We evaluated the effects of steady state flow and perfusion on end-organ function in a long-term calf model. The animal received a continuous-flow total artificial heart (CFTAH) that we created from two axial-flow ventricular assist devices. Pump flow, blood pressure, and other pump parameters were monitored throughout the study, as were arterial blood gas and hematologic values, including neurohormone levels. Some hematologic values were mildly abnormal transiently after surgery but returned to acceptable levels within the first week. During the 90-day study, the calf showed no signs of hemolysis or thrombosis. Its mental function remained normal, as evidenced by the animal’s interest in its surroundings and response to stimuli. End-organ and vasomotor function was not adversely affected by 90 days of steady state flow. This was the first study in which CFTAH support of an animal model was maintained for this duration. ASAIO Journal 2014; 60:15–18.

Key Words: continuous-flow total artificial heart, steady state perfusion, left ventricular assist devices, mechanical cardiac assistance, heart failure

We previously explored the physiologic effects of a continuous-flow total artificial heart (CFTAH) in a calf model over a 7-week period.1 In the current study, we demonstrated that such a device could support a calf for 90 days. By showing that continuous-flow or steady state perfusion can maintain normal physiologic parameters for this length of time in a calf, we gained new insights into the potential effects of long-term CFTAH use in mammals.

Materials and Methods

A 5-month-old, 88 kg, male Corriente cross calf was implanted with a CFTAH. The calf received humane care in compliance with the Principles of Laboratory Animal Care (National Society of Medical Research) and the Guide for the Care and Use of Laboratory Animals (National Institutes of Health Publication no. 85-23, revised 1996). Our Institutional Animal Care and Use Committee approved all the protocols used in this study.

Continuous-Flow Total Artificial Heart

Two customized axial-flow ventricular assist devices (HeartAssist 5, Continuous-Flow Left Ventricular Assist Devices; MicroMed Cardiovascular, Inc., Houston, TX) were specifically configured for integration into a CFTAH (Figure 1). The original curved pump inlet cannula was replaced with a short, straight inlet designed to attach to our custom-fabricated atrial cuffs made of silicone-impregnated Dacron. Fluid-filled pressure lines were directly attached to each atrial cuff to measure the left and right atrial pressure (LAP and RAP, respectively). The outflow portion of each pump consisted of a 12 mm gelatin-impregnated polyester graft (GelWeave; Vascutek, Inchinnan, Scotland) that was externally supported by a plastic casing to prevent kinking. Each pump had a flow probe (Transonic Systems, Inc., Ithaca, NY) on its outflow graft to measure systemic and pulmonary blood flow. In addition, each pump had its own driveline, which was tunneled through the chest wall and was connected to a controller, power supply, and system monitor.

Surgical Technique

Once general anesthesia was induced per our standard protocol,1 a tracheostomy was created as described elsewhere.2,3 A left thoracotomy was performed, and an incision was made in the left side of the neck. The fifth rib was then removed to allow better visualization of the cardiac structures. A fluid-filled pressure line was placed in the left internal mammary artery for arterial pressure (AoP) monitoring throughout the study. The pressure lines and the power cables for the right and left pumps were tunneled so as to exit the skin near the lumbar spine. After heparin (300 IU/kg intravenously [IV]) was systemically administered, arterial cannulas were placed in the left carotid artery and the descending thoracic aorta. Venous lines were placed in the left jugular vein and the inferior vena cava. Cardiopulmonary bypass (CPB) was initiated, and cross-clamps were applied to the origin of the innominate artery and the proximal descending thoracic aorta. The heart was excised by dividing the aorta and the pulmonary arteries just above their respective valves. The left and right atria were incised just cephalad to the atrioventricular (AV) grooves, enabling removal of the corresponding AV valves.

