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REVIEW TOPIC OF THE WEEK

Mechanisms Contributing to the Progression of Ischemic and Nonischemic Dilated Cardiomyopathy

Possible Modulating Effects of Paracrine Activities of Stem Cells

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ABSTRACT

Over the past 1.5 decades, numerous stem cell trials have been performed in patients with cardiovascular disease. Although encouraging outcome signals have been reported, these have been small, leading to uncertainty as to whether they will translate into significantly improved outcomes. A reassessment of the rationale for the use of stem cells in cardiovascular disease is therefore timely. Such a rationale should include analyses of why previous trials have not produced significant benefit and address whether mechanisms contributing to disease progression might benefit from known activities of stem cells. The present paper provides such a reassessment, focusing on patients with left ventricular systolic dysfunction, either nonischemic or ischemic. We conclude that many mechanisms contributing to progressive left ventricular dysfunction are matched by stem cell activities that could attenuate the myocardial effect of such mechanisms. This suggests that stem cell strategies may improve patient outcomes and justifies further testing.
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Over the past 1.5 decades, numerous stem cell trials have been performed in patients with cardiovascular disease, using both autologous and allogeneic stem cells, numerous stem cell types, and various strategies to administer the stem cells. Although many individual studies reported encouraging signals, these were all phase 1 or 2 studies with appropriately small numbers of patients, and their conclusions must therefore be considered preliminary. In an attempt to increase statistical robustness, a recent meta-analysis assessing the results of all randomized clinical trials of stem cell therapy for patients with acute myocardial infarction (AMI) was performed, demonstrating no net

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beneficial effects on outcomes, except for a small improvement in ejection fraction (1). A major review of patients with heart failure (HF) yielded similar conclusions (2).

Given these results, a reassessment of the rationale for the use of stem cells in cardiovascular disease is timely. There are powerful arguments for concluding that stem cell-based mechanisms exist that might be therapeutically efficacious, thereby making continued pursuit of stem cell strategies for treating cardiovascular disease reasonable. Such a rationale would have to include the likely mechanisms contributing to progression of the disease and the potential influences, if any, of stem cells on such mechanisms. The present paper is intended to provide such a rationale, focusing on the patient with HF and left ventricular (LV) systolic dysfunction, whether nonischemic cardiomyopathy (NICM) or ischemic cardiomyopathy (ICM).

By definition, the most conspicuous difference between ICM and NICM is the existence of atherosclerotic lesions of the epicardial coronary arteries in patients with ICM and the absence of such lesions in patients with NICM. This leads to 1 major difference in the initiation of the cardiomyopathic process and its progression to HF: most patients with ICM have had 1 or more previous clinically-recognized or clinically-silent myocardial infarctions (MIs), with the development of progressive remodeling and LV dysfunction occurring consequent either to an initial large injury to the LV or to smaller, repeated injuries occurring over time. Such a mechanism does not exist in NICM.

Despite this difference, disease progression in ICM can occur, even when the initial infarct does not result in severe LV dysfunction, and conversely, many patients with large infarcts do not develop such progression (3,4). These findings raise the possibility that a given patient may experience progressive deterioration of LV function on the basis of additional mechanisms independently of MI, which may be shared by patients with ICM and with NICM.

Another abnormality shared by both ICM and NICM patients, which might provide an important therapeutic target, is the presence of dysfunctional but viable myocardium. Patients with ICM invariably have areas of myocardial scar, usually extensive, whereas patients with NICM either do not or have it to a lesser extent. Bello et al. (5) observed, using cardiac magnetic resonance imaging, that whereas all patients with ICM had myocardial scar, only 12% of patients with NICM did so. Importantly, both ICM and NICM patients had areas of myocardial dysfunction due not to scar, but to dysfunctional *viable* myocardium (DVM).

Although more common and extensive in patients with NICM, DVM provides a potential target for therapeutic interventions in both ICM and NICM. If the dysfunctional tissue consists of viable rather than scarred myocardium, LV function can presumably be improved. **Figure 1**, adapted from Bello et al. (5), demonstrates these concepts—that LV ejection fraction can be improved, that the magnitude of improvement is related to the percent of the LV that is dysfunctional but viable, and that DVM is present in both ICM and NICM.

The concept of DVM may also help direct which patients may benefit most from stem cell therapy. As the stage of HF that may be considered too late for stem cell therapy is unclear, the presence of DVM may help guide the identification of those patients with the most potential to benefit.

