Published in final edited form as:
Prog Cardiovasc Dis. 2007 ; 50(1): 18–30.

The Bone Marrow - Cardiac Axis of Myocardial Regeneration

Ronglih Liao, **Otmar Pfister**, **Mohit Jain**, and **Frederic Mouquet**
Cardiac Muscle Research Laboratory, Cardiovascular Division, Department of Medicine, Brigham and Women's Hospital, Harvard Medical School, Boston, MA

Abstract

Congestive heart failure remains the leading cause of morbidity and mortality in the developed world. Current therapies do not address the underlying pathophysiology of this disease, namely the progressive loss of functional cardiomyocytes. The notion of repairing or regenerating lost myocardium via cell based therapies remains highly appealing. The recent identification of adult stem cells, including both cardiac stem/progenitor cells and bone marrow stem cells, has triggered an explosive interest in utilizing these cells for physiologically relevant cardiomyogenesis. Enthusiasm for cardiac regeneration via cell therapy has further been fueled by the many encouraging reports in both animals and human studies. Further intensive research in basic science and clinical arenas are needed in order to make this next great frontier in cardiovascular regenerative medicine a reality. In this review, we focus on the role of bone marrow derived stem cells and cardiac stem/progenitor cells in cardiomyocyte homeostasis and myocardial repair and regeneration, as well as provide a brief overview of current clinical trials utilizing cell-based therapeutic approaches in patients with heart disease.

Despite advances in the treatment of congestive heart failure (CHF), morbidity and mortality remain inappropriately high [1] This medical epidemic has only continued to escalate, given an overall aging population and the greater number of patients surviving an initial myocardial infarction (MI). The pathophysiology of post-MI heart failure is driven by the loss of cardiomyocytes, either due to acute ischemic necrosis or chronic apoptosis, and the inability of the remaining cardiomyocytes to adequately compensate. As such, the concept of repairing or regenerating lost myocardium via cell based therapies (so termed "cardiomyoplasty") remains highly appealing. Over the past decade, much research has focused upon identifying the ideal cell type with which to promote myocardial regeneration. Thus far, several cells types have been investigated in animal models including, but not limit to, fetal cardiomyocytes [2,3], fibroblasts, skeletal myoblasts [4-7] and endothelial progenitor cells [8,9]. Results with all these cell types have generally been encouraging with regards to beneficial post-MI remodeling; albeit none have resulted in definitive differentiation into physiologically-significant, force-generating cardiomyocytes. Five years ago, striking reports suggested, for the first time, that bone marrow derived stem cells (hematopoietic stem cells) may have the potential to regenerate significant amounts of lost myocardium in mice following MI [10,11], creating overwhelmingly enthusiasm and subsequent skepticism in the field of cardiac repair and regeneration. More recently, the identification of resident cardiac stem/progenitor cells by several groups, including ours [12-15], has brought about a second wave of the scientific interest. These findings have advanced our understanding of myocardial biology and physiology and have introduced

For Correspondence: Dr. Ronglih Liao, Cardiac Muscle Research Laboratory, Cardiovascular Division, Department of Medicine, Brigham and Women's Hospital, Harvard Medical School, 77 Avenue Louis Pasteur, New Research Building, NRB 431, Boston, MA 02115, USA, Email: rliao@rics.bwh.harvard.edu Ph: (617) 525-4866, Fax: (617) 525-4868.

the new paradigm of the heart as a non-terminally differentiated organ. Despite the inability of myocardial tissue to adequately 'self-heal' following acute injury, as well as controversy regarding the ideal cell type for therapy, and a lack of understanding regarding the underlying cellular mechanisms mediating cardiac regeneration, we remain cautiously optimistic that cell based therapies may be sufficiently developed to effectively regenerate myocardium following cardiac injury.

In this review, we focus on the role of bone marrow derived stem cells and cardiac stem/progenitor cells in cardiomyocyte homeostasis and myocardial repair and regeneration, as well as provide a brief overview of current clinical trials utilizing cell-based therapeutic approaches in patients with heart disease.

Cardiac Stem/Progenitor Cells

For decades, the adult heart has been thought to exist as a terminally differentiated organ with limited proliferative capacity. Cardiomyocytes undergo hypertrophy, rather then hyperplasia, in response to hemodynamic stress, in contrast to other tissues, such as liver, intestine, and skeletal muscle. These long-held tenets of myocardial biology have recently been challenged and a new paradigm of the heart as a partially self-renewing organ has been proposed. While evidence challenging the old belief has fermented for some time [16,17], the identification of resident cardiac stem/progenitor cells (summarized **in** Table 1) has brought this new concept to the scientific forefront [12,13,15,18-20] and suggests the capacity of adult myocardium to maintain physiological homeostasis, at least partially, through resident cardiac stem cells.

The report by Hierlihy and colleagues [18] in 2002 was the first identifying the presence of a stem cell-like population in adult hearts based on their specific ability to efflux Hoechst dye. Such Hoechst-effluxing capacity was first introduced to identify highly enriched hematopoetic stem cell populations, termed side population (SP) stem cells, from bone marrow [21]. Recently, this methodology has been utilized to identify tissue specific stem/progenitor cells in various adult organs including pancreas, pituitary, testis, mammary gland, lung, liver, skeletal muscle, liver, lung as well as heart (for review see [22,23]). Using immunohistochemistry analysis, Hierlihy et al found that adult myocardium retains a specific SP cells population, capable of tissue specific differentiate into cardiomyocytes, *in-vitro* [18]. In 2003, Beltrami et al thoroughly described a population of cardiac stem cells (c-kit+ cells) found in clusters and residing among cardiomyocytes in adult hearts [12]. *In-vitro*, cardiac c-kit+ cells appear to be clonogenic and were able to undergo self renewal and differentiation into cardiac cell lineages (cardiomyocytes, endothelial, smooth muscle cell). More importantly, these c-kit+ cell, when implanted in mouse hearts following MI, retained the capacity for differentiation into cardiomyocytes, *in-vivo*. These *in-vivo* data are of both scientific and clinical significance, as they strongly implicate the regeneration potential of cardiac stem cells in injured hearts. In the same year of Beltrami's report, Oh et al employed a different stem cell marker, Sca-1, to identify yet another population of resident cardiac progenitor cells in adult hearts [19]. Similarly, these Sca1+ cells were found to be capable of differentiation into cardiomyocytes, *in-vitro* and *in-vivo*, in response to 5-azacytidine and myocardial ischemia, respectively [19]. In addition to the initial observation identifying SP cells in adult myocardium, several groups, including ours, have confirmed the presence of such progenitor cells population in adult hearts [13,15]. Martin et al reported expression of α-sarcomeric actinin in cardiac SP cells co-cultured with other cardiac cells as well as demonstrated the presence of SP cells in human myocardium [13,18]. Work performed by Tomita and colleagues documented the generation of neurosphere like clusters, referred to as "cardiospheres", from neonatal cardiac SP cells [24]. Similar to the cardiospheres described by Messina et al [14], cardiospheres derived from cardiac SP cells have been shown to harbor clonogenic cells with remarkable multi-lineage differentiation potential [24]. These cardiospheres expressed cardiac, smooth muscle and interestingly, neuronal genes

and proteins. Data from our group demonstrated not only the capacity for biochemical, but also functional, cardiomyogenic differentiation in cardiac SP cells [15]. More importantly, our study demonstrated that among cardiac SP cells, cardiomyogeneic differentiation is restricted to cells negative for CD31 expression and positive for Sca-1 expression (CD31-/Sca-1+ SP cells) [15] While the *in-vitro* cardiomyogenic differentiation potential of cardiac SP cells has been consistently demonstrated, less is known about the ability of these cells to undergo cardiomyogeneic differentiation, *in-vivo*. Recently, Komuro and colleagues studied the homing and differentiation efficiency of intravenously injected cardiac SP cells in a myocardial cryoinjury rat model [25]. Neonatal rat cardiac SP cells were found to be able to home to areas of injured myocardium and undergo differentiation into cardiomyocytes, endothelial cells, smooth muscle cells and fibroblasts. Another potential marker for cardiac stem/progenitor cells, Isl-1 (LIM homeodomain transcription factor), was more recently reported by Laugwitz et al [20]. These cells were found to harbor similar cardiomyogenic potential *in-vitro*, though were phenotypically distinct from SP cells. As more information becomes available regarding cardiac stem/progenitor cells, a key question remains whether these seemingly unique progenitor populations described above are truly distinct from each other, or represent the same population of progenitor cells at different stages in the differentiation process.

