A fetal goat model of cardiopulmonary bypass with cardioplegic arrest and hemodynamic assessment

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Objective: Increasing evidence shows that some cardiac defects may benefit from fetal interventions, including fetal cardiac surgery. We attempted to develop an in vivo animal model of fetal cardiopulmonary bypass with cardioplegic arrest.

Methods: Operations were performed on 14 pregnant goats. The extracorporeal circulation circuit consisted of a centrifugal pump, silicone tubings with an inner diameter of 6 mm, a roller pump, and a reservoir. The placenta was the sole oxygenator. Cardiopulmonary bypass was maintained at a mean flow rate of 344 ± 68 mL/kg/min, including 30 minutes of cardiac arrest and 15 minutes of reperfusion. Mean arterial blood pressure and heart rate were monitored. Arterial blood samples were analyzed. The pulse index and resistance index of the fetal umbilical artery were monitored.

Results: Experiments were completed in 11 cases (79%), with the fetuses weighing 0.65 to 1.8 kg. Fetal mean arterial blood pressure and heart rate remained stable throughout the experiments. A decrease in partial pressure of oxygen with concomitant increase in carbon dioxide partial pressure was noted, but trends were relatively stable. Metabolic acidosis was recognized during and after cardiac bypass. The pulse index and resistance index of the umbilical artery increased significantly after 2 hours off bypass.

Conclusions: We confirmed the technical feasibility of establishing an in vivo model of fetal cardiac bypass with cardioplegic arrest. This fetal goat model provides reproducible data and is suitable to study clinically relevant problems related to fetal cardiopulmonary bypass, myocardial protection, and hemodynamics. (J Thorac Cardiovasc Surg 2011;142:1562-6)

Some severe fetal cardiac disease is progressive and involves secondary damage to the rapidly growing pulmonary bed and ventricles,[1-3] affecting postnatal treatment options and prognosis, and may require palliative rather than definitive surgical procedures. Some fetuses with cardiac abnormalities may also develop cardiac failure and fetal hydrops, leading to a higher rate of intrauterine fetal demise or early neonatal death. Theoretically, in utero therapy might improve the outcome. There is increasing evidence that some of these defects may benefit from fetal interventions, including fetal cardiac surgery.[4-6] If the primary lesion is corrected in the fetal period, secondary damages that might be irreversible and result in single ventricle physiology can be avoided.

There is no safe and effective method to gain access to perform open surgery in the fetus. A motionless and bloodless field is absolutely necessary to permit the meticulous intracardiac repair. Furthermore, little is known about the pathophysiologic effect of cardiopulmonary bypass (CPB) with cardioplegic arrest on the developing fetus.

The feasibility of safely supporting the fetus during bypass has been demonstrated, when placental dysfunction was prevented by overcoming the detrimental response of the placenta to bypass.[7-9] Studies on fetal myocardial protection have been carried out, but research is limited and principally relies on isolated heart models.[10,11] The requisite techniques to safely arrest and protect the fetal myocardium during cardiac interventions need to be developed.

By using existing beating-heart models of CPB in fetal goats, we developed a clinically relevant model of performing open surgery that allows administration of antegrade cardioplegia and crossclamping in the fetus. We also assessed the hemodynamics of fetal-placental circulation after fetal cardiac bypass. This novel model enables us to study the various pathophysiologic effects of CPB with cardioplegic arrest on hemodynamics and myocardial protection in vivo. Defining these mechanisms has significant

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Abbreviations and Acronyms

CPB = cardiopulmonary bypass
EDV = end-diastolic velocity
HR = heart rate
MAP = mean arterial pressure
PI = pulse index
RI = resistance index
SPV = systolic peak velocity
TAMAX = time-averaged maximum velocity

implications for the successful translation of fetal cardiac surgery in vivo.

MATERIALS AND METHODS

Animals

The study protocol was approved by the Animal Research Committee of Guangdong Provincial People's Hospital. All animals were handled in accordance with the Guidelines of Guangdong Provincial People's Hospital for Laboratory Animal Studies. Operations were performed on singleton and twin fetuses carried by mixed-breed, time-dated pregnant goats (110–130 days, term = 150 days).

Anesthesia and Monitoring

Pregnant goats (n = 14) were anesthetized by intramuscular injection of 10 to 20 mg/kg ketamine (Gutian Pharmacy Co, Fujian, China) and then intubated endotracheally. Arterial oxygen saturation was maintained at 100%, and arterial carbon dioxide tension was maintained at approximately 30 mm Hg. Catheters were placed in the goats' femoral artery and jugular vein for measurement of blood gases and delivery of intravenous fluids, respectively. Maternal arterial pressure and heart rate (HR) were monitored. Anesthesia was maintained with 5 mg/kg intravenous fentanyl per hour (Renfu Pharmacy Co, Yichang, China) and 0.1 mg/kg intravenous vecuronium per hour (Xianjun Pharmacy Co, Zhejiang, China).

