

# Six Month Follow-up after Transendocardial Autologous Fractionated Bone Marrow Shows Increased Ejection Fraction in Patients with Ventricular Dysfunction After Chronic Myocardial Infarction, an Open-Label, Non-Randomized, Single-Center Study (the TABMMI study)

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## Abstract:

Local autologous bone marrow (ABM) stem cell therapy has shown benefit in preclinical and clinical studies, although debate continues on the mechanism of action and the most appropriate methods for performing such therapies. We assessed the hypothesis that helical needle transendocardial (TE) delivery of ABM around regions of dyskinesia in chronic post-MI patients would be safe and possibly improve ejection fraction, as this system presents theoretical advantages over other TE delivery systems and was applied to promote a specific mechanism of action.

**Methods:** Stable post-MI Patients (N=10) with left ventricular (LV) dysfunction, defined as an ejection fraction (EF) <40% determined by echo prior to catheterization, who were not candidates for LV surgical restoration, AICD placement or valvular surgery were treated. Patients having evidence of infection, active malignancy, EF<30% at baseline, or a recent MI (< 4 w) were excluded. 50 cc of ABM cells were aspirated from the iliac crest before cardiac catheterization and isolated by Ficoll density gradient. Cells were delivered percutaneously with a TE helical needle catheter. 86 x 106 cells were injected into 7.1+/±3.1 sites around the infarct to target the peri-infarct zones. 2D echo LV EF measurements, 24 hour Holter, and exercise tolerance testing were performed at baseline, day of procedure, and at 1W, 3M, and 6M after procedure. Results: Of the 1.3 x 108 cells prepared for delivery 98% +/- 1% were viable, 2.2 +/- 1.2% were CD34+, and 0.03 +/- 0.02% were CD133/1+. There were no adverse events associated with the catheter based cell transplantation procedure. One hour after the procedure, there was a statistically significant rise in Troponin T levels (0.017 +/- 0.013 to 0.276 +/- 0.300, p<0.05, N=9) and CK MB (2.7 +/- 1.2 to 10.8 +/- 4.7, p<.05, N=10). At most recent follow-up of six months, there were statistically significant increases in LV EF on echo (35.4 +/- 4.1 to 40.8 +/- 4.5, p<0.05, N=10); with no acute events or sustained arrhythmias.

**Conclusion:** ABM cells delivered transendocardially with the helical needle in this pilot study improved ventricular wall motion in chronic myocardial infarct patients without arrhythmias, or other clinical complications.

## Methods:

This study was approved by the Argentina National Institute for Coordination of Ablation and Implantation, the Argentina National Administration for Medication and Medical Technology, and the institutional review committees at both the Argentina Swiss Clinic and Argentina Institution of Diagnostic and Treatment. All patients gave their informed consent.

## Population Studied

Patients were enrolled who had sustained a transmural myocardial infarction and showed echocardiographic evidence of left ventricular dysfunction having ejection fractions of <40%. These patients were not candidates for ventricular aneurysm surgery, AICD placement or valvular repair. Patients were 18 years of age or older, able to give informed consent, and had the approval of their cardiologist. Patients were excluded due to clinical evidence of infection, active malignancy, other cardiopulmonary disease, high grade atrioventricular block, sustained ventricular tachyarrhythmias, ejection fraction <30%, a recent MI (< 4 w), presence of artificial aortic valve, recent history of alcohol or drug abuse or evidence of other multisystem disease. Subjects were entered without regard to socioeconomic status, ethnic background, or source of referral.

## Marrow Harvest and Processing

Approximately four hours prior to the percutaneous catheterization, 50 cc of autologous bone marrow cells were collected under local anesthesia from the posterior iliac crest. Bone marrow mononuclear cells were isolated by density gradient on Ficoll-Paque Plus tubes (Amersham Biosciences). Cells were washed and filtered through 100 µm nylon mesh to remove cell aggregates, and resuspended in Ringers solution at a concentration of 1 x 10<sup>8</sup> cells/ml in a total volume of 1.3 ml. 10% of the final cell solution was used for cell counting and viability testing using trypan blue exclusion. Enumeration of CD34+ and CD133/1- cells was done by staining with antibodies CD34-FITC from Beckman Coulter and AC133/1-PE from Miltenyi Biotec, respectively, using a Coulter Profile flow cytometer, according to the standards proposed by International Society of Hematotherapy and Graft Engineering. ABM cells from each patient were preserved for future genome-wide expression analysis to identify possible mechanisms of action in positive responders.