Bone Marrow Derived Adult Stem Cells

The bone marrow is known to be excellent reservoir for many adult stem cells, and bone marrow derived stem cells have been employed to treat hematologic disorders for decades. Recent reports have demonstrated that bone marrow derived stem cells are able to traverse cell lineage boundaries and transdifferentiate into hepatocytes, endothelial cells, skeletal muscle, and neurons upon proper stimulation [26-28]. While the ability of bone marrow derived stem cells to transdifferentiate into cardiomyocytes remains highly controversial, much of the recent progress in regenerative cardiovascular research, both in animal and humans, has been achieved using bone marrow derived stem cell populations, including hematopoietic stem cells (HSC), mesenchymal stem cells (MSC), and endothelial progenitor cells (EPC).

Hematopoietic stem cells (HSC)

HSC can be isolated from bone marrow cells through selective sorting for a particular set of surface receptors (Lineage$^-$, c-kit+, Sca-1+, CD34^{lo}, CD38^{hi}) [29,30] and represent the prototypic adult stem cell population. The ability of HSC to reconstitute the hematopoietic system of a myeloablated host led to the first clinical application of adult stem cells more than three decades ago [31]. Despite the failure of studies to definitely prove differentiation of HSC into cardiomyocytes, *in-vitro*, several studies in mice have demonstrated the potential of HSC to differentiate into cardiomyocytes or vascular cells following cardiac injury, *in-vivo* [32-34].

Mesenchymal stem cells (MSC)

Within the bone marrow stroma resides a subset of non-hematopoietic cells that have the potential to differentiate into cells of mesenchymal origin [35,36]. These mesenchymal stem cells (MSC) represent approximately 0.001 to 0.01% of the total nucleated marrow cell population, a concentration 10-fold lower than their hematopoietic counterparts. MSC are self-renewing and expandable *in-vitro* using standard cell culture techniques. Immunophenotypically, MSC lack the typical hematopoietic antigens (CD45, CD34, CD14) but express specific adhesion molecules (ALCAM/CD44) and antigens (SH2/SH3/SH4/STRO-1) [37,38]. At first, MSC were thought to contribute solely to the formation of the stromal microenvironment in the bone marrow and maintain HSC survival and function. However, subsequent studies have suggested that MSC are themselves capable of multipotency, with differentiation into chondrocytes, osteoblasts, astrocytes, neurons, skeletal muscle and, notably, cardiomyocytes [26,39-41].

Endothelial progenitor cells (EPC)

Endothelial progenitor cells (EPC) represent a subset of hematopoietic stem cells that are able to acquire an endothelial phenotype, *in-vitro* [42-45]. EPC express the hematopoietic stem cell markers CD133, CD34 and the endothelial marker Flk-1 (VEGFR-2) [44]. EPC can be isolated directly from the bone marrow or from the peripheral circulation and expanded, *in-vitro*.

Cardiac Cellular Homeostasis: Physiological and Pathological States

The identification of resident cardiac stem/progenitor cells evokes a new understanding of the mechanisms by which the adult heart may maintain cellular homeostasis. It is still a matter of debate whether cardiac cellular homeostasis is maintained solely by endogenous stem/progenitor cells or via extra-cardiac sources, notably bone marrow derived stem cells. In particular, the observation of male (host) cells in male patients transplanted with female hearts (mix-gender donor hearts) [46,47] suggests the potential role of extra-cardiac stem cells in the turnover of the cardiac cells. Interestingly, it has been proposed that the chimerism observed in humans may possibly result from cardiac progenitor cells residing in host atria which were kept intact during cardiac transplantation, and not from circulating bone marrow stem cells [48]. More recent data in animal models, however, have suggested that bone marrow derived stem cells contribute little in maintaining the homeostasis of cardiac cells during normal post natal growth as well as normal adulthood [34,49,50]

In contrast, bone marrow derived stem cells likely play a significant role in maintaining cardiac cells homeostasis, including the turnover of cardiac stem/progenitor cells, cardiomyogenesis, and angiogenesis, following myocardial injury [49-51]. Jackson et al [34] demonstrated the ability of bone marrow SP cells to undergo cardiomyogenic differentiation, albeit at a very low frequency, and angiogenesis, following MI. Utilizing a murine model of GFP-labeled bone marrow, we also have found that bone marrow derived stem cells (SP cells) homed to areas of injured heart as early as three days following MI [49]. These bone marrow derived cells may not only contribute to active myocardial repair, as have been suggested by several groups [8,9,32,33,52-56], but also participate in the reconstitution of the cardiac progenitor cell pool [49]. This is further supported by additional recent work [50], which has utilized genetic mouse models to demonstrate an increase in cardiac c-kit+ cells, recruited from bone marrow, following MI [50] Moreover, using a rat model of heterotropic gender-mismatched cardiac transplantation, Wang et al also demonstrated that bone marrow derived stem cells are attracted to areas of myocardial ischemic injury and participate in cardiac repair [51] These experimental data were further supported by observations in human mix-gender cardiac transplants, which suggest greater cardiac chimerism may occur in patients with MI [57]. In summary, the current literature suggests that cardiac injury may serve as a necessary and potent stimulant for the recruitment and potential cardiomyogeneic differentiation of endogenous bone marrow derived stem cells.

Mobilization and Homing of Marrow Derived Stem Cell

It is well recognized that, despite the existence of cardiac stem/progenitor cells, this endogenous capacity for regeneration is insufficient to mediate repair following severe cardiac injury. Thus, the ability of injured myocardium to recruit extra-cardiac stem cells following injury is critical to aid in myocardial repair and regeneration. At least three major compartments can be thought of to regulate this complicated orchestra, the injured myocardium, the bone marrow, and the peripheral circulation. The injured myocardium is responsible for releasing the signals via peripheral blood to signal the mobilization of the extra-cardiac stem cells from the major reservoir, bone marrow, into peripheral circulation. Following mobilization, these circulating bone marrow-derived stem cells are then able to follow a trail marked by specific signals, subsequently exit the circulation, and home to injured sites to initiate the cardiac repair

process (Figure 1). These three players involved in mobilization and homing process must work together to achieve functional significant stem cell-mediated repair and regeneration.

