



hK_{ir}2.1-HEK
Stably Transfected Cell Line
Catalog Number CT6103

Related Services and Products

FastPatch[®] and ScreenPatch[™] automated patch clamp services
Replicating hK_{ir}2.1-CHO cell line. Cat. No. CT6127
Additional information available at www.chantest.com

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1 Cell Line Description

1.1 Background

K_{ir}2.1 is an inwardly-rectifying, K⁺-selective channel, widely distributed in various tissues including cardiac, smooth and skeletal muscle, and brain. In the heart, K_{ir}2.1 is responsible for the I_{K1} current that regulates resting potential and contributes to action potential repolarization.

1.2 Pore-forming subunit identifier: hK_{ir}2.1

Class: Inwardly rectifying potassium channel
Species: Human
Gene name: KCNJ2

1.3 Sequence Information

The cDNA sequence of the KCNJ2 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_000891.2.

1.4 Expression System

HEK293 (human embryonic kidney) cells, tetracycline-inducible 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

Table 1. Stability of hK_{ir}2.1 Current

Passage Number	Current Amplitude (pA)	n
37	6.78 ± 3.32	4
45	6.64 ± 1.49	5
53	4.54 ± 1.20	8
61	4.84 ± 1.51	5

A frozen vial at P28 was thawed and passaged for stability measurements. hK_{ir}2.1 current amplitudes recorded by IonWorks[®] Quattro[™] (mean ± standard deviation). Table 1 shows that current amplitude measured from hK_{ir}2.1-HEK remains stable for at least 33 passages beyond the original frozen vial.

2 Validated Test Platforms

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

Manual Patch Clamp
PatchXpress[®] (MDS-AT)

2.1 Manual Patch Clamp Validation Results

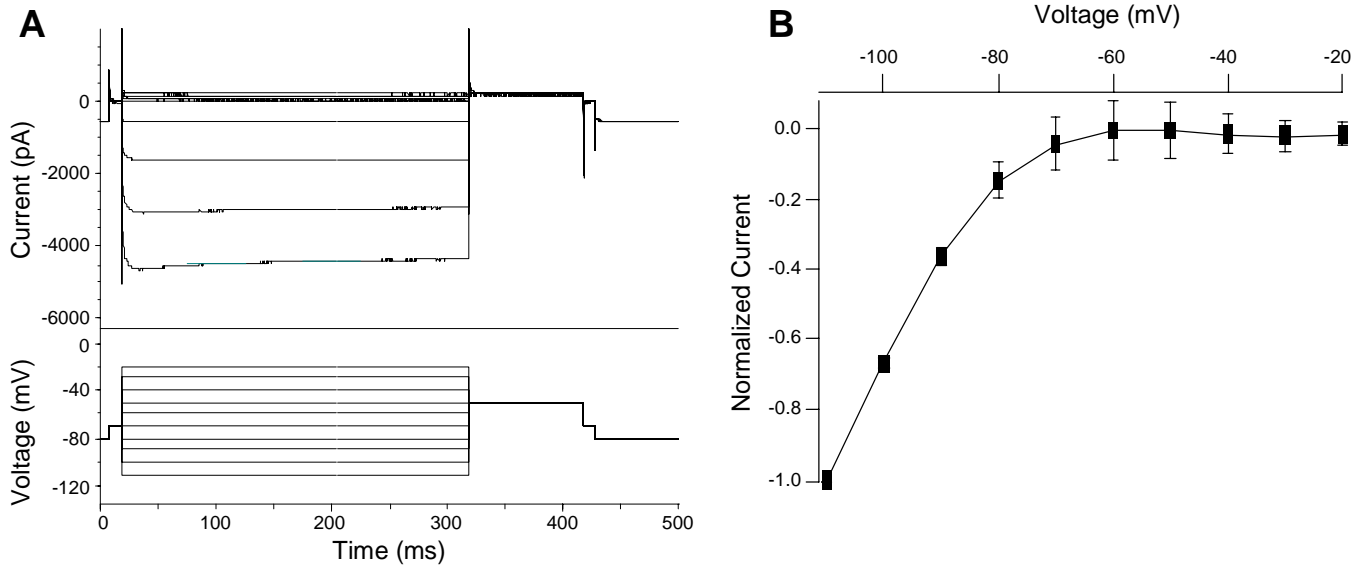


Figure 1. hK_{ir}2.1-HEK Gating Characteristics in Manual Patch Clamp

A: Current-voltage family of current traces (upper panel) elicited by hyperpolarizing test pulses (lower panel) from -20 to -120 mV in 10 mV increments, holding potential -80 mV. **B:** Current-voltage relationship at the end of the test pulse.

