TRPV1 channel[117] as well as attenuate the positive modulation of TRPV1-mediated behavioral responses by 5HT.[118] Modulation of TRPV1 may therefore be among the mechanisms by which sumatriptan has efficacy for migraine. Mechanisms contributing to activation of TRPV1 within the dura before or during a migraine are less well-defined. The channel is known to be activated by endocannabinoids such as anandamide,[119] endovanilloids such as *N*-arachidonoyl dopamine (NADA),[120] and various lipid products of the lipoxygenase pathway,[121] as well as modulated by bradykinin,[122] nerve-growth factor,[122] and prostaglandins.[123] Whether activation/modulation of dural TRPV1 by any of these substances contributes to migraine has yet to be determined.

The potential of TRPV1 for discovery of therapeutics has been an intense focus of the pharmaceutical industry since the molecular identification of the channel.[102] The structure of the channel along with its activation mechanisms have recently been revealed using electron cryomicroscopy[124,125] which may aid in

the development of novel agents targeting the channel. Potential therapies emanating from targeting TRPV1 include pain, urge incontinence, asthma, cough, irritable bowel syndrome, and others (see refs 126 and 127). Activation of TRPV1 initially produces sensations of heat and pain but can lead additionally to long lasting desensitization of afferent fibers preventing nociceptive signaling. The desensitization strategy has been exploited in numerous TRPV1 therapeutics including capsaicin cream (Zostrix) as well as high concentration patches (Qutenza). An alternate strategy is to block transduction at the channel with antagonists. Unlike agonists that promote inhibition of function in the entire nociceptor through desensitization,[127] TRPV1 antagonists only block activation of the channel and do not prevent the actions of other potential pronociceptive mediators. Multiple molecules have been developed as TRPV1 antagonists,[128,129] and some of these have advanced to human trials (e.g., refs 130 and 131). These studies have identified important potential limitations[132] to the use of TRPV1 antagonists including hyperthermia and blockade of thermosensation that may be dangerous to activities of daily living in patients.[133] Studies are onging to develop TRPV1 antagonists that may block the channel without producing a blockade of thermosensation or of hyperthermia.[127]

TRPV1-based therapies have nonetheless been developed for migraine. The intranasal TRPV1 agonist Civamide showed efficacy for migraine and reduces the frequency of cluster headache attacks.[134] Intranasal capsaicin also showed efficacy for migraine,[135] although the site of action is unknown. However, side effects due to TRPV1 activation within the nasal cavity[134] may ultimately limit the clinical potential of these agents. TRPV1 antagonists have also been tested in several preclinical models related to headache, but the results have been inconsistent. The TRPV1 antagonist capsazepine given systemically blocked dural vessel dilation due to capsaicin but not electrical stimulation,[113] while another TRPV1 antagonist, SB-705498, also given systemically, decreased neuronal activity in the n. caudalis following stimulation of the dura with mechanical, electrical, and chemical stimuli.[136] In contrast, systemic administration of the TRPV1 antagonist A993610 had no effect in similar experiments.[137] In a recent phase II clinical study, the TRPV1 antagonist SB-705498 was inferior to placebo against migraine headache, photophobia, and phonophobia.[138] Ultimately, the future of TRPV1-based therapies for migraine and other applications is unclear.

TRANSIENT RECEPTOR POTENTIAL V4 (TRPV4)

Dural afferents have been shown in many studies to be mechanically sensitive,[139−142] and they are also activated by changes in extracellular osmolarity,[140] implicating the mechano- and osmo-sensitive channel TRPV4[143−146] in processes contributing to headache. TRPV4 mRNA is expressed in trigeminal ganglion neurons,[147] and functional effects of TRPV4 activation can be measured in trigeminal neurons in vitro.[148,149] Mice lacking TRPV4 exhibited a loss in both osmotic and pressure sensation.[150,151] TRPV4 can be sensitized by pro-inflammatory mediators, and its mechanosensitivity may be enhanced under inflammatory conditions. Administration of inflammatory soup into the paw results in hypersensitivity to osmotic and mechanical stimulation that is present in wild-type but not TRPV4-null mice.[152] Intradermal injection of mildly *hypertonic* solutions causes paw flinching in rats that is increased 7-fold following sensitization with PGE_2.[153] This effect is decreased following antisense-mediated knockdown of TRPV4 expression and is not observed in TRPV4 knockout mice.[154] *Hypotonic* solutions excite 54% of saphenous nerve c-fibers in vivo following sensitization by PGE_2, an effect that is decreased by antisense knockdown of TRPV4 expression.[155] This effect is likely mediated via direct actions on neurons, since consistent effects are observed using intracellular Ca^{2+} measurements in trigeminal ganglion cultures taken from TRPV4 wild-type but not TRPV4 knockout mice.[156] Finally, support for a role of TRPV4 in mechanosensation in sensitized states is provided by a direct interaction between TRPV4, α1 integrin, and Src tyrosine kinase, which are additional factors thought to be important for mechanosensation.[152]

