Nonphysiologic blood flow triggers endothelial and arterial remodeling in vivo: Implications for novel left ventricular assist devices with a peripheral anastomosis

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Objectives: Less invasive circulatory support devices have been developed that require anastomosis to a peripheral artery. The Symphony Heart Assist System (Abiomed, Inc, Danvers, Mass) is a volume-displacement pump sewn to the subclavian artery to provide partial circulatory support. The surgical configuration produces nonphysiologic blood pressure and bidirectional flow in the subclavian artery. Our objective was to identify effects of altered hemodynamics on arterial structure and function.

Methods: In calves (n = 23; 80-100 kg), the Symphony pump was sewn end-to-side to the carotid artery. Acutely, carotid blood pressure and flow were recorded to evaluate hemodynamic changes. After medium-term support (1-4 weeks), carotid artery was studied. Histologic and molecular assays evaluated architectural changes. Quantitative real-time polymerase chain reaction evaluated gene expression of matrix metalloproteinase (MMP)-2, MMP-9, and connective tissue growth factor. In vitro carotid arterial-ring studies evaluated physiologic responses.

Results: During Symphony support, carotid arterial pressure was 200/15 mm Hg. Antegrade flow increased significantly ($P < .05$) from 1.40 ± 0.32 to 4.29 ± 0.33 L/min. Flow during native cardiac diastole reversed completely from 0.25 ± 0.05 to −4.15 ± 0.38 L/min in carotid artery proximal to the anastomosis. After medium-term support, the carotid artery was significantly dilated with significantly thinner tunica media and thicker tunica adventitia than in control carotid arteries. MMP-9 gene expression decreased significantly, connective tissue growth factor gene expression increased significantly, and collagen, elastin, and total extracellular matrix increased significantly. Endothelial cells were significantly hypertrophied and produced significantly more von Willebrand factor. Endothelial apoptosis increased significantly. Platelet-endothelial interactions decreased significantly. Endothelial-independent contraction decreased significantly, whereas endothelial-dependent relaxation increased modestly.

Conclusions: Assisted circulation with a left ventricular assist device triggered arterial remodeling that allowed a peripheral artery to accommodate the altered hemodynamics of a novel partial-support pump. Further delineation of remodeling pathways may be of significance for the emerging field of partial circulatory support. (J Thorac Cardiovasc Surg 2014;148:311-21)

Mechanical circulatory support has evolved into a standard therapy for adult patients with advanced heart failure. With nearly 4300 patients studied, the Interagency Registry for Mechanically Assisted Circulatory Support reported a recent 2-year survival of 74% with current devices.[1] However, concerns have been raised that abnormal hemodynamics in these patients may trigger unfavorable changes in myocardial structure.[2] In particular, prolonged unloading of the failing left ventricle may reduce myocardial mass and produce myocyte atrophy and ventricular stiffening.

Yet, arterial remodeling during left ventricular assist device (LVAD) support has not received the same attention. It is well known that blood pressure and blood flow influence arterial architecture and function.[3,4] However, if hemodynamics are maintained outside the normal physiologic range, the effect on arterial tissues is uncertain. Arteries exposed to supraphysiologic pressures,

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Abbreviations and Acronyms

CTGF = connective tissue growth factor
DAPI = 4′,6-diamidino-2-phenylindole
FITC = fluorescein isothiocyanate
LVAD = left ventricular assist device
MMP = matrix metalloproteinase
PCR = polymerase chain reaction
PSS = physiologic saline solution

flows, and shear stress from an LVAD may undergo changes in size, architecture, and function.

A number of novel circulatory support devices are being developed that require an end-to-side anastomosis to a peripheral artery.[5,6] This anatomic configuration forces a medium-sized artery to accommodate significant increases in blood flow, blood pressure, and, with certain devices, bidirectional blood flow. The effects on arterial structure and function are unknown.

