Journal of
Stem Cell Research & Therapy
ISSN: 2157-7633

Faulkner et al., J Stem Cell Res Ther 2011, S1
DOI: 10.4172/2157-7633.S1-004

Review Article Open Access

Stem Cell Therapy: A New Approach for Treatment of Myocardial Infarction

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Abstract

Stem cell therapy offers the opportunity of myocardial repair in patients who have suffered a myocardial infarction, a concept currently not feasible with present treatment options. Embryonic, adult, and induced pluripotent stem cells all offer a potential cell source for myocardial repair. Pre-clinical studies suggest that embryonic and induced pluripotent stem cells may be the most ideal cell type as they have the potential to differentiate into cardiomyocytes and restore a degree of functional recovery in animal models. Due to practical and ethical issues surrounding these cell types however, more focus has been on the use of adult stem cells, predominantly those of the bone-marrow. Pre-clinical studies suggest that bone-marrow stem cells can promote a degree of functional recovery through either differentiating into cardiomyocytes or acting in a paracrine manner to promote neoangiogenesis. The apparent success in pre-clinical models paved the way to a number of clinical trials to take place. Although mixed results have been reported, these trials have however shown that stem cell therapy is safe and feasible in humans. Many questions are still unanswered including, what is the optimal cell type, dose and timing of transplantation. This review highlights the benefits and limitations of each cell type and possible regenerative mechanisms.

Introduction

Cardiovascular disease is the leading cause of death in the Western world. In the United Kingdom alone, there are approximately 124,000 cases of myocardial infarction (MI) every year [1].

The main treatment option for these patients is revascularization therapy including percutaneous coronary intervention (PCI) and coronary artery bypass grafting (CABG). The aim of such procedures is to restore blood flow to the ischaemic myocardium in a timely manner in an attempt to limit the degree of myocardial damage and subsequent death. However, most patients who suffer an MI display impaired heart function and many go on to develop heart failure as myocardial performance declines, thus requiring additional intervention and hospitalization [2]. Therefore, new cost-effective treatments are urgently needed.

A new treatment strategy that aims to regenerate the damaged myocardium and restore myocardial function is now being sought, with stem cell therapy offering much promise. Embryonic stem cells, adult stem cells, and more recently induced pluripotent stem cells all demonstrate the ability to differentiate into cardiac-lineage cells *in vitro* [3-5]. As such, many research teams have focused on utilizing these cells as possible sources for myocardial repair in animal models of MI. The apparent success in a majority of these studies has paved the way for a number of human clinical trials to take place in order to translate these findings into the clinic.

This review therefore discusses the use of stem cell therapy in the treatment of MI and focuses on the pre-clinical and clinical studies that have taken place.

Methodology

Literature was obtained using the electronic database PubMed. Search terms used and number of hits used for in this review is summarized in Table 1.

For inclusion, papers must be primary literature, directly related to MI or cardiomyocyte differentiation, and peer-reviewed. Excluded were non-primary literature, studies related to non-myocardial ischaemia, articles in languages other than English, and articles pre-1998.

Pre-clinical studies

Adult cells: Adult stem cells (ASCs) are multipotent cells that have the ability to self-renew and differentiate into any cell type of a

Search Terms	No of Hits
Embryonic stem cells, myocardial infarction, transplantation	25
Embryonic stem cells, differentiation, cardiomyocytes, myocardial infarction	116
Induced pluripotent stem cells, myocardial infarction, cardiomyocytes	22
Induced pluripotent stem cells, myocardial infarction	38
Induced pluripotent stem cells, cardiomyocytes, cardiac lineage, differentiation	16
Induced pluripotent stem cells, cardiomyocytes	145
Fetal, cardiomyocytes, transplant, myocardial infarction	65
Skeletal myoblasts, cell transplantation, myocardial infarction, myocardial repair	61
Skeletal myoblasts, stem cells, myocardial infarction, humans, clinical trials	42
Bone-marrow-derived, stem cells, myocardial infarction, humans, clinical trials	91

Table 1:

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Received August 26, 2011; **Accepted** November 17, 2011; **Published** November 19, 2011

Citation: Faulkner A, Madeddu P (2011) Stem Cell Therapy: A New Approach for Treatment of Myocardial Infarction. J Stem Cell Res Ther S1:004. doi:10.4172/2157-7633.S1-004

particular organ or system in which they are found. They have been identified in a number of tissues including the bone-marrow, heart, skeletal muscle and adipose tissue. Their use as a cell source for myocardial regeneration has been widely discussed [6-8].

Foetal cardiomyocytes: Foetal cardiomyocytes were one of the first cell types to be studied for cardiomyoplasty after MI, predominantly because they are already destined to become adult cardiac tissue, therefore may integrate well with host tissue and successfully restore myocardial function. There have been mixed results when foetal cardiomyocytes have been transplanted into rodent models. Several studies report survival for up to six weeks post-transplant, improvement in left ventricular (LV) function and reduced mortality rates [9,10]. Additionally, some reports suggest that the donor cardiomyocytes are able to successfully integrate into host myocardium [9,11]. In contrast, a number of studies report low survival rates with limited or no integration and no LV improvement [12,13]. Another study suggests that the time of transplant influences the success rate [14]. They reported that the optimal transplantation time is 2-3 weeks post-infarct, when the inflammatory response has resolved but scar formation is limited [14]. Other issues limiting foetal cardiomyocytes include that most transplanted cells remain immature and structurally unorganised, there is a limited source of cells and, as they are non-autologous, present a possibility for immune rejection.

Skeletal myoblasts: Skeletal myoblasts (SMs) are a potential autologous cell source for regenerating the infarcted myocardium. They develop from skeletal muscle satellite cells and already have contractile functions. Transplantation of SMs into models of MI showed the capacity of these cells of replacing cardiomyocyte loss and restoring a degree of cardiac function [15-17]. Transplanted SMs appear to be short-lived, with a significant number of transplanted cells no longer being detectable after 72 hours post-grafting [18]. Those that survive develop patches of elongated and striated cells that retain skeletal muscle cell characteristics [15], unable to integrate into myocardial tissue, as evidenced by the failure to detect gap junctions between skeletal and myocardial cells [16]. This lack of integration poses the risk of ventricular arrhythmias [19].

Bone marrow-derived stem cells: Cells of the bone-marrow and circulation have received much attention as a therapeutic cell source. They include haematopoietic stem cells (HSCs), endothelial progenitor cells (EPCs), and mesenchymal stem cells (MSCs).

In 2001 Orlic and colleagues reported the first use of $lin^-/c\text{-}kit^+$ murine HSCs in regenerating the infarcted heart [20]. *In vitro* expanded HSCs were delivered via intramyocardial injection 3-5 hours after infarct initiation. They reported the presence of proliferating HSC-derived cardiomyocytes, endothelial cells (ECs) and smooth muscle cells (SMCs) occupying around 68% of the infarcted area 9 days post-transplantation, suggesting the ability of HSCs to trans-differentiate into cardiac and vascular-lineage cells [20]. These HSC-derived myocytes expressed connexin-43, suggesting electrical coupling and a degree of functional recovery was observed [20]. A study published in the same year was in line with Orlic's results in reporting that HSCs migrated to the infarcted myocardium are able to differentiate into cardiomyocytes and ECs thereby contributing to the formation of functional tissue [21]. Additionally, cytokines such as granulocyte-colony-stimulating-factor (G-CSF) and stromal-derived-factor-1α (SDF-1α) have been administered to improve mobilization and recruitment of HSCs to infarcted areas, initiating myocardial repair [22,23]. Many have disputed these results with reports of no trans-differentiation and no functional recovery taking place [24-26], meaning that cell fusion rather than trans-differentiation could be occurring.

EPCs have the potential to regenerate damaged/aged ECs and promote post-natal angiogenesis. The definition of EPC is not homogeneous. Based on antigenic and clonogenic characteristics, three populations, often referred to in the literature, can currently be distinguished: CFU-Hill, also sometimes referred to as CFU-EC, yield adherent colonies consisting of myeloid cells and T-cells. The second population, most commonly used in cardiovascular studies,

is now often referred to as circulating angiogenic cells (CACs) and comprise mainly myeloid cells, who are able to support angiogenesis in a paracrine way. Finally, endothelial colony forming cells (ECFC) give rise to cells not expressing myeloid markers anymore, but exhibiting phenotypic and antigenic characteristics of mature ECs. Recent studies showed that EPCs may have greater potential for cell therapy after an MI compared with other cell types [27,28].

The means for utilizing EPCs for MI therapy is to boost their numbers through the administration of cytokines or through their *in vitro* expansion and re-administration. G-CSF promotes the mobilization of EPCs from the bone-marrow. Many studies have isolated EPCs *in vitro* from G-CSF-mobilized mononuclear cells harvested from the circulation. When these EPCs have been reintroduced via intravenous, intramyocardial or intracoronary transfer into MI models, they appear capable of enhancing neovascularisation, through their differentiation into ECs, leading to improved infarct perfusion [29-31]. As a result, a reduction in cardiomyocyte apoptosis, increased survival of viable myocardium and improvements in myocardial function has been observed [29,30]. Additionally, the use of SDF-1α delivered to the infarct site can further improve EPC homing to the myocardium leading to increased neovascularisation [30,32].

MSCs are also found within the bone-marrow, peripheral and umbilical cord blood, and within tissues including adipose tissue and the adventitia of the vascular wall. They are capable of differentiating into ECs [33] and cardiomyocytes [34], both *in vitro* and *in vivo*, and more efficient in restoring myocardial function after MI compared with HSCs [35]. Our recent work shows that pericyte-like progenitor cells from saphenous vein leftovers of CABG patients improve recovery from myocardial infarction trough stabilization of vascularisation, containment of infarct extension and inhibition of fibrosis, by a mechanism involving the microRNA 132 and its target genes of Ras-GTPase activating protein and methyl-CpG-binding protein 2 (Katare et al., Circ Res in press).

