Structural determinants of pacemaker HCN channelsblockage by Ivabradine and its technological advancements

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Hyperpolarization-activated cyclic nucleotide-gated (HCN) channels are the molecular correlate of the I_h (or I_f) current, which plays a key role in controlling rhythmic activity in cardiac pacemaker cells and spontaneously firing neurons. We have recently obtained the structure of HCN4 with the pore in the open and closed conformation[1], thus advancing the understanding of permeation and conductance in HCN channels[1,2]. Being able to purify HCN4 molecules with the pore in the open state[1], we have now characterized, at atomic details, the action of Ivabradine, an open channel blocker specific to HCN channels and currently approved for clinical use in heart failure. By testing purified HCN4 proteins for the Ivabradine-induced shift in a thermal denaturation assay[3] we have obtained the first biochemical demonstration of Ivabradine binding to the open pore of HCN4and proceeded to solve the structure of the complex by single particle cryo-EM[4]. We can thus describe the pattern of contacts that Ivabradine develops with the residues facing the intracellular vestibule of the HCN pore[4]. Strikingly, molecular dynamics simulation experiments uncovered the key role of the tertiary ammine of Ivabradine in the mechanism of block, thus explaining the current dependency of the drug-induced block[4]. Moreover, our structural understanding of Ivabradine binding to the open pore of HCN channels let us to engineer a blue-light activable version of the drug able to optogenetically control HCN current both *ex vivo*and *in vivo*.