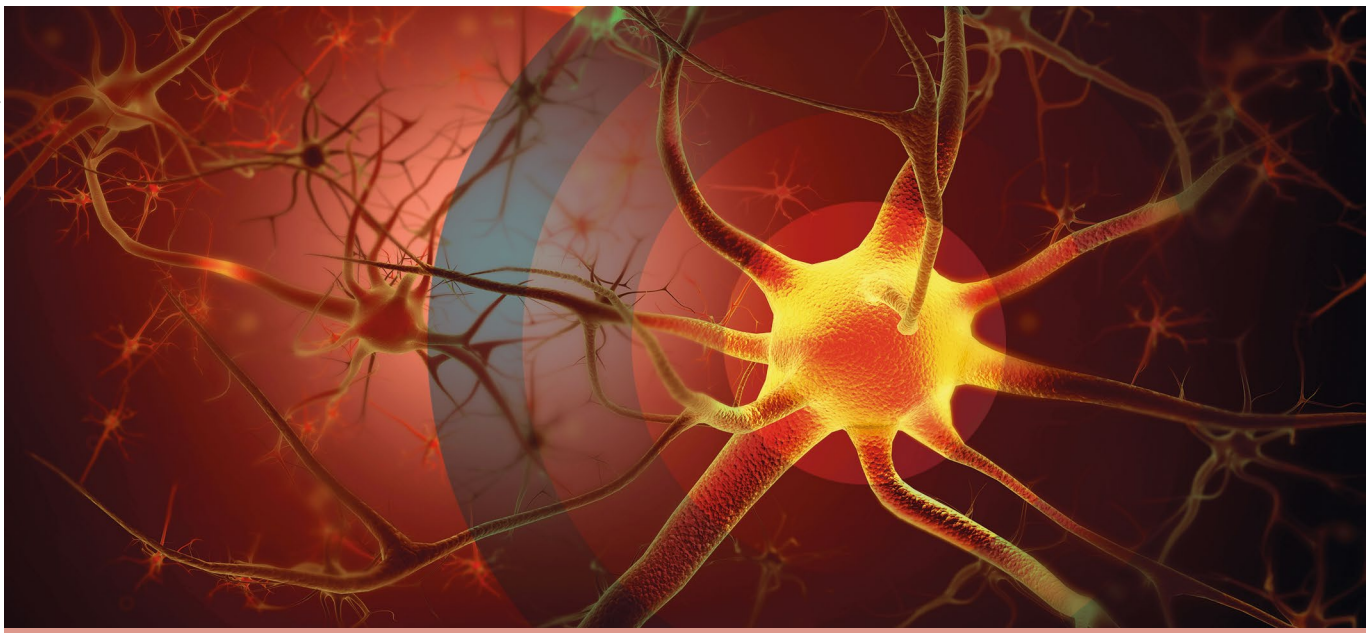

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Nav1.7 withholds its pain potential

Despite compelling genetic validation, drug developers are struggling to unlock the therapeutic promise of the Nav1.7 sodium channel as a pain target.

Katie Kingwell

When researchers reported in 2006 that patients with null mutations in the voltage-gated sodium channel Nav1.7 were impervious to pain, they kicked off an industry-wide hunt for novel analgesics. Genetic target validation was gaining traction as a means of improving drug development success rates, and Nav1.7 promised to be a poster child example of the drug development future. Instead, a slew of failures attest to how hard it can be to translate even the most compelling targets into therapeutics.

"Just because a target is genetically validated, it doesn't necessarily mean to say it's very druggable," says Andrea Houghton, executive director of pharmacology at Merck & Co. "Nav1.7 as a target is very hard to drug."

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The most recent setbacks were announced in October 2018. Biogen pulled its Nav1.7 blocker vixotrigine in painful lumbosacral radiculopathy following a failed phase II trial, leaving a question mark over the compound's chances in two other pain indications. Genentech meanwhile jettisoned its lead Nav1.7-targeted candidate, GDC-0310, licensed from Xenon Pharmaceuticals, prior to initiation of phase II testing (TABLE 1).

The biology of Nav1.7, as well as the inherent difficulties of developing pain drugs, is responsible for the headaches.

With the huge unmet need for new pain relief options, brought into sharp focus by the ongoing opioid crisis in the United States, many drug companies remain committed to Nav1.7. But they have tempered their views. "Originally there was a period of exuberance around the target. Now people have kind of taken a step back, realizing, 'OK, this is really difficult, but still possible,'" says David Hackos, senior scientist at Genentech.