Bone-marrow-derived MSCs have been cultured *ex vivo* and delivered to the infarcted heart *via* intramyocardial injection immediately, or shortly following, MI [36-38]. These cells were able to differentiate into ECs and cardiomyocytes and act as potent activators of angiogenesis through releasing VEGF. This led to increased vessel density in the infarct area and a restoration of myocyte mechanical function and overall LV function, as assessed by improved LV ejection fraction (LVEF) and fractional shortening (FS). Similar results are seen when MSCs are transplanted up to 1 week post-MI induction [39]. MSCs can also be mobilized by G-CSF and migrate to the infarcted heart where they can exert their regenerative effects [40,41]. Cytokines may therefore be useful as adjuvant of systemic MSC delivery [42].

Other sources of MSCs have also been investigated for use in myocardial repair. Adipose tissue is acquiring increasing attention as

an easily obtainable, autologous source of MSCs. When monolayered adipose tissue-derived MSCs were transplanted onto the scarred area 4 weeks post-MI induction, the engrafted sheet grew to form a thick stratum that included blood vessels, undifferentiated cells and a few cardiomyocytes [43]. The MSC sheet also acted in a paracrine fashion, producing VEGF to promote angiogenesis in the infarcted myocardium. This led to reduced wall thinning of the infarcted area and increased myocardial function [43]. More recently, adipose tissue-derived MSCs have been delivered via intramyocardial injection immediately following MI induction in mouse and pig models [44,45]. Both studies report an increase in vessel density, reduced scar size, and a functional improvement as evidenced by increased LVEF and improved FS. These results suggest that adipose-derived MSCs are just as capable of restoring function after MI as bone-marrow-derived MSCs.

Bone-marrow cells may therefore be capable of promoting myocardial repair after MI by contributing to neovascularisation and cardiomyogenesis, thereby limiting myocardial remodelling and preserving overall function.

Cardiac resident stem cells: The heart was long considered a terminally differentiated organ lacking the capacity to replace or repair its cells. In the last 10 years however this concept is slowly changing with the introduction of the potential of a cardiac resident stem/progenitor cell (CSC). Beltrami's study was the first to show that isolated c-kit$^+$/Lin$^-$ CSCs behave as self-renewing, clonogenic and multipotent cells that give rise to cardiomyocytes, ECs and SMCs [46]. It is thought that these CSCs are activated after heart injury and migrate to damaged regions to generate new cardiomyocytes [47]. More recently, it has been demonstrated that the human heart has a significant growth reserve and replaces its myocyte and non-myocyte compartments several times during a life-time [48]. These studies therefore support the idea that CSCs may be capable of repairing the heart after an MI.

Both murine and human CSCs have been cultured and expanded *in vitro* and delivered to the infarcted rodent heart by intravenous, intracoronary and intramyocardial injection [46,49-52]. These studies show that CSCs are capable of homing to the ischaemic-damaged heart where they promote increased vessel density, cardiomyocyte differentiation and reduced myocyte apoptosis at the infarct site. This leads to reduced infarct size and attenuation in cardiac remodelling and dyfunction. CSCs are reportedly capable of differentiating into the same cells *in situ* as can be obtained *in vitro* [51], therefore there is no need to pre-differentiate these cells prior to transplantation, thus reducing their time in culture. Cardiomyocyte differentiation of transplanted CSCs however seem to be quite low suggesting that the beneficial effects on myocardial function are through paracrine effects on neoangiogenesis and endogenous CSC activation/differentiation rather than their self-differentiation.

Most studies have transplanted CSCs immediately following or within a few hours of MI-onset. As it is more clinically desirable to use autologous CSCs, then delivery at MI-onset is not possible as it takes a number of weeks to expand these cells in culture [53]. The transplantation of autologous CSCs into old infarcts or the activation of endogenous CSCs, without the need for cell transplantation, need to be investigated. Tang delivered CSCs via intracoronary infusion one month after MI-induction [53]. After 35 days, more viable myocardium and reduced fibrosis was observed in non-infarcted regions and improved LV function was reported. Once again there was limited CSC differentiation suggesting a beneficial paracrine effect.

Endogenous CSCs express a number of growth-factor receptors and when their ligands are introduced there is significantly increased endogenous CSC proliferation and migration towards infarcted regions [54-56]. Once there, CSCs are able to differentiate into cardiomyocytes, replacing around 40% of the scar [53] and promote neoangiogenesis. These changes have led to increased myocardial performance and increased animal survival [54], suggesting that stimulating endogenous CSC activation/mobilization may be another treatment option for myocardial repair.

Embryonic stem cells

Embryonic stem cells (ESCs) are pluripotent cells derived from the inner-cell-mass of the blastocyst [57]. Their pluripotent nature means they can differentiate into any cell type of the three germ layers that make up the body, including cardiomyocytes [57]. This characteristic makes ESCs an attractive cell source for cell transplantation therapies for many diseases, including that of myocardial repair.

Murine ESCs have been demonstrated to survive, migrate and proliferate when injected into infarcted rodent myocardium up to one hour post-MI [58-60]. Indeed, these studies also demonstrated the potential of ESCs to provide some, albeit small, restoration of global LV function up to eight weeks post-transplant. One of the concerns to arise from these studies however was that the degree of cardiomyocyte differentiation was extremely low, with reports as low as 0.5% of the total number of cells delivered differentiating towards a cardiomyocyte lineage [59]. An additional issue was the high incidence of teratoma formation with reports of 100% of rats receiving mESCs developing teratomas [59]. These limitations are not confined to murine cells. It is also the case in rodent models receiving human ESCs (hESCs) [3]. When hESCs were injected into both healthy and infarcted myocardium, few cardiomyocytes were detected and teratomas had formed in approximately 50% of rats.

Two key issues have been highlighted that need addressing if ESCs are ever to be used in clinical practise, namely the extremely low rate of cardiomyocyte differentiation and the high incidence rate of teratomas. It is known that the risk of teratoma formation is reduced as cells become more specialised so in theory, if the rate of cardiomyocyte differentiation can be significantly increased, teratoma risk should reduce. Indeed, many research groups are now pre-differentiating ESCs into NKx2-5$^+$ cardiac-lineage progenitors (CPCs) or early cardiomyocytes *in vitro*, through methods such as embryoid-body formation, before transplanting into MI models [3,61]. The incidence of teratomas in these studies is considerably reduced with many reporting a complete absence of tumours [62,63]. When teratomas are present they are on average significantly lesser in volume compared with undifferentiated ESCs [60] and usually occur due to inefficient differentiation protocols, leading to the presence of undifferentiated cells within the transplant media. The majority of studies using mouse and human ESC-derived CPCs or cardiomyocytes have reported at least an attenuation of LV decline in the short term (4-8 weeks) through reducing the occurrence of LV remodelling and improving myocardial contractility [3,61,63,64]. Again, ESC-derived CPCs can survive the infarct environment and differentiate into immature cardiomyocytes expressing α-actinin, cardiac MHC and troponin-I around the infarct boarder-zones [61,64]. There is a possibility that these cells continue, at least for a short time, to proliferate as detected by Ki67 expression [3,62]. These cardiomyocytes are however low in number, small and immature but seem to be able to form gap-junctions between themselves [63]. Whether or not they can form connections

with host cardiomyocytes is still an area of controversy [65,66]. ESC-derived ECs have also been detected in transplanted hearts [61,67], suggesting a possible angiogenic response that may also contribute to the attenuation in LV decline.

ESCs show promise as a cell source for transplantation therapy after MI. They are capable of differentiating into cardiomyocytes *ex vivo* that can successfully survive within the infarct area and promote a degree of myocardial functional recovery in MI-models [3,61,64]. They do however come with a few, but key, limitations. The requirement of animal products for ESC culture poses a risk of zoonotic transfer. This can easily be overcome with the development of xeno-free media. Additionally, the low number of cells that can be produced and the inability to control teratoma risk makes them unsuitable for human transplantation therapy at present. Aside from the practical limitations they are surrounded by ethical controversy, mainly concerning the destruction of human embryos in order to harvest such cells. With the availability of ASCs and the discovery of being able to produce induced pluripotent stem cells, whether or not ESCs become part of clinical therapy in the future is far from certain.

Induced pluripotent stem cells

In 2006 Yamanaka and colleagues successfully reprogrammed murine somatic cells into a pluripotent ESC-like state by retroviral delivery of the transcription-factors OCT-4, SOX-2, Klf-4 and c-Myc [68]. These new, so called induced pluripotent stem cells (iPS cells), had a similar morphology and up-regulation of gene expression as ESCs, and also gave rise to teratomas consisting of cells of all three germ layers when injected into nude mice, demonstrating their pluripotency.

