ELSEVIER

The Journal of
Heart and Lung
Transplantation

http://www.jhltonline.org

Partial mechanical circulatory support in an ovine model of post-infarction remodeling

Jef H. Geens, MD,[a] Steven Jacobs, MD,[a] Piet Claus, MD, PhD,[b] Sander Trenson, MD,[a] Veerle Leunens,[a] Ingrid Vantichelen, DVM,[a] Filip R. Rega, MD, PhD,[a] Erik K. Verbeken, MD, PhD,[c] Daniel Burkhoff, MD, PhD,[d] and Bart Meyns, MD, PhD[a]

From the [a]Department of Cardiac Surgery, University Hospitals Leuven, Leuven, Belgium; [b]Department of Cardiac Imaging, University Hospitals Leuven, Leuven, Belgium; [c]Department of Morphology and Molecular Pathology, University Hospitals Leuven, Leuven, Belgium; and [d]CircuLite Inc., Saddle Brook, New Jersey.

KEYWORDS:
chronic heart failure;
partial support;
ventricular assist devices (VADs);
sheep model;
reverse remodeling;
myocardial infarction

BACKGROUND: Full unloading of the left ventricle (LV) in chronic heart failure (CHF) induces reversal of LV dilation and geometric distortion. In this study we describe the partial unloading effects in ischemic CHF.

METHODS: Six weeks after myocardial infarction, sheep were randomized to partial support ("pump," $n = 5$), as provided by the CircuLite Synergy micro-pump, or to no therapy ("sham," $n = 6$) for an additional 6 weeks. At baseline, and at 6 and 12 weeks after infarction, pressure–volume (PV) recordings were made. Systolic and diastolic functions were characterized by the end-systolic volume (ESV) where LV end-systolic pressure reached 90 mm Hg (V_{90}), and the end-diastolic volume (EDV) where LV end-diastolic pressure reached 15 mm Hg (V_{15}), respectively. Magnetic resonance imaging (MRI) was performed 6 and 12 weeks after infarction. During autopsy at 12 weeks, isolated LVs were weighed. Histologically, the degree of fibrosis in the non-infarcted area was assessed using systematic randomized sampling, and myocyte hypertrophy was measured by the mean linear intercept method.

RESULTS: At 6 weeks, PV measurements showed a V_{90} and V_{15} increase (p = NS between groups). Six weeks later, V_{90} and V_{15} increased in the sham group. In the pump group, V_{90} decreased but V_{15} did not change significantly. At 6 weeks, MRI indicated no significant difference between groups. Six weeks later, in the sham group, EDV and ESV increased significantly. In the pump group, EDV decreased significantly and ESV trended to decrease. Sphericity index increased in the sham group and decreased in the pump group, although not significantly. Explanted LV masses were significantly higher in the sham group than in the pump group. The pump group had a decrease in fibrosis and less myocyte hypertrophy.

CONCLUSION: Partial support 6 weeks after major myocardial infarction halts and reverses ventricular dilation and hypertrophy.

J Heart Lung Transplant 2013;32:815–822

Chronic heart failure (CHF) is a major health problem in industrialized countries.[1] Regardless the CHF etiology, remodeling and neurohormonal activation are key components of disease progression. Ventricular remodeling comprises changes ranging from structural (e.g., increased sphericity) to histologic (e.g., myocyte hypertrophy) to subcellular (e.g., decreased sensitivity of contractile proteins to Ca^{2+}).[2] Ventricular assist devices (VADs) are now in common use in patients with end-stage heart failure (HF). VADs provide hemodynamic unloading of the left ventricle (LV) and normalize the neurohormonal milieu. This breaks

Reprint requests: Jef H. Geens, MD, Department of Cardiac Surgery, University Hospitals Leuven, Herestraat 49, B-3000 Leuven, Belgium. Telephone: +32-16-337-839. Fax: +32-163-44616.
E-mail address: jefgeens1979@gmail.com

1053-2498/$ - see front matter
http://dx.doi.org/10.1016/j.healun.2013.05.019

the vicious circle of CHF progression, and induces a process known as *reverse remodeling*, which includes a variety of changes "back to normal," from the sub-cellular to the structural level.[3] Although this approach rarely results in recovery of native heart function to the point where the VAD can be explanted,[4–10] even modest improvements in LV strength have the potential to provide clinical benefit. This is particularly the case when a partial ventricular support device is used, because, by definition, body perfusion still depends partially on blood flow from the native heart. However, as the degree of LV unloading achieved during partial support is less than during full ventricular support, it is unclear whether partial support induces significant reverse remodeling.

