Review Article [Parikh *et al.*, 4(2): Feb., 2013]
CODEN (USA): IJPLCP **ISSN: 0976-7126**

INTERNATIONAL JOURNAL OF PHARMACY & LIFE SCIENCES

Stem cells as regenerative medicine targeting cardiovascular diseases: Covenant, Contingency and Challenges

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Abstract
Cardiovascular disease is the leading cause of morbidity and mortality worldwide. Repair of the heart is an old dream of physicians caring for patients with cardiac disease. There is now growing evidence that the human heart is capable of undergoing repair and in recent years there has been an increase in basic and clinical research with the aim of harnessing the regenerative properties of stem cells in order to facilitate restoration of myocardial function. Experimental studies suggest that cardiac transfer of stem and progenitor cells can have a favorable impact on tissue perfusion and contractile performance of the injured heart. For advancement several important aspects need to be addressed in carefully designed comparative studies which allow discriminating superior cell populations, time, dosage and delivery route and mode for different applications in patients with acute myocardial infarction, advanced coronary artery disease, and chronic heart failure. The overall clinical experience also suggests that stem cell therapy can be safely performed, if the right cell type is used in the right clinical setting. The future of stem cell research will require closer collaborative efforts between scientists and clinicians to understand how cell therapy works and to define the ideal cell type and method of delivery to be able to obtain maximum output.

Key-Words: Stem cells, Heart Failure, Cardiovascular Disease, Myocardial Infarction, Adult Stem Cell Therapy, Ischaemic heart disease

Introduction
Stem cells perform important functions in the establishment of embryonic tissues during development and, in some cases, are retained into adulthood where they support homeostasis through the continued replacement of senescent cells and regeneration of injured or diseased organs. The dogma of the heart as an organ composed of terminally differentiated myocytes incapable of regeneration is being challenged. The regenerative capacity of the human myocardium is, however, grossly inadequate to compensate for the severe loss of viable heart muscle that follows myocardial infarction (MI). Stem cells are capable of self-renewal, transformation into dedicated progenitor cells, and differentiation into specialized progeny. Traditionally, tissue-resident adult stem cells were believed to differentiate into progeny only within tissue lineage boundaries.

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Plasticity implies that stem cells can transdifferentiate into mature cell types outside their original lineage in response to microenvironmental cues. For example, hematopoietic stem cells (HSCs), when transplanted into the murine myocardium, may transdifferentiate into cardiomyocytes and blood vessels, thereby improving heart function and survival [1]. Existing therapies for heart failure, the leading cause of death worldwide, only serve to delay progression of the disease. More recent approaches have focused on replacement of injured myocardium with healthy cardiomyocytes, and the induction of neovascularization and significant effort has been invested in the search for the optimal embryonic or adult progenitor cells with which to replace damaged cells [2]. Evidence has been presented that a fraction of cardiomyocytes may be able to reenter the cell-cycle and that limited regeneration can occur through recruitment of resident and circulating stem cells [3]. We are standing on the merge to the era of biological repair in ischaemic cardiovascular disease after the

potential of cardiac repair by a variety of stem and progenitor cell populations has been revealed in pre-clinical and early clinical studies [4].

It has been proposed that stem cells release angiogenic ligands, protect cardiomyocytes from apoptotic cell death, induce proliferation of endogenous cardiomyocytes, and may recruit resident cardiac stem cells [5]. Regardless of the mechanisms, there appears to be general agreement that stem cell therapy has the potential to improve perfusion and contractile performance of the injured heart as shown in figure 1 [6]. However, the existence of endogenous repair mechanisms suggests that cardiac repair may be achieved therapeutically in these clinical settings will be reviewed in this article.

TYPES OF STEM CELLS USED FOR CARDIAC REPAIR

Conceptually, a variety of stem and progenitor cell populations could be used for cardiac repair. Each cell type has its own profile of advantages, limitations, and practicability issues in specific clinical settings. Studies comparing the regenerative capacity of distinct cell populations are scarce. Many investigators have therefore chosen a pragmatic approach by using unfractionated bone marrow cells (BMCs), which contain different stem and progenitor cell populations, including HSCs, endothelial progenitor cells (EPCs), and mesenchymal stem cells (MSCs) [7, 8].

Skeletal Myoblasts

Autologous skeletal myoblasts were among the first cell types tested in the context of cardiac regeneration [9], an obvious choice given their resistance to ischemia and ability to regenerate after injury [10]. They form myotubes in vivo but appear unable to differentiate into cardiomyocytes and yet are reported to improve ventricular function in animal studies. Human trials are ongoing, although, in some, lack of efficacy has resulted in their premature termination [11]. The main factor limiting the therapeutic use of skeletal myoblasts is their failure to integrate electrically with surviving cardiomyocytes [12], posing a greater risk of arrhythmia. Furthermore, skeletal myoblasts do not extravasate (transverse the vascular endothelium) and migrate to ischaemic areas [13] and may even obstruct distal microcirculation after intracoronary administration, leading to embolic myocardial damage [14].

Embryonic stem cells

Embryonic stem (ES) cells have the broadest developmental pluripotent potential since they can give rise to cells of all three embryonic germ layers and functionally intact cardiomyocytes have been generated from human ES cells in vitro [15]. In a mouse model ES cell-derived cardiomyocytes, when injected into infarcted myocardium, formed stable grafts and subsequently contracted in synchrony with adjacent cells [16]. However, the use of ES cells is associated with teratoma formation in animal models [17] which raises concerns regarding their malignant potential with the ethical and legal issues together surrounding the use of human ES cells, has hampered further research efforts and current focus is on other sources of stem cells for cardiac repair.

Induced Pluripotent Stem Cells

An exciting alternative to ES cells is emerging in the form of inducible Pluripotent Stem Cells (iPSCs) adult stem cells that have been successfully reprogrammed back to an undifferentiated pluripotent state by inserting four genes, Oct3/4, Sox2, KL4 and c-Myc, into differentiated somatic cells [18, 19. 20]. These cells have the morphological phenotype of ES cells and have been demonstrated in vivo and in vitro to have the same differentiation potential as ES cells (able to form all three germ layers). Functioning cardiomyocytes [21] have already been produced from iPSCs demonstrating their potential use in cardiovascular regenerative medicine although there remain theoretical concerns regarding tumor genesis.

Multipotent Adult Germline Stem Cells (maGSCs)

Adult spermatogonial stem cells were recently isolated from adult mouse testis and shown to acquire certain ES cell properties [22] including multipotency and germline transmission. These so-called multipotent adult germline stem cells (maGSCs) efficiently differentiate into ventricle-, atrial-, pacemaker-, and Purkinje-like cardiomyocytes, which exhibit rhythmic Ca^{+2} transients and beating [23]. Transplanted maGSCs were able to proliferate and differentiate in normal murine hearts, suggesting that maGSCs could provide a source of suitable cardiomyocytes for potential therapeutic application.

