Pulmonary trunk, ductus arteriosus and pulmonary arterial phasic blood flow interactions during systole and diastole in the fetus

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Running head: Fetal pulmonary and ductal blood flow interactions

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Abstract:

Although the distribution of average fetal pulmonary trunk (PT) blood flow favors the ductus arteriosus (DA) over the lungs, the phasic aspects of this distribution during systole and diastole are not well understood. Accordingly, flow profile and wave intensity (WI) analyses were performed at baseline and during brief flow increases accompanying an extrasystole (ES) in ten anesthetized late-gestation fetal sheep instrumented with PT, DA and left pulmonary artery (PA) micromanometer catheters and transit-time flow probes. At baseline, 83% of mean PT flow crossed the DA and 17% entered the lungs. However, early-systolic flow associated with a forward-running compression wave (FCW_{in}) was higher in the PA, and predominant DA flow only emerged in mid-systole when a large PA backward-running compression wave (BCW_{ms}), which reduced PA flow, was transmitted into the DA as a forward-running compression wave (FCW_{ms}) that increased flow. Subsequent proto-diastolic forward DA flow occurring during pulmonary valve closure was associated with substantial retrograde PA flow, but insignificant PT flow. Conversely, forward DA flow in the remainder of diastole occurred with forward PT, but near-zero PA flow. These flow and WI patterns, in conjunction with the results of mathematical modeling, suggest that 1) fetal PT flow preferentially passes into the PA during early-systole due to a lower PA-than-DA characteristic impedance, while DA flow predominates in mid- and late-systole due to flow effects arising from the PA BCW_{ms}, 2) forward DA flow is mainly sustained by reversal of PA flow in proto-diastole, but discharge of a more central reservoir in diastole.

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The junction of the pulmonary trunk (PT), ductus arteriosus (DA) and major pulmonary arteries plays a pivotal role in fetal circulatory physiology. Anatomically, the PT arises from the right ventricle (RV) and is contiguous distally with the DA, while a main pulmonary artery (PA) or its left and right major branches arise from the PT-DA junction (13). Although DA and PA cross-sectional dimensions are similar (15), flow measurements using radio-labeled microspheres in fetal lambs (3, 46, 47) or Doppler-echocardiography in human fetuses (20, 30, 31) indicate that only 10-40% of net PT flow (i.e. RV output) is distributed to the lungs via the PA, with the remainder passing to the placenta and lower fetal body via the DA and descending aorta (10, 33, 35, 39). The widely-accepted explanation for this flow inequality between the DA and PA is that, due to a high fetal pulmonary vascular resistance, PT flow is preferentially shunted across the widely patent DA to the lower resistance fetal body and placental circulations (12, 34, 35).

Two sets of findings suggest, however, that phasic PT, DA and PA interactions are not only more complex than suggested by average flow data, but also vary between systole and diastole. Thus, systolic flow profiles in the PT and PA differ strikingly from the DA, with the former peaking in early systole and falling abruptly in mid-systole, particularly in the main and major branch PA (7, 11, 33, 35, 43, 44), but the DA profile rising in early-systole and then increasing further to a delayed mid-systolic peak (7, 37, 42). A mechanism for these differing profiles has recently been defined using wave intensity (WI) analysis, a time-domain approach for characterizing the forward- and backward-running energy waves that accompany phasic changes in pressure (P) and flow/velocity waveforms (4, 25). Whereas the early flow rise in the PT, major PA and DA coincided with an initial systolic forward-running compression wave (FCW_s) generated by RV impulsive contraction (42), contrasting mid-systolic PT/PA and DA flow patterns were related to the divergent fates of an extremely large PA backward-running compression wave (BCW_ms) that arose from within the pulmonary microvasculature in response to the FCW_s. Specifically, this PA BCW_ms was partially transmitted into the PT as a BCW_ms that reduced blood velocity (43, 44), but into the DA as a mid-systolic forward-running compression wave (FCW_ms) that increased velocity (42). In contrast to systole, flow around the time of pulmonary valve closure (i.e. proto-diastole) and in diastole may be positive in the DA (7, 15, 37) but negative in major PA (7, 32, 37), with
these flow patterns interpreted as evidence that right-to-left DA shunting in this part of the cardiac cycle is mainly related to a reversal of PA flow (7, 34, 35, 37).

Given the apparent diversity of fetal PT, DA and major PA flow interactions, three complementary approaches are likely to facilitate their elucidation. The first is direct comparison of phasic blood flow changes between these sites throughout the cardiac cycle, an evaluation which is yet to undertaken. The second is beat-by-beat analysis of systolic inter-relationships between adjacent vascular segments (43) via use of a ventricular extrasystole (ES) to produce transient post-extrasystolic rises in cardiac contractility and stroke volume (6). Current data suggest that the PA BCW\textsubscript{ms} and its velocity effects are transmitted in a highly linear manner into the PT after an ES (43). However, it is unknown whether systolic transmission of PT waves into the DA and PA, and the PA BCW\textsubscript{ms} into the DA, exhibit similar relationships. The third approach is to characterize flow dynamics using a mathematical model, which permits independent assessment of factors potentially involved in hemodynamic interactions (22, 23). However, no previous model has specifically focused on the PT, DA and major PA region in the fetus.

This study therefore had three main aims. The first was to directly compare PT, DA and PA blood flow profiles within late-gestation fetal lambs in both systole and diastole. The second was to evaluate PT-PA-DA systolic interactions in the same experimental preparation, by examining WI inter-relationships and their associated flow effects at steady-state and after an ES. The third was to use mathematical modeling of PT-DA-PA dynamics to clarify the specific physiological mechanisms contributing to experimental WI and flow patterns.

Methods

Experiments were approved by the institutional Animal Ethics Committee and conformed to guidelines of the National Health and Medical Council of Australia.

\textit{Surgical preparation} The preparation was similar to that previously described (42-45). Briefly, 10 pregnant Border-Leicester cross ewes were anesthetised at a gestation of 139 ± 1 days (mean ± SD, term = 147 days) with intramuscular ketamine 5 mg/kg and xylazine 0.1 mg/kg, followed by inhalation of isoflurane (5%). After tracheal intubation, anesthesia was maintained with 2-3% isoflurane and nitrous oxide (10-20%) in oxygen-enriched air delivered...
via a volume-controlled ventilator (900C Servo, Siemens-Elema, Solna, Sweden), supplemented by intravenous infusion of ketamine (1-1.5 mg/kg/hr), midazolam (0.1-0.15 mg/kg/hr) and fentanyl (2-2.5 mg/kg/hr). Oxygen saturation was monitored continuously with a pulse-oximetry sensor (Oximas Dura-Y, Tyco Healthcare, Pleasanton, CA) applied to the ear. The right common carotid artery was cannulated for monitoring of blood pressure (90308 Multiparameter Monitor, Spacelabs. Medical, Redmond, Wash) and sampling for blood gas analysis (ABL 620, Radiometer, Copenhagen, Denmark), with ventilation adjusted to maintain arterial O₂ tension at ∼120 mmHg and arterial CO₂ tension at ∼40 mmHg.

