

Activation of Slo1 BK channels by Mg²⁺ coordinated between the voltage sensor and the RCK1 domains

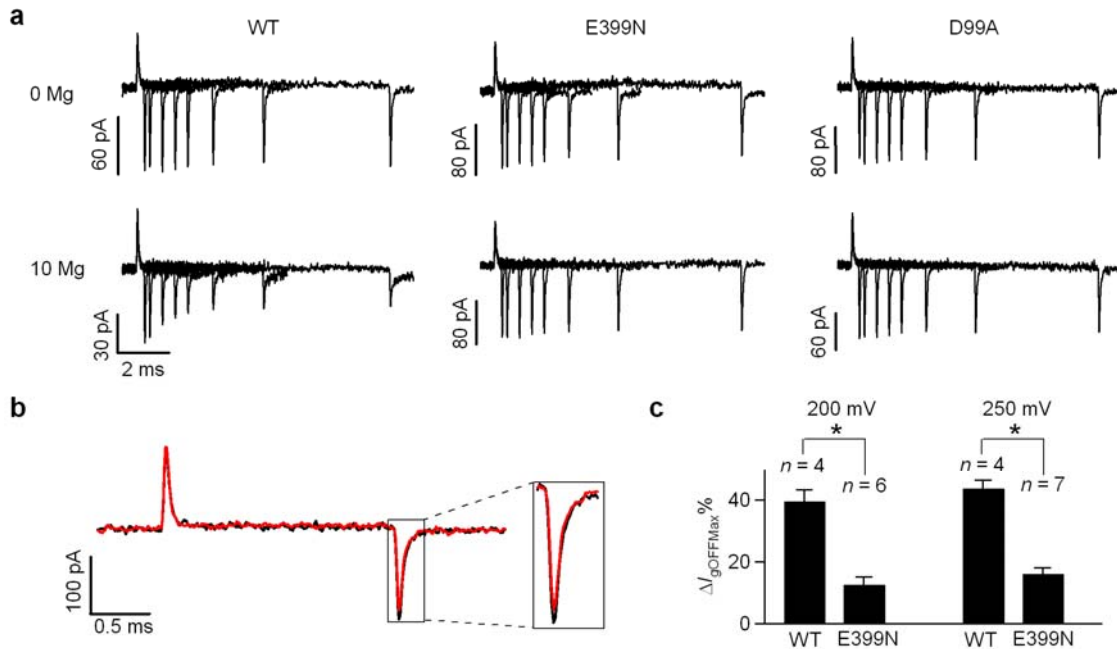
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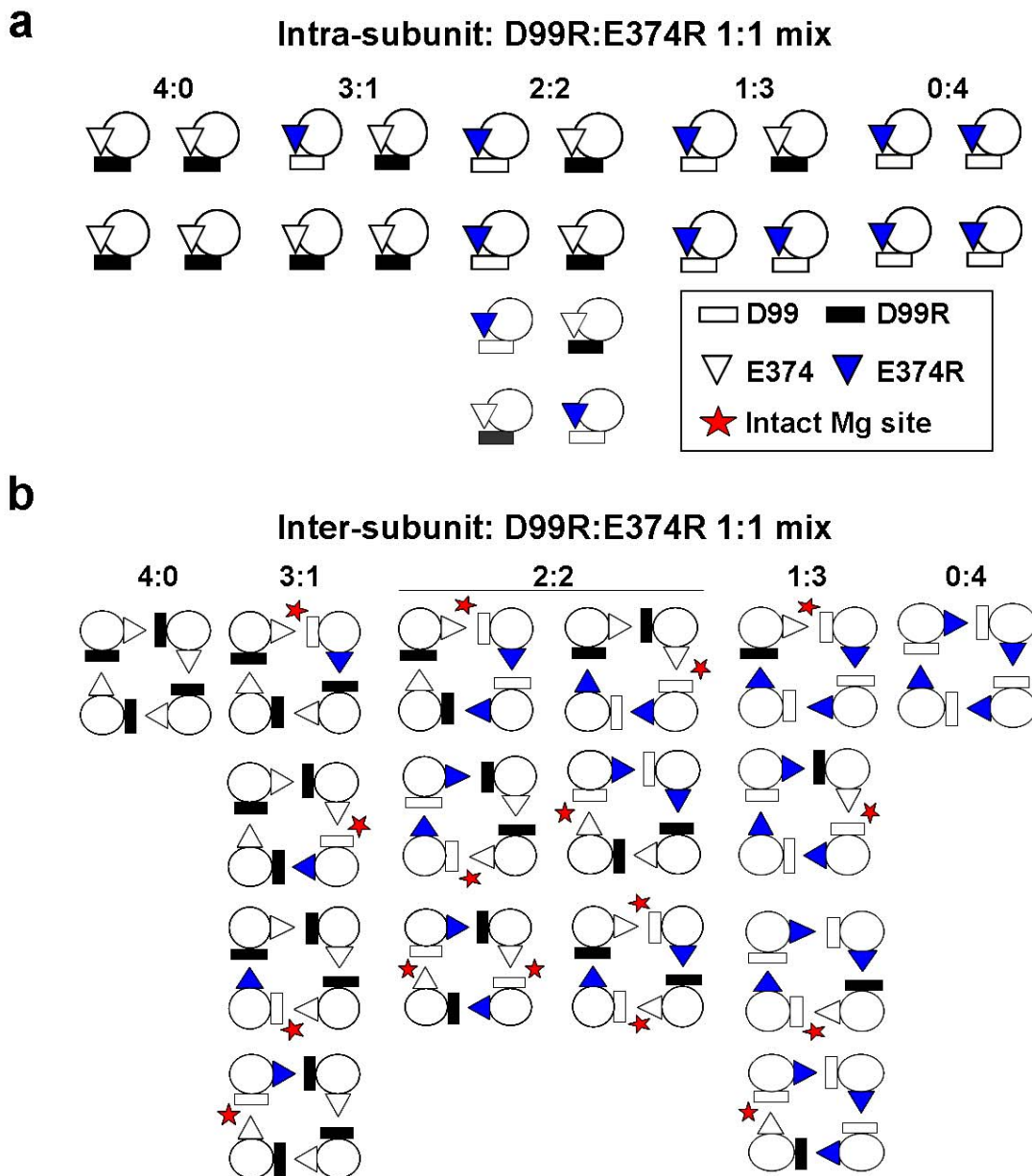
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Supplementary information.