2.2 PatchXpress[®]

2.2.1 Throughput Capability in PatchXpress[®]

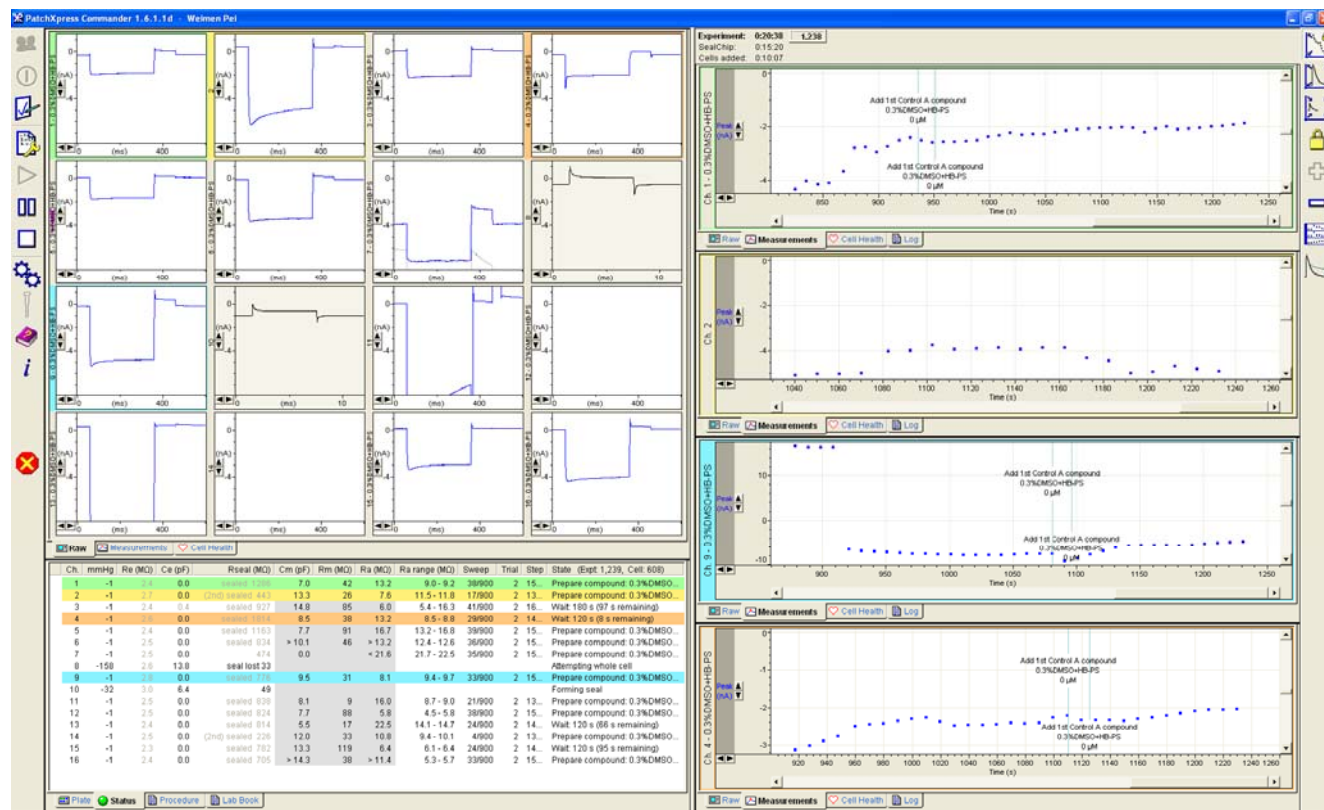


Figure 2. PatchXpress[®] hK_{ir}2.1-HEK Screen Capture.

Throughput capability in PatchXpress[®] depends upon many factors which may result in success rate variability. The screen capture shows a typical PatchXpress[®] experiment. In this example, cells were dispensed to 14 of the 16 seals were formed, whole-cell configuration was achieved in 14 cells, and 11 cells showed characteristic hK_{ir}2.1 current waveforms with little leak current and peak current amplitudes > 1.0 nA.