The potential of any therapy, including stem cells, to improve outcomes in ICM or NICM is related not only to its effects on restoring function to DVM, but also to its capacity to improve processes that contribute to

ABBREVIATIONS AND ACRONYMS

ECM = extracellular matrix

HF = heart failure

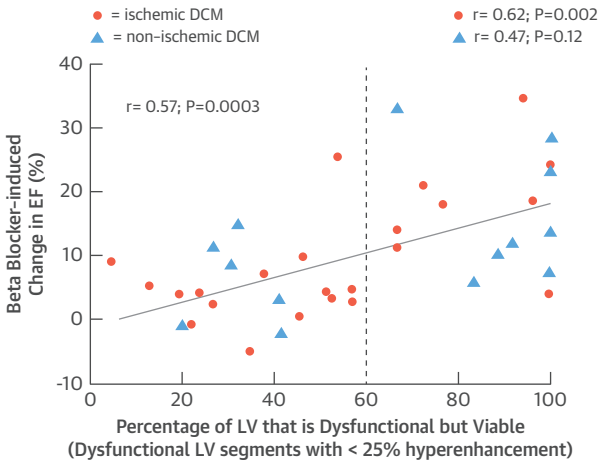
ICM = ischemic cardiomyopathy

MMP = matrix metalloproteinase

MSC = mesenchymal stem cells

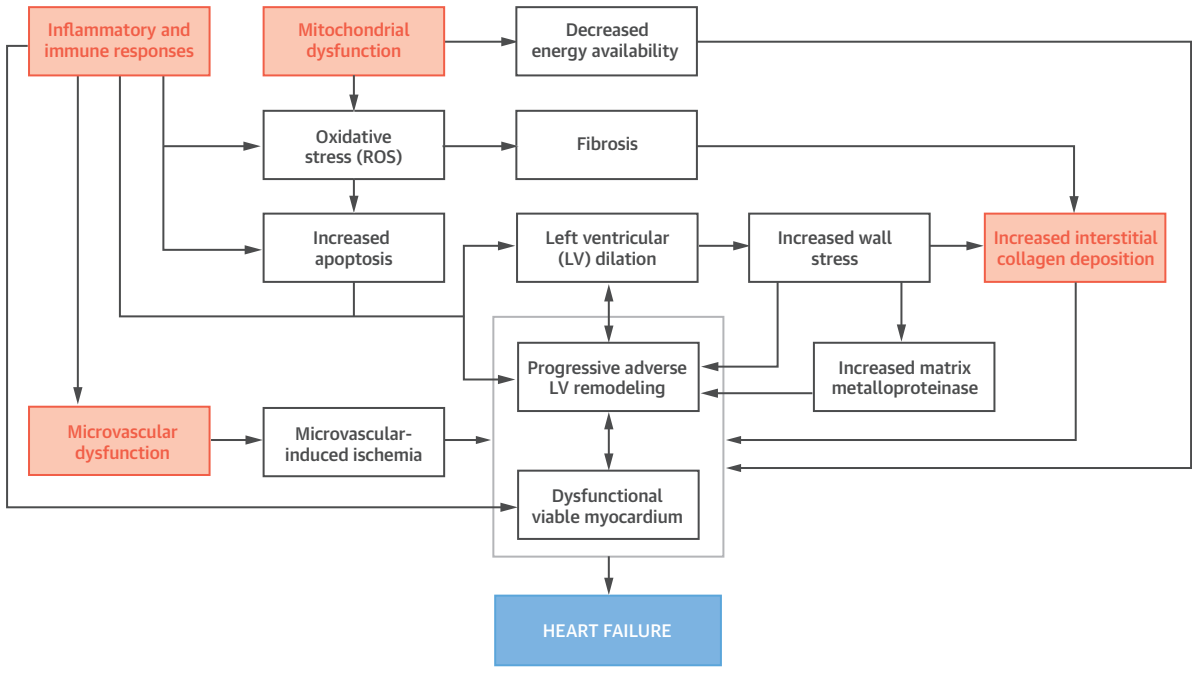
NICM = nonischemic cardiomyopathy

FIGURE 1 Ischemic or Nonischemic DCM: Identification of Dysfunctional But Viable Myocardium and Relation to Beta-Blocker–Induced Increase in EF



Dysfunctional myocardium refers to myocardium that is dysfunctional in the absence of delayed hyperenhancement by magnetic resonance imaging. The magnitude of improvement in LV EF that occurs following initiation of beta-blocker therapy is directly related to the percent of the LV that is dysfunctional, but still viable. Importantly, although compared with patients with ischemic cardiomyopathy, those with nonischemic cardiomyopathy have greater portions of the LV that are dysfunctional. However, viable, dysfunctional myocardium is present in both groups. Adapted with permission from Bello et al. (5). DCM = dilated cardiomyopathy; EF = ejection fraction; LV = left ventricle.