In 2007 two independent groups showed that iPS cells were able to be produced from human somatic cells [69,70]. These breakthrough experiments opened up the possibility of a new type of cell that can differentiate into any cell type of the body and can be generated from the patient's own somatic cells, making patient-specific treatment more feasible.

iPS cells can form embryoid-bodies with spontaneously contracting clusters [5] in the same way as ESCs. These contracting clusters have been shown to occur more readily with iPS cells than with ESCs, express cardiac mesoderm and cardiomyocyte markers and demonstrate electrophysiological properties [5]. iPS cell-derived cardiomyocytes display a similar genetic profile to ESC-derived cardiomyocytes [71,72] and demonstrate similar contractile properties, although they are still significantly different in comparison to ventricular tissue of comparable age [73], demonstrating their immaturity.

iPS cells have been assessed as a potential cell source for the treatment of acute MI. Spontaneously contracting embryoid-bodies were produced by the hanging-drop culture method and injected into the myocardium of mouse models 30 minutes after LAD ligation [74]. Over the four week follow-up, mice displayed increased LVEF and FS and a normalisation of systolic wall motion. Sinus rhythm was also maintained throughout with no ventricular tachycardia (VT) or ectopy. These results demonstrated a restoration of contractile performance, LV wall thickness and electrical stability [74]. As with ESCs, iPS cells pose a risk of teratoma formation due to their pluripotent nature. However, this study reported no teratoma formation in immunocompetent mice but did report tumour growth in immunosuppressed mice after subcutaneous injection of iPS cells [74]. Another study transplanted iPS cell-derived cardiomyocytes into acutely infarcted hearts of NOD-SCID mice. Cardiomyocytes were able to survive, showed no marker of pluripotency and did not form tumours [71] suggesting that like ESCs, the more differentiated the cells the less tumour risk they pose.

Research has become focused on boosting iPS-derived cardiomyocyte cell number through more efficient culture and differentiation protocols. A study has recently reprogrammed somatic cells into iPS cells without the use of c-Myc, promoting cardiac mesoderm lineage differentiation [75]. The authors report that c-Myc-dependent reprogramming produces progeny that consistently prolong the expression of Oct-4 and FGF-4 genes whilst suppressing cardiac differentiation. C-Myc-independent reprogramming however led to the up-regulation of pre-cardiac (CXCR4, Flk-1 and Mesp 1/2) and cardiac (NKx2.5, Mef2c and myocardin) gene expression. This progeny was reported to undergo early and robust cardiogenesis during *in vitro* differentiation [75]. Research teams are looking at improving differentiation protocols to generate higher cardiomyocyte cell numbers [76-78]. Key safety issues with iPS cells are the requirement for integrating viral-vectors for genetic reprogramming of somatic cells and the use of animal products in the culture media. A xeno-free culture medium has recently been described that can support human ESCs and iPS cells for prolonged periods of time whilst maintaining their pluripotent characteristics, allowing for the production of clinical-grade cells [79]. In recent years non-integrating viral-vectors [80], drug-inducible transgenic systems [81], transgene removal systems including the Cre-Lox system, non-viral methods [82,83], and even direct reprogramming [84] (removing the need for iPS cell stage) are being developed, however the reprogramming efficiency is low.

iPS cells represent an opportunity for providing patient-specific treatment after MI. iPS-derived cardiac-lineage cells may have potential in restoring myocardial function in a rodent model [74] however the small number of cardiomyocytes being produced with current differentiation protocols needs to be addressed. Efficient cardiomyocyte differentiation methods are essential for developing cardiac cells for regenerative medicine although recently, methods for direct reprogramming [84] may remove the need for acquiring an induced pluripotent phenotype altogether.

Clinical trials

ESCs and potentially iPS cells offer the most ideal cell type for myocardial regeneration due to their pluripotent nature but obvious practical, ethical and political issues currently prevent their progress as a clinical tool. Of the adult cells, SMs, bone-marrow-derived stem/progenitor cells, and possibly in the future, cardiac-resident stem/progenitor cells offer the most realistic means of cell-based cardiac regeneration. As such, clinical trials have focused on utilizing autologous SMs and bone-marrow cells.

Skeletal myoblasts: The first use of SMs being transplanted into a human patient was reported in 2001 [85]. The patient received injections of SMs in and around the non-viable areas of myocardium immediately following CABG surgery. After 5 months, the patients' clinical status improved from New York Heart Association (NYHA) class III to II and the LVEF increased from 21% to 30%. Additionally, myocardial contractility improved and no arrhythmias were detected on holter electrocardiogram analysis [85]. These results paved the way for a number of other phase-1 safety and feasibility trials to take place (Table 2).

These phase-1 trials mainly involved a small number of patients that were planned to undergo revascularization surgery. Each trial varied with regards to the number of cells being transplanted (Table

Study Reference	N° of Patients	N° Cells	Undergoing CABG	Delivery Route	Outcomes
[85,89]	1	$800x10^6$	Yes	IM	NYHA class, LVEF & tissue viability improved. No arrhythmias. Graft survived up to 1.5 years. Cells had skeletal muscle phenotype & aligned in parallel with cardiomyocytes. No connections formed.
[90,92]	8	$871x10^6$	Yes	IM	LVEF, tissue viability & contractility, & NYHA score improved. 5 patients developed VT.
[87]	4	$300x10^6$	No	IM	<1% myoblast survival, no inflammation, increased vessel density, cells aligned in parallel with myocardium. 4 patients developed arrhythmias.
[89]	11	$221x10^6$	Yes	IM	LVEF, tissue viability & contractility, & NYHA score improved. 1 patient developed VT.
[91]	9	$4x10^5$-$5x10^7$	Yes	IM	LVEF & contractility improved. VT observed in first 2 patients (7 patients received prophylaxis).
[88]	9	$1x10^8$	No	Percutaneous transcoronary-venous	Limited LVEF improvement. NYHA score improved. VT developed in 1 patient not receiving prophylaxis
[93] (randomized controlled)	n=12 SMs; n=11 controls	$30x10^6$-$600x10^6$	No	Endovascular transcatheter	1 patient in control and 2 patients in treatment group developed VT. NYHA score improved. No effect on myocardial viability or function.

SMs skeletal myoblasts; CABG coronary artery bypass graft; IM intramyocardial; NYHA New York heart association; LVEF left ventricular ejection fraction; VT ventricular tachycardia

Table-2: Key outcomes of phase-1 safety and feasibility trials with skeletal myoblasts.

2) and the length of follow-up (6-52 months). All trials report the potential of generating millions of cells from a small skeletal muscle biopsy with a myoblast purity consistently >60% and cell viability >90% [86-88] suggesting that it is feasible to generate large quantities of autologous cells for transplantation therapy. The study by Pagani [87] allowed for the analysis of graft success and provides an insight into how myoblasts behave in the human heart. Animal studies suggest that myoblasts are short-lived, maintain their skeletal phenotype and are unable to form connections with host cardiomyocytes [15,16,18]. Pagani reported similar findings, with less than 1% of the $300x10^6$ cells transplanted surviving and those that do survive maintain a skeletal phenotype [87]. They do however demonstrate that the majority of surviving myoblasts align themselves in parallel with host myocardial fibres, suggesting the potential for structural enhancement. Further to this, a post-mortem analysis in 2003 of the first patient to receive a myoblast transplant in 2001 revealed that the transplanted cells were unable to form connections with the myocardium but did display a more predominant expression of the slow, rather than the fast, myosin heavy-chain isoform that may allow them to sustain a cardiac workload [89].

Although not designed for looking at the efficacy of myoblast therapy a number of trials reported a significant beneficial effect on LV function, as assessed by a significant increase in LVEF, and an improvement in tissue viability and contractility within the infarct area after intramyocardial delivery [86,90,91]. For example, Menasche reported an increase in LVEF from 23.8% to 32.1% ($P<0.02$) and 62% of the tissue segments injected with myoblasts, associated with improved viability and contractility in six of the eight patients within the first 12 months [90]. These functional improvements translated to a positive impact in NYHA score ($P<0.0001$). These factors remained stable for up to 52 months post-transplant [92].

Siminiak [88] and Dib [93] have conducted trials using alternative methods of cell delivery, namely percutaneous trans-coronary-venous and endovascular trans-catheter injection respectively. Through these methods there was limited functional benefit but there was an improvement in NYHA score. The trial by Dib and colleagues was particularly interesting as it is the first randomized, controlled phase-1 trial to have taken place [93]. Additionally, these trials were the first which did not involve patients undergoing revascularization by CABG. Therefore the improvements in tissue viability and function in the previous uncontrolled studies could have been predominantly due to revascularization from the CABG rather than the myoblast transplantation. However it is important to note that many of the revascularization procedures were performed in areas distant from the transplant area.

At present only one randomized, placebo-controlled, double-blind phase-2 trial has taken place. The MAGIC trial [94] incorporated 121 patients with an indication for CABG. Immediately following CABG, myoblasts or placebo solution were injected into non-viable, akinetic myocardial segments. All patients received an internal defibrillator and started on amiodarone for the first 3 months post-transplant. At 6 months, tissue viability and global myocardial function was not significantly different between those receiving myoblasts and those receiving placebo [94] suggesting that myoblast transplantation is not related to any significant benefit in the infarcted heart.

One safety issue that has emerged from these trials is the incidence of ventricular arrhythmia. Most studies report a number of patients developing either sustained or non-sustained VT predominantly within the initial post-operative period [87,90,91,93]. Some studies therefore had to adapt the protocol to encompass prophylaxis for the remaining patients on the trials [91]. It is important to consider however that the patients enrolled in these studies are more prone to

developing arrhythmias. Indeed, the phase-1 randomized trial [93] and phase-2 MAGIC trial [94] report no significant difference between control or cell transplantation groups in VT incidence, however this area still remains a cause for concern.

Bone-marrow-derived stem cells

The success of bone-marrow cells to restore a degree of functional recovery in animal models of MI has been followed by a strong concentration of clinical trials. As with pre-clinical studies, the majority of trials have taken the form of collecting and re-introducing bone-marrow mononuclear cells (BMNCs) into the heart, or the mobilization of endogenous BMNCs by lone G-CSF therapy.

G-CSF is a non-specific stimulant of cell mobilization from the bone-marrow. In animal models it has successfully been used to stimulate bone-marrow stem/progenitor cell mobilization into the circulation [22,40]. These cells appear to be capable of restoring function by differentiating into cardiomyocytes and/or stimulating neoangiogenesis [29,34].

