Stem cell registry programme for patients with ischemic cardiomyopathy undergoing coronary artery bypass grafting: what benefits does it derive?

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Abstract

Aims Standardization of stem cell therapy requires application of appropriate methods to evaluate safety and efficacy, including long-term pharmacovigilance. To accomplish this objective, a long-term registry programme was installed.

Methods and results We analysed 150 patients with ischemic cardiomyopathy, who received intramyocardial CD133+ bone marrow mononuclear stem cell treatment combined with coronary artery bypass grafting (CABG) or CABG alone. The mortality rate, major adverse cerebral and cardiac events, and functional outcome parameters were evaluated for the time period up to 14 years follow-up. As a result, we have stratified the patient population (96 patients) into responders and non-responders. Furthermore, the analysis of relevant predictors of good response to CD133+ bone marrow mononuclear stem cell treatment was performed. Several positive tendencies related to stem cells transplantation were demonstrated. First, no significant difference in major adverse cardiovascular and cerebral events was observed between stem cell and control group up to 14 years follow-up. Second, an improvement of left ventricle ejection fraction (LVEF) in stem cell group retained for 5 years in contrast with CABG-only group, where no significant changes in LVEF after 2 years were observed. In addition, LVEF under 30% and left ventricle end diastolic diameter above 60 mm were independent predictors of functional response to CD133+ cell therapy.

Conclusions Participants with overt heart failure benefit most from CABG combined with intramyocardial injection of CD133+ bone marrow mononuclear cell within the group. An improvement LVEF in stem cell group remained for 5 years in contrast with the CABG-only group. The patients, in whom the improvement of both LVEF and LVED was observed, have benefited by increased life expectancy.

Keywords register; stem cells; CD133+; CABG; responder; intramyocardial injection

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Introduction

Stem cell therapy in heart disease was originally introduced to clinical practice in 2001 by Menasche et al. as an intramyocardial injection of skeletal myoblasts and Strauer et al. in a form of human autologous bone marrow stem cells delivered intracoronary.1,2 In parallel, Steinhoff and co-workers firstly initiated intramyocardial injection in a Phase I trial of bone marrow derived CD133+ purified cells in conjunction with coronary artery bypass grafting (CABG).3

Today, this approach is widely extended, and more than 600 clinical trials at different phases are mentioned in clinicaltrials.gov. An average period of patient’s observation in these studies is approximately 2 years after recruitment. This fact raises an important question about further perspectives for participants, which remain unclear. In addition, standardization of stem cell therapy requires application of appropriate methods to evaluate safety and efficacy, including long-term pharmacovigilance. Nevertheless, registry programme for stem cell patients is not obligatory, and stem
cell research clinics have to arrange high cost registries independently.

In the Reference and Translation Centre for Cardiac Stem Cell Therapy (RTC), University of Rostock, a registry programme has been functioning for more than 14 years. In the current study, we have analysed benefits and disadvantages of this programme by evaluating data of 150 participating patients, who suffered from chronic obstructive coronary disease and severe heart failure and therefore underwent intramyocardial CD133+ bone marrow stem cells (BMSCs) transplantation combined with CABG or CABG alone. Moreover, the most important parameters included in the register have been defined. In addition, the most efficient way of registry employment has been proposed.

Methods

Registry patients

The present analysis was performed using the registry data set collected at the University Medical Center Rostock. The registry programme includes the long-term follow-up of patients who previously had been treated with CD133+ stem cells in combination with CABG, mitral valve reconstruction or replacement, CD133+ stem cells alone, and patients with CABG alone (control group) since 2001. The current examination was performed with the data of 150 patients with ischemic cardiomyopathy, who underwent either an operation CABG and perioperative CD 133+ BMSC treatment or CABG alone. The baseline patient characteristics are summarized in Table 1. The inclusion criteria comprised: (i) chronic coronary disease suitable to CABG surgery, (ii) either reduced left ventricle ejection fraction (LVEF) visualized by transthoracic echocardiography (TTE) at rest or a distinct area of akinetic segments. The investigations conform to the principles outlined in the Declaration of Helsinki. All patients signed informed consent prior to inclusion in the study. Clinical trials were approved by the Ethics Committee of the Ärztekammer Mecklenburg-Vorpommern of the University Rostock (the Registration No. II HV 01/2001, the Registration II HV 08/2003).3,4