The precise time course, kinetics and factors stimulating bone marrow mobilization remain the subject of intense investigation; nonetheless, several crucial factors have been shown to promote the mobilization of bone marrow-derived stem cells into peripheral circulation, including granulocyte colony-stimulating factor (G-CSF), granulocyte / macrophage colony-stimulating factor (GM-CSF), stem cell factor (SCF), vascular endothelial growth factor (VEGF), hepatocyte growth factor (HGF) and erythropoietin (EPO) (for review, [58]). Myocardial ischemia is known to induce several classically 'mobilizing cytokines', including, but not limit to, G-CSF [59,60,61], SCF [59-61], VEGF [61-65], SDF-1 [59,61,65,66], and EPO [67,68] and these cytokines may be responsible for the observed homing of bone marrow-derived stem cells following MI. Mobilization of EPC through cytokine stimulants increases EPC concentration in the peripheral circulation substantially [68]. In addition to well-recognized HSC mobilizing agents such as G-CSF and SCF, VEGF, and EPO, statins have been shown to promote EPC recruitment [68-71]. Moreover, given the capacity of bone marrow-derived stem cells to home to sites of injury, it has been suggested that mobilization of bone marrow-derived stem cells through systemically delivered cytokine stimulants may represent a less invasive strategy to activate and deliver stem cells following MI. Therefore, these cytokines/factors and their respectively receptors can be targeted to promote stem cell mobilization and homing for therapeutics applications. Herein, we highlight several key signalling factors to demonstrate the potential of manipulating these signalling axises to achieve functionally significant cell-based cardiac repair.

G-CSF and SCF/c-kit

SCF, also known as steel factor, is a ligand for c-kit, a receptor expressed in stem cell and tissue progenitor cells, including resident cardiac stem cells. Similar to G-CSF and GM-CSF, SCF is a hematopoietic factor that is well known to regulate proliferation, differentiation and survival of bone marrow derived stem cells [72,73] Orlic et al was the first to use a combined therapy of G-CSF and SCF in a murine model of MI and demonstrated a significant improvement in LV remodelling, cardiac function, and animal survival with five days of treatment [33]. Improved outcome was associated with significant bone marrow derived cardiomyogenesis [33] These results, however, were not reproduced when G-CSF and SCF were given as a single dose at 4 hours following MI to non-human primates [74] While G-CSF and SCF/c-kit represent important factors for the recruitment of bone marrow derived stem cells following MI, actual results from various groups have been controversial at best; owing to the timing of cytokine administration and the dose utilized, as well as the actual model system. Nonetheless, the best "proof of concept" approach demonstrating the importance and involvement of the SCF/c-kit axis in bone marrow mobilization and cardiac repair has taken advantage of a transgenic mouse model overexpressing mutant c-kit (kit^{w}/kit^{w-v}) [50]. In kit^{w}/kit^{w-v} mice, the mobilization and homing of bone marrow-derived stem cells to the heart is markedly impaired following MI, despite elevated circulating levels of SCF. This deficiency further results in early cardiac failure and death. Intriguingly, this dysfunction can be rescued by bone marrow transplantation with wild type cells, thus restoring the capacity for homing by bone marrow derived stem cells. While certainly essential, proper manipulation of the SCF/c-kit axis for clinical benefit remains a goal of the future.

SDF-1/CXCR4

SDF-1, and its receptor CXCR4, have recently been suggested to also be important in regulating the mobilization of stem cells. Using gain and loss of function approaches, Moore et al has demonstrated that inhibition of SDF-1/CXCR4 by neutralizing antibodies retards the mobilization of stem cells [75]; while overexpression of CXCR4 augments the migration of

progenitor cells [76] More recently, several groups have suggested that the SDF-1/CXCR4 axis may be involved in the mobilization of bone marrow-derived stem cells and homing to injured myocardium following MI [77,78], whereas the mechanisms regulating this mobilization remain less clear. SDF-1 may upregulate secondary agents, including metalloproteinase-9 (MMP-9), causing the release of SCF and subsequently mobilization of c-kit+ cells into the circulation [79] Alternatively, Misao and colleagues have suggested that the beneficial effects of G-CSF mediated cardiac repair following MI is through the upregulation of SDF-1 and subsequently recruitment of CXCR4+ cells [80] Altogether, these reports demonstrate the complex interplay that exists between these mobilizing / homing signals. To take advantage of these secreted factors/cytokines to improve post-MI cardiac repair and regeneration, thorough investigation of the timing of the release and interactions among signalling factors is required.

Potential Mechanisms of Stem Cell-mediated Myocardial Repair/ Regeneration

Over the past decade, many groups have employed a spectrum of bone marrow-derived stem cell populations, including total bone marrow, HSC, MSC, and EPC for the treatment of post-MI heart failure in both animal models as well as in human clinical trials. Interestingly, while only few groups have observed differentiation of bone marrow derived stem cell into cardiomyocytes, most groups have reported a beneficial effect on post-MI remodelling. As such, these data are certainly both encouraging, given the improvement in objective measures such as cardiac structure and function, yet disappointing, as they fail to demonstrate physiologically relevant cardiac differentiation. This has brought into question the ultimate goal of cell-based therapies. Certainly our central objective is to improve cardiac function, and by doing so, patient outcomes. To that end, cell based therapies may be beneficial not only in the regeneration of lost myocardium, via direct trans-differentiation, but also in protecting existing viable myocardium or repairing damaged myocardium, via paracrine influences.

Stem Cells (Trans)differentiation

The foremost purpose of cell-based therapies remains the regeneration of lost cardiac cells via differentiation. Using genetic markers and/or labelled fluorescent dyes, several groups have reported the transdifferentiation of bone marrow derived HSC into cardiomycoytes [32-34] However, these results, subsequently, have been called into question by others, who have failed to identify HSC-derived cardiomyocytes [81-83]. In addition to HSC, MSC have also been suggested to retain the capacity for cardiomyogenic differentiation both *in-vitro* with proper stimulation and *in-vivo* [40,55,56,84,85]. Similar to criticisms voiced regarding HSC, the ability of MSC to trans-differentiate into cardiomyocytes have been challenged [86]. Furthermore, many studies have suggested that cell fusion, rather than trans-differentiation, of bone marrow derived cells may explain observed phenotypic changes [87-90] Regardless of the mechanism responsible, fusion vs. trans-differentiation, it is generally agreed that the number of reported cardiomyocytes derived from exogenously delivered bone marrow stem cells remains relatively low and cannot physically account for observed functional improvements. As such, one alternative proposed mechanism is stem cell-mediated paracrine effects (discussed in the following section). In contrast to cardiomyogenesis, by in large, most groups have observed bone marrow derived stem cells to contribute to angiogenesis, an observation made over 10 years by Asahara and colleagues [42,91] and more recently, by many other laboratories [8, 92-95]. Finally, an alternative mechanism by which bone marrow derived stem cell populations may contribute to myocardial repair is via maintenance of cardiac-specific stem cells pool following injury. Indeed, our group has recently found that bone marrow derived SP cells home to injured myocardium following MI and under go phenotypic changes to adapt a cardiac SP cell phenotype. Such cardiac SP cells, may in-turn, contribute to the capacity of the heart for long-term endogenous cardiac repair.

