

Stem Cell Therapy: A Primer for Interventionalists and Imagers



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In recent years, research advancement in stem cell therapy has been rapid. Accordingly, general clinical, scientific, and public attention to the application of stem cell therapy has been substantial. Promises are great, most notably with regard to the application of stem cell therapy for diseases that are currently difficult to treat or incurable such as Parkinson disease or diabetes mellitus. It is in the best interest of patient care for diagnostic and interventional radiologists to be actively involved in the development of these therapies, both at the bench and at the bedside in clinical studies. Specifically, the diagnostic radiologist can become an expert in imaging, tracking, and monitoring of stem cells and in the assessment of engraftment efficiency, whereas the interventionalist is a natural expert in targeted stem cell delivery by means of different routes (percutaneous, selective intravenous, or intraarterial). In addition, there is a potential role for the interventionalist to create engraftment territory and increase engraftment bed fertility with controlled intentional tissue destruction (eg, by means of thermal ablation) that might precede stem cell administration.

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Abbreviations: MS = multiple sclerosis, RF = radiofrequency, SPIO = superparamagnetic iron oxide

RESULTS of recent stem cell research have received much clinical, scientific, and public attention, and clinical applications of stem cell therapy will undoubtedly continue to expand in the future. The interventional and diagnostic radiologist should be familiar with the basic principles of stem cell

properties and potential therapeutic applications. Radiology may play a pivotal role in stem cell delivery, stem cell engraftment monitoring through imaging, and, potentially, improvement of engraftment conditions with use of minimally invasive procedures, as will be shown below.

Stem cells have the ability to divide and self-renew indefinitely as well as to differentiate into one or more cell types (1). It is relevant to differentiate between the various types of stem cells—discussed below in greater detail—and to distinguish between embryonic stem cells, which are obtained from the inner cell mass of the blastocyst, and adult stem cells, which are found in adult somatic tissue. The only types of stem cell that are pluripotent (ie, may differentiate into any cell type) are embryonic stem cells (Fig 1 [2]). Embryonic stem cells subsequently develop into partially differentiated stem cells that may in turn give rise to several different cell lines, but these cells can no longer become any type of cell (ie, they are multipotent stem cells) (Fig 1 [2]). Adult stem cells are multipotent cells as well and the result of further lineage progres-

sion. They are of more limited differentiation ability and destined to develop into cells of a specific organ, tissue, or organ system with the (potential) ability to fulfill corresponding functions (Fig 1 [2]). Adult stem cells can be harvested from bone marrow, adipose tissue, and umbilical cord blood. Examples of adult stem cells are hepatic progenitor cells, which have the capability to differentiate into hepatocytes, or type II pneumocytes, which have the potential to differentiate into parenchymal lung cells (Fig 1 [2]). It is notable that ethical concerns surrounding stem cell research are mostly related to embryonic stem cells. A comprehensive ethical discussion about the use of human embryonic stem cells is clearly beyond the scope of this article. Briefly, however, the pursuit of the undisputedly ethical end of alleviation or cure of human suffering conflicts in this case with the means of destruction of embryonic tissue—an act that many regard as an unacceptable violation of respect for human life.

Adult stem cells, such as those derived from bone marrow (subdivided into hematopoietic and mesenchymal/marrow stromal cells), are classically

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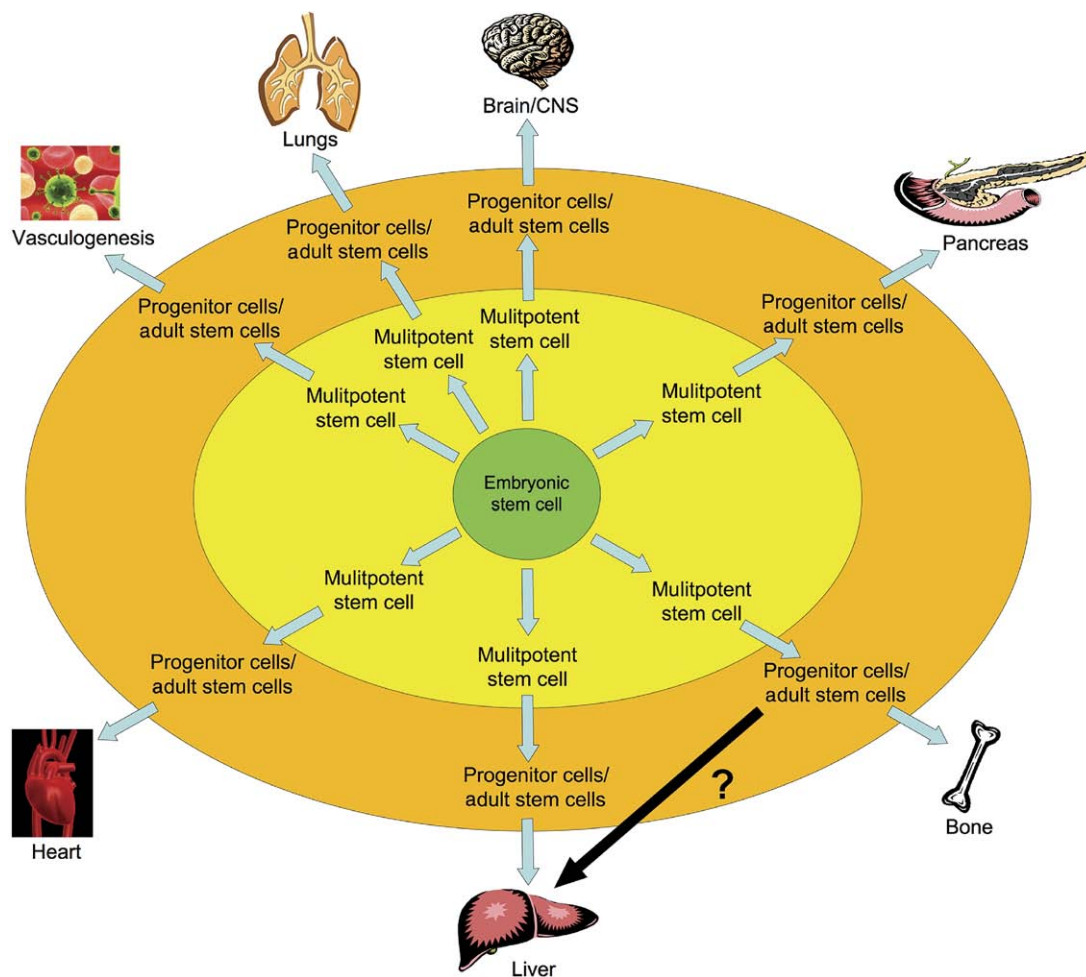