Bone marrow-derived progenitor/stem cells

The most widely studied of the adult stem cells has been bone marrow derived mononuclear cells (BMSCs) in part due to the ease of obtaining cells via a BM aspirate. In a landmark animal study, myocardial infarction was induced in a mouse model by coronary artery ligation following which BMSCs were injected directly into the contracting wall bordering the infarct [1]. The transplanted cells appeared to undergo trans-differentiation to cardiomyocytes with newly formed myocardium occupying a significant proportion of the infarcted area with significant improvement in the left ventricular ejection fraction (LVEF) just 9 days after cell transplantation. These results have been challenged by different groups, which have demonstrated that

transplanted cells do not acquire a cardiomyocyte phenotype but rather develop into haematopoietic cell types after transplantation [24, 25]. It is therefore possible that adult stem cell plasticity (i.e. ability to transdifferentiate into different cell types) has been overestimated particularly with regards to cardiomyogenic transdifferentiation. This has fuelled the ongoing debate regarding the mechanism of action by which stem cell therapy leads to cardiac repair and it is likely that the beneficial effects seen are multi-factorial in origin. Possible explanations include neovascularization by differentiation into an endothelial phenotype, paracrine effects of the cell infusate, cell fusion as well as myocardial regeneration [26].

Endothelial Progenitor Cells

Endothelial progenitor cells (EPCs) are a specialized subset of hematopoietic cells found in the bone marrow and peripheral circulation [27]. During development, endothelial and hematopoietic cells arise from a common progenitor, the hemangioblast, a cell type considered to be restricted to the embryo. Asahara *et al.* demonstrated that up to 20% of the CD34 population of peripheral blood in the adult are also vascular endothelial factor receptor (VEGFR) and phenotypically characterized by antigens usually associated with hematopoietic stem cells, including CD133, CD34, c-kit, VEGFR2, CD144 (cadherin), and Sca-1 [27]. EPCs are mobilized from bone marrow and recruited to foci of neovascularization, where they from new blood vessels in situ. EPCs are incorporated into injured vessels and develop into mature endothelial cells during the processes of reendothelialization and neovascularization [28]. On differentiation, CD133 expression is lost and EPCs begin to express vascular endothelial cadherin and von Willebrand factor [29]. EPCs have not been shown to differentiate into cardiomyocytes but appear to promote angiogenesis [30] and likely provide paracrine survival signals to cardiomyocytes [31]. Angiogenic growth factors including VEGF-A, VEGF-B, stromal cell– derived factor (SDF)-1, and insulin-like growth factor-1 are themselves secreted at high levels from EPCs, and these elicit a potent migratory response on mature endothelial cells and cardiac resident c-kit progenitor cells [32]. Thus, in addition to the physical contribution of EPCs to newly formed vessels, an equal, if not greater, mode of EPC action may, as with BMSCs, be the paracrine secretion of proangiogenic factors. The use of EPC populations for therapeutic purposes has rapidly progressed into clinical trials with promising preliminary results [33, 34].

Cardiac-derived Cardiovascular Stem Cells (CSCs)

Bergmann *et al.* [35] showed evidence for in-men cardiomyocytes renewal at a rate of 1% per year in younger adults and 0.5% in the elderly. In post-natal hearts, various subtypes of tissue-resident cardiac stem and progenitor cells (CSCs) classified by surface antigens and transcription markers have been reported, although it is undetermined whether these subtypes have clearly distinct phenotypes. Cardiac stem and progenitor cells, which have been suggested to be capable of creating cardiomyocytes and all surrounding cell types, are a promising candidate-at least in theory-to provide contractility and vascularization [36]. In the light of the fact that their genuine number is low, CSCs isolated from endomyocardial biopsies have successfully been expanded ex vivo to leverage this therapeutic concept [37]. Dr Marban's group has proposed a population of potential clinical relevance that has been identified by expanding CSCs from self-adherent clusters (cardiospheres) under certain conditions, i.e. cardiosphere-derived stem cells (CDCs) [38, 39]. There is still some controversy on the cardiomyogenic potential of cardiospheres [40, 41]. Dr Field's group had suggested by using genetic cell tracking that there are temporal limitations for the ability of cardiac-resident c-kit+ cells to acquire a cardiomyogenic phenotype, i.e. that the cardiomyogenic population is present in neonatal hearts but largely lost in adult mouse hearts and suggested that elucidation of the underlying molecular mechanisms may permit a more robust cardiomyogenic induction in adult-derived cardiac c-kit+ cells [42].

MECHANISMS OF STEM CELL DELIVERY TO VARIOUS PARTS OF THE HEART

Systemic delivery of stem cells as an effective therapy for the injured heart is dependent on successful homing and retention of cells before the secretion of paracrine factors and/or transdifferentiation. There are 2 ways in which cardiac progenitors can be delivered to the heart as shown in figure 2: either by an intracoronary arterial route or by injection into the ventricular wall via percutaneous endocardial, percutaneous transcoronary venous, or surgical epicardial approaches. Intracoronary delivery enables the application of a maximum dose of cells homogeneously to the site of injury although this mode is less efficient for delivery to nonperfused regions of the infarct related artery. Homing of intra-arterially applied progenitor cells requires their extravasation and migration to the surrounding ischemic tissue. Although BMSCs and hematopoietic stem cells can extravasate [43], and this has not been shown for all cell types and larger, less motile cells, such as skeletal myoblasts may even

obstruct the microcirculation, leading to embolic myocardial infarction [44]. Direct injection is the preferred delivery method for chronic heart failure patients with considerable scar tissue. Cell homing signals such as SDF-1 and VEGF are expressed at low levels at late stages of disease, limiting any homing potential following intracoronary application [45]. Injection into the injured myocardium is particularly suited for large cells such as myoblasts and mesenchymal stem cells and is not limited by cell uptake from the circulation or embolic risk. However, injection of progenitor cells into necrotic tissue, which lacks both blood flow to provide oxygen and nutrients and healthy surrounding cardiomyocytes to provide paracrine support, reduces graft survival and differentiation. The optimal delivery route for autologous cell transplantation not only varies according to the administered cell type but will be influenced in the future by our ability to enhance the migratory capacity of stem cells.

Hematopoietic Stem Cell Recruitment and Homing

During embryogenesis, blood-forming stem cells migrate from the fetal liver via the circulation, home to the bone marrow, and repopulate it with high numbers of immature and maturing blood cells of all lineages. These, in turn, are released into the circulation while maintaining a small pool of undifferentiated stem cells within the bone marrow [46]. HSC recruitment (mobilization) and homing are mirror processes regulated by the interplay of cytokines, chemokines, and proteases [47]. Essentially, HSC recruitment is characterized both by loss of cell– cell contacts (via downregulation of cell adhesion molecules) and a desensitization of chemokine signaling, notably the SDF-1α/CXCR4 axis, the fundamental signaling pathway underlying stem cell mobilization and homing during homeostasis and injury [48]. Conversely, stem cell homing requires upregulation of cell adhesion molecules and activation of the SDF-1α/CXCR4 axis.