In clinical studies to date, subcutaneous G-CSF administration has successfully led to significant increases in circulating $CD34^+$ cells and leukocytes from the patient's bone-marrow with only occasional mild side-effects [95-99] (Table 3). An early phase-1, non-randomized, trial incorporating 23 patients reported a significant improvement in wall motion score (WMS) and myocardial perfusion in the treatment group verses controls at 3 months ($P<0.0001$ for both) [100]. Additionally, there was a trend towards increased LVEF in the treatment group, although this did not reach significance [100]. Similar results were reported in the larger FIRSTLINE-AMI randomized, controlled trial involving 50 patients. LVEF, WMS, and tissue viability all significantly improved in the treatment group after 6 months [96]. These results suggest that $CD34^+$ cell mobilization by G-CSF administration could promote a degree of functional recovery after MI in humans. These trials however are small in number and could not evaluate the efficacy of the therapy, but only determine the safety and feasibility of mobilizing BMNCs with G-CSF in patients after an MI. These positive results therefore should be viewed with caution as many other small trials report no clinical benefit [101,102].

A number of randomized, placebo-controlled phase-2 trials have reported no significant benefit of lone G-CSF treatment. The REVIVAL-2 trial reported no significant improvement in LVEF and no significant reduction in overall infarct size in the treatment group between baseline and 4-6 months follow-up [98]. The double-blind, randomized, placebo-controlled G-CSF-STEMI [102] and STEMMI [99] trials report similar results (Table 3), suggesting that BMNC mobilization alone is not enough to evoke a clinically significant recovery in humans. Further larger, randomized, double-blind, placebo-controlled trials are therefore required.

Many more trials have centred on re-introducing autologous BMNCs through direct intramyocardial/intracoronary transfer. Although more invasive, these methods allow for the full complement of cells to reach the damaged myocardium as compared with the wide-spread, systemic mobilization gained through G-CSF alone.

Most studies have focused on acute myocardial infarction (AMI) and vary with regards to BMNC numbers, patient baseline characteristics and length of follow-up. Many trials demonstrate safety and feasibility of BMNC transplantation by intracoronary or intramyocardial transfer [103-108], however the clinical and functional improvement is far from clear with mixed results being reported [103-105,107-109].

The randomized, controlled TOPCARE-AMI trial reports that intracoronary infusion of either BMNCs or blood-derived progenitors 4 days post-AMI leads to significant improvements in global LVEF and WMS at the infarct boarder-zone from baseline to 4 months follow-up, a response that significantly differed from the control group [106].

Study Reference	Design	No of Patients	G-CSF Dose (μg/kg/day)	Treatment Initiated	Follow-Up	Outcomes
[95] (MAGIC Trial)	Phase-2, Randomised, Controlled	n=7 BMNC; n=3 GCSF only; n=1 control	10 for 4 days	4 days prior to PCI	6 months.	GCSF treatment is safe. Increased $CD34^+$ cell mobilization. GCSF alone has no functional benefit. BMNC transplantation significantly increased EF & ESV. Trial stopped due to significant restenosis rate.
[100]	Non-randomised, Open-labelled	n=14 GCSF; n=9 control	10 for 7 days	48 hours post-PCI	3 and 12 months.	GCSF is safe. Increased $CD34^+$ cells/leukocytes. Trend towards increased EF and significant increase in WMS & perfusion vs. control.
[96] (FIRSTLINE-AMI trial)	Randomised, Controlled	n=25 GCSF; n=25 control	10 for 6 days	90 minutes post-PCI	6 months.	GCSF is safe. Increased $CD34^+$ cell mobilization. EF, WMS & viability significantly improved vs. control. 4 & 5 patients in GCSF & control had restenosis, respectively.
[101]	Randomised, placebo-controlled	n=8 GCSF; n=8 control	5 for 4 days	37 hours post-PCI	6 months.	GCSF is safe. Increased mobilization of $CD34^+$ cells. No significant benefit with lone GCSF.
[102] (G-CSF-STEMI Trial)	Phase-2, Randomised, double-blind, placebo- controlled	n=19 GCSF; n=21 control	10 for 5 days	32 hours post-PCI	3 and 6 months.	GCSF is safe. Increased $CD34^+$ cells. No functional benefit with lone GCSF. No difference if restenosis rate vs. control.
[98] (REVIVAL-2 Trial)	Randomised, Placebo-controlled	n=56 GCSF; n=58 control	10 for 5 days	4-5 days post-PCI	4-6 months. Angiography at	GCSF is safe. Increased $CD34^+$ cells. No functional benefit with lone GCSF. No difference in restenosis rate.
[99] (STEMMI Trial)	Phase-2, randomised, double-blind, placebo-controlled	n=33 GCSF; n=37 control	10 for 6 days	10-60 hours post-PCI	6 months.	GCSF is safe. Increased $CD34^+$ cells and MSCs. No functional benefit with lone GCSF. No increase in restenosis rate with GCSF.

GCSF granulocyte-colony-stimulating-factor; EF ejection fraction; WMS wall motion score; PCI percutaneous coronary intervention; BMNC bone-marrow mononuclear cells; MSC mesenchymal stem cell

Table 3: Summary of trials utilizing G-CSF to mobilize bone-marrow cells for treatment of MI.

There was also a significant improvement in myocardial viability within the infarct area and a normalization of coronary-flow-reserve (CFR) in the infarct-related-artery in patients without restenosis. These initial reports were based on the first 20 patients enrolled, although when 59 patients were followed-up to 1 year there was also a significant improvement in LVEF and infarct size compared with the controls [106]. The results of this small trial suggest that BMNCs have the potential to significantly improve vessel and myocardial function after AMI.

The BOOST randomized, controlled trial support the observation that BMNCs can significantly improve LVEF and WMS up to 6 months [105]. When followed up to 18 months and 5 years however, the initial increase in LVEF could no longer be detected [109], suggesting that any functional benefit resulting from BMNC transplantation is short-lived.

The larger, randomized, double-blind, placebo-controlled REPAIR-AMI trial involving 202 patients found that global LVEF significantly increased at 4 months versus baseline after intracoronary infusion of either BMNCs or placebo [101]. The absolute change in LVEF however was significantly greater in the BMNC group versus the control (P=0.01). The same pattern was found with regards to myocardial contractility but no significant changes were observed in end-systolic volume (ESV) or end-diastolic volume (EDV) in either the control or treated groups at 4 months [103]. A small sub-study associated with the REPAIR-AMI trial also suggests that BMNCs can normalize CFR in the infarct-related-artery and reduce microvascular resistance, in agreement with the TOPCARE-AMI trial, leading the authors to believe that BMNCs may promote vascular repair [110].

The REGENT trial, another large randomized controlled trial, also reported a significant increase in LVEF in the BMNC group however found no significant difference in absolute change in LVEF between any of the treatment groups or controls at 6 months [111] which is in contrast to the previous trials.

An interesting find from both the REPAIR-AMI [103,104] and REGENT [111] trials is that there appears to be significantly greater improvement in LV function with cell therapy in patients with worse baseline LVEF scores. The REPAIR-AMI trial also report a progressive increase in BMNC-associated recovery of contractile function as the interval between reperfusion therapy and time of BMNC infusion increases (P=0.01), with the greatest results seen in those treated >4 days after infarct reperfusion [103,104]. The REGENT trial however found no such association [111].

No major adverse cardiac events (MACE) have been reported with intracoronary infusion in any of the trials discussed, suggesting that intracoronary infusion is safe. In fact, the REPAIR-AMI trial reported that the number of deaths, recurrent MI and revascularization was significantly lower in the treatment group versus control [103,104]. Additionally the combination of deaths, recurrent MI and hospitalisation for chronic heart failure (CHF) occurred less frequently in the treatment group.

Large, randomized, placebo-controlled, double-blind, multicentre trials are therefore warranted in order to clarify the functional and clinical effects of BMNCs after AMI. A trial of this kind has been awarded by the EU-FP7 and is going to recruit 3,000 patients in 17 clinical centres across Europe (the BAMI consortium). (Table 4)

Only a small number of trials have involved patients with CHF as a result of MI (Table 5). The largest trial to date is the STAR-heart study which enrolled 391 patients [112]. BMNCs were delivered via intracoronary infusion into the infarct-related-artery of each patient in the treatment group and followed-up for 5 years. No adverse events were associated with the procedure and at 3 months the treatment group displayed a significant increase in LV function and contractility, leading to a considerable improvement in exercise performance and NYHA score [112]. These improvements appear to be long-standing with the beneficial effects lasting up to five years, whereas LV function in the control group steadily declined. A more clinically important

Study Reference	Study Design	No of Patients	Cell Dose	Procedure Time	Follow-Up	Outcomes
[106,119] (TOPCARE-AMI Trial)	Randomized, controlled	n=29 BMNC; n=30 CPC; n=11 Control	7.35±7.31x10^6 $CD34^+/CD45^+$ Cells	~4 days post-AMI	4 months & 1 year	Cell therapy significantly improved LVEF and WMS at border-zone. No significant difference between cell therapy groups. CFR normalized and viability in infarct-zone significantly increased with cell therapy. At 1 year LVEF and infarct size significantly improved with cell therapy.
[105,109] (BOOST Trial)	Randomised controlled	n=30 BMNC; n=30 Control	24.6x10^8 nucleated cells, 9.5x10^6 $CD34^+$cells, 3.6x10^6 haemopoietic colony-forming cells	~5 days post-AMI	6 Months and 5 years	LVEF and WMS at border-zone significantly improved in BMNC group vs. controls at 6 months. EDV, ESV, LV mass index and myocardial injury did not significantly differ from control. No significant difference in MACEs or restenosis at 5 years. No significant improvement in LVEF at 5 years.
[103,104,110] (REPAIR-AMI Trial)	Randomized, double-blind, placebo-controlled, multicentred	n=101 BMNC; n=101 Control	236±174x10^6	3-7 days post-PCI	4 months & 1 year	Significant improvement in LVEF and regional contractility in control and BMNC groups. Absolute change in LVEF significantly greater in BMNC vs. control. Inverse relationship between baseline and absolute change in LVEF in treatment group. Trend for increased contractile function in patients treated >4 days post-PCI. CFR normalized and vascular resistance decreased in treatment group. No MACEs at 1 year.
[111] (REGENT Trial)	Randomised controlled, Multicentered	n=80 $CD34^+$/ $CXCR4^+$ cells; n=80 BMNC; n=40 control	1.90x10^6 $CD34^+$/ $CXCR4^+$ cells; 1.78x10^8 BMNC	7 days post-PCI	6 Months	Procedure is safe. No significant difference in absolute change in LVEF or EDV and ESV between groups. Inverse correlation between baseline LVEF and its change after cell therapy in patients treated with $CD34^+$/ $CXCR4^+$ cells but not with BMNCs. Significant improvement in patients receiving any cell therapy when baseline LVEF was <median. No significant difference in MACEs or restenosis between groups.

