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Effect of Intracoronary Delivery of Autologous Bone Marrow Mononuclear Cells 2 to 3 Weeks Following Acute Myocardial Infarction on Left Ventricular Function

The LateTIME Randomized Trial

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for the Cardiovascular Cell Therapy Research Network (CCTRN)

Context Clinical trial results suggest that intracoronary delivery of autologous bone marrow mononuclear cells (BMCs) may improve left ventricular (LV) function when administered within the first week following myocardial infarction (MI). However, because a substantial number of patients may not present for early cell delivery, the efficacy of autologous BMC delivery 2 to 3 weeks post-MI warrants investigation.

Objective To determine if intracoronary delivery of autologous BMCs improves global and regional LV function when delivered 2 to 3 weeks following first MI.

Design, Setting, and Patients A randomized, double-blind, placebo-controlled trial (LateTIME) of the National Heart, Lung, and Blood Institute–sponsored Cardiovascular Cell Therapy Research Network of 87 patients with significant LV dysfunction (LV ejection fraction [LVEF] ≤45%) following successful primary percutaneous coronary intervention (PCI) between July 8, 2008, and February 28, 2011.

Interventions Intracoronary infusion of 150×10^6 autologous BMCs (total nucleated cells) or placebo (BMC:placebo, 2:1) was performed within 12 hours of bone marrow aspiration after local automated cell processing.

Main Outcome Measures Changes in global (LVEF) and regional (wall motion) LV function in the infarct and border zone between baseline and 6 months, measured by cardiac magnetic resonance imaging. Secondary end points included changes in LV volumes and infarct size.

Results A total of 87 patients were randomized (mean [SD] age, 57 [11] years; 83% men). Harvesting, processing, and intracoronary delivery of BMCs in this setting was feasible. Change between baseline and 6 months in the BMC group vs placebo for mean LVEF (48.7% to 49.2% vs 45.3% to 48.8%; between-group mean difference, −3.00; 95% CI, −7.05 to 0.95), wall motion in the infarct zone (6.2 to 6.5 mm vs 4.9 to 5.9 mm; between-group mean difference, −0.70; 95% CI, −2.78 to 1.34), and wall motion in the border zone (16.0 to 16.6 mm vs 16.1 to 19.3 mm; between-group mean difference, −2.60; 95% CI, −6.03 to 0.77) were not statistically significant. No significant change in LV volumes and infarct volumes was observed; both groups decreased by a similar amount at 6 months vs baseline.

Conclusion Among patients with MI and LV dysfunction following reperfusion with PCI, intracoronary infusion of autologous BMCs vs intracoronary placebo infusion, 2 to 3 weeks after PCI, did not improve global or regional function at 6 months.

Trial Registration clinicaltrials.gov Identifier: NCT00684060.

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Several randomized trials have demonstrated that administration of autologous bone marrow mononuclear cells (BMCs) following acute myocardial infarction (MI) may result in improvement in left ventricular ejection fraction (LVEF)[1-3] or regional LV function,[4] and may be associated with decreased clinical adverse events.[5] However, the

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majority of trials[1-4,6-11] have administered BMCs within the first week following primary percutaneous coronary intervention (PCI). Because the optimal time to administer BMCs has not been determined, the National Heart, Lung, and Blood Institute (NHLBI)–sponsored Cardiovascular Cell Therapy Research Network (CCTRN) developed 2 prospective clinical trials, TIME[12] and LateTIME.[13] The TIME trial was designed to compare the effects of BMC delivery in patients with predominantly ST-segment elevation MIs at 3 vs 7 days post-MI, and the LateTIME trial was designed to explore whether delayed BMC delivery 2 to 3 weeks following MI could improve global and regional LV function.

The time frame of 2 to 3 weeks post-MI may be particularly important for those patients who present to centers that lack expertise in cell therapy or those patients initially too sick as a result of cardiogenic shock or other medical issues. These patients may particularly benefit from cell therapy given that several trials[1,6,14] have demonstrated that those patients with the most depressed LV function appear to derive the most improvement from BMC delivery.

LateTIME is a novel, randomized, double-blind, placebo-controlled trial designed to investigate the use and therapeutic efficacy of intracoronary autologous BMC delivery 2 to 3 weeks following MI using rigorous methods of cell isolation in conjunction with local cell processing.[15] It is the first BMC trial to our knowledge to deliver a standardized dose of cells following stenting of the infarct vessel during primary PCI.

METHODS

Organizational Structure and Oversight

The CCTRN was established by the NHLBI to develop, coordinate, and simultaneously conduct multiple collaborative trials testing the effects of cell therapy on cardiovascular disease. The CCTRN consists of 5 clinical research centers (Cleveland Clinic Foundation, Cleveland, Ohio; University of Florida, Gainesville; Minneapolis Heart Institute Foundation, Minneapolis, Minnesota; Texas Heart Institute, Houston; and Vanderbilt University, Nashville, Tennessee) and their satellite sites, a data coordinating center at the University of Texas School of Public Health, which provides trial management and data analysis, a cell processing quality control center, and 6 core laboratories.[16] All clinical centers participate in the selection and design of network protocols, which are also reviewed by an independent protocol review committee and a gene therapy/cell therapy data and safety monitoring board under the aegis of the NHLBI. Each clinical center and the data coordinating center have independent institutional review board approvals and oversight.

Study Design

The LateTIME trial is a phase 2, randomized, double-blinded, placebo-controlled trial developed to determine if delayed (2-3 weeks) intracoronary administration of 150×10^6 total nucleated cells to patients with predominantly anterior MIs can safely produce a measurable improvement in global and regional LV function as determined by cardiac magnetic resonance imaging (MRI) at 6 months compared with baseline. Patients with an LVEF of 45% or less by echocardiography post-PCI were randomized in a 2:1 ratio of BMC to placebo following successful stenting of the infarct-related coronary artery. All patients will be followed up for 2 years to assess clinical events.

