Stromal cell models of arrhythmogenic cardiomyopathy

L. Iengo1, A.S. Maione1, V. Meraviglia2, M. Rabino1, M. Chiesa1, V. Catto1, C. Tondo1, G. Pompilio1, M. Bellin3, E. Sommariva1

1Monzino Cardiology Centre, Milan, Italy
2Leiden University Medical Center, Leiden, Netherlands (The)
3University of Padua, Padova, Italy

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Background: Arrhythmogenic cardiomyopathy (ACM) is an inherited heart disease mainly associated with mutations in genes encoding cardiac desmosomes, most frequently Plakophilin-2 (PKP2). Primary cardiac mesenchymal stromal cells (CMSCs) were shown to contribute to the aberrant ACM cardiac remodeling based on their ability to differentiate into adipocytes and myofibroblasts, thus representing a useful in vitro model for the disease. The limitations associated with access to biopsies and finite passages in culture have been overcome by the advancement of stem cell technology, especially human induced pluripotent stem cells (hiPSCs) and their ability to differentiate towards cardiac stromal population. Specifically, CMSCs from hiPSCs provide a virtually unlimited cell sources that are genetically identical to patient cells of origin.

Purpose: The study compares hiPSC-derived stromal cells (hiPSC-D) and primary stromal cells (PRs), to explore the feasibility to use iPSC-D as an alternative in vitro model to study ACM.

Methods: PRs were isolated from ACM patients and healthy donors (HC). hiPSCs from a PKP2 mutated ACM patient (c.2013delC) and a sex/aged matched HC were used to produce iPSC-D. Flow cytometry analysis and specific differentiation induction (osteogenic, chondrogenic, adipogenic and pro-fibrotic) were performed to confirm mesenchymal lineage and multipotency. qRT-PCR and western blot analysis were performed to evaluate the expression of desmosomes, both gene and protein levels. Illumina transcriptomic profiling of both PRs and iPSC-D were compared and enriched pathways were identified by GSEA.

Results: Both PR and iPSC-D cells expressed mesenchymal surface markers and were able to differentiate towards osteogenic, chondrogenic, adipogenic and myofibroblast lineages. Desmosomal genes and proteins were expressed by both populations, showing a general trend toward lower expression of desmosomal markers in ACM compared to HC cells. Overall, higher expression of desmosomes was observed in iPSC-D stromal cells compared to their primary counterparts, independently on the genotype. As occurs for PRs, ACM iPSC-D exhibited an increased propensity to accumulate lipid droplets and collagen compared to HC cells. Moreover, both cell populations shared a highly similar transcriptomic profile, showing a significant linear correlation (R=0.85). Interestingly, PRs were enriched for genes associated with the immune response and differentiation processes, hinting to the hypothesis that an inflammatory stimulus received from the cardiac tissue might activate the innate immunity and drive the cell differentiation fates in the PRs.

Conclusion(s): Both PR and iPSC-D cardiac stromal cells obtained from ACM patients can be used to model the disease, where the choice for the most suitable cell populations can be determined by the experimental strategy and/or the availability of human primary material.