The IMPACT-CABG trial: A multicenter, randomized clinical trial of CD133$^+$ stem cell therapy during coronary artery bypass grafting for ischemic cardiomyopathy

Nicolas Noiseux, MD, MSc,[a,b,c] Samer Mansour, MD,[b,d,e] Richard Weisel, MD,[g,h] Louis-Mathieu Stevens, MD, PhD,[a,b,c] Shant Der Sarkissian, PhD,[b,c] Katherine Tsang, RN, MN,[g,h] Andrew M. Crean, MRCP,[g,i] Eric Larose, MD,[f] Shu-Hong Li, MD,[g,h] Bernd Wintersperger, MD,[g,i] Minh Quan Vu, MD,[b] Ignacio Prieto, MD,[a,c] Ren-Ke Li, MD, PhD,[g,h] Denis Claude Roy, MD,[c,j] and Terrence M. Yau, MD, MSc[g,h]

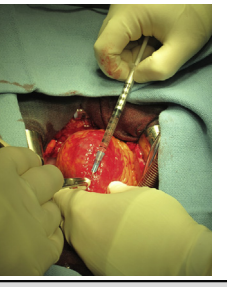
ABSTRACT

Objectives: The IMPACT-CABG trial is the first North American multicenter phase II randomized study of intramyocardial delivery of autologous CD133$^+$ stem cells in patients with chronic ischemic cardiomyopathy undergoing coronary artery bypass grafting. The primary objective was to demonstrate safety, including freedom from major adverse cardiac events. The secondary objective was to evaluate feasibility of same-day autologous cell preparation. Although the trial was not powered to evaluate LV function, exploratory data were collected.

Methods: After 7 open-label patients who received cells, patients randomly received stem cells or placebo (N = 40 total, 20 per center). After completion of coronary anastomoses, up to 10 million CD133$^+$, CD34$^+$, CD45$^+$ triple-positive cells or placebo were injected into the infarct and border zones. Patients were followed up clinically and underwent magnetic resonance imaging preoperatively and after 6 months.

Results: There were no procedural complications from bone marrow isolation and cell injection, no in-hospital mortality, and no protocol-related complications. Four patients had transient renal insufficiency, with 1 death during 6-month follow-up. Magnetic resonance imaging revealed that left ventricular volumes and ejection fractions improved in all patients (no difference between groups).

Conclusions: The trial successfully met both primary and secondary objectives, demonstrating that same-day isolation and autologous CD133$^+$ cell delivery with coronary artery bypass grafting is safe and feasible. The positive findings support a larger randomized, multicenter trial, with higher numbers of transplanted cells to demonstrate beneficial effects. The upcoming IMPACT-CABG II trial will evaluate higher cell doses and pharmacologic enhancement to determine whether these cells improve perfusion and myocardial function. (J Thorac Cardiovasc Surg 2016;152:1582-8)



Intramyocardial CD133$^+$, CD34$^+$, CD45$^+$ injection into infarct zone and penumbra during CABG.

Central Message

CD133$^+$ stem cell therapy for patients with ischemic cardiomyopathy is safe and feasible when using a same-day cell isolation protocol.

Perspective

The optimal cell type and broader applicability of cell therapies for ischemic cardiomyopathy remain controversial. Our findings suggest that same-day autologous CD133$^+$ cell therapy is safe and may be more widely applicable than has been possible in the past. Further studies are necessary to demonstrate myocardial repair for chronic ischemic cardiomyopathy and how to improve protocols.

See Editorial Commentary page 1589.

From the Divisions of [a]Cardiac Surgery and [d]Cardiology, Centre Hospitalier de l'Université de Montréal (CHUM); [b]Centre Hospitalier de l'Université de Montréal Research Center (CRCHUM); Departments of [c]Surgery and [e]Medicine, Université de Montréal; [f]Institut Universitaire de Cardiologie et de Pneumologie de Québec, Quebec City; [g]Peter Munk Cardiac Centre, University Health Network; and [h]Division of Cardiovascular Surgery, Department of Surgery, and [i]Division of Medical Imaging, University of Toronto, Toronto, Ontario; and [j]Department of Hematology, Hôpital Maisonneuve-Rosemont (HMR), Montréal, Québec, Canada.

Supported by Miltenyi Biotec, who provided financial and in-kind support for the conduct of this study but who had no role in the collection, analysis, or interpretation of the data, the writing of the report, or the decision to submit the report for publication. The authors attest that they have had full freedom to explore the data and analyze the results independently of the sponsor, and that they have had sole authority to make the final decision to submit the material for publication. The authors attest that they have no conflicts of interest related to the content of this article.

Institutional review board approval numbers and dates: Toronto, 11-0304B, August 26, 2011; Montreal, 08.147, November 27, 2008.

Clinicaltrials.gov registration numbers NCT01033617, NCT01467232.

Read at the 96th Annual Meeting of The American Association for Thoracic Surgery, Baltimore, Maryland, May 14-18, 2016.

Received for publication May 3, 2016; revisions received June 15, 2016; accepted for publication July 1, 2016; available ahead of print Sept 21, 2016.

Address for reprints: Terrence M. Yau, MD, MSc, Toronto General Hospital, University Health Network, 4N-470, 200 Elizabeth St, Toronto, Ontario M5G 2C4, Canada (E-mail: terry.yau@uhn.ca). or Nicolas Noiseux, MD, MSc, Centre Hospitalier de l'Université de Montréal (CHUM), 3840 Rue Saint-Urbain, Montréal, Québec H2W 1T8, Canada (E-mail: noiseuxn@videotron.ca).