Study Protocol

All patients provided written informed consent following broad discussions of the risks, benefits of the trial, and alternatives explained by the investigative team. Race/ethnicity was documented as self-described by participants. Demographic and clinical variables were determined by interview and the patient's medical record. Patients were randomized in a 2:1 ratio to cell therapy or placebo. All patients underwent bone marrow aspiration and intracoronary infusion of BMCs or cell-free solution (placebo). All caregivers and patients were blinded to treatment. Approximately 80 to 90 mL of bone marrow were aspirated from the iliac crest using standard techniques. The aspirate was processed at all sites with a closed, automated cell processing system (Sepax, Biosafe SA)[15] to ensure a uniform cellular product. After BMC enrichment, cells were washed 3 times and suspended in 5% human serum albumin/saline solution. The composition of CD34 and CD133 cells was determined by fluorescent activated cell sorting. After the cells passed stipulated lot release criteria, including viability (>70%) and sterility, randomization was performed by the data coordinating center. Treatment assignment was masked to all but 1 designated cell processing team member at each of the 5 centers who was not involved in patient care. The target dose for the treatment group was 150×10^6 total nucleated cells. Patients randomized to placebo received 5% human serum albumin/saline solution to which 100 µL of autologous blood was added to ensure that the color and consistency of the solution matched that of the BMC product.

Within 12 hours of aspiration, the BMCs or cell-free product was delivered to the infarct-related artery via a percutaneous transluminal coronary angioplasty catheter (Maverick, Boston Scientific Corporation) using the stop-flow technique in 6 aliquots of 5 mL each, administered over 2 minutes with balloon inflation at low pressures within the previously placed stent. Each infusion cycle was separated by balloon deflation and 2 minutes of reperfusion. All patients were treated with aspirin and 75 mg of clopidogrel, in addition to guideline-recommended post-MI medications.

Study End Points

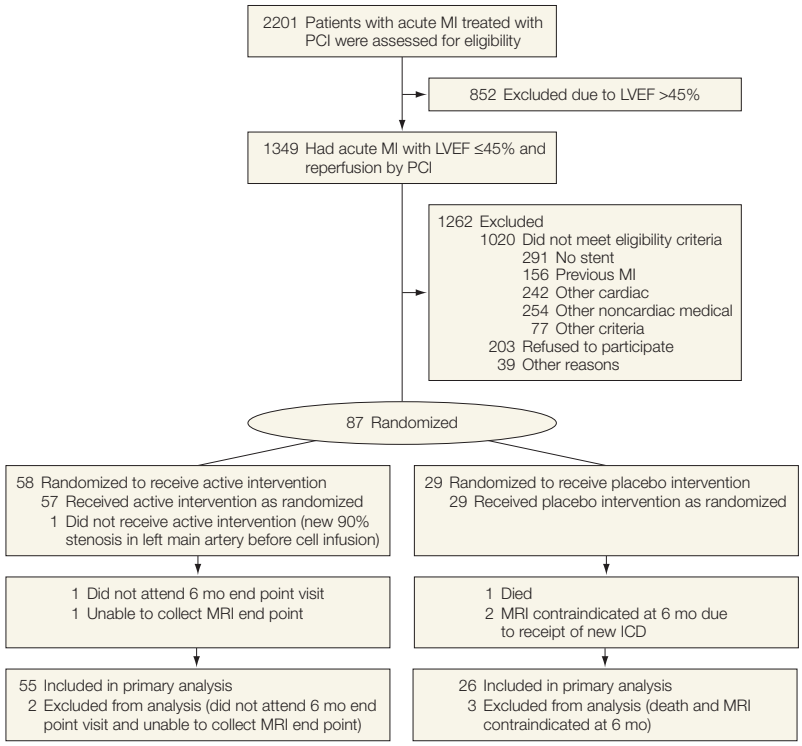
Wall Motion Imaging. All imaging was performed by using 1.5T MRI scanners with imaging protocols developed by the MRI core laboratory (Uni-

versity of Florida) certified before the study began. The MRI core laboratory was blind to study group assignment. Both global and segmental LV function measurements were obtained using a steady state free precession or fast gradient Echo technique. Long-axis cine images in the 2-chamber and 4-chamber projections were acquired. In addition, a set of contiguous short-axis slices (8-10 mm thick) were obtained from the mitral valve annulus through the apex of the LV throughout the cardiac cycle.

Data were analyzed using the Cardiovascular Angiography Analysis System/Magnetic Resonance Ventricular analysis software (PIE Medical Imaging BV). Global LV parameters assessed included end-diastolic volume, end-systolic volume, stroke volume, ejection fraction, and LV mass. Volumetric measurements were performed by direct planimetry on the contiguous short-axis images at both end systole and end diastole. Regional measurements include wall thickening and wall motion and were calculated using 100 chords spaced every 3.6 degrees originating from the centroid of the left ventricle for each short-axis image. Regional data were reported using the American Heart Association 17-segment model. The minimum spatial and temporal resolution requirements of the steady state free precession sequence are 2.5×2.5 mm voxels and 40 milliseconds, respectively.

Viability Imaging. Fifteen to 20 minutes following administration of a gadolinium-chelate contrast agent (0.05 mmol/kg intravenous), delayed-enhancement imaging was performed with a T1-weighted inversion-recovery prepared gradient-echo sequence (delayed-enhancement MRI). The inversion delay time was iteratively adjusted for optimal nulling of normal myocardium. Contrast-enhanced viability imaging was performed using the standard 2D technique in the short-axis projections, which acquires a single slice each breath hold using the same plane prescription as the functional short-axis cine series.

Figure 1. Flow Diagram of Patients



MI indicates myocardial infarction; PCI, percutaneous coronary intervention; LVEF, left ventricular ejection fraction; MRI, magnetic resonance imaging; ICD, implantable cardioverter defibrillator.

Regions of irreversible myocardial damage are manifested by "hyperenhancement" (bright white areas) on the images, and normal and/or viable tissue is "nulled" (black) on the acquired images. The presence, location, and extent of irreversibly damaged tissue was assessed and reported using the American Heart Association 17-segment model, in order to permit direct correlation with regional functional measurements. Pretherapy and posttherapy imaging, both cine wall motion and delayed-enhancement MRI, were carefully matched using internal landmarks including the insertion sites of the right ventricular free-wall and the papillary muscle insertions.

Safety Monitoring

All participants were closely monitored for adverse events and this information was transmitted to the US Food and Drug Administration, the NHLBI gene and cell therapy data and safety monitoring board, and institutional review boards of each center. A set of stopping rules was developed in consultation with the Food and Drug Administration. The data coordinating center was responsible for coordination of collection, standardization, integration, and analysis of study data from the various study components (enrolling sites and core facilities) and the preparation and distribution of the required reports to each of the safety oversight entities.