Figure 1. Simplified representation of stem cell differentiation. Cell differentiation progresses from the center toward the periphery. The green center includes blastocyst and pluripotent cells. The yellow area includes cells developed from embryonic stem cells, which have a capacity to differentiate into many different cell types but are more restricted than embryonic stem cells. Cells in the gold area are the result of further lineage progression, generally destined to develop into a certain cell type such as hepatic progenitor cells into hepatocytes or type II pneumocytes into parenchymal lung cells. ? denotes potential crossing of individual cell populations to differentiate into specialized cells of organs derived from other germ layers; this crossing ability is somewhat controversial but can likely occur under certain circumstances. Adapted and reprinted from *Biochimica et Biophysica Acta*, 1782, M. Oertel and D. Shafritz, Stem cells, cell transplantation and liver repopulation, 61–74, 2008, with permission from Elsevier.

harvested as autologous cells and are generally free of ethical controversy. With recent demonstration of the ability to reprogram human somatic cells to cells with embryonic stem cell type differentiation potential with transduction of certain defined transcription factors, ethical concerns can be expected to lessen and opportunities to use stem cells for therapy will likely expand in the future (3,4).

Although stem cells have received much research attention recently, the first successful transplantation of bone marrow–derived hematopoietic stem cells dates back to the late 1960s. Translational research later

led to Nobel Prize recognition of Joseph E. Murray's and E. Donnall Thomas' work in managing transplant rejection and graft-versus-host reaction as well as performance of transplantation of hematopoietic stem cells, respectively. Autologous or allogeneic hematopoietic stem cell transplantation is now a routine procedure and is successfully used clinically for the treatment of diseases such as lymphoma, multiple myeloma, leukemia, neuroblastoma, germ cell tumors, certain types of anemias such as sickle cell disease or aplastic anemia, autoimmune disorders such as systemic lupus erythem-

atosus, or amyloidosis as well as other blood dyscrasias. It is performed 30,000–40,000 times each year, increasing in use annually, and there are more than 20,000 individuals who have survived at least 5 years after hematopoietic stem cell transplantation (5).

However, not all forms of utilization of stem cells in clinical care have endured. Specifically, stem cell administration to overcome bone marrow toxicity from high-dose chemotherapy as practiced in the early 1990s has been largely abandoned due to short periods of therapeutic response and high mortality rates (6,7). More recent experi-

ments have focused on the use of stem cells for the therapy of different organs and organ systems beyond blood dyscrasias and autoimmune disorders (5). Multiple studies have shown the ability of stem cells of various origins to differentiate into specialized, fully functional parenchymal cells both in vivo and in vitro (8–12).

The general capacity of stem cells to repair damaged tissue and restore function that would otherwise be lost irreversibly has been demonstrated numerous times and has fueled recent research efforts to use undifferentiated living cells to maintain, improve, or recover organ function in lieu of organ transplantation or while lacking other treatment options. For instance, stem cells of various origins have successfully been used to alleviate pulmonary hypertension in dogs, restore biochemical function of the liver in an animal model of tyrosinemia 1, or regenerate axons through chronically denervated peripheral nerves (8,9,13).

General additional areas of research efforts include the use of stem cells for the treatment of degenerative neurologic disorders such as Parkinson and Alzheimer disease and motor neuron disorders, stroke, multiple sclerosis (MS), and acute injury of the spine (14–24). Stem cell utilization has been considered for musculoskeletal regeneration such as the repair of nonhealing fractures and rebuilding of degenerated cartilage or tendons (25–28). In addition, stem cell treatment of myocardial infarction and heart failure to improve cardiac function and performance has been explored (29,30). Stem cell-based alleviation or correction of liver disease has been sought (2,9,10,31–35). Stem cell potential is being tested to treat diabetes (36–45). Additionally, stem cell therapy has shown promise in promoting wound healing and improving perfusion in the setting of limb ischemia (46–49). Furthermore, stem cell administration has been contemplated to alleviate pulmonary disorders such as chronic obstructive pulmonary disease (50–53). The treatment of miscellaneous disorders such as scleroderma, retina degeneration, and inner ear as well as renal disorders has also been researched (54–57).

The current state of knowledge and type and extent of available data vary considerably in these mentioned fields of clinical and experimental stem cell applications and across specific entities. Although hu-

man clinical trials are occurring or are under way in some areas, only preliminary experimental data that oftentimes have been obtained from animals are currently available in others. For instance, stem cell therapy has been routinely used for some time in the treatment of leukemia and lymphoma (58) and is in the early stages of clinical investigation for the treatment of cardiac disease and diabetes mellitus (59,60) as well as certain neurologic disorders such as MS and stroke (22,61), whereas it has not (yet) undergone the translation from bench to bedside application for some other entities and organ systems such as certain lung diseases (52). Consequently, stem cell therapy may soon become routine clinical reality in some areas, whereas only hope and promise currently exist in others. As will be shown, a plethora of experimental data have produced partially inconsistent results, most of which have been attributed to methodologic differences such as variations in stem cell delivery route and stem cell administration timing. In fact, it appears that successes in stem cell therapy mandate profound knowledge of stem cell properties and harvesting, cell trafficking, and engraftment bed receptiveness as well as cell engraftment efficiency and subsequent engraftment monitoring. On the basis of this type of knowledge, it may be possible to properly select stem cell type, administration timing and delivery route for specific disease entities, anatomic areas, and physiologic circumstances to accomplish the distinction between generalized unsubstantiated claims pertaining to stem cell and reproducible experimental and clinical therapeutic successes.

A very interesting yet poorly developed area of stem cell research is the concept of utilization of stem cells as vehicles for gene and drug delivery. This area holds great promise because viable cells that are administered have the properties to adjust, multiply, migrate, and communicate with adjacent cells. For instance, Sha et al have shown that injected mouse neural precursor cells had the ability to migrate to the contralateral brain hemisphere and deliver cytotoxic tumor therapy to glioma foci, thereby reducing tumor growth (62,63).