We studied a novel partial-support LVAD (Symphony Heart Assist System; Abiomed, Inc, Danvers, Mass) in a bovine model. The Symphony is a 30-mL pump designed to deliver prolonged ambulatory partial circulatory support for months to years in patients with advanced heart failure. The pump is implanted posterior to the pectoralis major muscle, which precludes the need for a sternotomy or thoracotomy (Figure 1, *B*). A modified polytetrafluoroethylene graft is sewn to the subclavian artery. The patient's continuous electrocardiogram triggers real-time filling and emptying of the Symphony pump. The pump fills during native cardiac systole to reduce left ventricular afterload and ejects during native cardiac diastole to augment diastolic blood pressure. This results in improved coronary and systemic blood flow. By these mechanisms, the myocardial oxygen supply-demand relationship improves,[5] and end-organ perfusion increases. A clinical trial with the Symphony pump is currently underway in Canada and France.[7]

The anatomic configuration of the Symphony produces a unique hemodynamic profile near the anastomosis. The segment of the subclavian artery between the outflow graft and the aorta experiences a nonphysiologic pattern of alternating antegrade-retrograde blood flow during pump filling and emptying, respectively (Figure 1). The effects of altered pressure and bidirectional flow on arterial structure and function have not previously been studied. Consequently, we hypothesized that a nonphysiologic profile of blood flow triggered endothelial and arterial remodeling in vivo. We investigated structural and functional changes that occurred after 1 to 4 weeks of support with the Symphony pump.

MATERIALS AND METHODS

This study was conducted in accordance with the National Institutes of Health *Guide for the Care and Use of Laboratory Animals* (Guide for the Care and Use of Laboratory Animals). All experimental procedures were approved by the institutional animal care and use committee of the University of Louisville (Louisville, Ky).

Study Overview

Preclinical testing of implantable devices requires a large animal model that closely mimics human cardiovascular anatomy and physiology.[8] Male calves (n = 23; 80-100 kg) were used to evaluate the Symphony pump. Calves are the established industry standard to test safety, performance, reliability, and efficacy of LVADs.[9]

The study was performed according to Good Laboratory Practices (GLP) guidelines to determine preclinical safety of the pump. Animals were supported acutely (n = 4) or for 1 week (n = 10), 2 weeks (n = 7), or 4 weeks (n = 2) of uninterrupted support with the Symphony pump. Evaluation of the carotid artery near the anastomosis and contralateral carotid artery allowed each animal to be used as its own control.

The Symphony pump was implanted subcutaneously in the neck. An anastomosis was performed between the pump graft and the carotid artery as described. This approach was chosen because of the similar size and structure of the bovine carotid artery and human subclavian artery (approximately 8-10 mm diameter), as well as the similar distance from the anastomosis to the aortic valve in humans and in calves (approximately 8-10 cm).[10]

After completing the planned duration of support, animals were euthanized. Carotid artery proximal to the anastomosis and contralateral carotid artery were harvested. Initially, histologic and molecular analyses were performed on tissues from animals that underwent 1 week of support (n = 8). Interesting histologic changes in this group of animals led to more analyses in additional animals. We evaluated whether observed histologic changes may have resulted from changes in matrix metalloproteinase (MMP) and connective tissue growth factor (CTGF). Quantitative real-time polymerase chain reaction (PCR) for MMP and CTGF gene expressions was performed on carotid arteries from additional animals that underwent 1 week and 2 weeks of support (n = 9). Later, to determine whether architectural changes were associated with abnormal vessel reactivity, isolated arterial ring studies were performed with carotid arteries from animals that underwent 2 weeks and 4 weeks of support (n = 5). In these animals, live carotid artery sections were transferred to physiologic saline solution (PSS) for in vitro isolated ring preparations to determine endothelial-dependent and endothelial-independent vasoreactivity to pharmacologic challenge as described below. In addition to histologic and molecular analyses, carotid artery pressures and flows were measured acutely (n = 4) to document the hemodynamic changes associated with partial support through an end-to-side anastomosis to a peripheral artery.

Surgical Preparation and Hemodynamic Measurements

One day before surgery and continuing for the duration of the study, each animal received a daily oral dose of 75 mg clopidogrel. Six hours after surgery, animals were placed on heparin, which was titrated to maintain an activated clotting time longer than 200 seconds. Warfarin was initiated on postoperative day 3 and titrated to maintain an international normalized ratio (INR) of 2.5 to 3.5. Heparin was discontinued after the INR was at a therapeutic level.

Animals were anesthetized with 3% to 5% isoflurane and prepared for sterile surgery. Permanent fluid-filled catheters were placed in the right jugular vein for intravenous access and in the proximal right carotid artery for arterial blood pressure monitoring. A left fifth intercostal space minithoracotomy was performed to place screw-in epicardial electrocardiographic leads (Medtronic, Inc, Minneapolis, Minn).