Earlier this year, Houghton and others showcased continuing work on Nav1.7 inhibitors as alternatives for opioid analgesics at a symposium organised by the NIH's Helping to End Addiction Long-Term (HEAL) Initiative.

Manna from heaven

Nav1.7 is expressed on the surface of peripheral pain-sensing neurons, or nociceptors, where it conducts Na^+ currents in response to membrane depolarizations that are generated by potentially tissue-damaging events, triggering action potential firing and sending pain signals. "It basically sets the gain on pain-signalling neurons — it acts as a volume knob," explains Steve Waxman, a neurologist at Yale School of Medicine.

The first hint that Nav1.7 could have a central role in pain sensing came from a group of researchers in China in 2004, who showed that patients with an inherited persistent pain syndrome called erythromelalgia had point mutations in *SCN9A*, the gene that encodes Nav1.7. The following year, Waxman and colleagues discovered that such mutations caused gain-of-function alterations in Nav1.7, leading to pain hypersensitivity.

Around this time researchers in the laboratory of John Wood, a neurobiologist at University College London, showed that loss-of-function mutations abrogated pain perception. Mice with conditional knockout of Nav1.7 were insensitive to inflammatory pain.

Table 1 | **Selected Nav1.7 inhibitors**

Drug candidate	Sponsor	Modality	Development status
PF-05089771	Pfizer	Small-molecule inhibitor	Discontinued in 2015 after failed phase II trial in painful diabetic peripheral neuropathy
TV-45070	Teva/Xenon	Small-molecule inhibitor	Discontinued in 2017 after failed phase II trial in post-herpetic neuralgia
RG-6029/GDC-0310	Roche/ Genentech/Xenon	Small-molecule inhibitor	Discontinued in 2018 prior to phase II initiation
Vixotrigine	Biogen	Small-molecule inhibitor	Discontinued in painful lumbosacral radiculopathy after phase II failure in 2018; phase III trial planned in trigeminal neuralgia; phase II trial ongoing in small fibre neuropathy
BIIB-095	Biogen	Small-molecule inhibitor	Phase I trial for neuropathic pain ongoing
ST-2427	SiteOne	Small-molecule inhibitor	IND for post-operative pain
AM-6120, AM-8145 and AM-0422	Amgen	Peptide derived from tarantula venom	Discovery
Nav1.7-targeted mAb	Shionogi	mAb	Discovery
VY-NAV-01	Voyager Therapeutics	Gene therapy Nav1.7 knockdown	Discovery

IND, investigational new drug; mAb, monoclonal antibody.

The final piece of the puzzle fell into place in 2006 when medical geneticist Geoff Woods from the University of Cambridge reported in *Nature* that children from three families in Pakistan seemed incapable of experiencing pain. The children earned money as street performers, walking on hot coals and cutting their arms with blades. "A lot of these people have had fractures, and you can hear the bones grating, but it's not painful," says Woods. Genome sequencing revealed that these individuals had loss-of-function Nav1.7 mutations.

Together, these studies marked Nav1.7 out as a crucial mediator of pain. "It's one of very few targets that has what we think of as exquisite human genetic validation," says Bryan Moyer, a neuroscientist at Amgen. "It's very rare to have a gene that plays such a central role in disease pathophysiology, and you have proof-of-concept genetics going in both directions — gain of function causing pain, and loss of function causing lack of pain."

Crucially, individuals with Nav1.7-null mutations seemed otherwise healthy apart from a loss of a sense of smell. "The side effect profile was pretty attractive," says Woods. Existing pain medications are often hampered by dose-limiting side effects such as motor impairment, gastrointestinal liabilities or addiction.

"It just looked like Manna from heaven," agrees Wood. "Every drug company jumped on it."

Seeking selectivity

Several aspects of the channel's biology have since emerged as roadblocks, however.

For Moyer, these provide an important caveat to the enthusiasm for genetically validated targets. "Certainly having that validation is great in terms of general enthusiasm and alignment for drug discovery programmes. But in terms of delivering on a molecule, it doesn't give you a fast-forward to the clinic. You still have to do your job, do rigorous science to understand the biology and solve the problems."

The most widely acknowledged problem among drug developers is the need for channel subtype selectivity. "We need to be exceptionally selective for Nav1.7 in order to achieve a successful pain drug. That is the challenge," says Hackos.