Paracrine Influence

As suggested above, the beneficial effects of stem cell therapy on post-MI cardiac function remain disproportionate to the degree of cardiomyogeneic differentiation. Such observations have lead to the hypothesis that potential paracrine effects may hold a prominent role in stem cell therapy. Such paracrine influences may include secretion of factors that either attenuate apoptosis of endogenous cardiomyocytes [96,97] and EC [8], promote angiogenesis [52,98], and/or activate resident cardiac stem / progenitor cells [80]. Uemura and colleagues suggested that hypoxia-induced apoptosis may be attenuated in cardiomyocytes co-cultured with total bone marrow cells, in-vitro [97]. Moreover, mesenchymal stem cells also may produce angiogenic factors such as VEGF and bFGF, as well as chemotactic factors, including as MCP-1 and PGF, that serve to recruit monocytes and promote angiogenesis [99]. While the "paracrine hypothesis" of stem cell therapy seems rational given prior observations, to date, it remains unexplored with largely indirect supportive evidence.

Current Translational Approaches for Bone Marrow Based Therapies

While the ideal cell type for stem cell-based therapies remains to be determined, to date, bone marrow derived stem cells, isolated from whole bone marrow aspirate, remains the most commonly used cell type for human studies. This is largely due to its easy accessibility and well characterized properties by haematologists for over thirty years. Among bone marrow populations, total mononuclear bone marrow cells and circulating endothelial progenitor cells have recently or are currently being employed in many Phase I and/or II trials (Table 2). Current methods of delivery include direct intramyocardial injection, via both endocardial catheter-based and epicardial surgical-based approaches, and more recently, percutaneous (catheter-based) intracoronary injection. Alternatively, indirect mobilization has also been attempted with peripheral delivery of cytokines, notably G-CSF.

In just a few years, cell based therapy has evolved at an explosive pace, from early *in-vitro* cell studies to animal models of myocardial infarction, and now to several early phase clinical trials. Overall, most initial non-randomized clinical trials, while designed for safety rather than efficacy, have encouragingly suggested a moderate improvement in heart function following stem cell therapy. The first randomized trial of intracoronary bone marrow derived stem cells, the BOOST I trial [100], demonstrated an early benefit in left-ventricular ejection fraction at 6 months post-cell therapy as assessed by cardiac MRI. However, due to continued improvement in the control group, the benefit in treated patients relatively to the control group was lost at 18 months follow up [101]. Two larger clinical trials investigating intracoronary delivery of bone marrow cells have also been initiated, with early results presented at the Scientific Sections of American Heart Association in late 2005. In the ASTAMI trial, a randomized trial of one hundred patients [102] with acute MI, bone marrow mononuclear cells was delivered 6 days post-PTCA. At 6 months follow-up, no improvement in left ventricular ejection fraction or infarct size was observed. In somewhat contrast, the REPAIR-AMI [103], a randomized, placebo control trial with over two hundred patients following acute-MI, suggested a small, albeit significantly important, improvement in left ventricular ejection fraction by ventriculography. While both trials still are ongoing, many potential explanations for observed differences, including severity ventricular dysfunction, the timing of cell delivery, and the method cells isolation (quality of the cells), have been proposed and are currently undergoing investigation in animal models. Mobilization of stem cells from the bone marrow represents an alternative cell-based therapy that has also recently been investigated in clinical trials. The FIRSTINE-AMI [104] has demonstrated not only the safety and feasibility of bone marrow mobilization using G-CSF in MI patients after reperfusion, but also suggested a potential improvement in left ventricular ejection fraction and an attenuation of left ventricular dilation. Importantly, this trial showed that treatment with G-CSF did not augment post-percutaneous coronary intervention restenosis rate. However, subsequent randomized placebo controlled clinical

trials, REVIVAL II [105] and STEMMI [106], have failed to reproduce the benefits previously seen in early human studies. Although these trials failed to demonstrate positive outcomes, no adverse events, including vessel restenosis, were observed. Reasons for these negative results remain to be determined, though, inappropriate cytokine dosing and inadequate timing of the cytokine administration have been proposed as potential explanations.

Conclusions

Myocardial infarction results in cell death and it replacement of cardiomyocytes with non-contractile scar tissue. The optimal goal for cell-based cardiac repair is to restore cardiac structure and function through regeneration of functionally-competent cardiomyocytes. To rebuild a normal and functional cardiac tissue requires not only highly integrated cardiomyogenesis and angiogenesis, but also a proper matrix network system, to ensure synchronized contraction and relaxation with native myocardium. While the task of achieving such goal is daunting, the therapeutic potential of myocardial regeneration remains enormous. Further intensive research, in basic science and clinical arenas, as well as carefully constructed clinical trials, are needed in order to make this next great frontier in cardiovascular medicine a reality.

Acknowledgements

This work was supported by research grants funding from the National Institutes of Health (R.L.). O.P. is a recipient of a Swiss National Science Foundation Grant (PBBSB-101349), and F.M. is a recipient of a Federation Francaise de Cardiologie grant.

References

1. Braunwald E, Bristow MR. Congestive heart failure: fifty years of progress. Circulation 2000;102:IV14–23. [PubMed: 11080127]
2. Scorsin M, et al. Comparison of the effects of fetal cardiomyocyte and skeletal myoblast transplantation on postinfarction left ventricular function. J Thorac Cardiovasc Surg 2000;119:1169–75. [PubMed: 10838534]
3. Koh GY, et al. Stable fetal cardiomyocyte grafts in the hearts of dystrophic mice and dogs. J Clin Invest 1995;96:2034–42. [PubMed: 7560097]
4. Jain M, et al. Cell therapy attenuates deleterious ventricular remodeling and improves cardiac performance after myocardial infarction. Circulation 2001;103:1920–7. [PubMed: 11294813]
5. Taylor DA, et al. Regenerating functional myocardium: improved performance after skeletal myoblast transplantation. Nat Med 1998;4:929–33. [PubMed: 9701245]
6. Murry CE, Wiseman RW, Schwartz SM, Hauschka SD. Skeletal myoblast transplantation for repair of myocardial necrosis. J Clin Invest 1996;98:2512–23. [PubMed: 8958214]
7. Chiu RC, Zibaitis A, Kao RL. Cellular cardiomyoplasty: myocardial regeneration with satellite cell implantation. Ann Thorac Surg 1995;60:12–8. [PubMed: 7598572]
8. Kocher AA, et al. Neovascularization of ischemic myocardium by human bone-marrow-derived angioblasts prevents cardiomyocyte apoptosis, reduces remodeling and improves cardiac function. Nat Med 2001;7:430–6. [PubMed: 11283669]
9. Kawamoto A, et al. Intramyocardial transplantation of autologous endothelial progenitor cells for therapeutic neovascularization of myocardial ischemia. Circulation 2003;107:461–8. [PubMed: 12551872]
10. Orlic D. Adult BM stem cells regenerate mouse myocardium. Cytotherapy 2002;4:521–5. [PubMed: 12568987]
11. Orlic D. Adult bone marrow stem cells regenerate myocardium in ischemic heart disease. Ann N Y Acad Sci 2003;996:152–7. [PubMed: 12799293]
12. Beltrami AP, et al. Adult cardiac stem cells are multipotent and support myocardial regeneration. Cell 2003;114:763–76. [PubMed: 14505575]

