



Z-score reference ranges for pulsed-wave Doppler indices of the cardiac outflow tracts in normal fetuses

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Abstract

To determine Z-score equations and reference ranges for Doppler flow velocity indices of cardiac outflow tracts in normal fetuses. A prospective cross-sectional echocardiographic study was performed in 506 normal singleton fetuses from 18 to 40 weeks. Twelve pulsed-wave Doppler (PWD) measurements were derived from fetal echocardiography. The regression analysis of the mean and the standard deviation (SD) for each parameter were performed against estimated fetal weight (EFW) and gestational age (GA), in order to construct Z-score models. The correlation between these variables and fetal heart rate were also investigated. Strong positive correlations were found between the twelve PWD indices and the independent variables. A linear-quadratic regression model was the best description of the mean and SD of most parameters, with the exception of the velocity time interval (VTI) of ascending aorta against EFW, which was best fitted by a fractional polynomial. Z-score equations and reference values for PWD indices of fetal cardiac outflow tracts were proposed against GA and EFW, which may be useful for quantitative assessment of potential hemodynamic alternations, particularly in cases of intrauterine growth retardation and structural cardiac defects.

Keywords Z-score · Reference ranges · Doppler flow velocity waveform · Fetal echocardiography · Fetal cardiac outflow tract

Introduction

Doppler studies of blood flow through the fetal cardiac outflow tracts provide an insight into changes under various pathological conditions and can be helpful in the prenatal assessment of fetuses with congenital cardiac defects, intrauterine growth retardation (IUGR), anemia and other non-structural abnormalities [1–7]. Traditional nomograms for pulsed-wave Doppler (PWD) indices at the level of semi-lunar valves have been established by a number of studies [8–16], which typically were based on gestational age (GA).

However, most of these reference ranges included short GA ranges or small sample sizes. What's more, one cannot precisely quantify the measurements that fall outside the normal percentile ranges, since the standard deviation (SD) does not change smoothly with advancing GA [17]. Moreover, comparisons between subgroups or of longitudinal cardiovascular changes would be extremely difficult and problematic using centiles [18]. The Z-score, expressed as the number of SDs a measurement deviates from the mean, can be a more effective alternative. It offers simpler interpretation of abnormal values [18, 19] and allows comparisons between subgroups by adjusting values to the mean and variation at any given GA, or any direct parameter of fetal size [18, 20].

In a literature review, we found only two studies proposing Z-score models [8, 21] for Doppler indices of the fetal cardiac outflow tracts, focusing on peak systolic velocity (PSV), velocity–time integral (VTI) and cardiac output (CO). Therefore, we conducted this study to establish Z-score reference ranges for several PWD parameters of the fetal cardiac outflow tracts in the second half of pregnancy, based on a large number of cases and appropriate methodology. As the fetal body size can vary substantially by late

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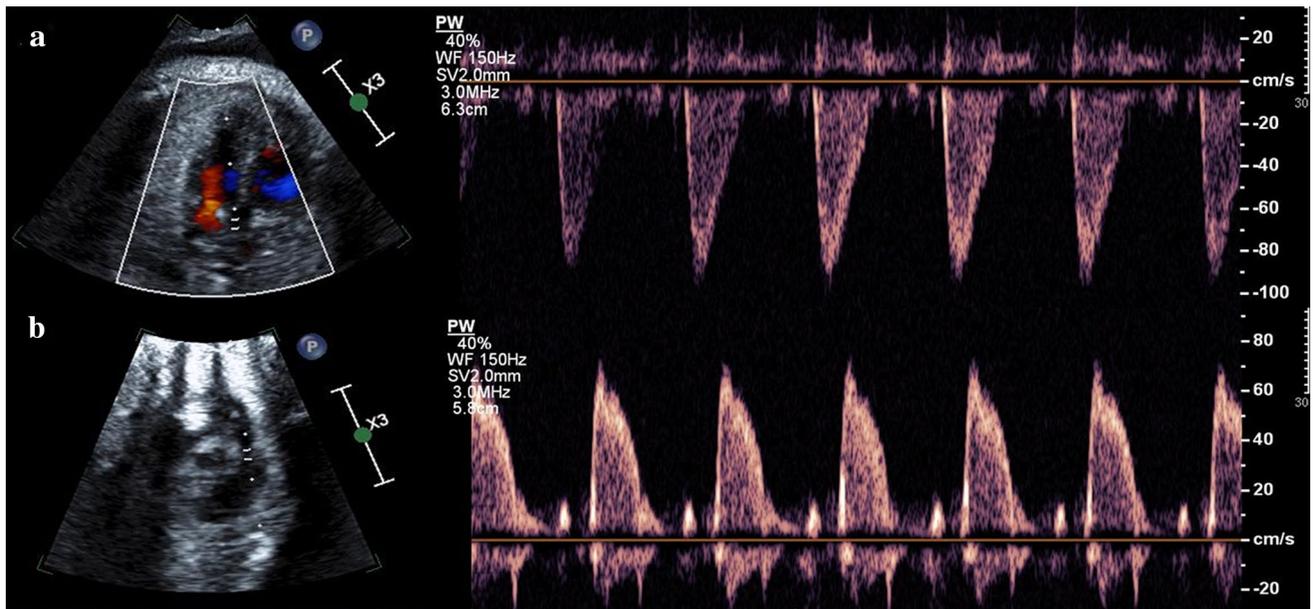


Fig. 1 **a** Blood flow velocity waveforms at the level of the aortic valve at 32 weeks of gestation. **b** Blood flow velocity waveforms across the pulmonary valve at 25 weeks' gestation

pregnancy, we also constructed normal ranges with respect to estimated fetal weight (EFW), which, to the best of our knowledge, have not been published before.

Materials and methods

Patients

This prospective cross-sectional study was conducted at Sir Run Run Shaw Hospital, Zhejiang University School of Medicine, between March 2016 and January 2017. The study protocol was approved by the local institute's Ethics Committee. All participants provided their written, informed consent to participate in the study. Healthy pregnant women referred for routine antenatal ultrasound examination were consecutively recruited in this study. All were singleton pregnancies from 18 to 40 weeks' gestation that met the following criteria: low-risk pregnancy without known obstetric or medical complications (pre-eclampsia, anemia, diabetes mellitus, and chronic hypertension, etc.); GA calculated from the menstrual history and confirmed by fetal biometric measurements at first trimester ultrasound [22]; absence of cardiac and extracardiac malformations detected on ultrasound; absence of amniotic fluid disorders. The exclusion criteria were as follows: chromosomal abnormalities (confirmed or suspected); abnormal intrauterine fetal growth; persistent non-sinus rhythm; abnormal placental function; abnormal neonatal outcome; inability to obtain adequate blood flow velocity waveforms of cardiac outflow tracts. All

the participants had postnatal echocardiography and neonatal clinical examination after birth to confirm normality. Each fetus was included only once in the data set.

Echocardiographic examinations

A systematic examination of the fetal heart was performed using a Philips EPIQ 7C (Philips, Andover, MA 01810, USA) ultrasound system with transducers of 5–1 MHz and 8–3 MHz by two investigators with extensive experience in fetal echocardiography (Bo Wen Zhao and Bei Wang).

