Impact of Cell Therapy on Myocardial Perfusion and Cardiovascular Outcomes in Patients With Angina Refractory to Medical Therapy
A Systematic Review and Meta-Analysis
Abdur Rahman Khan, Talha A. Farid, Asif Pathan, Aynish Tripathi, Shahab Ghafghazi, Marcin Wysoczynski, Roberto Bolli

Rationale: The effect of stem/progenitor cells on myocardial perfusion and clinical outcomes in patients with refractory angina remains unclear because studies published to date have been small phase I–II trials.

Objective: We performed a meta-analysis of randomized controlled trials to evaluate the effect of cell-based therapy in patients with refractory angina who were ineligible for coronary revascularization.

Methods and Results: Several data sources were searched from inception to September 2015, which yielded 6 studies. The outcomes pooled were indices of angina (anginal episodes, Canadian Cardiovascular Society angina class, exercise tolerance, and antianginal medications), myocardial perfusion, and clinical end points. We combined the reported clinical outcomes (myocardial infarction, cardiac-related hospitalization, and mortality) into a composite end point (major adverse cardiac events). Mean difference (MD), standardized mean differences, or odds ratio were calculated to assess relevant outcomes. Our analysis shows an improvement in anginal episodes (MD, −7.81; 95% confidence interval [CI], −15.22 to −0.41), use of antianginal medications (standardized MD, −0.59; 95% CI, −1.03 to −0.14), Canadian Cardiovascular Society class (MD, −0.58; 95% CI, −1.00 to −0.16), exercise tolerance (standardized MD, 0.331; 95% CI, 0.08 to 0.55), and myocardial perfusion (standardized MD, −0.49; 95% CI, −0.76 to −0.21) and a decreased risk of major adverse cardiac events (odds ratio, 0.49; 95% CI, 0.25 to 0.98) and arrhythmias (odds ratio, 0.25; 95% CI, 0.06 to 0.98) in cell-treated patients when compared with patients on maximal medical therapy.

Conclusions: The present meta-analysis indicates that cell-based therapies are not only safe but also lead to an improvement in indices of angina, relevant clinical outcomes, and myocardial perfusion in patients with refractory angina. These encouraging results suggest that larger, phase III randomized controlled trials are in order to conclusively determine the effect of stem/progenitor cells in refractory angina. (Circ Res. 2016;118:984-993. DOI: 10.1161/CIRCRESAHA.115.308056.)

Key Words: cell- and tissue-based therapy ■ meta-analysis ■ perfusion ■ randomized controlled trial ■ stem cells
to promote neovascularization and endothelial repair, thereby improving myocardial perfusion. Although several studies have been conducted to assess the effect of cell therapy in patients with angina refractory to conventional medical therapy and ineligible for coronary revascularization, the small size of these phase I–II trials and the lack of uniform primary end points make it difficult to discern an efficacy signal. As a result, the effect of stem/progenitor cells on myocardial perfusion and clinical outcomes in RFA remains unclear; there is no phase III trial underway to corroborate the findings of these proof-of-concept trials. A meta-analysis published in 2013 concluded that cell-based therapy is safe and effective in RFA. However, that study failed to evaluate all of the functional and clinical end points examined in the individual studies; importantly, the impact of cell therapy on myocardial perfusion was not assessed. Here, we present a comprehensive, current review, and meta-analysis of the safety and efficacy of cell-based therapy in RFA that expands on the previous analysis by including a new study, by assessing all of the functional and clinical end points measured, and by exploring the effect of cell therapy on myocardial perfusion.

**Methods**

**Data Sources and Search Strategy**

This systematic review was performed in accordance with the Preferred Reporting Items for Systematic Reviews and Meta-analyses guidelines. The search strategy and subsequent literature search were performed by an experienced medical reference librarian. The search strategies were developed in PubMed and translated to match the subject headings and keywords for Embase, Cochrane Central Register of Controlled Trials, and ISI Web of Science from inception to September 8, 2015. In addition, conference proceedings were searched for articles pertaining to the search criteria. The following MeSH, Emtree, and keyword search terms were used in combination: cardiac stem cell therapy, bone marrow derived mononuclear cells, cardiac progenitor cells, endothelial progenitor cells (EPCs), refractory angina, intractable angina, drug resistant angina, coronary heart disease, coronary perfusion, myocardial perfusion, myocardial ischemia, single photon emission computed tomography (SPECT), controlled trials, intervention study, and randomized controlled trials. The search accounted for plurals and variations in spelling with the use of appropriate wildcards. To identify further articles, we hand-searched related citations in review articles and commentaries. All results were downloaded into EndNote (Thompson Reuters), and duplicate citations were identified and removed.