13. Martin CM, et al. Persistent expression of the ATP-binding cassette transporter, Abcg2, identifies cardiac SP cells in the developing and adult heart. Dev Biol 2004;265:262–75. [PubMed: 14697368]
14. Messina E, et al. Isolation and expansion of adult cardiac stem cells from human and murine heart. Circ Res 2004;95:911–21. [PubMed: 15472116]
15. Pfister O, et al. CD31- but Not CD31+ cardiac side population cells exhibit functional cardiomyogenic differentiation. Circ Res 2005;97:52–61. [PubMed: 15947249]
16. McDonnell TJ, Oberpriller JO. The response of the atrium to direct mechanical wounding in the adult heart of the newt, Notophthalmus viridescens. An electron-microscopic and autoradiographic study. Cell Tissue Res 1984;235:583–92. [PubMed: 6713487]
17. Rumyantsev PP, Borisov A. DNA synthesis in myocytes from different myocardial compartments of young rats in norm, after experimental infarction and in vitro. Biomed Biochim Acta 1987;46:S610–5. [PubMed: 3435518]
18. Hierlihy AM, Seale P, Lobe CG, Rudnicki MA, Megeney LA. The post-natal heart contains a myocardial stem cell population. FEBS Lett 2002;530:239–43. [PubMed: 12387899]
19. Oh H, et al. Cardiac progenitor cells from adult myocardium: homing, differentiation, and fusion after infarction. Proc Natl Acad Sci U S A 2003;100:12313–8. [PubMed: 14530411]
20. Laugwitz KL, et al. Postnatal isl1+ cardioblasts enter fully differentiated cardiomyocyte lineages. Nature 2005;433:647–53. [PubMed: 15703750]
21. Goodell MA, Brose K, Paradis G, Conner AS, Mulligan RC. Isolation and functional properties of murine hematopoietic stem cells that are replicating in vivo. J Exp Med 1996;183:1797–806. [PubMed: 8666936]
22. Asakura A, Rudnicki MA. Side population cells from diverse adult tissues are capable of in vitro hematopoietic differentiation. Exp Hematol 2002;30:1339–45. [PubMed: 12423688]
23. Challen GA, Little MH. A Side Order of Stem Cells: The SP Phenotype. Stem Cells 2006;24:3–12. [PubMed: 16449630]
24. Tomita Y, et al. Cardiac neural crest cells contribute to the dormant multipotent stem cell in the mammalian heart. J Cell Biol 2005;170:1135–46. [PubMed: 16186259]
25. Wada H, Matsuura K, Oyama T, Iwanaga K, Takahashi T, Goto T, Nagai T, Komuro I. Homing and differentiation of intravenously transplanted cardiac side population cells in injured myocardium. Circulation 2005;111:801.
26. Ferrari G, et al. Muscle regeneration by bone marrow-derived myogenic progenitors. Science 1998;279:1528–30. [PubMed: 9488650]
27. Krause DS, et al. Multi-organ, multi-lineage engraftment by a single bone marrow-derived stem cell. Cell 2001;105:369–77. [PubMed: 11348593]
28. Mezey E, Chandross KJ, Harta G, Maki RA, McKercher SR. Turning blood into brain: cells bearing neuronal antigens generated in vivo from bone marrow. Science 2000;290:1779–82. [PubMed: 11099419]
29. Osawa M, et al. In vivo self-renewal of c-Kit+ Sca-1+ Lin(low/-) hemopoietic stem cells. J Immunol 1996;156:3207–14. [PubMed: 8617942]
30. Spangrude GJ, Heimfeld S, Weissman IL. Purification and characterization of mouse hematopoietic stem cells. Science 1988;241:58–62. [PubMed: 2898810]
31. Goldman JM, Horowitz MM. The international bone marrow transplant registry. Int J Hematol 2002;76(Suppl 1):393–7. [PubMed: 12430889]
32. Orlic D, et al. Bone marrow cells regenerate infarcted myocardium. Nature 2001;410:701–5. [PubMed: 11287958]
33. Orlic D, et al. Mobilized bone marrow cells repair the infarcted heart, improving function and survival. Proc Natl Acad Sci U S A 2001;98:10344–9. [PubMed: 11504914]
34. Jackson KA, et al. Regeneration of ischemic cardiac muscle and vascular endothelium by adult stem cells. J Clin Invest 2001;107:1395–402. [PubMed: 11390421]
35. Friedenstein AJ, Petrakova KV, Kurolesova AI, Frolova GP. Heterotopic of bone marrow.Analysis of precursor cells for osteogenic and hematopoietic tissues. Transplantation 1968;6:230–47. [PubMed: 5654088]

36. Tavassoli M, Crosby WH. Transplantation of marrow to extramedullary sites. Science 1968;161:54–6. [PubMed: 4871792]
37. Conget PA, Minguell JJ. Phenotypical and functional properties of human bone marrow mesenchymal progenitor cells. J Cell Physiol 1999;181:67–73. [PubMed: 10457354]
38. Pittenger MF, et al. Multilineage potential of adult human mesenchymal stem cells. Science 1999;284:143–7. [PubMed: 10102814]
39. Azizi SA, Stokes D, Augelli BJ, DiGirolamo C, Prockop DJ. Engraftment and migration of human bone marrow stromal cells implanted in the brains of albino rats--similarities to astrocyte grafts. Proc Natl Acad Sci U S A 1998;95:3908–13. [PubMed: 9520466]
40. Makino S, et al. Cardiomyocytes can be generated from marrow stromal cells in vitro. J Clin Invest 1999;103:697–705. [PubMed: 10074487]
41. Pereira RF, et al. Cultured adherent cells from marrow can serve as long-lasting precursor cells for bone, cartilage, and lung in irradiated mice. Proc Natl Acad Sci U S A 1995;92:4857–61. [PubMed: 7761413]
42. Asahara T, et al. Isolation of putative progenitor endothelial cells for angiogenesis. Science 1997;275:964–7. [PubMed: 9020076]
43. Gehling UM, et al. In vitro differentiation of endothelial cells from AC133-positive progenitor cells. Blood 2000;95:3106–12. [PubMed: 10807776]
44. Peichev M, et al. Expression of VEGFR-2 and AC133 by circulating human CD34(+) cells identifies a population of functional endothelial precursors. Blood 2000;95:952–8. [PubMed: 10648408]
45. Shi Q, et al. Evidence for circulating bone marrow-derived endothelial cells. Blood 1998;92:362–7. [PubMed: 9657732]
46. Quaini F, et al. Chimerism of the transplanted heart. N Engl J Med 2002;346:5–15. [PubMed: 11777997]
47. Murry CE, Whitney ML, Laflamme MA, Reinecke H, Field LJ. Cellular therapies for myocardial infarct repair. Cold Spring Harb Symp Quant Biol 2002;67:519–26. [PubMed: 12858578]
48. Schwartz RS, Curfman GD. Can the heart repair itself? N Engl J Med 2002;346:2–4. [PubMed: 11777996]
49. Mouquet F, et al. Restoration of cardiac progenitor cells after myocardial infarction by self-proliferation and selective homing of bone marrow-derived stem cells. Circ Res 2005;97:1090–2. [PubMed: 16269652]
50. Fazel S, et al. Cardioprotective c-kit+ cells are from the bone marrow and regulate the myocardial balance of angiogenic cytokines. J Clin Invest 2006;116:1865–77. [PubMed: 16823487]
51. Wang Y, et al. Combining pharmacological mobilization with intramyocardial delivery of bone marrow cells over-expressing VEGF is more effective for cardiac repair. J Mol Cell Cardiol 2006;40:736–45. [PubMed: 16603183]
52. Kamihata H, et al. Implantation of bone marrow mononuclear cells into ischemic myocardium enhances collateral perfusion and regional function via side supply of angioblasts, angiogenic ligands, and cytokines. Circulation 2001;104:1046–52. [PubMed: 11524400]
53. Kawamoto A, et al. Therapeutic potential of ex vivo expanded endothelial progenitor cells for myocardial ischemia. Circulation 2001;103:634–7. [PubMed: 11156872]
54. Mangi AA, et al. Mesenchymal stem cells modified with Akt prevent remodeling and restore performance of infarcted hearts. Nat Med 2003;9:1195–201. [PubMed: 12910262]
55. Shake JG, et al. Mesenchymal stem cell implantation in a swine myocardial infarct model: engraftment and functional effects. Ann Thorac Surg 2002;73:1919–25. [PubMed: 12078791]discussion 1926
56. Toma C, Pittenger MF, Cahill KS, Byrne BJ, Kessler PD. Human mesenchymal stem cells differentiate to a cardiomyocyte phenotype in the adult murine heart. Circulation 2002;105:93–8. [PubMed: 11772882]
57. Hocht-Zeisberg E, et al. Cellular repopulation of myocardial infarction in patients with sex-mismatched heart transplantation. Eur Heart J 2004;25:749–58. [PubMed: 15120885]
58. Lapidot T, Petit I. Current understanding of stem cell mobilization: the roles of chemokines, proteolytic enzymes, adhesion molecules, cytokines, and stromal cells. Exp Hematol 2002;30:973–81. [PubMed: 12225788]