Table 1 Clinical characteristics and pregnancy outcome of the study population

Parameter	Value
Clinical characteristics	
Maternal age (years)	29.2 ± 4.4
Maternal body mass index (kg/m ²)	23.1 ± 4.8
Nulliparous	319 (63)
Cigarette smoker	13 (2.6)
Pregnancy outcome	
GA at delivery (weeks)	39 ± 2
Cesarean section	158 (31.2)
Birth weight (g)	3341 ± 356
Birth-weight centile	51 ± 32
5 min Apgar score	9 ± 2

Data given as mean ± SD or n(%)

GA gestational age

Two-dimensional assessment of the intra-cardiac anatomy and great vessels was followed by Doppler flow interrogation, according to the guidelines of the International Society of Ultrasound in Obstetrics and Gynecology [23]. Flow velocity waveforms through the pulmonary and aortic valves were recorded from the left and right ventricular outflow tract views, respectively (Fig. 1). The sample volume was set at 2 mm and placed slightly distal to the pulmonary and aortic valve leaflets in the center of the vessel [11, 12]. The angle between the ultrasound beam and the direction of blood flow was $< 10^\circ$ [9]. Angle correction was not used. Pulse repetition frequency ranged from 2.5 to 5 MHz. The high-pass filter was set at 100–200 Hz. Doppler tracings were made in the absence of fetal breathing movements. The fetal heart rate (FHR) was within the normal range (mean 145 beats/min, SD 7.8 beats/min). At least five consecutive

uniform Doppler velocity waveforms were obtained, and the image was frozen and recorded on videotape. All Doppler recordings were analyzed by one of the authors (Li Zhou). The following indices were measured and averaged over three best cardiac cycles: peak systolic velocity (PSV), velocity time integral (VTI: defined as the area under the velocity spectral envelope), ejection time (ET: defined as the whole ejection time), and acceleration time (AT: measured as the time from the onset of ejection to peak velocity). The AT/ET ratio was also calculated. The opening and closure of the semilunar valves were identified by ‘valve clicks’ while measuring the time intervals [24]. Two-dimensional measurements of the pulmonary and aortic valve were also acquired during the end-systolic phase to compute cardiac outputs [8]. The left and right ventricular cardiac outputs (LVCO and RVCO) were calculated separately as: $VTI \times \pi$

Table 2 Regression analysis of PWD indices of fetal cardiac outflow tracts based on GA and EFW

Parameter	Mean	SD	r(P)	F test
GA (weeks)				
<i>Ascending aorta</i>				
PSV (cm/s)	38.089 + 1.463GA	4.227 + 0.239GA	0.626 (<0.0001)	356.030
VTI (cm)	-4.618 + 0.742GA - 0.00825GA ²	0.0361 + 0.0635GA	0.653 (<0.0001)	255.135
CO (mL/min)	24.238 - 10.563GA + 0.628GA ²	-125.7828 + 6.6236GA	0.895 (<0.0001)	2031.603
AT (ms)	27.745 + 1.0132GA	4.690 + 0.0854GA	0.662 (<0.0001)	407.732
ET (ms)	164.009 + 0.504GA	3.841 + 0.0695GA	0.483 (<0.0001)	152.626
AT/ET ratio	0.182 + 0.00470GA	0.0292 + 0.0000912GA	0.679 (<0.0001)	430.291
<i>Pulmonary artery</i>				
PSV (cm/s)	34.10 + 1.393GA	5.634 + 0.121GA	0.694 (<0.0001)	478.472
VTI (cm)	3.496 + 0.181GA	0.716 + 0.0164GA	0.692 (<0.0001)	483.296
CO (mL/min)	65.325 - 16.760GA + 0.932GA ²	-100.572 + 6.216GA	0.928 (<0.0001)	2260.228
AT (ms)	20.644 + 1.054GA	0.884 + 0.147GA	0.776 (<0.0001)	840.467
ET (ms)	162.328 + 0.397GA	2.742 + 0.0671GA	0.476 (<0.0001)	162.175
AT/ET ratio	0.1383 + 0.00537GA	0.00803 + 0.000748GA	0.734 (<0.0001)	631.624
EFW(g)				
<i>Ascending aorta</i>				
PSV (cm/s)	60.131 + 0.02163EFW - 0.00000370EFW ²	8.8807 + 0.00148EFW	0.581 (<0.0001)	184.283
VTI (cm)	12.533 + 491624.249EFW ⁻² - 3441.163EFW ⁻¹	1.273 + 0.000388EFW	0.598 (<0.0001)	211.432
CO (mL/min)	-7.986 + 0.223EFW - 0.0000185EFW ²	-2.6327 + 0.04724EFW	0.896 (<0.0001)	1804.306
AT (ms)	43.6494 + 0.0136 EFW - 0.00000215EFW ²	6.909 + 0.000148EFW	0.624 (<0.0001)	241.840
ET (ms)	172.605 + 0.00539EFW - 6.52E-07EFW ²	5.378 + 0.000288EFW	0.476 (<0.0001)	165.267
AT/ET ratio	0.254 + 0.0000665EFW - 1.08E-10EFW ²	0.0333 - 1.02E-06 EFW	0.633 (<0.0001)	270.710
<i>Pulmonary artery</i>				
PSV (cm/s)	57.0767 + 0.0166EFW - 0.00000234EFW ²	8.591 + 0.000359EFW	0.667 (<0.0001)	272.365
VTI (cm)	6.414 + 0.00232EFW - 0.000000356EFW ²	1.058 + 0.0000951EFW	0.652 (<0.0001)	253.202
CO (mL/min)	-6.822 + 0.334EFW - 0.0000308EFW ²	9.379 + 0.0488EFW	0.929 (<0.0001)	2417.690
AT (ms)	37.647 + 0.0134EFW - 0.00000203EFW ²	3.7666 + 0.0009622EFW	0.739 (<0.0001)	367.844
ET (ms)	168.388 + 0.00592EFW - 0.00000100EFW ²	4.177 + 0.000364EFW	0.438 (<0.0001)	171.343
AT/ET ratio	0.225 + 0.0000675EFW - 1.01E-08EFW ²	0.0228 + 4.71E-06EFW	0.701 (<0.0001)	281.699

AT acceleration time; CO cardiac output; EFW estimated fetal weight; ET ejection time; GA gestational age; PSV peak systolic velocity; PWD pulsed-wave Doppler; VTI velocity time integral

(aortic or pulmonary valve diameter/2)² × FHR. In each case, fetal biometric variables were measured on the same hospital visit. EFW was calculated according to the method of Hadlock et al. [25]. The GA was recorded in exact weeks.

To assess intra- and inter-observer variability, all measurements were taken in 30 randomly selected datasets by two independent investigators, who were blinded to each other's results. Each observer repeated the measurements on two separate occasions.

Statistical analysis

IBM SPSS package 22.0 (SPSS, Inc., Chicago, IL, USA) was used to perform the statistical analyses. The statistical model described by Royston et al. [26] and DeVore [27] was used to estimate Z-score reference intervals. First, regression analyses of the means and SDs of all measurements were performed using GA and EFW as the independent variables. The best-fitted regression equations were selected among linear, quadratic, cubic, logarithmic and fractional models. When two different regression curves achieved similar model fit, the one with lower residual SD and fewer terms would be preferable. The best-fitted models for the SD were derived from regression of scaled absolute residuals, which was determined as $1.25 \times$ absolute (measured value-predicted value). Second, the Z-score was calculated as (observed value-estimated mean value)/estimated SD to examine the model fit. The Z-scores were plotted against the independent variables to show whether they were randomly distributed above and below zero. Normality of Z-scores was validated using the Shapiro–Wilk W test and Q-Q plots. Statistical transformation was required only if the data were not distributed normally. Finally, we constructed Z-score models based on EFW and GA, with Z-scores of ± 2 representing two SDs above or below the mean (corresponding to the 97.7th percentile and the 2.3rd percentile).

Inter- and intra-observer variability was tested using intraclass correlation coefficients (ICC), with their respective 95% confidence intervals. Pearson's correlation coefficient was used to explore the correlations between PWD parameters and FHR. Statistical significance was defined as $P < 0.05$.