**Study Selection**

Two investigators (T.A.F. and A.P.) independently assessed the eligibility of the studies identified. The studies that were evaluated were RCTs that focused on the role of cell therapy in patients with angina refractory to medical therapy who were not eligible for coronary revascularization.

**Data Extraction**

From the included studies, 2 reviewers (A.R.K. and A.T.) independently extracted data on the population under study, patient characteristics, type of cell-based therapy used, and relevant outcomes. The outcomes measured in our analysis were the safety and efficacy of cell-based therapy. The main efficacy outcomes studied were indices of angina, myocardial perfusion, and clinical end points. Safety of cell-based therapy was measured by the adverse events reported in the included studies.

The indices of angina measured were changes in frequency of angina episodes, Canadian Cardiovascular Society (CCS) angina class, exercise tolerance, change in antianginal medications, and quality of life. Myocardial perfusion was determined by SPECT, which was the imaging modality used in individual studies. Clinical end points reported in the studies were myocardial infarction, cardiac-related hospitalization, and mortality. Because a small number of these outcomes were reported in a limited number of studies, and because none of the studies was powered to assess these clinical end points, we combined the cardiovascular outcomes into a composite cardiac end point (major adverse cardiac events [MACE]) that includes myocardial infarction, cardiac-related hospitalization, and mortality.

**Data Synthesis and Statistical Analysis**

Continuous data were reported either as mean (SD) or median (interquartile range). If the data were reported as median, mean and SD were estimated. For continuous variables, the mean change between end of follow-up and baseline was measured in both groups; the mean difference (MD) was calculated as the difference between the mean change in the cell-treated group and the mean change in the control group. The MD was used to estimate changes in frequency of angina episodes, CCS class, and use of antianginal medications. The standardized MD (SMD; calculated in an analogous manner) was used to assess changes in myocardial perfusion and exercise tolerance, in which different units of measurements were used in different studies. For dichotomous variables, an odds ratio was calculated for MACE and occurrence of adverse events. Meta-analyses were performed with a fixed-effects model; a random-effect model was used if heterogeneity was encountered. The F statistic was used to assess heterogeneity among studies. Publication bias was assessed by means of a funnel plot; the Beggs and Mazumdar test was used to assess funnel plot asymmetry and publication bias if needed. Sensitivity analysis was done to investigate the associated heterogeneity and the effect of individual studies on it.

**Quality Assessment**

Two reviewers (A.R.K. and A.P.) independently assessed the methodological quality of the selected studies using the Cochrane risk of bias tool. This scale is used to explore the adequacy of sequence generation, allocation sequence concealment, blinding of participants and caregivers, blinding for outcome assessment, incomplete outcome reporting, and other potential bias. Any disagreements between reviewers in study inclusion, data extraction, and quality assessment that could not be resolved by consensus were resolved by a third reviewer (R.B.). All analyses were conducted using the statistical software Review Manager (version 5.2).

**Results**

**Identification of Studies**

The literature search identified 1136 publications, of which 6 studies were eligible for our analysis (Figure 1). To reduce variability in the study population, studies of ischemic cardiomyopathy that did not report angina symptoms refractory to medical therapy were not included. One study reported outcomes with 2 different doses of cell-based therapy; to avoid duplication, only the dose (low-dose group) reported to have better outcome was included in the analysis. The risk of bias in all included studies was determined to be low because the studies were of sound methodological quality (Figure 2).
There was no allocation bias because adequate randomization was reported in all of the trials except 2. All assessments of outcomes measured were blinded, with a low risk of documented bias for both selection and reported outcomes. The RCTs included in the analysis had a low risk of bias because of attrition during follow-up. There was excellent agreement between the reviewers with respect to inclusion of the studies, data abstraction, and quality assessment.

Study Characteristics
Table 1 summarizes the characteristics of the included studies. Table 2 highlights the characteristics of the patient populations in these studies. A total of 6 trials comprising 353 participants (ranging from 24 to 112 in individual trials) were included in the analysis. The studies were conducted in centers located in the United States, Europe, Asia, and Australia. Four studies were performed in >1 center.

Figure 1. Flowchart of eligible studies. G-CSF indicates granulocyte colony-stimulating factor.

