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Molecular imaging: The key to advancing cardiac stem cell therapy

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Abstract

Cardiac stem cell therapy continues to hold promise for the treatment of ischemic heart disease despite the fact that early promising pre-clinical findings have yet to be translated into consistent clinical success. The latest human studies have collectively identified a pressing need to better understand stem cell behavior in humans and called for more incorporation of noninvasive imaging techniques into the design and evaluation of human stem cell therapy trials. This review discusses the various molecular imaging techniques validated to date for studying stem cells in living subjects, with a particular emphasis on their utilities in assessing the acute retention and the long-term survival of transplanted stem cells. These imaging techniques will be essential for advancing cardiac stem cell therapy by providing the means to both guide ongoing optimization and predict treatment response in humans.

Introduction

The development of stem cell therapy for ischemic heart disease has followed a growth pattern best described as premature enthusiasm followed by premature disappointments. Indeed, countless pre-clinical studies have initially reported encouraging findings for various cell types including skeletal myoblasts (SKMs), bone marrow-derived stem cells (BMCs), mesenchymal stem cells (MSCs), circulating progenitor cells (CPCs), embryonic stem cells (ESCs), and cardiac resident cells (CSCs) (Segers and Lee, 2008). However, before the working of these stem cells has been fully elucidated, recent large-scale clinical trials have already raised concerns over the untoward side-effects of SKM therapy (Menasche et al., 2008) and the marginal benefits of BMC therapy (Perin et al., 2012; Traverse et al., 2011, 2012). Although disappointing, these trials have revealed a pressing need to better understand stem cell behavior in humans.

The development of molecular imaging tools has enabled unprecedented opportunities to interrogate stem cells in living subjects (Chen and Wu, 2011). Using these tools, stem cell scientists are now capable of addressing some of the unanswered questions arising from recent clinical trials, including the optimal cell type, delivery route, dosing regimen, and timing of cell delivery (Fig. 1). In the present review, we (1) highlight various molecular imaging techniques developed to date for noninvasively tracking stem cells and (2) discuss

their utilities in assessing, optimizing, and guiding the clinical translation of stem cell therapy. Our hope is that a more widespread use of molecular imaging techniques in clinical trials will help further advance cardiac stem cell therapy in humans.

Molecular imaging techniques for tracking stem cells

Various imaging modalities have been validated for tracking stem cells, and these include fluorescence imaging (FI), bioluminescence imaging (BLI), positron emission tomography (PET), single-photon emission computed tomography (SPECT), magnetic resonance imaging (MRI), ultrasound (US), and computed tomography (CT). The selection of a given imaging modality depends on its strengths and weaknesses with respect to the intended application.

Cell imaging modalities

BLI has been the most popular imaging modality for small animal studies due to its superior imaging sensitivity (10^{-15} mol/L, compared to 10^{-12} , 10^{-11} , and 10^{-5} mol/L for PET, SPECT, and MRI, respectively) (Massoud and Gambhir, 2003). Despite its poor spatial resolution (3–5 mm), BLI has had unparalleled success in the high-throughput assessment of stem cell homing, engraftment, differentiation, and survival in small animal models (de Almeida et al., 2011). By comparison, planar FI has been limited to proof-of-principle studies, where imaging performance is not significantly compromised by its high background signal (Lin et al., 2007).

Imaging modalities such as PET, SPECT, and MRI allow tomographic assessment of cells in both small and large animals, as well as humans. PET and SPECT, when combined with CT, have been particularly useful in quantifying the whole-body distribution of cells after delivery, whereas MRI has seen more utility in determining the transmural location of stem cells due to its superb spatial resolution (10–100 μ m for small animal MRI; 0.5–1.5 mm for human MRI), about 10–20-fold greater than that of either PET or SPECT (Massoud and Gambhir, 2003).