0022-5223/$36.00

http://dx.doi.org/10.1016/j.jtcvs.2016.07.067

Abbreviations and Acronyms
CABG = coronary artery bypass grafting
LV = left ventricular
LVEF = left ventricular ejection fraction
MRI = magnetic resonance imaging
BM = bone marrow

Scanning this QR code will take you to the supplemental video, figure, and table for this article.

Cell therapy may improve healing of the heart, repopulate injured myocardium, restore cardiac function, and treat heart failure.[1] The feasibility and safety have been well established,[2,3] but varying cell lineages and protocols have yielded conflicting results.[4]

The multipotent hematopoietic progenitor $CD133^+$ cells have high engraftment, proangiogenic capacity, and antiapoptotic effects in ischemic tissue[5] via paracrine mechanisms and differentiation into newly forming vessels.[6] Numerous clinical trials have reported improved repair of infarcted hearts or ischemic limbs after infusion of $CD133^+$ stem cells.[7] The COMPARE-AMI trial, the first randomized, controlled trial to evaluate intracoronary injection of autologous $CD133^+$ stem cells into an acute myocardial infarction, noted no safety concerns in the patients receiving cells.[2,8] Data from the Intramyocardial Transplantation of Bone Marrow Stem Cells for Improvement of Postinfarct Myocardial Regeneration in Addition to CABG Surgery: A Controlled, Prospective, Randomized, Double Blinded Multicenter Trial (PERFECT) trial, which is randomly allocating patients undergoing coronary artery bypass grafting (CABG) to receive $CD133^+$ stem cells or placebo, are anticipated shortly.[9]

The main objective of the IMPACT-CABG trial[3,10] was to evaluate the safety and feasibility of intramyocardial injection of autologous $CD133^+$ cells in patients with chronic ischemic cardiomyopathy undergoing CABG in 2 high-volume academic cardiac surgery centers in Canada. Although the trial was not powered to evaluate left ventricular (LV) function, as a secondary objective we assessed changes in LV end-systolic and end-diastolic volumes and LV ejection fraction (LVEF).

MATERIALS AND METHODS

This study was approved by the research ethics boards of the respective institutions and by Health Canada. All patients provided written, informed consent. Patients eligible for the study were those with indications for CABG and moderately depressed LVEF (25%-45%) as a result of chronic ischemic cardiomyopathy. Detailed patient inclusion and exclusion criteria are presented in Table E1; the protocol and details have been published previously.[3,10]

Patients underwent routine preoperative clinical investigations and study-specific echocardiography and cardiac magnetic resonance imaging (MRI) preoperatively and 6 months postoperatively. The study interventions and follow-up are detailed in Figure E1.

The primary safety end point was defined as freedom from major adverse cardiac events, including freedom from cardiac death, myocardial infarction, repeat CABG or percutaneous intervention of a bypassed coronary artery, or severe cardiac arrhythmias (sustained ventricular tachycardia or resuscitation from an episode of sudden death) during 6 months of follow-up. The primary feasibility end point was to determine in what proportion of patients our $CD133^+$, $CD34^+$, $CD45^+$ cell–preparation protocol resulted in a final cell product meeting all prespecified and Health Canada–approved release criteria (Table 1). A secondary exploratory efficacy end point was to evaluate the effect of intramyocardial cell injections on changes in global LV function and volumes, as assessed by MRI.

Echocardiography

Echocardiography was performed at baseline to screen for patients with a LVEF between 25% and 45% and included parasternal long- and short-axis views, as well as apical 4- and 2-chamber views. Regional wall motion scores and global LVEF (by Simpson's biplane method) were assessed and LV dimensions measured from M-mode recordings in parasternal views.

Cardiac MRI

Cardiac MRI was performed on 1.5-T systems in short-axis orientation, including standard cine steady state free precession imaging and stress/rest first-pass perfusion imaging (3 short-axis slices) with a saturation-recovery spoiled gradient recalled echo technique with bolus injection of 0.05 mmol/kg gadolinium-based contrast agent. After a 0.1-mmol/kg gadolinium-based contrast agent top up bolus, late gadolinium enhancement imaging was acquired 10 to 15 minutes after the rest perfusion with a 2-dimensional segmented inversion recovery gradient recalled echo technique with individual inversion time optimization.[8,9] Images were transferred for off-line processing with dedicated algorithms (cvi42; Circle Cardiovascular Imaging, Calgary, Alberta, Canada).

Randomization and Blinding

Sequentially numbered envelopes containing computer-generated randomization sequences, stratified by site and in randomly permuted blocks of 4, were generated and used to determine group assignment for each patient. The patients, surgical team, study coordinators, and all personnel involved in patient and outcome assessment remained blinded until after the last patient had completed the 6-month follow-up evaluation. Random assignment was performed on the day of CABG during the cell-processing phase by the unblinded cell team.

Bone Marrow Harvesting and Cell Processing

On the morning of planned CABG surgery, patients were administered local anesthesia under light sedation, and 100 to 150 mL of bone marrow (BM) was aspirated from the posterior iliac crest (Video 1). $CD133^+$ stem cells were selected with the CliniMACS $CD133^+$ Reagent System (Miltenyi Biotec GmbH, Bergisch Gladbach, Germany) through the use of paramagnetic beads composed of iron-dextran particles conjugated with anti-CD133 monoclonal antibodies. After labeling, cells were separated with a high-gradient magnetic separation column. After purity and viability evaluation, negative endotoxin measurement, and initiation of microbiologic cultures, a cell preparation containing a minimum of 0.5 million and a maximum of 10 million $CD133^+$, $CD34^+$, $CD45^+$ triple-positive cells was delivered to the operating room for patients randomly assigned to receive cells; control patients received a preparation of patient-derived plasma.