59. Leone AM, et al. Endogenous G-CSF and CD34(+) cell mobilization after acute myocardial infarction. Int J Cardiol 2006;111:202–8. [PubMed: 16051386]
60. Dewald O, et al. Of mice and dogs: species-specific differences in the inflammatory response following myocardial infarction. Am J Pathol 2004;164:665–77. [PubMed: 14742270]
61. Wojakowski W, et al. Mobilization of CD34/CXCR4+, CD34/CD117+, c-met+ stem cells, and mononuclear cells expressing early cardiac, muscle, and endothelial markers into peripheral blood in patients with acute myocardial infarction. Circulation 2004;110:3213–20. [PubMed: 15533859]
62. Namiki A, et al. Hypoxia induces vascular endothelial growth factor in cultured human endothelial cells. J Biol Chem 1995;270:31189–95. [PubMed: 8537383]
63. Couffinhal T, et al. Mouse model of angiogenesis. Am J Pathol 1998;152:1667–79. [PubMed: 9626071]
64. Banai S, et al. Angiogenic-induced enhancement of collateral blood flow to ischemic myocardium by vascular endothelial growth factor in dogs. Circulation 1994;89:2183–9. [PubMed: 7514110]
65. Wang Y, et al. Changes in circulating mesenchymal stem cells, stem cell homing factor, and vascular growth factors in patients with acute ST elevation myocardial infarction treated with primary percutaneous coronary intervention. Heart 2006;92:768–74. [PubMed: 16251230]
66. Askari AT, et al. Effect of stromal-cell-derived factor 1 on stem-cell homing and tissue regeneration in ischaemic cardiomyopathy. Lancet 2003;362:697–703. [PubMed: 12957092]
67. Vogt C, Noe G, Rich IN. The role of the blood island during normal and 5-fluorouracil-perturbed hemopoiesis. Blood Cells 1991;17:105–21. [PubMed: 2018848]discussion 121-5
68. Heeschen C, et al. Erythropoietin is a potent physiologic stimulus for endothelial progenitor cell mobilization. Blood 2003;102:1340–6. [PubMed: 12702503]
69. Kalka C, et al. Vascular endothelial growth factor(165) gene transfer augments circulating endothelial progenitor cells in human subjects. Circ Res 2000;86:1198–202. [PubMed: 10864908]
70. Llevadot J, et al. HMG-CoA reductase inhibitor mobilizes bone marrow--derived endothelial progenitor cells. J Clin Invest 2001;108:399–405. [PubMed: 11489933]
71. Walter DH, et al. Statin therapy accelerates reendothelialization: a novel effect involving mobilization and incorporation of bone marrow-derived endothelial progenitor cells. Circulation 2002;105:3017–24. [PubMed: 12081997]
72. Ulich TR, et al. Hematologic effects of stem cell factor in vivo and in vitro in rodents. Blood 1991;78:645–50. [PubMed: 1713510]
73. Anderson DM, et al. Molecular cloning of mast cell growth factor, a hematopoietin that is active in both membrane bound and soluble forms. Cell 1990;63:235–43. [PubMed: 1698558]
74. Norol F, et al. Influence of mobilized stem cells on myocardial infarct repair in a nonhuman primate model. Blood 2003;102:4361–8. [PubMed: 12947003]
75. Petit I, et al. G-CSF induces stem cell mobilization by decreasing bone marrow SDF-1 and up-regulating CXCR4. Nat Immunol 2002;3:687–94. [PubMed: 12068293]
76. Kahn J, et al. Overexpression of CXCR4 on human CD34+ progenitors increases their proliferation, migration, and NOD/SCID repopulation. Blood 2004;103:2942–9. [PubMed: 15070669]
77. Kucia M, et al. Cells expressing early cardiac markers reside in the bone marrow and are mobilized into the peripheral blood after myocardial infarction. Circ Res 2004;95:1191–9. [PubMed: 15550692]
78. Abbott JD, et al. Stromal cell-derived factor-1alpha plays a critical role in stem cell recruitment to the heart after myocardial infarction but is not sufficient to induce homing in the absence of injury. Circulation 2004;110:3300–5. [PubMed: 15533866]
79. Heissig B, et al. Recruitment of stem and progenitor cells from the bone marrow niche requires MMP-9 mediated release of kit-ligand. Cell 2002;109:625–37. [PubMed: 12062105]
80. Misao Y, et al. Importance of recruitment of bone marrow-derived CXCR4(+) cells in post-infarct cardiac repair mediated by G-CSF. Cardiovasc Res 2006;71:455–65. [PubMed: 16769043]
81. Murry CE, et al. Haematopoietic stem cells do not transdifferentiate into cardiac myocytes in myocardial infarcts. Nature 2004;428:664–8. [PubMed: 15034593]
82. Balsam LB, et al. Haematopoietic stem cells adopt mature haematopoietic fates in ischaemic myocardium. Nature 2004;428:668–73. [PubMed: 15034594]