ROLE OF THE INTERVENTIONAL AND DIAGNOSTIC RADIOLOGIST

One may wonder what role the interventional radiologist and/or imager may have in research and clinical application of stem cell therapy. It seems prudent for all radiology professionals to approach stem cell research not in an attempt to capture a piece of the pie of an emerging field of promising research. Instead, radiologists should determine their respective roles as the most qualified expert contributor for certain steps in the process of stem cell research and clinical application of stem cell therapy. Even though stem cell research efforts have been scarce in all radiology, there are indeed such roles in this research field that is rapidly progressing and will inevitably further implement itself into clinical practice and patient care. Successful stem cell transplantation generally consists of adequate stem cell harvesting, trafficking to the desired target area, full stem cell differentiation, and significant—and ideally sustainable—contribution to organ function. The stem cell trafficking process has been subdivided into cell homing, that is, directed blood dispersion of stem cells, and interstitial migration, which generally occurs within a confined territory within a given organ (63). Clearly, the interventionalist should have a pivotal role in targeted stem cell delivery to certain organs such as the pancreas, liver, or kidney. The interventionalist's participation may overcome some of the challenges that are currently associated with stem cell homing. In particular, selective and direct stem cell release to the target organ by means of transcatheter intraarterial delivery may overcome potential mechanical barriers caused by liver cirrhosis and fibrosis that have been postulated to exist for nonselective stem cell delivery techniques (64). In this context, recipient organ perfusion with donor cells "via a radiologically placed catheter" has been recommended by a prominent stem cell expert (64). Indeed, in a recent clinical study, hepatic regeneration could be enhanced with portal venous infusion of bone marrow-derived stem cells into portal vein branches before portal vein embolization and partial hepatectomy (65).

As mentioned, some discrepancies of experimental results of significant

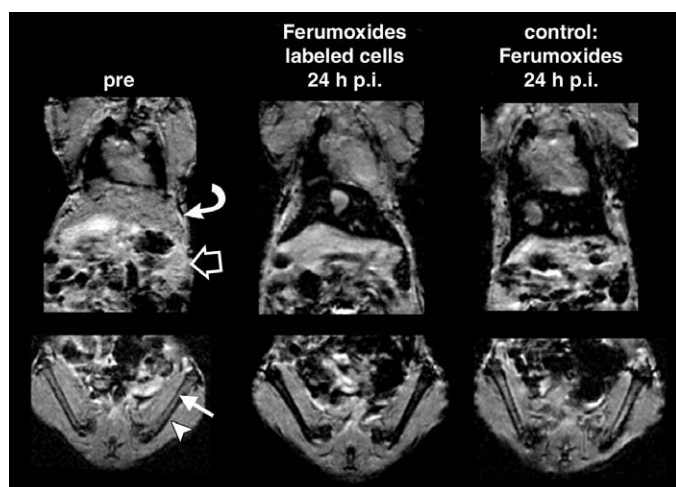


Figure 2. T2*-weighted MR images of one Balb/c mouse before injection, one Balb/c mouse 24 hours after injection (*p.i.*) of 3×10^7 ferumoxide-labeled progenitor cells, and one Balb/c mouse 24 hours after injection of ferumoxide (without cells). The labeled cells distribute differently than does the pure contrast medium. The amount of administered iron was 20 μg through injection of iron oxide-labeled cells and 25 μg through injection of the pure contrast medium (the latter applies to the usual clinical dose). MR images of the body show the liver with the left hepatic lobe extending across the midline (curved solid arrow) and spleen (open arrow). Below that, MR images were reconstructed along the long axes of the femora, in which the corticalis (arrowhead) and the bone marrow (straight solid arrow) can be clearly delineated. The ferumoxide-labeled cells caused a marked decrease in signal intensity in the liver, spleen, and bone marrow, whereas injection of the pure contrast medium caused visible signal intensity changes in the liver and spleen but not the bone marrow. Reprinted from *Radiology*, 234, H. Daldrup-Link, M. Rudelius, G. Piontek et al, Migration of iron oxide-labeled human hematopoietic progenitor cells in a mouse model: in vivo monitoring with 1.5-T MR imaging equipment, 197–205, 2005, with permission from Radiological Society of North America.

differentiation of administered stem cells have occurred and have partly been attributed to a nonselective route of stem cell administration (systemic vs transportal), further underscoring the relevance an interventionist could have in stem cell delivery (66–68).

An additional relevant parameter with regard to stem cell engraftment success is the target area receptiveness for stem cells after their respective administration. Depending on the type of target organ and stem cell type that is used, infliction of acute organ injury is known to be necessary for cell trafficking and differentiation, the former of which is triggered by the release of certain key cytokines (31,32,35). Interestingly, substantial mobilization of bone marrow-derived stem cells has been demonstrated after myocardial infarction in humans and liver injury in rodents, which has been interpreted as persistence of a more primitive self-repair mechanism of viable organisms

during moments of injurious stress to certain organs (69,70).

In the setting of acute organ injury, stem cell trafficking could likely be further enhanced by optimizing timing of stem cell administration, which likely occurs if it is synchronized with peak levels of certain key cytokines. However, the key cytokines that regulate stem cell trafficking for various target organs must also be further investigated in terms of their respective release mechanisms and role as well as their respective interaction with each other. A description and analysis of the various key cytokines is clearly beyond the scope of this article but has been described elsewhere (63,69). The types and respective roles of certain cytokines differ across different organs, and an excellent example of effects on expansion, proliferation, and/or mitogenesis of specific key cytokines on oval cells during activation of stem cells in hepatic disease is given by Bird et al (69).

A role for imaging of stem cell therapy is likewise developing. Numerous

studies have been performed for the purpose of stem cell tracking and have shown that superparamagnetic iron oxide-labeled stem cells can be visualized with magnetic resonance (MR) imaging by causing signal drop-out on T2*-weighted sequences and strong effects at R2* mapping, respectively (71–79) (Figs 2, 3 [79,80]). This tracking ability has been exploited to demonstrate glomerular homing of magnetically labeled stem cells in a rat model of nephropathy, in vivo imaging of magnetically labeled stem cells in the liver, and mapping and monitoring of injected stem cells in the setting of stroke and brain as well as spinal cord injury. In the context of these studies, the capacity of stem cells to migrate or home to the area of damaged renal and brain tissue has been demonstrated. Future study of MR imaging-based labeled stem cell tracking should further advance the knowledge of in vivo distribution, migration, and engraftment of stem cells and result in clinical monitoring of stem cell therapy in certain anatomic areas.

Modalities that have been used for cardiac stem cell imaging include optical imaging, single-photon emission computed tomography (SPECT), positron emission tomography (PET), MR imaging, and multimodality imaging, that is, the use of multimodality contrast media. Optical imaging encompasses bioluminescent and fluorescent techniques. With bioluminescence, light is generated by the enzyme luciferase (81–83). This technique is limited by the facts that only visible light is generated (400–700 nm), luciferase genes and substrates are associated with very high absorption and scatter, and no animals larger than rats have been imaged with this methodology with satisfactory accuracy (84,85). In fluorescence imaging, cells are labeled with organic (green fluorescence protein, small molecule polymethines) or organic/inorganic hybrid (quantum dots) agents for in vivo detection (86). Limitations with this technique include spatial limitations of imaging capabilities to a tissue depth of 4–10 cm, dilution of contrast signal due to subsequent cell divisions, and possible stem cell uptake by macrophages after stem cell death (87).