The Synergy micro-pump is a small, continuous mixed-flow pump that pumps at between 1.5 and 4.2 liters/min, depending on the pressure gradient and the rotational speed. In the clinical setting, the pump is generally set to supply ~3 liters/min and thus offers partial support, as evidenced by the fact that the aortic valve opens with every beat. Hemodynamic recordings from patients supported with this device show improvements in cardiac output (CO) and pulmonary arterial and capillary pressures that appear greater than could be expected by this degree of pump flow. It has therefore been hypothesized that reverse remodeling and some degree of functional recovery is achieved by partial support.[11]

To directly test this hypothesis, we developed an ovine model of post-myocardial infarct HF and implanted the Synergy pump. We quantified ventricular systolic and diastolic pump properties through pressure–volume analysis and compared the results to those of a control group with the same degree of post-infarct HF but did not receive a pump. Pressure–volume analysis has been shown to yield load-independent indexes of heart size and function that are sensitive for quantifying ventricular remodeling in the setting of HF, and reverse remodeling in the setting of effective therapies such as VADs.[12,13]

Other parameters that would support the presence and degree of reverse remodeling were obtained using magnetic resonance imaging (MRI), ex vivo passive diastolic pressure–volume analysis and heart weight.

Methods

This study was performed with the approval of and adherence to the guidelines of the animal ethics committee of the KU Leuven and according to the "Principles of Laboratory Animal Care," formulated by the National Society for Medical Research, and the *Guide for the Care and Use of Laboratory Animals*, prepared by the Institute of Laboratory Animal Resources (National Institutes of Health). Twenty-five female sheep (Texal crossbreed), weighing 52.3 ± 2.8 kg, were studied. Animals were sedated (15 mg/kg ketamine intramuscularly [IM]), induced (5% isoflurane, via face mask), intubated and mechanically ventilated. General anesthesia was maintained by isoflurane inhalation (2.5%). Peripheral arterial pressure was measured invasively with a catheter in an ear artery via a fluid-filled line connected to a pressure transducer (Maquet GmbH, Rastatt, Germany). Venous access was obtained via a deep venous line in the external jugular vein.

By way of an overview of the protocol (Figure 1), CHF was induced in all animals by acute coronary ligation. Six weeks after infarct, animals underwent invasive hemodynamic testing and MRI. Animals were then divided into 2 groups. The control group ($n = 6$) was observed for an additional 6-week period and did not receive any form of treatment. The partial support group ($n = 5$) underwent implantation of the Synergy pump and were also followed for 6 weeks. At the end of the 6-week follow-up period, the pump was explanted from the active group and all animals underwent repeat MRI, followed by invasive hemodynamic testing. Hearts were then explanted, and left and right ventricles were isolated and weighed.

Creation of myocardial infarction

Prior to the infarction procedure, the following anti-arrhythmics were administered: amiodarone (150 mg in 200 ml of 5% glucose) and lidocaine (250 mg in 200 ml of 0.9% NaCl) infused intravenously (IV) over 30 minutes plus $MgSO_4$ (2 g), given in a bolus injection. The heart was exposed via left thoracotomy. Then 10,000 IU of heparin was administered. The left anterior descending artery (LAD) was ligated permanently (using polypropylene 3/0) at the midpoint between the apex and base. The second diagonal was also ligated permanently at the same level. After a 15-minute equilibration period, the proximal LAD was occluded with a 3/0 Prolene snare for 1 hour. After hemodynamic stabilization, the thorax was closed. Animals were weaned from ventilation. Post-operative hypotension was treated with phenylephrine and, in some cases, milrinone (0.125 to 0.500 mg/kg/min). When present, ventricular ectopic beats were treated with lidocaine (2 mg/kg/h, IV) and amiodarone (0.5 mg/kg/h, IV). Piritramide (10 mg intramuscularly) was used for post-operative analgesia.

Pump implantation and sham thoracotomy 6 weeks after infarction

Six weeks after infarction, all animals underwent redo left thoracotomy. In control animals, the thoracotomy was immediately closed. Animals in the partial support group were implanted with a Synergy system. After exposure of the heart, 10,000 IU of heparin

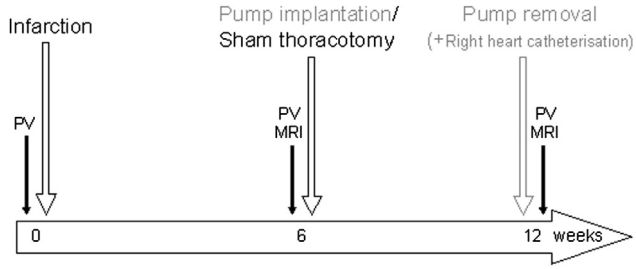

Figure 1 Time-points of surgical intervention and MRI. Pump group–specific events are indicated in gray. In both groups, PV measurements were performed before the infarction, at week 0. Six weeks after the infarction (i.e., "6 weeks"), PV measurements were repeated and MRI was performed. Subsequently, a thoracotomy was performed with and without pump implantation in the pump and sham groups, respectively. Six weeks later (i.e., "12 weeks" post-infarction), PV measurements and MRI were repeated: in the pump group, the pump had been explanted 0.5 to 1 day prior to these studies. Right heart catheterization was performed during pump removal.

was given. The inflow cannula, equipped with a flow probe, was inserted through the left atrial dome. The outflow graft was anastomosed to the descending aorta (5/0 polypropylene). Pump speed was set so that its flow rate, measured by the flow probe, was approximately half of the CO measured by thermodilution.