Table 1 Baseline characteristics of patients treated with stem cells and control patients

| Characteristics at baseline | Stem cell (n = 114) | Control (n = 36) | P-value |
|-----------------------------|---------------------|-----------------|---------|
| Male (%)                    | 105 (92%)           | 32 (89%)        | 0.513^a|
| Age (years)                 | 66.7 ± 8.3          | 68.6 ± 5.78     | 0.548^b|
| Infarct (%)                 | 89 (78%)            | 24 (69%)        | 0.293^c|
| LVEF (%)                    | 34.4 ± 9.3          | 36.7 ± 10.6     | 0.251^d|
| Median (25–75%Q) LVDD (mm) | 58.1 ± 6.4          | 58.9 ± 6.2      | 0.531^e|
| LVSD (mm)                   | 44.3 ± 8.2          | 46.4 ± 8.0      | 0.269^f|
| NYHA class                  | 3 (2–3)             | 3 (2–3)         | 0.964^g|
| Median (25–75%Q) CSS class | 3 (2–3)             | 3 (3–3)         | 0.687^h|
| Defibrillator (%)           | 8 (7%)              | 0 (0%)          | 0.2^i|
| Diabetes n (%)              | 51 (45%)            | 16 (44%)        | 0.565^j|
| Hypertonia n (%)            | 100 (88%)           | 32 (89%)        | 0.536^k|
| Smoking n (%)               | 29 (25%)            | 6 (17%)         | 0.264^l|
| Dyslipidemia n (%)          | 86 (75%)            | 25 (69%)        | 0.450^m|
| Coumadin n (%)              | 45 (40%)            | 12 (33%)        | 0.401^n|
| Aspirin n (%)               | 21 (18%)            | 8 (22%)         | 0.420^o|
| Beta-blockade n (%)         | 89 (78%)            | 30 (83%)        | 0.464^p|
| Statin n (%)                | 98 (86%)            | 29 (81%)        | 0.467^q|
| Diuretic n (%)              | 64 (56%)            | 17 (47%)        | 0.364^r|
| ACE inhibitor n (%)         | 87 (76%)            | 27 (75%)        | 0.536^s|

ACE, angiotensin-converting-enzyme; CSS, angina pectoris class; LVEF, left ventricle ejection fraction; LVDD, left ventricle diastolic diameter; LVSD, left ventricle systolic diameter; NYHA, New York Heart Association; NT-proBNP, N-terminal pro-brain natriuretic peptide.

*fisher’s exact test.
*Mann–Whitney U test.
*cTwo-sample t-test.

either 1 day before the CABG operation. CD133+ BMSC were isolated by magnetic separation with ferrite-conjugated antibody (Miltenyi CliniMacs System; Miltenyi Biotec, Bergisch Gladbach, Germany). Flow cytometry was performed to evaluate the quality of the stem cell product. Figure 1 shows the distribution of CD133+ BMSC (n = 114). The median CD133+ BMSC dose was over 3.9 × 10^6 cells in 1 mL (95% CI 4.0–5.3).

The cardiopulmonary bypass surgery was performed according to the standard of care. Before the aortic clamp was removed, 10–25 injections of 0.2 mL cell suspension were placed into the infarct border or hypokinetic myocardial segments.