CENTRAL ILLUSTRATION **Mechanisms Contributing to Progression of Ischemic and Nonischemic Dilated Cardiomyopathy**



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A display of some pathways believed to contribute to the development and progression of ischemic and nonischemic cardiomyopathy, which overlap with activities exerted by different types of stem cells. **Orange font** indicates possible primary causes. LV = left ventricular; ROS = reactive oxygen species.

progressive deterioration of LV structure and function (Central Illustration). On the basis of this conceptual framework, our paper explores the potential of stem cells to exert beneficial effects in ICM and NICM by considering the overlap between pathways believed to contribute to disease progression (other than atherosclerotic disease of the large coronary arteries) and the known activities of stem cells that could favorably influence these pathways.

MECHANISMS CONTRIBUTING TO PROGRESSIVE LV DYSFUNCTION THAT MIGHT BE TARGETED BY ADMINISTRATION OF STEM CELLS

The Central Illustration displays some of the pathways, exclusive of large vessel coronary artery disease, believed to (both individually and in combination) play a role in initiation/progression of LV dysfunction and adverse LV remodeling and, ultimately, in development of HF.

INFLAMMATORY AND IMMUNE RESPONSES. Patients with both ICM and NICM exhibit persistently elevated C-reactive protein levels, consistent with a chronic inflammatory state. Elevated C-reactive protein levels were found to be independent markers of mortality (6,7). In addition, DVM has been associated with the up-regulation of other inflammatory markers, such as inducible nitric oxide synthase and tumor necrosis factor alpha (8). In patients with HF, circulating levels of T regulatory cells are reduced and their suppressive function compromised, providing further evidence that an important mechanism modulating immune and inflammatory responses is deranged in this condition (9).

A persistent inflammatory state can contribute directly not only to the development of DVM and adverse LV remodeling (10), but also to increased reactive oxygen species, apoptosis, fibrosis, and microvascular dysfunction, which further exacerbate its effects on DVM and adverse LV remodeling (11–14) (Central Illustration). It has long been established that inflammation plays a critical role in progression of the *arterial wall component* of atherosclerotic heart disease (15). However, the deleterious effects of the inflammatory and immune systems are now recognized

as also contributing to adverse LV remodeling and progressive LV dysfunction following AMI (16,17). The potential importance of immune and inflammatory processes as key targets for disease modulation is evidenced by studies in progress testing whether interventions that modulate these processes favorably influence disease outcomes (18,19).

On the basis of these considerations, we and others propose to extend the concept of the causal role of inflammatory and immune pathways beyond their influences on plaques involving the large coronary arteries or induction of these pathways by AMI—that is, we hypothesize that a patient's particular immune and inflammatory responses are potential critical players in the progression to HF, even in the absence of large vessel disease. Such pathways, we propose, would be operative in patients with ICM, as well as in patients with NICM. We further suggest that the causal role played by such responses will vary from patient to patient, depending on multiple factors including genetic, epigenetic, and environmental.

MICROVASCULAR DYSFUNCTION. Animal models and human studies have shown microvascular dysfunction (MVD) in subjects without demonstrable cardiac disease and in patients with both NICM and ICM (20–25). Abnormalities include decreased capillary density, impaired vasculogenesis, and impaired endothelial function leading to a reduced vasodilator response to increased oxygen demand (24,26,27). In addition, Roura et al. (20) found defective vascularization in NICM patients that included decreased epicardial microvascular density. These abnormalities could be exacerbated by inflammatory and immune responses. Moreover, through myocardial ischemia at the microvascular level, MVD could lead either to DVM or to myocyte cell death and to progressive LV dysfunction and adverse LV remodeling (**Central Illustration**). In this regard, the magnitude of MVD has been correlated with the risk of adverse outcome in patients with NICM (24).

OXIDATIVE STRESS. Patients with ICM and NICM exhibit abnormalities in mitochondrial function, leading to energy deprivation and increased oxidative stress (6–8,24,28–33). As depicted in the **Central Illustration**, these abnormalities, through complex interactions with other pathways, likely contribute to progression of DVM and adverse LV remodeling, thereby playing a role in the development of HF.