After a midline laparotomy, the fetal head was exteriorized through a hysterotomy and placed into a saline-filled glove to prevent any loss of lung liquid. Cannulae were inserted into the left external jugular vein for fluid administration, and into the ascending aorta via the left common carotid artery for pressure monitoring and blood sampling. The left forelimb and upper thorax were exteriorized, a thoracotomy was performed in the 3rd left interspace, the pericardium incised and major vessels carefully dissected for placement of transit-time flow probes (Transonic Systems, Ithaca, NY) around the PT (10-14 mm ‘A series’), left PA (4-6 mm ‘S series’) and DA (8-10 mm ‘A series’). Polyvinyl tubing snares were placed around the DA and distal to the flow probe in 6 fetuses, and around the main PA in 4 fetuses. A catheter was inserted through a purse-string suture into the PT to measure pressure. Through separate purse string sutures, one 2.5F micromanometer catheter (Millar Instruments, Houston, TX) was inserted into the distal part of the PT and passed into the DA, a second was placed into the PT immediately distal to the PT flow probe, while a third was inserted into the PT close to the base of the main PA, and its tip advanced into the origin of the left PA (Fig. 1).

**Experimental protocol** Following completion of surgery, the thoracotomy was left open to prevent any distortion or compression of thoracic contents and hemodynamics were allowed to stabilize for 10-15 min. After withdrawal of an aortic blood sample for gas analysis and recording of baseline hemodynamics, physiological data were recorded in all fetuses during induction of an ES by application of a brisk, light tap to the RV infundibular region with forceps (43). This perturbation was repeated 1-3 times after hemodynamics had
returned to baseline, so that WI analyses could be performed in duplicate. At the end of the study, animals were killed with a pentobarbitone sodium overdose (100 mg/kg).

*Physiological data* Aortic and PT blood pressures were measured via the fluid-filled catheters with a transducer referenced to atmospheric pressure at the level of the left atrium and calibrated against a water manometer before each study. Signals from fluid-filled or micromanometer catheters and flow probes were digitized at a sampling rate of 1 kHz using programmable acquisition and analysis software (Spike2, Cambridge Electronic Design, Cambridge, UK). No data filtering was employed, apart from a 48 Hz low-pass filter at the time of analysis to remove electrical interference from signals.

To calibrate high-fidelity pressure signals, mean PT micromanometer pressure was first matched to the mean pressure of the fluid-filled PT catheter. The late-diastolic portions of DA and left PA micromanometer waveforms were then matched to the corresponding segment of the PT micromanometer pressure. A zero-offset calibration of all flow probes was performed prior to each experiment using a water bath. To check that offset calibrations of DA and left PA probes were maintained *in situ*, flow data were recorded while the DA or main PA were briefly (2-3 sec) and completely occluded by tightening of the snares around these vessels. During these occlusions, neither DA (0.02 ± 0.03 L/min, \( P = 0.2, n=6 \)) nor left PA flow (0.001 ± 0.009 L/min, \( P = 0.8, n=4 \)) differed from zero. To ensure that PT, PA and DA flows were internally consistent, RV output was derived in each fetus as the sum of steady-state mean DA flow and the combined left and right PA flow, calculated as the product of measured left PA flow and the total-to-left lung weight ratio (2.55 ± 0.14). The measured PT flow was then matched to the calculated RV output using a scaling factor (1.10 ± 0.17).

All subsequent hemodynamic and WI analyses were performed using this adjusted PT flow.

Blood flow measurements obtained from the PT, PA and DA comprised 1) peak early-systolic flow, 2) peak mid-systolic flow, 3) mean flow, 4) proto-diastolic flow and 5) average diastolic flow. Proto-diastolic flow comprised the average of the prominent negative peak evident in the PA flow profile in the period spanning the dicrotic notch of the PA pressure profile (and thus pulmonary valve closure), with PT and DA flows measured over the same interval. Average PT, PA and DA diastolic flows were measured from a point immediately
after the peak which followed the dicrotic notch, to the start of the ensuing pressure/flow systolic upstroke (Fig. 3A&B). In order to permit a direct comparison with PT and DA flows, reported PA flows refer to the calculated combined left and right PA flow values.

Wave intensity analysis  PT, DA and left PA blood flows were converted to velocity ($U$) using cross-sectional area derived from a caliper measurement of vessel diameter (42). For steady-state WI analysis, an ensemble average of $P$ and $U$ signals was generated from a median of 35 beats (range 25-60). Beat-by-beat WI analysis was performed in the two beats preceding the ES and in seven consecutive beats that followed the ES, an interval which spanned the return of post-ES variables to baseline levels (43). When present, the ES itself was not analyzed, as it varied considerably in magnitude and timing. Two sets of beat-by-beat analyses were performed in all fetuses, and comprised either runs with and without a distinct ES beat ($n = 6$) or duplicate runs with no distinct ES ($n = 4$). In all analyses, the rates of change of PT, left PA and DA blood pressure ($dP/dt$) and velocity ($dU/dt$), and the product of these differentials (i.e. “time-corrected” net WI), were calculated (26, 42-45).

Due to the extent of temporal overlapping of waves in central arteries of the fetus, accurate quantitation of wave magnitude requires separation of WI into forward and backward components (42-44). To perform this separation, wave speed ($c$) was obtained with the P-$U$ loop method using the relation $\rho c = dP/dU$ (17), where $\rho$ is blood density (assumed as 1050 kg/m$^3$). For both ensemble-averaged and individual beats, $dP/dU$ was calculated using least squares linear regression from the P-$U$ slope during early systole, when the contribution of backward-running waves is minimal (17, 18, 26, 43, 44). Time lags between $P$ and $U$ data points related to hardware-related delays (14) and any difference in the relative positions of flow probe and micromanometer measurement sites were corrected by aligning the peak second derivatives of these signals. As noted previously (42), all three sites displayed highly linear early-systolic P-$U$ relations ($R^2 \geq 0.99$).

As per convention (4, 25), waves propagating away from the ventricle were defined as forward-running and those arising from the vasculature as backward-running. Using established methodology (8, 26, 42-45), WI of forward-running ($WI_+$) and backward-running waves ($WI_-$) was calculated as $WI_{\pm} = (dP_{\pm}/dt)(dU_{\pm}/dt)$, where the pressure and velocity
differentials associated with these waves were given by \( \frac{dP_\pm}{dt} = \frac{1}{2} (dP/dt \pm \rho c \cdot dU/dt) \) and
\( \frac{dU_\pm}{dt} = \frac{1}{2} (dU/dt \pm 1/\rho c \cdot dP/dt) \) respectively. Waves causing a pressure increase (\( dP/dt > 0 \))
were classified as compression waves, while waves causing a pressure decrease (\( dP/dt < 0 \))
were classified as expansion waves (8, 26, 42-45). The forward and backward components of
pressure (\( P_\pm \)) and velocity (\( U_\pm \)) were obtained by integrating the appropriate \( P \) or \( U \)
differentials (21). Wave size was quantified with the cumulative intensity (CI), calculated as
the integral of \( W_{I\pm} \) over the duration of the wave (8, 26, 42-44). \( P \) and \( U \) changes related to
each wave (i.e. \( \Delta P \) and \( \Delta U \) respectively) were obtained as the difference in \( P_\pm \) or \( U_\pm \) measured
between the start and end of the wave (42-45). To analyze the relative distribution of PT flow
into the DA and major PA, \( \Delta U \) was converted back to a flow value (i.e. \( \Delta Q \)) via multiplication
by vessel cross-sectional area, with left PA \( \Delta Q \) then multiplied by the total-to-left lung weight
ratio to obtain the combined left and right PA \( \Delta Q \).