Brain natriuretic peptide measurements

Blood samples were taken on ice-cooled ethylene-diamine tetra-acetic acid (EDTA)-containing tubes at baseline and at 6 and 12 weeks. After centrifugation at 4°C (15 minutes at 6,000 rotations/min), plasma was immediately frozen at −80°C. After SepPak extraction, a radioimmunoassay procedure with ^{125}I porcine brain natriuretic peptide (BNP; ovine BNP26 is identical to ovine BNP) was performed.[14,15]

Pressure–volume loop recordings

Hemodynamic measurements were obtained at baseline and at 6 and 12 weeks after the infarction. A 7-French sheath (Cordis Corporation, Miami Lakes, Florida) was placed in the common carotid artery. An 8-French sheath (Cordis) and a custom-made balloon were inserted into the external jugular. The balloon is docked in the inferior vena cava. A Swan–Ganz catheter (Edwards Lifesciences Corporation, Irvine, California) was advanced to the pulmonary artery via the venous sheath. CO was measured by thermodilution; saline injections were repeated until at least 4 recordings within a 10% range of variation were obtained. A 12-electrode conductance catheter (Millar Instruments, Houston, Texas) was introduced via the arterial sheath and advanced to the LV apex. The catheter was connected to an Inca (CD Leycom, Zoetermeer) conductance signal conditioner. Pressure–volume loops were recorded during vena cava occlusion achieved by balloon inflation. Parallel conductance was estimated by bolus injection of hypertonic saline. The proportionality constant was determined by determining the ratio between thermodilution and conductance stroke volume.

In the pump group, right heart catheterization was performed at 12 weeks during surgery for pump removal. PV measurements at 12 weeks were performed 0.5 to 1 day after pump stoppage (after MRI; followed by killing and autopsy).

Magnetic resonance imaging

MRIs were performed under general anesthesia at 6 and 12 weeks after infarction. Animals were scanned in the right lateral decubitus with 3.0-Tesla MRI (Magnetom, Trio Tim; Siemens Medical Solutions, Erlangen, Germany) with a phased-array body coil wrapped over the heart. Images were acquired with electrocardiographic (ECG) gating and during suspended respiration. Cine images were acquired in vertical and horizontal long- and short-axis planes.

In the pump group, measurements at 12 weeks were performed 0.5 to 1 day after pump stop.

Ex vivo passive EDPVR

After explantation of the heart, a custom-made balloon was fitted into the LV and the mitral annulus was closed with a purse-string ligature. The balloon was progressively inflated while monitoring the pressure inside the balloon. Pressure was then plotted as a function of volume to assess ex vivo passive end-diastolic pressure–volume relations. Hearts of 5 healthy, weight-matched animals served as controls.

LV and RV mass

The right ventricular (RV) free wall was separated from the LV and both parts weighed. Hearts of 4 sheep that died at 6 weeks after myocardial infarction and 5 healthy, weight-matched animals were analyzed as well.

Fibrosis

An LV slice at the basal and the mid-ventricular level was fixed in 10% formalin followed by paraffin embedding, sectioning at 5-μm thickness and hematoxylin and eosin staining. If infarction was present, a region remote from the infarction was chosen. An 8-point grid with an area of 0.0625 mm^2 at 400× magnification was projected at 8 randomly chosen quadrants. The proportion of points hitting connective tissue equals the volume occupied by this compartment according to the principle of Delesse.[16] The hearts in the pump and sham groups, harvested at autopsy, were analyzed, as were the hearts of 4 sheep that died at 6 weeks after myocardial infarction.

Myocyte hypertrophy

Hematoxylin and eosin sections were analyzed by using the mean linear intercept method at the basal level in the remote area, free of fibrosis.[17] In brief, the number of myocytes transsected by a probe with a length of 500 μm at 200× magnification was counted on 10 fields per heart and then averaged. The number of transsected myocytes correlates with the inverse of myocyte volume and thus indexes myocyte hypertrophy. Sections of the pump and sham group were analyzed as were the 4 hearts harvested at 6 weeks after infarction.

Data analysis and statistics

Continuous variables were summarized by group using mean and standard deviation. Group differences were assessed by *t*-tests, with *p*-values adjusted for multiple comparisons when made. Conductance-derived pressure–volume loops were analyzed to obtain beat-by-beat pressure–volume points at end-diastole and end-systole (P_{es}, V_{es}, P_{ed}, V_{ed}) for construction of end-diastolic pressure–volume relations (EDPVRs) and end-systolic pressure–volume relations (ESPVRs), respectively. ESPVR data were fit by linear regression analysis to arrive at the slope (E_{es}, end-systolic elastance) and volume–axis intercept (V_o): $P_{es} = E_{es}(V_{es} - V_o)$. In addition, we calculated the volume on the ESPVR at which the pressure was 90 mm Hg ($V_{90} = 90/E_{es} + V_o$); this was used to index the position of the ESPVR on the pressure–volume plane for purposes of comparing relations from different time-points. EDPVR was linearized by logarithmic transformation of both end-diastolic pressures and end-diastolic volumes. Linear regression analysis yielded a slope (α) and an intercept [$\log(\beta)$] so that the EDPVR was quantified by $P_{ed} = \beta V_{ed}^{\alpha}$. In addition, we calculated the volume on the EDPVR at which the pressure was 15 mm Hg [$V_{15} = (15/\beta)^{1/\alpha}$]. MRI results were analyzed for LV end-diastolic and end-systolic volumes and for sphericity index using MATLAB (The MathWorks, Inc., Natick, Massachusetts).