Figure 2. Risk of bias of the included studies. LD indicates low-dose group.
### Table 1. Characteristics of Included Studies

<table>
<thead>
<tr>
<th>Study; Country</th>
<th>Study Design</th>
<th>Center</th>
<th>Inclusion Criteria</th>
<th>Exclusion Criteria</th>
<th>Cell Type; Route of Delivery</th>
<th>Outcome</th>
<th>Follow-Up</th>
</tr>
</thead>
<tbody>
<tr>
<td>Losordo et al; USA</td>
<td>RCT: double-blind, placebo-controlled</td>
<td>3</td>
<td>Age &gt;21 y, CCS III/IV, refractory to medical therapy, 2 antianginals, ineligible for revascularization, documented ischemia on nuclear perfusion</td>
<td>MI ≤30 d, revascularization ≤3 mo, joint disease/COPD/PVD, which may limit walking on treadmill severe AS/MS, CHF symptoms, life expectancy &lt;1 y, uncontrolled HTN</td>
<td>CD34+ cells; Intramyocardial (EMM, NOGA guided)</td>
<td>Angina frequency, NTG use, exercise tolerance, CCS class, SPECT imaging, QoL safety</td>
<td>1, 2, 4 wk; 2, 3, 6, 9, and 12 mo</td>
</tr>
<tr>
<td>Tae et al; Hong Kong, Australia</td>
<td>RCT: blind, placebo-controlled</td>
<td>2</td>
<td>CCS III/IV, refractory to medical therapy, ineligible for revascularization, documented viable myocardium on SPECT</td>
<td>Unprotected L main, decompensated CHF, LVEF ≤30%, ACS, or stroke within 3 mo, significant renal, liver, or hematomatologic abnormalities; AF/AS/LV thrombus/PAD prevent electromechanical LV mapping</td>
<td>BMMNC; Intramyocardial (EMM, NOGA guided)</td>
<td>Exercise treadmill, SPECT, cardiac MRI</td>
<td>3, 6 mo</td>
</tr>
<tr>
<td>van Ramshorst et al; Netherlands</td>
<td>RCT: double-blind, placebo-controlled</td>
<td>1</td>
<td>CCS III/IV despite medical therapy, documented ischemia in at least 1 myocardial segment on SPECT, ineligible for revascularization</td>
<td>LVEF &lt;35%, MI within 6 mo, GFR &lt;30 mL/min, unexplained hematologic abnormalities, malignancy</td>
<td>BMMNC; Intramyocardial (EMM, NOGA guided)</td>
<td>CCS class, QoL, exercise capacity, myocardial perfusion, LV function and volumes, arrhythmia</td>
<td>3, 6 mo</td>
</tr>
<tr>
<td>Wang et al; China</td>
<td>RCT: placebo-controlled</td>
<td>1</td>
<td>Age &gt;30 y, CCS III/IV, refractory to medical therapy, ineligible for revascularization, ischemia on nuclear perfusion, angina during baseline exercise</td>
<td>MI within 30 d, revascularization within 6 mo, TIA within 60 d, severe AS/MS, CHF symptoms, life expectancy &lt;1 y, uncontrolled HTN; joint disease/COPD/PVD, which may limit walking on treadmill</td>
<td>CD34+ cells; Intracoronary</td>
<td>Angina frequency, NTG use, exercise tolerance, CCS class, SPECT imaging; safety</td>
<td>1.2, 4 wk; 3 and 6 mo</td>
</tr>
<tr>
<td>Losordo et al; USA</td>
<td>RCT: double-blind, placebo-controlled</td>
<td>26</td>
<td>Age &gt;21 y, CCS III/IV, refractory to medical therapy, angina during baseline exercise, ischemia on nuclear perfusion, ineligible for revascularization</td>
<td>MI within 60 d, revascularization within 3 mo, LVEF ≤35%, predominant CHF symptoms, severe AS, prosthetic aortic valve, COPD which may limit walking on treadmill, creatinine &gt;2.5 mg/dL</td>
<td>CD34+ cells; Intramyocardial (EMM, NOGA guided)</td>
<td>Angina frequency, antianginal use, exercise tolerance, CCS class, SPECT imaging, MRI; safety</td>
<td>28 d; 3, 6 and 12 mo</td>
</tr>
<tr>
<td>Jimenez-Quevedo et al; Spain</td>
<td>RCT: single-blind, placebo-controlled</td>
<td>3</td>
<td>CCS III–IV, refractory to medical therapy, coronary anatomy not amenable to revascularization, documented ischemia on SPECT</td>
<td>MI within 3 mo, LV thrombus, aortic valve ds, hemorrhagic disorder, LV wall thickness &lt;8 mm, history of malignancy ≤5 y, pregnancy</td>
<td>CD133+ cells; Intramyocardial (EMM, NOGA guided)</td>
<td>CCS class, treadmill test, SPECT, antianginal medication, QoL, MRI; safety</td>
<td>1 wk; 1, 3 and 6 mo; 1 and 2 y</td>
</tr>
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</table>

ACS indicates acute coronary syndrome; AF, atrial fibrillation; AS, aortic stenosis; BMMNC, bone marrow mononuclear cells; CCS, Canadian Cardiovascular Society functional angina class; CHF, congestive heart failure; COPD, chronic obstructive pulmonary disease; EMM, electromechanical mapping; GFR, glomerular filtration rate; HTN, hypertension; LVEF, left ventricular ejection fraction; MI, myocardial infarction; MRI, magnetic resonance imaging; MS, mitral stenosis; NOGA, NOGA® XP Cardiac Navigation System; NTG, nitroglycerin; PAD, peripheral arterial disease, PVD, peripheral vascular disease; QoL, quality of life; RCT, randomized controlled trial; and SPECT, single photon emission computed tomography.