## Catheter Delivery of Cells

Prior to cell delivery, the infarct target zones were defined using ECG, baseline echocardiography, and previous orthogonal ventriculography data. Upon catheterization, the selected target regions were assessed using fluoroscopy. Three investigators established anatomic consensus regarding the helical needle placement in predefined target regions. Cells were delivered transendocardially with the two-catheter Helical Infusion System for transendocardial delivery (BioCardia, Inc., South San Francisco, CA).

Table 1. Study tests and procedures to be performed per the protocol.

Test or procedure	Baseline	Transplant	Follow-up visit windows: weekly visits (± 3 days); Monthly visits (± 7 days)				
			W1	W12	M6	M12	M24
ABM Aspiration		X					
Cell Implantation		X					
Physical Exam	X						
ECG	X		X	X	X	X	X
Echo	X	X	X	X	X	X	X
24 Hour Holter	X		X	X	X	X	X
Exercise Tolerance Testing	X		X	X	X	X	X
Blood Tests	X	X	X	X	X	X	X
Urinalysis	X	X	X	X	X	X	X
Adverse Events	X	X	X	X	X	X	X
Concomitant Meds	X	X	X	X	X	X	X

Figure 1. The Helix catheter for transendocardial delivery is advanced through a steerable guide (not shown). In addition to the therapeutic lumen which discharges at the distal tip of the helix, it has a second lumen which discharges from the base of the helix for delivering contrast to confirm engagement and positioning.

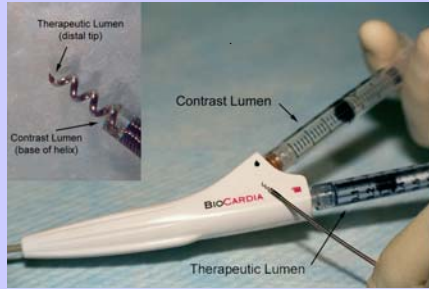


Figure 2. Cine image of the Helix catheter system showing contrast discharged at the Helix base to confirm engagement with the endocardium.

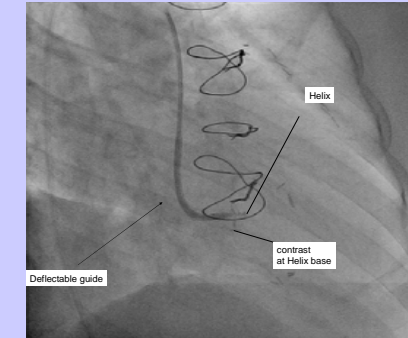


Figure 3: Ventricular Events on 24 Hour Holter. There are no statistical differences in these values. Error bars denote +/- stdev.

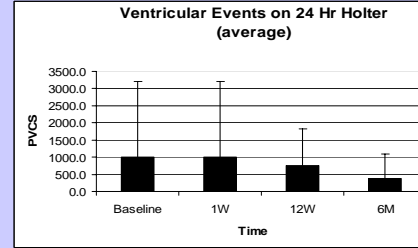


Figure 4: Changes in ETT are not statistically significant. Error bars denote +/- stdev.

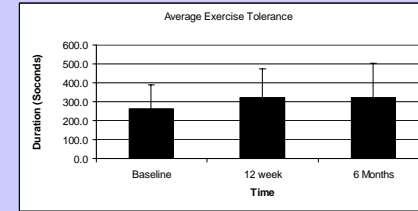


Figure 5: Changes in ejection fraction at both 12 W and 6M are statistically significant. Error bars denote +/- stdev

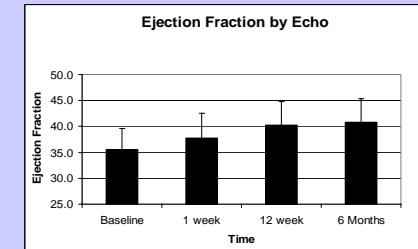
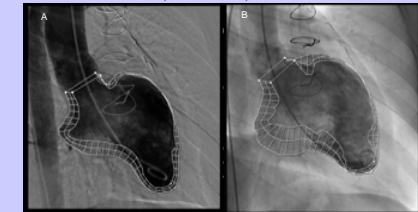


Figure 6: Right anterior oblique left ventriculogram on date of procedure (A) and at 6 month follow-up (B) showing ejection fraction (EF) changing from 37.9% to 50.1%, respectively. It should be noted that on echo this patient had an ejection fraction of 39% at baseline and 46% at 6 Month follow-up. A. End Systolic Volume (ESV) 81.9 ml, End Diastolic Volume (EDV) 131.8 ml, EF 37.9%. B. ESV: 92 ml, EDV 184.4 ml, EF: 50.1%.