Results

Mortality

Myocardial infarction was induced in 25 sheep, of which 11 completed the study. Nine sheep died peri-operatively due to

intractable arrhythmias. At the 6-week time-point, 1 sheep died at induction of anesthesia, 2 died at pump implantation, and 1 was found dead the day after sham thoracotomy. At 12 weeks, 1 sheep died at induction and another sheep died during pump removal (prior to hemodynamic measurements). One sheep had a superficial wound infection at the site of the pump pocket, which was successfully treated with amoxicillin (Duphamox; Pfizer) 750 mg/day intramuscularly for 10 days.

Hemodynamics and acute hemodynamic effects of partial support

Hemodynamic characteristics at baseline and 6 weeks after myocardial infarction are summarized in Table 1. Although baseline parameters were similar between the 2 groups, the HF degree in the partial support group was slightly worse, yet not statistically significant.

The acute effects of instituting partial support are illustrated in typical pressure–volume loops obtained from a representative animal (Figure 2). The blue loop was obtained in the HF state just prior to turning on the pump. The presence of HF is evidenced by the low systolic blood pressure (~60 mm Hg), the high end-diastolic pressure (just over 20 mm Hg), and the fact that the pressure–volume loop is situated on the steep portion of the EDPVR. The CO in this case was 2.7 liters/min. Upon initiation of pumping at 1.7 liters/min (i.e., 50% of the native cardiac output), the total CO determined by the Swan–Ganz catheter increased to 3.5 liters/min and there were significant changes in the pressure–volume loop. First, the end-diastolic pressure and volume decreased significantly (diastolic unloading), tracking down the EDPVR. Second, the loop was taller, showing the increase in blood pressure due to the increase in CO. Third, the width of the loop (which equates with the native heart stroke volume) became narrower, indicating that the heart was only partially contributing to the total CO. The other important point is that, unlike when full support was used, the aortic valve opened with every beat (i.e., the heart ejects with every beat) and there was significant pulse pressure.

In the partial support group, native CO averaged 3.1 ± 1.8 liters/min and pump speed was set so that pump flow averaged 1.8 ± 0.2 liters/min. The resulting total CO averaged 3.8 ± 0.9 liters/min ($p < 0.001$ vs CO pre-pump). Thus, the net increase in total CO (which averaged 0.7 liter/min, or a 22% increase) amounted to 40% of the flow rate of the pump, totally consistent with expectations.[18] The less than 1:1 ratio between change in total CO and pump flow was because the pump shunts blood away from the LV; with decreased filling, intrinsic LV output was decreased through the Frank–Starling relationship. Along with the changes in total CO, there were trends for reductions in pulmonary capillary wedge pressure (PCWP; 17.4 ± 4.2 vs 13.6 ± 2.1 mm Hg, $p = 0.1$) and increases in mean arterial blood pressure (MAP; 56.1 ± 11.5 vs 62.3 ± 13.7 mm Hg, $p = 0.15$). End-diastolic volume (EDV) decreased from 118 ± 27 to 107 ± 28 ($p = 0.001$), further evidence of partial unloading of the ventricle.

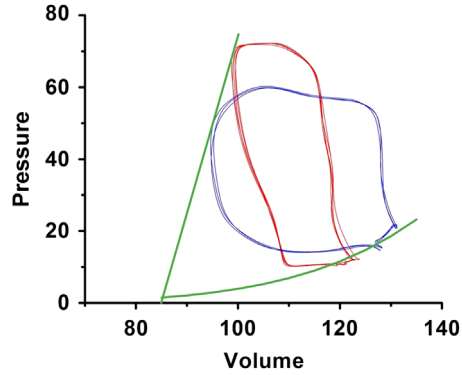

Figure 2 Acute effects of partial support on pressure–volume relations. The blue loop represents the heart-failing state immediately before starting the pump; the red loop was recorded after an equilibration period of 20 heart beats. On initiation of the pump (1.7 liters/min or half the total cardiac output), the end-diastolic pressure and volume decreased, and the end-systolic pressure increased. The loop became taller and more narrow, indicating that the pump partially supported the circulation.

At the time of pump explant, total CO during pumping averaged 3.1 ± 0.3 liters/min with the pump contributing 1.9 ± 0.2 liters/min. When the pump was turned off, CO decreased to 2.5 ± 0.4; this means that, similar to what was measured during the implant, the increase in CO due to partial support was ~32% of the pump's flow rate. With termination of pumping there were no detectable changes in PCWP (10.2 ± 3.6 vs 10.4 ± 2.4 mm Hg), but a trend for MAP to decrease (85.5 ± 10.3 vs 68.7 ± 14.9 mm Hg, $p = 0.07$).