Registry clinical and instrumental tests

The register in RTC includes parameters for the evaluation of functional outcomes, such as LVEF, left ventricle end diastolic diameter (LVEDD), and left ventricle end systolic diameter (LVESD), measured by the standard ultrasound method (Simpson, M-mode). All the measurements were performed by professional echocardiographers from the cardiology department (not blinded). In addition, 6 min walk test,
N-terminal pro-brain natriuretic peptide, heart failure class (New York Heart Association), and angina pectoris class were evaluated. Moreover, the register contains such safety parameters as results of laboratory tests (troponin, creatine kinase MB isoenzyme, creatine kinase, C-reactive protein, and leucocytes), electrocardiogram, mortality rate, and major adverse cardiovascular and cerebral events (MACCEs). MACCE was defined as an incidence of cardiac death, myocardial infarction, rehospitalization, and intensive care stays because of cardiac events and percutaneous or surgical revascularization, acute heart failure, ventricular arrhythmias, post-operative implantation of defibrillators or resynchronization therapy, and apoplexies. Furthermore, we studied the events of new tumour formation, immune diseases, and infections after stem cell transplantation procedure.

Statistics

For the processing and statistical analysis of data, the SPSS/PC software package was used (version 21.0, SPSS Inc, Chicago, USA). Descriptive statistics were computed for continuous and categorical variables. Data are provided as mean±standard deviations, or median (first and third quartiles). Testing for differences of continuous variables between two study groups created by responder/non-responder was performed by a two-sample t-test for independent samples or a Mann–Whitney U test by ranks, as appropriate. Data distributions of parameters were analysed using Kolmogorov–Smirnov tests. Testing for differences of continuous variables between different time points was accomplished by the paired t-test or the Wilcoxon test by ranks for paired data as appropriate. Categorical factors comparisons between groups were performed by Fisher’s exact tests. All P values resulted from two-sided statistical tests, and values of P < 0.05 were considered to be statistically significant.

Results

Major adverse cardiovascular and cerebral event and mortality rate

During the time period from 2001 to 2011, 150 patients were treated with either CABG (n = 36) or combination of CABG and autologous CD133+ BMSC cell transplantation (n = 114). MACCE and mortality rate were evaluated while patients were followed for 9.6 ± 2.8 years on average (4–14 years; total of 1058 patient years). In order to collect required information, either patients or their relatives (in case of patient’s death) were yearly surveyed. To date, all 150 patients have reached 1 and 4 years post-treatment time point, whereas 84 patients (58 stem cell/26 control) have reached 10 years follow-up.

During the whole observation period, 36 patients died: 28 from 114 (25%) patients in the stem cell treatment group and 8 from 36 (22%) in the control group (P = 0.827). Among them, no cases of deaths due to immune diseases in both groups were noted. At the same time, two patients (1.8%) have died from lung and bronchial cancer in the stem cell group after 50 and 86 months follow-up. No significant difference in MACCEs between the treatment groups was observed: 45 (39%) events in the CD133+ BMSC/CABG group vs. 17 (47%) recorded events in the CABG group (P = 0.442). Post-operative implantation of defibrillators or resynchronization therapy, ventricular arrhythmias, and apoplexies were the most frequent MACCE form noted. In particular, 19 patients (17%) from the stem cell group required the procedure of defibrillator implantation or resynchronization therapy. Moreover, half of them underwent it during the first 2 years after treatment with CD133+ stem cells injection in combination with CABG. In the control group, defibrillators were implanted or resynchronization therapy was performed in 14% of patients—half of them during the first year after CABG (P = 0.799). New episodes of ventricular arrhythmias occurred in 13% of cases in stem cell group and in 11% of cases in the matched control group (P = 1.000). However, the reduction of most frequent arrhythmias was observed in both groups 5 years after the procedure, and therefore, the correlation between stem cells injection and arrhythmias is not certain. In addition, the percentages of apoplexies during 14 year follow-up were almost equal in stem cell and control groups (8.8% and 8.3%, respectively, P = 1.000).
Rehospitalization and intensive care stays due to cardiac events and percutaneous or surgical revascularization took place in 6.1% and 8% of the stem cell group and in 5.5% and 5.5% of the control group, respectively.

