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Current methodologies for human-induced pluripotent stem cell-derived cardiomyocytes handling

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Human-induced pluripotent stem cells (hiPSC) were first generated by Yamanaka group in 2006. He developed a method to induce pluripotency in somatic cells, such as adult fibroblasts, by the use of core transcription factors that are responsible for the induction and maintenance of pluripotency phenotype. So far, many protocols have been introduced for hiPSC differentiation in several cell types, such as hepatocytes, cardiomyocytes, adipocytes and so on. This innovative model offers a plethora of translational applications. For instance, it allows to study a pathology by isolating fibroblasts from a donor patient in a non-invasive manner, thus maintaining its genetic background. Furthermore, it could be useful as a model for screening and testing of new molecules with potential pharmacological activity.

In our laboratory, we deal with hiPSC-derived cardiomyocytes (hiPSC-CMs), in collaboration with prof. Eschenhagen's group (Hamburg University), which performs the differentiation protocol.

HiPSC-CMs cryopreserved vials are stored in liquid nitrogen until the thawing procedure. The thawing protocol consists in gradually adapting cells to room temperature, in order to plate them in an adequate density on the multiwell support. Since on the day of thawing cells are at day 17 post-differentiation, to reach a complete maturation stage, hiPSC-CMs are maintained in the incubator for two weeks. At 28-35 days post-differentiation, hiPSC-CMs can be dissociated and plated in single cell or monolayer (ML) format. Then, electrophysiological recordings and Ca^{2+} measurements are performed.

By the use of current-clamp techniques, we record action potentials from single cells. Field stimulation recordings, instead, are performed in MLs to allow Ca^{2+} fluorescence measurements. In this way, we are able to investigate action potential shapes, whereas Ca^{2+} recordings could be useful to look at hiPSC-CMs Ca^{2+} dynamics. The outcome of our experiments, compared to data shown in literature, confirm that our culturing protocol is an efficient and valid method for hiPSC-CMs handling.