TABLE 1. Cell product release criteria

- Total volume of 2-2.5 mL of suspension.
- $CD133^+$ cell number lower limit of 0.5×10^6 and upper limit of 10×10^6.
- $CD133^+$ cell purity >30%.
- $CD133^+$ cell recovery >10%.
- Viability >70%.
- Negative results of endotoxin screen.

Samples from the cell preparation were also tested for sterility. Specimens were sent for culture, although the final results were not available before administration of the cell product.

Surgical Procedure

On the day of cell preparation, patients underwent conventional CABG surgery with cardiopulmonary bypass and cardioplegic arrest. After completion of the distal coronary anastomoses and before removal of the aortic crossclamp, cells were injected into the revascularized infarct and border zone, as identified preoperatively by MRI. Multiple (10-15) direct intramyocardial injections of 0.2-mL aliquots of the cell suspension or autologous plasma control preparation were performed.

Patients and Protocol Differences

The study protocol was originally conceived and written by the Montreal team (N.N.) and registered in 2009 on clinicaltrials.gov as NCT01033617. The protocol was subsequently modified for the Toronto patients (T.M.Y.). Differences in the protocols at the 2 sites included the number of initial open-label patients (5 in Montreal vs 2 in Toronto) and the randomization ratio (2:1 cells:placebo in Montreal vs 1:1 in Toronto). In Montreal, cell processing was performed in a separate cell-processing facility, whereas in Toronto the cell processing took place in a dedicated regenerative medicine laboratory within the operating suites. Because of these protocol differences, the Toronto arm of the trial was registered in 2011 as NCT01467232. There were no changes to the respective study protocols during the conduct of the trial.

Study Oversight and Monitoring

A separately constituted data safety and monitoring board reviewed the progress of the study, all serious adverse events, and blinded outcomes at 6-month intervals. The data safety and monitoring board recommended continuation of the trial after each review.

Statistical Analysis

Continuous variables are expressed as mean ± SD or median and interquartile range, and analyzed with the Student *t* test or Wilcoxon test as appropriate. Categoric variables are expressed as a number and percentage and compared using the Fisher exact test. Mixed effects models were used to assess changes in MRI measures from baseline to 6 months, accounting for the correlation between repeated measurements, and stratifying results by center (PROC MIXED, SAS software version 9.4; SAS Institute, Inc, Cary, NC). Between-group differences as well as changes with time were assessed. Patients with missing values for the 6-month MRI assessment were excluded from the analyses.

RESULTS

Patients and Study Flow

The flow of patients through the study is presented in Figure 1. A total of 59 patients were screened, of whom 41 (69%) were eligible. Among the eligible patients, all 41 agreed to participate in the trial. Seven of these patients (5 in Montreal, 2 in Toronto) received open-label treatment at the beginning of the study. In 1 open-label case, the cell preparation did not meet release criteria (complete description in a later portion of the Results section). This patient was withdrawn from the study and replaced according to our prespecified protocol, but constitutes part of the cohort analyzed for safety and feasibility. The randomized cohort comprised the remaining 33 patients: 15 in Montreal who were randomly allocated in a 2:1 ratio (10 cells and 5 placebo) and 18 in Toronto who were randomly allocated in a 1:1 ratio. The first patient was enrolled on November 30, 2010, and the last patient completed 6-month follow-up on December 24, 2014.

VIDEO 1. Bone marrow aspiration in the iliac crest is performed under local anesthesia and light sedation the morning of the surgery. The bone marrow is then transferred into a sterile bag containing heparin and sent to the cell preparation lab for $CD133^+$ stem cell isolation with the CliniMACS system (Miltenyi Biotec GmbH, Bergisch Gladbach, Germany). After completion of the coronary artery bypass grafts, during cardiopulmonary bypass with cardioplegic arrest, the stem cell or placebo preparation is injected into the infarct and peri-infarct regions (10-15 injections, 2.0 mL total volume). Video available at http://www.jtcvsonline.org/article/S0022-5223(16)30914-X/addons.

Demographic Data

The demographic and intraoperative data of the entire cohort of 41 patients enrolled, detailed by group assignment, are presented in Table 2. Most patients were male, and hypertension, diabetes, and peripheral vascular disease were prevalent. Most patients had New York Heart Association class 3 or 4 symptoms and had had one or more myocardial infarctions more than 6 months preoperatively. Mean LVEF, as measured by preoperative MRI and by echocardiography, was ≤40% in all groups.

BM Harvest and Cell Processing

BM harvesting was well tolerated and was performed successfully in all 41 patients, without any complications. Cell processing was performed in the open-label cases and for 19 patients randomly allocated to cell administration. In 1 open-label patient, the total number of $CD133^+$, $CD34^+$, $CD45^+$ cells fell below the minimum level to meet the release criteria. Review of the cell isolation process revealed no potential reasons for the low final cell