Although traditionally used for anatomical imaging, both ultrasound and CT have found new applications in direct imaging of stem cell engraftment using targeted microbubbles (Kuliszewski et al., 2009) and radiopaque microcapsules (Fu et al., 2010). The combined use of different imaging modalities (e.g., PET/MRI)—with the goal to utilize their individual strengths and complement their respective shortcomings—represents the latest trend in noninvasive imaging of stem cell therapy (Qiao et al., 2009).

Cell labeling approaches: direct versus reporter gene labeling

A critical aspect of stem cell imaging lies in the selection of an appropriate cell marker. Two commonly used cell labeling methods are *direct labeling* and *reporter gene labeling*. In general, direct labeling is suitable for applications where short-term imaging is adequate (e.g., imaging acute cell retention), whereas reporter gene labeling is ideal for longitudinal imaging (e.g., imaging cell survival kinetics).

Direct cell labeling involves incubating cells with a contrast agent, the choice of which depends on the imaging modality chosen. The most commonly used contrast agents include quantum dots for FI (Lin et al., 2007), ^{18}F -FDG for PET (Hofmann et al., 2005), $^{99\text{m}}\text{Tc}$ -HMPAO (Musialek et al., 2011) for SPECT, ^{111}In -oxine for planar gamma camera imaging (Hou et al., 2005), and superparamagnetic iron oxide (SPIO) nanoparticles for MRI (Chen et al., 2009). Direct cell labeling can be easily performed to achieve high cell labeling efficiency prior to implantation. However, because these labels can be diluted with cell division and potentially engulfed by resident macrophages if released after stem cell death

(e.g., SPIO), the cell signal obtained over time may not reflect true stem cell viability (Chen et al., 2009; Li et al., 2008). Therefore, direct cell labeling has found the greatest utility in applications where cell death is minimized (e.g., imaging of acute cell retention).

Reporter gene labeling of stem cells involves transfecting or transducing cells with a reporter gene using either a non-viral or a viral vehicle such that its transgene expression under the regulation of a chosen promoter can be imaged either with or without an imaging probe. The most commonly used reporter genes are fluorescent proteins for FI (Gyongyosi et al., 2008), firefly luciferase for BLI (Chen et al., 2009), Herpes Simplex Virus type 1 thymidine kinase (HSV1-tk) or its mutant variants for PET (Cao et al., 2006; Liu et al., 2012), sodium iodide symporter (NIS) for SPECT (Terrovitis et al., 2008), and ferritin for MRI (Campan et al., 2011). The main advantage of reporter gene labeling is that the cell signal obtained is specific to viable cells, as dead cells cannot express reporters to generate an imaging signal. When the reporter gene is integrated into the cell genome by means of stable plasmid transfection or viral vector-mediated integration, the cell signal will then be reflective of cell number. The potential drawbacks of reporter gene labeling include potential mutagenesis with non-specific vector integration and gene/promoter silencing. Despite these barriers, reporter gene labeling has been successfully performed in a recent human oncological trial (Yaghoubi et al., 2009) and will likely play a more pivotal role in the advancement of human cardiac stem cell therapy. Furthermore, recent advances in site-specific integration using zinc finger nucleases and phiC31 integrase are expected to mitigate the concerns of random genomic integration (Wang et al., 2012b; Lan et al., 2012).

Conceivably, both direct and reporter gene labeling can cause untoward perturbation to the stem cells. Fortunately, with careful titration of cell labels, cytotoxicity can be effectively minimized. For direct cell labeling, most contrast agents discussed herein, except for ^{111}In -oxine (Gholamrezaezhad et al., 2009) and debatably quantum dots (Lin et al., 2007), have been shown to not significantly affect cell viability or proliferation. The same safety profile holds true for commonly used reporter genes such as HSV1-tk or its mutant variant HSV1-sr39tk, firefly luciferase, NIS, and ferritin. More reassuringly, comprehensive microarray and proteomic studies have demonstrated only negligible transcriptional and translational changes associated with the overexpression of HSV1-sr39tk (Wu et al., 2006a, 2006b), a reporter gene that has been successfully and safely used in humans.