Results

A total of 525 singleton pregnancies were recruited for this study. Among these cases, 19 were excluded for the following reasons: inadequate images due to fetal position, movement, breathing, and maternal obesity ($n = 9$), cardiac abnormalities (left superior vena cava to coronary sinus, $n = 1$; atrial septal defect, $n = 3$, all detected postnatally), abnormal intrauterine fetal growth ($n = 4$), and abnormal neonatal

outcome ($n = 2$). Overall, 506 (96.4%) data sets with successful measurements were used for the final statistical analysis. The fetuses were scanned at a median GA of 28^{+5} (range,

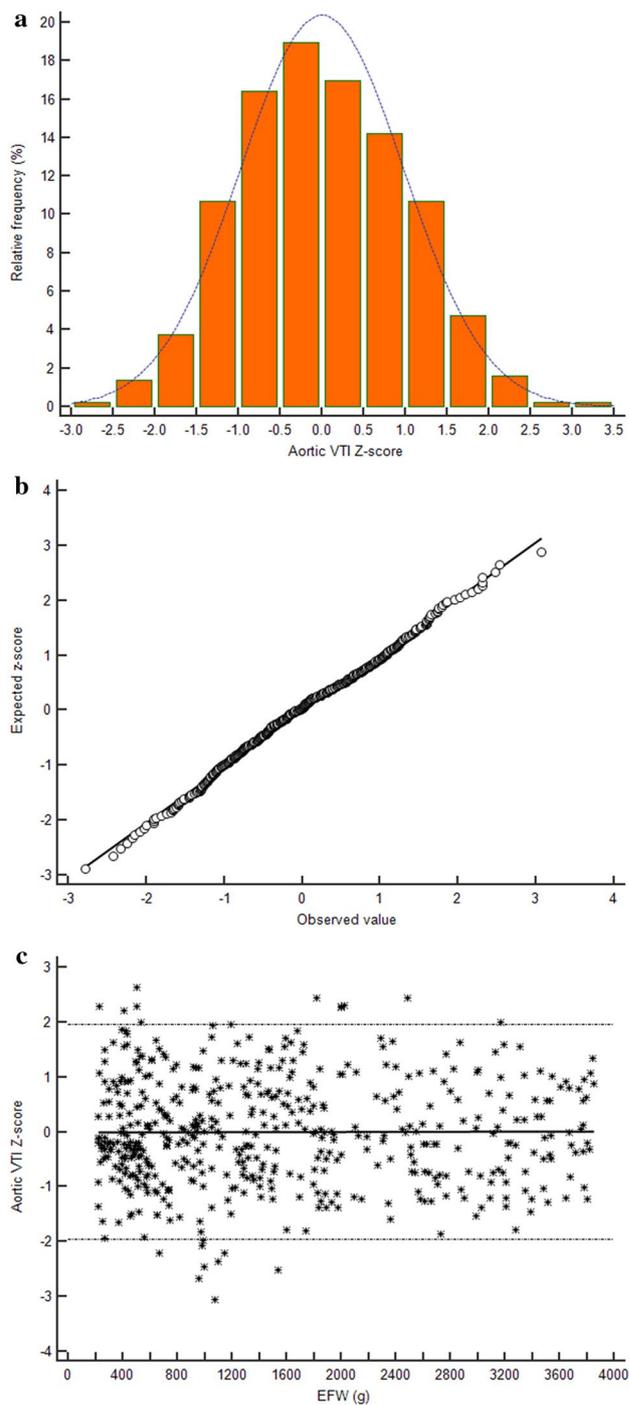


Fig. 2 **a** The frequency of the Z-score with an approximately normal distribution. **b** Normal plot of Z-scores from the fitted model for aortic VTI based on EFW. **c** A scatterplot of Z-scores for aortic VTI against EFW, with tramlines at 1.96, 0 and -1.96 . EFW, estimated fetal weight; VTI, velocity time integral

18⁺¹–40⁺⁵) weeks. Clinical characteristics and pregnancy outcome of the study population are shown in Table 1.

All PWD indices showed a progressive increase with GA and EFW, and good correlations were found between the studied parameters and independent variables (GA and EFW). A linear-quadratic regression model was the best description of the mean and SD of most parameters, with the exception of the mean aortic VTI against EFW, which

was best modeled by a fractional polynomial. The best-fitted regression equations, correlation coefficients and P values of the mean and SD of each parameter are presented in Table 2. These statistical models were validated by developing a Z-score for each variable. For example, the Z-score of aortic VTI based on EFW was calculated as (observed value – predicted mean)/predicted SD) according to the formulae presented in Table 2. The plot of VTI Z-scores against

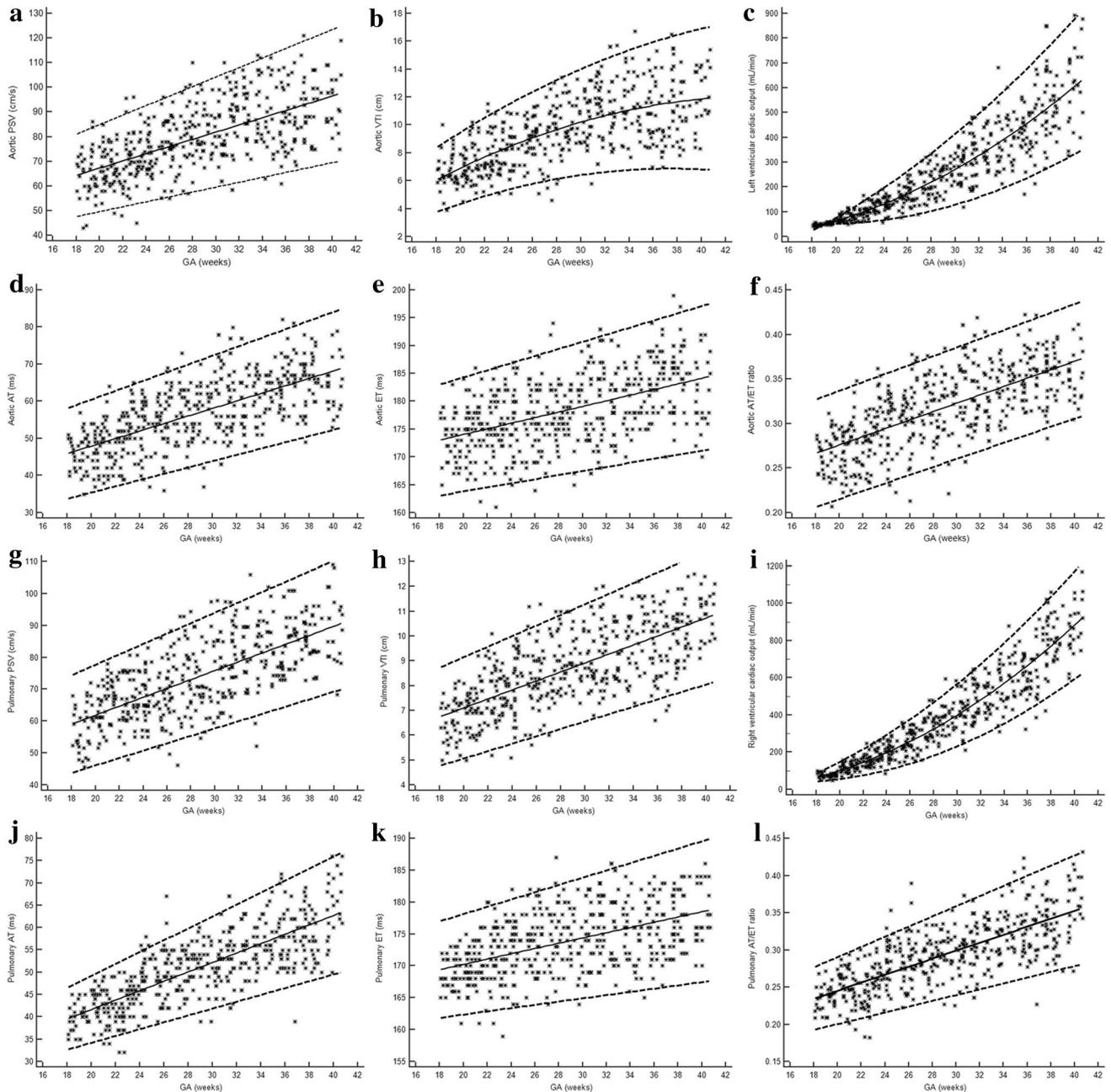


Fig. 3 Scatterplots of PWD indices of the left (a–f) and right (g–l) cardiac outflow tracts in normal fetuses based on gestational age (GA). The mean is indicated by a solid black line, the Z-score ± 2 is

indicated by broken black lines. AT, acceleration time; CO, cardiac output; ET, ejection time; PSV, peak systolic velocity; PWD, pulsed-wave Doppler; VTI, velocity time integral

EFW showed no obvious patterns or trends. Additionally, the number of Z-scores that fell outside the range of $\pm 2SD$ did not exceed the expected 10% of the values (Fig. 2). The normalcy of the Z-scores was evident in a Q-Q plot. The Shapiro–Wilk W test demonstrated normality in the distribution ($P=0.3215$). Thus, we concluded that this model was reasonable. All regression statistical models were found to fit the data well by assessing the validity of Z-scores. Figures 3 and 4 showed scatterplots of PWD indices against GA and

EFW, with Z-score boundaries and the mean curves superimposed. Reference ranges for these parameters by percentile as a function of GA and EFW are displayed in Tables 3, 4, 5 and 6, respectively. Table 7 summarized the available regression equations for fetal PWD measurements of the cardiac outflow tracts in the literature.