Repeated right heart catheterization at the moment of PV measurement revealed no significant hemodynamic changes.

BNP measurements

Baseline BNP values were 3.76 ± 0.34 pmol/liter and 3.54 ± 0.48 pmol/liter in the sham and pump groups, respectively. At 6 weeks after myocardial infarction, BNP values were significantly increased compared with baseline

Table 1 Hemodynamic Parameters at Baseline and 6 Weeks After Induction of Heart failure

	Control group ($n = 6$)			Partial support group ($n = 5$)		
	Baseline	6 wk	p	Baseline	6 wk	p
Mean arterial pressure (mm Hg)	60.1 ± 7.0	54.1 ± 8.8	0.264	65.5 ± 15.4	60.1 ± 12.4	0.513
Mean pulmonary pressure (mm Hg)	13.1 ± 2.4	15.3 ± 2.9	0.337	15.8 ± 3.7	21.8 ± 1.4	0.006
Mean pulmonary capillary wedge pressure (mm Hg)	8.3 ± 2.2	12.6 ± 1.2	0.004	10.1 ± 3.2	17.3 ± 3.0	0.000
Cardiac output (liters/min)	3.6 ± 0.9	2.8 ± 0.2	0.146	3.9 ± 0.5	3.1 ± 0.6	0.011

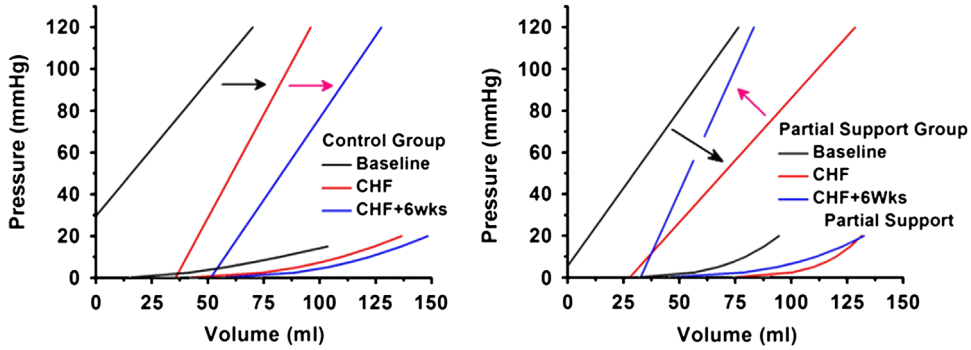

Figure 3 Pressure–volume relations at baseline, 6 weeks after the infarction, and after an additional 6 weeks without (left) and with (right) partial left ventricular support. In control animals (left panel) the mean ESPVR and EDPVR shift toward larger volumes 6 weeks after infarction (red line) and even further 6 weeks later (blue line). In the supported animals (right panel), the mean ESPVR and EDPVR are shifted equally toward the right after the infarction (red line). However, after 6 weeks of support (blue line), they are shifted back towards the left.

(7.14 ± 1.67 pmol/liter [$p < 0.001$] and 7.44 ± 1.44 pmol/liter [$p < 0.001$]). At 12 weeks, BNP increased further in the sham group, but did not change significantly in the pump group (10.07 ± 1.65 pmol/liter [$p = 0.03$] and 7.85 ± 0.06 pmol/liter [$p = 0.60$]) compared with 6 weeks.

Pressure–volume relations

The main findings of this study are summarized in Figure 3 and Table 2. The average results from the control group are shown in Figure 3A. Compared with baseline (shown in black) both ESPVR and EDPVR shift towards significantly larger volumes at 6 weeks after infarct induction (shown in red). E_{es} increased slightly, but V_o increased markedly, an indication of the decrease in contractile strength. The average EDPVR also shifted rightward, an indication that the heart had remodeled. After the additional 6-week follow-up period in the control group, both ESPVR and EDPVR shifted further rightward (shown in blue), indicative of additional remodeling and deterioration of systolic function. The degree of shifting of the EDPVRs and ESPVRs is further indexed in the progressive and significant increases in the values of V_{15} and V_{90}, respectively.

The findings in the partial support group are essentially the same as those for the baseline and 6-week follow-up measurements. The impact of 6 weeks of partial support is revealed by the pressure–volume relations, namely a *leftward* shift of the ESPVR (mainly due to an increase in E_{es} and minimal change in V_o). Consequently, there was a significant *reduction* in V_{90}. The EDPVR tends to shift towards *lower* volumes, but the reduction in V_{15} was not statistically significant. Note that these measurements were made *after* we explanted the pump, with the native heart fully supporting the circulation.

Magnetic resonance imaging

MRI was performed on animals at the 6- and 12-week follow-up time-points (after explantation of the pump in the treatment group). The results (Table 3) show that, in the control group, EDV, ESV and sphericity index increased substantially between 6 and 12 weeks, indicative of remodeling; there was no appreciable change in ejection fraction. In the partial support group, these volumes decreased, as did the sphericity index. Again, there was no appreciable change in ejection fraction.