AMI acute myocardial infarction; WMS wall motion score; LVEF left ventricular ejection fraction; CFR coronary-flow-reserve; EDV end diastolic volume; ESV end systolic volume; MACE major adverse cardiac events, PCI percutaneous coronary intervention; BMNC bone-marrow mononuclear cells

Table 4: Summary table of key trials utilizing direct transplantation of BMNCs in AMI.

Study	Study Design	No of Patients	Cell Dose	Procedure Time	Follow-Up	Outcomes
[120] (TOPCARE-CHD Trial)	Randomized, controlled	n=28 BMNC; n=24 CirPC; n=23 Control	$22x10^6 \pm 11x10^6$ CPCs; $205x10^6 \pm 110x10^6$ BMNCs	≥3 months post-MI	3 months	No MACE from procedure. LVEF significantly improved in BMNC group only. No other functional benefit in either groups. Significant improvement in NYHA score in BMNC group only.
[112] (STAR-heart Trial)	Controlled	n=191 BMNC; n=200 Control	$6.6\pm3.3x10^7$	8.5±3.2 years post-MI	5 years	No MACE. Cell therapy significantly increased exercise capacity. Significant increase in LVEF & contractility with reduced infarct size in cell group. NYHA score significantly improved in cell group. Improvements maintained up to 5 years. Mortality rates significantly reduced in cell group at 5 years and significantly differed from controls.
[113] (FOCUS-HF Trial)	Randomized, blinded, controlled	n=20 BMNC; n=10 Control	2 million	Not Stated	6 Months	No MACE. Cell therapy had no significant effect on myocardial function. Downward trend in infarct size with cell therapy. QOL and CCS, but not NYHA scores significantly improved with cell therapy. BMNC dysfunction with presence of CHF and increased age (60 years).

BMNC bone-marrow mononuclear cells; CHF chronic heart failure; CirPC circulatory-derived progenitor cell; MI myocardial infarction; MACE major adverse cardiac event; LVEF left ventricular ejection fraction; NYHA New York heart association; QOL quality of life; CCS Canadian cardiovascular society

Table 5: Summary table of key trials utilizing direct transplantation of BMNCs in CHF.

finding was the mortality rates in the treatment group significantly reduced and was significantly less than the control group at 5 years (0.75% per year in the treatment group vs. 3.68% for controls, P<0.01) [110].

Recently however, the randomized, blinded, controlled FOCUS-CHD study involving 20 patients receiving BMNCs via transendocardial injections reported no significant difference in LV function with cell therapy at 6 months [113]. Despite this, there were improvements in Canadian Cardiovascular Society (CCS) score, quality of life scores, and a trend towards an increase in myocardial viability and reduced infarct size. An additional aspect of this study looked at the characteristics of patients BM-cells. They found that this patient population showed reduced progenitor cell activity and patients <60 years had significantly increased MSCs (P=0.04) with greater proliferative capacity verses those >60 years [113], suggesting that the presence of chronic infarcts and increasing age leads to BM-cell dysfunction. This may explain the lack of any significant functional benefit in these patients.

A main safety concern with BMNCs is the potential contribution of these cells to atherosclerotic lesions. Indeed, the MAGIC trial was suspended early on due to the unacceptably high occurrence of restenosis in the infarct-related-artery of the two treatment groups [95], although cell therapy was initiated prior to PCI rather than after (Table 3). The majority of other trials however report no significant difference in restenosis rate between cell treatment groups and controls. A recent study looking for evidence of accelerated atherosclerosis with BMNC therapy has also reported no significant difference in maximal stenosis area and plaque volume from baseline to 9 months in BMNC-treated patients or in minimum lumen diameter and percentage of stenosis in the infarct-related-artery in those treated with BMNCs versus controls [114].

As there are a number of different cell types within the BMNC preparations, a number of teams are now isolating and expanding specific stem/progenitor cells, such as antigenically purified or functionally enriched HSCs or MSCs, before reintroducing them to the damaged heart [115,116]. Many of these trials are reporting that this therapy is safe and feasible, with the potential of having a beneficial functional effect in patients with both AMI and CHF [115-118] and therefore may be a safer and more beneficial option over BMNC transplantation. As pre-clinical studies suggest that MSCs, EPCs and HSCs all have therapeutic potential it is important that the most appropriate cell for promoting myocardial repair is identified and expanded to satisfactory numbers. At present, most appealing are MSCs as they can be isolated from easily-obtainable tissues, such as adipose tissue, and have the potential to differentiate into cardiomyocytes and promote neoangiogeneis in the infracted heart.

Pathways to go

The number of patients who underwent cell therapies to date is still in the order of thousands. Thus, although researchers have been investigating adult SCs since the 1940's and bone marrow transplants were introduced 50 years ago, the concept of cell therapy remains new. Since science cannot establish if any type of cell is therapeutically better than another one, research on all kinds of cell therapy should continue. In fact, this is a field where translational innovativeness overwhelms the common concept of scientific originality. Too many breakthroughs discoveries are dropped after publication with no further development toward clinical application. Intellectual property is certainly an issue to consider when making a strategic plan to develop those seminal results. While this is especially applicable to conventional medical products and in some case allogeneic cell therapy, autologous cell therapies offer less scope for IP coverage (since a patient's own cells cannot be patented). In the latter case, it is recommended to embed the exploitation plans in a national health service-based model to allow timely delivery to the patient. Public awareness and appreciation of cell therapy is high, although the support of lay persons varies according to societal, economic and ethical backgrounds across Europe. Confidence by scientists is also remarkable although most believe that the clear demonstration of therapeutic efficacy is still missing. Finally, opportunities for industry are soaring. Cell therapy alone had global sales of $410 million in 2008 and this is predicted to grow to $5.1 billion by 2014. Revenues might grow even faster with integration of cell products into current therapeutic programmes.

Conclusions

Stem cell therapy offers an opportunity to replace/regenerate the damaged myocardium and promote a significant functional recovery after MI, therefore preventing the decline into heart failure.

ESCs and iPS cells offer a potential pluripotent cell source for transplantation therapy after an MI for the future. They are capable of differentiating into cardiac-lineage cells *in vitro* which upon transplantation into MI models, can contribute to the restoration of

myocardial function through possible angiogenic and cardiomyogenic processes. They are however hampered by the low rate of cardiomyocyte differentiation, the potential for tumour formation, and in the case of ESCs the ethical controversy surrounding their use. These issues, among others, are currently being targeted by many research groups in an attempt to progress these cells into a realistic option for cell therapy.

ASCs, particularly bone-marrow-derived stem/progenitor cells, offer the most realistic source for cell therapy after MI in the near future. Many pre-clinical studies using cells, such as MSCs, demonstrate a potential angiogenic effect in the infarcted heart and possibly the ability to differentiate into cardiomyocytes and ECs. These effects have translated into a degree of functional recovery in animal models and as such a strong concentration of clinical trials utilizing autologous BMNCs has followed. Unfortunately these benefits observed in the pre-clinical studies have not translated into the clinical trials with mixed results being reported. This may be due to wide variation in study design and it is clear that larger, randomized, placebo-controlled trials are indicated. A consensus in terms of the ideal cell dose, time and method of delivery is also mandatory.