This is an example of Z-score calculation for aortic VTI based on EFW: For a fetus with an EFW of 1038 g, aortic VTI is measured at 15.1 cm. From Table 2, the predicted

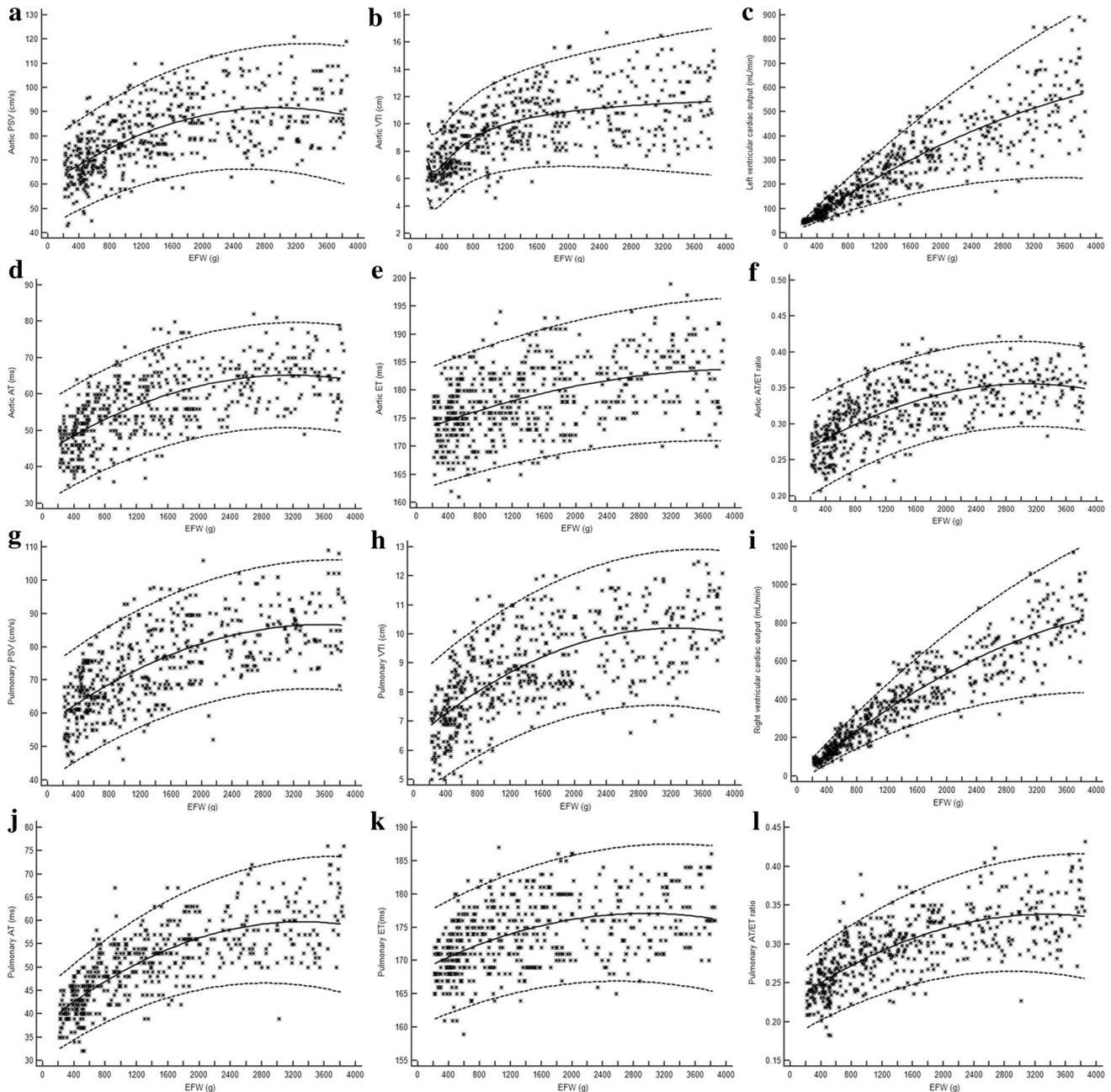


Fig. 4 Scatterplots of PWD indices of the left (a–f) and right (g–l) cardiac outflow tracts in normal fetuses based on estimated fetal weight (EFW). The mean is indicated by a solid black line, the

Z-score ± 2 is indicated by broken black lines. AT, acceleration time; CO, cardiac output; ET, ejection time; PSV, peak systolic velocity; PWD, pulsed-wave Doppler; VTI, velocity time integral

Table 3 Reference ranges of fetal PWD indices of left cardiac outflow tract based on GA

GA (weeks)	PSV (cm/s)			VTI (cm)			CO (mL/min)			AT (ms)			ET (ms)			AT/ET ratio		
	2.5th	Mean	97.5th	2.5th	Mean	97.5th	2.5th	Mean	97.5th	2.5th	Mean	97.5th	2.5th	Mean	97.5th	2.5th	Mean	97.5th
18	47.7	64.4	81.1	3.76	6.07	8.38	50.4	37.5	24.7	33.8	46.0	58.2	163.1	173.1	183.1	0.206	0.267	0.327
19	48.7	65.9	83.1	4.07	6.51	8.94	50.1	50.2	50.3	34.6	47.0	59.4	163.5	173.6	183.7	0.211	0.272	0.332
20	49.7	67.4	85.0	4.37	6.93	9.49	51.0	64.1	77.3	35.5	48.0	60.5	163.8	174.1	184.3	0.215	0.276	0.337
21	50.7	68.8	86.9	4.65	7.33	10.02	53.2	79.3	105.4	36.3	49.0	61.7	164.2	174.6	185.0	0.220	0.281	0.342
22	51.7	70.3	88.9	4.91	7.72	10.53	56.7	95.8	134.8	37.2	50.0	62.9	164.6	175.1	185.6	0.225	0.286	0.347
23	52.7	71.7	90.8	5.16	8.09	11.02	61.4	113.5	165.5	38.0	51.0	64.1	164.9	175.6	186.3	0.229	0.290	0.352
24	53.7	73.2	92.7	5.39	8.44	11.50	67.4	132.4	197.4	38.9	52.1	65.3	165.3	176.1	186.9	0.234	0.295	0.357
25	54.7	74.7	94.7	5.60	8.78	11.97	74.6	152.6	230.6	39.7	53.1	66.5	165.7	176.6	187.5	0.238	0.300	0.361
26	55.7	76.1	96.6	5.80	9.10	12.41	83.1	174.1	265.1	40.5	54.1	67.6	166.0	177.1	188.2	0.243	0.304	0.366
27	56.7	77.6	98.5	5.98	9.41	12.84	92.8	196.8	300.8	41.4	55.1	68.8	166.4	177.6	188.8	0.247	0.309	0.371
28	57.7	79.1	100.5	6.14	9.70	13.25	103.8	220.8	337.7	42.2	56.1	70.0	166.8	178.1	189.5	0.252	0.314	0.376
29	58.6	80.5	102.4	6.29	9.97	13.65	116.0	246.0	375.9	43.1	57.1	71.2	167.1	178.6	190.1	0.256	0.319	0.381
30	59.6	82.0	104.3	6.42	10.23	14.03	129.5	272.5	415.4	43.9	58.1	72.4	167.5	179.1	190.7	0.261	0.323	0.386
31	60.6	83.4	106.3	6.53	10.46	14.39	144.3	300.2	456.1	44.8	59.2	73.5	167.9	179.6	191.4	0.265	0.328	0.391
32	61.6	84.9	108.2	6.63	10.69	14.74	160.3	329.2	498.1	45.6	60.2	74.7	168.3	180.1	192.0	0.270	0.333	0.396
33	62.6	86.4	110.1	6.71	10.89	15.07	177.6	359.5	541.3	46.5	61.2	75.9	168.6	180.6	192.7	0.274	0.337	0.400
34	63.6	87.8	112.1	6.78	11.08	15.39	196.1	391.0	585.8	47.3	62.2	77.1	169.0	181.1	193.3	0.279	0.342	0.405
35	64.6	89.3	114.0	6.83	11.26	15.68	215.9	423.7	631.6	48.2	63.2	78.3	169.4	181.7	194.0	0.283	0.347	0.410
36	65.6	90.8	115.9	6.86	11.41	15.96	236.9	457.7	678.6	49.0	64.2	79.4	169.7	182.2	194.6	0.288	0.351	0.415
37	66.6	92.2	117.9	6.88	11.55	16.23	259.2	493.0	726.8	49.8	65.2	80.6	170.1	182.7	195.2	0.292	0.356	0.420
38	67.6	93.7	119.8	6.87	11.68	16.48	282.8	529.5	776.3	50.7	66.2	81.8	170.5	183.2	195.9	0.297	0.361	0.425
39	68.6	95.2	121.7	6.86	11.78	16.71	307.6	567.3	827.1	51.5	67.3	83.0	170.8	183.7	196.5	0.301	0.366	0.430
40	69.6	96.6	123.6	6.82	11.87	16.92	333.6	606.4	879.1	52.4	68.3	84.2	171.2	184.2	197.2	0.306	0.370	0.435