The patients included in the individual studies had symptoms of angina, CCS III–IV, refractory to medical therapy, and were not candidates for coronary revascularization. The patients were predominantly men with an average age >60 years in almost all studies. In the study by Wang et al,17 the patient population in the cell-based therapy group was...
relatively older than those in the control group. In all studies except that by Wang et al., the patient population had a high incidence of prior PCI and CABG (Tables 1 and 2).

There were 192 patients who received cell therapy along with the current standard of care and 161 patients who were on maximal medical therapy. Three studies used CD34+ cells, unfractionated bone marrow mononuclear cells, and CD133+ cells. The techniques to enrich and harvest the cells differed. Some studies used granulocyte colony-stimulating factor to treat patients before harvest. Three studies harvested the cells directly from the bone marrow, whereas 3 studies harvested the cells from peripheral blood. Three studies used magnetic sorting of cells, whereas 2 studies used density gradient centrifugation to enrich the cells after harvesting them. All but 1 study used electromechanical mapping with the NOGA® XP Cardiac Navigation System to deliver cells to the myocardium. In 1 study, the cells were infused into the left main and right coronary arteries during cardiac catheterization (Tables 1 and 2).

### Efficacy of Cell-Based Therapy

The efficacy of cell-based therapy was assessed by measuring changes in perfusion of the ischemic myocardium, changes in the indices of angina, and the composite cardiovascular end point. The indices used for the assessment of angina were frequency of angina episodes, CCS class, use of antianginal medications, exercise tolerance, and quality of life.

#### Myocardial Perfusion

SPECT was the imaging modality used to assess changes in myocardial perfusion, which were measured as the difference in SPECT scores between end of follow-up and baseline. Studies reported (1) both summed stress and summed rest scores, (2) both summed stress and difference scores, (3) summed stress only, or (4) summed difference score. Overall, 4 studies reported the change in summed stress scores, 2 reported the change in summed rest scores, and 2 reported the change in summed difference scores. All studies evaluated perfusion at 6 months except 2 studies, one of which reported SPECT scores both at 3 and 6 months whereas the other reported SPECT scores at 3 months. One study reported both automated and visually estimated scores; the scores with the lower reported improvement were included in the analysis. Given the limited number of studies that reported summed rest or difference scores, we pooled only summed stress scores to assess perfusion.

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**Table 2. Baseline Characteristics of the Included Patients**

<table>
<thead>
<tr>
<th>Study</th>
<th>Groups</th>
<th>n</th>
<th>Age, y</th>
<th>Men, %</th>
<th>DM, %</th>
<th>HTN, %</th>
<th>HPL, %</th>
<th>Smoking, %</th>
<th>CHF, %</th>
<th>MI Prior, %</th>
<th>PCI Prior, %</th>
<th>CABG Prior, %</th>
<th>BB Prior, %</th>
<th>ACEi Prior, %</th>
<th>CCB Prior, %</th>
<th>Nitrate Prior, %</th>
<th>Statin Prior, %</th>
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<tbody>
<tr>
<td>Losordo et al</td>
<td>18 Cell treated</td>
<td>62†</td>
<td>39</td>
<td>78</td>
<td>94</td>
<td>72</td>
<td>33</td>
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<td>NR</td>
<td>83</td>
<td>94</td>
<td>89</td>
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<td>Losordo et al</td>
<td>Cell treated</td>
<td>6</td>
<td>50</td>
<td>50</td>
<td>66</td>
<td>83</td>
<td>33</td>
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<td>NR</td>
<td>83</td>
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<tr>
<td>Tse et al</td>
<td>19 Cell treated</td>
<td>65</td>
<td>42</td>
<td>68</td>
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<td>NR</td>
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<tr>
<td>Tse et al</td>
<td>Control</td>
<td>9</td>
<td>69</td>
<td>88</td>
<td>63</td>
<td>75</td>
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<td>NR</td>
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<tr>
<td>van Ramshorst et al</td>
<td>25 Cell treated</td>
<td>64</td>
<td>52</td>
<td>48</td>
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<td>Wang et al</td>
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<td>Wang et al</td>
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<td>56</td>
<td>50</td>
<td>43</td>
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<td>61</td>
<td>47</td>
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<tr>
<td>Jimenez-Quevedo et al</td>
<td>19 Cell treated</td>
<td>70</td>
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<td>84</td>
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<td>15</td>
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</table>

*Shows combined data for use of ACEi and ARB (angiotensin receptor blocker).
†Mean age of both Cell treated and Control group; percentage of men in both Cell treated and Control group.