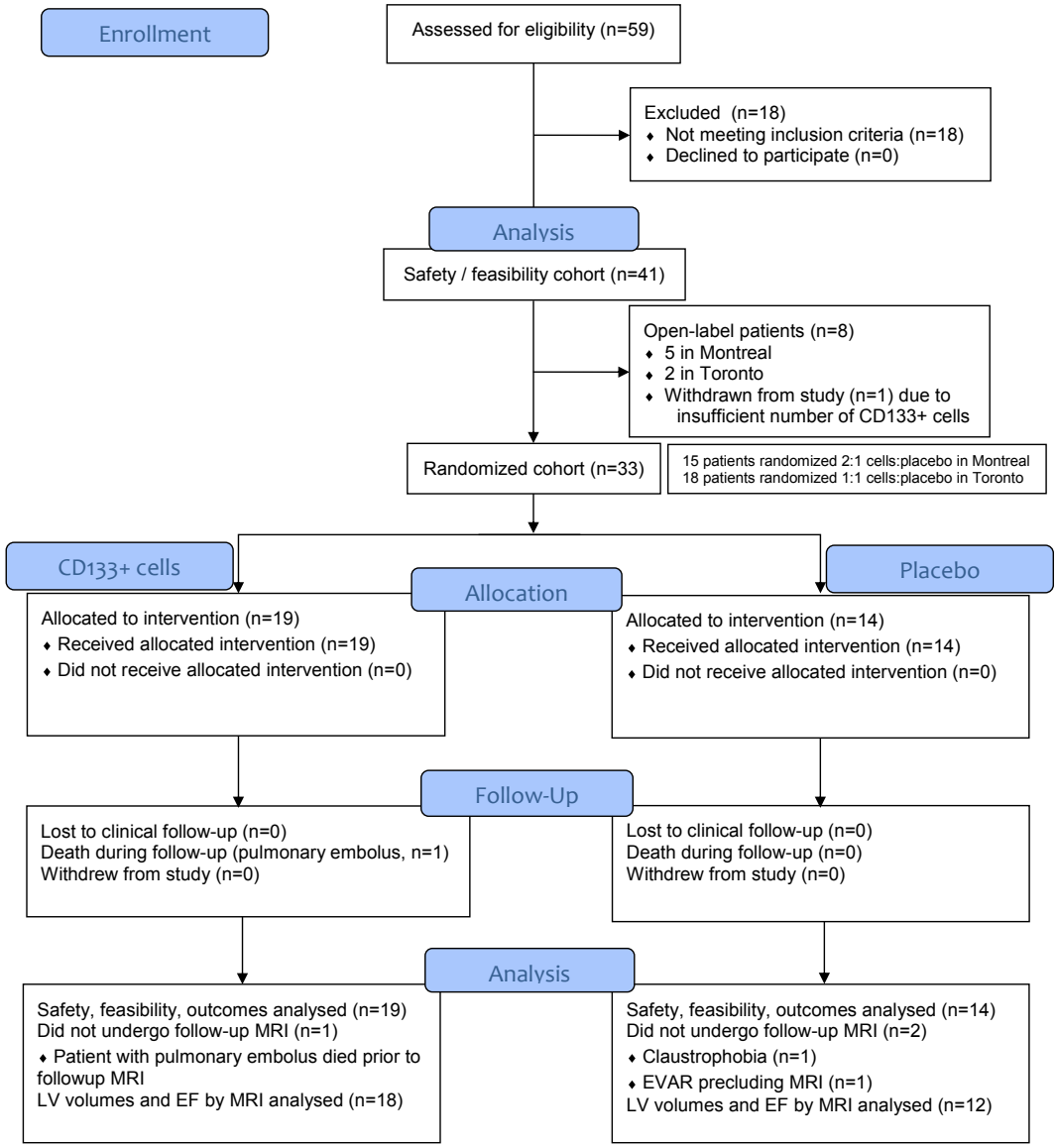


FIGURE 1. Study flow diagram indicating number of patients screened, enrolled, randomly allocated, and analyzed. *MRI*, Magnetic resonance imaging; *LV*, left ventricle; *EF*, ejection fraction; *EVAR*, endovascular aneurysm repair.

counts other than low initial prevalence of $CD133^+$ cells in the BM aspirate. For 26 of 27 patients (96%), a cell preparation meeting all release criteria was therefore obtained.

The number of $CD133^+$, $CD34^+$, $CD45^+$ cells after enrichment on the magnetic column was 10.5 ± 9.6 million (mean $\pm$ SD), for a purity of $63.3\% \pm 15.5\%$ and an enrichment of 165 ± 79 fold. The mean number of cells administered to patients was 6.5 ± 3.1 million (range, 0.8-9.8 million).

Perioperative Data and Clinical Outcomes

The durations of cardiopulmonary bypass and aortic crossclamping were similar in all groups, as were the number of grafts constructed and the number of cell injection sites (Table 2). In all cases, injections of cells or placebo were performed in areas of myocardium that also received a coronary bypass graft, with the exception of the single case of a patient (randomly allocated to the placebo group) with a chronically occluded, small right coronary artery.

There was no hospital mortality, stroke, perioperative myocardial infarction, or reexploration for bleeding in any patient (Table 3). One patient had a seizure in the early postoperative period but recovered with no neurologic sequelae, no recurrence, and no abnormality on computed tomographic and MRI scans of the brain. Four patients (9.8%) had a transient postoperative increase in serum creatinine, but no patient required dialysis. One patient died during the follow-up period, confirmed on