Stem cell imaging with SPECT is performed by detecting high-energy γ -rays emitted by technetium 99m ($^{99\text{Tc}}$), indium 111, or iodine 123, which are introduced by direct radiometal

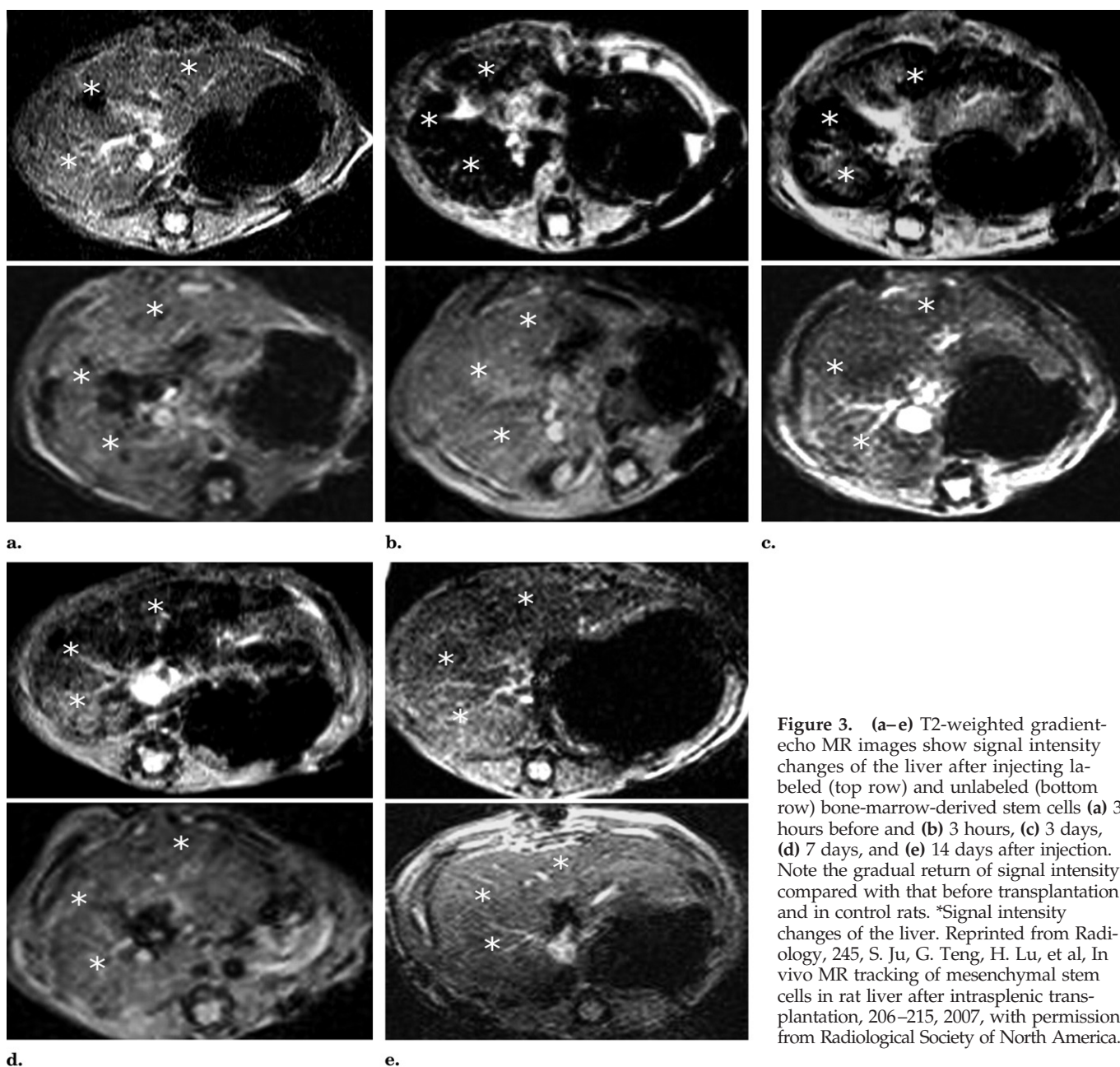


Figure 3. (a–e) T2-weighted gradient-echo MR images show signal intensity changes of the liver after injecting labeled (top row) and unlabeled (bottom row) bone-marrow-derived stem cells (a) 3 hours before and (b) 3 hours, (c) 3 days, (d) 7 days, and (e) 14 days after injection. Note the gradual return of signal intensity compared with that before transplantation and in control rats. *Signal intensity changes of the liver. Reprinted from Radiology, 245, S. Ju, G. Teng, H. Lu, et al, In vivo MR tracking of mesenchymal stem cells in rat liver after intrasplenic transplantation, 206–215, 2007, with permission from Radiological Society of North America.

loading, enzymatic conversion with retention of a radioactive substrate, or receptor-mediated binding (88–93). Although visualization of ^{99}Tc radiometal-loaded stem cells has been accomplished up to 4 hours after cell infusion in a rat model of myocardial infarction, limitations of this technique include the trade-off between half-life and long-term exposure to ionizing radiation as well as potential of the radiometal transfer to non-stem cells (87,90).

Enzymatic conversion and retention implies enzyme introduction through a transgene. This technique has been used for SPECT as well as PET imaging and is characterized by the ability of indefinite in vivo stem cell monitoring without effects of signal dilution by stem cell division but requires expression of a unique stable receptor (93,94). The sensitivity of SPECT is high when compared with that of optical imaging and MR imaging. Limited experience with

PET has shown only a fraction of stem cells (1.3%–2.6%) around the infarction border within 1–1½ hours after intracoronary injection. Attempts have been made to overcome the limited half-life of fluorine (110 minutes) by integrating a mutant herpes simplex type 1 thymidine kinase into stem cells followed by periodic intravenous injection of thymidine kinase substrate and serial image acquisition over time. This technique, however, is hampered by

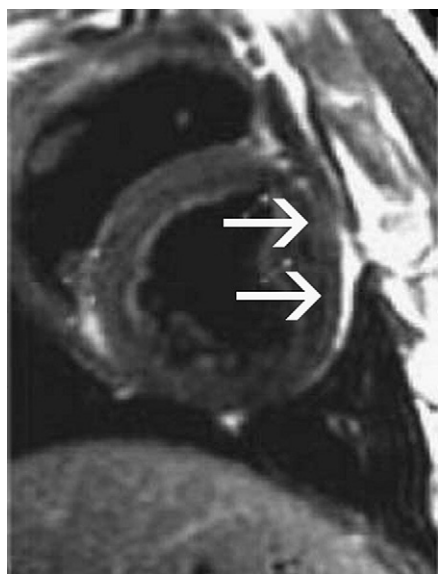


Figure 4. T2-weighted MR image of autologous pig bone marrow mesenchymal stem cells labeled with Feridex (Bayer, Leverkusen, Germany) (arrows) and injected in the anterior left ventricular wall. Image was obtained 4 weeks after injection. Reprinted from *Trends in Cardiovascular Medicine*, 15, E. Chemaly, R. Yoneyama, J. Frangioni, and R. Hajjar, Tracking stem cells in the cardiovascular system, 297–302, 2005, with permission from Elsevier.

stem cell radiation and the necessity of genetic stem cell manipulation.