Statistical Analyses

The statistical methods used in the LateTIME trial have been reported previously.[13] The primary end points were (1) change in global LV function over time and (2) change in regional function over time as assessed by change in wall motion in the infarct and border zones. The prespecified analyses for the primary end point compared the change

Table 1. Baseline Characteristics of BMC and Placebo Groups[a]

Characteristics	BMC (n = 58)	Placebo (n = 29)
Age, mean (SD), y	57.6 (11)	54.6 (11)
Female sex	12 (21)	3 (10)
Race/ethnicity		
White	51 (88)	24 (83)
Nonwhite	7 (12)	5 (17)
History		
Diabetes	11 (19)	7 (24)
High blood pressure	32 (55)	14 (48)
Hyperlipidemia	43 (74)	20 (69)
Angina	15 (26)	4 (14)
Smoking	36 (62)	15 (52)
Preinfarction angina	14 (24)	9 (31)
Height, mean (SD), in	68 (4)	70 (3)
Weight, mean (SD), lbs	183 (37)	194 (33)
BMI, mean (SD)	27.7 (5.5)	28.0 (4.3)
BP at initial discharge, mean (SD), mm Hg		
Systolic	111.1 (13.9)	110.1 (11.7)
Diastolic	68.0 (10.1)	68.4 (9.0)
Heart rate		
Initial at ED		
Mean (SD)	77.5 (18)	90.3 (26)[b]
Median (range)	74 (50-124)	85 (54-170)[c]
Initial discharge		
Mean (SD)	73.2 (12)	77.2 (9)
Median (range)	73 (52-100)	77 (57-99)
Qualifying LVEF (echocardiogram), mean (SD)[d]	36.4 (6.5)	35.0 (7.6)
Hemoglobin, mean (SD), gm/dL	13.5 (0.2)	13.1 (1.8)
hsCRP, mean (SD), mg/L	20.1 (6)	14.2 (4)
BNP, mean (SD)	273.9 (210)	528.8 (1291)
Peak creatine kinase MB, mean (SD)	234.0 (212)	318.2 (203)
Peak troponin, mean (SD)		
Troponin T	6.8 (6.3)	10.3 (5.9)
Troponin I	163.3 (197.6)	144.2 (129.5)
Time from chest pain to ED, median (IQR), h	1.95 (1.0-10.7)	2.01 (0.8-5.3)
Time from chest pain to PCI, median (IQR), h	3.4 (2.3-14.3)	3.3 (2.2-7.5)
Door-to-balloon time, median (IQR), h	1.73 (0.9-3.4)	1.52 (0.8-2.3)
Transferred from outside hospital after PCI	10 (17)	6 (21)
Time from bone marrow aspiration to infusion, median (IQR), h	8.5 (7.95-9.25)	8.6 (7.8-9.8)
Time from PCI to infusion, median (IQR), d	17.4 (15.5-20.0)	16.8 (15.8-17.8)
Drug-eluting stent	45 (78)	20 (69)
Stent region		
LAD	53 (91)	27 (93)
LAD only	49 (92)	24 (89)
LAD + LCX	1 (2)	2 (7)
LAD + RCA	3 (6)	1 (4)
LCX (only)	1 (2)	1 (3)
RCA (only)	4 (7)	1 (3)

Abbreviations: BMC, bone marrow mononuclear cell, BMI, body mass index (calculated as weight in kilograms divided by height in meters squared); BNP, brain natriuretic peptide; BP, blood pressure; ED, emergency department; hsCRP, high-sensitivity C-reactive protein; IQR, interquartile range; LAD, left anterior descending coronary artery; LCX, left circumflex coronary artery; LVEF, left ventricular ejection fraction; PCI, percutaneous coronary intervention; RCA, right coronary artery.

[a] Data are presented as No. (%) unless otherwise specified. BMC vs placebo group comparisons are not statistically significant unless otherwise noted. For preinfarction angina, n=57 for BMC. For heart rate at initial presentation in ED, n=54 for BMC and n=28 for placebo; and for initial discharge, n=55 for BMC and n=27 for placebo. For hsCRP, n=51 for BMC and n=25 for placebo. For BNP, n=52 for BMC and n=24 for placebo. For peak creatine kinase MB, n=38 for BMC and n=23 for placebo. For peak troponin T, n=23 for BMC and n=10 for placebo; and for peak troponin I, n=15 for BMC and n=14 for placebo. For time from chest pain to ED, n=53 for BMC and n=28 for placebo. For door-to-balloon time, n=28 for placebo. For transferred from outside hospital after PCI, time from bone marrow aspiration to infusion, and time from PCI to infusion, n=57 for BMC.

[b] For mean heart rate at initial presentation in ED, $P=.01$ (Wilcoxon rank sum).

[c] For median heart rate at initial presentation in ED, $P=.02$ (Wilcoxon rank sum).

[d] Qualifying was defined as an LVEF of 45% or less from myocardial infarction through consent (at any point during this 2-3 week period).

over 6 months in the end point for the BMC group with the same change over time in the placebo group.

Sample Size Consideration

The sample size was calculated using a 2-sample *t*-test statistic. During the design phase of the LateTIME trial, the literature[1,3] suggested that the placebo adjusted change in the global LVEF (change in global LVEF in the BMC group minus change in global LVEF in the placebo group) was 4% and common group standard deviation of the difference of LVEF over time was expected to be $\sigma_\Delta=6$. Anticipating that 5% of patients would be lost to follow-up, and a 2:1 (BMC: placebo) ratio, a trial size of 86 was required. For the regional function wall motion evaluation, we assumed a placebo-adjusted change of 6.7 with $\sigma_\Delta=9.5$ from the Boost trial[3]; again assuming 5% loss to follow-up and 2:1 randomization, the study sample size was 77 patients. To have adequate power for both end points, a final sample size of 87 patients was selected (58 patients in the BMC group and 29 patients in the placebo group), providing 83% power for the global and 87% power for the regional measures of LV function.

The regional LV function end point was defined as change in wall motion over time in the infarct and border zone of the infarct. The infarct zone was defined as the segments with the largest 2 signal intensity enhancement measures with gadolinium (using a 17-segment model). The border zone was defined as those regions adjacent to the infarct zone in which the signal intensity enhancement were in the 10% to 75% range.