Functional parameters

Transthoracic echocardiography was applied to analyze the data of 96 patients (n = 73 cell therapy and n = 23 control group), collected before the procedure and 12 months follow-up and further yearly up to 14 years. As a result, two different tendencies for short-term and long-term outcomes were revealed.

After 12 months follow-up, no significant difference in the efficiency of treatment was demonstrated in stem cell and control groups. The results showed that LVEF was increased by 5.3% in the stem cells group (35.7% ± 10.0 to 41.0 ± 9.0%; P < 0.001) and by 4.7% in the matched control group (37.2 ± 2.0% to 41.9 ± 2.0%; P = 0.05). In addition, the LVEDD decreased by 2.3 mm in the stem cell group (57.8 ± 6.1 to 55.5 ± 6.2 mm; P < 0.001) and by 2.5 mm in the control group (59.4 ± 1.2 to 56.9 ± 1.1 mm; P = 0.022) 12 months after procedure, which is not significant. Moreover, the New York Heart Association heart failure and angina pectoris class (CSS) decreased from Classes 3 to 1 in both groups 12 months follow-up.

At the same time, the continuous improvement of ejection fraction in patients of stem cell group was observed during 5 years after the therapy in comparison to pre-operative value (35.7 ± 10.0% to 45.3 ± 3.2%; + 9.6%, P < 0.05). In the CABG-only group, the LVEF, on the contrary, returned to baseline numbers after a moderate peak at 2 years follow-up. These tendencies are demonstrated on Figure 2. Such dimension parameters as LVEDD and LVESD showed no significant changes after 1 year follow-up in both stem cell and control, groups (data not shown).

Sub-stratification of cell therapy patient population based on therapy responsiveness

The response of different patients in the register to the injection of CD133+ BMSC varied considerably; that is, in certain cases, patients had a clear functional benefit, while in others, no apparent response or even a deterioration of function was noted after 1 year follow-up. Therefore, we stratified patient population into responders and non-responders. The changes in global LVEF more than 5% improvement and reduction of LVEDD more than 5 mm LVEDD at 12 month follow-up were chosen as main criteria of responsiveness. The 5% baseline in LVEF was chosen according to previously published meta-analyses: intramyocardial BMSC transplantation during CABG resulted in average increase of LVEF 5.8% compared with the control CABG group.5 No meta-analyses regarding changes in LVEDD were found. The analysis of cross-correlation between functional outcomes and mortality and MACCE in observation period, however, suggests that a proposed combination of LVEF and LVEDD criteria can define a beneficial long-term prognosis for CD133+ BMSC treatment combined with CABG or CABG-alone compared with stand-alone improvement in LVEF or LVEDD. This observation is illustrated by Table 2: the level of mortality was 0% for patients who had an improvement of both functional parameters after 12 months follow-up, whereas in the non-responder group, 24% of deaths were noted (P = 0.020). Consequently, responders were defined as patients who had an increase of LVEF more than 5% in combination with >5 mm decrease of LVEDD, while all the patients, who had an unchanged or even deteriorated LVEF and LVEDD at 12 month follow-up, were categorized as non-responders.

Table 2. Mortality and MACCEs in responder and non-responder patient groups

|                | Responder (n = 44) | Non-responder (n = 52) | P*       |
|----------------|-------------------|------------------------|----------|
| >+5% LVEF (%)  | 14 (32%)          | 25 (48%)               | 0.144    |
| Mortality n (%)| 5 (11%)           | 14 (27%)               | 0.073    |
| >5 mm LVEDD (%)| 13 (42%)          | 26 (40%)               | 0.999    |
| Mortality n (%)| 3 (10%)           | 16 (25%)               | 0.105    |

Mortality for responder and non-responder patients at 12 months follow-up. *Combined LVEF and LVEDD criteria.