ACEi indicates angiotensin-converting enzyme inhibitor; BB, β-blocker; CABG, coronary artery bypass grafting; CCB, calcium channel blocker; CHF, congestive heart failure; DM, diabetes mellitus; HPL, hyperlipidemia; HTN, hypertension; MI, myocardial infarction; NR, not reported; and PCI, percutaneous coronary intervention.
Khan et al  Cell Therapy in Refractory Angina  989

(SMD, −0.49; 95% confidence interval [CI], −0.76 to 0.21; P=0.0006; F=0%). There was no between-study heterogeneity (I²=0%; Figure 3). Publication bias was not assessed because of the limited number of studies in the analysis.

Anginal Episodes
The mean change in frequency of anginal episodes versus baseline was reported in 4 studies.14,17–19 The pooled analysis of the studies suggested an improvement in the number of anginal episodes in patients treated with cell therapy (MD, −7.81; 95% CI, −15.25 to 0.37; P=0.04). There was substantial between-study heterogeneity (I²=90%), and sensitivity analysis revealed that all the heterogeneity was secondary to 1 study.17 After removal of that study, heterogeneity dropped to 0% and the MD decreased as well (MD, −3.38; 95% CI, −6.56 to 0.19; P=0.04; Figure 4A).

Changes in Antianginal Medications
The mean change in the number of medications versus baseline was reported in 4 studies.14,17–19 Two studies reported mean changes in medications per week,14,17 whereas 1 study each reported mean changes per day18 or per month.19 A meta-analysis of these 4 studies suggested a decrease in the use of antianginal medications in the cell therapy group (SMD, −0.62; 95% CI, −1.05 to 0.18; P=0.006; F=59%). Sensitivity analysis revealed that all of the heterogeneity was secondary to 1 study.17 After removal of that study, heterogeneity dropped to 0% and the SMD decreased as well (SMD, −0.35; 95% CI, −0.67 to 0.03; P=0.03; Figure 4C).

Change in CCS Class
The mean change in functional class versus baseline was reported in all included studies except 1.15 However, 1 study reported improvement in number of patients rather than a change in class.18 A pooled analysis of 4 studies suggested an improvement in CCS class in patients who received cell therapy (MD, −0.58; 95% CI, −1.00 to 0.16; P=0.007; F=0%; Figure 4C).

Change in Exercise Tolerance
The mean change in exercise tolerance versus baseline was reported in all studies. Exercise capacity was measured by a treadmill test with the standard Bruce protocol in 2 studies,14,17 a treadmill test with a modified Bruce protocol in 2 studies,15,18 and a symptom-limited bicycle exercise test in 1 study.16 One study used a treadmill test to ascertain tolerance but did not report the protocol used.19 Pooled analysis of the included studies demonstrated an improvement in exercise tolerance in the cell therapy group when compared with the control population (SMD, 0.31; 95% CI, 0.08 to 0.55; P=0.008; F=0%; Figure 5A).

Left Ventricular Function
Three studies reported measurements of left ventricular (LV) ejection fraction.15,16,19 LV ejection fraction was measured by

Figure 4. Forest plot showing the weighted differences (mean difference [MD] or standardized mean difference [SMD]) between the mean changes from baseline in indices of angina in patients with refractory angina who received cell-based therapy when compared with those received maximal medical therapy. A, MD in the frequency of angina episodes. B, SMD in use of antianginal medications. C, MD in Canadian Cardiovascular Society (CCS) class. CI indicates confidence interval; IV, inverse variance; and LD, low-dose group.
cardiac magnetic resonance imaging in 2 studies\textsuperscript{15,16} and by 3 modalities—SPECT, echocardiography, and ventriculography—in 1 study.\textsuperscript{19} Pooled data from the 3 studies suggested an improvement in LV function in the cell therapy group when compared with controls (SMD, 4.14; 95% CI, 1.86−6.41; \(P = 0.004\); \(I^2 = 0\%\); Figure 5B).

