



Stably Transfected Cell Line - Product Data Sheet
hK_{Ca}3.1-CHO
Catalog Number CT6123

Related Services and Products

FastPatch[®] and ScreenPatch[™] automated patch clamp services
Additional information available at www.chantest.com

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1 Cell Line Description

1.1 Background

K_{Ca}3.1 (IK) is an intermediate-conductance (11 pS single channel conductance), potassium-selective, channel that is activated by increased intracellular Ca²⁺. Widely distributed, hK_{Ca}3.1 channels are therapeutic targets in several inflammatory and immune diseases.

1.2 Pore-forming subunit identifier: hK_{Ca}3.1

Synonym: IK channel

Class: Calcium-activated potassium channel

Species: Human

Gene name: KCNN4

1.3 Sequence Information

The cDNA sequence of the KCNN4 gene used to create this cell line was confirmed prior to transfection. The amino acid sequence encoded by the transfected cDNA is identical to the translated sequence for GenBank accession number NM_002250.2.

1.4 Expression System

CHO (Chinese hamster ovary cells), constitutive expression

1.5 Product Format

Cryopreserved cells, 1 x10⁶ cells/vial.

1.6 Mycoplasma Status: Negative

The absence of mycoplasma species in this cell line was confirmed with the MycoAlert Kit (Lonza Rockland, Inc.).

1.7 Cell Line Stability

Channel expression in this cell line has been stable for at least 52 passages.

2 Validated Test Platforms

Electrophysiological and pharmacological verification of the functional properties of the cloned channels was assessed in the following test platforms:

QPatch™ HT (Sophion)

IonWorks® Quattro™ (MDS-AT)

2.1 QPatch™ HT Representative Data

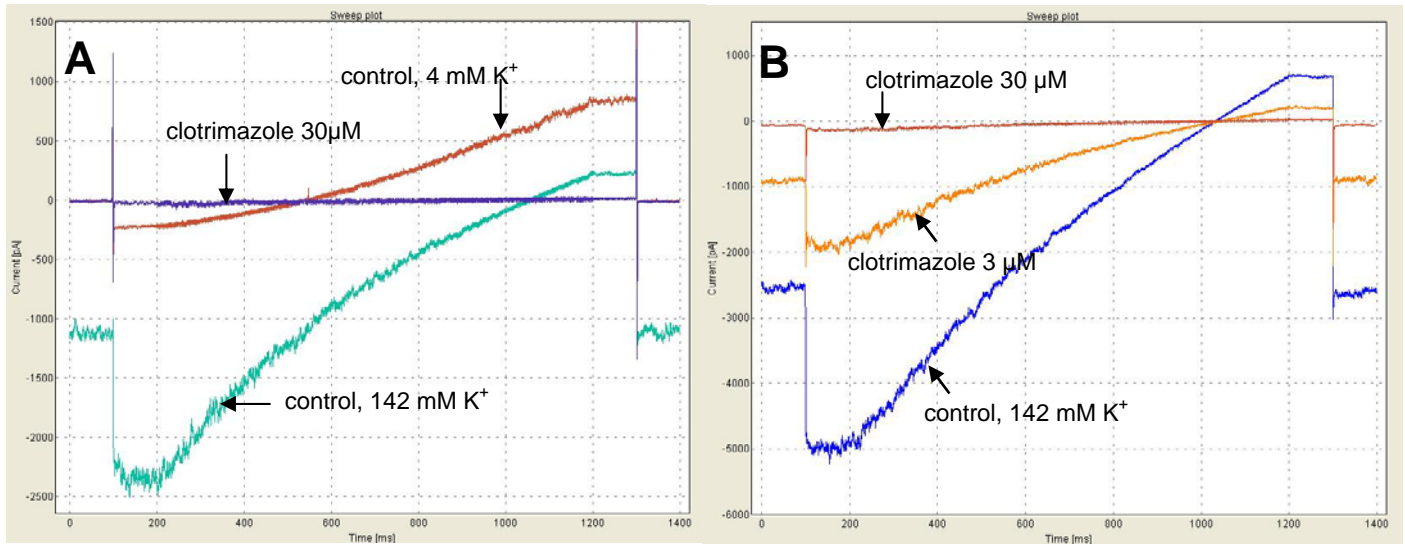


Figure 1. hK_{Ca}3.1 currents in QPatch™ HT.

A: hK_{Ca}3.1 activation by inclusion of 10 μM free Ca²⁺ in the intracellular solution. Test pulse currents evoked by voltage ramps (-110 to +40 mV, holding potential -60 mV) before (control) and after 30 μM clotrimazole application. Horizontal and vertical calibrations, 200 ms and 500 pA, respectively. **B:** Test pulse currents evoked by voltage ramps (-110 to +40 mV, holding potential -60 mV) with 10 μM free [Ca²⁺]_i and 142 mM [K⁺]_o, before (control) and after clotrimazole application. Horizontal and vertical calibrations, 200 ms and 1000 pA, respectively.