MR imaging is the most validated modality for cardiac stem cell tracking as well and has been applied in T2/T2* technique after stem cell labeling with superparamagnetic iron oxide (SPIO) particles (**Fig 4**) (95–97). In cardiac imaging, SPIO-labeled mesenchymal stem cells have generated optimal imaging results up to 8 weeks after delivery following myocardial infarction in the swine model (96,97). Limitations of SPIO labeling include SPIO-induced artifact, potential false-positive results caused by contrast medium uptake of macrophages after stem cell death, and recently alleged SPIO-related stem cell differentiation inhibitions (87).

Multimodality imaging is an attempt to overcome limitations of individual imaging techniques through the use of multimodality contrast media. In this context, optical/MR imaging agents have been developed that use visible wavelengths in conjunction with gadolinium 3+ chelators conjugated to dextran as well as nanoparticles that are simultaneously usable as

MR imaging, ultrasonographic, and fluorescence contrast media (98,99).

On a more cautionary note, a separate but also noteworthy area of stem cell research is the identification and recognition of cancer stem cells. The cancer stem cell paradigm postulates that a minority of cancer stem cells are tumorigenic and give rise to tumor relapse, metastasis, and de novo tumor formation whereas the more differentiated tumor bulk is nontumorigenic. Indeed, cancer stem cells have been found in malignancies of the breast, brain, colon, pancreas, and liver. In fact, α -fetoprotein, a well known and clinically routinely used marker for hepatocellular carcinoma, is a marker of fetal hepatocytes (69,100–103). Even though the existence of cancer stem cells offers great potential for more effective approaches to cancer therapy, it may also harbor substantial risks for the use of stem cells for the purpose of tissue engineering, particularly in the setting of benign disease. It is currently uncertain whether cancer stem cells originate from stem cells that are devoid of the regulation of proliferation or whether they arise from more differentiated progenitor cells that have transformed to self-renewing cells. The answer to this question is crucial to the safe use of stem cells, and the phenomenon of carcinogenesis of stem cells warrants great caution in the pursuit of therapeutic stem cell use. In addition, careful follow-up exclusion of any potential unwarranted development of malignancy along with every successful accomplishment of stem cell–induced tissue engineering is mandatory.

CURRENT STATUS OF KNOWLEDGE: SUCCESSES AND LIMITATIONS FOR SPECIFIC ORGANS AND DISEASE ENTITIES

Among the many areas of active stem cell research, the current status of knowledge is exemplified by a description of the application of stem cell research for liver, cardiac, and neurologic diseases/disorders and the pursuit of primary therapy of diabetes mellitus (ie, restoration or improvement of endocrine pancreatic function). In these areas, the therapeutic use of stem cells is most illustrative,

most advanced, and/or most relevant for the diagnostic and/or interventional radiologist.

Stem Cells for Liver Disease

The liver possesses tremendous regenerative potential, and mature hepatocytes in transgenic mice may divide at least 69 times (104,105). Hence, hepatocytes have a property similar to that of stem cells, although they do not share the characteristic of stem cell immortality. However, the remarkable regenerative potential of the liver may be blocked or insufficient in settings of intrinsic liver disease, in which case stem cell–based liver regeneration is a promising treatment alternative to liver transplantation because the latter is substantially morbid, is costly, is limited by inadequate donor supply, and necessitates life-long immunosuppression. Stem cell–based therapy for metabolic liver disease has been the focus of much attention, particularly since Lagasse et al (9) restored the biochemical function of the liver in an animal model of tyrosinemia. This therapeutic success had been accomplished with hematopoietic stem cells by Jang et al (106), who found hematopoietic stem cell conversion into liver cells within days. However, some experimental failures of significant hepatocytic differentiation of administered bone marrow–derived stem cells have also occurred. Although this has been attributed to inadequate timing of liver injury or a suboptimal route of stem cell administration (systemic vs portal), the potential for hepatic differentiation of hematopoietic stem cells has been generally questioned by some investigators (66–68,107).

Embryonic stem cells have the capacity to differentiate into hepatocytes in vitro and have been successfully transplanted into the acutely injured liver, leading to functional recovery (10,32). However, the use of embryonic stem cells generates ethical concerns and implies the risk of carcinogenicity. Last, liver progenitor cells may be considered for cell transplantation. Four types of progenitor cells have been described: oval cells, small hepatocytes, liver epithelial cells, and mesenchymal-like cells (108). Oval cells originate from the biliary tree after injury but have also been found in normal liver, are named after their shape in rodents, and have bipotent differentiation