Quality of Life
Quality of life was assessed with the Seattle Angina Questionnaire in four studies, all of which reported an improvement in LV function in the cell therapy group when compared with controls (SMD, 4.14; 95% CI, 1.86−6.41; \(P = 0.004\); \(I^2 = 0\%\); Figure 5B).

Composite Cardiac End Point
All included studies analyzed myocardial infarction or mortality either as a clinical outcome or as a safety measure. Cardiac-related hospitalization was reported in 2 studies,\textsuperscript{18,19} The occurrence of myocardial infarction was reported in 2 studies,\textsuperscript{15,16} cardiac-related hospitalization in 2 studies,\textsuperscript{18,19} and mortality in 3 studies.\textsuperscript{14,15,19} The composite cardiac end point of all of these outcomes suggested a decreased risk of occurrence of MACE (odds ratio, 0.49; 95% CI, 0.25−0.98; \(P = 0.04\); \(I^2 = 0\%\); Figure 6A) in patients who received cell-based therapy.

Safety
All included studies evaluated the safety of cell-based therapy. Adverse events reported during follow-up were mortality, congestive heart failure, angina exacerbation, respiratory arrest, tumor occurrence, bleeding, renal insufficiency, and arrhythmias. With the exception of MACE (vide supra) and arrhythmias (atrial and ventricular; vide infra), the other adverse events were not either consistently evaluated in all studies or found to differ between the 2 groups; therefore, no meta-analyses were performed for these safety parameters.

Five studies evaluated the occurrence of arrhythmias (both atrial and ventricular).\textsuperscript{14,17,19} A pooled analysis revealed a decreased risk of arrhythmias in the cell-based therapy group (odds ratio, 0.25; 95% CI, 0.06−0.98; \(P = 0.05\); \(I^2 = 0\%\); Figure 6B) when compared with maximal medical therapy. Analysis of the composite cardiac end point was also a measure of safety and, as indicated above, suggested a decreased risk of MACE in the cell-based therapy group (Figure 6A).

Discussion
Administration of stem/progenitor cells is a new therapeutic approach with immense potential. In this meta-analysis of patients with angina refractory to medical therapy and ineligible for revascularization, we found a significant improvement in several indices of angina, namely, decreased frequency of angina episodes, improvement in CCS class, and decreased use of antianginal medications in cell-treated patients. These clinical changes were associated with an improvement in myocardial perfusion (as demonstrated by SPECT imaging), exercise tolerance, and LV ejection fraction and a marked decrease in MACE. Moreover, cell-based therapy was found to be safe. The present work is the largest and most current meta-analysis of cell therapy trials in RFA to date; the results provide strong evidence supporting a beneficial effect of cell-based treatments and a robust rationale for larger, definitive phase III trials.

The importance of this analysis stems from the fact that, although the individual studies of RFA conducted heretofore reported a favorable trend, they were limited by their small sample size and by the use of different primary end points. As a result, the effect of stem/progenitor cells in this patient population...
is unclear. The present analysis advances the field because it indicates that when all available RCTs are pooled and the same end points are evaluated across studies, cell therapy has a statistically significant, beneficial effect on not only indices of angina and myocardial perfusion but also clinical outcomes.

Although our analysis revealed an improvement in indices of angina, exercise tolerance, LV function, and myocardial perfusion in cell-treated patients, it did not demonstrate a reduction in mortality. An effect of cell therapy on mortality, however, would be difficult to detect because the studies included in this analysis were not large enough to assess this end point; they were designed to establish safety and provide initial evidence of symptomatic efficacy, not to show a decrease in mortality. Moreover, mortality in patients with RFA is low, making it difficult to demonstrate a significant change even when the 6 studies were pooled (total of 353 patients). Nevertheless, our analysis does suggest a decreased risk of MACE in patients who received cell therapy (Figure 5A). The reduction in MACE in the absence of significant changes in mortality in this study is consistent with a recent analysis of a database of patients with RFA, which has shown a low mortality but a relatively high incidence of death, MI, and cardiac-related hospitalizations in this population. Together with the results of that study, the present investigation suggests that measures of morbidity and MACE are more appropriate end points than mortality when designing trials in this patient population.

The mechanism whereby cell therapy produces clinical improvement in patients with refractory angina is unclear. Several lines of evidence suggest that the salubrious effects of cell therapy are secondary to paracrine actions of transplanted cells that promote neovascularization and collateral perfusion. Even in the setting of a compromised macrovascular supply, improvement in microvascular and collateral perfusion can augment contractile function. The concept that cell therapy promotes neovascularization is supported by our finding that, in treated patients, there was an increase in myocardial perfusion, as assessed by SPECT (Figure 3). Although SPECT is a good tool to detect changes in myocardial perfusion, its ability to do so may be limited in this patient population because the frequent presence of multivessel disease reduces the relative magnitude of changes in perfusion. Despite these limitations, however, our analysis was able to show a beneficial effect of cell therapy on myocardial perfusion, as evidenced by the changes in summed stress scores. An abnormal perfusion scan has been reported to be a surrogate measure for adverse clinical outcomes.