Exact testing for categorical variables and Student *t* testing for continuous variables assessed the comparability of baseline variables between treatment groups. All hypotheses testing and all effect sizes with their 95% CIs were evaluated using the general mixed linear model (adjusted for heart rate) and unadjusted comparisons of treatment effects. The primary and sec-

ondary evaluations compared the randomized study groups using an intention-to-treat analysis. No adjustments for multiple comparisons were made, and $P < .05$ was used to assess statistical significance. An imputation analysis of the primary end points was also performed by including all patients with incomplete follow-up data by last observation carried forward method.

RESULTS

Screening and Enrollment

Screening commenced for the LateTIME trial on July 8, 2008, and the first patient was randomized on September 30, 2008. Between July 8, 2008, and February 28, 2011, 2201 patients were screened, with the majority excluded for having an LVEF of more than 45% (FIGURE 1). No statistically significant differences in baseline characteristics were observed between the BMC and placebo groups, except for heart rate on initial presentation at the emergency department, which was higher in the placebo group (TABLE 1). The median time from chest pain onset to PCI was 3.4 (interquartile range [IQR], 2.3-14.3) hours in the BMC group and 3.3 (IQR, 2.2-7.5) hours in the placebo group. The mean (SD) LVEF on the qualifying echocardiogram performed following PCI during the initial hospitalization was 36.4% (6.5%) in the BMC group and 35.0% (7.6%) in the placebo group. As expected, this was significantly less than the baseline LVEF obtained by cardiac MRI several weeks later, in part due to resolution of myocardial stunning.

Cell Processing

Bone marrow aspiration and intracoronary infusion were performed a median of 17.4 (IQR, 15.5-20.0) days in the BMC group and 16.8 (IQR, 15.8-17.8) days in the placebo group following primary PCI (Table 1). There were no complications associated with the bone marrow aspiration. All patient products underwent automated cell processing with Ficoll using the Sepax device.[15]

Table 2. Cell Characteristics of BMC and Placebo Groups[a]

Characteristics	BMC (n = 58)	Placebo (n = 29)
Total nucleated cells/product ($\times 10^6$), mean (SD)	147 (17)	NA
% Viability/product by trypan blue exclusion		
Mean (SD)	98.5 (1.3)	98.6 (1.2)
Median (range)	99.0 (94.0-100.0)	99.0 (96.0-100.0)
% Viability/product by 7-AAD staining		
Mean (SD)	95.2 (5.4)	95.7 (5.1)
Median (range)	97.4 (77.5-99.8)	97.0 (72.1-99.3)
% CD34 cells/product, mean (SD)[b]	2.6 (1.0)	2.7 (1.4)
% CD133 cells/product, mean (SD)[b]	1.2 (0.5)	1.2 (0.6)
Colony-forming units-Hill/product, mean (SD)[b,c]	139 (251)	194 (277)
Endothelial colony-forming cells/product, mean (SD)[b,d]	184 (250)	163 (218)

Abbreviations: BMC, bone marrow mononuclear cell; NA, not applicable.
[a] BMC vs placebo group comparisons are not statistically significant.
[b] Seven patients either declined participation or had insufficient product for the biorepository.
[c] n=45 for BMC and n=23 for placebo.
[d] n=38 for BMC and n=21 for placebo.

Intracoronary Infusion

The median time from bone marrow aspiration to intracoronary infusion was 8.5 hours in the BMC group (Table 1). All patients received 150×10^6 total nucleated cells (mean 60%-70% BMCs; mean [SD], 2.6% [1.0%] CD34 cells and 1.2% [0.5%] CD133 cells), except 3 patients who received a lower than target total nucleated cell dose due to the low cell numbers in the initial bone marrow (TABLE 2). The mean (SD) viability of the cell product was 98.5% (1.3%). One patient did not receive the BMC infusion due to the presence of a severe left main coronary stenosis, which was identified before infusion. This patient was referred for coronary artery bypass graft surgery. Two patients underwent additional stenting at the time of the intracoronary infusion (one for the discovery of a distal stent edge dissection related to the primary PCI procedure and another for a possible dissection related to the stop-flow procedure). One patient who had a postpartum spontaneous coronary dissection was found to have diffuse thrombus throughout the stented region of the left anterior descending artery. The patient successfully underwent aspiration thrombectomy with ultrasound-guided stent expansion followed by infusion of study product. No patients experienced a postprocedural increase in cardiac enzymes and the patients were routinely discharged the following day.

Table 3. Clinical/Safety Outcomes at 6-Month End Point Window

Outcomes	BMC (n = 58)	Placebo (n = 29)
Patients, No. (%)	3 (5)	5 (17)
No. of events		
Death	0	1
Reinfarction	1	0
Repeat revascularization	1	3
Target vessel	1	2
Nontarget vessel	0	1
Heart failure hospitalization	1	0
ICD placement	0	2
No. of total events	3	6

Abbreviations: BMC, bone marrow mononuclear cell; ICD, implantable cardiac defibrillator.

Safety

Despite a high-risk cohort of patients with moderate to severe LV dysfunction following predominantly large, anterior MIs, there were very few clinical events (TABLE 3). In the placebo group, 1 death occurred due to recurrent pancreatitis 3 months following randomization. Three patients underwent repeat revascularization and 2 patients received implantable cardiac defibrillators. The BMC group had fewer events than the placebo group did, with 1 reinfarction, 1 repeat revascularization, and 1 hospitalization for heart failure.

Analysis of Global and Regional LV Function

A total of 55 patients in the BMC group and 26 patients in the placebo

group had paired cardiac MRI data at baseline and 6 months available for analysis of global and regional LV function. Six patients were excluded from the global analysis (LVEF) due to the following reasons: (1) death (n=1), (2) withdrawal from study due to presence of severe left main stenosis before cell infusion (n=1), (3) placement of implantable cardiac defibrillators (n=2), and (4) lost to follow-up (n=2). One additional patient was excluded from the infarct zone regional analysis due to incomplete signal intensity enhancement data and 4 additional patients did not undergo regional measurements in the border zone because of lack of a signal intensity enhancement signal in the border zone.

