Clinical Recovery From End-Stage Heart Failure Using Left-Ventricular Assist Device and Pharmacological Therapy Correlates With Increased Sarcoplasmic Reticulum Calcium Content but Not With Regression of Cellular Hypertrophy

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Background—Left ventricular assist device (LVAD) treatment is known to lead to structural and functional cellular modifications in the heart. The relevance of these changes for clinical recovery is unknown.

Methods and Results—We compared properties of cardiomyocytes obtained from tissue taken at explantation of the LVAD in patients with clinical recovery with those obtained from hearts of patients who did not show clinical recovery, thus requiring transplantation. Compared with myocytes taken at implantation, both the recovery and nonrecovery groups showed ≈50% reduction in cell capacitance, an index of cell size. However, action potential duration shortened, L-type Ca2+ current fast inactivation was more rapid, and sarcoplasmic reticulum Ca2+ content was increased in the recovery compared with the nonrecovery group.

Conclusions—These results show that specific changes in excitation-contraction coupling, and not regression of cellular hypertrophy, are specifically associated with clinical recovery after LVAD and further identify sarcoplasmic reticulum Ca2+ handling as a key functional determinant in patients with heart failure. (Circulation. 2004;109:2263-2265.)

Key Words: mechanical devices ■ electrophysiology ■ heart failure ■ myocytes ■ sarcoplasmic reticulum

Heart transplantation is currently the only effective long-term treatment for patients in end-stage heart failure. Recently, left ventricular assist device (LVAD) treatment has been used successfully as a bridge to transplantation, as destination therapy in patients with contraindications to transplantation, or to obtain recovery in a small number of patients.2,3 We have shown that in some patients with idiopathic dilated cardiomyopathy in end-stage heart failure, combined LVAD and pharmacological therapy induced clinical recovery to allow explantation of the device without recurrence of heart failure.3

LVAD treatment results in numerous changes in cardiomyocyte phenotype, excitation-contraction coupling, gene expression, and function.2 Whether these changes are beneficial is not known. Regression of hypertrophy is thought to be an important factor in recovery.2 However, several studies have also shown that hypertrophy is not required as a compensatory response to pressure overload,4 and its regression is not necessarily associated with normalization of electrophysiological parameters.5

Cardiomyocytes isolated from biopsies taken at explantation of the LVAD in patients with clinical recovery showed reductions in cell volume and cell surface area. L-type Ca2+ current and sarcoplasmic reticulum (SR) Ca2+ content were increased in myocytes during recovery, suggesting a critical role for intracellular Ca2+ homeostasis.6 Increased SR Ca2+ uptake in cardiac tissue after LVAD treatment has also been described previously.7 However, whether these specific cellular changes are related to clinical recovery remained unclear.

In this study, we compared findings in cardiomyocytes obtained from tissue taken from patients who recovered with those obtained from patients who, despite identical treatment, did not. This allowed correlation between cellular electrophysiological properties and clinical outcome.

Methods

We studied myocytes from patients at the time of LVAD implantation (LVAD core; n=7), from patients showing recovery after combined LVAD (HeartMateI, Thoratec Inc) and pharmacological treatment (biopsies at the time of device removal; n=6), and from patients who failed to recover and required cardiac transplantation (tissue from left ventricle of explanted hearts; n=4).

Before LVAD implantation, echocardiography showed left ventricular end-diastolic diameter of 78±12 mm, end-systolic diameter of 68±13 mm, and ejection fraction of 14±8% (n=6). In the patients showing recovery, end-diastolic diameter (15 minutes off-pump) decreased from 71±19 mm (before implantation) to 56±10 mm, end-systolic diameter decreased from 63±20 to 39±8 mm, and ejection fraction increased from 16±12% to 65±10% (n=6). In the
patients who underwent transplantation, end-diastolic diameter was 74±8 and 59±10 mm, end-systolic diameter was 69±10 and 52±11 mm, and ejection fraction was 17±9% and 27±17% before implantation and before transplantation, respectively (n=4; recovery versus transplanted, P<0.001). The duration of LVAD treatment was 425±209 days (n=6) for the recovery and 468±116 days (n=4) for the transplanted group.

Ventricular myocytes were enzymatically isolated as described previously, placed on the stage of an inverted microscope, and superfused with a normal Tyrode’s solution containing (in mmol/L) NaCl 140, KCl 6, MgCl2 1, CaCl2 1, glucose 10, and HEPES 10, with pH adjusted to 7.4 with 2 mol/L NaOH. Electrophysiological assessment was performed using switch-clamping and high (20 to 30 MΩ)-resistance microelectrodes containing KCl 2 mol/L, EGTA 0.1 mmol/L, HEPES 5 mmol/L, pH 7.2. Action potentials (APs) were triggered by 1 to 1.2 nA current pulses (frequency, 1 Hz; duration, 5 ms) and measured in current clamp. Cell capacitance (index of cell size) was measured as reported. The L-type Ca2+ current was the 200 μmol/L Cd–subtracted current recorded on voltage clamp steps from −40 to 0 mV for 500 ms (1 Hz). Decay of the current was fitted using 2-exponential decay. A fast time constant,τf, and slow time constant,τs, were measured. The SR Ca2+ content was measured by integrating a 20 mmol/L caffeine-induced transient inward current. Statistical approval was obtained by the Royal Brompton and Harefield NHS Trust Ethics Committee. Informed consent was obtained from each patient. Data are expressed as mean±SD. Statistical differences between means were calculated with 1-way ANOVA comparisons and Bonferroni post hoc tests.