EXTRACELLULAR MATRIX PRODUCTION AND INCREASED APOPTOSIS. Myocardial remodeling involves a combination of changes in cardiac structure and function, including increased apoptosis (6,34) and dysfunctional collagen homeostasis. These may, in part, be due to ischemia-induced cardiomyocyte degeneration and replacement of sarcomeres with glycogen or mitochondria deposits (35). The underlying biology of dysfunctional collagen homeostasis in ICM and NICM, including extracellular matrix (ECM) expansion and interstitial fibrosis, is poorly understood (36). It is unclear if myocyte loss occurs first followed by replacement fibrosis, or if primary fibroblast activation and development of fibrosis then lead to myocyte loss through myocyte compression and compromised blood flow to myocytes (36). Further contributing to the abnormal collagen homeostasis is increased expression of matrix metalloproteinase activity, leading to adverse LV remodeling (37).

PARACRINE STEM CELL ACTIVITIES THAT COULD FAVORABLY INFLUENCE MECHANISMS OPERATIVE IN ICM AND NICM

Many types of adult stem cells have been used in clinical trials (2). In vitro and in vivo studies have defined detailed characteristics of these cells, which differ in their degree of differentiation, the patterns of the molecules they secrete, the tissue niche from which they derive, their potential for eliciting a host immune response, and their ability to engraft into the target tissue.

Although some adult stem cells differentiate to express markers characteristic of cardiomyocytes, endothelial cells, and smooth muscle cells (38), most investigators now believe that the potential benefit derived from stem cells obtained from adult donors does not result through their transdifferentiating into, and thereby regenerating, the target tissue. Rather, any potential beneficial effect derives from their paracrine activities; i.e., through secretion of numerous cytokines and growth factors, including those relating to inflammation, adverse LV remodeling, collagen deposition, angiogenesis, tissue healing, apoptosis, mitochondrial dysfunction, microvascular dysfunction, and collagen deposition (38–45). These multiple activities could attenuate the multiple pathways contributing to progression of DVM and adverse LV remodeling that ultimately lead to HF (**Central Illustration**).

We believe this multiplicity of effects can uniquely benefit clinical outcomes to a greater degree than a therapeutic agent with more targeted and, therefore, more limited activities. Furthermore, that different types of stem cells secrete different varieties of factors suggests that their particular patterns of secretory products influence their relative efficacy. Following is a more detailed description of stem cell paracrine activities that could influence processes leading

to progressive adverse LV remodeling and, ultimately, to HF.

ANTI-INFLAMMATORY, IMMUNOMODULATORY, AND ANTIOXIDANT ACTIVITIES. Mesenchymal stem cells (MSCs) exert immunosuppressive activity in vitro (46,47). In the presence of an inflammatory milieu, MSCs suppress proliferation and activation of T cells, dendritic cells (DCs), macrophages, and B lymphocytes and alter the secretory profile of DCs, T cells (T helper 1 [TH1] and TH2), and natural killer cells, thus inducing an anti-inflammatory and immunotolerant phenotype. Thus, MSCs stimulate DCs to decrease secretion of tumor necrosis factor alpha and increase secretion of interleukin-10, TH1 cells to decrease secretion of interferon (interferon) gamma, TH2 cells to increase secretion of interleukin-4, and natural killer cells to decrease secretion of interferon gamma. These cells also exert anti-inflammatory effects in vivo (48,49).

MSCs also exert antioxidant and anti-inflammatory activities in vivo. Although we are not aware of any demonstration of these activities in the setting of HF, compelling data using other models have demonstrated these activities in vivo. Thus, carbon tetrachloride-induced liver injury in rats damages the liver, at least in part, by reducing glutathione production and increasing free radical levels, triggering a cascade of reactions leading to liver inflammation and fibrosis. Treatment with MSCs increased glutathione levels and decreased hepatic necrosis, fatty changes, and inflammation (48). Similarly, in a rat liver model of ischemic reperfusion, administration of MSCs led to less liver damage, higher superoxide dismutase and glutathione peroxidase activities, fewer apoptotic hepatocytes, and lower levels of Bcl-2-associated X and caspase-3 proteins (49). The published data relating to whether other stem cell types have such activities is much more limited, although they may have such activities (50).

NEOVASCULARIZATION. Patients with ICM or NICM have coronary microvascular dysfunction, raising the possibility that angiogenic interventions might improve outcomes. Angiogenesis involves the coordinated expression of a very large number of factors, such as vascular endothelial growth factor, hepatocyte growth factor, and angiopoietin-1, which promote migration of endothelial progenitor cells to the target organ and are involved in regulation of the ECM (43,51). Many of these factors are secreted by stem cells, and multiple studies have suggested that stem cells can enhance angiogenesis by increasing capillary density (41,52,53). For example, when comparing 18 control subjects to 15 NICM patients, Roura et al. (20) found defective vascularization in NICM patients, characterized by short and smaller major coronary segments (when adjusted to LV mass) and decreased epicardial microvascular density (20).