Noninvasive imaging of acute cell retention

Early myocardial cell retention following cell delivery has been shown in pre-clinical models of myocardial ischemia to predict the long-term outcome of stem cell therapy (Liu et al., 2012). Yet, none of the large-scale clinical trials thus far has included a thorough investigation of cell engraftment, mainly because of the lack of tools for doing so. Owing to recent advances in molecular imaging, several pre-clinical and small-scale clinical studies have now been able to assess various parameters affecting acute cell retention using molecular imaging techniques.

Lessons learned from human imaging studies

Various imaging modalities have been used to evaluate acute myocardial cell retention, but only radionuclide imaging has been shown to be effective in humans. Whereas BLI is limited to small animals because of light attenuation, MRI using iron oxide as a cell label is better for determining the trans-mural location of transplanted cells, but not their whole-body distribution due to the lower imaging sensitivity of MRI compared to radionuclide-based imaging techniques (Kraitchman et al., 2005). Radionuclide imaging has been routinely used in the clinical setting for evaluating the trafficking of specific cell types (leukocytes, red blood cells) and therefore can be readily applied to studying stem cells. For

these reasons, all human imaging studies to date have employed PET, SPECT, or planar gamma camera imaging for monitoring acute cell retention (Table 1).

The feasibility of using PET to study acute myocardial cell retention was first performed in a small number of patients with acute myocardial infarction (AMI) (5–10 days post-PCI) who underwent either intracoronary (IC) infusion or intravenous (IV) delivery of ^{18}F -FDG-labeled BMCs (Hofmann et al., 2005). Myocardial ^{18}F -FDG uptake was weak (3%) and undetectable for IC and IV infusions, respectively, suggesting modest superiority of IC delivery. Notably, enrichment for the CD34+ marker led to a greater myocardial engraftment (26%). The superiority of IC over IV delivery was later recapitulated in a PET study of chronic MI (90 days post-MI) involving ^{18}F -FDG-labeled CPCs, in which poor cell retention was also observed (2% and 0% for IC and IV, respectively) (Kang et al., 2006). In fact, poor acute cell retention remains a common theme among all human imaging studies to date, irrespective of stem cell type (BMC or CPC), cell delivery route (IC or IV), cell dose (15–4000 million), timing of cell delivery (5–10 days or up to 17 years), and cell label (^{18}F -FDG, ^{111}In -Oxine, $^{99\text{m}}\text{Tc}$ -HMPAO) (Table 1). Considering that the persistence of engrafted cells is necessary for the sustained benefits of stem cell therapy (e.g., neovascularization, cardiac function) (Ziebart et al., 2008), the development of means to improve cell retention will be needed to maximize the efficacy of stem cell therapy.

Investigation of factors affecting acute cell retention

Numerous studies have shown that the cell delivery route can significantly influence acute cell retention. For instance, a porcine study showed that intramyocardial (IM) delivery of ^{111}In -Oxine-labeled peripheral blood stem cells 6 days post-MI led to 4-fold greater, albeit more variable, cell retention compared to IC or interstitial retrograde coronary venous (IRV) injection, while causing 2-fold less unintended retention in the lungs (Hou et al., 2005). Several human imaging studies thus far showed undetectable cell retention with IV delivery (Hofmann et al., 2005; Kang et al., 2006). Taken all together, IM delivery appears to be the most efficient delivery route with a modest risk of leakage to the systemic circulation. More imaging studies will be needed to compare and optimize the different cell delivery routes in humans.

Following cell delivery, the number of cells acutely retained in the myocardium should theoretically depend on the number of cells delivered. However, the nature of this relationship has not been well established in humans. Early clinical trials on IC infusion of BMCs in the setting of AMI failed to demonstrate a clear correlation between the number of cells delivered and the functional end points studied (Lunde et al., 2006; Meyer et al., 2006). However, a more recent trial showed a clear dose-response relationship for IC infusion of selected CD34+ BMCs in similar patients (Quyyumi et al., 2011). These inconsistent findings raised the possibility that the number of cells acutely retained in the myocardium might not have directly correlated with the number of cells delivered in the earlier studies, leading to an obscuration of a clear dose-response relationship. These conflicting studies highlighted the importance of using cell imaging techniques in human trials to quantify the actual number of cells acutely retained so as to better characterize the dose-response relationship and establish the optimal cell dose for maximal functional benefits.