## Discussion:

The patients were selected to provide a stable data set by the chronic unchanging nature of their LV dysfunction, and the absence of other clinical parameters that could account for late improvement. Further, the stable chronic infarct population with ejection fractions between 30 to 40% represent the subset for whom the advancement to congestive heart failure is a common progression.

One primary safety issue for cell therapy strategies is the potential to cause cardiac arrhythmias. The Holter measurements suggest that there are no arrhythmia safety issues within six months of such a procedure, although the presence of an episode of ventricular tachyarrhythmia warranting implantation of an AICD at 11 months in one patient is noted. This individual at baseline had an ejection fraction of less than 35% and would have warranted an AICD based on the recent MADIT II and SCD-HEFT trials. This patient did not demonstrate any significant Holter detected arrhythmias at baseline or follow-up, and the AICD has not noted any evidence of sustained VT or discharges since implantation.

The transthoracic echo measurements of ejection fraction demonstrate improvements in all patients at 6 months in this pilot study. However, it is noted that one patient had a precipitous fall in ejection fraction from baseline to 12 weeks and that this significant change reversed itself. These unpredicted improvements in EF at six months are not powered to confirm the clinical efficacy of this therapy but they are compelling in a safety pilot trial. In this regard, the echo measurements of end diastolic volume also trended downward, suggesting overall anatomic LV improvements despite the lack of statistical power. Baseline and follow up echocardiographies were performed by the same technician operator.

By using the helical fixation transendocardial delivery device, the investigators were provided with a means to target segmental dyskinesia with stability from myocardial fixation during the delivery process. This enables physician-operators to agree on the delivery location as well as to control the time course of delivery. The helix fixation needle may reduce back-leak into the ventricular chamber by providing a simplified direct means to confirm that one is positioned within the myocardium, a longer self sealing helical pathway into the tissue, and the ability to leave the helix in place after delivery to act as a barrier to obstruct backleak.

The obstruction of back-leak may be extremely important if cells or cell aggregates do have the ability to act as microemboli. Care in cell processing to ensure that cell aggregates are not present may mitigate this issue. However, backleak may also be important should future strategies seek to incorporate other more viscous materials to enhance retention.

The elevation in CKMB and Troponin T are statistically significant and believed to be due to the penetration of the myocardium. This is in line with increases seen in the studies performed in ischemic patients with straight needle TE catheter systems in stable patients. Although these values correspond to values that are clinically meaningful in a patient presenting with an acute infarct, elevation in these cases may be expected from penetrating the myocardium in this stable patient population. It is interesting to note that elevation of these markers is also only seen in the patients who are not experiencing a recent MI, suggesting the possibility that already elevated markers due to large infarcts may mask changes in these same markers that could be due to the presence of microinfarcts.

By selecting stable patients for this sole therapy safety study we maximized the signal to noise ratio associated with any damage or detriment to the patients treated. This was not intended to be an efficacy study, and the improvements in functional measures observed may be due to other clinical realities that results from frequent follow-ups such as increased compliance to medications.

## Limitations:

This study is limited in its design and scope from supporting any significant conclusions on the efficacy of the clinical approach presented. Although adverse events in three patients are unlikely to be due to the procedure performed, the absence of a control group prevents the elimination of this possibility. The results of this study suggest the need for a larger randomized trial.

## Conclusions:

In this open labeled uncontrolled safety trial of helical needle TE delivery of autologous bone marrow, the lack of procedural complications, the statistically significant improvements in LV ejection fraction, and trend towards benefit in end-diastolic volume, 24 hour Holter, and exercise tolerance support the safety of this technology and suggest the need for a larger trial to determine the overall ability to improve LV dysfunction after myocardial infarction.