INTRODUCING SIMULATED IK1 INTO HUMAN iPSC-CARDIOMYOCYTES USING DYNAMIC CLAMP ON AN AUTOMATED PATCH CLAMP SYSTEM

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Dynamic clamp is a powerful tool involving injection of real-time simulated currents into patch-clamped cells, and has been employed in conventional patch-clamp electrophysiology to inject I_{K1} current into human induced pluripotent stem cell-derived cardiomyocytes (hiPSC-CMs).

hiPSC-CMs are attractive cell types because of their unlimited availability and human origin. However, I_{K1} is expressed at low levels in those cells, hence they display a more depolarized membrane potential than adult cardiomyocytes (CM). Introducing simulated I_{K1} into hiPSC-CMs improves their resting potential and makes them a viable alternative to the scarcely available adult human CMs. Another limitation of hiPSC-CMs is that the ratio of seal resistance (R_{seal}) to membrane resistance (R_m) is small due to smaller capacitance and typically slightly lower R_{seal} in automated patch clamp (APC). Thus, membrane potential dissipation is more pronounced compared to patch-clamp recordings of adult CMs. To correct for that, we implemented a R_{seal} compensation (SRC) mechanism to inject current compensating for the potential dissipation through R_{seal}.

In this study, we combined dynamic clamp with an APC system to demonstrate that I_{K1} conductance can be added to hiPSC-CMs on this platform, while at the same time, applying automatic R_{seal} compensation. Our results show that virtual I_{K1} can be successfully injected into hiPSC-CMs in up to 4 cells simultaneously and that R_{seal} is correctly compensated avoiding overcompensation. Our approach results in more stable resting membrane potentials and improved action potential (AP) shape. Increased I_{K1} resulted in AP shortening and acceleration of the upstroke. We measured native, but small, Ba^{2+}-sensitive I_{K1} in voltage clamp mode in approximately 50% of these cells. Adding a Ca^{2+} channel activator (BayK 8644), or blocker (Nifedipine) caused an increase and decrease of the AP duration, respectively.

In conclusion, combining dynamic clamp and R_{seal} compensation with APC resulted in an enhanced, medium-throughput platform for safety pharmacology.