TABLE 2. Demographic and intraoperative data

	Total cohort	Randomly allocated to CD133$^+$ cells	Randomly allocated to placebo	*P* value CD133$^+$ cells vs placebo
Patients	41	19	14	
Baseline characteristics				
Age (y, mean ± SD)	65.2 ± 7.2	66.4 ± 6.5	63.1 ± 7.2	.18
Male	38 (92.7%)	17 (89.5%)	13 (92.9%)	.74
BMI (kg/m^2, mean ± SD)	28.2 ± 5.1	28.8 ± 6.2	29.1 ± 4.0	.88
Hypercholesterolemia	38 (92.7%)	18 (94.7%)	13 (92.9%)	.82
Currently smoking	9 (22.0%)	3 (15.8%)	4 (28.6%)	.66
Hypertension	34 (82.9%)	16 (84.2%)	10 (71.4%)	.37
Diabetes mellitus	17 (41.5%)	10 (52.6%)	3 (21.4%)	.07
Previous stroke	6 (14.6%)	2 (10.5%)	2 (14.3%)	.74
Peripheral vascular disease	11 (26.8%)	4 (21.1%)	4 (28.6%)	.62
Recent (<6 mo) MI	19 (46.3%)	7 (36.8%)	8 (57.1%)	.25
Old (>6 mo) MI	23 (56.1%)	11 (57.9%)	9 (64.3%)	.71
Atrial fibrillation	5 (12.2%)	1 (5.3%)	4 (28.6%)	.06
Chronic renal insufficiency	2 (4.9%)	1 (5.3%)	1 (7.1%)	.82
Anemia	16 (39.0%)	7 (36.8%)	6 (42.9%)	.73
euroSCORE (median and IQR)	3.8 (2.5-5.0)	4.0 (2.2-5.8)	3.2 (2.0-4.2)	.37
NYHA class III-IV symptoms	28 (68.3%)	12 (63.2%)	11 (78.6%)	.49
LVEF (mean ± SD)				
Echocardiography	39% ± 7%	40% ± 7%	40% ± 7%	.98
MRI	37% ± 6%	37% ± 6%	36% ± 7%	.57
Intraoperative characteristics				
No. of grafts (mean ± SD)	3.2 ± 0.8	3.4 ± 1.0	3.0 ± 0.7	.17
Incomplete revascularization	4 (9.8%)	2 (10.5%)	2 (14.3%)	.74
Crossclamp (min, mean ± SD)	63.3 ± 19.4	64.2 ± 20.7	64.1 ± 15.6	>.999
Cardiopulmonary bypass (min, mean ± SD)	82.8 ± 24.1	82.7 ± 24.2	81.9 ± 21.7	.91
No. of puncture sites (mean ± SD)	12.9 ±2.8	13.8 ± 2.2	12.8 ± 0.9	.09

Data are number and percentage except as noted. *SD*, Standard deviation; *BMI*, body mass index; *MI*, myocardial infarction; *IQR*, interquartile range; *NYHA*, New York Heart Association; *LVEF*, left ventricular ejection fraction; *MRI*, magnetic resonance imaging.

postmortem examination to be the result of a massive pulmonary embolus. There were no serious adverse events related to the BM harvest or cell injection during hospitalization or follow-up. All patients were in New York Heart Association functional class I or II at 6 months, with the exception of 1 patient in the cell-treated group who had New York Heart Association class III symptoms. Similarly, all patients were in Canadian Cardiovascular Society angina class I at 6 months of follow-up, with the exception of 1 patient in the placebo group and 1 patient in the cell-treated group.

TABLE 3. Clinical outcomes

Outcomes	Total cohort	CD133$^+$ cells	Placebo	*P* value
Patients	41	19	14	
Hospital mortality	0	0	0	>.999
Perioperative MI	0	0	0	>.999
Stroke	0	0	0	>.999
Seizure	1 (2.4%)	1 (5.3%)	0	.38
Increased creatinine	4 (9.8%)	1 (5.3%)	1 (7.1%)	.82
Resternotomy	0	0	0	>.999
Infection	1 (2.4%)	1 (5.3%)	0	.38
Delirium	4 (9.8%)	2 (10.5%)	1 (7.1%)	.74
Late mortality	1 (2.4%)	1 (5.3%)	0	.38
Late infection	1 (2.4%)	1 (5.3%)	0	.38

Data are number and percentage. *MI*, Myocardial infarction.

LV Volumes and Ejection Fractions

LV end-diastolic and end-systolic volume indices were significantly reduced 6 months postoperatively relative to preoperative values in patients receiving cells or placebo, with no difference between groups (Table 4). LVEF was significantly increased during the 6-month interval in both cell and placebo groups, to a similar degree. There was no correlation between the number of cells infused and changes in LV volumes or LVEF.

DISCUSSION

The main objective of the IMPACT-CABG trial was to demonstrate the safety of intramyocardial injection of autologous CD133$^+$, CD34$^+$, CD45$^+$ cells in patients with ischemic cardiomyopathy undergoing CABG. Despite initial concerns about BM aspiration in patients with ischemic cardiomyopathy before revascularization, all patients underwent BM aspiration with no undue discomfort, signs or symptoms of myocardial ischemia, hemodynamic

TABLE 4. Left ventricular dimensions and ejection fraction according to magnetic resonance imaging at baseline and 6 months

Randomization group	Time		*P* value	
	Baseline	6 mo	Baseline vs 6 mo	$CD133^+$ vs placebo
LVEDVI				.91
Placebo (n = 12)	107 ± 7	94 ± 7	.01	
$CD133^+$ cells (n = 18)	105 ± 5	93 ± 6	<.01	
LVESVI				.90
Placebo (n = 12)	67 ± 6	54 ± 5	.02	
$CD133^+$ cells (n = 18)	67 ± 4	53 ± 4	<.01	
LVEF				.14
Placebo (n = 12)	36 ± 2	48 ± 2	<.01	
$CD133^+$ cells (n = 18)	37 ± 2	44 ± 2	<.01	

Values are presented as estimate ± SE from a stratified mixed effect model with random effects for the patient and for the center. Only patients with data available at baseline and 6 months were included in this analysis. *LVEDVI*, Left ventricular end-diastolic volume index; *LVESVI*, left ventricular end-systolic volume index; *LVEF*, left ventricular ejection fraction.

changes, or hematoma formation. In 40 of 41 patients, direct intramyocardial injections of the cell product or placebo were carried out, with no evidence of intracoronary embolization, myocarditis, myocardial infarction or hematoma, malignant arrhythmias, or bleeding. Despite the high-risk profile of these patients, there was no hospital mortality, stroke or myocardial infarction. One patient died during the follow-up period, of a massive pulmonary embolus confirmed on postmortem examination. Thus there were no adverse events related to the protocol or any of the study interventions, and the safety of this approach to $CD133^+$ cell delivery appears to be excellent.