Chemokine Signaling

Chemokines are defined as small peptides that initiate the migration of effector cells. Although mobilization of HSCs by cytokines requires 5 to 6 days to attain peak response, chemokines induce mobilization within 30 minutes to a few hours. Mobilization of HSCs from bone marrow is achieved by the action of cytokines, such as granulocyte colony stimulating factor (G-CSF) [49] and the closely related granulocyte/ macrophage colony-stimulating factor (GM-CSF) [50], Flt-3 ligand [51], erythropoietin [52], and stem cell factor (SCF) (the ligand for c-kit) [53]; growth factors such as vascular endothelial growth factor (VEGF) [54], angiopoietin-1 [55], and placental growth factor [56], as well as several chemokines such as SDF-1 [57], interleukin (IL)-8 [58], growth-regulated oncogene-β [59], and macrophage inflammatory protein-2 [60]. The first suggestion that cytokine-induced stem cell mobilization may be used to enhance cardiac repair came from studies to increase EPC levels for neovascularization in hind limb ischemia. VEGF [61] and GM-CSF [62] were found to augment EPC levels and improve neovascularization. Hematopoietic stem cell–mobilizing factors G-CSF and SCF were subsequently shown to improve cardiac regeneration in mice and this and other small scale animal studies rapidly led to initiation of clinical trials to assess the ability of G-CSF to mobilize stem/progenitor cells in patients with coronary artery disease [63] and AMI [64]. G(M)-CSF–mobilized blood from patients contained 5- to 100-fold higher levels of HSCs, MSCs, and EPCs, compared with nonmobilized blood [62-64]; however, the ability of these cells to improve cardiac remodeling and function after AMI has been disappointing [65]. Although the idea that recruited BMSCs differentiate into cardiomyocytes to any significant degree is now generally discounted, G-CSF treatment has been shown to induce angiogenesis in the infarcted heart and to have a paracrine protective effect on cardiomyocytes [65]. Chemokine signaling for directing migration is an embryonic principle because directed cell movement is a fundamental requirement for tissue formation. Chemokine receptors, predominantly CXCR4, have been detected from embryonic day (E), coexisting spatially and temporally with SDF-1, and have been implicated in the organogenesis of cardiovascular, neuronal, hematopoietic, and craniofacial systems [66].

Homing of Stem Cells to the Heart

The term "homing" describes the migration of a circulating stem cell into a target tissue or the bone marrow. Homing constitutes a multistep cascade comprising: recognition and interaction with microvascular endothelium, transmigration through the endothelium, and, finally, migration and invasion of the target tissue, a process that relies on a complex interplay between cytokines, chemokines, adhesion molecules, and extracellular matrix– degrading proteases. The capacity of stem cells to migrate and invade is critical for functional integration even when cells are injected directly into the site of injury. Although the mechanisms of progenitor cell homing to sites of tissue injury are poorly understood, some insight can be gained from parallels with the homing of hematopoietic progenitor cells to bone marrow [67]. Kollet *et al.* first demonstrated that HSCs used SDF-1α for homing to damaged tissue [68]. They observed that

the level of HSC engraftment into liver was greatly enhanced following injury or viral inflammation by elevated MMP-9 activity, which in turn led to increased CXCR4 expression and SDF-1α –mediated recruitment of hematopoietic progenitors to the liver [68]. It is well documented that stress signals such as tissue injury or inflammation cause upregulation of SDF-1α in endothelial cells, which promotes the recruitment of stem cells, as demonstrated for heart [69, 70], kidney [70], and brain [71]. Systemic mobilization and homing to sites of cardiac injury was suggested to be confounded by the trapping of cells in organs such as the spleen [72]; however, a number of studies have since demonstrated that cytokine therapy can overcome the complications of trapping and demonstrate significant cardiac regeneration in nonsplenectomized animals [73, 74]. Malek *et al.* addressed the issue of whether cardiac inflammation plays an important role in successful homing of ES cells to the heart after intravenous delivery in a murine myocarditis model [75]. Maximal engraftment of ES cells occurred at a time of peak inflammatory cytokine production, most notably IL-6, supporting the notion that factors released from the myocardium during an inflammatory response, as occurs in MI, are important for enhancing the homing, migration, and implantation of systemically infused stem cells.

Stem Cell Migration within the Myocardium

Cardiac progenitor cells, whether resident or transplanted, migrate through the interstitium of the heart, although the extent of migration and the mechanisms involved are poorly understood. Bone marrow–derived HSCs, introduced into remote myocardium [76] and cardiosphere-derived cells injected into the border zone [77] migrated to the infarct region, illustrated by tracking of EGFP and lacZ expressing cells, respectively. Although the mechanism of migration was not investigated in these studies, their directional migration toward the scar likely results from secretion of factors by dead or hypoxic cells in a similar manner to stem cell homing. Recent advances in noninvasive imaging technologies have enabled the tracking of progenitor cells, labeled with F-18 fluorodeoxyglucose or iron particles, during homing from the peripheral circulation to ischemic tissues [78] or their migration within the heart following cardiac delivery [79]. As cell therapy is further developed for heart repair, the ability to track cells will prove essential for assessing their capacity to home and migrate to the injury zone.

Migration of Resident Epicardium–Derived Cells stimulated by Thymosin β4

Significant effort in the field of cardiovascular medicine has been invested in the search for adult cardiac progenitor cells that may replace damaged muscle cells and/or contribute to new vessel formation (neovascularization) and in the identification of key factors, which may induce such progenitor cells to contribute to myocardial repair and collateral vessel growth. Smart N *et al.* demonstrated that the actin monomer-binding protein, thymosin beta-4 (Tbeta-4), when secreted from the myocardium provides a paracrine stimulus to the cells of the epicardium-derived cells (EPDCs) to promote their inward migration and differentiation into endothelial and smooth muscle cells to form the coronary vasculature. Translating this essential role for Tbeta-4 in coronary vessel development to the adult, we found that treatment of cultured adult explants with Tbeta-4 stimulated extensive outgrowth of epicardin-positive epicardial cells, which, as they migrated away from the explant, differentiated into procollagen type I, SMalphaA, and Flk1-positive cells indicative of fibroblasts, smooth muscle, and endothelial cells; thus releasing the adult epicardium from a quiescent state and restoring pluripotency. The ability of Tbeta-4 to promote coronary vessel development and potentially induce new vasculature in the adult is essential for cardiomyocyte survival and could contribute significantly toward the reported Tbeta4-induced cardioprotection and repair in the adult heart. Tbeta-4 is currently subject to multicenter phase 1 clinical trials for treatment of cardiovascular disease, therefore, insight into the repair mechanism(s) induced by Tbeta-4 is an essential step toward harnessing therapeutic survival, migration, and repair properties of the peptide in the context of acute myocardial damage [80].

Migration of Endogenous Cardiac Progenitor Cells

Several populations of cardiac progenitors residing within the adult heart have now been characterized. However, to maintain stemness, progenitors are required to be retained within a supportive stem cell niche [81]. One of the limitations for cardiac regeneration is that the small progenitor populations within the heart reside in a quiescent state and require reactivation before they can promote regeneration. Indeed some of the factors associated with reactivation and restoration of pluripotency, such as Tβ4, become upregulated following MI [82], but even the induced levels are insufficient for regeneration and require exogenous application or therapeutic augmentation of endogenous induction to promote sufficient repair [83]. A rapidly evolving paradigm, and one that holds much

promise for therapeutic myocardial regeneration, is the identification of paracrine factors that stimulate endogenous cardiac stem cells to migrate to the site of injury within the heart and differentiate into the cardiomyocyte and vascular cells required to induce neovascularization and repair. Only a few such factors have hitherto been identified, including high-mobility group box protein (HMGB)1 and Tβ4. It is perhaps significant that the common feature shared by these proteins is the ability to promote cell migration and suggests that migration of progenitors away from their restrictive niche is sufficient to reactivate their proliferation and differentiation.

