Electrophysiological and arrhythmogenic effects of intramyocardial bone marrow cell injection in patients with chronic ischemic heart disease

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BACKGROUND Bone marrow cell injection has been introduced to treat patients with ischemic heart disease. However, focal application of bone marrow cells may generate an arrhythmogenic substrate.

OBJECTIVES To assess the electrophysiological and arrhythmogenic effects of intramyocardial bone marrow cell injection in patients with chronic myocardial ischemia.

METHODS Bone marrow was aspirated in 20 patients (65±11 years, 19 male) with drug-refractory angina and myocardial ischemia. Electroanatomical mapping (NOGA, Biosense-Webster, Waterloo, Belgium) was performed during mononuclear cell isolation. Areas for cell injection were selected based on the localization of ischemia on SPECT. These areas were mapped in detail to evaluate local bipolar electrogram duration, amplitude and fragmentation. Mononuclear cells were injected in the ischemic area with the NOGA system. SPECT and electroanatomical mapping were repeated at 3 months. Holter monitoring was repeated at 3 and 6 months.

RESULTS SPECT revealed a decrease in the number of segments with ischemia (3.5±2.5 vs. 1.1±1.0 at 3 months; $P<0.01$) and an increased left ventricular ejection fraction (44±13% vs. 49±17% at 3 months; $P=0.02$). The number of ventricular premature beats remained unchanged ($10\pm24\times10^2$/24h vs. $8\pm23\times10^2$/24h at 3 months (P=NS) and $12\pm30\times10^2$/24h at 6 months (P=NS)). At 3 months follow-up, bone marrow cell injection did not prolong electrogram duration (15.9±4.6 ms vs. 15.6±4.0 ms; P=NS), decrease electrogram amplitude (3.8±1.5 mV vs. 3.8±1.5 mV; P=NS), or increase fragmentation (2.0±0.5 vs. 1.9±0.4; P=NS).

CONCLUSION Intramyocardial bone marrow cell injection does not increase the incidence of ventricular arrhythmias and does not alter the electrophysiological properties of the injected myocardium.

KEYWORDS Bone marrow cells; Electrophysiology; Electroanatomical mapping; Arrhythmias; Ischemia; Angina.

ABBREVIATIONS CCS=Canadian Cardiovascular Society; CL=Cycle length; EP=Electrophysiology; ICD=Implantable cardioverter-defibrillator; SPECT=Single photon emission computed tomography.

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Introduction

Intramyocardial cell transplantation is currently being investigated as a potential therapy for chronic ischemic heart disease.[1] Through application of therapeutic cells to ischemically-damaged myocardium, this novel treatment modality aims to enhance left ventricular function and improve myocardial perfusion. At present, several cell types including bone marrow-derived cells and skeletal myoblasts have been used in clinical studies for this purpose.[2–8] The increased incidence of ventricular tachycardia in clinical studies using human skeletal myoblast transplantation highlighted the importance of a systematic assessment of the potential arrhythmogenic consequences of cell therapy.

A few pre-clinical studies addressed the electrophysiological characteristics of bone marrow cells. From these studies it appears that focal application of bone marrow cells can potentially generate an arrhythmogenic substrate. For example, Chang et al. recently reported that mixtures of bone marrow-derived mesenchymal stem cells and neonatal rat cardiomyocytes resulted in an arrhythmogenic substrate that is characterized by decreased conduction velocity and easily inducible sustained reentrant arrhythmia.[9] Similarly, we previously demonstrated that transmission of the electrical impulse across human adult bone marrow-derived mesenchymal stem cells is characterized by slow conduction, reduced depolarization rates and low-amplitude electrical activity decaying with distance.[10]

Conflict of interest: none. **Address reprint requests and correspondence:** Martin J. Schalij, Department of Cardiology, Leiden University Medical Center, Albinusdreef 2, 2333 ZA Leiden, The Netherlands. E-mail: m.j.schalij@lumc.nl. (Received August 25, 2006; accepted October 30, 2006)

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In the clinical setting, 5 studies evaluated the safety and feasibility of intramyocardial bone marrow cell injection for chronic myocardial ischemia.[2–6] Holter monitoring in these studies demonstrated no increase in the incidence of ventricular arrhythmias. However, no study thus far systematically evaluated the electrophysiological effects of intramyocardial bone marrow cell injection in patients with chronic myocardial ischemia.