Results

Compared with myocytes taken at implantation, both recovery and nonrecovery groups showed a similar reduction in cell capacitance (LVAD core: 290±61 pF; N=7 patients, n=30 cells; recovery: 142±28 pF; N=6, n=21; transplanted: 167±65 pF; N=4, n=14, P<0.001; Figure 1). APs recorded in myocytes from patients with recovery shortened significantly compared with myocytes from LVAD core. No such effect was observed in the nonrecovery group (time to 90% repolarization, LVAD core: 735±160 ms; N=7 patients, n=28; recovery: 454±58 ms; N=6, n=20; transplanted: 801±125 ms; N=4, n=13, P<0.01; Figure 1). This finding is supported by the observation that QTc intervals measured from ECGs of patients before LVAD implantation were longer compared with the recovery group. However, this effect was less pronounced in the patients undergoing transplantation (LVAD core: 473±50 ms; n=5; recovery: 397±2 ms, n=5; transplanted: 421±27 ms; n=4, P<0.05).

L-type Ca2+ current density measured in ventricular myocytes was not statistically different in the recovery versus the transplanted group (LVAD core: 1.32±0.49 pA/pF; N=6 patients, n=22; recovery: 2.62±0.79 pA/pF; N=5, n=16; transplanted: 2.13±1.98 pA/pF; N=4, n=12, P=NS). However, L-type Ca2+ current fast inactivation was significantly more rapid in recovery versus nonrecovery (τf, LVAD core: 16.4±3.3 ms; N=6, n=22; recovery: 9.5±1.7 ms; N=5, n=16; transplanted hearts: 19±5.5 ms; N=4, n=12; P<0.01, Figure 1), but the slow inactivation was unchanged (τs, LVAD core: 72±20 ms; N=6, n=22; recovery: 64±18 ms; N=5, n=16; transplanted hearts: 107±52 ms; N=4, n=12, P=NS). Finally, the SR Ca2+ content was increased in recovery compared with both the implantation and the nonrecovery groups (LVAD core: 30.3±16.9 μmol/L, nonmitochondrial (non mito) volume; N=7, n=25; recovery: 83.9±37 μmol/L; N=6, n=18; transplanted: 35±15 μmol/L; N=4, n=13; P<0.01; Figure 2).

Discussion

Our data strongly suggest that increased SR Ca2+ content is linked with clinical recovery by improving cardiac function. Lack of recovery is not accompanied by changes in SR Ca2+ content, AP duration, and Ca2+ current fast inactivation compared with myocardium from end-stage heart failure. In contrast, regression of cellular hypertrophy did not appear to influence recovery. These findings
further identify alterations in excitation-contraction coupling, and SR Ca\(^{2+}\) handling in particular, as a key functional determinant in patients with heart failure.

We hypothesize that an increased SR Ca\(^{2+}\) content is linked to clinical recovery by improving cardiac function, as previously shown in animal models.\(^{11}\) This can be obtained by increasing the gain of excitation-contraction coupling (more Ca\(^{2+}\) release for unitary trigger), resulting in larger Ca\(^{2+}\) transients and stronger cell contractions.\(^{12}\)

We also observed a faster inactivation of the Ca\(^{2+}\) current, and we suggest that this could be a consequence of a larger release of Ca\(^{2+}\) from the SR. This faster inactivation may possibly contribute to a shortening of the AP duration, which is known to occur in heart failure.\(^{14}\) In explanted hearts after LVAD treatment, AP shortening has been described previously.\(^{15}\) In this study, we show that this phenomenon is associated with clinical recovery. These results support the importance of electrical reverse remodeling in the clinical improvement observed in our patients.

In this study, a reduction in cell capacitance and, by inference, cell size was not always associated with clinical recovery. A reduction in size could be produced by regression of hypertrophy with restoration of function or by atrophy caused by unloading, with impairment of function. The balance between these 2 effects could explain the different outcome in the patients with or without recovery, and more investigation in this direction is required.

In summary, we have shown that clinical recovery from end-stage heart failure after mechanical and pharmacological treatment is linked with modifications in excitation-contraction coupling, and SR Ca\(^{2+}\) homeostasis in particular, and not to a reduction in cell size. These findings support the importance of SR Ca\(^{2+}\) regulatory mechanisms in the pathophysiology and treatment of severe heart failure in patients with dilated cardiomyopathy. Further research into the role of different forms of hypertrophy and/or atrophy in these patients is warranted.

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**References**


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