**Computer modeling** Flow dynamics between the PT, PA and DA were investigated
using three variants of a simple mathematical model, with full details of computational
techniques described elsewhere (22, 23). The basic design of each model consisted of three
one-dimensional (1D) segments representing the PT, PA and DA. The PA and DA segments
were each terminated in a 3-element windkessel (3Wk), i.e. a zero-dimensional (0D)
compartment representing the characteristic impedance of the downstream vessel (\( Z_{PA}, Z_{DA} \)),
as well as the resistance (\( R_{Lung}, R_{LBP} \)) and compliance (\( C_{Lung}, C_{LBP} \)) of the lungs and the
combined lower body and placenta (LBP) respectively (Fig. 2). \( Z_{PA} \) and \( Z_{DA} \) were calculated
to achieve well-matched coupling to the appropriate 1D segment (22), \( R_{Lung} \) and \( R_{LBP} \) were
computed on the basis of experimental mean pressure/flow data, \( C_{LBP} \) was estimated on the
basis of pulse pressure, while \( C_{Lung} \) was set to achieve the mean PA BCW ms/FCW ms CI ratio
obtained in the experimental studies. The boundary condition at the PT inlet was prescribed
as a forward-component of RV pressure combined with a numerical pulmonary valve (22,
23), and unless otherwise stated, any backward-running PT waves approaching the RV/PT
interface were assumed to be completely absorbed without reflection when the pulmonary
valve was open. The respective group-averaged wave speeds from experimental studies were
applied to the 1D segments, while segment diameters (1.2, 0.9 and 0.9 cm for the PT, main
PA and DA) were also obtained from experimental studies.
To assess the role of large vessel compliance and propagation delay between forward and backward waves, two model variants were compared. In the first (Model A), all 1D segments were assigned a very short length (0.2 cm), thus approximating a classical lumped-parameter (i.e. 0D) model containing two 3Wk compartments representing the PA/Lung and DA/LBP flow paths. In Model B, segment lengths (2.6, 7.0 and 2.6 cm for the PT, PA and DA respectively) and the locations at which pressure and flow were monitored (0.3, 1.2 and 1.27 cm from the junction of these vessels) were estimated on the basis of wave speed and wave timing information derived from experimental studies, thus accounting for both wave propagation effects and PT/DA/major PA compliance. As Model B did not fully reproduce WI features seen in experimental studies, a third model variant (Model C) additionally incorporated an impedance mismatch at the RV/PT interface, so that any backward-running waves were partially reflected with a reflection coefficient of 0.36, calculated from experimental data as the ratio of PT FCWms ΔP to PT BCWms ΔP. For all models, WI analysis was performed as described in the preceding section.

**Statistical Analysis**

Experimental data were analysed using Statistical Package for the Social Sciences Version 16.0 (SPSS Inc., Chicago), with logarithmic transformation performed before analysis where data were non-normally distributed. Baseline and beat-by-beat post-ES DA, PT and PA hemodynamic and WI data were analyzed using repeated measures analysis of variance, and specific comparisons evaluated by partitioning within-animal sums of squares into individual degrees of freedom. As the pattern of change in WI profiles was not different between duplicate ES datasets, ES data were pooled so that each fetus yielded one dataset for inclusion in the final analysis. WI and flow relationships amongst the PT, PA and DA were evaluated with least squares regression analysis, and because both X and Y datapoints were associated with an error component, the regression equation was calculated as the line of symmetry through the data (5). Individual animal relationships were then averaged to obtain an overall regression equation for the study group. Results are expressed as mean ± SD and significance was taken at *P* < 0.05.

**Results**

*Baseline blood gases, hemodynamics and WI analysis* Ascending aortic pH was 7.31 ±
0.02, Hb 12.4 ± 1.7 g/dL, Hb O_2 saturation 69 ± 7%, Po_2 24.5 ± 3.0 mmHg, Pco_2 48.9 ± 2.4 mmHg and base excess -2.4 ± 1.3 mmol/L.

Peak systolic and mean blood pressures in the PT and left PA were higher than in the DA, while 83% of mean PT flow crossed the DA and 17% entered the lungs via the PA. However, DA and PA phasic flow profiles displayed distinct temporal differences, with early systolic peak flow in the PA being ~50% higher than in the DA (P < 0.005) and predominant DA flow only emerging in mid-systole. Proto-diastolic flow was forward in the DA (0.52 ± 0.19 L/min, P < 0.001), retrograde in the PA (−0.39 ± 0.15 L/min, P < 0.001) and insignificant in the PT (−0.09 ± 0.17 L/min, P = 0.15). Conversely, average diastolic flow was forward in the DA (0.14 ± 0.10 L/min, P = 0.002) and PT (0.06 ± 0.03 L/min, P < 0.001), but not different to zero in the PA (−0.03 ± 0.07 L/min, P = 0.3; Fig. 3A&B, Table 1).

Wave speed differed between the three sites, such that DA > PT > PA. Although prominent in all sites, a FCW_is and its associated ΔQ were smaller in the DA than PA (P < 0.025). Moreover, a BCW_ms was substantial in the PT, particularly large in the PA, but quite small in the DA (P < 0.001), with correspondingly similar differences in the associated ΔP, ΔQ and BCW_ms/FCW_is ratios (all P < 0.001). By contrast, the DA contained a striking FCW_ms with a ΔQ similar to the DA FCW_is (P = 0.7) but larger than that of a FCW_ms present in the PT and PA (P < 0.001; Fig. 3C-E, Table 2).

Dynamic blood pressure and flow changes Small rises (~2 mmHg) in systolic PT, PA and DA blood pressures (P < 0.001) occurred for several beats after an ES, while mean and diastolic pressures fell by ~2 mmHg (P < 0.001) at all sites in the first post-ES beat (see data supplement). Mean PT, DA and PA flows all rose after an ES (P < 0.001), but the rise in the early systolic PA flow peak (1.01 ± 0.31 L/min) exceeded that of the DA (0.44 ± 0.23 L/min, P < 0.001). By contrast, the mid-systolic flow peak increased by 0.50 ± 0.22 L/min in the DA (P < 0.001), but was unaltered in the PA (change 0.08 ± 0.25 L/min, P > 0.3; Fig. 4).

Dynamic changes in WI analysis Apart from minor (≤10%) and transient changes in the PT and PA, wave speed was unaltered after an ES (see supplemental data). As shown in Fig. 5, FCW_is and BCW_ms CI increased in the first post-ES beat (all P < 0.001), with proportionally similar rises in the PT (155-160%), DA (144-146%) and left PA (146-175%). However, the increase in DA FCW_ms CI (56%, P < 0.001) was similar to the rise in PT.
FCW<sub>ms</sub> CI (53%, \( P < 0.001 \)), but less than half of the increment in DA FCW<sub>is</sub> CI (\( P = 0.002 \)). The CI of FCW<sub>is</sub>, FCW<sub>ms</sub> and BCW<sub>ms</sub> returned to control levels by the 3<sup>rd</sup>-4<sup>th</sup> post-ES beat. By contrast, FEW<sub>is</sub> CI fell only in the initial post-ES beat in the PT (−19%) and PA (−24%, both \( P < 0.001 \)), and was unchanged in the DA (\( P = 0.5 \)).