Other studies have indirectly implicated TRPV4 in migraine. Sensitization of threshold mechanical responses of dural afferents in vivo was found following activation of the protease-activated receptor 2 (PAR2).[157] PAR2 is activated by its N-terminus, which is cleaved by extracellular proteases including tryptase. One likely source of these proteases (in addition to other pro-inflammatory mediators) is mast cells, which have been previously implicated in the pathogenesis of migraine.[51,158,159] Consistent with a downstream role for TRPV4 following mast-cell degranulation and release of proteases is a prior study indicating that PAR2 agonists sensitize TRPV4 currents in DRG neurons and lead to mechanical hyperalgesia in the hindpaw and hypersensitivity to colorectal distension that is dependent on expression of this channel.[160−162]

The most direct evidence supporting a role for TRPV4 in headache thus far comes from a recent study that found TRPV4-like currents on retrogradely labeled dural afferents in response to hypotonic solutions and the TRPV4 activator 4αPDD.[163] Activation of dural TRPV4 in rats also produced behavioral responses consistent with headache.[163] Whether changes in osmolarity contribute to headache is not known but changes in intracranial pressure due to coughing, sneezing, routine physical activity, or simple changes in position or posture as well as mechanical stimulation of the dura following sudden head movements is known to worsen headache in migraine patients.[59] TRPV4 may contribute to these responses and thus TRPV4-based therapies may provide relief to mechanically induced pain in migraine patients but this awaits further investigation.

TRANSIENT RECEPTOR POTENTIAL MELASTATIN 8 (TRPM8)

TRPM8 is activated by cool temperatures (below 26 °C) and also by the chemical activators menthol and icilin. The mRNA for TRPM8 is expressed in a small fraction of sensory neurons,[164,165] including those of the trigeminal ganglion,[166] and mice deficient in TRPM8 have deficits in cold sensation, implicating this channel as the endogenous sensor of external cold temperature.[167−169] Trigeminal expression of TRPM8 mediates sensory input from cold stimuli in and around the oral cavity[170] and was shown to mediate sensitivity to volatile odorants.[171] However, TRPM8 is also expressed on sensory afferents innervating deep tissues such as the colon and bladder[172,173] that are not exposed to cold temperatures, suggesting an endogenous sensory role for this channel. The endogenous activator or sensitizer is unclear but may be various lipid mediators or the growth factor artemin.[174,175] What information is signaled by TRPM8-expressing neurons in response to these mediators is not yet known.

Little preclinical data exists thus far for a role of TRPM8 in headache and the only studies performed have examined expression, but not function, of TRPM8 on dural afferents.

One study found minimal expression of TRPM8 on dural afferents,[112] while another found region-specific innervation,[176] so whether and where TRPM8 is expressed on these afferents is still unclear. Despite the lack of preclinical data, TRPM8 has turned out to be one of the most consistent findings among the genome-wide association studies performed on migraine patients. TRPM8 gene variants have been found in seven separate groups of migraine patients,[177−182] making this one of the most replicated findings in all of the genetic analyses performed on humans to date. To date, it is unknown how these genetic variants alter channel function or expression and what role this may play in migraine. The mutations are found both in/around coding regions as well as in the 5′ untranslated regions (5′UTR), so it is not known whether they impact channel function or expression. These data nonetheless provide much excitement for future TRPM8-based migraine therapeutics.

A question that has yet to be answered is whether future therapeutics should be TRPM8 agonists or antagonists. This likely depends on how the mutations impact channel function/expression, that is, whether channel expression/function is increased or decreased. Adding to this uncertainty is that TRPM8 is implicated as a sensor of environmental cold (as well as cold allodynia following nerve injury, see also ref 183), but activation of TRPM8 is required for the well-known property of agonists such as menthol to be analgesic.[184] Thus, TRPM8 activation can be either pro- or antinociceptive and it is not yet clear which approach would be more likely have potential efficacy for migraine. Future work further clarifying how TRPM8 mutations impact the channel may provide better clues to which direction pharmacological approaches should take, activating or blocking channel function.