When compared with a previous meta-analysis of cell therapy in RFA, our study is based on a comprehensive literature search that includes the largest number of relevant studies to date. Our study expands on that previous study by including more functional and clinical end points, by demonstrating improved myocardial perfusion, and by assessing the effect of cell-based therapy on a composite clinical cardiovascular end point. Thus, the present work adds substantially to the existing evidence in support of cell therapy in RFA.

Some limitations of the present analysis need to be discussed. The robustness of the evidence suggesting a beneficial effect of cell therapy was negatively affected by the small sample sizes and, in some cases, the short (<1 year) follow-up periods of the studies included, as well as by the variance in the results of individual studies. This variation may be partly explained by differences in cell type, cell dosage, cell isolation protocols, cell delivery methods, perfusion scores measured, and definition of successful outcome, as well as by the heterogeneity encountered in some of the analyses. All of the heterogeneity was accounted for by 1 study, which differed from the other studies with respect to patients' baseline
characteristics, coexisting diseases, need for prior coronary revascularization, and use of concomitant medications. Removal of that study affected the magnitude of the beneficial effect of cell-based therapy but did not change the direction of the results, which remained in favor of the treated group.

It should be pointed out that although the present study supports the utility of stem/progenitor cell administration in RFA, meta-analyses cannot demonstrate therapeutic efficacy. Larger phase III trials are needed to provide definitive evidence, evidence sufficient to lead to Food and Drug Administration approval and routine use of cell-based therapy in this clinical setting. Nonetheless, when evaluating the role of cell-based therapy in RFA, the lessons learned from the early-phase clinical trials reviewed herein will be important for designing future studies. These phase I/II studies used either cells that expressed markers of endothelial progenitor cells (CD34+ and CD133+) or unfractoned bone marrow mononuclear cells. On the basis of current evidence, both of these cell types show promise, and it would be difficult to formulate a recommendation as to which type should be tested further. MSCs could be another option, as they have been reported to have a beneficial effect in RFA even when used alone and can be obtained from healthy donors and used in an allogeneic manner because of their immunoprivileged status.

Furthermore, additional studies should be designed to include standardized quantitative assessment of myocardial perfusion, quality of life measures, and MACE as measures of efficacy. Although most of the included studies used SPECT imaging to assess changes in myocardial perfusion, this method is limited by poor spatial resolution, long acquisition protocols, and, most importantly, the occurrence of balanced flow reduction (eg, in multivessel disease). Other available modalities, such as positron emission tomography and cardiac magnetic resonance imaging, can be used with better capabilities to detect regional and global myocardial perfusion. It would seem more appropriate to use multimodality imaging with a combination of positron emission tomography and cardiac magnetic resonance imaging to assess both anatomic and functional changes after cell-based therapy. In addition to objective evidence of changes in myocardial perfusion, it is important to demonstrate improvement in clinical outcomes. Relevant measures of efficacy should be not only indices of angina but also quality of life measures. Moreover, as mentioned above, a composite cardiac end point that includes cardiac mortality, myocardial infarction, and cardiac-related hospitalization would be most useful to assess efficacy.

In conclusion, the clinical trials of cell therapy conducted heretofore in patients with angina refractory to medical treatment and not amenable to revascularization (no-option angina) are limited by the small sample size and, in some cases, the short-follow-up period, making it difficult to discern an efficacy signal. Individually, these studies have been mostly inconclusive. These limitations can be overcome, in part, by a meta-analysis. The present analysis, based on a total of 353 patients from 6 RCTs, suggests that cell-based therapies are not only safe but also lead to an improvement in indices of angina, exercise tolerance, LV function, and myocardial perfusion and a decrease in MACE. These encouraging results provide a strong rationale for conducting larger, rigorous RCTs to conclusively determine the efficacy of cell-based therapy in this problematic patient population in which few, if any, options are currently available.

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Disclosures
None.