As evident in Fig. 6, the magnitude of FCW<sub>is</sub> and BCW<sub>ms</sub> \( \Delta Q \) increased in the first post-ES beat (all \( P < 0.001 \)), with proportionally similar rises in the PT (52-69%), DA (39-62%) and PA (44-54%). By contrast, FCW<sub>ms</sub> \( \Delta Q \) rose by only 16% in the PT and 28% in the DA (both \( P < 0.001 \)). The FCW<sub>is</sub>, FCW<sub>ms</sub> and BCW<sub>ms</sub> \( \Delta Q \) returned to control levels by the 3<sup>rd</sup>-4<sup>th</sup> post-ES beat. However, the absolute value of FEW<sub>is</sub> \( \Delta Q \) in the PT and PA fell by 6% (\( P < 0.025 \)) and 18% (\( P < 0.001 \)) respectively in the initial post-ES beat, but was unchanged in the DA (\( P > 0.8 \)).

Highly linear relationships were present between the CI of PT FCW<sub>is</sub> (X) and the PA or DA FCW<sub>is</sub> (Y), with ~60% greater average slope for the PT-PA FCW<sub>is</sub> CI relation (0.79 ± 0.25 vs. 0.48 ± 0.17, \( P < 0.01 \), Fig. 7A&B). The overall slope of the corresponding PT-PA FCW<sub>is</sub> \( \Delta Q \) relation (0.55 ± 0.29) was double that of the PT-DA FCW<sub>is</sub> \( \Delta Q \) relation (0.26 ± 0.07, \( P < 0.01 \), Fig. 7C&D).

Linear relationships were evident between the CI of PA BCW<sub>ms</sub> (X) and PT BCW<sub>ms</sub> or DA FCW<sub>ms</sub> (Y), with the average slope of the former (0.35 ± 0.12) nearly double that of the latter (0.19 ± 0.12, \( P < 0.005 \), Fig. 8A&B). In addition, the overall slope of the PA-PT BCW<sub>ms</sub> \( \Delta Q \) relation (1.34 ± 0.40) was 9-fold that of the PA BCW<sub>ms</sub>-DA FCW<sub>ms</sub> \( \Delta Q \) relation (0.15 ± 0.06, \( P < 0.001 \), Fig. 8C&D).

**Mathematical modeling** While the 0D approximation (Model A) produced a higher DA-than-PA mean flow, flow and WI profiles (Fig. 9) were not in accord with experimental data (Table 1, Fig. 3). In particular, 1) phasic PA flow never exceeded DA flow, 2) PT flow did not fall abruptly in mid-systole, 3) the rise in DA flow was monophasic, 4) the PT and PA BCW<sub>ms</sub> occurred entirely within the time-span of the FCW<sub>is</sub>, 5) the DA WI profile did not contain a FCW<sub>ms</sub> and 6) DA flow was near-zero in proto-diastole and negative (i.e. retrograde) throughout diastole.

Inclusion of vessel compliance and wave propagation effects (Model B) unmasked a
greater transmission of waves from the PT into the PA, resulting in flow and WI patterns (Fig. 9) that were much closer to experimental data (Table 1, Fig. 3). Specifically, 1) PA flow exceeded DA flow in early systole, 2) PT flow fell abruptly in mid-systole, 3) the increase in DA flow was bi-phasic, 4) a very large PA BCWms was partially transmitted into the PT as a BCWms and into the DA as a FCWms, 5) proto-diastolic flow was positive in the DA, negative in the PA and near-zero in the PT and 6) diastolic flow was positive in both the DA and PT, and near-zero but slightly negative in the PA.

Model B did not, however, predict a PT FCWms, which may explain why the PT flow reduction between early and mid-systolic peaks (46%) was more pronounced than in experimental data (27%, Table 1). The additional incorporation of an impedance mis-match at the RV-PT interface (Model C) had four main consequences (Fig. 9): 1) a FCWms appeared in the PT WI profile, ameliorating the PT mid-systolic flow reduction to a level close to that seen experimentally, 2) the DA FCWms became broader, with a rise in the DA FCWms/PA BCWms CI ratio to near-experimental values, indicating that this wave arose partly as a direct antegrade transmission of the PA BCWms, and partly as an antegrade transmission of the PT FCWms arising from reflection of the BCWms at the PT-RV interface, 3) the augmented DA FCWms was accompanied by an increase in its flow effect, and 4) the PT FCWms was partially transmitted into the PA where it partly counteracted the flow-reducing effect of the PA BCWms. Overall, the PT, PA and DA flow and WI profiles obtained from Model C were very similar to in vivo waveforms (Fig. 3).

A visual representation of PT, PA and DA flow and wave interactions using Model C is provided in a video (see data supplement), where 1) the instantaneous direction and magnitude of flow is indicated by arrows in each vessel, 2) the propagation of waves is apparent from the pressure-dependent variations in vessel diameter, which have been amplified ten-fold for purposes of visual clarity, and 3) corresponding flow and wave intensity signals are shown in separate panels.

**Discussion**

Three main findings have emerged from this study, where flow profile and WI analyses performed simultaneously in the fetal PT, PA and DA at steady-state and after an ES were
complemented with mathematical modeling of flow dynamics. First, although average PT blood flow mainly passed across the DA, most of the initial systolic component of PT flow related to FCW_{is} entered the major PA, and a predominance of DA flow only emerged with appearance of the PA BCW_{ms} and DA FCW_{ms} in mid-systole. Second, forward DA flow in proto-diastole was accompanied by substantial retrograde PA flow. Third, subsequent forward DA flow in diastole occurred without significant retrograde PA flow.

Our data indicated that the various phases of the cardiac cycle were associated with quite specific interactions between the fetal PT, DA and major PA. In early systole, interactions mainly occurred along the PT-to-DA and PT-to-PA axes. Thus, an early-systolic flow peak in PT, DA and PA flow profiles was associated with a FCW_{is}, a wave which arises from the initial ventricular impulse that provides forward momentum to blood in the ventricle (49). However, despite mean PA flow comprising only 17% of RV output, three lines of evidence indicated that this RV-derived FCW_{is} and its flow effects were preferentially transmitted into the major PA rather than the DA: 1) in early systole, PA peak flow was higher than in the DA (Table 1, Figs. 3&4), 2) both PA FCW_{is} CI and its ΔQ component were greater than their DA counterparts (Table 2, Figs. 5&6) and 3) the slopes of the PT-PA FCW_{is} CI and ΔQ relations exceeded those of the corresponding PT-DA relations (Fig. 7). Importantly, despite the large size of the DA, such preferential passage of the FCW_{is} CI and ΔQ into the PA strongly suggests that characteristic impedance (i.e. the ΔP/ΔQ ratio in the absence of reflection) in the DA was higher than in the PA. This proposition was not only supported by modeling results, but also by the observation that the magnitudes of PT and PA FEW_{ls} CI and ΔQ all fell in the first beat after an ES, whereas the DA FEW_{ls} CI and ΔQ did not change (Figs. 5&6).