2.2.2 Representative Data

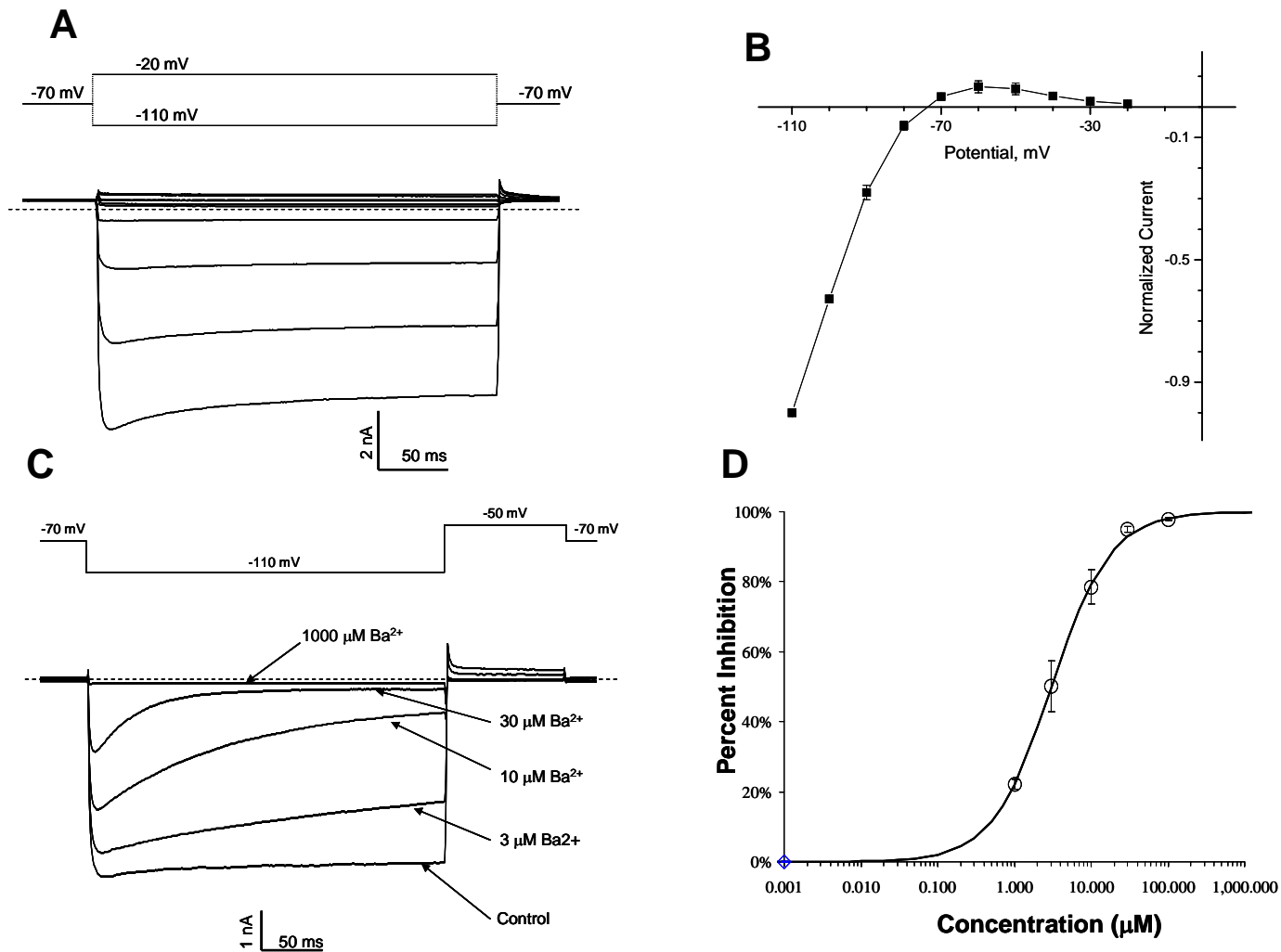


Figure 3. Gating and Pharmacological Characterization of hK_{ir}2.1-HEK in PatchXpress[®]

A: Voltage-dependent activation. Currents elicited by 300-ms test pulses from -110 to +20 mV in 10 mV increments, holding potential -70 mV. **B:** Current-voltage relationship exhibited strong inward rectification with reversal potential -75 mV. **C:** Ba²⁺-induced inhibition of hKir2.1 current evoked by hyperpolarizing pulses to -110 mV. **D:** Concentration-response relationship (Mean ± SEM, IC₅₀ = 3 μM).

3 References

Kubo Y et al., International Union of Pharmacology. LIV. Nomenclature and molecular relationships of inwardly rectifying potassium channels. *Pharmacol Rev.* 2005 57:509-526.