References

1. Scarborough P,Bhatnagar P, Wickramasinghe K, Smolina K, Mitchell C (2010) Coronary heart disease statistics 2010. In British Heart Foundation heart statistics publications. British Heart Foundation.
2. Velagaleti RS, Pencina MJ, Murabito JM, Wang TJ, Parikh NI, et al. (2008) Long-term trends in the incidence of heart failure after myocardial infarction. In Circulation 118: 2057-2062.
3. Caspi O, Huber I, Kehat I, Habib M, Arbel GA, et al. (2007) Transplantation of human embryonic stem cell-derived cardiomyocytes improves myocardial performance in infarcted rat hearts. In J Am Coll Cardiol 50: 1884-1893.
4. Xu W, Zhang X, Qian H, Zhu W, Sun X, et al. (2004) Mesenchymal stem cells from adult human bone marrow differentiate into a cardiomyocyte phenotype in vitro. In Exp Biol Med (Maywood) 229: 623-631.
5. So KH, Han YJ, Park HY, Kim JG, Sung DJ, et al. (2010) Generation of functional cardiomyocytes from mouse induced pluripotent stem cells. In Int J Cardiol.
6. Choi YH, Kurtz A, Stamm C (2011) Mesenchymal stem cells for cardiac cell therapy. Hum Gene Ther 22**:** 3-17.
7. Bollini S, Smart N, Riley PR (2011) Resident cardiac progenitor cells: at the heart of regeneration. In J Mol Cell Cardiol 50: 296-303.
8. Dimmeler S, Burchfield J, Zeiher A (2008) Cell-based therapy of myocardial infarction. Arteriosclerosis, thrombosis and vascular biology 28**:** 208-216.
9. Roell W, Lu ZJ, Bloch W, Siedner S, Tiemann K, et al. (2002) Cellular cardiomyoplasty improves survival after myocardial injury. Circulation 105**:** 2435-2441.
10. Skobel E, Schuh A, Schwarz ER, Liehn EA, Franke A, et al. (2004) Transplantation of fetal cardiomyocytes into infarcted rat hearts results in long-term functional improvement. Tissue Eng 10: 849-864.
11. Rubart M, Pasumarthi KB, Nakajima H, Soonpaa MH, Nakajima HO, et al. (2003) Physiological coupling of donor and host cardiomyocytes after cellular transplantation. Circ Res 92: 1217-1224.
12. Sakakibara Y, Tambara K, Lu F, Nishina T, Nagaya N, et al. (2002) Cardiomyocyte transplantation does not reverse cardiac remodeling in rats with chronic myocardial infarction. Ann Thorac Surg 74: 25-30.
13. Etzion S, Battler A, Barbash IM, Cagnano E, Zarin P, et al. (2001) Influence of embryonic cardiomyocyte transplantation on the progression of heart failure in a rat model of extensive myocardial infarction. In J Mol Cell Cardiol: 1321-1330.
14. Li RK, Mickle DA, Weisel RD, Rao V, Jia ZQ (2001) Optimal time for cardiomyocyte transplantation to maximize myocardial function after left ventricular injury. Ann Thorac Surg 72: 1957-1963.
15. Taylor DA, Atkins BZ, Hungspreugs P, Jones TR, Reedy MC, et al. (1998) Regenerating functional myocardium: improved performance after skeletal myoblast transplantation. Nat Med 4: 929-933.
16. Reinecke H, Murry CE (2000) Transmural replacement of myocardium after skeletal myoblast grafting into the heart. Too much of a good thing? In Cardiovasc Pathol 9: 337-344.
17. Horackova M, Arora R, Chen R, Armour JA, Cattini PA, et al. (2004) Cell transplantation for treatment of acute myocardial infarction: unique capacity for repair by skeletal muscle satellite cells. Am J Physiol Heart Circ Physiol 287: H1599-1608.
18. Suzuki K, Murtuza B, Beauchamp JR, Smolenski RT, Varela-Carver A, et al. (2004) Dynamics and mediators of acute graft attrition after myoblast transplantation to the heart. FASEB J 18: 1153-1155.
19. Fernandes S, Amirault JC, Lande G, Nguyen JM, Forest V, et al. (2006) Autologous myoblast transplantation after myocardial infarction increases the inducibility of ventricular arrhythmias. Cardiovasc Res 69: 348-358.
20. Orlic D, Kajstura J, Chimenti S, Jakoniuk I, Anderson SM, et al. (2001) Bone marrow cells regenerate infarcted myocardium. Nature 410: 701-775.
21. Jackson KA, Majka SM, Wang H, Pocius J, Hartley CJ, et al. (2001) Regeneration of ischemic cardiac muscle and vascular endothelium by adult stem cells. J Clin Invest 107: 1395-1402.
22. Tang YL, Qian K, Zhang YC, Shen L, Phillips MI (2005) Mobilizing of haematopoietic stem cells to ischemic myocardium by plasmid mediated stromal-cell-derived factor-1alpha (SDF-1alpha) treatment. Regul Pept 125: 1-8.
23. Fujita J, Mori M, Kawada H, Ieda Y, Tsuma M, et al. (2007) Administration of granulocyte colony-stimulating factor after myocardial infarction enhances the recruitment of hematopoietic stem cell-derived myofibroblasts and contributes to cardiac repair. Stem Cells 25: 2750-2859.
24. Murry CE, Soonpaa MH, Reinecke H, Nakajima H, Nakajima HO, et al. (2004) Haematopoietic stem cells do not transdifferentiate into cardiac myocytes in myocardial infarcts. Nature 428: 664-668.
25. Deten A, Volz HC, Clamors S, Leiblein S, Briest W, et al. (2005) Hematopoietic stem cells do not repair the infarcted mouse heart. Cardiovasc Res 65: 52-63.
26. Norol F, Bonnet N, Peinnequin A, Chretien F, Legrand R, et al. (2007) GFP-transduced CD34+ and Lin- CD34- hematopoietic stem cells did not adopt a cardiac phenotype in a nonhuman primate model of myocardial infarct. Exp Hematol 35: 653-661.
27. Dubois C, Liu X, Claus P, Marsboom G, Pokreisz P, et al. (2010) Differential effects of progenitor cell populations on left ventricular remodeling and myocardial neovascularization after myocardial infarction. J Am Coll Cardiol 55: 2232-2243.
28. Suuronen EJ, Price J, Veinot JP, Ascah K, Kapila V, et al. (2007) Comparative effects of mesenchymal progenitor cells, endothelial progenitor cells, or their combination on myocardial infarct regeneration and cardiac function. J Thorac Cardiovasc Surg 134: 1249-1258.
29. Kocher AA, Schuster MD, Szabolcs MJ, Takuma S, Burkhoff D, et al. (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.
30. Schuh A, Liehn EA, Sasse A, Hristov M, Sobota R, et al. (2008) Transplantation of endothelial progenitor cells improves neovascularization and left ventricular function after myocardial infarction in a rat model. Basic Res Cardiol 103: 69-77.
31. Li CJ, Gaol RL, Yang YJ, Hu FH, Yang WX, et al. (2010a) Effect of intracoronary infusion of bone marrow mononuclear cells or peripheral endothelial progenitor cells on myocardial ischemia-reperfusion injury in mini-swine. Chin Med Sci J 25 :176-181.
32. Atluri P, Panlilio CM, Liao GP, Hiesinger W, Harris DA, et al. (2010) Acute myocardial rescue with endogenous endothelial progenitor cell therapy. Heart Lung Circ 19: 644-654.
33. Oswald J, Boxberger S, Jorgensen B, Feldmann S, Ehninger G, et al. (2004)

Mesenchymal stem cells can be differentiated into endothelial cells in vitro. Stem Cells 22: 377-384.

34. Toma C, Pittenger MF, Cahill KS, Byrne BJ, Kessler PD (2002) Human mesenchymal stem cells differentiate to a cardiomyocyte phenotype in the adult murine heart. Circulation 105: 93-98.

35. Arminan A, Gandia C, Garcia-Verdugo JM, Lledo E, Trigueros C, et al. (2010) Mesenchymal stem cells provide better results than hematopoietic precursors for the treatment of myocardial infarction. J Am Coll Cardiol 55: 2244-2253.

36. Imanishi Y, Saito A, Komoda H, Kitagawa-Sakakida S, Miyagawa S, et al. (2008) Allogenic mesenchymal stem cell transplantation has a therapeutic effect in acute myocardial infarction in rats. J Mol Cell Cardiol 44: 662-671.

37. Chacko SM, Khan M, Kuppusamy ML, Pandian RP, Varadharaj S, et al. (2009) Myocardial oxygenation and functional recovery in infarct rat hearts transplanted with mesenchymal stem cells. Am J Physiol Heart Circ Physiol 296: H1263-1273.

38. Li Q, Turdi S, Thomas DP, Zhou T, Ren J (2010b) Intra-myocardial delivery of mesenchymal stem cells ameliorates left ventricular and cardiomyocyte contractile dysfunction following myocardial infarction. In Toxicol Lett 195: 119-126.

39. Tang J, Xie Q, Pan G, Wang J, Wang M (2006) Mesenchymal stem cells participate in angiogenesis and improve heart function in rat model of myocardial ischemia with reperfusion. Eur J Cardiothorac Surg 30: 353-361.

40. Kawada H, Fujita J, Kinjo K, Matsuzaki Y, Tsuma M, et al. (2004) Nonhematopoietic mesenchymal stem cells can be mobilized and differentiate into cardiomyocytes after myocardial infarction. Blood 104: 3581-3587.

41. Cheng Z, Liu X, Ou L, Zhou X, Liu Y, et al. (2008) Mobilization of mesenchymal stem cells by granulocyte colony-stimulating factor in rats with acute myocardial infarction. Cardiovasc Drugs Ther 22: 363-371.

42. Barbash IM, Chouraqui P, Baron J, Feinberg MS, Etzion S, et al. (2003) Systemic delivery of bone marrow-derived mesenchymal stem cells to the infarcted myocardium: feasibility, cell migration, and body distribution. In Circulation 108: 863-868.

43. Miyahara Y, Nagaya N, Kataoka M, Yanagawa B, Tanaka K, et al. (2006) Monolayered mesenchymal stem cells repair scarred myocardium after myocardial infarction. Nat Med 12: 459-465.

44. Yu LH, Kim MH, Park TH, Cha KS, Kim YD, et al. (2010) Improvement of cardiac function and remodeling by transplanting adipose tissue-derived stromal cells into a mouse model of acute myocardial infarction. Int J Cardiol 139: 166-172.

45. Alt E, Pinkernell K, Scharlau M, Coleman M, Fotuhi P, et al. (2010) Effect of freshly isolated autologous tissue resident stromal cells on cardiac function and perfusion following acute myocardial infarction. Int J Cardiol 144: 26-35.

46. Beltrami AP, Barlucchi L, Torella D, Baker M, Limana F, et al. (2003) Adult cardiac stem cells are multipotent and support myocardial regeneration. In Cell 114: 763-776.

47. Gonzalez A, Rota M, Nurzynska D, Misao Y, Tillmanns J, et al. (2008) Activation of cardiac progenitor cells reverses the failing heart senescent phenotype and prolongs lifespan. Circ Res 102: 597-606.

