ORIGINAL ARTICLE

The impact of left ventricle assist device on circulating endothelial microparticles – pilot study

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Abstract

OBJECTIVES: Recent technological breakthroughs in the design of reliable systems for long term non-pulsatile mechanical heart support offer the possibility to study the effect of continuous blood flow in the vascular system. Generally, it is assumed that the absence of physiological pulsatile flow leads to prothrombogenic and proatherogenic changes. We investigated the change in the circulating endothelial microparticle concentration as a marker of endothelial damage in patients implanted with a continuous-flow left ventricle assist device (LVAD).

METHODS: Endothelial microparticles were measured in 8 males (mean age 54.1±11.5 years) with terminal heart failure before and 3 months after implantation of an LVAD. The group consisted of 3 patients with dilated cardiomyopathy, 3 patients with ischemic cardiomyopathy, 1 patient with both conditions and 1 patient with congenital valvular disease. The concentration of endothelial microparticles was determined by ELISA Zymutest MP activity test.

RESULTS: We did not observe a significant change in the concentration of circulating endothelial microparticles measured before and 3 months after implantation (p=0.669). High inter-individual variability in response to implantation was found. However, no association between a change in endothelial microparticle concentration and heart failure aetiology or a significant clinical complication attributed to LVAD implantation was observed.

CONCLUSION: Results from this preliminary pilot study do not indicate that LVADs contribute to short-term vascular damage as defined by an increase in circulating endothelial microparticles.

Abbreviations:

ConVD	- congenital valvular disease
DCM	- dilated cardiomyopathy
IHD	- ischemic heart disease
LVAD	- left ventricular assist device
nMPS	- nano moles per litre relative to phosphatidylserine

INTRODUCTION

In patients with terminal heart failure, one crucial therapeutic option is the use of a mechanical heart support/left ventricular assist device (LVAD). These implantable pumps typically draw blood from the left ventricle and pump into the aorta, in this respect replacing the function of the failing heart/left ventricle. Recently, LVADs have mainly served as a bridge to heart transplant, but with their increasing long term reliability, their use as a long term solution for terminal heart failure (destination therapy) is expected (Coyle *et al.* 2010; Kirklin *et al.* 2012; Park *et al.* 2012). This approach could further extend the life of patients, especially those who are not eligible for various reasons to receive a heart transplant (Long 2008; Lund *et al.* 2010; Milla *et al.* 2012; Miller *et al.* 2007). The technological breakthrough leading to an increase in the long term reliability of LVADs was the introduction of rotary pumps with a minimum of moving parts (Pagani *et al.* 2009; Slaughter *et al.* 2009). This innovation enabled the miniaturisation of the system and reduced the risk of mechanical failure. Although these systems generate a continuous flow, this mostly non-pulsatile circulation is surprisingly well tolerated in the short- and medium-term (Rogers *et al.* 2010). However, the potential untoward impact of continuous flow has raised further questions regarding the long term effect this therapy may impose on the vascular system and its potential contribution to end-organ damage (Pirbodaghi *et al.* 2012a;b). As it turns out, continuous and pulsatile flow have different effects on hemodynamics and the vascular system in general (Chiu & Chien 2011). Reduced pulsatility and reduced vascular cyclic stress can cause atrophy of the vascular wall or reduce the calibre of vessels. In addition, the absence of cyclic strain on the endothelium, non-pulsatile circulation may lead to endothelial dysfunction (Zieman *et al.* 2005). Close and complex follow up of vascular and hemodynamic changes in patients with implanted LVADs could reveal the consequences of non-pulsatile versus pulsatile flow in greater detail. Recent preliminary data from the Mayo Clinic show a temporary improvement in endothelial function after implantation of mechanical support but also demonstrate decreasing endothelial reactivity (measured by changes in reactive hyperaemia) during extended follow up in the non-pulsatile treated group (Hasin *et al.* 2012b). In addition, complications due to LVADs such as cerebrovascular damage as well as peripheral vascular compromise are also of concern (Rose & Park 2005; Potapov *et al.* 2011). Non-invasive methods of measuring vascular damage have been highly sought after; one newly observed indicator of vascular damage is circulating endothelial microparticles. These complexes may represent the end products of apoptosis of endothelial cells but may also act as strong disregulators of endothelial function (Dignat-George & Boulanger 2011; Chironi *et al.* 2010; Robert *et al.* 2012). These microparticles are produced from membranes, cytoplasmic and nuclear constituents from precursor cells; their production is part of normal cell function, but production increases in cells under stress, including endothelial cells. Additionally, their levels have been shown to increase in several pathological conditions including transplantation (Amabile & Boulanger 2011; Boulanger 2010; Brodsky *et al.* 2012). Use of these markers of endothelial dysfunction and vascular damage could circumvent technical difficulties encountered during measurements of artery dilation in the presence of non-pulsatile flow and could expand our knowledge in this field. Therefore, we studied the short term impact of mechanical cardiac support on the concentration of circulating endothelial microparticles in a prospective study.