83. Limbourg FP, et al. Haematopoietic stem cells improve cardiac function after infarction without permanent cardiac engraftment. Eur J Heart Fail 2005;7:722–9. [PubMed: 16158493]
84. Tomita S, et al. Autologous transplantation of bone marrow cells improves damaged heart function. Circulation 1999;100:II247–56. [PubMed: 10567312]
85. Hakuno D, et al. Bone marrow-derived regenerated cardiomyocytes (CMG Cells) express functional adrenergic and muscarinic receptors. Circulation 2002;105:380–6. [PubMed: 11804996]
86. Davani S, et al. Mesenchymal progenitor cells differentiate into an endothelial phenotype, enhance vascular density, and improve heart function in a rat cellular cardiomyoplasty model. Circulation 2003;108(Suppl 1):II253–8. [PubMed: 12970242]
87. Terada N, et al. Bone marrow cells adopt the phenotype of other cells by spontaneous cell fusion. Nature 2002;416:542–5. [PubMed: 11932747]
88. Ying QL, Nichols J, Evans EP, Smith AG. Changing potency by spontaneous fusion. Nature 2002;416:545–8. [PubMed: 11932748]
89. Alvarez-Dolado M, et al. Fusion of bone-marrow-derived cells with Purkinje neurons, cardiomyocytes and hepatocytes. Nature 2003;425:968–73. [PubMed: 14555960]
90. Vassilopoulos G, Russell DW. Cell fusion: an alternative to stem cell plasticity and its therapeutic implications. Curr Opin Genet Dev 2003;13:480–5. [PubMed: 14550412]
91. Asahara T, et al. Bone marrow origin of endothelial progenitor cells responsible for postnatal vasculogenesis in physiological and pathological neovascularization. Circ Res 1999;85:221–8. [PubMed: 10436164]
92. Urbich C, et al. Relevance of monocytic features for neovascularization capacity of circulating endothelial progenitor cells. Circulation 2003;108:2511–6. [PubMed: 14581410]
93. Camargo FD, Green R, Capetanaki Y, Jackson KA, Goodell MA. Single hematopoietic stem cells generate skeletal muscle through myeloid intermediates. Nat Med 2003;9:1520–7. [PubMed: 14625546]
94. Zhao Y, Glesne D, Huberman E. A human peripheral blood monocyte-derived subset acts as pluripotent stem cells. Proc Natl Acad Sci U S A 2003;100:2426–31. [PubMed: 12606720]
95. Wang JS, Shum-Tim D, Chedrawy E, Chiu RC. The coronary delivery of marrow stromal cells for myocardial regeneration: pathophysiologic and therapeutic implications. J Thorac Cardiovasc Surg 2001;122:699–705. [PubMed: 11581601]
96. Takahashi T, et al. Ischemia- and cytokine-induced mobilization of bone marrow-derived endothelial progenitor cells for neovascularization. Nat Med 1999;5:434–8. [PubMed: 10202935]
97. Uemura R, Xu M, Ahmad N, Ashraf M. Bone marrow stem cells prevent left ventricular remodeling of ischemic heart through paracrine signaling. Circ Res 2006;98:1414–21. [PubMed: 16690882]
98. Fuchs S, et al. Transendocardial delivery of autologous bone marrow enhances collateral perfusion and regional function in pigs with chronic experimental myocardial ischemia. J Am Coll Cardiol 2001;37:1726–32. [PubMed: 11345391]
99. Kinnaird T, et al. Marrow-derived stromal cells express genes encoding a broad spectrum of arteriogenic cytokines and promote in vitro and in vivo arteriogenesis through paracrine mechanisms. Circ Res 2004;94:678–85. [PubMed: 14739163]
100. Wollert KC, et al. Intracoronary autologous bone-marrow cell transfer after myocardial infarction: the BOOST randomised controlled clinical trial. Lancet 2004;364:141–8. [PubMed: 15246726]
101. Meyer GP, et al. Intracoronary bone marrow cell transfer after myocardial infarction: eighteen months' follow-up data from the randomized, controlled BOOST (BOne marrOw transfer to enhance ST-elevation infarct regeneration) trial. Circulation 2006;113:1287–94. [PubMed: 16520413]
102. Lunde K, et al. Autologous stem cell transplantation in acute myocardial infarction: The ASTAMI randomized controlled trial. Intracoronary transplantation of autologous mononuclear bone marrow cells, study design and safety aspects. Scand Cardiovasc J 2005;39:150–8. [PubMed: 16152682]
103. Schachinger V, Tonn T, Dimmeler S, Zeiher AM. Bone-marrow-derived progenitor cell therapy in need of proof of concept: design of the REPAIR-AMI trial. Nat Clin Pract Cardiovasc Med 2006;3 (Suppl 1):S23–8. [PubMed: 16501626]
104. Ince H, et al. Prevention of left ventricular remodeling with granulocyte colony-stimulating factor after acute myocardial infarction: final 1-year results of the Front-Integrated Revascularization and

Stem Cell Liberation in Evolving Acute Myocardial Infarction by Granulocyte Colony-Stimulating Factor (FIRSTLINE-AMI) Trial. Circulation 2005;112:I73–80. [PubMed: 16159869]

105. Zohlnhofer D, et al. Stem cell mobilization by granulocyte colony-stimulating factor in patients with acute myocardial infarction: a randomized controlled trial. Jama 2006;295:1003–10. [PubMed: 16507801]
106. Ripa RS, et al. Stem cell mobilization induced by subcutaneous granulocyte-colony stimulating factor to improve cardiac regeneration after acute ST-elevation myocardial infarction: result of the double-blind, randomized, placebo-controlled stem cells in myocardial infarction (STEMMI) trial. Circulation 2006;113:1983–92. [PubMed: 16531621]
107. Urbanek K, et al. Intense myocyte formation from cardiac stem cells in human cardiac hypertrophy. Proc Natl Acad Sci U S A 2003;100:10440–5. [PubMed: 12928492]
108. Linke A, et al. Stem cells in the dog heart are self-renewing, clonogenic, and multipotent and regenerate infarcted myocardium, improving cardiac function. Proc Natl Acad Sci U S A 2005;102:8966–71. [PubMed: 15951423]
109. Matsuura K, et al. Adult cardiac Sca-1-positive cells differentiate into beating cardiomyocytes. J Biol Chem 2004;279:11384–91. [PubMed: 14702342]
110. Strauer BE, et al. Repair of infarcted myocardium by autologous intracoronary mononuclear bone marrow cell transplantation in humans. Circulation 2002;106:1913–8. [PubMed: 12370212]
111. Assmus B, et al. Transplantation of Progenitor Cells and Regeneration Enhancement in Acute Myocardial Infarction (TOPCARE-AMI). Circulation 2002;106:3009–17. [PubMed: 12473544]
112. Britten MB, et al. Infarct remodeling after intracoronary progenitor cell treatment in patients with acute myocardial infarction (TOPCARE-AMI): mechanistic insights from serial contrast-enhanced magnetic resonance imaging. Circulation 2003;108:2212–8. [PubMed: 14557356]
113. Schachinger V, et al. Transplantation of progenitor cells and regeneration enhancement in acute myocardial infarction: final one-year results of the TOPCARE-AMI Trial. J Am Coll Cardiol 2004;44:1690–9. [PubMed: 15489105]
114. Janssens S, et al. Autologous bone marrow-derived stem-cell transfer in patients with ST-segment elevation myocardial infarction: double-blind, randomised controlled trial. Lancet 2006;367:113–21. [PubMed: 16413875]
115. Chen SL, et al. Improvement of cardiac function after transplantation of autologous bone marrow mesenchymal stem cells in patients with acute myocardial infarction. Chin Med J (Engl) 2004;117:1443–8. [PubMed: 15498362]