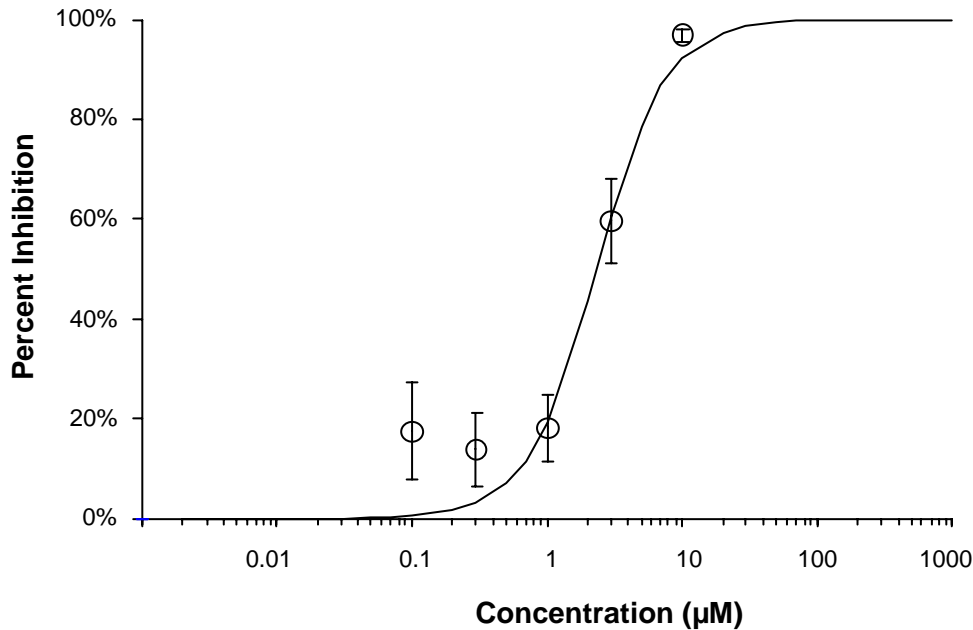


Figure 2. Clotrimazole Concentration-Response Relationship

Mean ± SEM, n = 2 - 6 cells/concentration. IC₅₀ = 2.3 μM.

2.2 IonWorks[®] Quattro[™] Representative Data

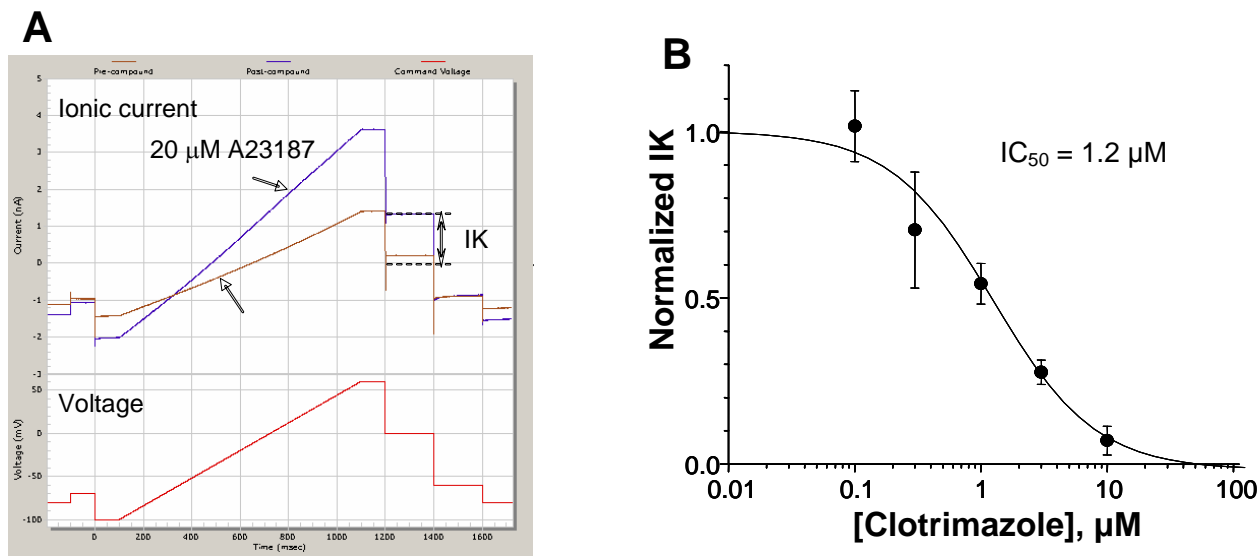


Figure 3. Clotrimazole Inhibition of hK_{Ca}3.1-CHO in IonWorks[®] Quattro[™]

A: Ca²⁺-sensitive potassium channels were activated by exposure of the cells to a Ca²⁺ ionophore (20 μM A23187). Ionic currents were elicited by the voltage pattern shown in the lower panel. The upper panel shows typical signals before (basal current) and after activation. To minimize the contribution of leak and endogenous currents, K_{Ca}3.1 current amplitude (IK) was measured at a test potential of 0 mV. All current amplitudes were normalized to the mean value of the control current (0 μM clotrimazole, 20 μM A23187). Horizontal and vertical calibrations, 200 ms, and 1 nA or 50 mV, respectively. **B:** Co-administration of clotrimazole produced dose-dependent inhibition of the A23187-activated IK. The calculated IC₅₀ value for clotrimazole-induced block was 1.2 μM; data presented as Mean ± SD (n = 4 replicates/concentration).

3 References

Jensen BS, et al. 1998. Characterization of the cloned human intermediate-conductance Ca²⁺-activated K⁺ channel. *Am J Physiol* 275:C848-C856.

Wei AD, et al. International Union of Pharmacology. LII. 2005. Nomenclature and molecular relationships of calcium-activated potassium channels. *Pharmacol Rev.* 57:463-472.