A functional genomics pipeline for mammalian heart regeneration

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The neonatal mammalian heart can regenerate following multiple forms of cardiac injury, which is in stark contrast to the extremely limited regenerative potential of the adult heart (Porrello *et al.* 2011; Porrello *et al.* 2013). Regeneration of the neonatal heart is associated with induction of cardiomyocyte proliferation, angiogenesis, immune cell infiltration and fibrotic scar regression but the molecular basis for neonatal heart regeneration remains poorly defined. Here, we perform the first comprehensive analysis of cellular transcriptomes during heart development and regeneration by comparing RNA-seq data derived from isolated cardiomyocytes, fibroblasts, endothelial cells and leukocytes from infarcted and sham-operated mice at postnatal day 1 (P1; regenerative) and postnatal day 56 (P56; non-regenerative). These studies reveal specific genes regulated during mammalian cardiac regeneration, as well as exquisitely controlled cell-specific transcriptional networks governing heart development and regeneration. In addition, a number of protein-coding genes and non-coding RNAs that are implicated in cardiomyocyte proliferation and extracellular matrix remodeling were identified, including miR-29a, which was found to be specifically up-regulated in the regenerative neonatal (but not adult) heart following myocardial infarction (MI). Gain- and loss-of-function studies in neonatal and adult mice revealed a critical anti-fibrotic role for the miR-29 family following MI. Liposome-mediated delivery of miR-29a to the adult heart following MI resulted in improved cardiac function, reduced fibrosis, prevented adverse cardiac remodelling and was associated with repression of a number of fibrotic genes including Col1a1 and *Col3a1*. Finally, *in vitro* studies using bioengineered human heart tissue confirmed the therapeutic potential of miR-29a in a human setting. These findings define a transcriptional signature for neonatal heart regeneration, identify miR-29a as a potential therapeutic target for heart repair and provide a framework for the identification of key genes for cardiovascular regeneration.

Porrello ER, Mahmoud AI, Simpson E, Hill JA, Richardson JA, Olson EN, Sadek HA. (2011) Transient regenerative potential of the neonatal mouse heart. *Science* **331,** 1078-80.

Porrello ER, Mahmoud AI, Simpson E, Johnson BA, Grinsfelder D, Canseco D, Mammen PP, Rothermel BA, Olson EN, Sadek HA. (2013) Regulation of neonatal and adult mammalian heart regeneration by the miR-15 family. *Proc Natl Acad Sci USA* **110,** 187-92.