Paracrine-Mediated Systemic Anti-Inflammatory Activity of Intravenously Administered Mesenchymal Stem Cells

A Transformative Strategy for Cardiac Stem Cell Therapeutics

Stephen E. Epstein, Dror Luger, Michael J. Lipinski

Stem cell therapy as a treatment option for acute myocardial infarction or heart failure caused by ischemic or nonischemic cardiomyopathy has focused on direct cardiac delivery of stem cells to facilitate cardiac engraftment. Once engrafted, it was believed that these cells would either transdifferentiate into new functioning myocytes or stimulate the expansion of resident myocardial stem cells. However, a sea change in thinking about cell therapy in cardiovascular disease has occurred as mounting evidence indicates that (1) inflammation is a major mechanism contributing to the progressive myocardial dysfunction seen in patients post-acute myocardial infarction and in patients with cardiomyopathy, and (2) systemic paracrine-mediated anti-inflammatory effects of stem cells can drive beneficial cardiac effects in these diseases. These concepts lead to a potentially transformative strategy that intravenous delivery of stem cells, through systemic anti-inflammatory mechanisms, improves myocardial function and thereby obviates the need for invasive methods of stem cell delivery.

Until recently, the prevailing view of the mechanism responsible for any potential benefit of stem cells in patients with acute myocardial infarction (AMI) or with heart failure (HF) caused by ischemic or nonischemic cardiomyopathy (ICM/NICM) was that benefit derived from local effects—once engrafted in damaged myocardium, the stem cells either transdifferentiate into functional myocardium, stimulate resident myocardial stem cells to expand, and repopulate the heart with functioning myocytes or secrete substances leading to myocardial healing. This mechanistic perspective implied that the greater the number of engrafted cells in the myocardium, the greater the cardiac benefit. Because few intravenously administered stem cells engraft in injured myocardium, invasive strategies providing direct delivery of stem cells to the heart were uniformly adopted. This necessarily involved either catheter-based delivery (intracoronary or transendocardial injection) or surgical delivery (direct intramyocardial injection).

A transformative concept relating to stem cell treatment of patients with AMI or ICM/NICM has recently evolved—that is, the intravenous administration of stem cells will improve left ventricular (LV) function in patients with AMI or with ICM/NICM. The concept's validity derives from compelling preclinical data and from 2 new mechanistic insights: first, that the progressive deterioration of cardiac function seen both in AMI and ICM/NICM is in part caused by an excessive, persistent inflammatory response[1–4]; and second, that if stem cells improve cardiac function, improvement is not caused by repopulating the myocardium with new myocytes, but rather by stem cells secreting numerous molecules with a diverse array of activities.[5,6] These paracrine activities include those related to angiogenesis, tissue healing, apoptosis, mitochondrial dysfunction, microvascular dysfunction, collagen deposition,[1] and, perhaps most importantly, through potent systemic anti-inflammatory actions.[1,2,7] These anti-inflammatory effects cause in vivo suppression of both innate and adaptive immune responses to AMI.[1–4,7]

An interesting concept on initiation and propagation of the inflammatory response resides in the concept of a cardiosplenic axis (Figure). Myocardial injury (indeed, injury involving any tissue) signals the spleen, either through neural or humoral pathways, causing increased splenic inflammatory cell populations, with subsequent release of these cells into the circulation where they then home to sites of injury.[8] A cardiosplenic axis may also participate in HF, as spleens in mice with ICM are enlarged, and splenectomy not only decreases cardiac macrophages and dendritic cells but also attenuates adverse LV remodeling.[9]

A critical issue solved by the intravenous administration of stem cells arises from the improbability that the progressive cardiac dysfunction experienced by patients with AMI or with ICM/NICM will be cured by a single injection of any therapeutic. Although inflammation and other causally contributory mechanisms can be transiently suppressed, they likely persist long term with the capacity to cause continuing damage. Thus, to counteract progressive pathophysiologic changes, multiple stem cell injections will probably be needed. Recent preclinical studies suggest the validity of this concept. Thus, Bolli laboratory demonstrated that 3 administrations of c-kit^{POS} cardiac progenitor cells, 35 days apart, into the LV cavity of rats with 30-day-old myocardial infarctions had cumulative beneficial effects on LV function. Similar results were observed in mice with 3-week-old infarct.[10]

The opinions expressed in this article are not necessarily those of the editors or of the American Heart Association.