The potential of stem cells to improve collateral development adds an important mechanism by which they could contribute to improved outcomes in ICM and NICM. This is highly relevant in DVM, as neovascularization may allow increased perfusion and improvement of myocardial function. Conversely, a small recent study (n = 6) examining the mechanisms of bone marrow-derived cell therapy in patients during LV assist device placement did not show evidence of increased vascular tissue, as assessed by density of $CD31^{+}$ endothelial cells and number of manually-counted blood vessels (54). As the authors duly noted, the results may be discordant from prior studies due to the unique physiological properties of LVADs on the myocardium, as well as the small study sample and older age of the donor cells.

ADVERSE REMODELING, ECM PRODUCTION, AND INHIBITION OF APOPTOSIS. Myocardial remodeling involves a combination of changes in cardiac structure and function, including apoptosis and an altered ECM. Among the changes in ECM is its expansion, resulting from collagen overproduction. The underlying biology of ECM expansion with associated development of myocardial fibrosis is poorly understood. It is unclear if primary fibroblast activation induces reactive fibrosis, thereby causing myocyte loss, or if myocyte injury occurs first and is then followed by fibroblast activation and replacement fibrosis (36). Interestingly, the abnormalities of ECM-related processes are improved by renin-angiotensin-aldosterone modulation drugs that also improve HF outcomes (36). This raises the possibility that the diverse molecular pathways involved in the processes leading to an altered ECM and to reversibility of DVM might also benefit from the multiple stem cell-released paracrine factors.

As an example, injection of MSCs can alter matrix metalloproteinase (MMP) expression (43), which could enhance their ability to migrate through tissue and thereby to contribute to tissue remodeling. In addition, their specific microenvironment appears to determine their overall activity. Recent studies have clarified MSC-related effects on MMPs and their inhibitors, the tissue inhibitors of metalloproteinase (TIMPs) (55,56). Thus, MSCs exposed to proinflammatory cytokines and hypoxia secrete TIMP-2 and -1, thereby reducing levels of MMP-2 and -9 (56). These activities could inhibit a key mechanism involved in adverse remodeling.

Numerous other studies suggest that stem cell-released factors inhibit cardiomyocyte apoptosis. The likelihood that these effects are due to paracrine

activity is evidenced by a study in rats, where bone marrow mononuclear-derived cells were cultured under hypoxic conditions and the obtained supernatant was associated with improved cardiac function, decreased apoptosis, and enhanced angiogenesis when injected into ischemic rat hearts (44). Nagaya et al. (57), using a rat model of NICM, injected MSCs into the myocardium. There was increased expression of MMP-2 and -9, and increased vascular endothelial growth factor, hepatocyte growth factor, and insulin growth factor-1, which are associated with angiogenesis and antiapoptotic and antifibrotic properties (57). Zhang et al. (53), using a rat model of diabetic cardiomyopathy, injected bone marrow mononuclear-derived cells into the femoral vein and demonstrated increased expression of MMP-2, decreased expression of an apoptotic factor, MMP-9, increased arteriolar density, and decreased collagen volume in myocardium. Similar findings were seen in rats with hypertensive heart disease (58). The published ICM data has also extensively demonstrated the reduction of infarct size and inhibition of apoptosis of chronic ischemic HF post-stem cell injection (59,60).

CARDIAC REGENERATION. Numerous types of adult progenitor cells have been hypothesized as having potential for cardiac regeneration, either by transdifferentiation or by recruitment or activation of resident cardiac stem cells. Although the former is now regarded as an unlikely therapeutic mechanism for adult stem cells, the latter 2 concepts are under active consideration. Two widely cited clinical trials in patients with ICM, SCIPIO (Stem Cell Infusion in Patients with Ischemic Cardiomyopathy) (61) and CADUCEUS (Cardiosphere-Derived Autologous Stem Cells) trials (62), reported that stem cell administration led to regeneration of functioning myocardium. However, these 2 trials involved very few patients (10 randomized patients in SCIPIO, 25 in CADUCEUS). (Note: an "Expression of Concern" was issued by *The Lancet* [63] relating to the SCIPIO paper, and a critical paper authored by the senior investigator of the laboratory responsible for the stem cell aspect of this study was retracted [64,65].) Pre-clinical data published after SCIPIO further questioned the contribution of the type of stem cell used (c-kit-positive cardiac stem cells), as endogenous c-kit-positive cells did not produce significant cardiomyocytes within the heart in a mouse model (66).