Supplementary Figure 1. Eliminating Mg^{2+} binding alters the response of gating currents (I_g) to Mg^{2+} . **(a)** I_g traces for WT, E399N and D99A mutant channels in 0 (upper panel) and 10 mM $[Mg^{2+}]_i$ (lower panel) elicited by voltage pulses to 200 mV with durations of 0.3, 0.5, 1, 1.5, 2, 3, 5 and 10 ms. I_g traces in 0 and 10 mM $[Mg^{2+}]_i$ were recorded from separate patches. **(b)** Gating current (I_g) traces for E399N mutant channels with (red) and without (black) 10 mM $[Mg^{2+}]_i$ in response to a 2-ms, 250 mV depolarizing pulse. I_g traces of the same patch were first recorded in the absence of Mg^{2+} , and then 10 mM $[Mg^{2+}]_i$. **(c)** Effects of 10 mM $[Mg^{2+}]_i$ on the reduction of peak OFF gating currents ($I_{gOFFMax}$) in response to 2-ms, 200 mV or 250 mV depolarizing pulses. $\Delta I_{gOFFMax} \% = (I_{gOFFMax}(0Mg) - I_{gOFFMax}(10Mg)) / I_{gOFFMax}(0Mg)$. * indicates $p \leq 0.001$.



Supplementary Figure 2. Binomial distribution of D99R:E374R mixture in 1:1 ratio according to the intra- (a) and inter-subunit (b) Mg^{2+} binding site models. (a) The intra-subunit Mg^{2+} binding site model does not predict any intact Mg^{2+} binding site. In this model, the putative Mg^{2+} coordinates in the VSD and the cytoplasmic domain are from the same subunit. (b) The inter-subunit Mg^{2+} binding site model predicts 25% probability to form intact Mg^{2+} binding sites. In this model, the putative Mg^{2+} coordinates in the VSD and the cytoplasmic domain are from neighboring subunits.