The second objective was to determine the feasibility of our same-day approach, in which BM harvesting is performed on the morning of the day of the operation. The cell isolation process takes approximately 4 to 5 hours, before which the preparation is delivered to the operating room for the patient to undergo CABG in the afternoon of the same day. A consequence, however, is that the number of $CD133^+$, $CD34^+$, $CD45^+$ cells that can be delivered to the patient depends on the volume of BM aspirated and the prevalence of the desired cell subtypes within the marrow. The ability to isolate cells was quite variable among patients and highlights the need for additional studies to define the nature of cells with reparative capacity. Autologous cell therapy is limited by the availability of cells and their reparative capacity, which reflects patient age,[11] sex, pharmacologic therapies, and comorbidities.[12,13] In 26 of 27 patients in whom cell isolation was performed (96.3%), we were nevertheless able to obtain a cell product meeting all release criteria. The mean number of cells administered in our study was 6.5 ± 3.1 million, which is comparable to the mean of 5.1 million $CD133^+$ stem cells reported in the Bypass Surgery and CD133 Marrow Cell Injection for Treatment of Ischemic Heart Failure (Cardio133) trial.[14] The feasibility of this approach therefore appears to be excellent as well.

We selected $CD133^+$, $CD34^+$, $CD45^+$ hematopoietic progenitor cells because they have vasculogenic properties that may improve perfusion in ischemic cardiomyopathy.[15-17] Whether improved perfusion alone can lead to improved function in patients with established myocardial infarction and chronic ischemic cardiomyopathy is as yet unclear. Two previous randomized trials have reported that $CD133^+$[18] or $CD34^+$[19] autologous BM stromal cells injected into the myocardium of patients undergoing CABG increased LVEF 6 months postoperatively relative to controls. These trials were not double-blind, however, nor were they controlled with placebo injections to compensate for angiogenesis that may result from needle trauma alone.[18,20] In addition, in these trials, LV function was quantitated by echocardiography rather than MRI, the current criterion standard. More recently, the Cardio133 trial (NCT00462774), which randomly assigned 60 patients undergoing CABG to receive injections of $CD133^+$ stem cells or placebo,[14] observed some improvements in scar size and regional perfusion but no effect on clinical symptoms or global LV function except in patients with a posterior myocardial infarction. The multicenter phase III PERFECT trial (NCT00950274),[9] which will also evaluate the effect of $CD133^+$ progenitor cell injection during CABG, is currently enrolling, and further information on the effect of these cells may be forthcoming soon.

The IMPACT-CABG trial was limited by a relatively small number of patients, which has been a common limitation of stem cell trials. We maintained strict inclusion and exclusion criteria, and our 2-center approach allowed us to increase the power of our study. This type of stem cell study requires highly selected patients, thus limiting enrolment. For this reason, Stamm and colleagues[18] had to modify inclusion criteria in their trial to accelerate recruitment. Recently, a similar trial investigating $CD133^+$ stem cells in patients undergoing CABG was terminated for lack of recruitment (NCT01721902). It is clear that carefully designed and adequately powered double-blind, randomized studies are needed to confirm the promising findings from early studies. Moreover, both cardiac repair and clinical outcomes must be assessed. In this regard, the An Efficacy, Safety, and Tolerability Study of Ixmyelocel-T Administered Via Transendocardial Catheter-Based Injections to Subjects With Heart Failure Due to Ischemic Dilated Cardiomyopathy (IDCM) (ixCELL DCM) study demonstrated, in the largest cell therapy trial to include patients with heart failure, a significant reduction in clinical cardiac events relative to placebo, but no improvement in LV dimensions or function (LVEF).[21]

Clearly, more work is needed to address further questions, including the most appropriate stem cell population and dose, and methods to improve the survival, engraftment and functionality of administered cells.[22,23] The upcoming

IMPACT-CABG II trial will evaluate a higher cell dose, but more importantly, it will examine how pharmacologic priming of the cells may improve cell retention and functional recovery.

In summary, the IMPACT-CABG trial has successfully demonstrated that a same-day approach to autologous $CD133^+$, $CD34^+$, $CD45^+$ hematopoietic progenitor cell therapy in patients with ischemic cardiomyopathy and moderately depressed LV systolic function undergoing CABG is safe, without adverse events related to cell harvesting or delivery, and feasible, with a 96% success rate in obtaining a cell preparation meeting stringent release criteria. Further investigation and additional trials will be required for careful elucidation of the effect of these cells on myocardial perfusion and function. If beneficial effects can be consistently demonstrated, however, this same-day autologous cell approach to stem cell therapy for myocardial repair may significantly increase the clinical application of stem cell therapies beyond the realm of only large institutions with dedicated good manufacturing process–compliant cell-processing facilities.

Conflict of Interest Statement

N.N., L.M.S., and S.M. are scholars of the Fonds de la Recherche en Santé du Québec (FRQS). T.M.Y. holds the Angelo & Lorenza DeGasperis Chair in Cardiovascular Surgery Research. All other authors have nothing to disclose with regard to commercial support. Partial funding for the study in Toronto was provided by the Innovation Fund of the Peter Munk Cardiac Centre. Miltenyi Biotec GmbH (Bergisch Gladbach, Germany), provided in-kind support for the trial in the form of the disposable cell isolation kits for the CliniMACS cell separation system for both the Montreal and Toronto sites. In addition, Miltenyi Biotec provided per-patient financial support for the trial in Montreal. The trial, data management and analysis, and publications were performed entirely by the investigators.

We thank Joannie Dionne, Bernard Lemieux, MD, Sowmya Viswanathan, MD, Matthew Scaife, and Laura Tumiati for their invaluable assistance, as well as the Centre of Excellence for Cellular Therapy (CETC) Hôpital Maisonneuve-Rosemont (HMR), Fonds de la Recherche du Québec Santé (FRQS)/Réseau de Thérapie Cellulaire et Tissulaire du FRQS (ThéCell).

