

# Cyclic Nucleotide-Gated (CNG) and Hyperpolarization Activated Cyclic Nucleotide-Gated (HCN) Channels

## Key References

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## Overview

Cyclic nucleotide-regulated cation channels are classified into two principal subfamilies, the cyclic nucleotide-gated (CNG) channels and the hyperpolarization-activated cyclic nucleotide-gated (HCN) channels. The two channel families differ from each other with regard to their mode of activation. CNG channels are typical ligand-gated channels because their activation requires the binding of cAMP or cGMP. In contrast, HCN channels are principally operated by voltage. These channels open at hyperpolarized membrane potentials and close on depolarization. However, apart from their voltage sensitivity, HCN channels are also activated directly by cyclic nucleotides, which act by shifting voltage dependence of channel activation to more positive potentials.

CNG and HCN channels are members of an extended superfamily of cation channels that is characterized by a principal building unit containing six transmembrane helices (S1-S6) and an ion conducting reentrant pore-loop between S5 and S6. CNG conduct calcium and a variety of monovalent cations whereas HCN channels only pass monovalents. Regulation by cyclic nucleotides is conferred by a cyclic nucleotide-binding domain (CNBD) present in the carboxy terminus. Activation of HCN channels by hyperpolarization is controlled by the positively charged S4 helix carrying nine regularly spaced arginine or lysine residues at every third position.

CNG channels are expressed in retinal photoreceptors and olfactory neurons and play a key role in visual and olfactory signal transduction. Defects in CNG channel function cause retinal diseases such as retinitis pigmentosa and total colorblindness (achromatopsia). In addition, CNG channels are found at low density in some

other cell types and tissues such as brain, testis and kidney. The physiological role of CNG channels in these tissues is not known yet. In vertebrates the CNG channel family comprises six homologous members. Based on phylogenetic relationship, these proteins are divided into two subfamilies, the A subunits (CNGA1-4) and the B subunits (CNGB1 and CNGB3). Native CNG channels are heterotetramers with different heteromers displaying distinct nucleotide sensitivity, ion selectivity and modulation by calcium. The subunit composition and stoichiometry has been determined for three native channels: the rod and cone photoreceptor channels and the olfactory channel.

Several drugs have been reported to block CNG channels, although not with very high affinity. The most specific among these drugs is L-*cis* diltiazem which blocks CNG channels in a voltage-dependent manner at micromolar concentrations. High affinity binding of L-*cis* diltiazem is only seen in heteromeric CNG channels containing the CNGB subunits. CNG channels are also moderately sensitive to blockage by some other inhibitors of the L-type calcium channel (e.g. nifedipine), the local anesthetic tetracaine and calmodulin antagonists.

HCN channels represent the molecular correlate of the hyperpolarization-activated cation current, I<sub>h</sub>. The channels play a central role in the initiation and control of the heart beat. Enhancement of channel activity by binding of cAMP represents the major mechanism by which norepinephrine and other adrenergic agonists increase heart rate. In brain, HCN channels serve to support multiple functions including sleep-wake cycle, motor learning, and dendritic signal integration. The HCN channel family comprises four homologous members

(HCN1-4). These subunits assemble to homomeric and heteromeric tetramers with distinct activation thresholds, opening kinetics and responsiveness to cAMP.

There is significant therapeutic potential for drugs that modulate HCN channels. Dysfunction of HCN channels has been linked to cardiac dysrhythmia, ataxia, absence epilepsy, and neuropathic pain syndromes. Blockers of HCN channels have been pursued as potential bradycardiac, antiepileptic and analgesic agents. The most extensively studied blockers of HCN channels are ZD7288 and ivabradine. Both agents block HCN channels in the low micromolar range and reduce heart rate in a variety of species including man.