FUTURISTIC DIRECTIONS FOR CELL BASED THERAPIES

Many strategies have been proposed to support stem cells in the host environment of ischaemic tissue characterized by ischaemia, acidosis, inflammation, and oxidative stress.

Bionanotechnology to support cell-based therapies

The rapidly evolving field of bionanotechnology allows to specifically design biomaterials to support transplanted cells within the ischaemic environment [84]. Herein, the structure, dimensions, and shape of constructs are pivotal to better mimic the native architecture of extracellular matrix. An optimal biomaterial to support cell therapy should provide a three-dimensional environment to enhance biomechanical properties of extracellular matrix; a purpose for which controlled organization at nano-scale is needed. In some biomatrices, bioactive signals can be incorporated to specifically modulate stem cell biology while supporting them structurally [85]. This strategy to support cell transfer has rapidly gained attention triggered by exciting pre-clinical data. In murine models, nanofibres self-assemble into a matrix recruited endogenous progenitors to the myocardium and support the transplantation of cardiomyocytes providing a particular microenvironment [86]. In principle, biomaterials can be custom-designed to optimally fit the organ-specific microenvironment [87]. Furthermore, bioactive signals can be incorporated in some biomaterials to additionally enhance cell survival, retention, proliferation, and differentiation. Padin-Iruegas *et al.* [88] reported that an insulin-like growth factor carrying nanofibre enhances CSC-dependent repair of cardiac injury [89]. In our hands, the combination of a linage-specific optimized, self-assembling nanofibre enhances the potency of cell-therapy in ischaemic tissue repair [90]. Also, bioactive sequences of biologically attractive paracrine factors, e.g. SDF-1, can more effectively be presented via biomaterials with the aim to recruit endogenous into or support exogenously applied cells in ischaemic myocardium [91]. Although emerging results for the role of bionanomaterials in cell-based ischaemic tissue repair are promising, but still there has not been any applications in humans.

Priming of stem and progenitor cells to enhance their therapeutic efficacy

The concept to pre-treat or modify stem/progenitor cells before application (priming) and thereby enhance their therapeutic potency has evolved from earlier pre-clinical observations [92, 98]. These strategies basically target any function step that influences cell fate from the application on: adhesion/transmigration, homing, migration, engraftment, survival, cell–cell interaction, repair capacity, differentiation, and retention. Potential tools for modification include drugs, small molecules, naked and vector facilitated plasmids, and epigenetic reprogramming [99, 100]. Priming of dysfunctional autologous cells from cardiovascular patients via any of these tools may allow for a 'resetting of impaired biopotency'.

Among multiple targets stemming from pre-clinical evaluation, the following examples are under clinical investigation: we and others have identified a reduced endothelial NO synthase dependent NO production as an important mechanism limiting the functional repair capacity of endogenous progenitor cells in patients with diabetes or hypertension [101].

Conclusion

The past decade has seen an explosion in clinical studies investigating the safety and efficacy of stem cell therapy for heart diseases. The safety of this therapy has been demonstrated uniformly in the vast majority of the studies despite heterogeneity in study design. In terms of efficacy there does seem to be some beneficial effects of cell therapy in the settings of AMI, chronic ischaemic heart failure and DCM. However, the magnitude of benefit is less impressive than was seen in the previous animal models. The future of this area of research will rely on elucidating the reasons for this difference which will require closer collaboration between basic scientists and clinical researchers. There is also a need for larger randomized controlled trials with longer term follow-up assessing morbidity and mortality as primary outcome. An understanding of the mechanisms involved in initiation and regulation, along with identification of factors that direct these processes, may offer approaches to enhance present strategies involving stem cell engraftment. Moreover, approaches requiring the introduction of exogenous stem cells are hampered by inefficient homing and immune rejection. In the absence of an effective intervention to optimize migration of transplanted cells,

a preferable therapeutic strategy might be to stimulate one or more of the identified populations of endogenous cardiac stem cells to initiate repair in situ. Before any proliferation or differentiation, they key initial event required to unleash the potential of these cells is their migration from the stem cell niche to the site of injury. The search is underway, therefore, to identify factors that can revive the potential of these cells to achieve efficient cardiac regeneration.

Acknowledgements

The corresponding author would like to thank Dr. Bakul Desai for his wisdom support and gracious guidance for the completion of this review.

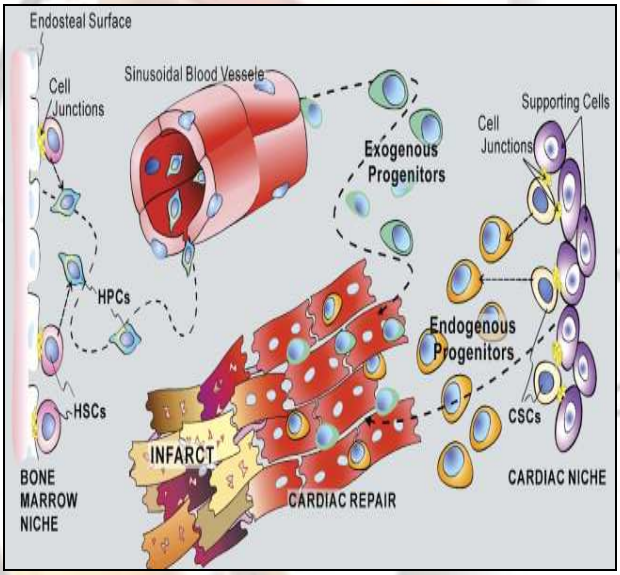

Fig. 1: Working hypothesis of therapeutic stem cell transplantation for myocardial regeneration. Stem and progenitor cell transplantation can have a favorable impact on tissue perfusion and contractile performance by promoting vascularization and myocyte formation. Improved vascularization may facilitate beneficial effects in the myocyte compartment.

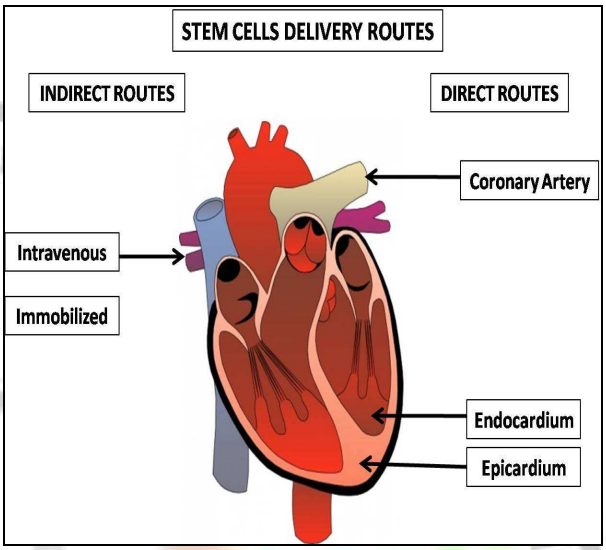

Fig. 2: Stem Cells delivery routes to various localities in the heart for Stem Cell Therapy.