Lastly, the timing of cell delivery after AMI has been implicated in the differential outcomes of human stem cell therapy. However, no large-scale trial to date has been specifically powered to study this question. Subgroup analysis of an early trial showed a lack of response in patients receiving IC infusion of BMCs within 24 h of MI (Janssens et al., 2006), perceivably due to the abundant cytotoxic inflammatory cytokines released after MI. In fact, a meta-analysis of 7 trials afterwards suggested that an optimal window of 4–7 days post-MI for cell delivery could lead to better LVEF because of the peaking of chemotactic

signals around that time frame (Zhang et al., 2009). A human SPECT study showed that IC delivery of ^{111}In -oxine-labeled CPCs within 14 days led to the greatest myocardial cell retention (6.3%) compared to injection after 14 days, and up to 1 year (4.5%) (Schachinger et al., 2008). This is consistent with the finding of a recent trial showing IC infusion of BMCs 2–3 weeks after the index MI led to no improvement in LVEF at 6 months (Traverse et al., 2011). Although revealing, the aforementioned studies had different study designs which made an unbiased interpretation difficult. To reconcile this, a large multi-center randomized placebo-controlled trial is now underway to help determine the optimal timing of cell delivery (5–7 days versus 3–4 weeks) for maximal therapeutic benefits (Surder et al., 2010). Hence future incorporation of cell imaging techniques into these studies will be needed to control for acute cell retention by excluding mis-injection as a potential confounding factor for analysis.

Optimization of acute cell retention

Most attempts to specifically improve acute cell retention have primarily focused on altering the physical properties of the cell graft. A rat study involving PET imaging of ^{18}F -FDG-labeled CDCs intramyocardially injected into the peri-infarct myocardium showed that both slowing the ventricular rate with adenosine to reduce cell “washout” and co-injecting cells with fibrin glue to minimize back cell leakage during injection can improve acute cell retention by 2-fold (35–38%) (Terrovitis et al., 2009). Enrichment of human amniotic fluid stem cells (hAFSCs) with methylcellulose hydrogel could similarly increase cell retention in rats by as much as 5-fold due the larger effective size of the composite graft for entrapment within the myocardial interstitium (Lee et al., 2011). In a recent proof-of-principle study, pigs co-injected intramyocardially with Matrigel and BMCs expressing HSV1-sr39tk led to a greater PET cell signal, compared to only background signal for cells injected alone (Willmann et al., 2009). These latter findings suggested that the employment of a “scaffold” to physically congeal injected cells can be highly effective and should warrant further efficacy testing in large animals and humans.

Noninvasive imaging of long-term stem cell survival

The long-term behavior of stem cells following implantation is largely unknown in humans due to the lack of imaging tools to longitudinally image them. The aforementioned imaging techniques using direct radioactive cell labeling are limited by the short half-lives of the radioisotopes used and therefore can only be used to track cells for a finite duration. The dilution of radioactive labels with cell division further renders these techniques unsuitable for quantification of cell number. Reporter gene-based cell tracking techniques overcome these limitations and have been the mainstay for noninvasive investigation of long-term stem cell survival in pre-clinical models of myocardial ischemia.

Lessons learned from pre-clinical imaging studies

Consistent with other studies based on post-mortem histology or PCR, rodent studies using BLI or microPET have demonstrated poor long-term cell survival after implantation for most cell types (i.e., less than 10% of the initially delivered cells survive by 3 weeks) (van der Bogt et al., 2008). The reasons for the poor cell survival have been attributed to ischemia, ischemia-reperfusion injury, inflammation, immune rejection, and apoptosis. This phenomenon also carries over to large animal studies. In one porcine study, PET imaging of HSV1-sr39tk-expressing human MSCs intramyocardially injected into the peri-infarct myocardium showed barely detectable cell signal after 7 days (Gyongyosi et al., 2008). Interestingly, with about 20-fold more cells injected in another porcine study, the cell signal could be detected up to 5 months, with an estimated cell survival of 40–50% (Perin et al., 2011). These studies suggested that long-term stem cell survival could be species-dependent

and argued for the use of reporter gene-based imaging techniques in human trials to correlate stem cell survival with functional outcomes.