References
8. Kinnaird T, Stabile E, Burnett MS, Lee CW, Barr S, Fuchs S, Epstein SE. Marrow-derived stromal cells express genes encoding a broad spectrum of arteriogenic cytokines and promote in vitro and in vivo arteriogenesis through paracrine mechanisms. Circ Res. 2004;94:678–685. doi: 10.1161/01.RES.0000118601.37875.AC.
16. van Ramshorst J, Bax JJ, Beeres SL, Dilbets-Schneider P, Roes SD, Stokkel MP, de Roos A, Fibbe WE, Zwaginga JJ, Boersma E, Schalij MJ, Atzema DF. Intramyocardial bone marrow cell injection for chronic myocardial

Novelty and Significance
What Is Known?
• Cell-based therapies have the potential to promote neovascularization and endothelial repair and consequently myocardial perfusion.
• Several phase I–II studies have been conducted to assess the effect of cell therapy in patients with angina refractory to conventional medical therapy and ineligible for coronary revascularization (refractory or no-option angina).
• These early phase I–II studies are limited by their small size and lack of uniform primary end points and clinical outcomes, making it difficult to discern an efficacy signal.
• There is no phase III trial currently underway; thus, the effect of cell-based therapies on myocardial perfusion and clinical outcomes in refractory angina remains unclear.

What New Information Does This Article Contribute?
• When the same end points are evaluated across studies, cell therapy has a significant beneficial effect on not only indices of angina and myocardial perfusion but also clinical outcomes.
• As none of the studies performed to date was powered to assess clinical end points, we combined the cardiovascular outcomes into a composite cardiac end point (major adverse cardiac events); our analysis shows that cell-based therapies lead to an improvement in clinical outcomes as demonstrated by a reduction in major adverse cardiac events.
• Our results provide strong evidence supporting a beneficial effect of cell-based treatments and a robust rationale for larger, definitive phase III trials.

Cell-based therapies have shown safety and efficacy in several early proof-of-concept studies. However, these studies, although showing safety and efficacy, are limited because they evaluated different end points and also were not powered enough to assess clinical outcomes. Results of our meta-analysis suggest that cell-based therapies lead to improvement in not only myocardial perfusion but also functional indices of angina and to reduction in a composite cardiac end point that includes myocardial infarction, cardiac-related hospitalization, and mortality. Although the present analysis supports the utility of cell-based therapies in ischemic heart disease and refractory angina, it cannot demonstrate therapeutic efficacy. Larger phase III studies are needed to provide definitive evidence of cell therapy in patients with refractory angina.
Impact of Cell Therapy on Myocardial Perfusion and Cardiovascular Outcomes in Patients With Angina Refractory to Medical Therapy: A Systematic Review and Meta-Analysis
Abdur Rahman Khan, Talha A. Farid, Asif Pathan, Avnish Tripathi, Shahab Ghafghazi, Marcin Wysoczynski and Roberto Bolli

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Supplementary Table I. Change in SPECT Scores Between the Two Groups

<table>
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<th>Study</th>
<th>Month</th>
<th>Cell-based Therapy Group</th>
<th>Maximal Medical Therapy Group</th>
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<tr>
<td><strong>Change in Summed Stress Scores</strong></td>
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<td></td>
<td></td>
</tr>
<tr>
<td>Tse(^1)</td>
<td>6 Months</td>
<td>-0.5 ± 5</td>
<td>2.4 ± 6.9</td>
</tr>
<tr>
<td>Ramshorst(^2)</td>
<td>3 Months</td>
<td>-3.4 ± 6.6</td>
<td>-1.1 ± 7.7</td>
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<tr>
<td>Losordo(^3) (LD)</td>
<td>6 Months</td>
<td>-117.4 ± 221.2</td>
<td>0.1 ± 161.1</td>
</tr>
<tr>
<td>Quevedo(^4)</td>
<td>6 Months</td>
<td>-0.5 ± 2.2</td>
<td>0 ± 1.9</td>
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<tr>
<td><strong>Change in Summed Rest Scores</strong></td>
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<td></td>
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</tr>
<tr>
<td>Ramshorst(^2)</td>
<td>3 Months</td>
<td>-0.6 ± 5.5</td>
<td>-0.3 ± 7.2</td>
</tr>
<tr>
<td>Quevedo(^4)</td>
<td>6 Months</td>
<td>-0.5 ± 2.7</td>
<td>-0.5 ± 1.3</td>
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<tr>
<td><strong>Change in Summed Difference Scores</strong></td>
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<td></td>
<td></td>
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<tr>
<td>Losorno(^5)</td>
<td>3 Months</td>
<td>-2.4 ± 4.4</td>
<td>-1.5 ± 1.6</td>
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<tr>
<td></td>
<td>6 Months</td>
<td>-2.3 ± 3.1</td>
<td>-0.7 ± 2</td>
</tr>
<tr>
<td>Tse(^1)</td>
<td>6 Months</td>
<td>-1.8 ± 3.6</td>
<td>0.8 ± 4.9</td>
</tr>
</tbody>
</table>

*Abbreviations: SPECT, single photon emission computed tomography; LD, Low dose*