AT acceleration time; CO cardiac output; ET ejection time ;GA gestational age; PSV peak systolic velocity; PWD pulsed-wave Doppler; VTI velocity time integral

Table 4 Reference ranges of fetal PWD indices of right cardiac outflow tract based on GA

GA (weeks)	PSV (cm/s)			VTI (cm)			CO (mL/min)			AT (ms)			ET (ms)			AT/ET ratio		
	2.5th	Mean	97.5th	2.5th	Mean	97.5th	2.5th	Mean	97.5th	2.5th	Mean	97.5th	2.5th	Mean	97.5th	2.5th	Mean	97.5th
18	43.9	59.2	74.5	4.78	6.76	8.74	43.5	65.7	87.8	32.7	39.6	46.5	161.9	169.5	177.0	0.193	0.235	0.277
19	45.0	60.6	76.1	4.93	6.94	8.95	49.0	83.4	117.7	33.5	40.7	47.9	162.1	169.9	177.6	0.197	0.240	0.284
20	46.2	62.0	77.8	5.08	7.12	9.16	56.4	103.0	149.5	34.2	41.7	49.2	162.4	170.3	178.2	0.201	0.246	0.291
21	47.3	63.4	79.4	5.22	7.30	9.38	65.7	124.4	183.2	35.0	42.8	50.5	162.6	170.7	178.7	0.204	0.251	0.298
22	48.5	64.8	81.0	5.37	7.48	9.59	76.9	147.8	218.7	35.8	43.8	51.9	162.9	171.1	179.3	0.208	0.256	0.304
23	49.7	66.2	82.7	5.52	7.66	9.80	89.9	172.9	256.0	36.5	44.9	53.2	163.1	171.5	179.9	0.212	0.262	0.311
24	50.8	67.5	84.3	5.67	7.84	10.02	104.7	200.0	295.3	37.3	45.9	54.6	163.4	171.9	180.4	0.216	0.267	0.318
25	52.0	68.9	85.9	5.82	8.02	10.23	121.5	228.9	336.4	38.1	47.0	55.9	163.7	172.3	181.0	0.220	0.272	0.325
26	53.1	70.3	87.6	5.97	8.21	10.44	140.1	259.7	379.3	38.8	48.0	57.2	163.9	172.8	181.6	0.224	0.278	0.332
27	54.3	71.7	89.2	6.12	8.39	10.66	160.5	292.3	424.1	39.6	49.1	58.6	164.2	173.2	182.2	0.228	0.283	0.339
28	55.4	73.1	90.8	6.27	8.57	10.87	182.8	326.8	470.8	40.4	50.1	59.9	164.4	173.6	182.7	0.232	0.289	0.345
29	56.6	74.5	92.4	6.42	8.75	11.08	207.0	363.2	519.4	41.1	51.2	61.3	164.7	174.0	183.3	0.236	0.294	0.352
30	57.7	75.9	94.1	6.57	8.93	11.29	233.1	401.4	569.8	41.9	52.3	62.6	164.9	174.4	183.9	0.240	0.299	0.359
31	58.9	77.3	95.7	6.72	9.11	11.51	261.0	441.5	622.1	42.7	53.3	63.9	165.2	174.8	184.4	0.243	0.305	0.366
32	60.1	78.7	97.3	6.86	9.29	11.72	290.8	483.5	676.2	43.4	54.4	65.3	165.4	175.2	185.0	0.247	0.310	0.373
33	61.2	80.1	99.0	7.01	9.47	11.93	322.4	527.3	732.2	44.2	55.4	66.6	165.7	175.6	185.6	0.251	0.315	0.380
34	62.4	81.5	100.6	7.16	9.66	12.15	355.9	573.0	790.1	45.0	56.5	68.0	165.9	176.1	186.2	0.255	0.321	0.386
35	63.5	82.9	102.2	7.31	9.84	12.36	391.3	620.6	849.9	45.7	57.5	69.3	166.2	176.5	186.7	0.259	0.326	0.393
36	64.7	84.3	103.9	7.46	10.02	12.57	428.6	670.0	911.5	46.5	58.6	70.6	166.5	176.9	187.3	0.263	0.331	0.400
37	65.8	85.7	105.5	7.61	10.20	12.79	467.7	721.3	974.9	47.3	59.6	72.0	166.7	177.3	187.9	0.267	0.337	0.407
38	67.0	87.1	107.1	7.76	10.38	13.00	508.6	774.5	1040.3	48.0	60.7	73.3	167.0	177.7	188.4	0.271	0.342	0.414
39	68.1	88.4	108.8	7.91	10.56	13.21	551.5	829.5	1107.5	48.8	61.7	74.7	167.2	178.1	189.0	0.275	0.348	0.421
40	69.3	89.8	110.4	8.06	10.74	13.43	596.2	886.3	1176.5	49.6	62.8	76.0	167.5	178.5	189.6	0.279	0.353	0.427

AT acceleration time; CO cardiac output; ET ejection time; GA gestational age; PSV peak systolic velocity; PWD pulsed-wave Doppler; VTI velocity time integral

Table 5 Reference ranges of fetal PWD indices of left cardiac outflow tract based on EFW

