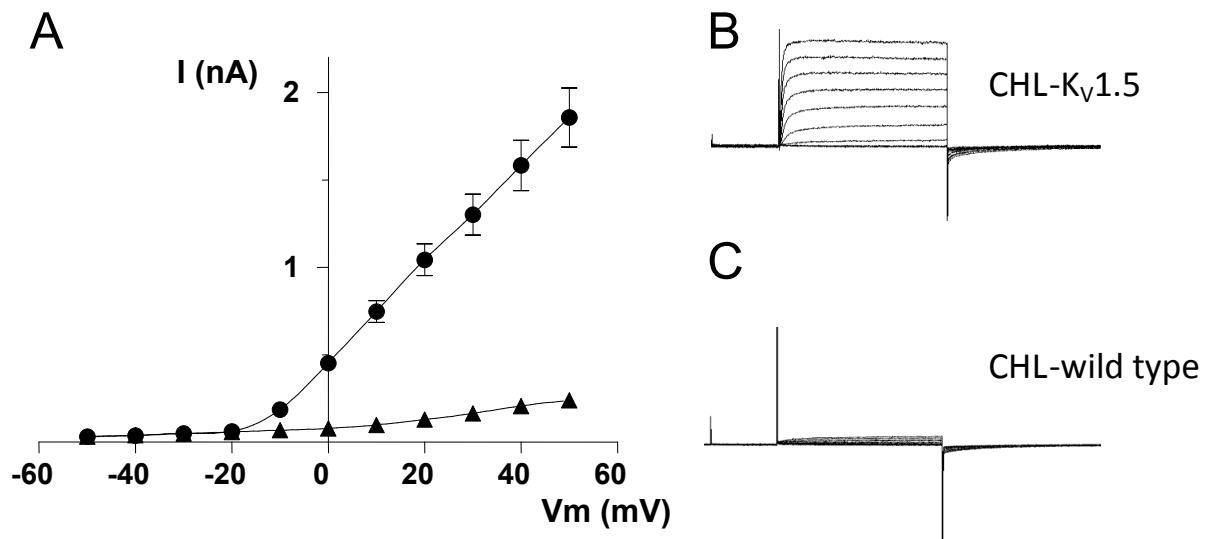


hK_v1.5 automated patch-clamp electrophysiology assay

- **Assay** hK_v1.5 automated patch clamp electrophysiology
- **Channel** voltage-gated K⁺ channel, K_v1.5 (*Shaker*-related family)
- **Gene Name** KCNA5 (Ref Sequence NM_002234.2)
- **Synonyms** HK2, HCK1, PCN1, ATFB7, HPCN1, KV1.5
- **Assay format** 384-well IonWorks^{HT} Population Patch Clamp electrophysiology
- **Cell Host** Chinese Hamster Lung (CHL)
- **Stimulus** Repeated gating steps, V_h -80mV, V_{step} +40mV, 1 Hz
- **Controls** Quinidine 1 mM, 0.3% DMSO



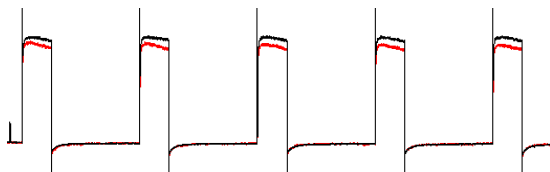
Biophysics

Current-voltage relationship for hK_v1.5 channels stably expressed in CHL cells (A). Representative recordings from CHL-K_v1.5 (B) and CHL-wild type cells (C). Experiments were conducted from a holding potential of -80mV. The depolarizing step length was 200ms.

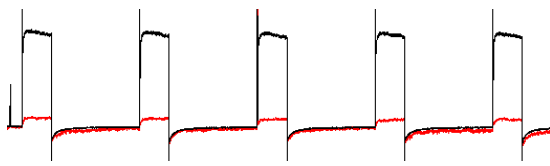
Background $K_v1.5$ voltage-gated K^+ channels are thought to underlie the ultra-rapidly activating delayed rectifier K^+ current (I_{kur}) found in human atrial myocytes. $K_v1.5$ is also expressed in the human ventricle where it is possible that it contributes to the K^+ current through formation of heteromultimeric K^+ channels with other K_v -alpha subunits. Inhibition of K_v channels increases atrial refractory period in man – this is clinically valuable for the treatment of patients suffering atrial fibrillation, but undesirable for drugs designed to have inert cardiovascular profiles.

Screening Protocol Repeated gating protocol to determine inhibition of h $K_v1.5$ K^+ currents

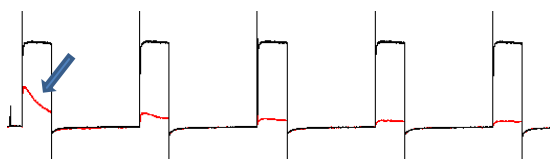
A Control



B + Psora-4 11 μ M



C + CP-339818 30 μ M



Compound	IC ₅₀ - 1 st Pulse Peak (μ M)	IC ₅₀ - 5 th Pulse Peak (μ M)
Psora-4	1.8	1.8
CP-339818	32	3.2
DPO-1	7.5	5.3
Mephetyl Tetrazole	13	4.9
Bupivacaine	39	26
Quinidine	50	47

Repeated gating steps, V_h -80mV, V_{step} +40mV, 1 Hz, pulse duration 250ms. Control traces in black, test compounds in red. IC₅₀ values were obtained by fitting concentration-response curves to values for inhibition of the 1st and 5th pulse in the train (μ M, shown in table). Use-dependent and open channel block can be assessed within this protocol (note blue arrow).

Assay QC Cell, Curve fit, Solubility & Plate QC parameters applied:

Cell: $I_{hKv1.5}$ >800pA, baseline & seal resistance stability filters

Plate QC: Minimum 350 cells, Quinidine pIC₅₀ 4.4 - 5.0 (1st pulse) and 4.4 – 5.0 (5th pulse)

Typical assay precision: compound pIC₅₀ values \pm 0.25 log units

Why Essen? The protocol for the hERG assay takes into account several critical factors which can affect compound potency. These include careful compound preparation/handling, standardized cell preparation procedures, and recording solutions and voltage protocols designed to maximize sensitivity. The assay is designed to accurately assess the widest array of pharmacological and chemical entities, including use-dependent and hard-to-handle (sticky) compounds. We offer high data fidelity, competitive pricing and turn-around times difficult to match by CRO's who are simply end-user's of the IonWorks technology.

References Wang Z, Fermini B, Nattel S. (1993). Sustained depolarization-induced outward current in human atrial myocytes. Evidence for a novel delayed rectifier K^+ current similar to $K_{v1.5}$ cloned channel currents. *Circ Res.* **73**:1061-76.

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