A family of nine Nav channels, Nav1.1–Nav1.9, are expressed in several organs, with roles in cardiovascular, respiratory and neuronal function. Although non-selective Nav blockers can provide useful pain relief — local anaesthetics such as the generic drug lidocaine, for example — the generalized nerve block and unwanted side effects such as dizziness underscore the need for enhanced selectivity to tap wider pain markets.

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Lack of selectivity could explain the recent struggles of vixotrigine, which was first discovered by GlaxoSmithKline and later bought by Biogen via its acquisition of Convergence Pharmaceuticals. Waxman, who was involved in the compound's development, points to mouse studies that indicate that vixotrigine hits several Nav channels in addition to Nav1.7. "That's a very dirty compound — it's not at all specific," says Wood.

Biogen did not respond to requests for comment on their Nav1.7-targeted pipeline.

The crux of the challenge lies in the high structural similarity of the Nav subtypes, which cuts down the binding site options.

"There are fairly limited small-molecule opportunities," says Houghton. "If we look at all the screening people have done, most of the publications and structures are quite closely related to each other. There's not a lot of novelty in that space."

The voltage-sensing domain 4 (VSD4) of Nav1.7 has emerged as one means of achieving selectivity. Many small-molecule Nav1.7 candidates past and present bind here, and are mainly derivatives of aryl sulphonamides, including an early series developed by Pfizer.

To sidestep the chemical constraints, companies including Amgen, Merck and Genentech are now also exploring peptide approaches. In the search for lead compounds, they have independently screened libraries of tarantula venom peptides and found candidates that bind to a different domain, VSD2.

Toxins produced by animals to trap prey and defend against would-be predators often block ion channels, providing a rich seam of selective compounds to work with. Indeed, ω-conotoxin, a calcium channel blocker produced by marine cone snails, is the active ingredient in TerSera Therapeutics' ziconotide, an intrathecally administered analgesic that was approved by the FDA for severe chronic pain in 2004.

Last year, Amgen reported that preclinical candidates from their tarantula venom-derived series inhibited Nav1.7 with high selectivity and blocked histamine-induced scratching in mice. In a recent publication in *Cell*, researchers at Genentech used tarantula toxins to probe the structural basis of Nav1.7 inhibition, in the hope of accelerating the design of their next-generation modulators.

Translating efforts such as these to the clinic will, however, require drug developers to overcome the usual pitfalls of peptide natural products, including short half-lives and anti-drug antibodies.

Other modalities, including Nav1.7-targeted antibodies and gene therapies, are also garnering some interest, although these are still in the discovery stages.

But even as investigators attempt to get a handle on selectivity, success remains elusive. Notably, Pfizer abandoned its lead sulphonamide, PF-05089771, after it failed phase II trials in painful diabetic neuropathy. This compound is thought to be highly selective for Nav1.7 over other Nav channels, suggesting that other challenges have yet to be resolved.

Unknown territory

Another big question that the community is still grappling with revolves around target engagement: how much Nav1.7 inhibition is needed for efficacy? Moyer thinks that the levels that have been achieved in the clinic so far have dampened Na^+ currents but just haven't been sufficient to prevent action potential firing, enabling pain signalling to persist. This issue could account for the failures of clinical efforts to date.

"Has there been enough target engagement to expect an effect on pain behaviour? That's where it gets a bit grey," says Moyer. "When people say 'failures' I'm a bit uncomfortable with that term because I think the field can do a better job in making sure we truly deliver enough drug to engage the target and block a pain response."

Some researchers, including Wood, posit that up to 100% block of Nav1.7 is needed to have a clinical effect on pain — a tall order for any drug. And even if that is achievable, whether it would be desirable is another matter. "It's really two questions," says Moyer. "What do you need to block pain, and what do you need to safely block pain? You certainly don't want to get to a point of blocking so much Nav1.7 that you don't retain protective pain." Whether a tractable therapeutic window exists is a pressing question in the field.

Researchers are also unsure exactly where, anatomically, Nav1.7 blockade is needed for therapeutic efficacy. The channel is expressed along the length of dorsal root ganglion neurons, which are found predominantly in the periphery but that also cross the blood–brain barrier (BBB) to synapse with spinal neurons, where Nav1.7 is thought to have a role in neurotransmitter release.

Some drug developers have specifically designed their candidates not to cross the BBB to avoid off-target toxicity at other Nav channels in the brain. But if Nav1.7 block at spinal nerve terminals is essential for clinical efficacy, such efforts could hit a wall early on.