■ TRANSIENT RECEPTOR POTENTIAL A1 (TRPA1)

One of the most intriguing TRP channels proposed to contribute to migraine is TRPA1. This channel was originally named ANKTM1 due to the presence of numerous N-terminal ankyrin repeats in the protein[185] but was later renamed as the only member of the TRPA subfamily. There are numerous cysteine residues in these ankyrin repeats at the N-terminus[186−189] that appear to contribute to activation of the channel when covalently modified by various substances (see below for examples). TRPA1 is expressed on peripheral sensory neurons,[190] including those that innervate the colon[191] and neurons innervating the airways that signal irritation of the respiratory system.[192,193] It is often coexpressed in sensory neurons with TRPV1, and like TRPV1 its activation can also promote the release of substance P and CGRP.[106] Although it was originally proposed to be a sensor of noxious cold,[185] this has been a controversial issue[194] and most studies now focus on a sensory role of this channel outside of temperature. TRPA1 has also been linked to mechanosensation, although it appears to mostly contribute to mechanical hypersensitivity of afferents after inflammation.[195] It should be noted that TRPA1 is expressed on the central terminals of primary afferents where its activation may enhance release of transmitter from afferents.[196] Activation of peripheral sensory neurons is thought to release both lipoxygenase and cytochrome P450-epoxygenase products at the central terminals and both can activate TRPA1.[197,198] Consistent with a role for central terminal TRPA1 in pain, intrathecal administration of TRPA1 antagonists has shown efficacy in several preclinical pain models.[196]

Numerous studies have focused on a role for TRPA1 and pain,[195] and it is now also believed to contribute to the sensation of itch.[199] Activation of the channel can be demonstrated by a variety of exogenous and endogenous substances including the environmental irritants formaldehyde,[200] acrolein,[201] chlorine,[202] and cigarette smoke extract,[203] natural products such as isothiocyanates,[190] cinnamaldehyde,[204] and allicin,[205] and endogenous byproducts of oxidative and nitrative stress such as nitro-oleic acid,[206] 4-hydroxynonenal,[207] and reactive prostaglandins.[208] It is reasonable to conclude that some of these TRPA1 activators may have a role in promoting migraine and, indeed, some of the environmental TRPA1 activators are well-known migraine triggers.[209−212] Interestingly, a gain-of-function mutation in TRPA1 has been identified in humans with familial episodic pain syndrome, a rare disorder characterized by pain in the upper limbs, often preceded by a prodrome phase, and triggered by fasting and physical stress.[213] Triggers for this disorder are among those commonly reported to trigger migraine, although headache is not reported in these patients. This study nonetheless links a mutation in TRPA1 to a pain state induced by common migraine triggers and suggests that there may be some role for the channel in other types of pain due to these stimuli.

There is a growing list of preclinical evidence implicating TRPA1 in the pathophysiology of headache. TRPA1 immunoreactivity[112] and functional TRPA1-like currents[214] have been found on identified dural afferents in rodents. Distribution of the channel has been reported in both unmyelinated and thinly myelinated axons with terminations in superficial lamina of the trigeminal nucleus caudalis providing an anatomical basis for TRPA1-dependent orofacial nociception.[215] Application of the TRPA1 agonist mustard oil as well as the environmental irritant acrolein to the nasal cavity can increase blood flow in the dura, an effect that is blocked by dural application of either a CGRP receptor or a TRPA1 antagonist.[216] These findings suggest that TRPA1 may contribute to environmental irritant-induced neuronal activation, and the mechanism may be via access of irritants to the meninges through the nasal route and subsequent activation of TRPA1 on meningeal afferents. However, these studies do not provide functional evidence that activation of meningeal TRPA1 contributes to headache. In a preclinical behavioral model of migraine, application of the TRPA1 activator mustard oil to the dura produced cutaneous facial and hindpaw allodynia as well as a decrease in exploratory behavior, both of which were blocked following oral administration of a TRPA1 antagonist.[214] Thus, preclinical data support the possibility that activation of TRPA1 within the dura contributes to increased dural blood flow, neurogenic vasodilation (via CGRP release), and headache.