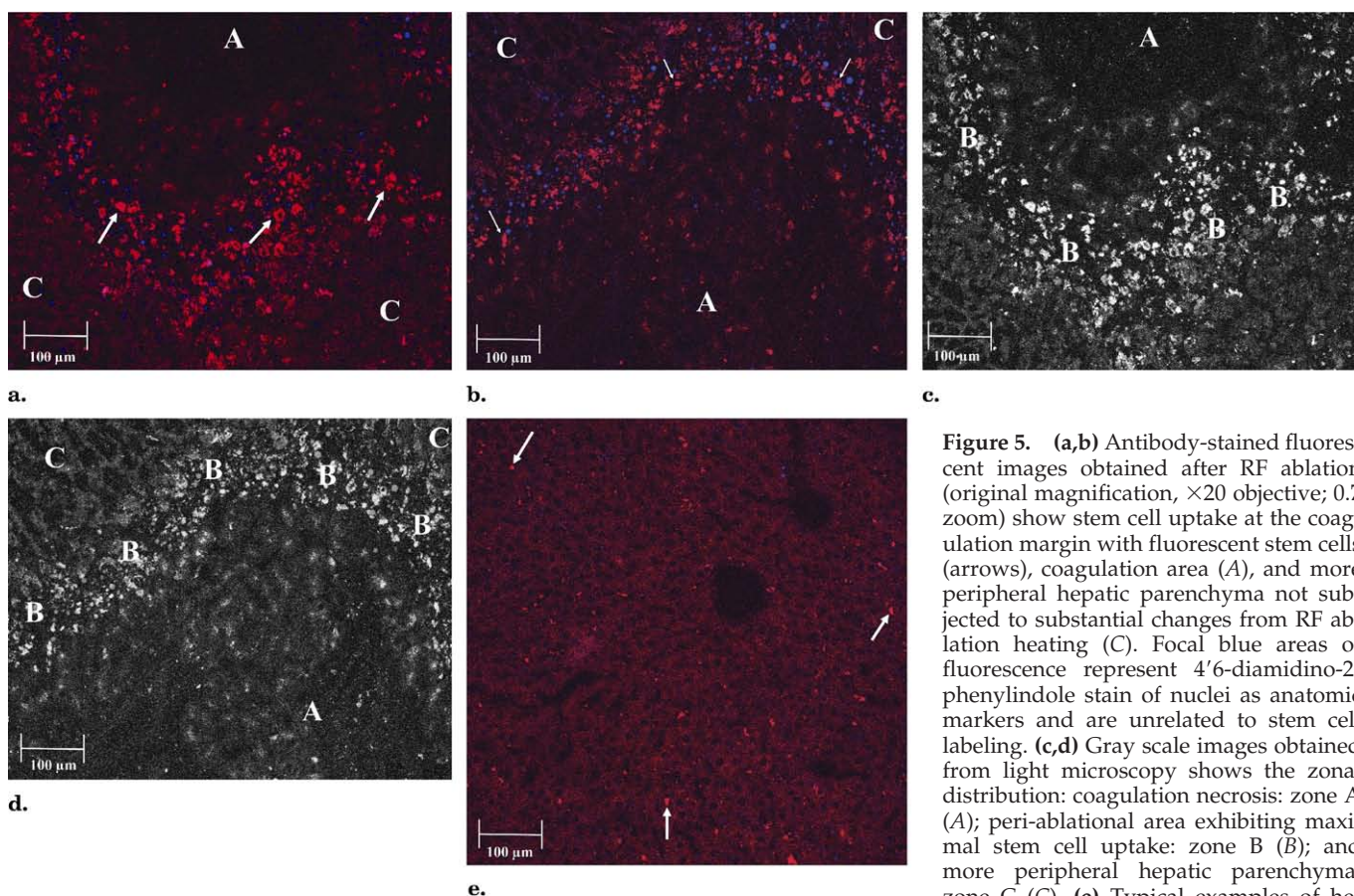


Figure 5. (a,b) Antibody-stained fluorescent images obtained after RF ablation (original magnification, $\times 20$ objective; 0.7 zoom) show stem cell uptake at the coagulation margin with fluorescent stem cells (arrows), coagulation area (A), and more peripheral hepatic parenchyma not subjected to substantial changes from RF ablation heating (C). Focal blue areas of fluorescence represent 4'6-diamidino-2-phenylindole stain of nuclei as anatomic markers and are unrelated to stem cell labeling. (c,d) Gray scale images obtained from light microscopy shows the zonal distribution: coagulation necrosis: zone A (A); peri-ablational area exhibiting maximal stem cell uptake: zone B (B); and more peripheral hepatic parenchyma: zone C (C). (e) Typical examples of hepatic control tissue obtained from the same animal as in (a) and (b) (magnification, $\times 20$ objective; 0.7 zoom). Fluorescent stem cells (arrows) are identified in a random pattern and are much less concentrated than that seen around an area of RF ablation (cf a,b). Focal blue areas of fluorescence represent 4'6-diamidino-2-phenylindole stain of nuclei as anatomic markers and are unrelated to stem cell labeling. Images are from the same animal but from different lobes: radiofrequency ablation area, right hepatic lobe; and control tissue, left hepatic lobe. Reprinted from *Journal of Vascular and Interventional Radiology*, 20, N. Boris, E. Mostafa, M. Pawel, et al, The effect of hepatic radiofrequency ablation on stem cell trafficking in the rat model, 640–647, 2009, with permission from Elsevier and Society of Interventional Radiology.

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potential into liver as well as biliary cells. Liver epithelial cells have similar differentiation characteristics as oval cells and are likewise found in healthy liver (109). Mesenchymal-like cells have broader differentiation potential, exhibit high levels of proliferation, and have been isolated from adult liver (110). Small hepatocytes have been found in healthy adult liver as well and have hepatocyte differentiation as well as high proliferation potential (111).

Interestingly, the presence of acute liver injury is known to greatly promote stem cell differentiation and engraftment within the liver. This finding, in conjunction with the dual blood supply and the substantial tolerance to injury of this organ, makes the liver an ideal target for stem cell delivery technique, stem cell trafficking, and stem cell en-

graftment research investigations to which the interventional radiologist could greatly contribute. Hence, stem cell trafficking to an injurious site has been exploited by creating target organ injury models, such as carbon tetrachloride injection or performance of partial hepatectomy in rodents (10,33,112). Partial hepatectomy, however, subtracts from the overall liver volume and is highly invasive and carbon tetrachloride injection is difficult to control, is carcinogenic, and has no therapeutic application. In pursuit of a more controllable, minimally invasive, clinically well-established and repeatable technique of causing liver injury, radiofrequency (RF) ablation has been performed and coupled with subsequent stem cell injection, resulting in proof of the concept that stem cells are trafficked to the RF

ablation area in significantly higher numbers than to the remaining liver—specifically to the peri-ablational margin (Fig 5). Hence, the interventionalist may also be able to enhance stem cell trafficking through performance of minimally invasive procedures in addition to assuming a pivotal role in targeted stem cell delivery by stem cell delivery via the transarterial route (113).

Cardiovascular Applications

Cardiac stem cell research in recent years has focused on bone marrow-derived and embryonic stem cells and has been driven by attempts to address the unmet clinical need to treat myocardial infarction and heart failure (114). Initial studies had indeed demonstrated transdifferentiation of bone marrow

stem cells into cardiomyocytes and restoration of cardiac function in mice after the occurrence of myocardial infarction (115). However, more recently bone marrow stem cell-to-host-cardiomyocyte fusion rather than stem cell differentiation was found to be the underlying engraftment mechanism, and recent randomized controlled clinical trials have failed to show significant increases in left ventricular ejection fraction after the injection of autologous bone marrow-derived stem cells (116–119). Nevertheless, infarct remodeling and exercise capacity showed improvement after bone marrow stem cell therapy for the first 4–6 months, with patients with the largest infarcts benefiting the most (120).