Figure 2 Changes in left ventricle ejection fraction during 5 years follow-up in stem cell and control groups. *Paired t-test. n1 = 73, n2 = 44, n3 = 35, n4 = 26, and n5 = 25. n1 = 23, n2 = 8, n3 = 7, n4 = 9, and n5 = 7 (n, number of patients; c, stem cell; c, control; 1, year).
Further, we compared the characteristics at a baseline between responders and non-responders to identify predictors of good response to stem cell therapy. Figure 3 clearly demonstrates that an improvement in LVEF and LVEDD depended on pre-operative values. The probability of positive response to stem cell therapy was higher for participants with worse baseline parameters; that is, patients that responded to CD133+ cell therapy had an average pre-operative LVEF of 27%, LVEDD of 63 mm and LVESD of 49 mm, while non-responding patients had a prior LVEF of 38%, LVEDD of 56 mm and LVESD of 42 mm ($P < 0.001$, $P < 0.001$, and $P = 0.017$). In addition, N-terminal pro-brain natriuretic peptide levels were higher in patients that responded to stem cell therapy. Interestingly, the number of CD133+ stem cells was not associated with the responsiveness of patients ($P = 0.311$). Moreover, the correlation between such factors as concomitant diseases or given medication and patient’s response to the therapy was not found. Detailed comparison of different baseline criteria for responders and non-responders is shown in Table 3.

### Discussion

Stem cell therapy is a non-standardized therapeutic approach for the treatment of multiple disorders including cardiovascular diseases. Standardization of stem cell therapy requires long-term pharmacovigilance, which contains safety and efficacy evaluation. In Commission Directive (2009/120/EC) from 14 September 2009, it is stated that ‘A strategy for the long-term follow-up of safety and efficacy shall be included in the risk management plan’. However, currently, the average period of patient observation after stem cell application in clinical trials lasts approximately for 2 years, and participants are usually not observed after the study closure. Such an approach as a registry programme, where patients are studied for their lifetime, can be adopted to improve this current situation. Present analysis was performed to define the most useful parameters of the register to find an optimal way to employ it.

In all the aspects of good clinical practice, the safety of participants of clinical trials is of considerable importance. Therefore, MACCEs, which represent the main parts of safety evaluation process, could be selected as the most relevant parameter of the register. In this case, arrhythmias, infarctions, apoplexies, and so forth can be monitored closely after stem cell application, and their comparison with the control group can be carried out. Furthermore, in the long-term follow-up, late probable complications such as calcifications and tumours are revealed. As a final result, the patient’s safety is maintained, which is a prerogative for any clinical trial.

Importantly, the efficiency of stem cell therapy can also be evaluated by studying multiple functional parameters, included in a register. Nevertheless, analysis of registry data demonstrated that the evaluation of functional parameters after long-term follow-up is a daunting task. The heterogeneity of patient’s cohort has to be acknowledged for the register. Therefore, registry data cannot fully replace the results of randomized clinical trials evaluation of long-term functional parameters. For example, the present study showed no difference between stem cell and control groups after 12 months follow-up. In contrast, previously conducted placebo-controlled Phase II study clearly showed the benefits of stem cell therapy for this particular time point. To conclude, to obtain higher reliability, the long-term analysis can be applied to different groups of patients with the same pre-operative parameters (recruited for one study) within the registry programme. The comparison of tendencies within the groups, stem cell treated and control, however, proved to be a solid tool for the evaluation of stem cell therapy functional outcomes.

#### Figure 3
Comparison of functional parameters between responders and non-responders. Pre-operative results. *Two-sample t-test.