48. Kajstura J, Urbanek K, Perl S, Hosoda T, Zheng H, et al. (2010) Cardiomyogenesis in the adult human heart. Circ Res 107: 305-315.

49. Dawn B, Stein AB, Urbanek K, Rota M, Whang B, et al. (2005) Cardiac stem cells delivered intravascularly traverse the vessel barrier, regenerate infarcted myocardium, and improve cardiac function. In Proc Natl Acad Sci U S A 102: 3766-3771.

50. Wang X, Hu Q, Nakamura Y, Lee J, Zhang G, et al. (2006) The role of the sca-1+/CD31- cardiac progenitor cell population in postinfarction left ventricular remodeling. Stem Cells 24: 1779-1788.

51. Smits AM, van Laake LW, den Ouden K, Schreurs C, Szuhai K, et al. (2009) Human cardiomyocyte progenitor cell transplantation preserves long-term function of the infarcted mouse myocardium. Cardiovasc Res 83: 527-535.

52. Oh H, Bradfute SB, Gallardo TD, Nakamura T, Gaussin V, et al. (2003) Cardiac progenitor cells from adult myocardium: homing, differentiation, and fusion after infarction. Proc Natl Acad Sci U S A 100: 12313-12318.

53. Tang XL, Rokosh G, Sanganalmath SK, Yuan F, Sato H, et al. (2010) Intracoronary administration of cardiac progenitor cells alleviates left ventricular dysfunction in rats with a 30-day-old infarction. Circulation 121: 293-305.

54. Urbanek K, Rota M, Cascapera S, Bearzi C, Nascimbene A, et al. (2005) Cardiac stem cells possess growth factor-receptor systems that after activation regenerate the infarcted myocardium, improving ventricular function and long-term survival. Circ Res 97: 663-673.

55. Rota M, Padin-Iruegas ME, Misao Y, De Angelis A, Maestroni S, et al. (2008) Local activation or implantation of cardiac progenitor cells rescues scarred infarcted myocardium improving cardiac function. Circ Res 103: 107-116.

56. Ellison GM, Torella D, Dellegrottaglie S, Perez-Martinez C, Perez de Prado A, et al. (2011) Endogenous Cardiac Stem Cell Activation by Insulin-Like Growth Factor-1/Hepatocyte Growth Factor Intracoronary Injection Fosters Survival and Regeneration of the Infarcted Pig Heart. J Am Coll Cardiol 58: 977-986.

57. Thomson JA, Itskovitz-Eldor J, Shapiro SS, Waknitz MA, Swiergiel JJ, et al. (1998) Embryonic stem cell lines derived from human blastocysts. Science 282: 1145-1147.

58. Nelson TJ, Ge ZD, Van Orman J, Barron M, Rudy-Reil D, et al. (2006) Improved cardiac function in infarcted mice after treatment with pluripotent embryonic stem cells. Anat Rec A Discov Mol Cell Evol Biol 288: 1216-1224.

59. Qiao H, Zhang H, Zheng Y, Ponde DE, Shen D, et al. (2009) Embryonic stem cell grafting in normal and infarcted myocardium: serial assessment with MR imaging and PET dual detection. In Radiology 250: 821-829.

60. Lin Q, Fu Q, Zhang Y, Wang H, Liu Z, et al. (2010) Tumourigenesis in the infarcted rat heart is eliminated through differentiation and enrichment of the transplanted embryonic stem cells. Eur J Heart Fail 12: 1179-1185.

61. Christoforou N, Oskouei BN, Esteso P, Hill CM, Zimmet JM, et al. (2010) Implantation of mouse embryonic stem cell-derived cardiac progenitor cells preserves function of infarcted murine hearts. PLoS One 5: e11536.

62. Menard C, Hagege AA, Agbulut O, Barro M, Morichetti MC, et al. (2005) Transplantation of cardiac-committed mouse embryonic stem cells to infarcted sheep myocardium: a preclinical study. Lancet 366: 1005-1012.

63. Tomescot A, Leschik J, Bellamy V, Dubois G, Messas E, et al. (2007) Differentiation in vivo of cardiac committed human embryonic stem cells in postmyocardial infarcted rats. Stem Cells 2200-2205.

64. Min JY, Yang Y, Converso KL, Liu L, Huang Q, et al. (2002) Transplantation of embryonic stem cells improves cardiac function in postinfarcted rats. J Appl Physiol 92: 288-296.

65. Laflamme MA, Chen KY, Naumova AV, Muskheli V, Fugate JA, et al. (2007) Cardiomyocytes derived from human embryonic stem cells in pro-survival factors enhance function of infarcted rat hearts. Nat Biotechnol 25: 1015-1024.

66. Qiao H, Zhang H, Yamanaka S, Patel VV, Petrenko NB, et al. (2011) Long-term improvement in postinfarct left ventricular global and regional contractile function is mediated by embryonic stem cell-derived cardiomyocytes. Circ Cardiovasc Imaging 4: 33-41.

67. Blin G, Nury D, Stefanovic S, Neri T, Guillevic O, et al. (2010) A purified population of multipotent cardiovascular progenitors derived from primate pluripotent stem cells engrafts in postmyocardial infarcted nonhuman primates. J Clin Invest 120: 1125-1239.

68. Takahashi K, Yamanaka S (2006) Induction of pluripotent stem cells from mouse embryonic and adult fibroblast cultures by defined factors. In Cell 126: 663-676.

69. Takahashi K, Tanabe K, Ohnuki M, Narita M, Ichisaka T, et al. (2007) Induction of pluripotent stem cells from adult human fibroblasts by defined factors. Cell 131: 861-872.

70. Yu J, Vodyanik MA, Smuga-Otto K, Antosiewicz-Bourget J, Frane JL, et al. (2007) Induced pluripotent stem cell lines derived from human somatic cells. Science 318: 1917-1920.

71. van Laake LW, Qian L, Cheng P, Huang Y, Hsiao EC, et al. (2010) Reporter-

based isolation of induced pluripotent stem cell- and embryonic stem cell-derived cardiac progenitors reveals limited gene expression variance. Circ Res 107: 340-347.

72. Gupta MK, Illich DJ, Gaarz A, Matzkies M, Nguemo F, et al. (2010) Global transcriptional profiles of beating clusters derived from human induced pluripotent stem cells and embryonic stem cells are highly similar. BMC Dev Biol 10: 98.

73. Xi J, Khalil M, Shishechian N, Hannes T, Pfannkuche K, et al. (2010) Comparison of contractile behavior of native murine ventricular tissue and cardiomyocytes derived from embryonic or induced pluripotent stem cells. FASEB J 2739-2751.

74. Nelson TJ, Martinez-Fernandez A, Yamada S, Perez-Terzic C, Ikeda Y, et al. (2009) Repair of acute myocardial infarction by human stemness factors induced pluripotent stem cells. Circulation 120: 408-416.

75. Martinez-Fernandez A, Nelson TJ, Ikeda Y, Terzic A (2010) c-MYC independent nuclear reprogramming favors cardiogenic potential of induced pluripotent stem cells. J Cardiovasc Transl Res 3: 13-23.

76. Zwi-Dantsis L, Mizrahi I, Arbel G, Gepstein A, Gepstein L(2011) Scalable production of cardiomyocytes derived from c-Myc free induced pluripotent stem cells. Tissue Eng Part A 17:1027-1037.

77. Fujiwara M, Yan P, Otsuji TG, Narazaki G, Uosaki H, et al. (2011) Induction and enhancement of cardiac cell differentiation from mouse and human induced pluripotent stem cells with cyclosporin-A. PLoS One 6: e16734.

78. Burridge PW, Thompson S, Millrod MA, Weinberg S, Yuan X, et al. (2011) A universal system for highly efficient cardiac differentiation of human induced pluripotent stem cells that eliminates interline variability. PLoS One 6: e18293.

79. Rajala K, Lindroos B, Hussein SM, Lappalainen RS, Pekkanen-Mattila M, et al. (2010) A defined and xeno-free culture method enabling the establishment of clinical-grade human embryonic, induced pluripotent and adipose stem cells. PLoS One 5: e10246.

80. Zhou W, Freed CR (2009) Adenoviral gene delivery can reprogram human fibroblasts to induced pluripotent stem cells. Stem Cells 27: 2667-2674.

81. Wernig M, Lengner CJ, Hanna J, Lodato MA, Steine E, et al. (2008) A drug-inducible transgenic system for direct reprogramming of multiple somatic cell types. In Nat Biotechnol 26: 916-924.

82. Zhou H, Wu S, Joo JY, Zhu S, Han DW, et al. (2009) Generation of induced pluripotent stem cells using recombinant proteins. Cell Stem Cell 4: 381-384.

83. Yu J, Hu K, Smuga-Otto K, Tian S, Stewart R, et al. (2009) Human induced pluripotent stem cells free of vector and transgene sequences. Science 797-801.

84. Szabo E, Rampalli S, Risueno RM, Schnerch A, Mitchell R, et al. (2010) Direct conversion of human fibroblasts to multilineage blood progenitors. Nature 468: 521-526.

85. Menasche P, Hagege AA, Scorsin M, Pouzet B, Desnos M, et al. (2001) Myoblast transplantation for heart failure. Lancet 357: 279-280.

86. Herreros J, Prosper F, Perez A, Gavira JJ, Garcia-Velloso MJ, et al. (2003) Autologous intramyocardial injection of cultured skeletal muscle-derived stem cells in patients with non-acute myocardial infarction. Eur Heart J 24: 2012-2020.

87. Pagani FD, DerSimonian H, Zawadzka A, Wetzel K, Edge AS, et al. (2003) Autologous skeletal myoblasts transplanted to ischemia-damaged myocardium in humans. Histological analysis of cell survival and differentiation. J Am Coll Cardiol 41: 879-888.