Baseline and follow-up end point measures for the primary end points are shown in TABLE 4 and FIGURE 2. The mean (SD) difference in the change from baseline to 6 months in the BMC group compared with the placebo group for LVEF was not different (48.7% [12.0%] to 49.2% [13.0%] vs 45.3% [9.9%] to 48.8% [7.8%]; between-group difference, –3.00; 95% CI, –7.05 to 0.95; *P*=.14). The mean (SD) difference in the change in wall motion in the infarct zone for the BMC group vs the placebo group was also not different (6.2 [6.5] to 6.5 [6.8] mm vs 4.9 [4.8] to 5.9 [5.7] mm; between-group difference, –0.70; 95% CI, –2.78 to 1.34; *P*=.49). Similarly, the mean (SD) difference in the change in wall motion in the border zones between the BMC and placebo groups was not different (16.0 [9.9] to 16.6 [9.6] mm vs 16.1 [10.0] to 19.3 [10.9] mm; between-group difference, –2.60; 95% CI, –6.03 to 0.77; *P*=.13). An analysis using a mixed linear model adjusted for heart rate did not change these findings. Inclusion of patients lacking paired cardiac MRI data by last observation carried forward imputation had no effect on the primary results. Left ventricular ejection fraction measured by echocardiography was consistent with the cardiac MRI findings in showing no treatment effect (mean [SD], 44.3% [8.4%] to 47.6% [11.0%] for BMC and 42.4% [6.5%] to 46.5% [8.0%] for placebo; *P*=.64).

Secondary end points of LV volumes demonstrated a small but nonsignificant increase in LV end diastolic volume index and end systolic volume index in the BMC group at 6 months (Table 4). Infarct volume uniformly decreased in both groups without significant difference (mean [SD] within-group change, –3.5 [19.0] mL for BMC and –2.0 [14.4] mL for placebo; between-group difference in 6-month change, –1.50; 95% CI, –9.89 to 6.88; *P*=.73).

Table 4. End Point Analyses of Global and Regional LV Function Between Baseline and 6 Months

	BMC		Placebo		Analysis	
	No.	Mean (SD)	No.	Mean (SD)	Between-Group Difference in 6-Month Change (95% CI)	*P* Value
Global LV Function						
LVEF, %						
Baseline	55	48.7 (12.0)	26	45.3 (9.9)		
Follow-up	55	49.2 (13.0)	26	48.8 (7.8)		
Within-group change	55	0.5 (8.2)	26	3.6 (9.3)	–3.00 (–7.05 to 0.95)	.14
Regional LV Function						
Infarct zone wall motion, mm						
Baseline	55	6.2 (6.5)	25	4.9 (4.8)		
Follow-up	55	6.5 (6.8)	25	5.9 (5.7)		
Within-group change	55	0.3 (4.3)	25	1.0 (4.5)	–0.70 (–2.78 to 1.34)	.49
Border zone wall motion, mm						
Baseline	53	16.0 (9.9)	23	16.1 (10.0)		
Follow-up	53	16.6 (9.6)	23	19.3 (10.9)		
Within-group change	53	0.5 (7.2)	23	3.2 (6.3)	–2.60 (–6.03 to 0.77)	.13
Global LV Function						
End diastolic volume index, mL/m^2						
Baseline	55	89.1 (23.9)	26	82.8 (26.4)		
Follow-up	55	92.5 (32.7)	26	85.5 (22.7)		
Within-group change	55	3.4 (23.4)	26	2.7 (18.1)	0.70 (–9.47 to 10.91)	.89
End systolic volume index, mL/m^2						
Baseline	55	47.8 (21.4)	26	46.3 (20.4)		
Follow-up	55	48.0 (25.1)	26	44.0 (14.8)		
Within-group change	55	0.2 (14.0)	26	–2.3 (14.7)	2.50 (–4.11 to 9.15)	.46
Infarct volume, mL						
Baseline	55	34.2 (23.7)	25	33.3 (15.6)		
Follow-up	55	30.7 (15.8)	25	31.4 (15.1)		
Within-group change	55	–3.5 (19.0)	25	–2.0 (14.4)	–1.50 (–9.89 to 6.88)	.73

Abbreviations: BMC, bone marrow mononuclear cell; LV, left ventricular; LVEF, left ventricular ejection fraction.

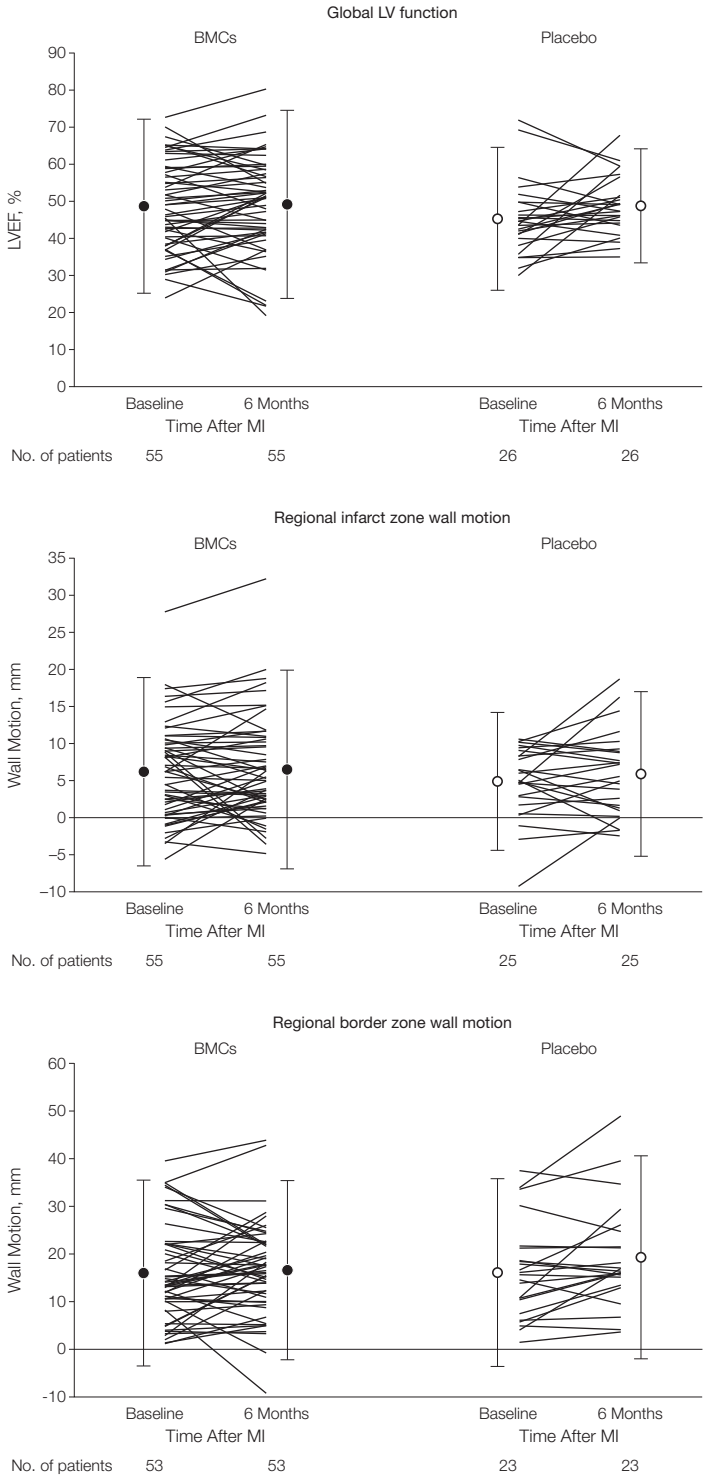
Several predetermined subgroup analyses were performed in the treatment group. In contrast with previous studies,[1,6,14] no observed improvement was observed in recovery of LV function in the group of patients with the most depressed LVEF at baseline. No difference was observed in global or regional function in patients stratified by ischemic time. Patients who were older than 65 years demonstrated a small but nonsignificant decrease in LVEF at 6 months following cell therapy (P=.11).