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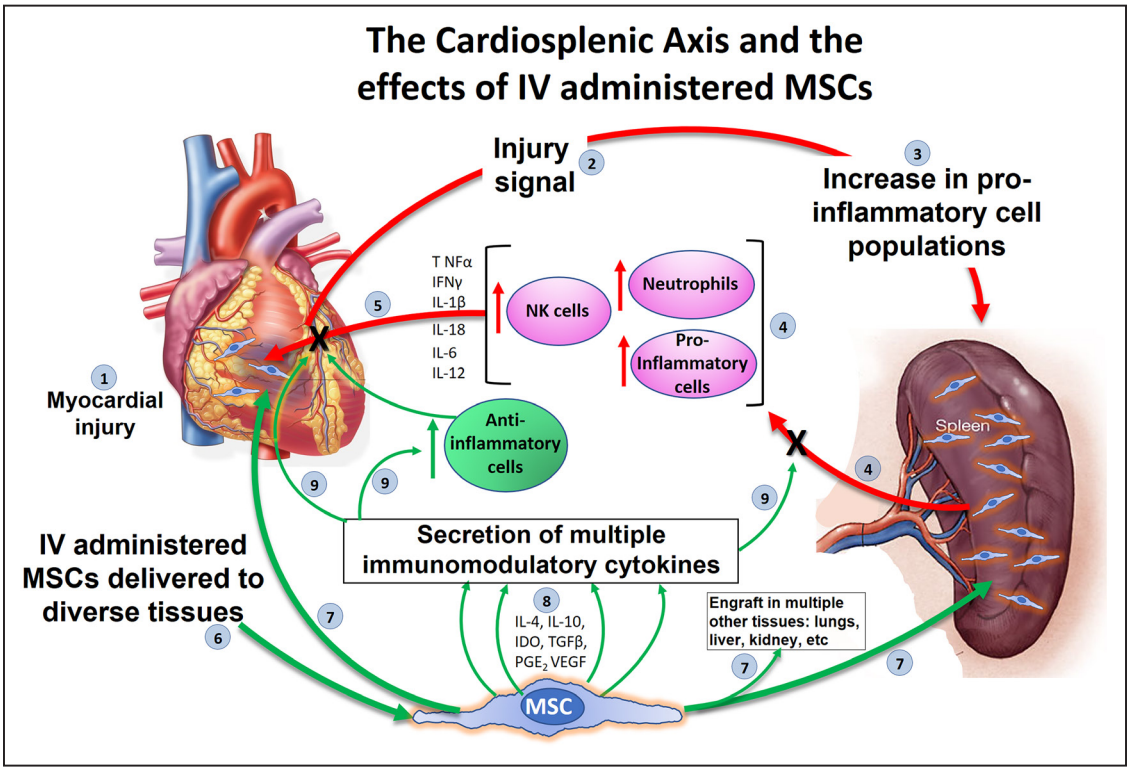

Figure. Inflammatory and immune responses, influencing a diverse array of pathways, importantly contribute to progressive myocardial dysfunction and adverse left ventricular (LV) remodeling observed in patients post-acute myocardial infarction (AMI) or with cardiomyopathy. Intravenously administered mesenchymal stem cell (MSCs) inhibit many of these proinflammatory pathways, an effect constituting one of the major mechanisms responsible for the beneficial myocardial effects of this intervention. IL-1β indicates interleukin 1 beta; IL-6, interleukin 6; IV, intravenous; NK, natural killer; PGE2, prostaglandin E2; TNF-α, tumor necrosis factor alpha; and VEGF, vascular endothelial growth factor.

Anti-Inflammatory Effects of Stem Cells in AMI

Mesenchymal stem cell (MSCs) have emerged as an interesting candidate cell type for the therapeutic agent of choice because of the coupling of 2 important concepts: inflammation importantly contributes to the progressive decline in cardiac function in patients with AMI and with HF,[1] and MSCs are potent modulators of the inflammatory responses occurring after an AMI. This latter activity has been particularly well documented for MSCs,[5,7,11] less so for other cell types. Our remarks will therefore focus mainly on the potential efficacy of MSCs.