In contrast to early systole, mid-systolic interactions predominantly occurred in the PA-to-PT and PA-to-DA directions, and were closely linked to the fates of a very large PA BCW_{ms}. Thus, in the PT, a mid-systolic reduction in flow (Fig 3, Table 1) was accompanied by a large BCW_{ms} that appears to be derived primarily from retrograde transmission of the PA BCW_{ms} into the PT (42, 44), as it is abolished by occlusion of the main pulmonary artery (11, 44). Moreover, baseline (Table 2) and ES data (Fig. 8) suggested that about one-third of the PA BCW_{ms} CI, but most of its ΔQ effects, were transmitted into the PT.
Unlike the PT, the DA flow profile displayed a mid-systolic augmentation associated with a large FCW_{ms} (Fig 3). As the DA FCW_{ms} is abolished by occlusion of the main PA, we previously concluded that this wave arose as an antegrade transmission of the PA BCW_{ms} (42). However, additional calculation of $\Delta Q$ in the present study suggests that this transmission has both direct and indirect components. Specifically, if all the DA FCW_{ms} was derived directly from the PA BCW_{ms}, then the $\Delta Q$ of PA BCW_{ms} should be similar to the sum of the PT BCW_{ms} and DA FCW_{ms} $\Delta Q$. However, the combined $\Delta Q$ of PT BCW_{ms} and DA FCW_{ms} exceeded the PA BCW_{ms} $\Delta Q$ (Table 2), implying that only a portion of the DA FCW_{ms} arose directly as an antegrade transmission from the PA BCW_{ms}. A plausible basis for the remainder of the DA FCW_{ms} was that the PT BCW_{ms} (which arises from the PA BCW_{ms}) was partially reflected at the PT/RV interface as a FCW_{ms} that was then transmitted into the DA. Such a dual origin of the DA FCW_{ms} is consistent with the magnitude of the PT FCW_{ms} $\Delta Q$ (Table 2), the finding that the PT FCW_{ms} is markedly attenuated after occlusion of the main PA (42, 44), and the results of our modeling simulations (Fig. 9).

Under baseline conditions, DA FCW_{ms} CI and $\Delta Q$ were \(-40\%\) and \(-50\%\) respectively that of the PA BCW_{ms} (Table 2). However, the average slopes of the PA BCW_{ms}-DA FCW_{ms} CI and $\Delta Q$ relations were 0.19 and 0.15 (Fig 8), implying that there was only a limited capacity for transient increases in the energy and flow effects of PA BCW_{ms} to be transmitted into the DA. Given our conclusion that the DA FCW_{ms} in part arose as a transmission of the PT FCW_{ms}, one likely contributor to this phenomenon was an attenuated rise in the PT FCW_{ms} CI and $\Delta Q$ observed after an ES. The latter in turn implies that the degree of reflection of the PT BCW_{ms} at the RV-PT interface transiently fell after an ES.

The fetal PA flow profile characteristically displays a prominent negative peak around the time of the dicrotic notch, (1, 19, 32, 34, 35, 37), and may also exhibit negative (i.e. retrograde) PA flow in the main part of diastole (2, 7, 27, 32, 37). No distinction has previously been made between these two periods of negative PA flow, with proposed mechanisms including an elastic recoil of major PA related to coupling of low compliance large arteries with a high pulmonary vascular resistance (34, 35, 37) and a reflection of pressure waves from a vasoconstricted pulmonary vascular bed (7). Delineation of the specific mechanism underlying proto-diastolic and diastolic PA flow patterns is particularly
relevant as persistence of forward DA flow in both these phases of the cardiac cycle has been attributed to a reversal of PA flow (7, 37). That this explanation might not be entirely correct, however, is suggested by published figures which indicate that PA flow in the main part of diastole is not always negative, but may be near-zero (19, 28) or even positive (35).

Concurrent measurement of PT, PA and DA flows in the present study strongly suggest that differing mechanisms underpinned forward DA flow in proto-diastole and diastole. Thus, the pattern of forward DA, retrograde PA but insignificant PT flow, together with the magnitudes of DA and PA flow (Table 1, Fig. 3), was consistent with reversal of PA flow accounting for the major portion (∼75%) of forward DA flow in proto-diastole. On the other hand, the presence of forward DA and PT, but near-zero PA flow, indicated that such flow reversal was not a major contributor to right-to-left DA shunting in diastole. Instead, the latter flow pattern was consistent with transuductal discharge of blood stored during systole in a more central reservoir, consisting of the PT and probably the main PA and proximal DA. This explanation accords with modeling data, where incorporation of large vessel compliance produced diastolic flow patterns (Fig. 9) similar to experimental findings (Fig. 3). Note that our explanation does not preclude a potential contribution from branches beyond the major PA to forward DA diastolic flow, but the variability of diastolic PA flow evident in published figures (2, 7, 19, 27, 28, 32, 35, 37) implies that the extent of this contribution is dependent on the balance between proximal and distal run-off.

A notable feature of the downstroke of the prominent proto-diastolic negative peak in PA flow was that it occurred in conjunction with the PA FEWls (Fig. 3). As the FEWls is a manifestation of a rarefaction or “suction” wave arising from ventricular relaxation (49), this coincidence not only indicates that proto-diastolic reversal of PA flow was intimately associated with RV relaxation, but also provides further insight into the mechanism underlying this reversal. Specifically, our data suggest that the elastic recoil previously proposed for the negative proto-diastolic PA flow peak (34, 35) is related to an active release of systolic PA pressure by the RV (via the FEWls). The extra blood volume stored in the major PA during systole is then primarily distributed towards the path of least resistance (i.e. the DA), thereby resulting in retrograde PA flow. Intriguingly, although the PA FEWls is a transmission of the PT FEWls, our measurements of proto-diastolic PT, DA and PA flows
(Table 1) indicated that the PA flow reversal initiated by the PA FEWs mainly passed into the DA, rather than the PT, and thereby ameliorated falls in DA flow related to the DA FEWs. This effect is clearly evident in the video depicting interactions between flow and wave dynamics in the PT, PA and DA (data supplement).

A greater distribution of average fetal PT blood flow into the DA compared with the PA (3, 20, 30, 31, 46, 47) has been explained on the basis that the high resistance and low compliance of the pulmonary circulation leads to preferential shunting of PT flow across a widely patent DA to the lower resistance/higher compliance fetal body and placental circulations (12, 34, 35). In terms of a mathematical model, this schema can be represented by a ‘classical’ lumped-parameter model (Fig 9, Model A), which produced appropriate levels of mean PT, DA and PA flow, but not the typical phasic features of flow or the WI patterns seen in vivo. These shortcomings were largely rectified by representing the PT, PA and DA as one-dimensional compliant vessels with physiological lengths and wave speeds (Fig 9, Model B), thus accounting for the delay between forward- and backward-running waves (i.e. ‘wave propagation effects’). Importantly, this modification revealed a greater transmission of waves (and flow) into the PA in early systole, compared with the DA, which was caused by a lower PA-than-DA characteristic impedance. The further incorporation of an impedance mismatch at the PT-RV interface (Fig 9, Model C) led to a PT FCWms that was transmitted into the DA and PA, resulting in simulated PT, PA and DA flow waveforms that were very similar to those obtained in vivo (Fig. 3). This final model therefore highlighted the hitherto unrecognized role of three factors in the interaction of fetal PT-PA-DA flow dynamics, namely 1) wave propagation effects, 2) a high DA but low PA characteristic impedance and 3) reflection of backward-running arterial waves at the PT-RV interface. Note that the similarity of the DA flow profile in this final model, which does not account for backward-running waves in the DA, to that observed in vivo (Fig. 3B), as well as the relatively small size of such waves in the DA WI profile (Fig. 3E), implies that backward-running waves originating from the left ventricle and ascending aorta do not normally make a significant contribution to fetal DA hemodynamics.

