## SUPPLEMENTAL MATERIAL



+80 mV

+40

200 pA

2 ms

100

-80 -40

С 400-

-80



Figure S1. Gating currents recorded from hERG channels expressed in mammalian cells. (A; left) Representative traces showing that no current could be recorded from untransfected tsA201 cells using symmetrical 140 mM NMDG<sup>+</sup> solutions. (Middle) Family of gating currents for a series of pulses from -80 to +80 mV in steps of 10 mV from a holding potential of -80 mV and repolarized back to -80 mV. Neither leak subtraction nor capacitance compensation was used. (Right) Representative gating currents evoked using the same protocol as in the middle panel, except that membrane was repolarized back to -100 mV after leak-current subtraction (P/6) and membrane capacitance compensation. (B) Repolarization-induced currents with uncompensated capacitance (A, middle) were integrated over time and plotted as a function of depolarizing pulse potential. The linear components (dashed lines) represent the linear capacitance of the cell and electrode voltage-clamp system. The nonlinear component, which is related to gating charge movement, was subtracted from the bottom linear regression and fit with a

double Boltzmann function ( $V_{1/2-1} = -36.6 \text{ mV}$ ,  $k_1 = 8.3 \text{ mV}$ ,  $V_{1/2-2} = 42.8 \text{ mV}$ ,  $k_2 = 15.5 \text{ mV}$ , and  $r^2 = 0.99$ ). (C) *Q*-*V* plot of repolarization-induced currents (*Q*-*V*<sub>0ff</sub>), using the same protocol as in A (right). The curve was fitted with a double Boltzmann function ( $V_{1/2-1} = -31.8$ mV,  $k_1 = 8.5$  mV,  $V_{1/2-2} = 30.6$  mV,  $k_2 = 14.1$  mV, and  $r^2 = 0.998$ ).

-30 mV

+10

-+60, +80

m٧

V<sub>1/2-2</sub>: 30.6 mV

-31.8 mV

8.5 mV

14.1 mV

250 pA

k<sub>2</sub>:

1/2-1

 $k_1$ :

Voltage (mV)

0 40 80 120

5 ms

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