Ex vivo passive EDPVR

The ex vivo passive EDPVR curve of the hearts that underwent 6 weeks of partial support is situated at lower volumes compared with the sham group, but at higher volumes than healthy controls (Figure 4).

Table 2 PressureVolume Analysis

	Control group			Partial support group		
	Baseline	6 wk	12 wk	Baseline	6 wk	12 wk
ESPVR						
E_{es}	1.3 ± 0.4	1.9 ± 1.0	1.5 ± 0.2	1.5 ± 0.1	1.1 ± 0.3	2.3 ± 1.4
V_o	−22.9 ± 15.9	35.5 ± 31.6	51.1 ± 17.4	−3.6 ± 17.0	27.6 ± 7.9	32.5 ± 37.9
V_{90}	53.5 ± 13.2	98.4 ± 12.1	116.6 ± 21	56.6 ± 16.3	117.7 ± 23.4	86.6 ± 14.5
p (6 wk vs 12 wk)		0.04			0.02	
EDPVR						
Alpha	2.2 ± 1	4.0 ± 1.8	5.8 ± 3.4	4.2 ± 1.1	8.3 ± 2.6	4.5 ± 1.6
Beta	0.085 ± 0.205	0.007 ± 0.017	0.005 ± 0.014	2.1E-5 ± 4.6E-5	6.4E-11 ± 1.4E-10	1.6E-5 ± 3.6E-5
V_{15}	103.4 ± 14.0	124.3 ± 14.5	136.3 ± 19.5	87.9 ± 14.8	126.5 ± 12.5	123.1 ± 14.0
p (6 wk vs 12 wk)		0.05			0.55	

Table 3 Magnetic Resonance Imaging

	Control group				Partial support group				
	6 wk	12 wk	Delta[a]	p^b	6 wk	12 wk	Delta	p^b	p^c
EDV (ml)	131.2 ± 24.9	153.8 ± 33.4	22.6 ± 12.5	0.016	146.2 ± 24.0	131.8 ± 22.7	−14.4 ± 5.3	0.004	0.000
ESV (ml)	95.6 ± 23.8	115.4 ± 26.4	19.8 ± 9.7	0.010	110.8 ± 24.5	103.6 ± 23.2	−7.2 ± 7.2	0.092	0.001
EF (%)	27.8 ± 7.9	25.0 ± 2.9	−2.8 ± 5.9	0.354	24.8 ± 6.6	22.0 ± 6.2	−2.8 ± 3.3	0.135	1.000
Sphericity	0.54 ± 0.08	0.60 ± 0.13	0.06 ± 0.07	0.135	0.52 ± 0.10	0.46 ± 0.10	−0.06 ± 0.08	0.153	0.033

[a]Delta, change between 12 wk and 6 wk within each group.
[b]p for comparison between 6 wk and 12 wk within each group.
[c]p for comparison of delta values between groups.

LV and RV mass

LV and RV mass was significantly higher at 6 weeks after the infarction, compared with healthy controls. At 12 weeks, LV mass was significantly lower in the partial support group. RV mass was also lower, but not as significantly reduced as in the LV (Table 4).

Fibrosis

The degree of fibrosis was 5.6 ± 1.7% of the non-infarcted LV mass in the 4 sheep at 6 weeks after infarction. At 12 weeks, fibrotic tissue accounted for 2.9 ± 1.9% and 6.4 ± 2.3% in the pump and sham groups, respectively ($p = 0.031$ between groups).

Myocyte hypertrophy

The number of transsected myocytes was 23.2 ± 2.6 and 19.1 ± 0.6 per 0.5 mm in the non-fibrotic areas of the pump and sham groups, respectively ($p = 0.004$). This was 18.3 ± 1.3 in the 4 sheep at 6 weeks after infarction ($p = 0.011$ vs pump group, p = NS vs sham group).

Discussion

We have created an infarct-related ovine model of CHF, as evidenced by hemodynamics (increased wedge pressure and decrease in CO) and a significant increase in BNP, to test whether partial support provided by the Synergy micro-pump results in reverse remodeling. Six weeks after myocardial infarction, significant rightward shifts of ESPVR and EDPVR were observed, indicating remodeling. The significantly increased LV mass of 4 sheep at 6 weeks after infarction compared with 5 healthy controls supports this finding. After a sham thoracotomy and another 6 week observation period in the control group, both ESPVR and EDPVR shifted further rightward, particularly the ESPVR. In the partial support group, pump flow rates were adjusted at the time of implant to ~50% of each animal's native heart CO.