COMMENT

The CCTRN was created by the NHLBI to accelerate development of cell-based therapies in the United States, using a network approach to facilitate patient recruitment, standardization of cell processing, and development of core laboratories for outcome measures analysis. LateTIME is the first trial to be completed from the CCTRN,[13] and it was developed to test the hypothesis that delayed delivery of autologous BMCs following MI would improve global and regional LV function when measured 6 months later by cardiac MRI. However, we observed that BMC delivery 2 to 3 weeks following MI resulted in no detectable improvement in LV function over that observed in the placebo group. The findings were consistent in demonstrating a lack of benefit in both global and regional wall motion in the infarct and border zone and stand in contrast with several studies[1-3] that demonstrated benefit in LV function when BMCs were administered within the first week following MI. Additionally, measurement of infarct size, which may be a more sensitive marker of cell therapy efficacy, decreased by a similar amount in both groups.

Patients recruited to the LateTIME trial constituted a high-risk cohort with depressed LV function that persisted several weeks following successful revascularization with stenting. Although retrospective analyses suggest that these patients may derive the most benefit from cell therapy in this set-

Figure 2. Primary End Point Analysis of Global LV Function and Regional Infarct and Border Zone Wall Motions



LV indicates left ventricular; LVEF, left ventricular ejection fraction; BMCs, bone marrow mononuclear cells; MI, myocardial infarction. Solid and open circles represent the means at baseline and 6 months of BMCs and placebo, respectively, and error bars represent 95% CIs.

ting,[1,6,14] no improvement in LV function was noted, even in the subgroup with the most depressed LVEF. The LateTIME trial is the first BMC trial in the MI population to deliver a uniform number of cells to its cohort in a dose thought sufficient to modify LV function.[17] However, our results suggest that intracoronary BMC delivery at this later period is not effective.

Timing of Cell Delivery Following Acute MI

The majority of randomized cell therapy trials using BMCs in the setting of MI have delivered cells within the first 7 days following ST-segment elevation MI.[1-4,6-10] In a subgroup analysis, the REPAIR-AMI trial[1] observed that the most favorable effects on LV function were observed with BMC delivery on days 5 to 7 post-MI. However, no trial has been specifically designed to identify the optimum time of cell delivery. The LateTIME trial was designed to determine if delayed delivery of BMCs to patients following MI would be safe and effective in improving LV function.

It is likely that the timing of cell delivery post-MI may have a major influence on treatment effect and, ultimately, may have contributed to our negative findings. Following MI, significant temporal changes occur in the myocardium and bone marrow that may affect engraftment and retention of delivered cells.[18-24] As a result, the myocardial environment during later treatment as in the LateTIME trial is likely to be considerably different from that present during the first week post-MI when BMCs were delivered in other trials. In the first few days following MI, there is an extensive inflammatory response triggered by infiltration of neutrophils and other cells that may lead to an increase in cytokines, such as tumor necrosis factor, interleukin-1 (IL-1), and reactive oxygen species that may adversely influence delivered cells.[18,19,21] Microvascular obstruction in the infarct zone may impair inflow of oxygen and nutrients to support stem cell survival. This is countered by an increased expression of stromal-derived factor-1[25] in the first few days following MI that may increase stem cell trafficking and engraftment. During the next 1 to 2 weeks, there is a transition from an inflammatory phase to a proliferative phase in which extracellular matrix is formed and neovascularization is increased.[18] The net effect of these changes may have negatively influenced the potential beneficial effects of BMC delivery.[26]

Changes in Circulating Progenitor Cells Derived From Bone Marrow Following Acute MI

Within hours of MI, a well-documented increase in circulating progenitor cells released from bone marrow occurs that may contribute to myocardial repair.[22-24,27] These include release of hematopoietic stem cells, endothelial progenitor cells, mesenchymal stem cells, and very small embryonic-like cells, a novel progenitor cell with pluripotent properties that express early markers of cardiomyocyte differentiation.[22] Many of these cells contain a variety of cell surface markers, including CD34, c-kit, c-met, vascular endothelial growth factor-2R, and CXCR4 that actively participate in ischemic tissue repair, in part through homing in response to gradients of vascular endothelial growth factor-2R, hepatocyte growth factor-1, and stromal-derived factor-1. The peak release of these bone marrow–derived cells has been measured in hours to days,[22-24,27] but would appear to be outside the window of when cells were delivered in our study. Thus, a possible synergistic effect between intracoronary delivered and circulating progenitor cells may not have been possible in our study.

With the rapid egress of stem cells shortly after MI, it is possible that the bone marrow was relatively depleted of progenitor cells when the bone marrow aspiration was performed in our study. As a result, the reparative quality of the BMC product may be different from cells delivered in the first week post-MI. However, a recent preclinical study in mice observed that BMCs are significantly more potent at 21 days compared with those obtained at 3 to 5 days following MI due to IL-1–mediated inflammatory changes in the bone marrow.[19] These effects will be studied by the CCTRN Biorepository Core Laboratory,[28] in the future through analysis of peripheral blood and bone marrow of patients enrolled in our study and the ongoing TIME trial,[12] that performed bone marrow aspirations and intracoronary infusions in patients randomized to days 3 or 7 following acute MI.