References

1. Marquis-Gravel G, Stevens LM, Mansour S, Avram R, Noiseux N. Stem cell therapy for the treatment of nonischemic cardiomyopathy: a systematic review of the literature and meta-analysis of randomized controlled trials. *Can J Cardiol*. 2014;30:1378-84.
2. Qiu F, Maehara A, El Khoury R, Généreux P, LaSalle L, Mintz GS, et al. Impact of intracoronary injection of $CD133^+$ bone marrow stem cells on coronary atherosclerotic progression in patients with STEMI: a COMPARE-AMI IVUS substudy. *Coron Artery Dis*. 2016;27:5-12.
3. Forcillo J, Stevens LM, Mansour S, Prieto I, Salem R, Baron C, et al. Implantation of $CD133^+$ stem cells in patients undergoing coronary bypass surgery: IMPACT-CABG pilot trial. *Can J Cardiol*. 2013;29:441-7.
4. Afzal MR, Samanta A, Shah ZI, Jeevanantham V, Abdel-Latif A, Zuba-Surma EK, et al. Adult bone marrow cell therapy for ischemic heart disease: evidence and insights from randomized controlled trials. *Circ Res*. 2015;117:558-75.
5. Quirici N, Soligo D, Caneva L, Servida F, Bossolasco P, Deliliers GL. Differentiation and expansion of endothelial cells from human bone marrow CD133(+) cells. *Br J Haematol*. 2001;115:186-94.
6. Belotti D, Gaipa G, Bassetti B, Cabiati B, Spaltro G, Biagi E, et al. Full GMP-compliant validation of bone marrow-derived human CD133(+) cells as advanced therapy medicinal product for refractory ischemic cardiomyopathy. *Biomed Res Int*. 2015;2015:473159.
7. Bongiovanni D, Bassetti B, Gambini E, Gaipa G, Frati G, Achilli F, et al. The $CD133^+$ cell as advanced medicinal product for myocardial and limb ischemia. *Stem Cells Dev*. 2014;23:2403-21.
8. Mansour S, Roy DC, Bouchard V, Stevens LM, Gobeil F, Rivard A, et al. One-year safety analysis of the COMPARE-AMI trial: comparison of intracoronary injection of CD133 bone marrow stem cells to placebo in patients after acute myocardial infarction and left ventricular dysfunction. *Bone Marrow Res*. 2011;2011:385124.
9. Donndorf P, Kaminski A, Tiedemann G, Kundt G, Steinhoff G. Validating intramyocardial bone marrow stem cell therapy in combination with coronary artery bypass grafting, the PERFECT Phase III randomized multicenter trial: study protocol for a randomized controlled trial. *Trials*. 2012;13:99.
10. Forcillo J, Stevens LM, Mansour S, Prieto I, Roy DC, Noiseux N. IMPACT-CABG trial: implantation of CD133(+) stem cells in patients undergoing coronary bypass surgery—presentation of the first treated patient. *Case Rep Transplant*. 2011;2011:685394.
11. Lee MM, Li RK, Yau TM. Stem cell therapy for heart failure: out with the new and in with the old? *J Thorac Cardiovasc Surg*. 2015;150:1035-7.
12. Forbes SJ, Rosenthal N. Preparing the ground for tissue regeneration: from mechanism to therapy. *Nat Med*. 2014;20:857-69.
13. Mason C, Dunnill P. Assessing the value of autologous and allogeneic cells for regenerative medicine. *Regen Med*. 2009;4:835-53.
14. Nasseri BA, Ebell W, Dandel M, Kukucka M, Gebker R, Doltra A, et al. Autologous $CD133^+$ bone marrow cells and bypass grafting for regeneration of ischaemic myocardium: the Cardio133 trial. *Eur Heart J*. 2014;35:1263-74.
15. Agbulut O, Vandervelde S, Al Attar N, Larghero J, Ghostine S, Léobon B, et al. Comparison of human skeletal myoblasts and bone marrow-derived $CD133^+$ progenitors for the repair of infarcted myocardium. *J Am Coll Cardiol*. 2004;44:458-63.
16. Bartunek J, Vanderheyden M, Vandekerckhove B, Mansour S, De Bruyne B, De Bondt P, et al. Intracoronary injection of CD133-positive enriched bone marrow progenitor cells promotes cardiac recovery after recent myocardial infarction: feasibility and safety. *Circulation*. 2005;112(9 Suppl):I178-83.
17. Simonetti OP, Kim RJ, Fieno DS, Hillenbrand HB, Wu E, Bundy JM, et al. An improved MR imaging technique for the visualization of myocardial infarction. *Radiology*. 2001;218:215-23.
18. Stamm C, Kleine HD, Choi YH, Dunkelmann S, Lauffs JA, Lorenzen B, et al. Intramyocardial delivery of $CD133^+$ bone marrow cells and coronary artery bypass grafting for chronic ischemic heart disease: safety and efficacy studies. *J Thorac Cardiovasc Surg*. 2007;133:717-25.
19. Patel A, Geffner L, Vina RF, Saslavsky J, Urschel HC Jr, Kormos R, et al. Surgical treatment for congestive heart failure with autologous adult stem cell transplantation: a prospective randomized study. *J Thorac Cardiovasc Surg*. 2005;130:1631-8.
20. Shahzad U, Li G, Zhang Y, Yau TM. Transmyocardial revascularization induces mesenchymal stem cell engraftment in infarcted hearts. *Ann Thorac Surg*. 2012;94:556-62.
21. Patel AN, Henry TD, Quyyumi AA, Schaer GL, Anderson RD, Toma C, et al. Ixmyelocel-T for patients with ischaemic heart failure: a prospective randomised double-blind trial. *Lancet*. 2016;387:2412-21.
22. Gaffey AC, Chen MH, Venkataraman CM, Trubelja A, Rodell CB, Dinh PV, et al. Injectable shear-thinning hydrogels used to deliver endothelial progenitor cells, enhance cell engraftment, and improve ischemic myocardium. *J Thorac Cardiovasc Surg*. 2015;150:1268-76.
23. Zhang GW, Gu TX, Sun XJ, Wang C, Qi X, Wang XB, et al. Edaravone promotes activation of resident cardiac stem cells by transplanted mesenchymal stem cells in a rat myocardial infarction model. *J Thorac Cardiovasc Surg*. 2016;152:570-82.