#### Table 3 Comparison between responder and non-responder patients (pre-operative results)

| Characteristics at baseline | Responder ($n = 15$) | Non-responder ($n = 58$) | $P$ |
|-----------------------------|----------------------|--------------------------|-----|
| NT-proBNP (pg/mL) Median (25–75%)Q | 2558 (1756–1810) | 762 (453–2456) | 0.102a |
| Number of stem cells (mean ± SD) | 3.8 ± 2.3 | 4.7 ± 3.2 | 0.311c |
| Diabetes n (%) | 7 (47%) | 24 (41%) | 0.7744a |
| Hypertonia n (%) | 15 (100%) | 54 (93%) | 0.5755a |
| Smoking n (%) | 6 (40%) | 15 (26%) | 0.341a |
| Dyslipidemia n (%) | 13 (87%) | 49 (85%) | 1.000a |
| Anti coagulantia n (%) | 5 (33%) | 23 (40%) | 0.770a |
| Aspirin n (%) | 4 (27%) | 8 (14%) | 0.253a |
| Beta blocker n (%) | 11 (73%) | 48 (83%) | 0.4674a |
| Statin n (%) | 14 (93%) | 47 (81%) | 0.438a |
| Diuretic n (%) | 11 (73%) | 29 (50%) | 0.1484a |
| ACE inhibitor n (%) | 10 (66%) | 44 (76%) | 0.5164a |

ACE, angiotensin-converting-enzyme; NT-proBNP, N-terminal pro-brain natriuretic peptide; SD, standard deviation. 
aFisher’s exact test.  
bTwo-sample t-test.  
cMann–Whitney U test.
Moreover, it has been known for a while that the response of different subsets of patients to BMSC therapy varies. The first report on this observation was published by Panovsky et al. in 2010, where only one-third of the study participants responded to autologous intramyocardial BMSC transplantation. In this work, patients were considered as responding to stem cell therapy when at least three from four of the following criteria were fulfilled: more than 10% decrease in perfusion defect size, more than 5% increase of LVEF, more than 10% decrease in left ventricle end systolic volume as quantified by methoxy isobutyl isonitrile single-photon emission computed tomography (MIBI SPECT), and more than 10% increase in peak systolic velocity measured by TTE. In the RTC, the combination of a minimum of 5% increase in LVEF and more than 5 mm decrease in LVEDD was selected as optimal to classify an improvement of patient’s health as a positive response to stem cell transplantation after 12 months follow-up. The current study confirmed the reliability of these proposed criteria for all selected responders showing no mortality up to 14 years follow-up ($P = 0.02$) and half the MACCEs compared with the non-responder group ($P = 0.173$). Hence, a combination of these functional parameters is used in the RTC to reveal patients, who had no response to cell therapy. Further non-responders are hospitalized for detailed examinations and intensive treatment.

In addition, baseline characteristics, which predict a beneficial response to stem cell therapy, can be defined using data of the registry. This application was first proposed by Rodrigo et al. in 2014. This research group has shown that diabetes and considerable numbers of ischemic segments in myocardium are predictors of significant response to BMSC injections in patients with refractory angina and chronic ischemia. Notwithstanding demonstrated correlation, our data have not confirmed the connection between diabetes and positive outcome of cell therapy. Moreover, patients’ response to stem cell transplantation was not associated with cardiovascular risk factors or baseline medication. In addition, we observed no influence of number of transplanted CD133+ BMSC (in the range of 0.5–20 × 10^6) on responsiveness. These findings are supported by the work of Bai et al., who found no clear correlation between the number of intracoronary delivered BMSC and changes in LVEF.

Such parameters as LVEF below 30%, or severe heart dilatation with a LVEDD over 60 mm, however, were predictors of responsiveness to the stem cell therapy in patients suffering from ischemic cardiomyopathy. This observation was earlier confirmed by Wen Y. et al., who demonstrated an enhanced improvement of ejection fraction after bone marrow-derived mononuclear cell therapy in patients with more severe heart damage (ischemic heart failure) compared with patients with ischemic heart disease. Moreover, the meta-analysis by Jeevanantham et al. showed that stem cell treatment in patients with low baseline LVEF (less than 40%) resulted in greater improvement in left ventricle end systolic volume and LVEDV ($P = 0.0004$ and $P = 0.01$, respectively). To summarize, these results, obtained because of registry programme, can be applied as criteria for the selection of candidates suitable for stem cell therapy.