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FAMILY	HCN	CNGA	CNGB
<b>SUBTYPES/STRUCTURAL INFORMATION</b>	HCN1 (910 aa [mouse]) HCN2 (889 aa [human]) HCN3 (779 aa [mouse]) HCN4 (1203 aa [human])	CNGA1 (690 aa [human]) CNGA2 (664 aa [mouse]) CNGA3 (694 aa [human]) CNGA4 (575 aa [rat])	CNGB1a (1251 aa [human]) CNGB1b (858 aa [rat]) CNGB3 (809 aa [human])
<b>TISSUE EXPRESSION</b>	Brain, heart	A1: rod photoreceptors A2: olfactory neurons A3: cone photoreceptors, some olfactory neurons, sperm A4: olfactory neurons, vomero-nasal organ A1-A4: low level expression in various neuronal and non-neuronal cell types	B1: rod photoreceptors (B1a), olfactory neurons (B1b), testis B2: olfactory neurons (B2b), testis B3: cone photoreceptors, testis
<b>STRUCTURE OF NATIVE CHANNELS</b>	Homo- and heterotetrameric (subunit stoichiometry not known)	Heterotetrameric: Rod photoreceptors: 3xCNGA1/1xCNGB1a Cone photoreceptors: 2xCNGA3/2xCNGB3 Olfactory neurons: 2xCNGA2/1xCNGA4/1xCNGB1b	Heterotetrameric: Rod photoreceptors: 3xCNGA1/1xCNGB1a Cone photoreceptors: 2xCNGA3/2xCNGB3 Olfactory neurons: 2xCNGA2/1xCNGA4/1xCNGB1b
<b>ACTIVATION</b>	Hyperpolarization, binding of cAMP shifts activation curve to more positive voltages	Heteromers and homomeric CNGA1-3 channels: activation by binding of cGMP or cAMP very weak voltage dependence CNGB1 subunits and CNGA4 do not express as functional homomeric channels	Heteromers and homomeric CNGA1-3 channels: activation by binding of cGMP or cAMP very weak voltage dependence CNGB1 subunits and CNGA4 do not express as functional homomeric channels
<b>ION SELECTIVITY</b>	K <sup>+</sup> , Na <sup>+</sup> (p <sub>Na</sub> /p <sub>K</sub> ~0.2)	Heteromers and homomeric CNGA1-3 channels: Ca <sup>2+</sup> >K <sup>+</sup> ~Na <sup>+</sup>	Heteromers and homomeric CNGA1-3 channels: Ca <sup>2+</sup> >K <sup>+</sup> ~Na <sup>+</sup>
<b>BLOCKERS</b>	ZD7288, ivabradine, cilobradine, zatebradine	L-cis diltiazem (high affinity binding requires presence of CNGB subunits), tetracaine ( <b>T7383</b> ), W-7 ( <b>A3281</b> ), pimozone ( <b>P1793</b> ), pseudochetoxin	L-cis diltiazem (high affinity binding requires presence of CNGB subunits), tetracaine ( <b>T7383</b> ), W-7 ( <b>A3281</b> ), pimozone ( <b>P1793</b> ), pseudochetoxin
<b>PHENOTYPE OF GENE-DEFICIENT MICE</b>	HCN1: deficit in motoric learning HCN2: absence epilepsy, cardiac dysrhythmia HCN4: lethal at embryonic day 10.5; impaired formation of sinoatrial pacemaker cells	A2: anosmia A3: achromatopsia and cone degeneration A4: decelerated adaptation of olfactory neurons to odor stimulation	B1: loss of rod-mediated vision, progressive photoreceptor degeneration, dysfunctional response to odorants
<b>HUMAN CHANNELOPATHIES</b>	HCN4-573X: sick sinus node disease HCN4-D553N: cardiac arrhythmia	CNGA1: missense mutations: retinitis pigmentosa CNGA3: missense mutations: achromatopsia	CNGB1a-G993V: retinitis pigmentosa CNGB3: missense mutations: achromatopsia

## Abbreviations

**W-7** : N-(6-Aminohexyl)-5-chloro-1-naphthalenesulfonamide hydrochloride

**ZD7288** : (N-Ethyl-N-phenylamino)-1,2 dimethyl-6-(methylamino) pyridinium chloride

## FOOTNOTES