**LINEAGE MARKERS IN TRACKING CARDIAC FIBROBLAST FATE IN VIVO AND IN VITRO**

**C.S. Long**

Denver Health Medical Center, Denver, CO, USA

We have utilized two transgenic lines first reported in order to investigate heterogeneities among cardiac fibroblast and myofibroblast cell phenotypes in hepatic fibrosis (Hepatology40:1151, 2004). These are novel transgenic mice that express lineage-sensitive promoter:reporter constructs wherein the smooth muscle actin promoter drives the dsRed Fluorescent Protein (SMA:RFP) and the collagen promoter drives the expression of the enhanced Green Fluorescent Protein (Coll I:EGFP). Using these lines we have investigated changes in fibroblast phenotype both in vivo as well as in cell culture. Freshly isolated cardiac fibroblasts from have been analyzed by flow cytometry from GFP+RFP+ double-lineage reporter mice following ischemia-reperfusion. Notably, the median intensity of GFP+ cell fluorescence is significantly increased in the I-R injured mouse, reflecting enhanced Coll I promoter activity. The corresponding flow cytometric analysis for non-injured versus post-injury RFP+ reporter mice indicates that I-R injury induces an increase in the population of RFP positive cells, consistent with observations of increased myofibroblasts post-injury. Phenotypes of cardiac fibroblasts from double lineage reporter mice have also placed in culture and exposed to cytokines known to be expressed in the post-injury heart. Notably, GFP+RFP- cells preserve their phenotype over this time frame. By contrast, GFP+RFP- cells from post-injury heart show robust SMA:RFP+ activation within this culture interval and this is increased following TGFbeta treatment. We believe this model is ideal for understanding the phenotypic modulation of fibroblast phenotype in response to injury and therapies designed to improve post-injury remodeling.