In conclusion, registry documentation of cardiac stem cell patients provides a method of long-term pharmacovigilance. Owing to yearly MACCE registrations, a register allows that late probable complications are carefully followed as well as unexpected complications reported, which is required to ensure patient’s safety as the main aspect of good clinical practice. Therefore, it is expedient to establish an obligatory common register for all centres carrying out stem cell studies. Moreover, this may be used for a standardized pharmacovigilance reporting to regulatory authorities. Nevertheless, the register by itself cannot replace randomized clinical trials. Registry data, however, can be used for long-term efficiency and safety evaluation in standardized patient groups. Furthermore, a register could help to improve patient selection revealing predictors of good response.

Conflict of interest
None declared.

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References
1. Menasche P, Hagège AA, Scorsin M, Pouzet B, Desnos M, Duboc D, Schwartz K, Vilquin JT, Marolleau JP. Myoblast transplantation for heart failure. Lancet 2001; 357: 279–280.
2. Strauer BE, Brehm M, Zeus T, Gattermann N, Hernandez A, Sorg RV, Kögl G, Wernet P. Intracoronary, human autologous stem cell transplantation for myocardial regeneration following myocardial infarction. Dtsch Med Wochenschr 2001; 126: 932–938.
3. Stamm C, Kleine HD, Choi YH, Dunkelmann S, Lauffs JA, Lorenzen B, David A, Liebold A, Nienaber C, Zurakowski D, Freund M, Steinhoff G. 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: 717–725.

DOI: 10.1002/ehf2.12132
4. Yerebakan C, Kaminski A, Westphal B, Donndorf P, Glass A, Liebold A, Stamm C, Steinhoff G. Impact of preoperative left ventricular function and time from infarction on the long-term benefits after intramyocardial CD133(+) bone marrow stem cell transplant. *J Thorac Cardiovasc Surg* 2011; **142**: 1530–9.e3.

5. Donndorf P, Kundt G, Kaminski A, Yerebakan C, Liebold A, Steinhoff G, Glass A. Intramyocardial bone marrow stem cell transplantation during coronary artery bypass surgery: a meta-analysis. *J Thorac Cardiovasc Surg* 2011; **142**: 911–920.

6. Jeevanantham V, Butler M, Saad A, Abdel-Latif A, Zuba-Surma E.K, Dawn B. Adult bone marrow cell therapy improves survival and induces long-term improvement in cardiac parameters: a systematic review and meta-analysis. *Circulation* 2012; **126**: 551–568.

7. Tian T, Chen B, Xiao Y, Yang K, Zhou X. Intramyocardial autologous bone marrow cell transplantation for ischemic heart disease: a systematic review and meta-analysis of randomized controlled trials. *Atherosclerosis* 2014; **233**: 485–492.

8. Panovsky R, Vasku A, Meluzin J, Kaminek M, Mayer J, Janousek S, Kincl V, Groch L, Navratil M. Association of polymorphisms of zinc metalloproteinases with clinical response to stem cell therapy. *Herz* 2010; **35**: 309–316.

9. Rodrigo SF, van Ramshorst J, Mann I, Leong DP, Cannegieter SC, Al Younis A, Dibbets-Schneider P, de Roos A, Fibbe WE, Zwaginga JJ, Bax JJ, Schalij MJ, Beeres SL, Atsma DE. Predictors of response to intramyocardial bone marrow cell treatment in patients with refractory angina and chronic myocardial ischemia. *Int J Cardiol* 2014; **175**:539–544.

10. Bai Y, Sun T, Ye P. Age, gender and diabetic status are associated with effects of bone marrow cell therapy on recovery of left ventricular function after acute myocardial infarction: a systematic review and meta-analysis. *Ageing Res Rev* 2010; **9**: 418–23.

11. Wen Y, Chen B, Wang C, Ma X, Gao Q. Bone marrow-derived mononuclear cell therapy for patients with ischemic heart disease and ischemic heart failure. *Expert Opin Biol Ther* 2012; **12**: 1563–1573.