References

[1] Orlic D., Kajstura .J, Chimenti S., Jakoniuk I., Anderson S. M., Li B., Pickel J., McKay R., Nadal-Ginard B, Bodine D.M., Leri A., Anversa P. (2001). Bone marrow cells regenerate infarcted myocardium. Nature. 410: 701–705.

[2] Nicola S., Paul R. R. (2008). The Stem Cell Movement. Circ Res. 102: 1155-1168.

[3] Nadal-Ginard B., Kajstura J., Leri A., Anversa P. (2003). Myocyte death, growth, and regeneration in cardiac hypertrophy and failure. Circ Res. 92: 139–150.

[4] Jörn T., Douglas W. L., Ulf L. (2011). Stem and progenitor cell-based therapy in ischaemic heart disease: promise, uncertainties, and challenges. European Heart Journal. 32: 1197–1206.

[5] Kai C. W., Helmut D. (2005). Clinical Applications of Stem Cells for the Heart. Circ Res. 96:151-163,

[6] Kocher A. A., Schuster M. D., Szabolcs M. J., Takuma S., Burkhoff D., Wang J., Homma S.,

Edwards N. M., Itescu S. (2001). Neovascularization of ischemic myocardium by human bone-marrow-derived angioblasts prevents cardiomyocyte apoptosis, reduces remodeling and improves cardiac function. Nat Med. 7:430–436.

[7] Strauer B. E., Brehm M., Zeus T., Kostering M., Hernandez A., Sorg R. V., Kogler G., Wernet P. (2002). Repair of infarcted myocardium by autologous intracoronary mononuclear bone marrow cell transplantation in humans. Circulation. 106:1913–1918.

[8] Assmus B., Schachinger V., Teupe C., Britten M., Lehmann R., Dobert N., Grunwald F., Aicher A., Urbich C., Martin H., Hoelzer D., Dimmeler S., Zeiher A. M. (2002). Transplantation of Progenitor Cells and Regeneration Enhancement in Acute Myocardial Infarction (TOPCARE-AMI). Circulation. 106:3009 –3017.

[9] Menasche P. (2007). Skeletal myoblasts as a therapeutic agent. Prog Cardiovasc Dis. 50:7–17.

[10] Shi X., Garry D. J. (2006). Muscle stem cells in development, regeneration, and disease. Genes Dev. 20:1692–1708.

[11] Cleland J. G., Coletta A., Abdellah A. T., Nasir M., Hobson N. F., Freemantle N. F., Clark A. L. Clinical trials update from the Am Heart Association 2006: OAT, SALT 1 and 2, MAGIC, ABCD, PABA-CHF, IMPROVE-CHF, and percutaneous mitral annuloplasty. Eur J Heart Fail. 9:92–97.

[12] Laflamme M. A., Murry C. E. (2005). Regenerating the heart. Nat Biotech. 23:845–856.

[13] Aicher A., Brenner W., Zuhayra M., Badorff C., Massoudi S., Assmus B., Eckey T., Henze E., Zeiher A. M., Dimmeler S. (2003). Assessment of the tissue distribution of transplanted human endothelial progenitor cells by radioactive labeling. Circulation. 107:2134 –2139.

[14] Dimmeler S, Zeiher AM, Schneider MD. (2005) Unchain my heart: the scientific foundations of cardiac repair. J Clin Invest. 115:572–583.

[15] Xu C., Police S., Rao N. (2002). Characterization and enrichment of cardiomyocytes derived from human embryonic stem cells. Circ Res. 91:501–8.

[16] Klug M. G., Soonpaa M. H., Koh G. Y. (1996). Genetically selected cardiomyocytes from differentiating embronic stem cells form stable intracardiac grafts. J Clin Invest. 98:216–24.

[17] Lee A. S., Tang C., Cao F. (2009). Effects of cell number on teratoma formation by human embryonic stem cells. Cell Cycle. 8:2608–12.

[18] Takahashi K., Yamanaka S. (2006). Induction of pluripotent stem cells from mouse embryonic and adult fibroblast cultures by defined factors. Cell 126:663–76.

[19] Aasen T., Raya A., Barrero M. J. (2008). Efficient and rapid generation of induced pluripotent stem cells from human keratinocytes. Nat Biotechnol. 26:1276–84.

[20] Takahashi K, Tanabe K, Ohnuki M. (2007). Induction of pluripotent stem cells from adult human fibroblasts by defined factors. Cell. 131:861–72.

[21] Zhang J., Wilson G. F., Soerens A. G. (2009). Functional cardiomyocytes derived from human induced pluripotent stem cells. Circ Res 104:30–41.

[22] Guan K., Nayernia K., Maier L. S., Wagner S., Dressel R., Lee J. H., Nolte J., Wolf F., Li M., Engel W., Hasenfuss G. (2006). Pluripotency of

spermatogonial stem cells from adult mouse testis. Nature. 440:1199 –1203.

[23] Guan K., Wagner S., Unsold B., Maier L. S., Kaiser D., Hemmerlein B., Nayernia K., Engel W., Hasenfuss G. (2007). Generation of functional cardiomyocytes from adult mouse spermatogonial stem cells. Circ Res. 100:1615–1625.

[24] Balsam L. B., Wagers A. J., Christensen J. L. (2004). Haematopoietic stem cells adopt mature haematopoietic fates in ischaemic myocardium. Nature. 428:668–73.

[25] Murry C. E., Soonpaa M. H., Reinecke H. (2004). Haematopoietic stem cells do not transdifferentiate into cardiac myocytes in myocardial infarcts. Nature. 428:664–8.

[26] Dimmeler S., Zeiher A. M., Schneider M. D. (2005). Unchain my heart: the scientific foundations of cardiac repair. J Clin Invest. 115:572–83.

[27] Asahara T., Murohara T., Sullivan A., Silver M., van der Z. R., Li T., Witzenbichler B., Schatteman G., Isner J. M. (1997). Isolation of putative progenitor endothelial cells for angiogenesis. Science. 275:964 –967.

[28] Miller-Kasprzak E., Jagodzinski P. P. (2007). Endothelial progenitor cells as a new agent contributing to vascular repair. Arch Immunol Ther Exp. 55:247–259.

[29] Hristov M., Erl W., Weber P. C. (2003). Endothelial progenitor cells: mobilization, differentiation, and homing. Arterioscler Thromb Vasc Biol. 23: 1185–1189.

[30] Rubart M., Field L. J. (2006). Cardiac regeneration: repopulating the heart. Annu Rev Physiol. 68:29–49.

[31] Narmoneva D. A., Vukmirovic R., Davis M. E., Kamm R. D., Lee R. T. (2004). Endothelial cells promote cardiac myocyte survival and spatial reorganization: implications for cardiac regeneration. Circulation. 110: 962–968.

[32] Urbich C., Aicher A., Heeschen C., Dernbach E., Hofmann W. K., Zeiher A. M., Dimmeler S. (2005). Soluble factors released by endothelial progenitor cells promote migration of endothelial cells and cardiac resident progenitor cells. J Mol Cell Cardiol. 39:733–742.