Optimization of long-term stem cell survival

Recognizing poor cell survival as a major problem facing stem cell therapy, various investigators have looked into means of improving cell survival. Some early studies have shown that co-delivery of cells with drugs (e.g., antioxidants (Rodriguez-Porcel et al., 2010), immunomodulators (Pearl et al., 2011), or growth factors (Xie et al., 2007) can promote cell survival via various mechanisms (e.g., counteracting oxidative stress, stimulating angiogenesis, maximizing stem cell differentiation, and minimizing rejection). However, the survival benefits tend to be short-lived (1–2 weeks) with these approaches due to the short half-lives of the co-injectants. To circumvent this problem, stem cells have been genetically engineered to persistently express growth factors (Deuse et al., 2009) or microRNAs (Hu et al., 2011) capable of countering hypoxia or apoptosis. This approach has the advantage of not perturbing the effective size of the cell graft and therefore can be used with all known delivery routes.

Besides genetic modification, tissue engineering-based approaches have also been explored as a means to augment graft function as a composite entity. At the simplest level, co-injection of cells intermixed with extracellular matrix materials (e.g., matrigel (Laflamme et al., 2007) or fibrin (Terrovitis et al., 2009)) has been found to improve cell survival in rodents via promotion of cell–host interaction. Micro-encapsulation of stem cells in semi-permeable RGD-modified alginate spheres is yet another approach to improve cell survival by providing physical support, immune protection, and substrates for therapeutic angiogenesis (Yu et al., 2010). Common to these approaches is the need to incorporate additional acellular components, which can increase either the viscosity or the effective size of the composite grafts. For this reason, these techniques will likely play a stronger role in conditions where IM delivery is more ideal (e.g., chronic ischemia, where IC delivery may be limited by total/sub-total occlusions).

Molecular imaging of the efficacy of stem cell therapy

Clinical trials thus far have relied on the use of conventional imaging techniques such as echocardiography, delayed-enhancement MRI, and nuclear myocardial perfusion imaging to evaluate the efficacy of stem cell therapy. Their popularity stems from the fact that the measurements obtained (e.g., LVEF) are predictive of cardiovascular mortality (Solomon et al., 2005). However, some reports have challenged the use of LVEF for evaluating BMC therapy, noting its inadequacy especially in evaluating remodeled ventricles (Williams et al., 2011). Regional contractility, LV size, and infarct size were thought to be better indicators of favorable outcomes. The caveat with these indices is that their changes often do not become apparent in humans until a few months into the therapy, by which time the opportunity to modify treatment strategies for those destined to fail may have already passed. Conceivably, the use of molecular imaging to detect early molecular events associated with cell therapy may help identify biomarkers which can serve as predictors of long-term outcome and be used to guide therapy.

Predicting the functional outcome of stem cell therapy

The previously described cell imaging techniques represent prime examples of attempts to identify biomarkers that can be used to predict the outcome of stem cell therapy. This point is well demonstrated in a recent PET study of cardiac progenitor cell therapy for murine models of AMI, in which early cell signal (1 day post-injection), a surrogate marker for acute cell retention, was found to predict the improvement in LVEF at 2 weeks (Liu et al.,

2012) (Fig. 2). Likewise, a recent randomized clinical trial of CD34+ BMC therapy for dilated cardiomyopathy showed that acute cell retention at 2 and 18 h post-intracoronary infusion, as assessed by SPECT imaging of ^{99m}Tc-HMPAO-labeled cells, could predict significant LVEF improvement at 3- and 12-months follow-ups (Vrtovec et al., 2012).