Several methodological issues require comment. First, the use of transit-time flow probes to measure distribution of PT flow into the DA and major PA was associated with
several potential sources of error. Thus, calculation of the combined left and right PA flow
from measured left PA flow assumed identical weight-corrected flow to the left and right
lungs. However, our scaling factor (2.55) is entirely in accord with the finding that left PA
flow comprises 40% of total lung flow in fetal lambs (19). Furthermore, even though the
recorded flow at all sites was associated with measurement error, our method of calculating
RV output assumed that all such error resided in the PT flow. Despite this, however, the
average adjustment applied to PT flow (10% increase) was in line with the ±10% absolute
accuracy of flow measurements obtained with ‘A series’ transit-time probes (48).

Second, wave speed was higher in the DA (5.2 m/s) than the PT (3.9 m/s), which
contrasts with our previous study (42), where DA (5.0 m/s) and PT wave speed (4.3 m/s) were
not significantly different. The main factor responsible for this differing result appears to be
the upward scaling of PT blood flow to ensure that it was internally consistent with the sum of
the DA and PA flows. This scaling skewed the PT P-U loop to the right, thereby decreasing
the slope of this relation in early-systole, and thus calculated PT wave speed.

Third, because of the extent of surgical instrumentation required for physiological
measurements, it was necessary to perform experiments under general anesthesia and with the
fetus partially exteriorized. However, blood gas and pressure data were within the normal
range reported in unanesthetized, chronically-instrumented late-gestation fetal lambs (3, 9, 16,
19, 24, 29, 36, 38, 41, 46, 47). Furthermore, the morphology of the left PA flow profile (Fig.
3) closely resembled that of chronically-instrumented fetal sheep (2, 7, 19, 27, 28, 32, 35, 37),
while the proportion of RV output distributed to the lungs was near the upper end of the range
of 10-16% measured in chronic studies (3, 46, 47). Finally, this experimental preparation also
exhibits the predominance of RV output (45) observed in chronically-instrumented fetal
lambs (3, 33, 38, 40, 46). It is thus unlikely that the qualitative features of findings were
affected by our experimental approach. However, we cannot exclude the possibility that wave
transmission and reflection characteristics may have been quantitatively altered by general
anesthesia, as well as by perivascular dissection and subsequent placement of flow probes,
although the magnitude of any such effect is likely to be quite minor.
In summary, the results of this study suggest that 1) a net preferential passage of fetal RV output into the DA is overlaid by a distinct systolic pattern favoring flow distribution to the PA in early systole, but the DA in mid- and late-systole, and 2) while DA right-to-left shunting around the time of pulmonary valve closure is mainly due a reversal of PA flow, continued DA shunting in diastole is primarily related to discharge of a more central vascular reservoir filled in systole. These phasic flow patterns were accompanied by a characteristic impedance which was higher in the DA than the PA. The combination of low PA characteristic impedance but high pulmonary microvascular resistance observed in the fetus is highly unusual, but appears to be a key element in the reservoir function of large PA. We speculate that this unusual combination may also facilitate the rapid birth-related rise in pulmonary blood flow (7, 27), as this rise could be achieved primarily through a reduction in pulmonary microvascular resistance, without any need for major changes in the vascular properties of conduit PA.
Acknowledgements

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Figure legends

Figure 1. Schematic diagram of fetal instrumentation. **Abbreviations:** AA, ascending aorta; Ao F, aortic fluid-filled catheter; DA FP, ductus arteriosus flow probe; DA M, ductus arteriosus micromanometer catheter; DA S, ductus arteriosus snare; LA, left atrium; LPA FP, left pulmonary artery flow probe; LPA M, left pulmonary artery micromanometer catheter; LV, left ventricle; PA S, main pulmonary artery snare; PT F, pulmonary trunk fluid-filled catheter; PT FP, pulmonary trunk flow probe; PT M, pulmonary trunk micromanometer catheter; RV, right ventricle.

Figure 2. Schematic of the basic mathematical model, which has three variants (Models A-C, see text). In each, the PT, main PA and DA are represented as 1D segments (higher wave speed is represented by thicker lines), a forward-component of RV pressure is applied at the inlet of the PT via a numerical pulmonary valve, and 3-element windkessel compartments represent the bulk properties of the lungs and the combined fetal lower body and placenta (LBP). Note that values of characteristic impedance ($Z$), resistance ($R$), compliance ($C$) and asymptotic pressure ($P_\infty$) have been derived from experimental data. Other abbreviations are as in Fig 1.

Figure 3. Net blood pressure (panel A) and flow (panel B) in the PT (thick black line), PA (thin black line) and DA (thin gray line), as well as separated forward and backward wave intensities in the PT (panel C), PA (panel D) and DA (panel E). Note early-systolic (solid arrow) and mid-systolic (dashed arrow) flow peaks. **Abbreviations:** AvD, average diastolic region; PD, proto-diastolic region; other abbreviations are as in Table 2.

Figure 4. Changes in fetal peak early systolic (panel A), peak mid-systolic (panel B) and mean blood flow (panel C) in the PT (●), PA (■) and DA (□) at baseline (B) and in seven consecutive beats (P1-P7) following a ventricular extrasystole.

Figure 5. Cumulative intensity (CI) of FCW_{is} (panel A), BCW_{ms} (panel B), FCW_{ms} (panel C) and FEW_{ls} (panel D) in the PT (●), PA (■) and DA (□) at baseline (B) and in seven consecutive beats (P1-P7) following a ventricular extrasystole. Abbreviations are as in Table 2.
Figure 6. Blood flow change ($\Delta Q$) related to $FCW_{is}$ (panel A), $BCW_{ms}$ (panel B), $FCW_{ms}$ (panel C) and $FEW_{is}$ (panel D) in the PT (●), PA (■) and DA (□) at baseline (B) and in seven consecutive beats (P1-P7) following a ventricular extrasystole. Abbreviations are as in Table 2.

Figure 7. Individual (thin lines) and average (thick line) linear regression relationships between the PT and PA $FCW_{is}$ CI (panel A) and $\Delta Q$ (panel C), and between the PT and DA $FCW_{is}$ CI (panel B) and $\Delta Q$ (panel D). Abbreviations are as in Table 2. Equations refer to average regression line.

Figure 8. Individual (thin lines) and average (thick line) linear regression relationships between the PA and PT $BCW_{ms}$ CI (panel A) and absolute value of $\Delta Q$ (panel C), and between the PA $BCW_{ms}$ and DA $FCW_{ms}$ CI (panel B) and absolute value of $\Delta Q$ (panel D). Abbreviations are as in Table 2. Equations refer to average regression line.

Figure 9. Flow (whole profile and zoom of the zero-axis region) and separated WI profiles in the PT, main PA and DA of three progressively more complex variants of a mathematical model, where wave propagation effects have been ignored by reducing 1D segment lengths to a small value (Model A), wave propagation effects have been included by incorporating physiological PT, PA and DA lengths (Model B), and an impedance mismatch has been added at the RV-PT interface (Model C). Note that 1) $FEW_{ms}$ is a mid-systolic forward-running expansion wave arising from reflection of PA $BCW_{ms}$ at the PT-PA-DA junction, and other abbreviations are as in Table 2, 2) the DA WA profile lacks backward-running waves, which arise from the thoracic aorta and have not been accounted for in the model, and 3) because vascular segments are very short in Model A, the $BCW_{ms}$ within the PT and PA WI profiles occurs almost entirely during the period of the $FCW_{is}$. 
**TABLE 1.** Baseline regional blood pressure and flow data in fetal lambs.