EFW (g)	PSV (cm/s)			VTI (cm)			CO (mL/min)			AT (ms)			ET (ms)			AT/ET ratio		
	2.5th	Mean	97.5th	2.5th	Mean	97.5th	2.5th	Mean	97.5th	2.5th	Mean	97.5th	2.5th	Mean	97.5th	2.5th	Mean	97.5th
200	46.3	64.3	82.3	4.97	7.62	10.26	22.5	35.9	49.3	32.7	46.3	59.9	163.0	173.7	184.3	0.202	0.267	0.332
300	48.0	66.3	84.6	3.80	6.52	9.25	34.7	57.3	79.9	33.9	47.5	61.2	163.5	174.2	184.9	0.208	0.273	0.338
400	49.6	68.2	86.8	4.20	7.00	9.80	46.4	78.3	110.2	35.1	48.8	62.4	163.9	174.7	185.4	0.214	0.279	0.343
500	51.2	70.0	88.9	4.74	7.62	10.49	57.8	99.0	140.1	36.2	49.9	63.6	164.3	175.1	186.0	0.220	0.285	0.349
600	52.6	71.8	90.9	5.21	8.16	11.11	68.9	119.3	169.7	37.3	51.1	64.8	164.7	175.6	186.5	0.226	0.290	0.354
700	54.0	73.5	92.9	5.59	8.62	11.65	79.5	139.2	198.8	38.4	52.1	65.9	165.1	176.1	187.0	0.231	0.295	0.359
800	55.3	75.1	94.8	5.90	9.00	12.10	89.8	158.7	227.6	39.4	53.2	67.0	165.5	176.5	187.5	0.237	0.300	0.364
900	56.6	76.6	96.6	6.14	9.32	12.50	99.7	177.9	256.1	40.4	54.2	68.0	165.9	176.9	188.0	0.242	0.305	0.369
1000	57.8	78.1	98.4	6.33	9.58	12.84	109.3	196.7	284.1	41.3	55.1	69.0	166.2	177.4	188.5	0.246	0.310	0.373
1100	58.8	79.4	100.0	6.48	9.81	13.14	118.4	215.1	311.8	42.2	56.0	69.9	166.6	177.8	188.9	0.251	0.314	0.377
1200	59.9	80.8	101.7	6.60	10.01	13.41	127.2	233.2	339.1	43.0	56.9	70.8	166.9	178.1	189.4	0.255	0.318	0.381
1300	60.8	82.0	103.2	6.69	10.18	13.66	135.7	250.9	366.1	43.8	57.7	71.7	167.2	178.5	189.8	0.259	0.322	0.385
1400	61.7	83.2	104.6	6.77	10.33	13.88	143.8	268.2	392.7	44.6	58.5	72.5	167.6	178.9	190.2	0.263	0.326	0.388
1500	62.5	84.2	106.0	6.82	10.46	14.09	151.5	285.2	418.9	45.3	59.3	73.2	167.8	179.2	190.6	0.267	0.329	0.391
1600	63.2	85.3	107.3	6.86	10.57	14.29	158.8	301.8	444.8	46.0	60.0	74.0	168.1	179.6	191.0	0.270	0.332	0.394
1700	63.9	86.2	108.6	6.89	10.68	14.47	165.7	318.0	470.2	46.6	60.6	74.7	168.4	179.9	191.4	0.273	0.335	0.397
1800	64.4	87.1	109.7	6.91	10.77	14.64	172.3	333.8	495.3	47.2	61.2	75.3	168.7	180.2	191.8	0.276	0.338	0.400
1900	64.9	87.9	110.8	6.92	10.86	14.80	178.5	349.3	520.1	47.7	61.8	75.9	168.9	180.5	192.1	0.279	0.341	0.402
2000	65.4	88.6	111.8	6.92	10.94	14.95	184.4	364.4	544.4	48.2	62.3	76.4	169.1	180.8	192.5	0.282	0.343	0.404
2100	65.7	89.2	112.7	6.91	11.01	15.10	189.9	379.2	568.4	48.7	62.8	77.0	169.3	181.1	192.8	0.284	0.345	0.406
2200	66.0	89.8	113.6	6.90	11.07	15.24	195.0	393.5	592.1	49.1	63.2	77.4	169.5	181.3	193.1	0.286	0.347	0.408
2300	66.2	90.3	114.4	6.89	11.13	15.37	199.7	407.5	615.3	49.4	63.6	77.8	169.7	181.6	193.4	0.288	0.349	0.410
2400	66.3	90.7	115.1	6.87	11.18	15.50	204.1	421.2	638.2	49.8	64.0	78.2	169.9	181.8	193.7	0.290	0.350	0.411
2500	66.4	91.1	115.7	6.84	11.23	15.63	208.1	434.4	660.7	50.0	64.3	78.6	170.1	182.0	194.0	0.291	0.352	0.412
2600	66.4	91.3	116.3	6.81	11.28	15.75	211.7	447.3	682.9	50.3	64.6	78.9	170.2	182.2	194.2	0.292	0.353	0.413
2700	66.3	91.5	116.8	6.78	11.33	15.87	215.0	459.8	704.7	50.5	64.8	79.1	170.4	182.4	194.5	0.294	0.353	0.413
2800	66.1	91.7	117.2	6.74	11.37	15.99	217.9	472.0	726.1	50.6	65.0	79.3	170.5	182.6	194.7	0.294	0.354	0.414
2900	65.9	91.7	117.6	6.71	11.40	16.10	220.4	483.8	747.1	50.7	65.1	79.5	170.6	182.8	195.0	0.295	0.354	0.414
3000	65.6	91.7	117.8	6.67	11.44	16.22	222.6	495.2	767.8	50.8	65.2	79.6	170.7	182.9	195.2	0.295	0.355	0.414
3100	65.2	91.6	118.0	6.62	11.47	16.32	224.4	506.2	788.1	50.8	65.3	79.7	170.8	183.1	195.4	0.295	0.355	0.414
3200	64.7	91.4	118.1	6.58	11.51	16.43	225.8	516.9	808.0	50.8	65.3	79.7	170.9	183.2	195.5	0.295	0.354	0.413
3300	64.2	91.2	118.2	6.53	11.54	16.54	226.8	527.2	827.6	50.7	65.2	79.7	170.9	183.3	195.7	0.295	0.354	0.412
3400	63.6	90.9	118.2	6.48	11.56	16.64	227.5	537.1	846.8	50.6	65.1	79.7	171.0	183.4	195.9	0.294	0.353	0.412
3500	62.9	90.5	118.1	6.43	11.59	16.74	227.8	546.7	865.6	50.5	65.0	79.6	171.0	183.5	196.0	0.294	0.352	0.410
3600	62.2	90.0	117.9	6.38	11.61	16.85	227.7	555.9	884.1	50.3	64.9	79.4	171.0	183.6	196.2	0.293	0.351	0.409
3700	61.3	89.5	117.6	6.33	11.64	16.95	227.3	564.7	902.2	50.0	64.7	79.3	171.0	183.7	196.3	0.292	0.350	0.407

Table 5 (continued)

EFW (g)	PSV (cm/s)			VTI (cm)			CO (mL/min)			AT (ms)			ET (ms)			AT/ET ratio		
	2.5th	Mean	97.5th	2.5th	Mean	97.5th	2.5th	Mean	97.5th	2.5th	Mean	97.5th	2.5th	Mean	97.5th	2.5th	Mean	97.5th
3800	60.4	88.9	117.3	6.28	11.66	17.04	226.5	573.2	919.9	49.8	64.4	79.1	171.0	183.7	196.4	0.290	0.348	0.406
3900	59.4	88.2	116.9	6.22	11.68	17.14	225.3	581.3	937.2	49.4	64.1	78.8	171.0	183.7	196.5	0.289	0.346	0.404

AT acceleration time; CO cardiac output; EFW estimated fetal weight; ET ejection time; PSV peak systolic velocity; PWD pulsed-wave Doppler; VTI velocity time integral

value of VTI = $12.533 + 491624.249 \times 1038^{-2} - 3441.163 \times 1038^{-1} = 9.7$ cm; predicted SD of VTI = $1.273 + 0.000388 \times 1038 = 1.68$, and Z-score = $(15.1 - 9.7) / 1.68 = 3.21$. Therefore, this fetus had an increased aortic VTI at 3.21 SD above the predicted mean for a fetus of 1038 g.

Results of the reproducibility study are presented in Table 8, and good reliability was observed ($P < 0.0001$). Table 9 shows correlations between each PWD parameter and FHR. FHR ranged between 122 and 162 beats/min and decreased significantly with advancing GA ($P < 0.0001$). Within the normal physiological range, there was a weak but significant inverse relationship between the time intervals (ET and AT) and FHR in both outflows (right: $r = -0.144, -0.135$; left: $r = -0.119, -0.169$, respectively, all $P < 0.01$). However, no association was found between volume blood flow (PSV, VTI and CO) and FHR (right: $P = 0.926, 0.489, 0.291$; left: $P = 0.591, 0.638, 0.397$, respectively).

Discussion

The present study established Z-score models for PWD parameters of the cardiac outflow tracts in normal fetuses, based on not only GA but also EFW. To the best of our knowledge, normal ranges in relation to EFW have not been reported before. We provided a relatively large sample (506 cases) of PWD measurements over a wide range of GAs, from the 18th week of pregnancy to term. Additionally, these Z-scores were carefully examined to ascertain the normality.