Besides imaging cell retention and viability, recent studies have shown the feasibility of using novel reporter gene constructs to image stem cell differentiation (e.g., Tie2 expression during endothelial differentiation from MSCs (Wang et al., 2012a)). Conceivably, by quantifying the degree of stem cell differentiation into the desired cell type, it would be feasible to predict the treatment response to stem cell therapy, assuming that greater stem cell differentiation will directly translate into greater benefits from the working of differentiated cells.

Another potentially promising imaging biomarker is the endothelial α 3-integrin, whose expression has been implicated in the early steps of stem cell-mediated angiogenesis. A recent porcine study using an ¹²³I-labeled integrin-binding probe (¹²³I-Gluco-RGD) showed the feasibility of quantifying stimulated angiogenesis (up to 1.7-fold of control) for subjects undergoing plasmid-mediated VEGF therapy (Johnson et al., 2008). The same technique should also be useful for assessing the effectiveness of stem cell therapy.

Lastly, the molecular events underpinning post-MI ventricular remodeling could be explored as imaging markers, as BMCs are known to reduce ventricular remodeling (Dixon et al., 2009). Various molecular imaging techniques for studying the molecular events associated with LV remodeling (e.g., metalloproteinase activity, apoptosis, collagen deposition, angiotensin receptor 2 expression) have already been extensively validated (Chen and Wu, 2011; Nguyen et al., 2011). Therefore, the potential is immense for combining molecular and conventional imaging techniques to both predict and assess the efficacy of stem cell therapy.

Conclusion

Reminiscent of early development of gene therapy for ischemic heart disease, cardiac stem cell therapy has enjoyed early pre-clinical success only to be followed by equivocal results in clinical trials. The common barrier of these molecular therapies in humans is the lack of imaging tools to fully understand their actions in humans after administration. Thanks to recent development of molecular imaging techniques for tracking stem cells, scientists are now more equipped than ever to unravel the mysteries underlying stem cell failure in humans. As recent phase I trials of stem cell therapy using cardiac stem cells (CSCs) and cardiosphere-derived cells (CDCs) seemed to yield more promising results than those with BMCs (Bolli et al., 2011; Makkar et al., 2012), now is the time to make use of molecular imaging to thoroughly optimize various stem cell therapies in humans so as to ensure persistent clinical success. The imaging techniques presented herein should also help accelerate the ongoing development of novel cell therapies based on the inducible pluripotent stem cell (iPSC) technology, which carries tremendous hope for safe and effective large-scale myocardial repair with patient-specific (non-immunogenic) stem cell derivatives in the future (Pearl et al., 2012).

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Fig. 1.

A flow diagram of important steps for performing image-guided stem cell therapy. There are unanswered questions regarding the choice of stem cell type, optimal cell labeling method, cell delivery route, means to assess and promote acute cell retention or long-term survival, as well as methods or indices for best assessing the efficacy of stem cell therapy.

Abbreviations: ^{18}F -FDG, ^{18}F -fluorodeoxyglucose; $^{99\text{m}}\text{Tc}$ -HMPAO, $^{99\text{m}}\text{Tc}$ -hexamethylpropyleneamine oxide; SPIO, superparamagnetic iron oxide; USPIO, ultrasmall superparamagnetic iron oxide; MPIO, microsized particles of iron oxide; HSV1-tk/HSV1-sr39tk, wild type/mutant Herpes Simplex Virus type 1 thymidine kinase; D2R/D2R80a, wild type/mutant dopamine type 2 receptor; NIS, sodium-iodide symporter; wk, week; HIF-1, hypoxia-inducible factor-1; VEGF, vascular endothelial growth factor; Bcl-2, B cell lymphoma 2; ECM, extracellular matrix; PET, positron emission tomography; SPECT, single-photon emission computed tomography; GCI, planar gamma camera imaging; MRI, magnetic resonance imaging; BLI, bioluminescence imaging; US, ultrasound; CT, computed tomography.

Fig. 2.