Operation

The uterus was exposed through a midline laparotomy. The number and orientation of the fetus were determined, and 50 mg fentanyl and 0.2 mg vecuronium were administered intramuscularly to the selected fetus. A hysterotomy was performed to expose a forelimb in which the axillary artery was cannulated to monitor fetal arterial blood pressure and HR. Anesthesia was maintained with ketamine 5 mg/h via the axillary vein.

Fetal Cardiac Pulmonary Bypass

The extracorporeal circulation circuit consisted of a nonpulsatile centrifugal pump (BP50; Medtronic, Inc, Minneapolis, Minn), silicone tubing with an inner diameter of 6 mm for the arterial and venous line (length 90 cm), another silicone tubing coupled to a roller pump (Stockert Instrumente GmbH, Munich, Germany), and cardiotomy reservoir with filter via a Y connector into the venous line of the extracorporeal circuit to confirm the left heart venting (Figure 1). The placenta was the sole oxygenator. Approximately 50 mL of the heparinized maternal blood containing 10 mg heparin, 50 mg methylprednisolone, and 5 mL 5% sodium bicarbonate was used to prime the pump circuit.

A midline fetal sternotomy was performed, and the pericardium was opened. Heparin (10 mg; Shanghai No 1 Biochemistry Pharmacy Co, Shanghai, China) was administered intravenously before cannulation. A fetal cardiac bypass was instituted after cannulation of the main pulmonary artery using an 8F DLP arterial cannula (Medtronic, Inc) and the right atrium using a 16F straight venous cannula (Medtronic, Inc). After ensuring the absence of any air bubbles within the circuitry, CPB was initiated. Because of the need for high flows when the placenta is incorporated into the circulation for oxygenation, pump flows were maintained at the maximum achievable flow in each animal (the mean value for a stable pump flow is 344 ± 68 mL/kg/min). Fetal body warming was ensured with an infrared lamp and gauze soaked in warm saline. Fetal body temperature was satisfactorily maintained during and after bypass, and the maximum gradient between pre-bypass and bypass temperatures did not exceed 3°C.

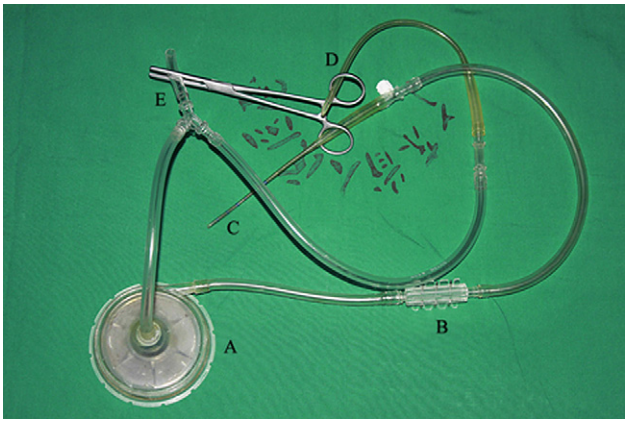

FIGURE 1. Extracorporeal circuit consisted of centrifugal pump (*A*), flow probe (*B*), arterial cannula (*C*), venous cannula (*D*), and silicone tubing with an inner diameter of 6 mm for the arterial and venous lines. Another silicone tubing coupled to a roller pump and reservoir with filter was inserted via a Y connector (*E*) into the venous line of the extracorporeal -circuit for left heart venting (not shown).

Fetal Cardiac Arrest

Immediately on reaching stable target bypass flow rates (2–3 minutes), the aorta was crossclamped and cardioplegic arrest was induced by cold modified St Thomas cardioplegia (4°C) without calcium solution to a total of 25 mL/kg estimated fetal body weight. Topical cooling using ice slush was used. A left heart venting catheter through the left auricle coupled to a reservoir and roller head pump was used for decompression of the fetal heart. After 30 minutes of myocardial ischemia, the aortic crossclamp was released and the heart was reperfused. After clamp removal, the fetal heart spontaneously resumed normal sinus rhythm. Bypass continued for 15 minutes before the animals were weaned from CPB. Heparin was not antagonized routinely. After cessation of CPB, the animals were decannulated. The fetuses were then monitored for 2 hours. After the completion of study, the fetuses were killed for autopsy and measurement of fetal weight.

Ultrasonographic Measurements

For ultrasonographic examinations, the ultrasound system GE LOGIQ book XP (GE, Wuxi, China) equipped with a (4–10 MHz) linear transducer probe for B-mode and color-coded and pulsed Doppler measurements was used. The umbilical artery was first visualized by means of color-coded Doppler. Then a pulsed Doppler flow velocity waveforms sample volume was placed exactly in the center of the color-coded blood flow, and the waveforms of at least 3 consecutive cardiac cycles were recorded; thus, the measurements of 3 different waveforms for each vessel were taken. The systolic peak velocity (SPV), end-diastolic velocity (EDV), time-averaged maximum velocity (TAMAX), pulse index (PI), and resistance index (RI) were automatically calculated for each waveform (PI = SPV – EDV/TAMAX, RI = SPV – EDV/SPV).