The atrial cuffs were sewn to the atrial remnants with 2.0 polypropylene sutures reinforced with felt strips (Bard Peripheral Vascular, Tempe, AZ). The pump inflow conduits were then inserted into each cuff and secured with zip ties. Once the GelWeave graft had been cut to the appropriate length for each pump, the left pump and right pump outflow grafts were anastomosed to...
data obtained from the left and right atrial cuffs. Obtained from lines placed in the left carotid artery and pressure of the left and right pumps; it was also based on pressure data flows, as measured by flow probes placed on the outflow grafts. The optimal pump speed was based on systemic and pulmonary pressures in determining the appropriate pump speed.

Figure 1. Two customized MicroMed axial-flow ventricular assist devices were specifically configured for integration into a continuous-flow total artificial heart. The pulmonary artery (A) and the ascending aorta (B) are attached, respectively, to the right and left outflow grafts (E, F), which are threaded through the pulmonary and systemic Doppler flow probes (C, D). The attachments of the drivelines to the right and left pumps (H, I) are partially visible, as is the chimney portion (G) of the Dacron and silicone inflow cuff attached to the right atrial remnant. Note that the outflow graft of the pulmonary pump crosses anterior to the outflow from the systemic pump.

As the calf was weaned from CPB, the pump speeds were gradually increased. Determination of pump speeds was based on systemic and pulmonary flows, as measured by flow probes placed on the outflow grafts of the left and right pumps; it was also based on pressure data obtained from lines placed in the left carotid artery and pressure data obtained from the left and right atrial cuffs.

Figure 2. The components of the continuous-flow total artificial heart before implantation. The modified inflow cannulas (A, B) of the right and left pumps, respectively, are inserted through the chimney portions (E, F) of the right and left atrial cuffs. Doppler flow probes (C, D) are placed over the pump outflow grafts (not shown) to measure pulmonary and systemic flow. Drivelines (G, H) to the two pumps are tunneled through the calf’s flank. Two pressure lines (I, J) are brought through the chest wall to allow continuous monitoring of the right and left atrial pressures, which are essential measurements in determining the appropriate pump speed.

Follow-Up Observation and Data Collection

On postoperative day (POD) 40, the heparin infusion was replaced with warfarin (2.5–5.0 mg orally). We had debated about maintaining the animal on heparin throughout the 90 day study because of difficulty in obtaining stable anticoagulation with oral warfarin in some of our previous animals. In this instance, however, anticoagulation with warfarin was not problematic. The dose was adjusted as necessary to maintain the international normalized ratio at 1.5 to 2.0, which is twice the baseline value. Recorded pulmonary and systemic pump data included pump rotational speed, pump power consumption, flow probe data, and pressure data from the LAP, RAP, and AoP lines. Throughout the 90 day study, pressure data were continuously recorded by a 16 channel data acquisition system (Ponemah System, version 5.0; Data Sciences International, St. Paul, MN). Arterial blood gases were evaluated hourly during the recovery period and later as needed throughout the study. Hematologic values, including hemoglobin levels, hematocrit, platelet count, and white blood cell count, were measured at least weekly to assess liver and kidney function and to detect anemia, hemolysis, or infection. Neurohormone levels were also evaluated, including renin, angiotensin-converting enzyme, epinephrine, norepinephrine, atrial natriuretic peptide (ANP), brain natriuretic peptide (BNP), and endothelin-1 levels. Fluid intake and output were monitored daily throughout the study.

During the recovery period, hypertension was treated with isoproterenol, nitroprusside, and nesiritide as discussed above, as well as with dopamine (800 mg/250 ml D5W at 1.25–12.12 μg/kg/min). Captopril (12.5 mg orally) was administered starting on POD 69. The calf was given an analgesic regimen of butorphanol (10 mg IV, 0.05–0.20 mg/kg) and flunixin meglunime.
(150 mg IV; 1.1–2.2 mg/kg) every 4 hours, each drug alternating with the other. A broad-spectrum antibiotic (cefazolin, 2 g slow push IV; 15–30 mg/kg) was administered every 8 hours for the duration of the study. Diuretic agents (furosemide, 100 mg IV, or bumetanide, 5 mg IV) were given as needed to increase urine output. Omeprazole (450 mg orally) and probiotic gel (15 g orally) were administered daily to maintain optimal ruminal flora. In addition, megestrol acetate (400 mg orally) was given three times daily to stimulate and maintain the animal’s appetite. Ranitidine (150 mg orally) and sucralfate (1 g orally) were given to help protect the gastrointestinal tract from irritation. A nitroglycerin infusion was administered when needed at 0.08 to 2.00 μg/kg/min.