Three-dimensional electroanatomical catheter mapping could potentially contribute in the evaluation of the electrophysiological effects of bone marrow cell injection. Based on the analysis of bipolar electrograms, this technique allows accurate identification of zones of slow conduction and characterization of an arrhythmogenic substrate.[11–13]

The present study investigated the electrophysiological and arrhythmogenic effects of intramyocardial bone marrow cell injection in patients with chronic myocardial ischemia and subsequent drug-refractory angina. Electroanatomical mapping of the injected ischemic myocardial region was performed to assess changes in local bipolar electrogram characteristics and 24-hour Holter monitoring was used to evaluate the incidence of ventricular arrhythmias.

Methods

Patients

Patients with severe angina pectoris (Canadian Cardiovascular Society (CCS) class III or IV) despite optimal medical therapy were included in the current study if stress-rest technetium-99m tetrofosmin single photon emission computed tomography (SPECT) revealed the presence of stress-inducible ischemia. All patients were ineligible for percutaneous or surgical revascularization as assessed by coronary angiogram. The exclusion criteria were: acute myocardial infarction within 6 months of enrollment, history of malignancy, renal dysfunction (serum creatinine >200 μmol/L) and unexplained hematological or biochemical laboratory abnormalities. The study protocol was approved by the institutional ethics committee, and written informed consent was obtained from all patients.

Study protocol

Baseline evaluation included assessment of the patients' clinical status according to the CCS classification and 24-hour Holter monitoring to document heart rhythm, heart rate, ventricular premature beats and the occurrence of (non-) sustained ventricular tachycardia. In order to identify the location of stress-inducible myocardial ischemia and assess left ventricular ejection fraction, stress-rest technetium-99m tetrofosmin gated SPECT was performed.

Immediately before bone marrow cell injection, 3-dimensional electroanatomical mapping of the left ventricle was performed using the NOGA system (Biosense-Webster, Waterloo, Belgium). The electroanatomical map was used to evaluate the local bipolar electrogram characteristics of the myocardial region with ischemia and guide the bone marrow cell injections. Immediately after the procedure, 2-day continuous heart rhythm monitoring was started.

Follow-up evaluations performed 3 and 6 months after the injection procedure consisted of a clinical assessment (CCS classification) and 24-hour Holter recording to monitor ventricular arrhythmia. At 3 months follow-up, electroanatomical mapping was repeated to reassess the local electrophysiological characteristics of the injected myocardium. In addition, stress-rest gated SPECT was repeated at 3 months follow-up to reassess myocardial ischemia and left ventricular ejection fraction. Cardiovascular medication remained unchanged during the 6 months follow-up period.

Electroanatomical mapping and bone marrow cell injection

At the day of the injection procedure, bone marrow was aspirated from the iliac crest under local anesthesia. During mononuclear cell isolation (Ficoll density gradient), patients underwent non-fluoroscopic mapping with the NOGA system, which has been described in detail previously.[14,15] All patients received an intravenous dose of 7,500 U heparin. After left ventricular angiography, left ventricular endocardial mapping was performed via femoral artery access and a retrograde aortic approach using a 7-French NOGAStar catheter (2 mm tip, 2 electrodes, interelectrode distance 0.5 mm; Biosense-Webster). The high-pass filters were set at 30 Hz and the low-pass filters at 500 Hz.

Landmark points outlining the endocardial left ventricular boundaries (apex, aortic outflow, mitral inflow) were acquired under fluoroscopic guidance. The catheter was dragged over the endocardial surface to record electrograms at different sites and to simultaneously determine the shape and volume of the left ventricle. Points were only acquired when the catheter tip had a stable position (using local stability, loop stability and cycle length stability as specified by the manufacturer)[14,15] perpendicular to the myocardial wall. In order to provide high spatial resolution and minimize interpolation between actual data points by the system, the filling threshold was set at 15 mm. This setting allows the NOGA system to fill triangles between acquired points using an interpolation algorithm, so that distances between points larger than 15 mm away from each other are left blank until points in between are sampled. After completion of the map, the mapping points of the internal sites of the left ventricle (distance from fitting plane >35 mm) and mapping points with unsatisfactory stability were removed.

Based on the localization of ischemia on technetium-99m tetrofosmin SPECT, the region of interest (= myocardial area with ischemia on SPECT at which cell injections were targeted) was delineated on the NOGA map. Therefore, by definition, the region of interest comprised ischemic but viable (unipolar voltage $\geq$6.9 mV, bipolar voltage $\geq$1.5 mV)[13,16,17] myocardium.

For further evaluation of the local electrophysiological characteristics of the region of interest, 25–30 additional points were acquired in this region. Accordingly, a high-density map of the region of interest was derived. In order to allow post-procedural off-line analysis, the additional electrograms recorded in the region of interest were simul-