MATERIALS AND METHODS

Our study population was composed of 8 males indicated for LVAD due to terminal heart failure and dependence on permanent inotropic support. Three patients were diagnosed with dilated cardiomyopathy (DCM), 3 with ischemic heart disease (IHD), 1 patient was diagnosed with both conditions (DCM/IHD), and 1 patient suffered from congenital valvular heart disease (ConVD). Two patients with IHD, 1 patient with DCM and the patient with ConVD had severe pulmonary hypertension. In all patients, the LVAD was used as a bridge to heart transplant.

All patients were anticoagulated with heparin and later with dicumarols prior to and after implantation. All patients were on inotropic/catecholamine support before LVAD implantation. No substantial changes in oral vasoactive drugs were made prior to and after implantation. However, before implantation, patients with IHD were treated with acetylsalicylic acid and statins in contrast to patients with other diagnoses.

In all participants prior to and 3 months after implantation of LVADs, the concentrations of circulating endothelial microparticles were recorded.

All patients received continuous-flow LVAD – HeartMate II (Thoratec Corp., Pleasanton, California) implantation. Analysis of the microparticles was performed in a specialised lipid laboratory, which was under the continuous external quality control of the CDC Atlanta, USA. The concentration of endothelial microparticles was determined by an ELISA Zymutest MP activity test (Hyphen Biomed, France) according to already established methodology (Slavik 2010) and expressed as nano moles per litre relative to phosphatidylserine (nMPS). The microparticles were measured in duplicate, and the mean of two measurements was used for further analyses. In addition, important clinical data, which included history and causes of heart failure, medication use, traditional cardiovascular risk factors, body mass index and systolic blood pressure measured in the arms by a continuous-wave Doppler ultrasound device with 10 MHz probe (Sonovit SV-1, Schiller AG,

Switzerland) and a standardised mercury sphygmomanometer with the appropriate sized cuff, were collected according to an established protocol. Other analysed data included the ejection fraction as established by echocardiography and laboratory parameters of importance including parameters of renal function and quantification of brain natriuretic peptide levels. The ethics committee of the Institute approved this study, and all participants provided their signed informed consent.

All data were stored in an electronic database. For continuous data, the mean and standard deviation were employed, and for categorical data, percentages were used. The statistical analysis used t-tests for continuous variables and a chi-square test for categorical variables. Differences in monitored parameters over time were evaluated using a paired t-test. STATA statistical software was used for data processing.

RESULTS

In Table 1, the main clinical characteristics of our study patients are presented before and after implantation of the device. In the entire group, significant improvement in ejection fraction established by echocardiography and a significant decrease in brain natriuretic peptide were observed 3 months after implantation. No significant difference in the circulating endothelial microparticles was found before and after implantation of the device (p=0.67) (Table 1). However, substantial inter-individual variability among our 8 patients was observed in response to implantation (Figure 1).

Prior to implantation, circulating endothelial microparticles in 3 patients with a diagnosis of DCM were not significantly higher than in patients with a diagnosis of IHD (5.98±4.23 versus 2.37±1.42 nM PS; p=0.27). After implantation, the change in circulating endothelial microparticles was also similar and non-significant in both patients with DCM (6.07±5.68 nM PS; p for change = 0.97) and in patients with IHD (4.04±3.56 nMPS; p for change = 0.57). Complications during follow up were as follows: 1 patient (IHD) suffered a blocked pump due to thrombosis; 1 patient (DCM/IHD) suffered right ventricular failure which was sufficiently managed by medication; 1 patient (ConVD) experienced a moderately severe haemorrhage; and 1 patient (IHD) experienced a thoracic empyema, which was successfully treated. In the remaining patients, no incidents of serious bleeding, infections, thromboembolic events, no signs of right ventricular failure, no evidence of serious end organ damage, and no malfunction of the pump were observed during 3 months of follow up. After implantation, circulating endothelial microparticles were not significantly lower in patients with clinical complications than in patients without complications (3.46±1.74 versus 6.57±4.75 nMPS, respectively; p=0.26). The change in circulating endothelial microparticles after 3 months was also not significantly different in patients with clinical complications (p=0.57) or in patients without clinical complications (p=0.52).

Tab. 1. Main characteristics of patients prior to and 3 months after implantation of left ventricular assist device (LVAD).

	Prior to implantation	After implantation	p-value (t-test)
Age	54.1±11.5	NA	NA
Endothelial microparticles (nMPS)	4.48±3.21	5.02±3.71	0.669
Current smokers (n)	0	NA	NA
Hypertension (n)	0	NA	NA
History of dyslipidemia (n)	2	NA	NA
Diabetes mellitus (n)	2	NA	NA
Ejection fraction established by echocardiography (%)	20±4	28±8	0.007
Body mass index (kg/m^2)	24.5±3.2	25.8±2.1	0.09
Systolic blood pressure (mm Hg)	84.9±15.8	82.4±8.3	0.43
Pulsatility of LVAD	NA	5.50±0.49	NA
Brain natriuretic peptide (ng/l)	1579.5±1215.1	423.7±162.1	0.048
Plasma creatinine (μmol/l)	119.4±31.3	99.7±48.9	0.330
Free hemoglobin after implantation (mg/dl)	NA	296.8±159.9	NA

means ± SD if not stated differently; LVAD-left ventricular assist device; nMPS- nanomoles of endothelial microparticles per liter relative to phosphatidylserine; NA – non -applicable.

