



The promise of adult mesenchymal stem cells for acute myocardial infarction

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The treatment of acute myocardial infarction (AMI) has evolved substantially over the past 3 decades. After an initial improvement in the mortality rate of AMI due to thrombolytics for pharmacologic reperfusion [1], further clinical advances were made employing primary percutaneous intervention [2,3]. While major effort is expended on reducing door-to-balloon time, reperfusion is rarely achieved within the first 'golden hour' of symptoms so as to actually abort AMI. As a result, most patients are left with a burden of infarcted myocardium, and therefore the substrate for ventricular remodeling, heart failure and malignant arrhythmias. In addition, much of the current therapy for patients post-AMI, including antiplatelet therapy, statins, β -adrenergic antagonists and modulators of the renin-angiotensin axis, are aimed at the prevention of recurrent cardiovascular events and the amelioration of left ventricular remodeling. Until recently, a pathophysiologic paradigm held that necrotic myocardial tissue lacked any chance of functional recovery. This paradigm is currently undergoing revision with the advent of stem cell therapy, which holds the promise of regeneration of viable myocardial tissue.

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Over the past 10 years, various cell-based therapies have undergone preclinical and early, small clinical studies with the hopes of regenerating viable myocardial tissue. The cell preparations include embryonic stem cells, skeletal myoblasts, autologous whole adult bone marrow, endothelial progenitor cells, umbilical cord blood stem cells, resident cardiac stem cells and mesenchymal stem cells (MSCs) [4]. Although there is considerable variability in the early studies, including the types of cells

used, the method of delivery, dosing of cells and time course of treatment, early results have been moderately encouraging, with a 3–4% improvement in left ventricular ejection fraction (LVEF) [5]. Intracoronary infusion of enriched bone marrow-derived progenitor cells (BMCs), which resulted in a 2–3% improvement in LVEF in patients with AMI, actually yielded more impressive clinical outcomes, reducing the combination of death, reinfarction and the need for heart failure hospitalization by an absolute 10% reduction [6,7]. However, the use of BMCs is limited by the need to obtain a bone marrow aspirate from each individual patient, and there is interpatient variability in the quality of BMCs and thus the clinical response. MSCs are found in bone marrow, muscle, skin and adipose tissue, and have the potential to differentiate into tissues of mesenchymal origin, including muscle, fibroblasts, bone, tendon, ligament and adipose tissue [8]. Precultured bone marrow-derived human MSCs (hMSCs) represent an alternative approach to BMCs. In comparison with autologous BMCs, allogeneic MSCs have various advantages that make them an attractive vehicle for cardiovascular cell therapy. MSCs lack major histocompatibility complex II cell surface antigens and costimulatory molecules, which make them a candidate for use as an allogenic graft [9]. Therefore, allogeneic hMSCs may be prepared *a priori* from healthy donors and administered any number of ways, including intravenous, intracoronary and by direct endomyocardial injection. Preclinical studies suggest that the level of cell retention in the heart after intravenous infusion is low [10]; nonetheless, it remains an attractive delivery option given the ease of administration. It is theorized that MSCs migrate and home to areas of injury, based on various mechanisms, including the stromal cell-derived factor-1/chemokine CXC ligand axis and the expression



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of various inflammatory and chemotactic markers including TNF- α , TNF-related apoptosis-inducing ligand and osteoprotegerin. The ability of MSCs to migrate to injured myocardium forms the basis for their intravenous infusion in post-myocardial infarction (MI) patients [11,12]. Mechanistically, we still continue to elucidate the exact mechanisms by which MSCs may lead to myocardial regeneration whether it is by a direct cellular response [13,14] or by a paracrine-mediated phenomena [15,16], inhibiting scar formation and promoting endogenous healing. Recent work in swine models correlates MSC engraftment to a decrement in infarct size and functional recovery, with trilineage differentiation into myocytes with coupling to host myocardium by gap junctions, as well as vascular and endothelial components [13]. Engraftment and differentiation may occur to the greatest extent in the 'penumbra' border zones between infarcted tissue and viable myocardium [13,17].