Measurements made both at the time when support was initiated and at the end of the follow-up period when support was terminated showed the characteristic, expected impact of left atrial-to-arterial partial support on the pressure–volume loop.[18] These consisted of reductions in end-diastolic pressures and volumes (diastolic unloading), narrowing of the loop (indicating decreased stroke volume of the native LV) and increases in peak pressure (due to the increased total output). These changes in the pressure–volume loop are significantly different from what is observed when blood is pumped directly from the ventricle, such as in the currently used VADs, during which there is generally more pre-load reduction, less increase in arterial pressure, and loss of isovolumic periods due to continuous pumping of blood throughout the cardiac cycle.[12] Thus, the nature and magnitude of ventricular unloading achieved with the Synergy system differ from what is achieved with the majority of currently used VADs. It was previously unknown whether partial support with a left atrial-to-arterial flow path induces reverse remodeling.

After 6 weeks of partial support, there was indeed a leftward shift of the in vivo ESPVR and, even after removal of the pump, there were significantly lower values of EDV by MRI. Ex vivo passive EDPVRs showed a shift to the left in the partial support group compared with the control group; LV mass was decreased and, histologically, there

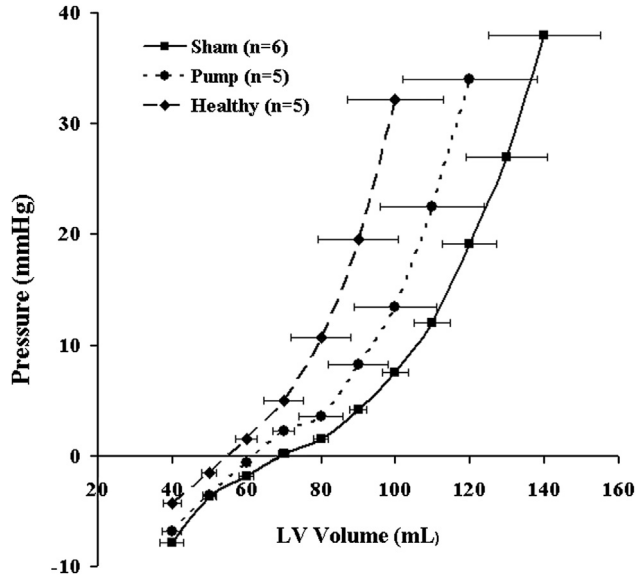

Figure 4 Ex vivo passive EDPVRs of explanted hearts. The mean ex vivo end-diastolic pressure–volume relationship of the pump group (dotted line) is situated at lower volumes compared with the sham group (solid line), but at higher volumes when compared with healthy control hearts (broken line).

Table 4 LV and RV Mass

	Healthy ($n = 5$)	6 weeks ($n = 4$)	Sham ($n = 6$)	Pump ($n = 5$)
LV mass (g)	90.2 ± 8.8	127.9 ± 15.6[a]	141.0 ± 11.2	103.7 ± 9.8[b]
RV mass (g)	33.0 ± 3.5	43.5 ± 5.5[c]	40.6 ± 5.1	36.8 ± 3.1[c]

[a] $p < 0.001$ vs healthy controls.
[b] $p = 0.001$ vs sham group.
[c] $p = 0.022$ vs sham group.

was less hypertrophy with partial support. Taken together, these findings indicate *reverse remodeling*.

At the level of the extracellular matrix, we found a lower degree of fibrosis in the pump group. Studies concerning connective tissue changes after mechanical support in patients yielded conflicting results: some reported an increase in fibrosis; some reported a decrease in fibrosis; and others reported no change in the total collagen content.[3]

We consider the reverse remodeling effects as probably not durable. Because it is highly unlikely that the pump induced changes in infarct mass, we expect that the spontaneous evolution after explantation of the pump would be a progressive loss of this reverse remodeling effect in a process called *recurrent remodeling*.[19]

Earlier studies of reverse remodeling during mechanical support have been primarily derived from retrospective studies of biologic material obtained from patients with devices that provide full support and more significant LV unloading.[3] Performing these studies in animals offers several advantages over human studies, including ability to have a more uniform HF degree between animals, the availability of a concurrent control group, and capacity to regulate background medical therapies (in the present study no other therapy was provided). However, studies have been limited by the lack of a suitable animal model in which VADs can be implanted for long periods. The current studies were facilitated by the small size of the Synergy pump, which was suitable for long-term studies.

Naturally, there are several limitations of this model and of the present study. First, there was a small number of animals that completed the study due to the high rate of mortality. This high mortality (~50%) is typical for large-animal HF models.[20–22] Second, compared with a clinical setting, the current study was conducted over short time periods, both with regard to the period allowed for establishing the HF state (6 weeks) and for the duration of support. The results from the control group show that there was still progressive remodeling between 6 and 12 weeks and, therefore, the Synergy system was introduced at a time when the heart was still actively remodeling. Studies of the time dependence of various aspects of reverse remodeling during full VAD support suggest that 6 weeks may be enough time to establish a near steady state for molecular and structural reverse remodeling, but not likely long enough to see the final degree of changes in the extracellular matrix. It is unknown whether the time course of reverse remodeling is slower with partial support. Because of these limitations, the current results should not be translated quantitatively to the clinical setting but should instead be considered a proof of concept that partial support can result in reverse remodeling.