Early cell retention predicts long-term myocardial functional improvement. A cohort of SCID beige mice underwent experimental myocardial infarction followed by intramyocardial injection of human cardiac progenitor cells expressing a mutant thymidine kinase (A168H TK) reporter gene. Serial PET and MR imaging were performed afterwards on these mice to assess cell engraftment and left ventricular systolic function. (A) A representative coronal PET image is shown for a mouse with low cell retention on day 1 (top), compared to that of a mouse with high cell retention (bottom). (B) Representative short-axis MR images of their hearts in both diastole and systole at day 1 and week 4 after cell delivery. (C) The average MRI-derived ejection fraction is greater at both weeks 2 and 4 for the mouse cohort with high initial cell retention, compared to that of the cohort with low cell retention ($p < 0.001$; $n = 19$ and 18 for mouse cohorts with high and low initial cell retention, respectively). Figures adopted from Liu et al. (2012).

Table 1

Human stem cell studies to date that focus on imaging acute cell retention or homing in patients with ischemic cardiomyopathy.

Authors/year	N	Cell type	Delivery route	Timing of delivery post-MI/ ^a PCI	Total cells delivered (10 ⁶)/% labeled	Cell imaging modality	Cell labeling technique	Maximum myocardial cell retention (%)
Hofmann et al. (2005)	9	1. BMC 2. BMC CD34+	IC/IV	* ₅ –10 d	1. 2486/5 2. 1754/100	PET	¹⁸ F-FDG	1. IC: 3 (50–75 min) IV: UD (50–75 min) 2. IC: 26 (50–75 min)
Karpov et al. (2005)	44	BMC	IC	7–21 d	89/100	SPECT	^{99m} Tc-HMPAO	NQ/96 8 (30 min)
Kang et al. (2006)	20	CPC	IC/IV	3–>300 d	450/100	PET	¹⁸ F-FDG	73/NQ IC: 2 (2 h) IV: UD (2 h)
Goussetis et al. (2006)	8	BMC CD133++BMC CD133–CD34+	IC	45±36 mo	16/100	GCI	^{99m} Tc-HMPAO	29/>95 9 (1 h)
Blocklet et al. (2006)	6	CPC CD34+	IC	7–21 d	15/40	PET WB GCI	¹¹¹ In-Oxine + ¹⁸ F-FDG	65, 6/NQ 6 (1 h)
Penicka et al. (2007)	10	BMC	IC	* ₁ . 3–10 d	3934/20	SPECT	^{99m} Tc-HMPAO	90/97 1. 3 (2 h)
Caveliers et al. (2007)	8	CPC CD133+	IC	2. 2–5 yr >12 mo	8/35	WB GCI WB GCI	¹¹¹ In-Oxine	2. 2 (2 h) 7 (1 h)
Schachinger et al. (2008)	19	CPC	IC	5 d–17 yr	15/10	WB GCI	¹¹¹ In-Oxine	29/90 7 (1 h)
Dedobbeleer et al. (2009)	12	CPC CD34+	IC	20±2 mo	23/9	PET	¹⁸ F-FDG	5/96 3 (1 h)
Musialek et al. (2011)	24	BMC CD34+	IC	* ₆ –14 d	4/100	SPECT	^{99m} Tc-HMPAO	6/96 5 (1 h)

Abbreviations: BMC, bone marrow-derived cells; CPC, circulating progenitor cells; IC, intracoronary; IV, intravenous; PCI, percutaneous coronary intervention; MI, myocardial infarction; WB GCI, whole-body gamma camera imaging; ¹⁸F-FDG, ¹⁸F-fluorodeoxyglucose; ^{99m}Tc-HMPAO, ^{99m}Tc-hexamethylpropyleneamine oxime; NQ, not quantified; UD, undetectable;

* post-PCI;

min, minute; h, hour; d, day; mo, month; yr, year; PET, positron emission tomography; SPECT, single photon emission computed tomography; GCI, gamma camera imaging. All original values have been rounded to full digits. Only average values are reported unless otherwise specified. Table modified from Chen and Wu (2011).