CLINICAL TRIALS WITH STEM CELL THERAPY IN HF

OVERVIEW. Definitive data supporting proof of stem cell clinical efficacy are still lacking. This is not surprising, given that adequately powered pivotal trials have not yet been reported. In this regard, 2 recently published comprehensive reviews, 1 focusing on stem cell treatment of AMI and the other on stem cell treatment of HF, have focused on whether efficacy has been attained.

Analyzing the results in patients with AMI, Clifford et al. (1) published a meta-analysis of all clinical trials that were randomized and published through 2012 (39 trials, 1,765 participants) (1). Although there was a significant increase in ejection fraction with stem cell therapy (as determined by cardiac magnetic resonance imaging), it averaged only 1.78%. This magnitude of improvement is of unknown clinical consequence. There were no significant changes in mortality or morbidity. Sanganalmath and Bolli (2) reached the same conclusion in a review of HF studies published through 2012 (37 trials; 17 randomized). Thus, both studies concluded that, viewed as a totality, evidence of efficacy is not compelling.

Meta-analyses provide a powerful tool for taking groups of clinical trials that individually have too few patients to yield statistically robust results and, by grouping the patients, markedly increase statistical power. However, when the individual trials being analyzed use different cell types, have very different cell delivery protocols, and vary in their patient accrual criteria, the grouping of all studies and the analysis provided would miss individual studies using particular cells and modes of administration that might be effective. At this time, no adequately powered individual studies convincingly demonstrate efficacy. Although individual studies may report encouraging "signals," until adequately powered randomized trials are completed, we have to recognize that efficacy has not yet been demonstrated.

AUTOLOGOUS VERSUS ALLOGENEIC STEM CELLS

With rare exceptions, all published stem cell studies for cardiovascular indications have used autologous cells. This may be a critically important reason why the overall results of stem cell trials have been disappointing (1,2). Patients with cardiovascular disease are usually older and have numerous risk factors—characteristics that compromise stem cell function in both humans and experimental animals (40,67). More recently, studies using allogeneic stem cells have been initiated (**Table 1**). This may be a critically important change in stem cell therapeutic strategy that could lead to clinical benefit.

TABLE 1 Ongoing Clinical Trials of Stem Cell Therapy in Nonischemic and Ischemic Dilated Cardiomyopathy*