Our results confirmed previous data showing a strong positive correlation between PWD parameters and GA. We also performed further regression analysis of the SD separately, which is essential for Z-score assessment [26]. As shown in Table 4, the fetal PSV is the best-studied PWD parameter in the literature. Compatible with most previous studies [9, 10, 12, 13], our results developed a best-fitted linear regression relationship for the mean PSV against GA. However, Gagnon et al. [8] provided a cubic regression equation for the MPA PSV, and a quadratic regression equation for the ascending aortic PSV, as well as Z-scores. In another study of fetal MPA PSV [14], a best-fitted cubic mean and quadratic SD was produced. With regard to the fetal MPA VTI, our study as well as previous results [8, 13] have consistently developed linear growth curves. Whereas, the ascending aortic VTI in relation to GA was best fitted by a second-order polynomial, similar to what Gagnon et al. [8] observed. Concerning the fetal CO, we found only three studies [8, 16, 21] reporting reference values. Gagnon et al. [8] and Mielke and Benda [16], provided quadratic and cubic regression equations on a log scale for the mean CO against GA, respectively. In a recent study [21], fractional polynomials with log transformation were the best-fitting curves

Table 6 Reference ranges of fetal PWD indices of right cardiac outflow tract based on EFW

EFW (g)	PSV (cm/s)			VTI (cm)			CO (mL/min)			AT (ms)			ET (ms)			AT/ET ratio		
	2.5th	Mean	97.5th	2.5th	Mean	97.5th	2.5th	Mean	97.5th	2.5th	Mean	97.5th	2.5th	Mean	97.5th	2.5th	Mean	97.5th
200	43.3	60.3	77.3	4.75	6.86	8.98	21.3	58.8	96.3	32.5	40.2	48.0	161.2	169.5	177.9	0.192	0.238	0.285
300	44.8	61.9	78.9	4.95	7.08	9.21	43.6	90.7	137.8	33.5	41.5	49.4	161.7	170.1	178.5	0.197	0.245	0.292
400	46.2	63.4	80.5	5.14	7.29	9.44	65.3	122.0	178.6	34.5	42.7	50.8	162.1	170.6	179.1	0.202	0.251	0.299
500	47.6	64.8	82.0	5.32	7.49	9.65	86.4	152.6	218.9	35.5	43.8	52.2	162.5	171.1	179.6	0.207	0.256	0.306
600	49.0	66.2	83.5	5.49	7.68	9.87	106.9	182.7	258.5	36.4	45.0	53.5	163.0	171.6	180.2	0.212	0.262	0.312
700	50.2	67.6	84.9	5.66	7.87	10.07	126.7	212.1	297.5	37.3	46.0	54.7	163.3	172.0	180.7	0.216	0.268	0.319
800	51.5	68.9	86.3	5.82	8.05	10.27	146.0	240.9	335.9	38.2	47.1	56.0	163.7	172.5	181.2	0.221	0.273	0.325
900	52.7	70.2	87.6	5.98	8.22	10.46	164.6	269.1	373.6	39.0	48.1	57.1	164.1	172.9	181.7	0.225	0.278	0.331
1000	53.8	71.4	88.9	6.12	8.38	10.64	182.6	296.7	410.8	39.8	49.0	58.3	164.4	173.3	182.2	0.229	0.283	0.337
1100	54.9	72.6	90.2	6.26	8.54	10.82	200.0	323.7	447.3	40.5	49.9	59.4	164.7	173.7	182.6	0.233	0.287	0.342
1200	56.0	73.7	91.4	6.39	8.69	10.99	216.8	350.0	483.2	41.2	50.8	60.5	165.0	174.0	183.1	0.236	0.292	0.348
1300	57.0	74.8	92.5	6.52	8.83	11.15	233.0	375.7	518.5	41.8	51.6	61.5	165.3	174.4	183.5	0.239	0.296	0.353
1400	58.0	75.8	93.6	6.64	8.97	11.31	248.5	400.9	553.2	42.4	52.4	62.5	165.5	174.7	183.9	0.242	0.300	0.358
1500	58.9	76.8	94.7	6.75	9.10	11.45	263.4	425.4	587.3	43.0	53.2	63.4	165.7	175.0	184.2	0.245	0.304	0.362
1600	59.8	77.7	95.7	6.85	9.22	11.59	277.7	449.2	620.7	43.5	53.9	64.3	165.9	175.3	184.6	0.248	0.308	0.367
1700	60.6	78.6	96.7	6.95	9.34	11.73	291.4	472.5	653.6	44.0	54.6	65.2	166.1	175.5	184.9	0.251	0.311	0.371
1800	61.4	79.5	97.6	7.04	9.44	11.85	304.5	495.2	685.8	44.4	55.2	66.0	166.3	175.8	185.2	0.253	0.314	0.376
1900	62.1	80.3	98.4	7.12	9.55	11.97	317.0	517.2	717.4	44.8	55.8	66.8	166.5	176.0	185.5	0.255	0.317	0.380
2000	62.8	81.0	99.3	7.19	9.64	12.09	328.8	538.6	748.4	45.2	56.3	67.5	166.6	176.2	185.8	0.257	0.320	0.383
2100	63.4	81.7	100.0	7.26	9.73	12.19	340.0	559.4	778.8	45.5	56.8	68.2	166.7	176.4	186.1	0.259	0.323	0.387
2200	64.0	82.4	100.8	7.32	9.81	12.29	350.7	579.6	808.5	45.8	57.3	68.8	166.8	176.5	186.3	0.260	0.325	0.390
2300	64.5	83.0	101.5	7.37	9.88	12.38	360.7	599.2	837.6	46.0	57.7	69.5	166.8	176.7	186.5	0.262	0.328	0.393
2400	65.0	83.6	102.1	7.42	9.94	12.46	370.0	618.1	866.2	46.2	58.1	70.0	166.9	176.8	186.7	0.263	0.330	0.396
2500	65.5	84.1	102.7	7.46	10.00	12.54	378.8	636.4	894.1	46.4	58.5	70.6	166.9	176.9	186.9	0.264	0.331	0.399
2600	65.9	84.6	103.2	7.49	10.05	12.61	387.0	654.2	921.4	46.5	58.8	71.1	166.9	177.0	187.0	0.264	0.333	0.402
2700	66.2	85.0	103.7	7.52	10.10	12.67	394.5	671.3	948.0	46.6	59.0	71.5	166.9	177.0	187.1	0.265	0.334	0.404
2800	66.5	85.4	104.2	7.54	10.13	12.73	401.4	687.7	974.1	46.6	59.3	71.9	166.9	177.1	187.3	0.265	0.336	0.406
2900	66.8	85.7	104.6	7.55	10.16	12.78	407.7	703.6	999.5	46.6	59.5	72.3	166.8	177.1	187.3	0.265	0.337	0.408
3000	67.0	86.0	104.9	7.55	10.18	12.82	413.4	718.9	1024.4	46.6	59.6	72.6	166.8	177.1	187.4	0.265	0.338	0.410
3100	67.2	86.2	105.2	7.55	10.20	12.85	418.5	733.5	1048.6	46.5	59.7	72.9	166.7	177.1	187.5	0.265	0.338	0.412
3200	67.3	86.4	105.5	7.54	10.21	12.88	422.9	747.5	1072.2	46.4	59.8	73.2	166.6	177.0	187.5	0.265	0.339	0.413
3300	67.4	86.6	105.7	7.52	10.21	12.90	426.7	760.9	1095.1	46.2	59.8	73.4	166.4	177.0	187.5	0.264	0.339	0.414
3400	67.4	86.7	105.9	7.50	10.20	12.91	430.0	773.7	1117.5	46.0	59.8	73.6	166.3	176.9	187.5	0.263	0.339	0.415
3500	67.4	86.7	106.0	7.46	10.19	12.92	432.6	785.9	1139.2	45.7	59.7	73.7	166.1	176.8	187.5	0.262	0.339	0.416
3600	67.3	86.7	106.1	7.42	10.17	12.91	434.5	797.5	1160.4	45.4	59.6	73.8	165.9	176.7	187.4	0.261	0.339	0.416
3700	67.2	86.7	106.1	7.38	10.14	12.91	435.9	808.4	1180.9	45.1	59.5	73.8	165.7	176.5	187.4	0.259	0.338	0.417

Table 6 (continued)

EFW (g)	PSV (cm/s)			VTI (cm)			CO (mL/min)			AT (ms)			ET (ms)			AT/ET ratio		
	2.5th	Mean	97.5th	2.5th	Mean	97.5th	2.5th	Mean	97.5th	2.5th	Mean	97.5th	2.5th	Mean	97.5th	2.5th	Mean	97.5th
3800	67.1	86.6	106.1	7.33	10.11	12.89	436.7	818.7	1200.8	44.7	59.3	73.8	165.5	176.4	187.3	0.257	0.337	0.417
3900	66.9	86.4	106.0	7.26	10.07	12.87	436.8	828.4	1220.0	44.3	59.1	73.8	165.2	176.2	187.2	0.256	0.336	0.417