In summary, left atrial-to-aortic partial ventricular support unloads the heart, improves blood pressure and CO and induces reverse remodeling. It is unknown how the degree of reverse remodeling achieved with this approach compares with what could be achieved in this model by full support.

Because the control group exhibited active remodeling (further ventricular dilation and increase in LV mass at 6 weeks after the infarction) and no third group receiving full support was included, we are unable to draw definitive conclusions regarding the inherent effect of the partial support modality of mechanical circulatory support. Nevertheless, the current model offers the possibility to study, in a large-animal model, the impact of concomitant pharmacologic and device-based treatment on reverse remodeling.

Disclosure statement

D.B. is Chief Medical Officer of CircuLite, Inc. The other authors have no conflicts of interest to disclose. This work was partially supported by the Belgian Fund for Cardiac Surgery and the European Union Seventh Framework Programme "Sensorart."

References

1. Bui AL, Horwich TB, Fonarow GC. Epidemiology and risk profile of heart failure. Nat Rev Cardiol 2011;8:30-41.
2. Opie LH, Commerford PJ, Gersh BJ, et al. Controversies in ventricular remodelling. Lancet 2006;367:356-67.
3. Klotz S, Jan Danser AH, Burkhoff D. Impact of left ventricular assist device (LVAD) support on the cardiac reverse remodeling process. Prog Biophys Mol Biol 2008;97:479-96.
4. Dandel W, Weng Y, Siniawski H, et al. Heart failure reversal by ventricular unloading in patients with chronic cardiomyopathy: criteria for weaning from ventricular assist devices. Eur Heart J 2011;32: 1148-60.
5. Torre-Amione G, Loebe M. Myocardial recovery following prolonged mechanical support. In: Frazier OH, Kirklin JK, editors. ISHLT monograph series. Mechanical circulatory support. New York: Elsevier; 2006. p. 155-69.
6. Young JB. Healing the heart with ventricular assist device therapy: mechanisms of cardiac recovery. Ann Thorac Surg 2001;71(suppl): S210-9.
7. Ferrar DJ, Holmann WR, McBride LR, et al. Long-term follow up of Thoratec ventricular assist device bridge-to-recovery patients successfully removed from support after recovery of ventricular function. J Heart Lung Transplant 2002;21:516-21.
8. Levin HR, Oz MC, Catanese KA, et al. Transient normalization of systolic and diastolic function after support with a left ventricular assist device in a patient with dilated cardiomyopathy. J Heart Lung Transplant 1996;15:840-2.

9. Müller J, Wallukat G, Weng Y, et al. Weaning from mechanical cardiac support in patients with dilated cardiomyopathy. Circulation 1997;96: 542-9.
10. Frazier OH, Myers TJ. Left ventricular assist system as a bridge to myocardial recovery. Ann Thorac Surg 1999;68:734-41.
11. Meyns BP, Simon A, Klotz S, et al. Clinical benefits of partial circulatory support in New York Heart Association class IIIB and early class IV patients. Eur J Cardiothorac Surg 2011;39: 693-8.
12. ten Brinke EA, Klautz RJ, Tulner SA, et al. Clinical and functional effects of restrictive mitral annuloplasty at midterm follow-up in heart failure patients. Ann Thorac Surg 2010;90:1913-20.
13. Steendijk P, Tulner SA, Bax JJ, et al. Hemodynamic effects of long-term cardiac resynchronization therapy: analysis by pressure-volume loops. Circulation 2006;113:1295-304.
14. Aitken GD, Raizis AM, Yandle TG, et al. The characterization of ovine genes for atrial, brain, and C-type natriuretic peptides. Domest Anim Endocrinol 1999;16:115-21.
15. Pemberton CJ, Yandle TG, Charles CJ, et al. Ovine brain natriuretic peptide in cardiac tissues and plasma: effects of cardiac hypertrophy and heart failure on tissue concentration and molecular forms. J Endocrinol 1997;155:541-50.
16. Delesse A. Procédé mécanique pour déterminer la composition des roches. CR Acad Sci Paris 1847;25:544-5.
17. Dunnill MS. Quantitative methods in the study of pulmonary pathology. Thorax 1962;17:320-8.
18. Morley D, Litwak K, Ferber P, et al. Hemodynamic effects of partial ventricular support in chronic heart failure: results of simulation validated with in vivo data. J Thorac Cardiovasc Surg 2007;133:21-8.
19. Helman DN, Maybaum SW, Morales DL, et al. Recurrent remodeling after ventricular assistance: is long-term myocardial recovery attainable? Ann Thorac Surg 2000;70:1255-8.
20. Huang Y, Kawaguchi O, Zeng B, et al. A stable ovine congestive heart failure model. A suitable substrate for left ventricular assist device assessment. ASAIO J 1997;43:M408-13.
21. Nakajima H, Nakajima HO, Hammond RL, et al. Chronic changes of end-systolic pressure-volume relationship after regional myocardial infarction. J Card Surg 1995;10:652-64.
22. Devlin G, Matthews K, McCracken G, et al. An ovine model of chronic stable heart failure. J Card Fail 2000;6:140-3.