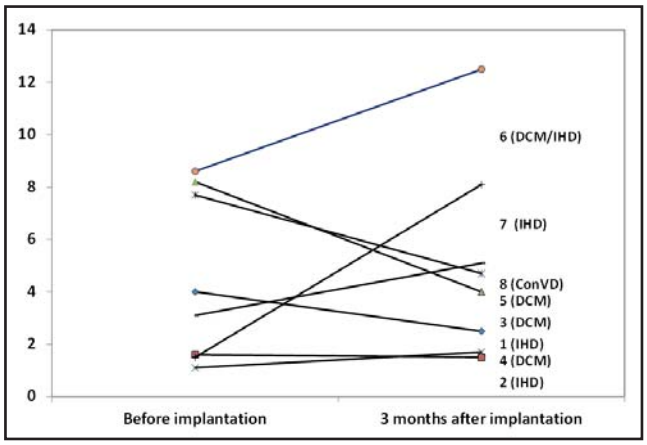

Fig. 1. Individual changes of circulating endothelial microparticles prior to and 3 months after implantation of left ventricular assist device (LVAD). Units: nMPS- nanomoles of endothelial microparticles per liter relative to phosphatidylserine, DCM - dilated cardiomyopathy, IHD – ischemic cardiomyopathy, ConVD – congenital valvular heart disease.

DISCUSSION

In this pilot study, we did not detect significant changes in circulating endothelial microparticles 3 months after implantation of LVAD with non-pulsatile flow in 8 patients with terminal heart failure. To the best of our knowledge, this is the first study that assessed endothelial function/vascular damage using the methods described herein. In previous studies, endothelial function was assessed by standard methods and impaired endothelial function was found (Hasin *et al.* 2012b). Interestingly, the same group in another study described improved renal function in patients with LVAD potentially due to increased renal perfusion (Hasin *et al.* 2012a). However, the results gathered using these methods could be strongly affected by non-pulsatile flow. The measurement of circulating endothelial microparticles is another predictor of endothelial dysfunction and avoids some of the disadvantages of methods based on changes in the reactivity of peripheral vessels. In our study, we detected substantial variability among individual patients with regard to changes in the concentration of circulating endothelial microparticles after implantation of a LVAD. These findings appeared to be independent of heart failure aetiology, and clinical complications attributed to LVAD implantation. Our study is limited by the low number of participants and their clinical heterogeneity. Another potential confounder is the complex natural history of circulating endothelial microparticles; it is still not clear if these particles are derived exclusively from endothelium, or also from circulating blood cells or platelets (Burger & Touyz 2012; Dignat-George & Boulanger 2011; Morel *et al.* 2011; VanWijk *et al.* 2003). As previously described, treatment by acetylsalicylic acid (Bulut *et al.* 2011) and statins (Huang *et al.* 2012) in IHD patients could decrease the number of microparticles; in the case of statins, this interaction may be more complex (Tramontano *et al.* 2004; Mobarez *et al.* 2012). Dietary habits could also have an impact on circulating endothelial microparticles and may demonstrate different results in patients with ischemic heart disease (Vafeiadou *et al.* 2012). However, we did not find strong evidence to suggest a difference between patients with ischemic disease and patients with dilated cardiomyopathy, neither did we find a significant change in circulating endothelial microparticles between patients diagnosed with DCM or IHD. This prospectively designed study provides a novel assessment of circulating endothelial microparticles in a unique patient population prior to and after LVAD implantation with non-pulsatile flow. The methodology used in this study mitigates the difficulties inherent in the interpretation of results gathered from dilation of peripheral vessels with non-pulsatile flow. Our results indicate that non-pulsatile flow does not cause damage to the endothelium and/or other circulating cells in the short term. Clinical complications attributable to LVAD implantation observed in half of our patients also did not substantially change the concentration of circulating endothelial microparticles during 3 months of follow up. Because of the uniqueness of our study population, these results on the effect of LVADs on vascular health add to our understanding of the complex nature of vascular changes after the implantation of non-pulsatile devices. Complex evaluation of the hemodynamic changes and biological markers during the management of terminal heart failure may provide important and unique information for monitoring and treating patients with implanted LVADs in the longer term. In conclusion, we did not detect an adverse effect of mechanical heart support on endothelial or vascular function assessed by circulating endothelial microparticles. However, these findings may well be dependent on the complex characteristics of individual patients and should be further evaluated in larger patient populations with extended follow up.

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