characterized by chronic progressive weakness whose affected members harbor a novel Ca$_{1.1}$ R1242G mutation affecting the third arginine in S4 of domain IV. Whole-cell patch clamp recordings of the R1242G mutant showed a strong hyperpolarized shift of steady-state inactivation in GLT mouse myotube expression system. The facilitated inactivation resulting in the reduction of channel availability may account for the chronic progressive weakness of the patients. In addition, we found an outward gating pore current through the mutant voltage sensor of R1242G which might induce a reduction in the amplitude and duration of the action potential and cause the failure of muscle fiber excitability.

1834-Plat
Bio-Inspired Voltage-Dependent Calcium Channel Blockers
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Blocking voltage-dependent Ca$_{1.1}$/Ca$_{2.2}$ channels is a prevailing or potential therapy for myriad diseases ranging from hypertension to Parkinson’s disease, but a major limitation is lack of selective small-molecule inhibitors for distinct Ca$_{1.1}$/Ca$_{2.2}$ channels. Here, we report a general bio-inspired approach towards developing novel Ca$_{1.1}$/Ca$_{2.2}$ channel blockers (CCBs). We discovered that different proteins (Ca$^{2+}$/β, 1,4-3, cannulin, and calmodulin-dependent protein kinase IID) that bind to widely distinct regions of pore-forming α-subunit intracellular loops can be converted into CCBs, with tunable selectivity and potency, simply by their anchoring to the plasma membrane. The principle is extendable to small molecules—engineering FKBP into specific sites within Ca$_{1.1}$/Ca$_{2.2}$ intracellular loops permitted heterodimerization-initiated channel inhibition with rapamycin (Fig). The results reveal a universal method for developing novel genetically-encoded and small-molecule CCBs.

1835-Plat
Novel Modulatory Action of Calmodulin Complexation with L-Type Channels
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L-type Ca$^{2+}$ channels convey vital Ca$^{2+}$ inflow, which is critically downregulated in intracellular Ca$^{2+}$. This Ca$^{2+}$-dependent inactivation (CDI) is only present in channels initially preassociated with Ca$^{2+}$-free calmodulin (apoCaM), where subsequent Ca$^{2+}$ binding to this ‘resident’ calmodulin (CaM) inactivates channels (Nature463:968). Channels lacking preassociated apoCaM fail to undergo CDI. Canonical L-type channels bind apoCaM so avidly that nearly all channels are ‘charged’ with apoCaM. However, natural variants feature potentially lower apoCaM affinity, so fluctuations in ambient apoCaM may tune overall CDI. Here, we demonstrate that channel preassociation with apoCaM does more than simply enable CDI, and strikingly enhances permanent charge (of acidic side chains) was varied. Substantial competition we used isothermal titration calorimetry to determine the affinity of both CaM and CaBP1 in both calcium-bound and apo-states for the IQ domain. Additionally, the IQ domain introduces by Nonner and Eisenberg, computing electric forces from all distances of permanent charge. Different conduction states had different selectivity, one resembling L-type Ca$_{1.1}$ and the other RyR channels. We speculate that thermally activated switching between conductance values could produce some types of spontaneous gating. Below is calcium current J as a function of permanent charge Qf and calcium concentration. Details at arXiv.org 1209.2581

1837-Plat
Pharmacological Correction of Gating Defects in the Voltage-Gated Cav2.1 Ca$^{2+}$ Channel due to a Familial Hemiplegic Migraine Mutation
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Voltage-gated ion channels exhibit complex forms of gating, which can be targeted in pharmacological therapies for disease. Here, we report that the pro-oxidant (1,2-dialkyl dihydroquinone (BHQ)), modulates Cav2.1 Ca$^{2+}$ channels in ways that oppose defects in channel gating and synaptic transmission resulting from a familial hemiplegic migraine mutation (S218L). BHQ inhibits voltage-dependent activation and unmasks slow deactivation that is enhanced by Ca$^{2+}$ and more prevalent in S218L mutant than in wild-type channels. These actions of BHQ help reverse the gain-of-function and reduced Ca$^{2+}$-dependent facilitation of Cav2.1 channels with the S218L mutation. Transgenic expression of the mutant channels at the Drosophila neuromuscular junction causes abnormally elevated evoked postsynaptic potentials and impaired synaptic plasticity, which are largely restored to the wild-type phenotypes by BHQ. Our results reveal a new mechanism by which Cav2.1 gating modifiers can correct defects associated with disease-causing mutations in Cav2.1.

1838-Plat
Comparison between Calcium Sensors on the Voltage-Gated Calcium Channel Cav1.2
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Voltage-gated calcium channel (CaV) activity is regulated by calcium sensors including calmodulin (CaM) and calcium-binding protein 1 (CaBP1). CaBP1 inhibits CaM-mediated calcium-dependent inactivation (CDI). We investigated the origins of functionally important differences between CaM and CaBP1 by creating a number of CaM/CaBP1 chimeras which suggest that both calcium sensors use their C-lobes for high affinity binding to the pore-forming CaV alpha-subunit, while their N-terminal lobe are responsible for the stark functional differences of the calcium sensors. CaBP1 and CaM are thought to modulate CaV function by competing for binding to the CaV C-terminal IQ-domain, but this assumption has never been tested directly. By determining Cav1.2 CDI in Xenopus oocytes under conditions with different ratios of CaM and CaBP1, we demonstrate direct competition between both calcium sensors for their Cav1.2 binding site. In order to extend our analysis of CaBP1/CaM competition we used isothermal titration calorimetry to determine the affinity of both CaM and CaBP1 in both calcium-bound and apo-states for the IQ domain, suggesting that competition occurs mainly in the apo-state. Overall, our data provide a framework for understanding how CaBP1 and CaM differentially regulate CDI on CaV1.2.

1839-Plat
Optically-Resolved Conformational Rearrangements of the Voltage-Sensing Domains of the Human Ca$_{1.2}$ Channel
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Voltage-gated Ca$^{2+}$ channels (Ca$_{3}$) consist of four tandem transmembrane repeats (I-IV), each including two pore-forming helices and four segments that make up a voltage-sensing domain (VSD). To understand voltage-dependent