Trial	Origin of CM	ClinicalTrials.gov NCT #	Status	Trial Type	Estimated Patient Enrollment	Cell Source	Delivery Method
DYNAMIC	NICM	NCT02293603	Recruiting	Phase 1 RCT	42	Allogeneic CDC	IC
NOGA-DCM	NICM	NCT01350310	Recruiting	Phase 2 RCT	60	Allogeneic HSC	IM IC
POSEIDON-DCM	NICM	NCT01392625	Recruiting	Phase 1 and 2 RCT	36	Autologous MSC Allogeneic MSC	TE
RIMECARD	NICM	NCT01739777	Recruiting	Phase 1 and 2 RCT	30	Umbilical cord MSC	IV
Maranon	NICM	NCT01957826	Recruiting	Phase 1 and 2 RCT	70	Autologous MSC	TE
CardioCell	NICM	NCT02123706	Recruiting	Phase 2 RCT	20	Allogeneic MSC	IV
Iniciativa Andaluza	NICM	NCT02033278	Recruiting	Phase 2 and 3 RCT	51	Autologous BMMC	IC
REMEDIUM	NICM	NCT02248532	Recruiting	Phase 2 and 3 RCT	80	Autologous CD34$^+$ BMMC	IM
Aghdami	NICM	NCT02256501	Recruiting	Phase 1 RCT	32	Autologous BMMC	IC
Ageless Regenerative	NICM	NCT01502501	Recruiting	Phase 1 and 2 RCT	10	Autologous ADSC	IM IV
Teva Pharmaceutical	NICM	NCT02032004	Recruiting	Phase 3 RCT	1,730	Allogeneic MSC	TE
Royan Institute	ICM	NCT01758406	Recruiting	Phase 2 RCT	50	Autologous CSC	IC
Ageless Regenerative	ICM	NCT01502514	Recruiting	Phase 1 and 2 RCT	10	Autologous ADSC	IM IC
AHEPA	ICM	NCT01753440	Recruiting	Phase 2 and 3 RCT	30	Allogeneic MSC	IM
Hebein	ICM	NCT01946048	Not yet recruiting	Phase 1 RCT	10	Allogeneic umbilical cord MSC	IM
WJ-ICMP	ICM	NCT02368587	Not yet recruiting	Phase 2 RCT	160	Umbilical MSC	IC IV
ISCIC	ICM	NCT01615250	Recruiting	Phase 1 RCT	50	Peripheral CD34$^+$ SC	IM
AHEPA	ICM	NCT01759212	Recruiting	Phase 2 and 3 RCT	5	Allogeneic MSCs	IM
Malheiros	ICM	NCT01913886	Recruiting	Phase 1 and 2 RCT	10	Autologous MSCs	IC
AlsterMACs	ICM	NCT01337011	Recruiting	Phase 1 and 2 RCT	64	Autologous CD133$^+$ BMC	IC IM
HUC-Heart	ICM	NCT02323477	Recruiting	Phase 1 and 2 RCT	79	Allogeneic umbilical MSCs Autologous BMC	IM
IMPACT-CABG	ICM	NCT01033617	Recruiting	Phase 2 RCT	20	CD133+ BMC	IM
ESCORT	ICM	NCT02057900	Recruiting	Phase 1 RCT	6	Embryonic SC	EPI
ASSURANCE	ICM NICM	NCT00869024	Recruiting	Phase 1 and 2 RCT	24	Autologous BMC	IM
CSCC_ASC	ICM	NCT02387723	Recruiting	Phase 1 RCT	10	Allogeneic ADSC	IM
ESTIMATION	ICM	NCT01394432	Recruiting	Phase 3 RCT	50	Autologous MSC	TE
TRIDENT	ICM	NCT02013674	Ongoing	Phase 2 RCT	30	Allogeneic MSC	TE
RACE-STEMI	ICM	NCT02323620	Not yet recruiting	Phase 3 RCT	200	Autologous BMC	IC
REPEAT	ICM	NCT01693042	Recruiting	Phase 2 and 3 RCT	676	Autologous BMC	IC
BAMI	ICM	NCT01569178	Recruiting	Phase 3 RCT	300	Autologous BMC	IC

*Last checked April 2, 2015 on Clinicaltrials.gov. Studies with an unknown status (status that had not been verified for more than 2 years), completed, terminated, suspended, or withdrawn were excluded.