Study Termination

At the end of the study, the calf was anesthetized and returned to the operating room. Contrast dye was injected through the left jugular vein, and a fluoroscopic image of the CFTAH was obtained. The calf was then humanely euthanized, and a complete necropsy was performed.

Results

Postoperative Course

The animal recovered from anesthesia uneventfully, was alert within 1 hour, and was able to stand with assistance within 2 hours. During the first week, pump rotational speeds were adjusted to maintain stable atrial pressures within the normal physiologic range. After that week, pump speeds were rarely adjusted. The mean left pump speed was set at 11.9 ± 0.58 kRPM and the mean right pump speed at 9.2 ± 1.03 kRPM. Based on our observations in previous studies with the CFTAH, we elected to oscillate the rotational speed of the right pump as we felt that this provided some element of protection from right pump dysfunction as a result of entrained venous emboli from elsewhere in the body. To that end, we used a specialized controller provided by MicroMed that alternated the speed from 10.6 to 7.6 kRPM, using a square-wave modulation with a frequency of 0.5 Hz. In previous studies, this produced a sine-wave appearance in the pulmonary artery pressure (PAP) tracing. However, no PAP line was used in this study.

Both pulmonary and systemic blood flows remained steady (left pump flow, 9.92 ± 2.37 L/min; right pump flow, 10.7 ± 1.13 L/min) (Figure 3). On POD 2, the HeartAttendant (MicroMed Cardiovascular), which measured the flow, was recalibrated because we discovered that the flow values recorded for the first 2 days were 25% lower than the actual values. After being recalibrated, the system measured the blood flows accurately.

In the early postoperative period, the LAP fluctuated depending on the position of the animal but eventually stabilized. During the first 60 days of the study, the calf had an AoP of 101.9 ± 10.4 mm Hg, an LAP of 14.4 ± 6.3 mm Hg, and an RAP of 12.9 ± 4.7 mm Hg (Figure 4). After POD 60, neo-intimal obstruction of the openings of the left and right atrial lines made LAP and RAP readings unreliable. Fluid levels were closely monitored to provide fluid balance; when necessary, water was rationed, and diuretic agents were given.

The calf’s respiratory rate was slightly elevated at 25 to 30 breaths per minute throughout the study, but the animal never showed signs of respiratory distress. On POD 5, the calf was successfully weaned from ventilatory support, and the tracheostomy tube was removed. A nasal oxygen cannula was placed, and oxygen administration was started at 5 L/min. The tracheal stoma was loosely covered with sterile gauze. The nasal cannula remained in place until POD 14, when it was removed for the duration of the study. Starting on POD 35, the calf produced an occasional dry cough. Although it did not worsen or cause any distress to the animal, the cough persisted until termination of the study.

Hematuria was present on POD 2 but quickly resolved without intervention. Blood urea nitrogen and creatinine levels...
increased immediately after surgery but quickly returned to normal (7.7 ± 2.1 and 0.7 ± 0.2 mg/dl, respectively).

Throughout the study, the animal responded appropriately to his caretakers and stood intermittently throughout the day. He exhibited a healthy appetite, an affinity for molasses, and normal bowel and bladder function. There was no clinical or laboratory evidence of infection or gastrointestinal hemorrhage.

