The Estrogen-related Receptor Gamma Promotes Maturation of iPSC-derived Cardiac Myocytes

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The mammalian heart has a tremendous energy requirement to support normal contractile function. Recently, our group discovered that the estrogen-related receptor γ (ERRγ) coordinately controls oxidative energy metabolism and the type I fiber program in skeletal muscle. We hypothesized that this circuit is also operative in heart, serving to match capacity to produce ATP with energy utilizing processes such as contraction and ion transport. To identify downstream targets and pathways of ERRγ in cardiac myocytes, RNA-sequencing (RNA-seq) was conducted in neonatal rat ventricular myocytes (NRVMs) overexpressing ERRγ. ERRγ induced the expression of genes involved in mitochondrial oxidative phosphorylation, fatty acid oxidation, and the TCA cycle. Interestingly, ERRγ also induced expression of a broad program of adult cardiac contractile and structural genes including *Myh6* (αMHC) and Ca^{2+} handling genes such as *Atp2a2* (SERCA2A). ERRγ-mediated activation of adult genes occurred concomitant with the suppression of fetal cardiac and embryonic/perinatal fast skeletal isoform contractile protein gene expression including *Myh7* (βMHC), *Myl1* and *Myh3/8*. These results prompted us to explore the role of ERRγ in differentiation and maturation of human iPS-derived cardiac myocytes (iPSC-CM). The expression level of ERRγ was markedly upregulated during the transition to cardiac lineage commitment in parallel with induction of cardiogenic transcription factors (e.g. NKX2.5 and GATA4). Forced expression of ERRγ in iPSC-CM activated the broad adult cardiac myocyte program including oxidative metabolism and adult contractile gene expression. Of note, ERRγ increased *MYH6* expression while decreasing expression of *MYH7* and the fetal gene markers *NPPA* (ANF) and *NPPB* (BNP). The expression of *TNNI3*, thought to be a marker of mature adult cardiomyocytes, was also induced by ERRγ. Conversely, CRISPR-Cas9n deletion of ERRγ in iPSC-CM resulted in diminished mitochondrial energy metabolic and adult cardiac contractile gene expression. These data provide evidence that ERRγ is a key nodal regulator of cardiac myocyte determination and terminal maturation. ERR signaling is a candidate target for driving iPSC-CM to the “adult” cardiac myocyte phenotype.

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