"The scientific community is still split on whether you need penetration beyond the BBB into the dorsal horn of the spinal cord to achieve clinical pain relief. That question from my point of view is still unanswered," says Waxman.

Opioid input

Findings emerging from academia over the past few years, spear-headed by Wood, could show a different way forward. Studies by Wood and other groups point to a crucial role for endogenous opioid (enkephalin) signalling in pain-insensitive Nav1.7 knockouts, suggesting another possible cause for the clinical failures of potent and selective Nav1.7 inhibitors.

"It certainly has raised the eyebrows of many folks in the pain field," says Moyer, adding that several groups are currently attempting to reproduce the findings.

According to the research, Na^+ can act as a second messenger in sensory neurons, such that deletion of Nav1.7 leads to upregulation of enkephalin expression and potentiation of opioid receptor signalling in mice. "This channel is clearly doing a lot of stuff," says Wood. He also reported that a Nav1.7-null patient was re-sensitized to thermal pain when treated with the opioid antagonist naloxone. And subtherapeutic doses of opioids can synergize with otherwise ineffective Nav1.7 blockers to provide profound analgesia in mice, his group has found.

"This is actually good news because if we had an opiate-sparing drug which allows opiates to give fantastic analgesia but at a dose that didn't give you the gastrointestinal side effects and the addiction propensity, that would be a major hit," says Woods, who was not involved in the research.

Houghton thinks that Nav1.7 inhibitors and opioids can certainly have additive effects in pain models, but she has not been able to reproduce the naloxone-driven reversal of analgesia in the context of Nav1.7 blockade in animal models. Hackos adds that the time course of analgesia onset in mice treated with Genentech's Nav1.7 inhibitors, which happens within about an hour of dosing, is "well before endogenous opioid upregulation has occurred". So in their preclinical studies, "it does not appear that we have to engage the opioid system in order to have a Nav1.7 inhibitor that can block pain," he says.

A more fundamental question is whether acute blockade with a drug can achieve the same effect as a lifelong lack of a channel. "There has probably been a lot of compensation that has gone on along the way [in Nav1.7-null patients]," says Houghton. "What you get in the human phenotype might not predict entirely what you see when you give a small-molecule inhibitor."

This is a potential caveat for other genetically validated targets, including new research that has highlighted a possible mutation in an endocannabinoid enzyme in an individual with insensitivity to pain.

Pain points

The broader translational challenges of pain drug development complicate matters further still. Pain models are poor, and were a major focus of the NIH's recent opioid crisis symposium. And it can be hard to tease therapeutic effect out from placebo effect in clinical trials. Emerging research also suggests that there may be sex-specific differences in pain signalling, with implications for preclinical and clinical work. But the field is making slow progress.

Researchers at Merck are developing non-human primate models of pain that measure analgesic effect using microneurography and quantitative sensory testing methods that are similar to what is already used in the clinic, in the hope of improving the translatability of preclinical studies.

And at the University of Oxford, researchers have used induced pluripotent stem cell technology to create nociceptors from Nav1.7-knockout patients, which they propose could be used as a platform to validate the specificity of putative Nav1.7 blockers. Their results also question the selectivity of Biogen's vixotrigine.

Waxman emphasises the importance of developing biomarkers of pain, pointing to the efforts of researchers in the UK and in the United States to develop functional brain imaging markers.

In the meantime, Waxman thinks trigeminal neuralgia is a good disease model for future trials because patients experience discrete attacks of pain. "It's like an axe hitting you in the face," he says, making it easier to count and quantify outcomes. He was involved in the phase II trial of Biogen's vixotrigine in trigeminal neuralgia, which failed to meet its primary end point. A phase III trial is planned to begin by the end of 2019.

Nav1.8 and Nav1.9, which are also involved in pain signalling by nociceptors, are attracting their own attention. Vertex Pharmaceuticals recently announced positive phase II data for their lead Nav1.8 inhibitor, VX-150, in small fibre neuropathy, adding to similar findings in osteoarthritis and acute pain.

Waxman, as a result, is guardedly optimistic. "I remain enthusiastic about the potential that we may be able to target Nav1.7 and achieve pain relief devoid of central nervous system side effects and addiction in at least some patients," he says. "And worst case, if that fails, I regard Nav1.8 and Nav1.9 as tractable targets."