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While preclinical studies of MSCs were promising, there remained clinical questions of efficacy and safety with the use of allogeneic MSCs. Some particular safety concerns with the use of stem cells include the risk of tumorigenesis and ectopic tissue formation, direct organ toxicity from microvascular dysfunction, arrhythmogenesis and adverse effect on pulmonary function. Small human studies using intracoronary MSCs in post-MI patients demonstrated a significant improvement in global and regional left ventricular function with a significant reduction in the size of the perfusion defect as compared with placebo [18]. To further evaluate the safety and efficacy of MSCs in the treatment of AMI, a recent Phase I randomized, doubleblind, placebo-controlled study was performed in 60 AMI patients using intravenous allogeneic adult human MSCs, Prochymal (Osiris Therapeutics, Inc., MA, USA), within 10 days of the AMI [19]. The safety of intravenous mesenchymal cell therapy at various doses was established. The adverse event rate was lower in the MSC-treated cohort. In addition, the MSC-treated cohort actually demonstrated a lower rate of ventricular arrhythmias and improved pulmonary function when compared with the placebo-treated group. At 3 months, echocardiography demonstrated a greater improvement in

LVEF in hMSC-treated patients as compared with placebo-treated patients ($5.9 \pm 1.8\%$; $p = 0.003$ vs $4.4 \pm 1.8\%$; $p = 0.021$ in placebo). In the subgroup of patients with anterior AMI, this treatment effect was magnified ($7.3 \pm 3.4\%$; $p = 0.044$ vs $3.4 \pm 3.4\%$; $p =$ not significant in placebo) at 6 months. In a cardiac MRI substudy, hMSC treated patients experienced a $5.2 \pm 1.9\%$ ($p < 0.003$) improvement in LVEF at 12 months, which was accompanied by an improved overall global assessment, as compared with no improvement in the placebo-treated group.

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Going forward with clinical trials of MSCs for AMI, there are various questions that remain to be answered. First, what is an adequate and clinically meaningful end point to evaluate the efficacy of therapy? Acute cardiovascular mortality after AMI has significantly diminished given current treatment strategies. As the treatment of AMI improves and mortality decreases, it is becoming harder for emerging strategies to demonstrate an incremental mortality benefit above current standard of care; clinical trials are becoming larger, requiring more patients, and are becoming more technically challenging and costly. Therefore, secondary end points are often used as a marker of clinical benefit. Acute survival and long-term cardiovascular event rates are clearly associated with residual left ventricular systolic function (LVSF) post-AMI [20]. LVSF and infarct size have become the *de facto* primary end point for many of the trials. Furthermore, MRI may supplant echocardiography or ventriculography as the gold standard for the evaluation LVSF and infarct size. Although prior studies demonstrated a modest improvement in LVSF, what is the clinical significance of a 2–3% improvement in LVEF? Since the prevention of remodeling is the pathophysiological target, measures of left ventricular chamber dimension may be superior to LVEF as global markers of efficacy. Going forward, clinical end points will become essential to prove that even if there is an improvement in LVSF and/or infarct size by echocardiography or MRI, these are clinically relevant results. The Reinfusion of Enriched Progenitor Cells

and Infarct Remodeling in Acute Myocardial Infarction (REPAIR-AMI) study suggests that there are clinical benefits, including a reduction in the composite of mortality, MI, need for repeat revascularization and heart failure hospitalizations [6]. Follow-up functional studies such as a metabolic stress test and or 6-min-walk test would also be helpful to objectively quantify any symptomatic improvement.

Do all AMI patients benefit equally from stem cell therapy? If not, which patients stand to benefit the most? It is reasonable to expect that the highest risk patients, specifically those with large AMIs, anterior infarcts and late presenters with evidence of significant left ventricular systolic dysfunction and/or infarct size, would stand to benefit the most from MSCs therapy. Indeed, in prior studies, it appears that the patients with lower LVEF at baseline, as well as anterior MI, demonstrate the greatest improvement in LVEF after treatment [6,19]. Should trials be conducted exclusively for these high-risk subgroups? It may be these patients who stand to benefit the most from any incremental therapy.

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What is the optimal timing for administration of the stem cells? In the REPAIR-AMI trial, there was a timing-related effect, with only

patients injected after 4 days demonstrating a benefit [6]. The myocardial milieu and signaling cascades at the time of administration of the cells is critical for engraftment and differentiation of the cells to viable myocardial tissue. But how this relates to timing of stem cell delivery, and if and how these signals can be modified safely, still continues to be elucidated by ongoing research.

Looking forward, there are many questions yet to be answered concerning the clinical use of MSCs, and stem cells in general, for AMI. Hopefully, some answers to these questions will be provided by both ongoing bench research and clinical trials. In addition, several studies are looking at the use of MSCs not only in AMI, but for myocardial regeneration in other conditions including chronic ischemia and nonischemic dilated cardiomyopathy. Importantly, the Osiris MSCs are currently being tested in a Phase II study. If MSCs hold to their promise, we are at the brink of a whole new treatment modality for myocardial regeneration in acute and chronic myocardial disease.

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