ADSC = adipose-derived stem cell; Ageless Regenerative I = Safety and Efficacy of Adipose Derived Stem Cells for Non-Ischemic Congestive Heart Failure I; Ageless Regenerative II = Safety and Efficacy of Adipose Derived Stem Cells for Congestive Heart Failure II; Aghdami = Intracoronary Transplantation of Bone Marrow Derived Mononuclear Cells in Pediatric Cardiomyopathy; AHEPA = Allogeneic Stem Cells Implantation Combined With Coronary Bypass Grafting in Patients With Ischemic Cardiomyopathy; AHEPA = Left Ventricular Assist Device Combined With Allogeneic Mesenchymal Stem Cells Implantation in Patients With End-stage Heart Failure; AlsterMACs = Intra-coronary Versus Intramyocardial Application of Enriched CD133pos Autologous Bone Marrow Derived Stem Cells; ASSURANCE = Stem Cell Therapy in Patients With Severe Heart Failure & Undergoing Left Ventricular Assist Device Placement; BAMI = The Effect of Intracoronary Reinfusion of Bone Marrow-derived Mononuclear Cells(BM-MNC) on All Cause Mortality in Acute Myocardial Infarction; BMC = bone marrow cell; BMMC = bone marrow mononuclear cell; Cardiocell = A Study to Assess the Effect of Intravenous Dose of (aMBMC) to Subjects With Non-ischemic Heart Failure; CDC = cardiosphere-derived cell; CM = cardiomyopathy; CSC = cardiac stem cell; CSCC_ASC = CSCC_ASC Therapy in Patients With Severe Heart Failure; DYNAMIC = Dilated cardiomYopathy iNtervention With Allogeneic MyocardIally-regenerative Cells; EPI = epicardial; ESCORT = Transplantation of Human Embryonic Stem Cell-derived Progenitors in Severe Heart Failure; ESTIMATION = "ESTIMATION Study" for Endocardial Mesenchymal Stem Cells Implantation in Patients After Acute Myocardial Infarction; Hebein = Umbilical Cord Derived Mesenchymal Stem Cells Therapy in Ischemic Cardiomyopathy; HSC = hematopoietic stem cell; HUC-Heart = Human Umbilical Cord Stroma MSC in Myocardial Infarction; IC = intracoronary; ICM = ischemic cardiomyopathy; IM = intramyocardial; IMPACT-CABG = IMPACT-CABG Trial: IMPlantation of Autologous CD133+ sTem Cells in Patients Undergoing CABG; Inciativa Andaluza = Infusion Intracoronary of Mononuclear Autologous Adult no Expanded Stem Cells of Bone Marrow on Functional Recovery in Patients With Idiopathic Dilated Cardiomyopathy and Heart Failure; ISCIC = Implantation of Peripheral Stem Cells in Patients With Ischemic Cardiomyopathy; IV = intravenous; Malheiros = Mesenchymal Stem Cells to Treat Ischemic Cardiomyopathy; Maranon = Mesenchymal Stem Cells for Idiopathic Dilated Cardiomyopathy; MSC = mesenchymal stem cell; NCT = National Clinical Trial; NICM = nonischemic cardiomyopathy; NOGA-DCM = Safety and Efficacy Study of Intramyocardial Stem Cell Therapy in Patients With Dilated Cardiomyopathy; POSEIDON-DCM = PercutaneOus StEm Cell Injection Delivery Effects On Neomyogenesis in Dilated CardioMyopathy (The POSEIDON-DCM Study); RACE-STEMI = Impact of Intracoronary Injection of Autologous BMMC for LV Contractility and Remodeling in Patients With STEMI; RCT = randomized controlled trial; REMEDIUM = Repetitive Intramyocardial CD34+ Cell Therapy in Dilated Cardiomyopathy; REPEAT = Compare the Effects of Single Versus Repeated Intracoronary Application of Autologous Bone Marrow-derived Mononuclear Cells on Mortality in Patients With Chronic Post-infarction Heart Failure; RIMECARD = Randomized Clinical Trial of Intravenous Infusion Umbilical Cord Mesenchymal Stem Cells on Cardiopathy; Royan Institute = Transplantation of Autologous Cardiac Stem Cells in Ischemic Heart Failure; SC = stem cell; TE = transendocardial; Teva Pharmaceutical = The Purpose of This Study is to Evaluate the Efficacy and Safety of a Allogeneic Mesenchymal Precursor Cells (CEP-41750) for the Treatment of Chronic Heart Failure; TRIDENT = The TRansendocardial Stem Cell Injection Delivery Effects on Neomyogenesis Study; WJ-ICMP = Intracoronary or Intravenous Infusion Human Wharton' Jelly-derived Mesenchymal Stem Cells in Patients With Ischemic Cardiomyopathy.

FUTURE IMPLICATIONS

Many questions about stem cell therapy for HF remain unanswered. We do not know the most effective cell type for cell therapy. This is compounded by the utilization of different cell types and either autologous or allogeneic cells in different studies. As mentioned earlier, there are distinct differences between stem cells derived from older patients with risk factors versus those derived from young, healthy donors. Other questions remain, such as the optimal dose, whether repeated administration is necessary, the ideal stage of HF for implementation, and the most effective route of administration. We also do not yet know whether optimal efficacy of stem cells requires a strategy to stimulate the cells to augment secretion of factors that might attenuate the many mechanisms involved in HF progression. Finally, if stem cell modification is necessary, we do not know whether this can best be achieved by genetic modification, by altering their growth conditions, or by some other strategy.

An additional question arises: assuming that the dysfunctional myocardium is not actively ischemic (which may not be true), are sufficient homing signals present in the damaged myocardium of these patients to induce stem cell engraftment? Evidence suggests that this signal exists (68,69), but the issue needs further study. Alternatively, if cells do not adequately home to the injured tissue, is it possible that during their residence in other distant tissues (lungs, spleen, liver) they secrete factors that either: 1) are delivered via the circulatory system to the myocardium, where they exert direct beneficial effects; and/or 2) exert systemic effects by modulating processes that cause progressive myocardial injury, such as activation of the inflammatory and immune systems?

The field of stem cell research in HF is extremely active and robust (**Table 1**). Studies are assessing various cell types of either autologous or allogeneic origin and various delivery routes. Thus, despite the remaining issues, we believe that the outcomes data and mechanistic knowledge accumulated from the many pre-clinical and clinical studies performed to date, and the knowledge we will accumulate from studies in progress, have the potential to provide insights that will eventually lead stem cell therapeutics to become a highly successful strategy in improving outcomes in patients with HF.

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