Part 1
Placental anastomoses
Chapter 3

Assessment of feto-fetal transfusion flow through placental arterio-venous anastomoses in a unique case of twin-to-twin transfusion syndrome

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Abstract

Objective: In vivo measurements of blood flow through arterio-venous anastomoses in monochorionic twin placentas have recently been attempted with Doppler ultrasound, but the accuracy is questionable. We present a new method to determine the arterio-venous anastomotic blood flow.

Methods: Detailed description of a unique twin-to-twin transfusion syndrome case treated with fetoscopic laser surgery and subsequently with an intrauterine blood transfusion. Prospective measurements of decreasing hemoglobin levels between the intrauterine transfusion and birth allowed us to assess the net blood flow through the residual anastomoses.

Results: A case of twin-to-twin transfusion syndrome was treated with fetoscopic laser surgery at 27 weeks’ gestation. The ex-recipient subsequently became severely anemic and was treated with an intrauterine blood transfusion at 29 weeks’ gestation. After birth, a placental injection study identified five residual unidirectional arterio-venous anastomoses from the ex-recipient to the ex-donor without arterio-venous anastomoses in the opposite direction. The net feto-fetal blood flow through the five residual arterio-venous anastomoses was determined to be 27.9 mL/24h.

Conclusions: We found the blood flow across a single arterio-venous anastomosis at 29 weeks’ gestation to be 5.6 mL/24h, much lower than previously measured with Doppler ultrasound. This finding may also explain the inaccuracy of Doppler flow measurements, as such low flow velocities cannot possibly be detected with current Doppler techniques.
Introduction

Twin-to-twin transfusion syndrome (TTTS) is the most common complication in monochorionic twin pregnancies and is associated with high perinatal morbidity and mortality rates\textsuperscript{186}. TTTS is attributed to imbalanced inter-twin blood transfusion through placental anastomoses, leading to hypovolemia and oligohydramnios in the donor twin and hypervolemia and polyhydramnios in the recipient twin. Despite major advances in this field, the exact pathogenesis of TTTS remains incompletely understood\textsuperscript{186}. Lack of a suitable experimental animal model has hampered further investigation on the development of TTTS. Computer modeling for TTTS has helped to elucidate several pathophysiological mechanisms\textsuperscript{187}. In vivo assessment of blood flow through an arterio-venous (AV) anastomosis would help to further unravel the complex pathophysiology of TTTS. Recently, several attempts have been made to measure anastomotic blood flow using Doppler ultrasound\textsuperscript{188,189}. However, the results of these studies are highly discordant and lead to a fierce debate on the “correct” blood flow down an AV anastomosis\textsuperscript{190}. In a case of TTTS treated with fetoscopic laser surgery, the ex-recipient became severely anemic due to residual AV anastomoses and required an intrauterine blood transfusion 48 hours before emergency delivery. This offered the opportunity to quantify the inter-twin blood flow through the unidirectional AV anastomoses with a novel method by analyzing the decrease in hemoglobin (Hb) concentrations at different time points.

Case presentation

A 31-year-old gravida 1 para 0 was referred to our institution at 27 + 0 weeks’ gestation with signs of TTTS (Quintero stage II) and was treated with fetoscopic laser coagulation of vascular anastomoses. Serial ultrasound examinations after laser surgery showed normalization of amniotic fluid in both sacs and normal bladder filling in both fetuses. However, Doppler measurement of the middle cerebral artery peak systolic velocity (MCA-PSV) in the ex-recipient gradually increased up to 120 cm/sec (> 2 Multiples of Median)\textsuperscript{191} suggesting the presence of severe fetal
anemia. An intrauterine blood transfusion was performed at 29 + 3 weeks’ gestation. A total volume of 53 ml of blood with an Hb concentration of 27.2 g/dL was transfused during 30 minutes. Hb concentrations in the ex-recipient before and after the transfusion were 3.0 g/dL and 11.2 g/dL, respectively. However, within 48 hours after the intrauterine transfusion, MCA-PSV Doppler studies showed again signs of severe fetal anemia and cardiotocography demonstrated a sinusoidal pattern. A caesarean section was performed 48 hours after the intrauterine transfusion. The first-born twin (ex-donor) was plethoric and weighed 1210 g. The second-born twin (ex-recipient) was pale and weighed 1527 g. Hb concentrations in twin 1 and twin 2 were 24.3 g/dL and 7.7 g/dL, respectively. Blood pressures at birth in twin 1 and twin 2 were 53/32 mmHg and 47/27 mmHg, respectively.

Macroscopic examination of the placenta revealed a fetoscopic hole in the membranes of twin 2, confirming that the second-born twin was the ex-recipient. Injection with color-latex showed four small residual unidirectional AV anastomoses from the ex-recipient to the ex-donor with a diameter of about 0.5 mm (Figure 1). A residual arterio-arterial (AA) anastomosis was also detected. However, this anastomosis was initially not detected after dye injection, but became patent only after injection with increased pressure and forced manual compression of the dye. Placental casting showed additionally a shared cotyledon underneath the placental surface connecting an ex-recipient chorionic artery with an ex-donor chorionic vein, hence a fifth unidirectional AV anastomosis.