Are BMCs the Proper Cell Type to Be Used in the Setting of Acute MI?

Our decision to use unselected BMCs for this late post-MI therapy was based on extensive preclinical data indicating that no specific cell type clearly exceeded another in regards to enhanced potency for altering ventricular remodeling and function.[29-31] Use of a closed and standardized commercial device, with central quality control for cell processing, facilitated provision of a uniform cell product. Extensive preclinical testing demonstrated a product with similar composition but less variability to that obtained by traditional manual separation methods with Ficoll.[15] Accordingly, autologous bone marrow–derived stem and progenitor cells could be produced at most hospitals and would be more convenient for transplantation by intracoronary infusion, if this strategy were successful. The CCTRN developed several satellite centers of cell delivery, including 1 center located more than 100 miles from the center where cell processing occurred. The quality control, processing metrics, and out of body times were equivalent, even at this distance, supporting the feasibility of this approach.

In addition to timing, an ongoing concern is the ultimate effectiveness of BMCs to improve LV function following MI. The LateTIME trial was developed at a time when several studies demonstrated significant improvement in LV function with BMC therapy,[1,3] and meta-analyses confirmed a small but significant improvement in LVEF and attenuation of LV re-

modeling.[17,32,33] Since these initial publications, several studies have failed to demonstrate that BMCs improve LV function in this setting.[6-11] Indeed a recent analysis found that the improvement in LVEF following BMCs was less than 1% when cardiac MRI was used for measurement of LVEF in nearly 700 patients.[34] Although concerns of cell processing were initially thought to contribute to some of the negative findings,[35] these concerns have largely been allayed.[36] Furthermore, it is well understood that the reparative properties of autologous stem and progenitor cells derived from bone marrow are negatively influenced by a variety of factors, including advanced age, diabetes, and other cardiovascular risk conditions, all common to patients enrolled in clinical trials. Whether newer cell types under investigation, including mesenchymal stem cells,[37] mesenchymal precursor cells,[38] multipotent adult progenitor cells (clinicaltrials.gov Identifier: NCT00677222), adipose-derived cells (clinicaltrials.gov Identifier: NCT00442806), or encompassing allogeneic products obtained from young healthy donors, can improve LV function to a greater degree than BMCs following MI remains to be determined.

Limitations

The LateTIME trial is the first cell therapy trial in the MI population to use a standardized, automated, closed system of cell processing. Rigorous preclinical testing determined that this system produced a similar cell product compared with manual Ficoll separation in regards to cell recovery, viability, and colony-forming unit formation.[17] However, because the cell product was not tested in vivo, we cannot discard that unknown modifications occurred in the cell product that could have contributed to our negative findings. In addition, although there are several different approaches to measure myocardial strain (myocardial tagging, DENSE [Displacement Encoding with Stimulated Echo], etc), these were not used in our study due to the need for specialized expertise that may not have been available at all of the clinical sites that collected the MRI data.

CONCLUSION

Among patients with MI and LV dysfunction following reperfusion with PCI, intracoronary infusion of autologous BMCs compared with intracoronary placebo infusion 2 to 3 weeks after PCI did not improve global or regional LV function at 6 months.

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REFERENCES

1. Schächinger V, Erbs S, Elsässer A, et al. Intracoronary bone marrow-derived progenitor cells in acute myocardial infarction. *N Engl J Med*. 2006;355(12):1210-1221.

2. Huang RC, Yao K, Zou YZ, et al. Long term follow-up on emergent intracoronary autologous bone marrow mononuclear cell transplantation for acute inferior-wall myocardial infarction. *Zhonghua Yi Xue Za Zhi*. 2006;86(16):1107-1110.

3. Wollert KC, Meyer GP, Lotz J, et al. Intracoronary autologous bone-marrow cell transfer after myocardial infarction: the BOOST randomised controlled clinical trial. *Lancet*. 2004;364(9429):141-148.

4. Janssens S, Dubois C, Bogaert J, et al. Autologous bone marrow-derived stem-cell transfer in patients with ST-segment elevation myocardial infarction: double-blind, randomised controlled trial. *Lancet*. 2006;367(9505):113-121.

5. Schächinger V, Erbs S, Elsässer A, et al. Improved clinical outcome after intracoronary administration of bone-marrow-derived progenitor cells in acute myocardial infarction: final 1-year results of the REPAIR-AMI trial. *Eur Heart J*. 2006;27(23):2775-2783.

6. Tendera M, Wojakowski W, Ruzyłło W, et al. Intracoronary infusion of bone marrow-derived selected CD34+CXCR4+ cells and non-selected mononuclear cells in patients with acute STEMI and reduced left ventricular ejection fraction: results of randomized, multicentre Myocardial Regeneration by Intracoronary Infusion of Selected Population of Stem Cells in Acute Myocardial Infarction (REGENT) Trial. *Eur Heart J*. 2009;30(11):1313-1321.

7. Lunde K, Solheim S, Aakhus S, et al. Intracoronary injection of mononuclear bone marrow cells in acute myocardial infarction. *N Engl J Med*. 2006;355(12):1199-1209.

8. Hirsch A, Nijveldt R, van der Vleuten PA, et al. Intracoronary infusion of mononuclear cells from bone marrow or peripheral blood compared with standard therapy in patients after acute myocardial infarction treated by primary percutaneous coronary intervention: results of the randomized controlled HEBE trial. *Eur Heart J*. 2011;32(14):1736-1747.

9. Traverse JH, McKenna DH, Harvey K, et al. Results of a phase 1, randomized, double-blind, placebo-controlled trial of bone marrow mononuclear stem cell administration in patients following ST-elevation myocardial infarction. *Am Heart J*. 2010;160(3):428-434.

10. Wöhrle J, Merkle N, Mailänder V, et al. Results of intracoronary stem cell therapy after acute myocardial infarction. *Am J Cardiol*. 2010;105(6):804-812.

11. Roncalli J, Mouquet F, Piot C, et al. Intracoronary autologous mononucleated bone marrow cell infusion for acute myocardial infarction: results of the randomized multicenter BONAMI trial. *Eur Heart J*. 2011;32(14):1748-1757.

12. Traverse JH, Henry TD, Vaughan DE, et al. Rationale and design for TIME: a phase II, randomized, double-blind, placebo-controlled pilot trial evaluating the safety and effect of timing of administration of bone marrow mononuclear cells after acute myocardial infarction. *Am Heart J*. 2009;158(3):356-363.