Embryonic stem cells, which can be transdifferentiated into cardiomyocytes *in vitro*, have been successfully used to restore atrioventricular conduction in pigs and guinea pigs with atrioventricular blocks (121,122). Although evaluation for the optimal stem cell delivery route is ongoing, current concepts favor combined tissue cell intramyocardial stem cell injection over single cell line suspension intracoronary injection because the latter has been associated with the occurrence of microinfarctions (123).

Peripheral Arterial Disease

Peripheral arterial disease of all stages has an estimated prevalence of 4.2%–35% (124). It progresses to critical limb ischemia in 4.3%–9.6% of cases, conveying quality of life indexes similar to those of terminal cancer patients and the eventual undesirable outcome of amputation (124,125). Diabetic patients are at highest risk for the development of gangrene and necessity of amputation (126). About 40% of patients with critical limb ischemia are not candidates for revascularization procedures, performance of amputation portends an even worse prognosis where it appears indicated, and no effective pharmacologic therapy is available for these patients (127,128). At the same time, bone marrow-derived endothelial progenitor cells have shown promise in providing neovascularization and have been found to contribute to wound healing (46).

Vascular cells develop from embryonic stem cells through mesodermal differentiation into cardiovascular progenitor cells or hemangioblasts (11).

Cardiovascular progenitor cells become cardiomyocytes or vascular progenitor cells, the latter of which can progress into smooth muscle cells (pericytes) or endothelial progenitor cells. Hemangioblasts may mature to hematopoietic or endothelial progenitor cells (11). Blood flow that is required for wound healing may be derived from angiogenesis, a process of wound-adjacent resident endothelial cell migration followed by neovessel creation that is accomplished in concert with mature resident stromal cells (47). Angiogenesis occurs naturally but is often insufficient to allow for wound healing. Vasculogenesis, conversely, is defined as *de novo* phenomena initiated by progenitor stem cells giving rise to a surrogate vascular network (47). Although it was previously believed that vasculogenesis only occurs during embryonic development, endothelial progenitor cells have subsequently been found in peripheral blood of adults and harvested from peripheral blood or bone marrow for performance of various clinical trials with the intent to use endothelial progenitor cells for the treatment of critical limb ischemia (47). The first report in 2002 (129) demonstrated the safety and efficacy of intramuscular injection of bone marrow cells in patients with chronic limb ischemia and significantly improved ankle-brachial pressure indexes, transcutaneous oxygen pressures, and pain-free walking times while reducing rest pain at 4- and 24-week follow-up. Multiple following studies confirmed these initial results, with two studies also demonstrating improvement of endothelial function and increased blood perfusion as assessed with ^{99m}Tc tetrofosmin perfusion scintigraphy (130–137). Granulocyte stimulating factor, which is known to mobilize hematopoietic and endothelial progenitor cells from the bone marrow into the circulation, has been used to stimulate bone marrow-derived stem cells before harvesting and therapeutic administration of these cells and likewise found improvement in critical limb ischemia and peripheral vascular parameters with additional improvement of glucose metabolism in one study (138). Currently reported results for stem cell utilization for peripheral vascular disease are limited by the lack of randomized, controlled studies as well as longer-term clinical follow-up. Although the vast majority of investigators have opted for the intramuscular

delivery route, the most efficient way of cell delivery is uncertain as is the optimal dose and the cell type and cell (sub) population that should be used for the treatment of critical limb ischemia. Hence, these aspects should be the focus of future investigations.

Neurologic Diseases

The *in vivo* neural stem and progenitor cell has been identified as an astroglial cell (139–141). However, mesenchymal stem cells, which are grown on soft matrices, have also been found to give rise to neuronal cells. This finding has caused a paradigm shift from a dogma of cell lineage restriction with regard to further cell differentiation to one of potential crossing of individual cell populations to other germ layers. More important, bone marrow-derived stem cells have created neurons *in vivo* (142,143). Consequently, granulocyte colony stimulating factor has been used to stimulate the release of endogenous bone marrow stem cells for angio- and neurogenesis and, in addition, reduces neuronal apoptosis and stimulates neural progenitor cells in the treatment of acute stroke (144,145). Engraftment efficiency comparisons and potential complementary effects of catheter-directed selective delivery of exogenous stem cells and endogenous stem cell mobilization would be one ideal realm of research for the neurointerventionalist.

Stem cell therapy has been applied to a variety of neurologic disorders and entities that may be categorized as chronic degenerative (eg, Parkinson and Alzheimer disease), acute traumatic (eg, cord injury), metabolic, and autoimmune (eg, MS) disorders. Stem cell therapy of neurologic disorders will be exemplified by a discussion of stem cell treatment of stroke, Parkinson disease, and MS (21,146–148).

Stroke.—Bone marrow-derived—specifically mesenchymal stem cells—and human umbilical cord blood stem cells have been tested for potential therapeutic applications in stroke. Multiple studies have demonstrated functional outcome improvement after intravenous, intracerebral, or intraarterial stem cell application in animal experiments even 4 weeks after the ischemic insult (146–148). Notably, a neurorestorative effect through stem cell administration is accomplished by the promotion of angiogenesis, neurogenesis, and synapto-

genesis rather than transdifferentiation of stem cells to fully functional neuronal cells (149). The angiogenetic effect of stem cells is not surprising because stem cells have been identified as potent producers of vascular endothelial growth factor, a positive regulator and promoter of vessel formation (150,151). There has also been one clinical trial in which the study group received mesenchymal stem cells intravenously after middle cerebral artery infarction and showed improved functional recovery compared to a control group during 1-year follow-up (152). Human umbilical cord blood cells via the intravenous as well as intrastriatal (directly into the globus pallidus/putamen) delivery route have also been used for stroke treatment. Although results have not been entirely consistent, successes have been attributed to the immunosuppressive as well as angiogenetic effects of the administered cells (61,153–155). As in other areas of stem cell treatment, the timing of cell administration is likely quintessential because the later stage of stroke evolution results in an intraaxial cavity that is poorly accessible for exogenous cells. Although an approach of intracavity stem cell transplantation via a biodegradable scaffold has been developed, stem cell delivery to the infarcted territory is likely more effective if performed before the late stage of infarct evolution. Nevertheless, on the basis of current knowledge, stem cell therapy has the potential to expand the current 3–6 hour postevent treatment window (for thrombolytic therapy) to several weeks. Although some of the initial experimental results for stroke treatment with stem cells are promising, the notion that stem cells may restore complex functional anatomy seems to be an overreaching expectation at the current time. Rather, stem cell therapy may be a functional recovery facilitator through angiogenesis, neurogenesis, and synaptogenesis promotion.