**Calculation of blood transfusion**

As a result of the intrauterine transfusion, the ex-recipient received 53 mL blood of 27.2 g/dL, which equals 14.4 g of Hb, resulting in an increase in Hb concentration from 3.0 g/dL to 11.2 g/dL. We used the method described by Hoogeveen et al to calculate the dilution of fetal Hb, with a post-transfusion feto-placental blood volume supposed to return to the pre-transfusion volume, i.e. 149 mL. The AV transfusion rate can then be calculated from the known amount of Hb transfused from ex-recipient to ex-donor, assuming that the AV flow and the ex-recipient’s blood volume
remain unaltered during the 48 hours following intrauterine transfusion. This is by standard physics as follows:

The change of the ex-recipient’s Hb concentration, \( d[Hb] \), at time \( t \) (at \( t = 0 \), and birth, at \( t = 48 \) hours), in an infinitesimal short period of time, \( dt \), equals the amount of ex-recipient Hb concentration \( [Hb] \) transfused to the other twin by the AV in time period \( dt \). The ex-recipient’s decrease in Hb concentration is the grams of Hb transfused, i.e. \( [Hb] \) times \( AV_{flow} \) times \( dt \), divided by the ex-recipient’s blood volume, or

FIGURE 1 Monochorionic placenta after dye-injection (blue or green for arteries and orange or yellow for veins). The white arrows indicate the residual AV anastomoses from the ex-recipient (left side) to the ex-donor (right side). Details of the four residual anastomoses between arteries of the ex-recipient (blue) and veins of the ex-donor (orange) are shown in the top-right and bottom-right corner. The green stars show the obliterated anastomoses after laser coagulation. The white star indicates the obliterated but during dye-injection fiercely re-opened arterio-arterial anastomosis.
Twin-to-twin transfusion syndrome: from placental anastomoses to long-term outcome

\[ d[Hb]_t = - \frac{AV_{\text{flow}}}{\text{BloodVol}} \cdot [Hb]_t \cdot dt \]  

(1)

The minus sign just implies that the increase in \([Hb]_t\) which is \(d[Hb]_t\) is negative. This equation actually represents the following standard differential equation (i.e. dividing by \(dt\))

\[ \frac{d[Hb]_t}{dt} = - \frac{AV_{\text{flow}}}{\text{BloodVol}} \cdot [Hb]_t \]  

(2)

with solution

\[ [Hb]_t = [Hb]_{t=0} \exp \left( - \frac{AV_{\text{flow}}}{\text{BloodVol}} \cdot t \right) \]  

(3)

Dividing by \([Hb]_{t=0}\), taking the natural logarithm of both sides, and solving for the \(AV_{\text{flow}}\) (in mL/h) gives

\[ AV_{\text{flow}} = \frac{\text{BloodVol}}{48} \cdot \ln \left( \frac{[Hb]_{t=48}}{[Hb]_{t=0}} \right) \]  

(4)

When the begin-Hb and end-Hb concentrations, i.e. at the moment of transfusion \([Hb]_{t=0}\) and at birth \([Hb]_{t=48}\), and the blood volume are substituted in equation 4, i.e. \([Hb]_{t=0} = 11.2, [Hb]_{t=48} = 7.7, \text{BloodVol} = 1.49 \, \text{dL}\), the solution of the combined AV flow yields

\[ AV_{\text{flow}} = 27.9 \, \text{mL/24h} \]  

(5)

Assuming that the blood flow through each of the five AV anastomoses was equal, the net transfusion flow through a single AV anastomosis was 5.6 mL/24h.
Comment

This study reports a unique case of TTTS that allowed us to assess the anastomotic blood flow through the placental anastomoses. Unfortunately, we did not measure the percentage of transfused adult red cells or adult Hb in the ex-donor after birth. This would have given us yet another method to calculate the feto-fetal transfusion during the last 48 hours before birth and thus to confirm the above calculations. Through the calculations in this paper, we found the blood flow across a single AV anastomosis at 29 weeks to be 5.6 mL/24 h.

This flow is in amazing agreement with the flow through an arterio-arterial anastomosis at 28 weeks (7.6 ± 4.0 x 10^{-8} L/s, which equals 6.6 ± 4.2 mL/24h)\textsuperscript{188,189}, which approximately equals the oppositely directed AV transfusion during steady state\textsuperscript{100}. In contrast, Nakata \textit{et al} measured in vivo AV blood flows of up to 25 mL/min (36 L/24 h) using Doppler flow during a fetoscopic procedure\textsuperscript{189}. Feto-fetal blood flow of this magnitude, however, would lead to fatal acute hemorrhagic shock in the donor fetus within a few minutes and is thus physiologically implausible\textsuperscript{190}. Previously, using micro-bubble contrast angiography, Denbow et al. measured an inter-twin transit time of 65 s\textsuperscript{193} in a placenta that included an anastomotic pattern of two AVs and one AA (Dr Mark L Denbow, personal communication). In our case of unidirectional AVs, assuming a 4 cm anastomotic length and a diameter of 0.5 mm, the transit time would be about 120 s. Interestingly, our TTTS mathematical model predicts AV flows that cause severe, i.e. Quintero stage IV, TTTS of about 10 to 15 mL/24 h\textsuperscript{187}, in excellent agreement with the value reported in this paper.

In conclusion, by direct measurement of Hb concentration decrease in a unique case of TTTS with known anastomotic pattern, we were able to estimate the feto-fetal transfusion flow through placental arterio-venous anastomoses. This information can now be used to further develop improved computer models and to help elucidate the pathophysiology of TTTS.