13. Traverse JH, Henry TD, Vaughan DE, et al. LateTIME: a phase-II, randomized, double-blinded, placebo-controlled, pilot trial evaluating the safety and effect of administration of bone marrow mononuclear cells 2 to 3 weeks after acute myocardial infarction. *Tex Heart Inst J*. 2010;37(4):412-420.

14. Miettinen JA, Ylitalo K, Hedberg P, et al. Determinants of functional recovery after myocardial infarction of patients treated with bone marrow-derived stem cells after thrombolytic therapy. *Heart*. 2010;96(5):362-367.

15. Gee AP, Richman S, Durett A, et al. Multicenter cell processing for cardiovascular regenerative medicine applications: the Cardiovascular Cell Therapy Research Network (CCTRN) experience. *Cytotherapy*. 2010;12(5):684-691.

16. Simari RD, Moyé LA, Skarlatos SI, et al. Development of a network to test strategies in cardiovascular cell delivery: the NHLBI-sponsored Cardiovascular Cell Therapy Research Network (CCTRN). *J Cardiovasc Transl Res*. 2010;3(1):30-36.

17. Martin-Rendon E, Brunskill SJ, Hyde CJ, et al. Autologous bone marrow stem cells to treat acute myocardial infarction: a systematic review. *Eur Heart J*. 2008;29(15):1807-1818.

18. Dobaczewski M, Gonzalez-Quesada C, Frangogiannis NG. The extracellular matrix as a modulator of the inflammatory and reparative response following myocardial infarction. *J Mol Cell Cardiol*. 2010;48(3):504-511.

19. Wang X, Takagawa J, Lam VC, et al. Donor myocardial infarction impairs the therapeutic potential of bone marrow cells by an interleukin-1-mediated inflammatory response. *Sci Transl Med*. 2011;3(100):ra90.

20. Higuchi T, Bengel FM, Seidl S, et al. Assessment of alphavbeta3 integrin expression after myocardial infarction by positron emission tomography. *Cardiovasc Res*. 2008;78(2):395-403.

21. Frangogiannis NG. The immune system and cardiac repair. *Pharmacol Res*. 2008;58(2):88-111.

22. Wojakowski W, Landmesser U, Bachowski R, Jadczyk T, Tendera M. Mobilization of stem and progenitor cells in cardiovascular diseases. *Leukemia*. doi:10.1038/leu2011.184.

23. Leone AM, Rutella S, Bonanno G, et al. Mobilization of bone marrow-derived stem cells after myocardial infarction and left ventricular function. *Eur Heart J*. 2005;26(12):1196-1204.

24. Massa M, Rosti V, Ferrario M, et al. Increased circulating hematopoietic and endothelial progenitor cells in the early phase of acute myocardial infarction. *Blood*. 2005;105(1):199-206.

25. Askari AT, Unzek S, Popovic ZB, et al. Effect of stromal-cell-derived factor 1 on stem-cell homing and tissue regeneration in ischaemic cardiomyopathy. *Lancet*. 2003;362(9385):697-703.

26. Schächinger V, Aicher A, Döbert N, et al. Pilot trial on determinants of progenitor cell recruitment to the infarcted human myocardium. *Circulation*. 2008;118(14):1425-1432.

27. Shintani S, Murohara T, Ikeda H, et al. Mobilization of endothelial progenitor cells in patients with acute myocardial infarction. *Circulation*. 2001;103(23):2776-2779.

28. Cogle CR, Pettway R, Zierold C, et al. Time from onset to acute myocardial infarction negatively influences bone marrow cell function: preliminary data from the NHLBI CCTRN ancillary studies [abstract]. *Circulation*. 2009;120(18):S1132.

29. Orlic D, Kajstura J, Chimenti S, et al. Bone marrow cells regenerate infarcted myocardium. *Nature*. 2001;410(6829):701-705.

30. Amado LC, Saliaris AP, Schuleri KH, et al. Cardiac repair with intramyocardial injection of allogeneic mesenchymal stem cells after myocardial infarction. *Proc Natl Acad Sci U S A*. 2005;102(32):11474-11479.

31. Yoon YS, Wecker A, Heyd L, et al. Clonally expanded novel multipotent stem cells from human bone marrow regenerate myocardium after myocardial infarction. *J Clin Invest*. 2005;115(2):326-338.

32. Abdel-Latif A, Bolli R, Tleyjeh IM, et al. Adult bone marrow-derived cells for cardiac repair: a systematic review and meta-analysis. *Arch Intern Med*. 2007;167(10):989-997.

33. Lipinski MJ, Biondi-Zoccai GG, Abbate A, et al. Impact of intracoronary cell therapy on left ventricular function in the setting of acute myocardial infarction: a collaborative systematic review and meta-analysis of controlled clinical trials. *J Am Coll Cardiol*. 2007;50(18):1761-1767.

34. Traverse JH, Henry TD, Moye' LA. Is the measurement of left ventricular ejection fraction the proper end point for cell therapy trials? an analysis of the effect of bone marrow mononuclear stem cell administration on left ventricular ejection fraction after ST-segment elevation myocardial infarction when evaluated by cardiac magnetic resonance imaging. *Am Heart J*. 2011;162(4):671-677.

35. Seeger FH, Tonn T, Krzossok N, Zeiher AM, Dimmeler S. Cell isolation procedures matter: a comparison of different isolation protocols of bone marrow mononuclear cells used for cell therapy in patients with acute myocardial infarction. *Eur Heart J*. 2007;28(6):766-772.

36. van Beem RT, Hirsch A, Lommerse IM, et al. Recovery and functional activity of mononuclear bone marrow and peripheral blood cells after different cell isolation protocols used in clinical trials for cell therapy after acute myocardial infarction. *EuroIntervention*. 2008;4(1):133-138.

37. Hare JM, Traverse JH, Henry TD, et al. A randomized, double-blind, placebo-controlled, dose-escalation study of intravenous adult human mesenchymal stem cells (prochymal) after acute myocardial infarction. *J Am Coll Cardiol*. 2009;54(24):2277-2286.

38. Perin EC, Dib N, DeMaria A, et al. A phase II dose-escalation study of allogenic mesenchymal precursor cells in patients with ischemic and non-ischemic heart failure. Paper presented at: 2011 Scientific Sessions of the American Heart Association; November 14, 2011; Orlando, FL.