88. Siminiak T, Fiszer D, Jerzykowska O, Grygielska B, Rozwadowska N, et al. (2005) Percutaneous trans-coronary-venous transplantation of autologous skeletal myoblasts in the treatment of post-infarction myocardial contractility impairment: the POZNAN trial. Eur Heart J 26: 1188-1195.

89. Hagege AA, Carrion C, Menasche P, Vilquin JT, Duboc D, et al. (2003) Viability and differentiation of autologous skeletal myoblast grafts in ischaemic cardiomyopathy. Lancet 361: 491-492.

90. Menasche P, Hagege AA, Vilquin JT, Desnos M, Abergel E, et al. (2003) Autologous skeletal myoblast transplantation for severe postinfarction left ventricular dysfunction. J Am Coll Cardiol :1078-1083.

91. Siminiak T, Kalawski R, Fiszer D, Jerzykowska O, Rzezniczak J, et al. (2004) Autologous skeletal myoblast transplantation for the treatment of postinfarction myocardial injury: phase I clinical study with 12 months of follow-up. In Am Heart J 148: 531-537.

92. Hagege AA, Marolleau JP, Vilquin JT, Alheritiere A, Peyrard S, et al. (2006) Skeletal myoblast transplantation in ischemic heart failure: long-term follow-up of the first phase I cohort of patients. Circulation 114: I108-113.

93. Dib N, Dinsmore J, Lababidi Z, White B, Moravec S, et al. (2009) One-year follow-up of feasibility and safety of the first U.S., randomized, controlled study using 3-dimensional guided catheter-based delivery of autologous skeletal myoblasts for ischemic cardiomyopathy (CAuSMIC study). In JACC Cardiovasc Interv 2: 9-16.

94. Menasche P, Alfieri O, Janssens S, McKenna W, Reichenspurner H, et al. (2008) The Myoblast Autologous Grafting in Ischemic Cardiomyopathy (MAGIC) trial: first randomized placebo-controlled study of myoblast transplantation. Circulation 117: 1189-1200.

95. Kang HJ, Kim HS, Zhang SY, Park KW, Cho HJ, et al. (2004) Effects of intracoronary infusion of peripheral blood stem-cells mobilised with granulocyte-colony stimulating factor on left ventricular systolic function and restenosis after coronary stenting in myocardial infarction: the MAGIC cell randomised clinical trial. Lancet 363: 751-756.

96. Ince H, Petzsch M, Kleine HD, Schmidt H, Rehders T, et al. (2005b) Preservation from left ventricular remodeling by front-integrated revascularization and stem cell liberation in evolving acute myocardial infarction by use of granulocyte-colony-stimulating factor (FIRSTLINE-AMI). Circulation 112: 3097-4106.

97. Ince H, Petzsch M, Kleine HD, Eckard H, Rehders T, et al. (2005a) 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 112: I73-80.

98. Zohlnhofer D, Ott I, Mehilli J, Schomig K, Michalk F, et al. (2006) Stem cell mobilization by granulocyte colony-stimulating factor in patients with acute myocardial infarction: a randomized controlled trial. JAMA 295: 1003-1010.

99. Ripa RS, Jorgensen E, Wang Y, Thune JJ, Nilsson JC, et al. (2006) 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. In Circulation 113: 1983-1992.

100. Kuethe F, Figulla HR, Herzau M, Voth M, Fritzenwanger M, et al. (2005) Treatment with granulocyte colony-stimulating factor for mobilization of bone marrow cells in patients with acute myocardial infarction. In Am Heart J 150: 115.

101. Valgimigli M, Rigolin GM, Cittanti C, Malagutti P, Curello S, et al. (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.

102. Engelmann MG, Theiss HD, Hennig-Theiss C, Huber A, Wintersperger BJ, et al. (2006) Autologous bone marrow stem cell mobilization induced by granulocyte colony-stimulating factor after subacute ST-segment elevation myocardial infarction undergoing late revascularization: final results from the G-CSF-STEMI (Granulocyte Colony-Stimulating Factor ST-Segment Elevation Myocardial Infarction) trial. J Am Coll Cardiol 48: 1712-1721.

103. Schachinger V, Erbs S, Elsasser A, Haberbosch W, Hambrecht R, et al. (2006a) Intracoronary bone marrow-derived progenitor cells in acute myocardial infarction. N Engl J Med 355: 1210-1221.

104. Schachinger V, Erbs S, Elsasser A, Haberbosch W, Hambrecht R, et al. (2006b) Improved clinical outcome after intracoronary administration of bone-marrow-derived progenitor cells in acute myocardial infarction: final 1-year results of the REPAIR-AMI trial. In Eur Heart J 27: 2775-2783.

105. Wollert KC, Meyer GP, Lotz J, Ringes-Lichtenberg S, Lippolt P, et al. (2004)

Intracoronary autologous bone-marrow cell transfer after myocardial infarction: the BOOST randomised controlled clinical trial. Lancet 364: 141-148.

106. Assmus B, Schachinger V, Teupe C, Britten M, Lehmann R, et al. (2002) Transplantation of Progenitor Cells and Regeneration Enhancement in Acute Myocardial Infarction (TOPCARE-AMI). Circulation 106: 3009-3017.

107. Stamm C, Westphal B, Kleine HD, Petzsch M, Kittner C, et al. (2003) Autologous bone-marrow stem-cell transplantation for myocardial regeneration. Lancet 361: 45-46.

108. Grajek S, Popiel M, Gil L, Breborowicz P, Lesiak M, et al. (2010) Influence of bone marrow stem cells on left ventricle perfusion and ejection fraction in patients with acute myocardial infarction of anterior wall: randomized clinical trial: Impact of bone marrow stem cell intracoronary infusion on improvement of microcirculation. Eur Heart J 31: 691-702.

109. Meyer GP, Wollert KC, Lotz J, Pirr J, Rager U, et al. (2009) Intracoronary bone marrow cell transfer after myocardial infarction: 5-year follow-up from the randomized-controlled BOOST trial. Eur Heart J 30: 2978-2984.

110. Erbs S, Linke A, Schachinger V, Assmus B, Thiele H, et al. (2007) Restoration of microvascular function in the infarct-related artery by intracoronary transplantation of bone marrow progenitor cells in patients with acute myocardial infarction: the Doppler Substudy of the Reinfusion of Enriched Progenitor Cells and Infarct Remodeling in Acute Myocardial Infarction (REPAIR-AMI) trial. Circulation 116: 366-374.

111. Tendera M, Wojakowski W, Ruzyllo W, Chojnowska L, Kepka C, et al. (2009) Intracoronary infusion of bone marrow-derived selected CD34+CXCR4+ cells and non-selected mononuclear cells in patients with acute STEMI and reduced left ventricular ejection fraction: results of randomized, multicentre Myocardial Regeneration by Intracoronary Infusion of Selected Population of Stem Cells in Acute Myocardial Infarction (REGENT) Trial. In Eur Heart J 30: 1313-1321.

112. Strauer BE, Yousef M, Schannwell CM (2010) The acute and long-term effects of intracoronary Stem cell Transplantation in 191 patients with chronic heARt failure: the STAR-heart study. In Eur J Heart Fail 12: 721-729.

113. Perin EC, Silva GV, Henry TD, Cabreira-Hansen MG, Moore WH, et al. (2011) A randomized study of transendocardial injection of autologous bone marrow mononuclear cells and cell function analysis in ischemic heart failure (FOCUS-HF). Am Heart J 161: 1078-1087 e3.

114. Arnold RA, Villa H, Gutierrez PL, Sanchez F, Gimeno ME et al. (2010) Absence of accelerated atherosclerotic disease progression after intracoronary infusion of bone marrow derived mononuclear cells in patients with acute myocardial infarction--angiographic and intravascular ultrasound--results from the TErapia Celular Aplicada al Miocardio Pilot study. In Am Heart J 159: 1154 e1-8.

115. Katritsis DG, Sotiropoulou PA, Karvouni E, Karabinos I, Korovesis S, et al. (2005) Transcoronary transplantation of autologous mesenchymal stem cells and endothelial progenitors into infarcted human myocardium. Catheter Cardiovasc Interv 65: 321-329.

116. Hare JM, Traverse JH, Henry TD, Dib N, Strumpf RK, et al. (2009) A randomized, double-blind, placebo-controlled, dose-escalation study of intravenous adult human mesenchymal stem cells (prochymal) after acute myocardial infarction. J Am Coll Cardiol 54: 2277-2286.

117. Katritsis DG, Sotiropoulou P, Giazitzoglou E, Karvouni E, Papamichail M (2007) Electrophysiological effects of intracoronary transplantation of autologous mesenchymal and endothelial progenitor cells. Europace 9: 167-171.

118. Mohyeddin-Bonab M, Mohamad-Hassani MR, Alimoghaddam K, Sanatkar M, Gasemi M, et al. (2007) Autologous in vitro expanded mesenchymal stem cell therapy for human old myocardial infarction. Arch Iran Med 10: 467-473.

119. Schachinger V, Assmus B, Britten MB, Honold J, Lehmann R, et al. (2004) Transplantation of progenitor cells and regeneration enhancement in acute myocardial infarction: final one-year results of the TOPCARE-AMI Trial. J Am Coll Cardiol 19: 1690-1699.

120. Assmus B, Honold J, Schachinger V, Britten MB, Fischer-Rasokat U, et al. (2006) Transcoronary transplantation of progenitor cells after myocardial infarction. In N Engl J Med 355: 1222-1232.

This article was originally published in a special issue, **Cardiovascular Cell Therapy** handled by Editor(s). Dr. Martin Rodriguez-Porcel, Medicine Mayo Medical School, USA; Dr. Rosalinda Madonna, USA