As examples of the anti-inflammatory effects of MSCs, intramyocardial injection of MSCs in a rat model of AMI reduces myocardial levels of tumor necrosis factor alpha, interleukin 1 beta, interleukin 6, matrix metalloproteinase 1, and tissue inhibitor of metalloproteinase 1 and reduces collagen deposition. These changes are associated with reduced infarct size and improved LV function.[11] In addition, it was shown in mice with AMI that intravenously administered MSCs improve LV function and decrease inflammation through factors secreted by MSCs lodged in organs outside the heart.[12] Because immunocompromised (non-obese diabetic/severe combined immunodeficiency) mice were used in this study, it is uncertain what the paracrine-mediated immunomodulatory and salutary cardiac effects of the MSCs would be in an immunocompetent animal.

Our group addressed this issue and demonstrated in immunocompetent mice that intravenously administered human MSCs 24 hours after AMI decreased splenic and myocardial natural killer cells, as well as myocardial neutrophils. These anti-inflammatory effects were associated with significantly attenuated adverse LV remodeling in mice with large infarcts.[2]

Thus, compelling preclinical evidence indicates that intravenously administered MSCs improve cardiac outcomes in AMI and appear to do so, at least in part, through anti-inflammatory activity. The beneficial myocardial functional effects of the paracrine-mediated anti-inflammatory activities have their greatest impact on large infarcts, where inflammation is greatest.[2]

The capacity of MSCs to decrease natural killer (NK) cell activity and other components of the innate immune system in the setting of AMI is well known. We were particularly interested in the question of whether MSC-induced reduction in NK cells played a causal role in improving myocardial function after AMI, as NK cells play a major role in coordinating the innate immune response. We proved the validity of this hypothesis by demonstrating that when NK cells were depleted, not by MSCs but by an anti-NK cell antibody administered 24 hours before AMI, infarct size significantly decreased and LV function significantly improved.[2]

Whereas NK cells and neutrophils are early responders to AMI, mononuclear cells play a critical role later in the reparative process. Interestingly, intravenously injected human MSCs in immune-deficient mice 48 hours after AMI significantly decreased proinflammatory macrophages and increased the portion of pro-reparative macrophages in the circulation and within the heart.[12]

Anti-Inflammatory Effects of MSCs in HF

Persistent inflammation seems to be a major contributor to progressive cardiac dysfunction not only in patients after AMI, but also in those with HF.[1,4] Importantly, intravenous MSC administration significantly improves LV function in a murine model of ICM.[2] This was accompanied by significant decreases in splenic myeloid cells, suggesting that MSCs reduce inflammatory processes in chronic cardiomyopathy. There was also a trend toward reduced splenic NK cells.

An immunomodulatory effect of MSCs was evident in a recent clinical trial of patients with NICM. Intravenously administered MSCs significantly reduced peripheral NK cells; most importantly, the degree of NK cell reduction correlated with the degree of improvement in LV function.[13] On the basis of our studies, we speculate that, as a regulator of both the innate and adaptive immune response in cardiovascular disease, NK cell modulation or depletion may be a target for new therapeutics.

An important study relating to the immunomodulatory effects of MSCs was recently published by Naftali-Shani et al.[14] These investigators demonstrated that MSCs derived from the hearts of mice with persistent LV dysfunction, when tested in vitro, changed to an inflammatory phenotype, and when administered into the periinfarction region of mice 28 days after AMI did not improve LV function—they actually worsened anterior wall thinning. Importantly, MSCs derived from a noninflamed tissue—subcutaneous fat—did not switch to an inflammatory phenotype. Of relevance to this is the fact that the vast majority of MSCs administered intravenously to mice with AMI embed in tissues other than the heart.[2] Although highly speculative at this point, these observations suggest that intravenously administered MSCs actually may lead to greater myocardial benefit than would direct delivery of the cells to the myocardium.

Conclusions

Compelling data indicate that chronic inflammation contributes to the progressive myocardial dysfunction occurring in patients post-AMI and in patients with ICM or NICM. It has now also been definitively demonstrated in preclinical models that intravenously delivered MSCs to mice with either AMI or ICM improves myocardial function, an effect caused at least in part by systemic MSC-induced anti-inflammatory effects. If these preclinical results are proven translatable to clinical settings, the strategy of intravenous delivery of MSCs to patients with AMI or with ICM/NICM for improving myocardial function or preventing its progression, as well as for providing a safe and efficient means for repeated administration of the MSCs, will be transformative to the field of cardiovascular stem cell therapeutics.

Disclosures

S.E. Epstein is a consultant to and holds equity interest in CardioCell, LLC. The other authors report no conflicts.

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