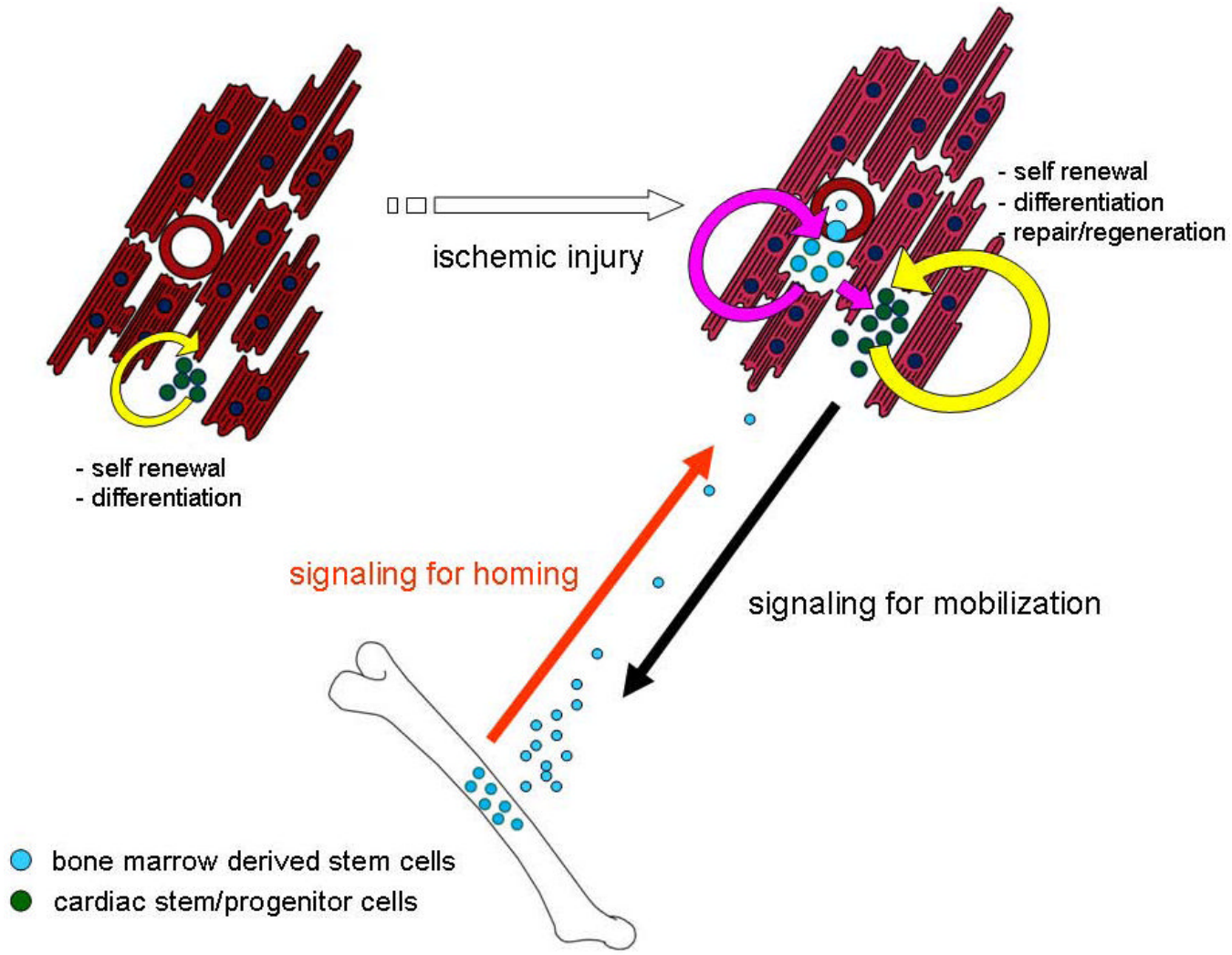

Figure 1.
A schematic representation of cell-based myocardial repair. Signals for mobilization and homing must work in an integrated fashion among the myocardium, peripheral blood, and bone marrow to achieve functionally significant stem cell-mediated repair and regeneration.

Table 1

Summery of reported cardiac stem/progenitor cells

	Surface Markers	Gene Expression	Self renewal	*In-vitro* differentiation	*In-vivo* differentiation	Species	Reference
SP	Sca-1+, CD31+, CD34 -, c-kit-*, CD45-, Isl1-	Nkx2.5, GATA4, MEF2C, Tie2	Yes	CM, CE, SMC	CM, EC	Mice, pig, human	Hierlihy et al 2002 [18] Pfister et al 2005 [15] Mouquet et al 2005 [49] Martin et al 2004 [13]
c-kit	Sca-1+, SP+** Lin-, CD45-, CD31-, CD34-	Nkx2.5, GATA4, MEF2C	Yes	CM, EC, SMC	CM, EC, SMC	Mice, rat, dog, pig, human,	Messina et al 2003 [14] Urbanek et al 2003 [107] Beltrami et al 2003 [12] Linke et al 2005 [108]
Sca-1	CD31+, CD34- Lin-, CD45-, c-kit-***	Nkx2.5***, GATA4, MEF2C, Tie2	ND	CM	CM	Mice, dog	Oh et al 2003 [19] Matsuura et al 2004 [109]
Isl-1	Lin-, CD45-, c-kit-, CD34-, CD31-, SP-	Nkx2.5, GATA4	Yes	CM	ND	Mice, rat, human	Laugwitz et al 2005 [20]

CM: cardiomyocyte. EC: endothelial cell. SMC: smooth muscle cell. ND: not determined

* C-kit might be cleaved by cardiac tissue enzymatic digestion. Martin et al found no expression of CD31, while Pfister et al reported two sub populations of CD31+ and CD31- among CSP. These differences may due to slight variation in isolation procedures used between groups.

** SP phenotype was not addressed through Hoechst exclusion but through MDR-1 expression.

*** Matsuura et al reported 40% and 10% of Sca-1 cells expressing CD45 and CD34, respectively.

Table 2

Summary of Major Cell-Based / Cytokine Clinical Trials

Study	Method of delivery	Patients treated/ control	Placebo/ control	Cell type Cell number or Dose	Time of cell delivery (days post-MI)	Results	Reference
Strauer	Intracoronary	10/10 30 (CPC)	control	BM-MNC 9×10^6 to 2.8×10^7 CPC	7	Improved contractility and reduced infarct size	Strauer et al 2002 [110]
TOPCARE-AMI	Intracoronary	29 (BM-MNC)	N/A	1.3×10^7 BM-MNC 2.4×10^8	3 to 7	Improved EF and reduced infarct size	Assmus et al 2002 [111] Britten et al 2003 [112] Schachinger et al 2004 [113]
BOOST	Intracoronary	30/30	control	BM-MNC 24×10^9	6	Improved EF at 6 months No difference at 18 months	Wollert et al 2004 [100] Meyer et al 2006 [101]
Janssens	Intracoronary	33/34	placebo	BM-MNC 3.0×10^8 cells	1	No effect	Janssens et al 2006 [114]
Chen	Intracoronary	34/35	placebo	MSC 48×10^{10} to 60×10^{10}	18	Improved and perfusion at 3 months	Chen et al 2004 [115]
REPAIR-AMI	Intracoronary	102/102	placebo	BM-MNC 2.4×10^8	4	Improved EF and reduced infarct size at 4 months	Schachinger et al 2006 [103]
ASTAMI	Intracoronary	100	control	BM-MNC 8.7×10^7	5 to 8	No difference at 6 months	Lunde et al 2006 [102]
FIRSTLINE-AMI	Mobilization	25/25	control	G-CSF 10μg/Kg BW	0 to 6 1QD	Improved EF and remodeling at 4 months	Ince et al. 2005 [104]
STEMMI	Mobilization	39/39	placebo	G-CSF 10μg/Kg BW	0 to 6 1QD	No difference at 6 months	Ripa et al 2006 [106]
REVIVAL II	Mobilization	56/58	placebo	G-CSF 10μg/Kg BW	0 to 5 1QD	No difference at 6 months	Zohlnhoefer et al 2006 [105]

BM-MNC: unfractionated bone marrow mononuclear cells; CPC: Circulating progenitor cells; MSC: mesenchymal stem cells; 1QD: one subcutaneous G-CSF injection per day; EF: left ventricular ejection fraction.