[33] Schachinger V., Assmus B., Honold J., Lehmann R., Hofmann W. K., Martin H., Dimmeler S., Zeiher A. M. (2006). Normalization of coronary blood flow in the infarct-related artery after intracoronary progenitor cell therapy: intracoronary Doppler substudy of the TOPCARE-AMI trial. Clin Res Cardiol. 95:13–22.

[34] Strauer B. E., Brehm M., Zeus T., Kostering M., Hernandez A., Sorg R. V., Kogler G., Wernet P. (2002). Repair of infarcted myocardium by autologous intracoronary mononuclear bone marrow cell transplantation in humans. Circulation. 106:1913–1918.

[35] Bergmann O., Bhardwaj R. D., Bernard S., Zdunek S., Barnabe-Heider F., Walsh S., Zupicich J., Alkass K., Buchholz B. A., Druid H., Jovinge S., Frisen J. (2009). Evidence for cardiomyocytes renewal in humans. Science 324:98–102.

[36] Beltrami A. P., Barlucchi L., Torella D., Baker M., Limana F., Chimenti S., Kasahara H., Rota M., Musso E., Urbanek K., Leri A., Kajstura J., Nadal-Ginard B., Anversa P. (2003). Adult

cardiac stem cells are multipotent and support myocardial regeneration. Cell.114:763–776.

[37] Bearzi C., Rota M., Hosoda T., Tillmanns J., Nascimbene A., De Angelis A., Yasuzawa-Amano S., Trofimova I., Siggins R. W., Lecapitaine N., Cascapera S., Beltrami A. P., D'Alessandro D. A., Zias E., Quaini F., Urbanek K., Michler R. E., Bolli R., Kajstura J., Leri A., Anversa P. (2007). Human cardiac stem cells. Proc Natl Acad Sci USA. 104:14068–14073.

[38] Smith R. R., Barile L., Cho H. C., Leppo M. K., Hare J. M., Messina E., Giacomello A., Abraham M. R., Marban E. (2007). Regenerative potential of cardiosphere-derived cells expanded from percutaneous endomyocardial biopsy specimens. Circulation. 115:896–908.

[39] Johnston P. V., Sasano T., Mills K., Evers R., Lee S. T., Smith R. R., Lardo A. C., Lai S., Steenbergen C., Gerstenblith G., Lange R., Marban E. (2009). Engraftment, differentiation, and functional benefits of autologous cardiosphere-derived cells in porcine ischemic cardiomyopathy. Circulation. 120:1075–1083.

[40] Andersen D. C., Andersen P., Schneider M., Jensen H. B., Sheikh S. P. (2009). Murine 'cardiospheres' are not a source of stem cells with cardiomyogenic potential. Stem Cells. 27:1571–1581.

[41] Davis D. R., Ruckdeschel Smith R., Marban E. (2010). Human cardiospheres are a source of stem cells with cardiomyogenic potential. Stem Cells. 28:903–904.

[42] Zaruba M. M., Soonpaa M., Reuter S., Field L. J. (2010). Cardiomyogenic potential of C-kit(+)-expressing cells derived from neonatal and adult mouse hearts. Circulation. 121:1992–2000.

[43] Aicher A., Brenner W., Zuhayra M., Badorff C., Massoudi S., Assmus B., Eckey T., Henze E., Zeiher A. M., Dimmeler S. (2003). Assessment of the tissue distribution of transplanted human endothelial progenitor cells by radioactive labeling. Circulation. 107:2134 –2139.

[44] Dimmeler S., Zeiher A. M., Schneider M. D. (2005). Unchain my heart: the scientific foundations of cardiac repair. J Clin Invest. 115:572–583.

[45] Perin E. C., Dohmann H. F., Borojevic R., Silva S. A., Sousa A. L., Mesquita C. T., Rossi M. I., Carvalho A. C., Dutra H. S., Dohmann H. J., Silva G. V., Belem L., Vivacqua R., Rangel F. O., Esporcatte R., Geng Y. J., Vaughn W. K., Assad J. A., Mesquita E. T., Willerson J. T. (2003). Transendocardial, autologous bone marrow cell transplantation for severe, chronic ischemic heart failure. Circulation. 107:2294 – 2302.

[46] Morrison S. J., Uchida N., Weissman I. L. (1995). The biology of hematopoietic stem cells. Annu Rev Cell Dev Biol. 11:35–71.

[47] Gazitt Y. (2004). Homing and mobilization of hematopoietic stem cells and hematopoietic cancer cells are mirror image processes, using similar signaling pathways and occurring concurrently: circulating cancer cells constitute an ideal target for concurrent treatment with chemotherapy and antilineage-specific antibodies. Leukemia. 18:1–10.

[48] Lapidot T., Dar A., Kollet O. (2005). How do stem cells find their way home? Blood. 106:1901–1910.

[49] Winter J. N., Lazarus H. M., Rademaker A., Villa M., Mangan C., Tallman M., Jahnke L., Gordon

L., Newman S., Byrd K., Cooper B. W., Horvath N., Crum E., Stadtmauer E. A., Conklin E., Bauman A., Martin J., Goolsby C., Gerson S. L., Bender J., O'Gorman M. (1996). Phase I/II study of combined granulocyte colony-stimulating factor and granulocyte-macrophage colony stimulating factor administration for the mobilization of hematopoietic progenitor cells. J Clin Oncol. 14:277–286.

[50] Gianni A. M., Siena S., Bregni M., Tarella C., Stern A. C., Pileri A., Bonadonna G. (1989). Granulocyte-macrophage colony-stimulating factor to harvest circulating haemopoietic stem cells for autotransplantation. Lancet. 2:580 –585.

[51] Fukuda S., Broxmeyer H. E., Pelus L.M. (2005). Flt3 ligand and the Flt3 receptor regulate hematopoietic cell migration by modulating the SDF- 1alpha (CXCL12)/CXCR4 axis. Blood. 105:3117–3126.

[52] Heeschen C., Aicher A., Lehmann R., Fichtlscherer S., Vasa M., Urbich C., Mildner-Rihm C., Martin H., Zeiher A. M., Dimmeler S. (2003). Erythropoietin is a potent physiologic stimulus for endothelial progenitor cell mobilization. Blood. 102:1340 –1346.

[53] Bodine D. M., Seidel N. E., Zsebo K. M., Orlic D. (1993). In vivo administration of stem cell factor to mice increases the absolute number of pluripotent hematopoietic stem cells. Blood. 82:445– 455.

[54] Gill M., Dias S., Hattori K., Rivera M. L., Hicklin D., Witte L., Girardi L., Yurt R., Himel H., Rafii S. (2001). Vascular trauma induces rapid but transient mobilization of VEGFR2(+)AC133(+) endothelial precursor cells. Circ Res. 88:167–174.

[55] Hattori K., Dias S., Heissig B., Hackett N. R., Lyden D., Tateno M., Hicklin D. J., Zhu Z., Witte L., Crystal R. G., Moore M. A., Rafii S. (2001). Vascular endothelial growth factor and angiopoietin-1 stimulate postnatal hematopoiesis by recruitment of vasculogenic and hematopoietic stem cells. J Exp Med. 193:1005–1014.