Hepatic Hematology

Liver function remained within normal parameters, as indicated by a serum glutamic oxaloacetic transaminase level of 285.3 ± 393.5 IU/L and a serum glutamate pyruvate transaminase level of 23.5 ± 21.3 IU/L. Destruction of red blood cells was not indicated (average plasma-free hemoglobin level, 3.02 ± 1.73 mg/dl). Whereas lactate dehydrogenase values remained elevated throughout most of the study (mean, 1260 ± 518 U/L), they steadily returned to baseline after POD 42.

Catecholamine levels remained at baseline (adrenaline, 1.15 pg/ml; noradrenaline, 3.8 pg/ml) until POD 7, when they increased to 6.3 and 76.9 pg/ml, respectively. The levels peaked on POD 14, then returned to near baseline by POD 28 and remained slightly elevated until the end of the study (adrenaline, 1.9 ± 2.0 pg/ml; noradrenaline, 19.8 ± 23.1 pg/ml). Natriuretic peptide levels showed a pattern similar to that of the catecholamines, except that the peptide levels decreased rather than increased. The natriuretic peptide levels decreased to below baseline (mean BNP level, 66.1 ± 12.0 pg/ml; ANP level, 43.5 ± 12.7 pg/ml). Endothelin-1 levels were also measured throughout the study and showed a mean of 33.91 ± 8.76 pg/ml. Angiotensin-converting enzyme and renin values were also measured (mean, 26.8 ± 5.5 U/L and 1.42 ± 0.19 pg/ml, respectively).

Fluoroscopy, performed just before study termination, revealed brisk flow through both sides of the CFTAH without evidence of kinking, twisting, or other impediment to flow. The necropsy further confirmed the acceptable position of both pumps, with widely patent inflow and outflow geometry. There was no evidence of hemomedastinum or hemothorax. The trachea had an approximately 50% stenosis at the site of the tracheotomy.

At necropsy, the animal weighed 90.9 kg. The CFTAH site was well healed, and all components were acceptably configured. The inner aspects of the two atrial Dacron cuffs were lined with a shiny flat layer of neoendothelium that had grown over the ostia of the LAP and RAP lines. Both pump inlets were clean and free of thrombus or other debris. All anastomoses were covered with neoendothelium as well, and the anastomoses with the atria and great vessels were unremarkable.

The lungs were somewhat stuck to the device with adhesion, but the parenchyma was completely normal, without evidence of microemboli, edema, or other adverse parenchymal processes. Similarly, the liver, kidneys, stomach, small and large intestines, adrenal glands, and pancreas were grossly normal. Histologic evaluation of the liver and kidneys revealed normal parenchyma without interstitial edema, inflammation, or infarcts. No arteriovenous malformations were identified in the gastrointestinal tract.

Discussion

In this study, we demonstrated that a calf implanted with a CFTAH could function normally with steady state blood flow and pressure for 90 days. To date, this is the only calf study done with a CFTAH to have a 90-day duration. The advantages of this device include its mechanical simplicity, small size, and improved power efficiency. By eliminating the need for inlet and outlet valves, as well as flexible membranes and bladders, devices like the CFTAH will likely prove more reliable and durable than the pulsatile, volume-displacement devices that preceded them. Without the volume-displacement mechanism, there is no need for compliance chambers or external venting. In addition, the CFTAH’s mechanical simplicity results in better power efficiency. Moreover, a theoretical advantage of the CFTAH derives from the pressure sensitivity that all rotary pumps exhibit. At a fixed rotational speed, flow will increase and decrease with rising and falling pump inlet pressures, respectively. This may allow a total artificial heart (TAH) constructed from two rotary pumps to maintain some degree of autonomous balance between the systemic and the pulmonary circulations under various physiologic conditions. In contrast, the challenges associated with achieving such a balance in pulsatile, volume-displacement TAHs are significant. Additional research is ongoing to determine the degree to which this autonomous balance between two rotary blood pumps can be leveraged in a CFTAH.

Despite these numerous potential advantages, little is known about the long-term effects of steady state, pulseless perfusion. In our calf model, the presence of normal end-organ histology and function after 90 days of support is further evidence that mammalian physiology can be supported by a CFTAH.

References