A series of recent studies has taken a backward translation approach using the observation that exposure of susceptible individuals to the "headache tree" *Umbellularia californica* can trigger violent headache crises.[212] One of the major constituents of extracts from the headache tree is a substance known as umbellulone, which can evoke nociceptive behavior, meningeal vasodilation, and calcitonin gene-related peptide (CGRP) release via TRPA1 activation.[212] Using the same rat behavioral migraine model described above, dural application of umbellulone produced cutaneous facial and hindpaw allodynia as well as a decrease in exploratory behavior similar to that seen with mustard oil, and these responses were blocked by an oral TRPA1 antagonist.[214] The ability of umbellulone to gain access to the meninges via intranasal delivery in humans is unclear, but intranasal delivery of both mustard oil and umbellulone in preclinical studies caused meningeal vasodilatation due to TRPA1 activation.[201] Together with the studies described

above, these data argue that environmental irritants produce headaches via activation of TRPA1 in the meninges. They also suggest that endogenous activation of TRPA1 may contribute to headache and that TRPA1-based therapeutics may have efficacy for environmental irritant induced headaches as well as other headache disorders such as migraine.

A similar backward translation approach stemmed from the observation that the feverfew herb is commonly used as a migraine therapeutic in humans. One of the major constituents of feverfew, parthenolide, was recently identified as a partial agonist for TRPA1.[217] In contrast to other TRPA1 agonists, parthenolide can desensitize the channel, leading to a decrease in neuropeptide release from TRPA1-containing nerve endings, and pretreatment of rats with parthenolide attenuated subsequent headache-like responses to dural application of mustard oil,[217] indicating functional desensitization of the channel. These studies raise the question of whether antagonists or desensitizing agonists are a better therapeutic strategy, but in addition they further implicate TRPA1 in headache using preclinical experiments based on human observations.

CONCLUSION

Migraine is a disabling neurological disorder, ranking among the top 10 in causes of disability worldwide. In addition to the moderate to severe headache pain, heightened sensitivity to environmental sensory stimuli including light, sound, and odors, along with nausea and vomiting contribute to this often debilitating condition. For some individuals, migraine progresses to a chronic condition, where the afflicted individual can expect to have headaches more than half the time if not continuously. Clearly, this condition significantly impacts quality of life in a profound and negative way. Yet in spite of the serious impact of migraine, and especially chronic migraine, novel therapeutics against this disorder have not been introduced into the market recently, and chronic migraine is still treated on a trial by error basis with therapeutics discovered serendipitously.

The past decade has seen a veritable explosion of studies revealing potential mechanisms through which migraine may be generated, providing optimism that the discovery of new medicines will be possible. Migraine is no longer considered primarily a vascular disorder, but a brain disorder. Activation and sensitization of dural afferents likely occur resulting in release of mediators that maintain the sensitized state and sustain pain. It is now clear that blocking the actions of CGRP, released from dural afferent terminals and central endings in the brainstem, is effective in migraine therapy. More recent studies point to a potential role of the TRP channels in activation and sensitization of dural afferents as well as in postsynaptic pathways. A noted example demonstrating the prominent role of TRPA channels is how umbellulone from the "headache tree" can precipitate severe migraine in susceptible individuals. However, the ability of feverfew to treat headache, potentially via desensitization of TRPA1 with parthenolide, raises new questions. Namely, whether continual desensitization of TRPA1 with desensitizing agonists would provide more efficacy or better tolerability than a standard antagonist. This is particularly relevant, as it would inform whether these drugs are designed as prophylactic or abortive agents, for example, desensitizing agonists would more likely be developed as prophylactics. This question should be the subject of future work as the answer is not yet clear. Additionally, given that side effects may limit the therapeutic utility of TRPV1 antagonists, it is important to determine whether continual exposure to TRPA1-modulating drugs has adverse effects. If TRP-based therapies have efficacy equal to that of triptans and opiates, but they do not lead to MOH, they could gain an important place in the migraine therapeutic tool kit. Clearly, many questions remain. Nevertheless, as the role of the TRPA channels as well as possible roles of other TRP channels in producing or maintaining sensitization of trigeminal dural afferent nerves are elucidated, the potential for development of novel, selective, and safer medications against migraine may be realized.

AUTHOR INFORMATION

Corresponding Author

*Telephone: 520-626-7421. Fax: 520-626-4182. E-mail: frankp@u.arizona.edu.

Author Contributions

All authors contributed equally to the preparation of this manuscript.

Notes

The authors declare no competing financial interest.

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