[56] Hattori K., Heissig B., Wu Y., Dias S., Tejada R., Ferris B., Hicklin D. J., Zhu Z., Bohlen P., Witte L., Hendrikx J., Hackett N. R., Crystal R. G., Moore M. A., Werb Z., Lyden D., Rafii S. (2002). Placental growth factor reconstitutes hematopoiesis by recruiting VEGFR1 stem cells from bone-marrow microenvironment. Nat Med. 8:841– 849.

[57] Hattori K., Heissig B., Tashiro K., Honjo T., Tateno M., Shieh J. H., Hackett N. R., Quitoriano M. S., Crystal R. G., Rafii S., Moore M. A. (2001). Plasma elevation of stromal cell-derived factor-1 induces mobilization of mature and immature hematopoietic progenitor and stem cells. Blood. 97: 3354–3360.

[58] Laterveer L., Lindley I. J., Hamilton M. S., Willemze R., Fibbe W. E. (1995). Interleukin-8 induces rapid mobilization of hematopoietic stem cells with radioprotective capacity and long-term myelolymphoid repopulating ability. Blood. 85:2269 –2275.

[59] Pelus L. M., Bian H., King A. G., Fukuda S. (2004). Neutrophil-derived MMP-9 mediates synergistic mobilization of hematopoietic stem and progenitor cells by the combination of G-CSF and the chemokines GRObeta/ CXCL2 and GRObetaT/CXCL2delta4. Blood. 103:110–119.

[60] Wang J., Mukaida N., Zhang Y., Ito T., Nakao S., Matsushima K. (1997). Enhanced mobilization of

hematopoietic progenitor cells by mouse MIP-2 and granulocyte colony-stimulating factor in mice. J Leukoc Biol. 62: 503–509.

[61] Asahara T. F., Takahashi T. F., Masuda H. F., Kalka C. F., Chen D. F., Iwaguro H. F., Inai Y. F., Silver M. F., Isner J. M. (1999). VEGF contributes to postnatal neovascularization by mobilizing bone marrow-derived endothelial progenitor cells. EMBO J. 18:3964 –3972.

[62] Takahashi T., Kalka C., Masuda H., Chen D., Silver M., Kearney M., Magner M., Isner J. M., Asahara T. (1999). Ischemia- and cytokine-induced mobilization of bone marrow-derived endothelial progenitor cells for neovascularization. Nat Med. 5:434–438.

[63] Powell T. M., Paul J. D., Hill J. M., Thompson M., Benjamin M., Rodrigo M., McCoy J. P., Read E. J., Khuu H. M., Leitman S. F., Finkel T., Cannon R. O. (2005). Granulocyte colony-stimulating factor mobilizes functional endothelial progenitor cells in patients with coronary artery disease. Arterioscler Thromb Vasc Biol. 25:296 –301.

[64] Valgimigli M., Rigolin G. M., Cittanti C., Malagutti P., Curello S., Percoco G., Bugli A. M., Della P. M., Bragotti L. Z., Ansani L., Mauro E., Lanfranchi A., Giganti M., Feggi L., Castoldi G., Ferrari R. (2005). Use of granulocyte-colony stimulating factor during acute myocardial infarction to enhance bone marrow stem cell mobilization in humans: clinical and angiographic safety profile. Eur Heart J. 26:1838 –1845.

[65] Kurdi M., Booz G. W. (2007). G-CSF-based stem cell therapy for the heartunresolved issues part A: paracrine actions, mobilization, and delivery. Congest Heart Fail. 13:221–227.

[66] McGrath K. E., Koniski A. D., Maltby K. M., McGann J. K., Palis J. (1999). Embryonic expression and function of the chemokine SDF-1 and its receptor, CXCR4. Dev Biol. 213:442–456.

[67] Papayannopoulou T. (2003). Bone marrow homing: the players, the playfield, and their evolving roles. Curr Opin Hematol. 10:214 –219.

[68] Kollet O., Shivtiel S., Chen Y. Q., Suriawinata J., Thung S. N., Dabeva M. D., Kahn J., Spiegel A., Dar A., Samira S., Goichberg P., Kalinkovich A., renzana-Seisdedos F., Nagler A., Hardan I., Revel M., Shafritz D. A., Lapidot T. (2003). HGF, SDF-1, and MMP-9 are involved in stress-induced human CD34+ stem cell recruitment to the liver. J Clin Invest. 112:160 –169.

[69] Askari A. T., Unzek S., Popovic Z. B., Goldman C. K., Forudi F., Kiedrowski M., Rovner A., Ellis S. G., Thomas J. D., DiCorleto P. E., Topol E. J., Penn M. S. (2003). Effect of stromal-cell-derived factor 1 on stem-cell homing and tissue regeneration in ischaemic cardiomyopathy. Lancet. 362:697–703.

[70] Ratajczak M. Z., Kucia M., Reca R., Majka M., Janowska-Wieczorek A., Ratajczak J. (2004). Stem cell plasticity revisited: CXCR4-positive cells expressing mRNA for early muscle, liver and neural cells ‘hide out’ in the bone marrow. Leukemia. 18:29–40.

[71] Stumm R. K., Rummel J., Junker V., Culmsee C., Pfeiffer M., Krieglstein J., Hollt V., Schulz S. (2002). A dual role for the SDF-1/CXCR4 chemokine receptor system in adult brain: isoform-selective regulation of SDF-1 expression

modulates CXCR4-dependent neuronal plasticity and cerebral leukocyte recruitment after focal ischemia. J Neurosci. 22:5865–5878.

[72] Aicher A., Brenner W., Zuhayra M., Badorff C., Massoudi S., Assmus B., Eckey T., Henze E., Zeiher A. M., Dimmeler S. (2003). Assessment of the tissue distribution of transplanted human endothelial progenitor cells by radioactive labeling. Circulation. 107:2134 –2139.

[73] Dawn B., Guo Y., Rezazadeh A., Huang Y., Stein A. B., Hunt G., Tiwari S., Varma J., Gu Y., Prabhu S. D., Kajstura J., Anversa P., Ildstad S. T., Bolli R. (2006). Postinfarct cytokine therapy regenerates cardiac tissue and improves left ventricular function. Circ Res. 98:1098 –1105.

[74] Misao Y., Takemura G., Arai M., Ohno T., Onogi H., Takahashi T., Minatoguchi S., Fujiwara T., Fujiwara H. (2006). Importance of recruitment of bone marrow-derived CXCR4 cells in post-infarct cardiac repair mediated by G-CSF. Cardiovasc Res. 71:455– 465.

[75] Malek S., Kaplan E., Wang J. F., Ke Q., Rana J., Chen Y., Rahim B., Li M., Huang Q., Xiao Y. F., Verheugt .F, Morgan J., Min J. Y. (2006). Successful implantation of intravenously administered stem cells correlates with severity of inflammation in murine myocarditis. Pflugers Arch Eur J Physiol. 452:268 –275.

[76] Orlic D., Kajstura J., Chimenti S., Jakoniuk I., Anderson S. M., Li B., Pickel J., McKay R., Nadal-Ginard B., Bodine D. M., Leri A., Anversa P. (2001). Bone marrow cells regenerate infarcted myocardium. Nature. 410: 701–705.

[77] Balsam L. B., Wagers A. J., Christensen J. L., Kofidis T., Weissman I. L., Robbins R. C. (2004). Haematopoietic stem cells adopt mature haematopoietic fates in ischaemic myocardium. Nature. 428:668–673.

