Coronary artery disease (CAD) remains the leading cause of death in the western hemisphere, in part because of the still-limited options for the treatment of diffuse CAD, myocardial infarction, and congestive heart failure. Cardiac stem cell therapy, which theoretically repopulates otherwise permanently scarred myocardium with contractile cells, has been embraced as a new approach to treating end-stage heart disease, but potential limitations to this strategy include extensive (>90%) loss of implanted cells and limited integration of these exogenous cells into the host myocardium. Moreover, an ideal adult cardiac stem-cell implant has not yet clearly been identified, even though embryonic stem cells appear to be the most aptly suited for use as a cardiac-implant cell type. The creation of induced pluripotent stem cells (iPSCs) and the generation of cardiomyocyte-like cells from iPSCs appear to have been breakthroughs in this field, but recent iPSC tumorogenicity and immunogenicity might ultimately indicate limits to the clinical applicability of iPSCs.

The recent discovery that a trio of “cardio-differentiating” transcription factors (Gata4, Mef2c, and Tbx5 [GMT]) can be used to generate “induced cardiomyocyte” (iCM) cells directly from somatic cells offers the exciting new possibility of generating autologous cells that possess characteristics at least consistent with those of cardiomyocyte phenotype production. Perhaps more important, this novel “cellular reprogramming” regenerative strategy offers the intriguing potential to convert myocardial scar fibroblasts into functional iCMs in situ, avoiding the challenges of exogenous cell-host integration and perhaps returning regions of myocardial infarction into regions of functional myocardium.

Given that ischemia has been shown to adversely affect the function or survival of stem cells implanted into regions of myocardial infarction and ischemia, we have postulated and have subsequently been able to show that angiogenic “prevascularization” of the infarct zone—via adenovirus encoding of all 3 major isoforms of vascular endothelial growth factor (VEGF)—leads to better cellular reprogramming efficacy than does the administration of transdifferentiating transcription factors alone. More specifically, VEGF-mediated scar vascularization, followed by the administration of a (lentivirus) cocktail of GMT transcription factors to induce in situ iCM generation, led to better functional improvements in a small-animal myocardial infarction model than GMT administration alone. In these studies, GMT administration also reduced by half the extent of fibrosis (in comparison with control groups) and reduced by 4-fold the number of scar-producing myofibroblasts detected in the infarct zone. The GMT-treated animals similarly manifested a greater density of cardiomyocyte-specific marker MYH7+ cells than did the control animals.

Left ventricular ejection fraction (LVEF) was also significantly improved after GMT administration, with an average improvement in LVEF of approximately 0.20 in treated animals, compared with a decrease in LVEF in untreated animals. Moreover, this improvement in LVEF was 4 times greater in animals receiving GMT with VEGF pretreatment than in those without VEGF pretreatment.

The dramatic increases in ventricular function and decreases in fibrosis seen in treated animals—both in our study and in earlier studies—appear to far exceed increases in the number of newly generated iCMs. This suggests that changes in wall stress or reductions in fibrosis might also contribute to the significant improvements in LVEF observed in treated animals. On the basis of our preliminary data, we might speculate that a paracrine effect of a relatively limited number of iCMs underlies the reduction in fibrosis; this could be due to the expression, by iCMs or host cells, of chemokines such as basic fibroblast growth factor and tissue inhibitor of matrix metalloproteinase.
ase (TIMP)-2, which have been reported to limit or reduce fibrosis.

Alternatively, it is conceivable that cellular reprogramming or the administration of VEGF transgenes diverts resident fibroblasts away from their normal postinfarct differentiation into myofibroblasts (known to produce fibrosis via expression of collagen and other extracellular matrix components) and directs them toward a more benign role as iCMs. The 4-fold reduction of myofibroblast populations that we observed in GMT-treated animals versus control animals, consistent with the trend in the reduction of fibrosis, supports this supposition. Theoretically, alternative processes such as myofibroblast apoptosis and repressed function (that is, decreased extracellular matrix component expression) could also play a role in such mechanisms.

Although these results are encouraging, the relatively inefficient transdifferentiation rate of fibroblasts into iCMs might present a barrier to the successful implementation of a cellular reprogramming clinical intervention. Recent findings that human fibroblasts are more resistant than rat or mouse cells to transdifferentiation appear to present a similar barrier. Yet if these initial cellular reprogramming studies are ultimately fully validated in additional preclinical and then clinical trials, in situ cellular reprogramming will offer a potentially important alternative to cardiac transplantation and mechanical support in the treatment of patients with end-stage heart failure.

References