KCC2 escape from neuropathic pain

By Benjamin Boettner, Assistant Editor

A Canadian team has published preclinical proof of concept that activating a transporter called KCC2 represents a new mechanism for treating neuropathic pain.[1] The group expects that compounds against the ion transporter will be safer than marketed neuropathic pain drugs because its effects are restricted to malfunctioning neurons.

The next step is developing more drug-like molecules against KCC2 (solute carrier family 12 potassium-chloride transporter member 5; SLC12A5) as the first iteration of activators had bioavailability issues.

Neuropathic pain is caused by insults to the nervous system that disrupt the balance between excitatory, pain-causing stimuli and inhibitory, pain-suppressing stimuli. This imbalance can persist long after the initial insult.[2]

The condition, also called hyperalgesia, is treated with a variety of compounds against a range of targets. These include **Pfizer Inc.**'s Lyrica pregabalin, a GABA receptor agonist; **Teva Pharmaceutical Industries Ltd.**'s Effentora, a μ-opioid receptor (OPRM1; MOR) agonist; **NeurogesX Inc.**'s Qutenza capsaicin, a transient receptor potential vanilloid 1 (TRPV1; VR1) agonist; **Elan Corp. plc**'s Prialt ziconotide, an N-type Ca^{2+} channel blocker; and **Eli Lilly and Co.**'s selective serotonin and norepinephrine reuptake inhibitor, Ariclaim duloxetine.

Most of the drugs have limited efficacy and can elicit side effects on neuronal functions including impaired motor abilities, numbing and physical dependency.

KCC2 maintains the low intracellular Cl^- levels needed for normal neuronal function by constantly pushing Cl^- ions out of the cell. KCC2 impairment causes the Cl^- gradient to collapse.

Previous work has shown that inhibitory stimuli in GABAergic neurons can be lost when KCC2 activity is compromised and that this can lead to neuropathic pain.[3] In rats, microglial cells induce Kcc2 activity as a mechanism to counter Cl^- imbalances caused by nerve injury.[4]

Now, a team led by **Laval University**'s Yves De Koninck has identified a compound that stimulates KCC2 activity and shown that it relieved pain in rats.

De Koninck is a professor of psychiatry and neuroscience at Laval and scientific director at the **Hospital Center of Quebec Research Center**.

Using a fluorescent Cl^- extrusion assay, the researchers identified and optimized a compound, CLP257, that had nanomolar activity and was highly selective for KCC2 over related Cl^- transporters.

In spinal slices taken from rats with neuropathic pain, CLP257 enhanced Kcc2 activity and normalized measures of pain sensation, including electrophysiological nerve responses and mechanical withdrawal behavior.

Next, the team developed a prodrug called CLP290 that provided higher serum levels of CLP257 than the original compound. In rats, the analgesic effect of CLP290 was similar to that of Lyrica. Unlike Lyrica, the prodrug did not numb motor functions.

Findings were reported in *Nature Medicine*.

"KCC2 represents a fresh avenue to pain medication because stimulating KCC2 normalizes endogenous pain inhibition," said De Koninck. "In normal neurons, Cl^- levels are kept very low. Therefore, the effect of KCC2 enhancers will mainly touch on troubled neurons with elevated Cl^- levels."

"The strategy could have significant advantages over other therapeutic agents like opioids, Lyrica pregabalin and calcium channel blockers, which mostly regulate ion channels and not active transporters," added De Koninck.

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> ***—Allan Basbaum, University of California, San Francisco***

"The mantra is that the best drugs for neuropathic pain—the gabapentinoids—are only effective in about 30% of patients, and those patients on average only get about 30% relief. So clearly new drugs are essential," said Allan Basbaum, chair of the Department of Anatomy at the **University of California, San Francisco**.

Targeting KCC2

Based on earlier KCC2 findings, De Koninck cofounded Chlorion Pharma Inc. to develop KCC2 agonists for neuropathic pain.

The company raised C$6 million ($6.1 million) in 2007, but Chlorion's first series of activators ran into bioavailability issues in a preclinical study in dogs. The molecules did show high KCC2 specificity and no on-target toxicity.

The company set about fixing the bioavailability issue and identified an undisclosed series of KCC2 agonists but ultimately ran out of money and closed its doors in 2011. Rights to the activators reverted back to Laval University.

De Koninck said that his team since has shown markedly improved bioavailability for the new agonists and is in talks with industry.

Regardless of bioavailability, Michael Gold, a professor of anesthesiology at the **University of Pittsburgh**, said that it will be important to examine KCC2 stimulation in assays that better reflect ongoing pain such as the conditioned place preference assay.

Ronald Burch, CMO of **Naurex Inc.**, said that it will also be

important to investigate KCC2 stimulation in multiple neuropathic pain models. The reason, he said, is that the condition can have many causes, including physical nerve injury and metabolic injury in diabetic neuropathy, as well as inflammatory and infectious diseases like postherpetic neuralgia.

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—Yves De Koninck, Laval University

Gold agreed, noting that inflammatory pain models in particular do not rely on spontaneously incurred physical injury. Although the models in the paper used pain that sets in after an initial physical injury subsides, inflammatory pain models likely involve different pain mechanisms altogether and would help show that KCC2 stimulation works more broadly.

Naurex has completed a Phase I trial of GLYX-13, an NMDAR modulator, to treat neuropathic pain.

Ruth McKernan, CSO of Pfizer's Neusentis unit, told *SciBX* that "it will be important to quantitatively assess target engagement at relevant sites of action *in vivo* to aid in predicting human exposure required for both efficacy and side effects. This would be a high priority for the target class since we really do not know what is required in man and how preclinical results will translate to clinical studies."

Basbaum and Gold also wanted to see more detailed dose-response curves for KCC2 stimulation in assays of neuropathic pain and motor functions. Such data would help define the therapeutic window of KCC2 stimulation.

Finally, Gold noted that it will be necessary to determine "how long the drug effects last and whether the drug can be repeatedly administered and still take effect. Time-course studies can show whether the drug-induced Cl^- shift persists or whether the cells compensate for it long term."

Basbaum added, "As the best drugs for neuropathic pain are generally also anticonvulsants, it is of interest to learn whether the new compound acts in a fashion similar, at least clinically. If not, then one can conclude that clearly it is working via a very different mechanism. That also argues that some combination of the gabapentinoids and the new compound could prove especially valuable."

Wider horizons

Besides neuropathic pain, other neurological disorders with imbalances in Cl^- homeostasis, like epilepsy, migraine or anxiety, could benefit from KCC2 stimulation.

De Koninck plans to study KCC2-targeted compounds in an epilepsy model in which epilepsy-related neurological changes develop before they eventually trigger epileptic episodes. The model will provide insights on the effects of KCC2 agonists on seizure-evoking hyperexcited neurons and on changes in neuronal networks.

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COMPANIES AND INSTITUTIONS MENTIONED

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