<table>
<thead>
<tr>
<th></th>
<th>PT</th>
<th>DA</th>
<th>PA</th>
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<tr>
<td><strong>Blood pressure (mmHg)</strong></td>
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<tr>
<td>Peak systolic</td>
<td>72.8 ± 7.6</td>
<td>70.4 ± 7.6&lt;sup&gt;b&lt;/sup&gt;</td>
<td>72.0 ± 7.6</td>
</tr>
<tr>
<td>Mean</td>
<td>59.3 ± 5.9</td>
<td>58.8 ± 5.9&lt;sup&gt;a&lt;/sup&gt;</td>
<td>59.0 ± 5.9</td>
</tr>
<tr>
<td>Minimum diastolic</td>
<td>49.9 ± 5.2</td>
<td>50.0 ± 5.2</td>
<td>50.0 ± 5.3</td>
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<tr>
<td><strong>Blood flow (L/min)</strong></td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>Early systolic peak</td>
<td>3.18 ± 0.40&lt;sup&gt;c&lt;/sup&gt;</td>
<td>1.08 ± 0.18&lt;sup&gt;f&lt;/sup&gt;</td>
<td>1.61 ± 0.37</td>
</tr>
<tr>
<td>Mid systolic peak</td>
<td>2.32 ± 0.87</td>
<td>2.14 ± 0.62</td>
<td>0.33 ± 0.20&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>Proto-diastolic</td>
<td>-0.09 ± 0.17&lt;sup&gt;c&lt;/sup&gt;</td>
<td>0.52 ± 0.19</td>
<td>-0.39 ± 0.15</td>
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<tr>
<td>Diastolic</td>
<td>0.06 ± 0.03</td>
<td>0.14 ± 0.10&lt;sup&gt;b&lt;/sup&gt;</td>
<td>-0.03 ± 0.07&lt;sup&gt;d&lt;/sup&gt;</td>
</tr>
<tr>
<td>Mean</td>
<td>0.78 ± 0.14</td>
<td>0.65 ± 0.13&lt;sup&gt;e&lt;/sup&gt;</td>
<td>0.13 ± 0.08&lt;sup&gt;c&lt;/sup&gt;</td>
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</table>

Data are expressed as mean ± SD; n = 10. Abbreviations: DA, ductus arteriosus; PT, pulmonary trunk; PA, pulmonary artery. <sup>a</sup> P < 0.05, <sup>b</sup> P < 0.025, <sup>c</sup> P < 0.001 compared to other vascular sites; <sup>d</sup> P < 0.025<sup>e</sup> P < 0.01 compared to PT; <sup>f</sup> P < 0.001 compared to PA. Note that 1) proto-diastolic flow value is not directly comparable to diastolic or mean flows, as it represents an average flow during a brief period surrounding closure of the pulmonary valve, 2) absolute values of proto-diastolic and diastolic flows have been compared between sites.
## TABLE 2. Baseline regional wave intensity data in fetal lambs.

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<th>PT</th>
<th>DA</th>
<th>PA</th>
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<tr>
<td><strong>Wave speed (m/s)</strong></td>
<td>3.9 ± 0.6</td>
<td>5.2 ± 1.5&lt;sup&gt;b&lt;/sup&gt;</td>
<td>3.1 ± 0.7&lt;sup&gt;c&lt;/sup&gt;</td>
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<tr>
<td><strong>CI (Wm&lt;sup&gt;-2&lt;/sup&gt;s&lt;sup&gt;-1&lt;/sup&gt; x10&lt;sup&gt;4&lt;/sup&gt;)</strong></td>
<td></td>
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<tr>
<td>FCW&lt;sub&gt;is&lt;/sub&gt;</td>
<td>2.37 ± 0.67&lt;sup&gt;b&lt;/sup&gt;</td>
<td>1.19 ± 0.27&lt;sup&gt;e&lt;/sup&gt;</td>
<td>1.91 ± 0.85 &lt;sup&gt;&lt;/sup&gt;</td>
</tr>
<tr>
<td>FCW&lt;sub&gt;ms&lt;/sub&gt;</td>
<td>0.18 ± 0.22</td>
<td>0.89 ± 0.72&lt;sup&gt;b&lt;/sup&gt;</td>
<td>0.14 ± 0.12 &lt;sup&gt;&lt;/sup&gt;</td>
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<tr>
<td>BCW&lt;sub&gt;ms&lt;/sub&gt;</td>
<td>0.73 ± 0.30</td>
<td>0.05 ± 0.06&lt;sup&gt;b&lt;/sup&gt;</td>
<td>2.15 ± 1.45&lt;sup&gt;d&lt;/sup&gt;</td>
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<tr>
<td>FEW&lt;sub&gt;ls&lt;/sub&gt;</td>
<td>1.57 ± 0.79&lt;sup&gt;a&lt;/sup&gt;</td>
<td>1.22 ± 0.76</td>
<td>1.12 ± 0.68 &lt;sup&gt;&lt;/sup&gt;</td>
</tr>
<tr>
<td><strong>CI ratio</strong></td>
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<tr>
<td>BCW&lt;sub&gt;ms&lt;/sub&gt;/FCW&lt;sub&gt;ls&lt;/sub&gt;</td>
<td>0.30 ± 0.05</td>
<td>0.05 ± 0.04&lt;sup&gt;b&lt;/sup&gt;</td>
<td>1.07 ± 0.32&lt;sup&gt;d&lt;/sup&gt;</td>
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<tr>
<td><strong>ΔP (mmHg)</strong></td>
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<tr>
<td>FCW&lt;sub&gt;is&lt;/sub&gt;</td>
<td>13.4 ± 2.1&lt;sup&gt;b&lt;/sup&gt;</td>
<td>9.9 ± 1.2</td>
<td>10.4 ± 1.5 &lt;sup&gt;&lt;/sup&gt;</td>
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<tr>
<td>FCW&lt;sub&gt;ms&lt;/sub&gt;</td>
<td>2.8 ± 2.1</td>
<td>9.6 ± 4.6&lt;sup&gt;b&lt;/sup&gt;</td>
<td>1.9 ± 1.5 &lt;sup&gt;&lt;/sup&gt;</td>
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<tr>
<td>BCW&lt;sub&gt;ms&lt;/sub&gt;</td>
<td>7.7 ± 1.7</td>
<td>1.7 ± 1.2&lt;sup&gt;b&lt;/sup&gt;</td>
<td>10.7 ± 1.9&lt;sup&gt;d&lt;/sup&gt;</td>
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<tr>
<td>FEW&lt;sub&gt;ls&lt;/sub&gt;</td>
<td>-8.0 ± 1.8</td>
<td>-8.5 ± 2.6</td>
<td>-5.9 ± 1.4&lt;sup&gt;b&lt;/sup&gt;</td>
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<tr>
<td><strong>ΔP ratio</strong></td>
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<tr>
<td>BCW&lt;sub&gt;ms&lt;/sub&gt;/FCW&lt;sub&gt;ls&lt;/sub&gt;</td>
<td>0.57 ± 0.07</td>
<td>0.18 ± 0.13&lt;sup&gt;b&lt;/sup&gt;</td>
<td>1.02 ± 0.12&lt;sup&gt;d&lt;/sup&gt;</td>
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<tr>
<td><strong>ΔQ (L/min)</strong></td>
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<tr>
<td>FCW&lt;sub&gt;is&lt;/sub&gt;</td>
<td>3.13 ± 0.51&lt;sup&gt;b&lt;/sup&gt;</td>
<td>1.02 ± 0.14&lt;sup&gt;f&lt;/sup&gt;</td>
<td>1.71 ± 0.30 &lt;sup&gt;&lt;/sup&gt;</td>
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<tr>
<td>FCW&lt;sub&gt;ms&lt;/sub&gt;</td>
<td>0.64 ± 0.55</td>
<td>0.96 ± 0.41&lt;sup&gt;b&lt;/sup&gt;</td>
<td>0.31 ± 0.26&lt;sup&gt;c&lt;/sup&gt;</td>
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<td>BCW&lt;sub&gt;ms&lt;/sub&gt;</td>
<td>-1.78 ± 0.33</td>
<td>-0.18 ± 0.14&lt;sup&gt;b&lt;/sup&gt;</td>
<td>-1.80 ± 0.44 &lt;sup&gt;&lt;/sup&gt;</td>
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<tr>
<td>FEW&lt;sub&gt;ls&lt;/sub&gt;</td>
<td>-1.87 ± 0.49&lt;sup&gt;b&lt;/sup&gt;</td>
<td>-0.86 ± 0.22</td>
<td>-0.99 ± 0.18 &lt;sup&gt;&lt;/sup&gt;</td>
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Data are expressed as mean ± SD; n = 10. 
Abbreviations: DA, ductus arteriosus; PT, pulmonary trunk; PA, pulmonary artery; FCW<sub>is</sub>, initial systolic forward compression wave; FCW<sub>ms</sub>, mid-systolic forward compression wave; BCW<sub>ms</sub>, mid-systolic backward compression wave; FEW<sub>ls</sub>, late-systolic forward expansion wave; CI, cumulative wave intensity; ΔP, blood pressure change; ΔQ, blood flow change. <sup>a</sup>P < 0.025, <sup>b</sup>P < 0.001 compared to other vascular sites; <sup>c</sup>P < 0.025 <sup>d</sup>P < 0.001 compared to PT; <sup>e</sup>P < 0.005, <sup>f</sup>P < 0.001 compared to PA.
**Figure 1.** Schematic diagram of fetal instrumentation. **Abbreviations:** AA, ascending aorta; Ao F, aortic fluid-filled catheter; DA FP, ductus arteriosus flow probe; DA M, ductus arteriosus micromanometer catheter; DA S, ductus arteriosus snare; LA, left atrium; LPA FP, left pulmonary artery flow probe; LPA M, left pulmonary artery micromanometer catheter; LV, left ventricle; PA S, main pulmonary artery snare; PT F, pulmonary trunk fluid-filled catheter; PT FP, pulmonary trunk flow probe; PT M, pulmonary trunk micromanometer catheter; RV, right ventricle.
Figure 2. Schematic of the basic mathematical model, which has three variants (Models A-C, see text). In each, the PT, main PA and DA are represented as 1D segments (higher wave speed is represented by thicker lines), a forward-component of RV pressure is applied at the inlet of the PT via a numerical pulmonary valve, and 3-element windkessel compartments represent the bulk properties of the lungs and the combined fetal lower body and placenta (LBP). Note that values of characteristic impedance ($Z$), resistance ($R$), compliance ($C$) and asymptotic pressure ($P^\infty$) have been derived from experimental data. Other abbreviations are as in Fig 1.