[78] Hoehn M., Kustermann E., Blunk J., Wiedermann D., Trapp T., Wecker S., Focking M., Arnold H., Hescheler J., Fleischmann B. K., Schwindt W., Buhrle C. (2002). Monitoring of implanted stem cell migration in vivo: a highly resolved in vivo magnetic resonance imaging investigation of experimental stroke in rat. Proc Natl Acad Sci USA. 99:16267–16272.

[79] Stuckey D. J., Carr C. A., Martin-Rendon E., Tyler D. J., Willmott C., Cassidy P. J., Hale S. J. M., Schneider J. E., Tatton L., Harding S. E., Radda G. K., Watt S., Clarke K. (2006). Iron particles for noninvasive monitoring of bone marrow stromal cell engraftment into, and isolation of viable engrafted donor cells from, the Heart. Stem Cells. 24:1968 –1975.

[80] Smart N., Risebro C. A., Melville A. A., Moses K., Schwartz R. J., Chien K. R., Riley P. R. (2007). Thymosin beta-4 is essential for coronary vessel development and promotes neovascularization via adult epicardium. Ann N Y Acad Sci. 1112:171-88.

[81] Spradling A., Drummond-Barbosa D., Kai T. (2001). Stem cells find their niche. Nature. 414:98 –104.

[82] Smart N., Risebro C. A., Melville A. A. D., Moses K., Schwartz R. J., Chien K. R., Riley P. R. (2007). Thymosin β4 induces adult epicardial progenitor mobilization and neovascularization. Nature. 445:177–182.

[83] Bock-Marquette I., Saxena A., White M. D., Dimaio J. M., Srivastava D. (2004). Thymosin 4 activates integrin-linked kinase and promotes

cardiac cell migration, survival and cardiac repair. Nature. 432:466–472.

[84] Laflamme M. A., Murry C. E. (2005). Regenerating the heart. Nat Biotechnol 23: 845–856.

[85] Tongers J., Webber M. J., Losordo D. W. (2009). Bioengineering to enhance progenitor cell therapeutics. Tex Heart Inst J. 36:140–144.

[86] Davis M. E., Motion J. P., Narmoneva D. A., Takahashi T., Hakuno D., Kamm R. D., Zhang S., Lee R. T. (2005). Injectable self-assembling peptide nanofibers create intramyocardial microenvironments for endothelial cells. Circulation. 111:442–450.

[87] Davis M. E., Hsieh P. C., Grodzinsky A. J., Lee R. T. (2005). Custom design of the cardiac microenvironment with biomaterials. Circ Res. 97:8–15.

[88] Padin-Iruegas M. E., Misao Y., Davis M. E., Segers V. F., Esposito G., Tokunou T., Urbanek K., Hosoda T., Rota M., Anversa P., Leri A., Lee R. T., Kajstura J. (2009). Cardiac progenitor cells and biotinylated insulin-like growth factor-1 nanofibers improve endogenous and exogenous myocardial regeneration after infarction. Circulation 2009;120:876–887.

[89] Davis M. E., Hsieh P. C., Takahashi T., Song Q., Zhang S., Kamm R. D., Grodzinsky A.J., Anversa P., Lee R. T. (2006). Local myocardial insulin-like growth factor 1 (IGF-1) delivery with biotinylated peptide nanofibers improves cell therapy for myocardial infarction. Proc Natl Acad Sci USA. 103:8155–8160.

[90] Tongers J., Webber M. G., Renault M. A., Roncalli J. G., Jujo K., Wu X., Klyachko E., Thorne T., Stupp S. I., Losordo D. W. (2009). Abstract 4931: RGDS-epitope presenting peptide amphiphile nanofibers enhance therapeutic potency of cell-based strategies in ischemic tissue. Circulation. 120:S1023-c-1024.

[91] Segers V. F., Tokunou T., Higgins L. J., MacGillivray C., Gannon J., Lee R. T. (2007). Local delivery of protease-resistant stromal cell derived factor-1 for stem cell recruitment after myocardial infarction. Circulation. 116:1683–1692.

[92] Sorrentino S. A., Bahlmann F. H., Besler C., Muller M., Schulz S., Kirchhoff N., Doerries C., Horvath T., Limbourg A., Limbourg F., Fliser D., Haller H., Drexler H., Landmesser U. (2007). Oxidant stress impairs in vivo reendothelialization capacity of endothelial progenitor cells from patients with type 2 diabetes mellitus: restoration by the peroxisome proliferator-activated receptor-gamma agonist rosiglitazone. Circulation. 116:163–173.

[93] Murasawa S. , Llevadot J., Silver M., Isner J. M., Losordo D. W., Asahara T. (2002). Constitutive human telomerase reverse transcriptase expression enhances regenerative properties of endothelial progenitor cells. Circulation. 106:1133–1139.

[94] Yamaguchi J., Kusano K. F., Masuo O., Kawamoto A., Silver M., Murasawa S., Bosch-Marce M., Masuda H., Losordo D. W., Isner J.M., Asahara T. (2003). Stromal cellderived factor-1 effects on ex vivo expanded endothelial progenitor cell recruitment for ischemic neovascularization. Circulation. 107:1322–1328.

[95] Assmus B. , Urbich C., Aicher A., Hofmann W. K., Haendeler J., Rossig L., Spyridopoulos I., Zeiher A. M., Dimmeler S. (2003). HMG-CoA

reductase inhibitors reduce senescence and increase proliferation of endothelial progenitor cells via regulation of cell cycle regulatory genes. Circ Res. 92:1049–1055.

[96] Vasa M., Fichtlscherer S., Adler K., Aicher A., Martin H., Zeiher A. M., Dimmeler S. (2001). Increase in circulating endothelial progenitor cells by statin therapy in patients with stable coronary artery disease. Circulation. 103:2885–2890.

[97] Dimmeler S., Aicher A., Vasa M., Mildner-Rihm C., Adler K., Tiemann M., Rutten H., Fichtlscherer S., Martin H., Zeiher A. M. (2001). HMG-CoA reductase inhibitors (statins) increase endothelial progenitor cells via the PI 3-kinase/Akt pathway. J Clin Invest. 108:391–397.

[98] Bahlmann F. H., de Groot K., Mueller O., Hertel B., Haller H., Fliser D. (2005). Stimulation of endothelial progenitor cells: a new putative therapeutic effect of angiotensin II receptor antagonists. Hypertension. 45:526–529.

[99] Penn M. S., Mangi A. A. (2008). Genetic enhancement of stem cell engraftment, survival, and efficacy. Circ Res. 102:1471–1482.

[100] Chavakis E., Koyanagi M., Dimmeler S. (2010). Enhancing the outcome of cell therapy for cardiac repair: progress from bench to bedside and back. Circulation. 121: 325–335.

[101] Giannotti G., Doerries C., Mocharla P. S., Mueller M. F., Bahlmann F. H., Horvath T., Jiang H., Sorrentino S. A., Steenken N., Manes C., Marzilli M., Rudolph K. L., Luscher T. F., Drexler H., Landmesser U. (2010). Impaired endothelial repair capacity of early endothelial progenitor cells in prehypertension: relation to endothelial dysfunction. Hypertension. 55:1389–1397.