Parkinson disease.—Parkinson disease was once believed to be an ideal target for stem cell therapy because it requires replacement of only one distinct cell population in the substantia nigra, unlike in the therapy of stroke, which is characterized by the territorial demise of numerous different types of cell populations. However, stem cell-based treatment attempts

for Parkinson disease have generated inconsistent results (156–158). Lessons learned from previous research in this area include the fact that the use of embryonic stem cells for this application may be inefficient and entail the risk of teratoma formation, an unwarranted ability of embryonic stem cells that has also been observed in other anatomic areas and seems to be site-dependent (159). Consequently, mesenchymal cells that had been cultured to assume characteristics of dopaminergic neurons have been used more successfully for Parkinson disease therapy in one study (160).

A very interesting and more elegant approach is stem cell-mediated delivery of glial cell line-derived neurotrophic factor, a protein that promotes dopaminergic neuron preservation and differentiation. This type of stem cell use as a vehicle for gene therapy has resulted in glial cell line-derived neurotrophic factor expression *in vivo* and led to prolonged neuron survival as well as functional improvement (161). Notably, intraarterial catheter-directed administration of adult stem cells into the posterior circulation has recently been presented as a beneficial and efficient way of improving symptoms in patients with Parkinson disease in a clinical trial encompassing 47 patients (162). In summary, currently available research suggest that stem cell-based therapy of Parkinson disease is most successful if cells are used that have undergone predifferentiation *in vitro* and possess dopaminergic properties, an approach that may also be most useful in many other anatomic areas. The use of genetically modified stem cells that express glial cell line-derived neurotrophic factor as vehicles for gene therapy is likewise promising and has thus far been free of associated tumor formation (161).

MS.—MS is an inflammatory autoimmune disease that may be characterized by a progressive or relapsing course. Autologous bone marrow-derived stem cells have also been found most suitable for stem cell-based therapy for this entity as well and have been most successful at an early stage of the disease and if performed as nonmyeloablative therapy. The rationale for stem cell therapy for this disease is to create an immunologically “naïve” state through autologous hematopoietic stem cells (22). A second approach that has generated promising results

has been intravenous and intraventricular administration of neural progenitor cells (24). These cells have been found to be therapeutic through anti-inflammatory effects in a pro-inflammatory environment and neuroregenerative effectiveness in a neurodegenerative environment, consistent with effective targeting of MS in its acute and chronic stages, respectively (23,163). The utility of stem cells in the treatment of MS underscores and exemplifies the potential immunosuppressive effect of stem cells in autoimmune diseases.

Cell-based Therapy for the Treatment of Diabetes Mellitus

The treatment of diabetes mellitus that is refractory to medical therapy has been a clinical challenge and resulted in the emergence of pancreas transplantation as a therapeutic option. Pancreas transplantation, however, is associated with limited donor supply and substantial morbidity and costs and requires lifelong immunosuppression. Cell-based therapy, conversely, is more elegant, more cost effective, and less invasive. The initial approach of cell-based therapy entailed harvesting of islet cells from brain-dead donors, a strategy that is still characterized by a limited donor supply. Among the various target areas and delivery routes of stem cell injection (percutaneous intrasplenic, subcapsular renal, intra-omental, subcutaneous, and into the celiac artery), intraportal venous delivery has emerged as the technically most feasible, least complicated, and most efficient way of islet cell delivery even though occurrence of bleeding complications as well as portal vein thrombosis that is apparently related to intraprocedural increases in portal venous pressure have been described (59,164). The investigative team from the University of Alberta must be credited with pioneering work of successful portal vein infusion of islet cells in patients with medically refractory type I diabetes mellitus, which effectively controlled blood glucose levels for 1 year before disease recurrence (59).

Stem cell-based therapeutic investigations have been conducted in animal experiments but have been compounded by the lack of a clearly identifiable type of pancreatic stem cell. Embryonic stem cells have been transformed into cells with β cell properties, although controversy sur-

rounds the issue as to whether these cells produce or simply absorb insulin (165–167). Nevertheless, transplanted embryonic stem cells have been successfully used to improve or cure diabetes in rodents (43–45). The ability of hematopoietic and bone marrow–derived stem cells to differentiate into functional islet cells has also been accomplished in rodents (168,169). Subcapsular renal transplantation of bone marrow–derived cells that had been differentiated toward insulin-expressing cells in vitro resulted in glucose level correction in rodents, which was reversed after removal of the grafted kidney (170,171).

Like in other organs, in vitro differentiation of toti- or pluripotent cells into endocrine pancreatic cells before transplantation seems most efficient. The intraportal venous delivery route appears to best combine engraftment efficiency with low complication rates and technical feasibility.

CONCLUSIONS AND FUTURE PROSPECTS

For many years, stem cell therapy has been accepted as the first- or front-line therapy for disease entities such as blood dyscrasias or certain autoimmune diseases. In other anatomic areas, the use of stem cells for treatment is at various stages of research and clinical application but research activity is rich in all, progress is rapid, and implementation into clinical routine will likely eventually occur in most—if not all—areas of current research. Consequently, any review about stem cell therapy tends to be outdated quickly. Nevertheless, distinct trends are identifiable at the current time that can be exploited for the future and may define the role of the interventional and diagnostic radiologist in the area of stem cell therapy. For instance, the interventional radiologist should advance knowledge of stem cell engraftment kinetics and optimize administration timing and delivery route through participation in—or primary investigation of—various research projects of tissue engineering. In vitro cell line differentiation toward the desired cell lineage before in vivo administration seems to result in most efficient stem cell engraftment in most anatomic areas.

In addition, engraftment bed fertility could be increased by the interventional radiologist by ablating and devascular-

izing dysfunctional or malignant tissue by means of RF ablation. Periablational hyperemia and/or ingrowing granulation tissue may then provide a vascular environment that allows for highly efficient stem cell trafficking to the selected target area and subsequent stem cell engraftment. In addition, the role of devascularization of dysfunctional tissue through transarterial catheter embolization could be explored. Both techniques (ie, RF ablation and transarterial catheter embolization) may be followed by subsequent targeted transarterial cell delivery to improve stem cell trafficking efficiency when compared to peripheral intravenous cell administration.

The imaging radiologist should continue to develop and refine imaging techniques that allow for stem cell tracking (eg, by means of SPIO labeling of stem cells) and take an active role in exploring occurrences and extent of stem cell engraftment, migrations, and stem cell effect on organ (system) functionality based on imaging.

In pursuit of these objectives, both the diagnostic radiologist and the interventional radiologist will be firmly integrated in this promising and still-developing field of medicine and become valuable partners for basic science researchers and clinicians alike.

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