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Value 1</th>
<th>Value 2</th>
<th>Unit 1</th>
<th>Unit 2</th>
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<td>$Z_{DA}$</td>
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<td>mmHg.s/mL</td>
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<td>22.0</td>
<td>mmHg.s/mL</td>
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<tr>
<td>$R_{Lung}$</td>
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<td>mmHg.s/mL</td>
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<tr>
<td>$C_{LBP}$</td>
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<td>mL/mmHg</td>
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<tr>
<td>$C_{Lung}$</td>
<td></td>
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<td>mL/mmHg</td>
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<tr>
<td>$P^\infty$</td>
<td>35</td>
<td></td>
<td>mmHg</td>
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Figure 3. Net blood pressure (panel A) and flow (panel B) in the PT (thick black line), PA (thin black line) and DA (thick gray line), as well as separated forward and backward wave intensities in the PT (panel C), PA (panel D) and DA (panel E). Note early systolic (solid arrow) and mid-systolic (dashed arrow) flow peaks. Abbreviations: AvD, average diastolic region; PD, proto-diastolic region; other abbreviations are as in Table 2.
Figure 4. Changes in fetal peak early systolic (panel A), peak mid-systolic (panel B) and mean blood flow (panel C) in the PT (●), PA (■) and DA (○) at baseline (B) and in seven consecutive beats (P1-P7) following a ventricular extrasystole.
Figure 5. Cumulative intensity (CI) of FCWsil (panel A), BCWms (panel B), FCWms (panel C) and FEWls (panel D) in the PT (●), PA (■) and DA (□) at baseline (B) and in seven consecutive beats (P1-P7) following a ventricular extrasystole. Abbreviations are as in Table 2.
Figure 6. Blood flow change (ΔQ) related to FCW$_{ls}$ (panel A), BCW$_{ms}$ (panel B), FCW$_{ms}$ (panel C) and FEW$_{ls}$ (panel D) in the PT (●), PA (■) and DA (○) at baseline (B) and in seven consecutive beats (P1-P7) following a ventricular extrasystole. Abbreviations are as in Table 2.
Figure 7. Individual (thin lines) and average (thick line) linear regression relationships between the PT and PA FCW_{is CI} (panel A) and ΔQ (panel C), and between the PT and DA FCW_{is CI} (panel B) and ΔQ (panel D). Abbreviations are as in Table 2. Equations refer to average regression line.
Figure 8. Individual (thin lines) and average (thick line) linear regression relationships between the PA and PT BCW<sub>ms</sub> CI (panel A) and absolute value of ΔQ (panel C), and between the PA BCW<sub>ms</sub> and DA FCW<sub>ms</sub> CI (panel B) and absolute value of ΔQ (panel D). Abbreviations are as in Table 2. Equations refer to average regression line.
Figure 9. Flow (whole profile and zoom of the zero-axis region) and separated WI profiles in the PT, main PA and DA of three progressively more complex variants of a mathematical model, where wave propagation effects have been ignored by reducing 1D segment lengths to a small value (Model A), wave propagation effects have been included by incorporating physiological PT, PA and DA lengths (Model B), and an impedance mismatch has been added at the RV-PT interface (Model C). Note that 1) FEW_{ms} is a mid-systolic forward-running expansion wave arising from reflection of PA BCW_{ms} at the PT-PA-DA junction, and other abbreviations are as in Table 2, 2) the DA WA profile lacks backward-running waves, which arise from the thoracic aorta and have not been accounted for in the model, and 3) because vascular segments are very short in Model A, the BCW_{ms} within the PT and PA WI profiles occurs almost entirely during the period of the FCW_{ls}.