Key Words: stem cell therapy, ischemic cardiomyopathy, randomized clinical trial, myocardial repair, $CD133^+$ cells

	Before Surgery Evaluation	Bone Marrow Aspiration	Day of Surgery	Post-op Prior to Discharge	First 3 Weeks After Discharge	Months					
						1	2	3	6	12	18
Medical History	X			X		X		X	X	X	X
Physical Examination	X	X		X		X		X	X	X	X
Questionnaires	X			X		X		X	X	X	X
ECG	X	X	X	X		X		X	X	X	X
Blood work (cardiac enzymes)	X	X	X	X							
Blood work (per-routine)	X	X	X	X		X		X	X	X	X
Holter ECG	X					X			X		X
Echocardiogram	X					X		X	X	X	X
Magnetic Resonance Imaging	X								X		
Bone Marrow Aspiration			X								
Heart Surgery & Cells/placebo Injection			X								
Telephone Follow-up					Every week		X				

FIGURE E1. Protocol-specified investigations and interventions. *ECG*, Electrocardiogram.

TABLE E1. Inclusion and exclusion criteria

Inclusion criteria

- Age ≥18 y and ≤75 y.
- Patients with severe chronic ischemic cardiomyopathy manifested by CCS class II or greater angina and/or NYHA class II or greater dyspnea, AND who have undergone diagnostic coronary angiography demonstrating ≥70% diameter narrowing of at least 2 major coronary arteries or branches or ≥50% diameter narrowing of the left main coronary artery.
- Significant left ventricular systolic dysfunction evaluated by echocardiography or LV angiography (LV ejection fraction ≤45% but ≥25%) from previous myocardial infarction. This area of LV dysfunction should be akinetic or severely hypokinetic, not dyskinetic or aneurysmal, when assessed by echocardiography or LV angiography.
- No contraindications or exclusions.
- Willingness to participate and ability to provide informed consent.

Exclusion criteria (patients who satisfy ≥1 of the following criteria are not considered for initial inclusion in the study):

- Contraindications to MRI including presence of an ICD or PPM, or cases in which it is anticipated that an ICD or PPM will be implanted before 6-mo follow-up, or patients with claustrophobia (thus precluding performance of follow-up MRI scans).
- Need for urgent or emergency revascularization.
- Anticipated for concomitant surgical procedure at the time of CABG (eg, valve repair or replacement, aneurysm resection).
- Hemodynamically unstable patients, as defined by heart rate ≤40 beats/min or ≥100 beats/min, and/or systolic blood pressure <90 mm Hg or ≥200 mm Hg, and/or ongoing need for intravenous inotropic or vasopressor medications.
- Patients with confirmed myocardial infarction within 14 days, and/or rising cardiac biomarker proteins (ie, CK-MB or troponin), and/or worsening ECG changes.
- Previous CABG surgery.
- Stroke within 3 months before planned CABG.
- Immunosuppressive medication (eg, prednisone, cyclophosphamide, etanercept).
- Severe chronic renal insufficiency (serum creatinine ≥200 mmol/dL or need for dialysis), liver disease, (diagnosis of cirrhosis, chronic hepatitis, or elevated serum transaminases ≥3 times the upper limit of normal), cerebrovascular disease requiring concomitant carotid endarterectomy, peripheral arterial disease (claudication as the primary factor limiting activity), active nondermatologic malignancy requiring ongoing treatment, or any other condition that would place the patient at increased risk for complications during the first 6 mo after the procedure in the judgment of the attending cardiologist or cardiac surgeon.
- Contraindication to bone marrow aspiration (thrombocytopenia <50,000 cells/mm^3, INR >2.0).
- Hemoglobin <10 g/dL, white blood cell count <4000 cells/mm^3, absolute neutrophil count <1500 cells/mm^3.
- Active infection, with a temperature >37.5°C within 48 h before surgery and an unexplained white blood cell count >10,000 cells/mm^3.
- Myelodysplastic syndrome.
- Significant cognitive impairment.
- Any condition associated with a life expectancy <6 mo.
- Known allergic reaction or contraindication to any of the components of the $CD133^+$-enriched cells.
- Participation in other studies.
- History of severe ventricular tachyarrhythmia requiring treatment.
- Positive laboratory test results for or a history of syphilis, hepatitis B virus, hepatitis C virus, human T-lymphotropic virus type 1 and 2, or human immunodeficiency virus.
- Pregnant woman.
- Inability or unwillingness to provide written, informed consent.

CCS, Canadian Cardiovascular Society; *NYHA*, New York Heart Association; *LV*, left ventricular; *MRI*, magnetic resonance imaging; *ICD*, implantable cardiac defibrillator; *PPM*, permanent pacemaker; *CABG*, coronary artery bypass grafting; *CK-MB*, creatine kinase isoenzyme MB; *ECG*, electrocardiographic; *INR*, international normalized ratio.