

Understanding Atomic Interactions to Achieve Well-being

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Voltage-gated sodium channels are among the most active and ubiquitous molecular machines found in animals. Residing in the cell membranes of excitable and other cells, they derive



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energy for their opening and closing from changes in membrane potential. In some cells, particularly those in sensory information-encoding structures of the central and peripheral nervous systems, these changes take place repetitively every few milliseconds, making the channel gates some of the most conformationally versatile structures of nature. The ion channel-lining domain, also called the α subunit, is particularly prone to mutations that alter channel production and targeting, ion conduction, or gate action. Particularly interesting are mutations that render ion channel gates more or less prone to opening. These mutations are a significant cause of abnormal pain sensation in man, a largely unmet medical need and a fascinating biophysical phenomenon. Channels are inherently capable of opening and closing, allowing or blocking sodium ions from traveling through the pore. In fact, they can do so spontaneously, traversing a variety of different conformational states over time. Some states are energetically more favored than others; however, such that little or no significant opening is detectable at rest (when cells are hyperpolarized). What membrane potential does, in the case of sodium channels, is favor some of these states, resulting in the net dwelling of the ion channel in open or closed conformation.

The channel structures involved in voltage-dependent opening and closing have been the subject of much research since the first sodium channel was cloned in 1984.¹ At the time, so little was known about how such structures may regulate gating that what would be later known as the voltage-sensing structure of the channel protein was drawn outside of the cell membrane, thus paradoxically preventing the voltage sensor from interacting with the membrane potential. Today, we know that, while voltage-dependence of opening of channels is initiated by the movement of conspicuous charges embedded in the channel protein, many other permissive interactions change elsewhere in the channel protein. It is these otherwise unsuspected interactions that are coming to the forefront of ion channel biophysics and of neurology.

Such an interaction is exemplified by the pair of residues S241 and V400. Located in disparate areas of the linear channel protein, S241 and V400 are part of α -helices that interact to stabilize the closed conformation of the channel $\text{Na}_v1.7$.² In fact, the S241 residue is located only within 2.8 Å of the V400 locus in the mature, folded protein. Nevertheless, the asso-

ciation between the residues (a hydrophilic serine and a hydrophobic valine) cannot be deduced a priori from any features of the channel protein and only becomes clear via a combination of site-directed mutagenesis and computational modeling of the observed effect on the channel protein. This interaction becomes more relevant because mutations in $\text{Na}_v1.7$ (one of which involves S241 in particular) cause several debilitating, treatment-refractory pain disorders such as congenital indifference to pain, paroxysmal extreme pain disorder, and inherited erythromelalgia.³

In erythromelalgia, the main site of pathology is the dorsal root ganglion cell, which harbors hyperactive voltage-gated sodium channels of the $\text{Na}_v1.7$ type. Such gain of function (if the function of the sodium channel is to depolarize the cell, making it more excitable) leads to an infrequent but disabling disorder consisting of frequent episodes of severe pain triggered by a variety of stimuli. Patients resort to cooling their painful limbs in ice water, with resulting damage. S241T leads to such a syndrome. By subtly disrupting the S241-V400 interaction, the channel opens more often, leading to excess depolarizing sodium influx in the dorsal root ganglion cells and consequent activation of neural relay structures including the cerebral cortex. This is the correlate of what the individual perceives as pain in this syndrome.

In this issue of *JAMA Neurology*, Geha et al⁴ cleverly exploit the susceptibility of the V400 locus to carbamazepine, a sodium-channel stabilizer, when mutated into V400M. The drug leads to stabilization of the V400M channel, reversing the pathological gain of function. Because V400 and S241 interact at the level of their side chains (this is how α -helices generally interact with one another), the authors reasoned that if carbamazepine stabilizes V400M, it should also stabilize its interacting partner S241T. To test this, 2 patients with the S241T mutation were recruited. Because pain is invisible to persons and technology, a variety of lines of evidence were pursued to demonstrate therapeutic efficacy. This started, foremost, with a controlled assessment of the sensation of pain by the 2 patients. A subjective measurement of pain used both a diary and a potentiometer, which reported the degree of pain experienced on controlled triggering. Then, the patients received blood oxygen level-dependent functional magnetic resonance imaging to ascertain that pain-related brain areas were indeed activated while experiencing pain and that this pattern changed while carbamazepine exerted its therapeutic effect. Last, to complement these powerful observations, recordings from dorsal root ganglion cells expressing the mutant sodium channels subjected to carbamazepine action documented a favorably restored excitability profile.

This study⁴ provides an intelligent practical demonstration of the growing value of molecular neurological reasoning. For the sake of rendering the problem treatable, the first step was reducing the patients' syndrome to an abnormal molecular mechanism, one that the group first suggested only in 2009.⁵ Next came a search for a drug that could alter such a mechanism. By knowing the effect of the drug on the molecule, the authors were able to predict that a pathological gain of function would be effectively palliated without any obvious collateral consequences. Understanding the site where the

mechanism resided allowed for a mechanistic proof that linked molecular mechanism with symptom relief. To my knowledge, there are still relatively few examples in medicine where molecular reasoning is rewarded with a comparable degree of success, such that treatment development for most diseases remains an arcane combination of epidemiological and toxicological efforts coated with some mechanistic varnish to lend credibility to an otherwise largely trial-and-error enterprise. The study by Geha et al⁴ points the way in a different, refreshing, and much more rewarding direction.

ARTICLE INFORMATION

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