AT acceleration time; CO cardiac output; EFW estimated fetal weight; ET ejection time; PSV peak systolic velocity; PWD pulsed-wave Doppler; VTI velocity time integral

against GA. However, in our study, the best-fitted models for CO were quadratic regression equations. For fetal cardiac time intervals, our results confirmed previous data [9, 11–13] demonstrating a linear increase in AT and AT/ET ratio throughout gestation for both vessels. The assumption that a short AT or a lower AT/ET ratio correlates with a high arterial pressure has been used in the human fetus to indirectly determine arterial pressures in both outflows [12, 13, 28, 29]. Therefore, the significant changes of AT and the AT/ET ratio with advancing GA is probably the consequence of the decrease of the afterload to both ventricles during the second half of the pregnancy [9], namely the placental vascular resistance [30], cerebral vascular resistance [31] and pulmonary vascular resistance [13], as well as the improving myocardial function [32]. Regarding the fetal ET, we agreed with Chaoui et al. [13] that this parameter increases linearly with GA, with higher correlation coefficients, while Kurihara et al. [11] and Hata et al. [12] reported that ET of both the left and right sides remained constant across GA. Reference values for these PWD indices observed in our study were slightly different from those previously reported. These conflicting results could be due to the variations in sample sizes [8–10, 12, 13], gestational periods [15, 33] or methodology [11]. Besides, the differences in ethnicity and ultrasound equipment may also have contributed to the discrepancies.

In this study, normal ranges adjusted for EFW are also provided, with different patterns to those based on GA. The rationale for providing EFW-adjusted curves is that these measurements are size-dependent [21, 34]. Although it may be reasonable to compare to GA as a surrogate of fetal size in the second trimester, there is significant variability between GA and fetal size by late pregnancy [35]. The use of GA-adjusted reference ranges might lead to biased estimates, especially in fetuses with IUGR. Recent studies on IUGR fetuses have demonstrated that velocity-based indices such as CO values maintained at normal levels when adjusted for fetal weight [21], which was in contrast to what was observed previously [6]. Therefore, it is essential that these PWD measurements be normalized to fetal size to overcome this limitation. Among different surrogates of fetal size, we chose to adjust measurements by EFW, which theoretically better represents the fetal size. A quadratic regression model was the best description of the mean and SD of most parameters, with the exception of the mean aortic VTI, which was best modeled by a fractional polynomial.

With regard to the correlation between FHR and the studied parameters, no association could be found between FHR and the velocity-related parameters (PSV, VTI and CO, respectively), whereas a significant increase of ET and AT with reducing FHR was found due to the lengthening systolic phase of the cardiac cycle. This is in agreement with previous observations [9, 11, 14], indicating that

Table 7 Summary of the available reference values for PWD measurements of cardiac outflow tracts in normal fetuses

References	n	GA (weeks)		Regression equations
van der Mooren et al. [9]	40	18–38	Normal fetuses	Right: $PSV = 60.1 + 1.18(GA-30) + 10.4$; $AT = 51.7 + 1.96(GA-30) + 19.8$ Left: $PSV = 74.8 + 1.35(GA-30) + 21.8$; $AT = 60.6 + 1.37(GA-30) + 19.6$
Chaoui et al. [13]	86	18–38	Normal fetuses	Right: $PSV = 30.8 + 1.59GA$; $VTI = 2.39 + 0.23GA$; $ET = 162 + 0.5GA$; $AT = 11.6 + 0.62GA$; $AT/ET = 0.086 + 0.0027GA$
Mielke and Benda [14]	222	13–41	Normal fetuses	Right PSV: third-order polynomial regression based on GA (SD: quadratic regression)
Gagnon et al. [8]	104	18–39	Normal fetuses	Right: $PSV = -2.7E - 05GA^3 + 0.00244GA^2 - 0.0523GA + 0.904$ ($SD = 0.00260GA + 0.0379$) $VTI = 0.00298GA + 0.0140$ ($SD = 0.000565GA - 0.00105$) $\ln(CO) = -0.00381GA^2 + 0.339GA - 0.621$ ($SD = -0.00466GA + 0.356$) Left: $PSV = -0.000365GA^2 + 0.0457GA - 0.0413$ ($SD = 0.00403GA + 0.019$) $VTI = -0.000085GA^2 + 0.00777GA - 0.0397$ ($SD = 0.000629GA - 0.00126$) $\ln(CO) = -0.000365GA^2 + 0.0457GA - 0.0413$ ($SD = 0.00403GA + 0.0189$)
Kurihara et al. [11]	359	17–38	Normal fetuses	Right: $AT = 0.781GA + 21.661$; Left: $AT = 0.652GA + 35.620$ ET showed no correlations with GA in both ventricles
Rozmus-Warcholinska et al. [33]	198	11–14	Normal fetuses	Right: $PSV = 0.324CRL + 15.15$ Left: $PSV = 0.172CRL + 23.62$
Hata et al. [10]	57	16–40	Normal fetuses	Right: $PSV = 1.25GA + 32.6$ Left: $PSV = 1.47GA + 29$
Hata et al. [12]	88	16–41	Normal fetuses	Right: $PSV = 1.3GA + 28.7$; $AT = 1.04GA + 17.7$; $AT/ET = 0.005GA + 0.112$ Left: $PSV = 1.27GA + 34.9$; $AT = 0.615GA + 28.8$ $AT/ET = 0.003GA + 0.154$ No correlations between aortic and pulmonary ET and gestation
Mielke and Benda [16]	222	13–41	Normal fetuses	CO of both ventricles: Cubic regression on log scale based on GA (SD: quadratic regression)
Vimpeli et al. [15]	143	11–20	Normal fetuses	CO of both ventricles: Fractional polynomials in relation to GA with box-Cox transformation
Kiserud et al. [21]	212	18–41	Normal fetuses	Right: $\ln(CO) = 5.825 - 722.668GA^{-2} + 0.0000236GA^3$ ($SD = 0.276 - 0.000172GA$) Left: $\ln(CO) = 6.252 - 974.141GA^{-2} + 0.00008087GA^3$ ($SD = 0.257 + 0.00137GA$)

AT acceleration time; CO cardiac output; CRL crown–rump length; ET ejection time; GA gestational age; PWD pulsed-wave Doppler; SD standard deviation; PSV peak systolic velocity; VTI velocity time integral

Table 8 Intraclass correlation coefficients (ICC) for intra-observer and inter-observer agreement with their 95% CIs

Parameter	Ventricles	Intraobserver ICC (95% CI)	Interobserver ICC (95% CI)
PSV	Right	0.989 (0.968–0.996)	0.981 (0.945–0.994)
	Left	0.988 (0.963–0.996)	0.976 (0.930–0.992)
VTI	Right	0.973 (0.922–0.991)	0.965 (0.899–0.988)
	Left	0.952 (0.865–0.984)	0.893 (0.713–0.963)
CO	Right	0.911 (0.763–0.969)	0.891 (0.723–0.963)
	Left	0.851 (0.622–0.947)	0.837 (0.592–0.942)
AT	Right	0.968 (0.907–0.989)	0.868 (0.653–0.954)
	Left	0.956 (0.874–0.985)	0.905 (0.740–0.967)
ET	Right	0.965 (0.900–0.988)	0.950 (0.859–0.983)
	Left	0.976 (0.931–0.992)	0.974 (0.926–0.991)

AT acceleration time; CO cardiac output; ET ejection time; PSV peak systolic velocity; VTI velocity time integral

Table 9 Correlations between fetal heart rate (FHR) and PWD parameters

Parameter	Ventricles	FHR	<i>P</i>
PSV	Right	−0.004	0.926
	Left	−0.024	0.591
VTI	Right	0.031	0.489
	Left	−0.021	0.638
CO	Right	−0.047	0.291
	Left	−0.038	0.397
AT	Right	−0.135	0.002
	Left	−0.169	0.0001
ET	Right	−0.144	0.001
	Left	−0.119	0.007

AT acceleration time; CO cardiac output; ET ejection time; PSV peak systolic velocity; PWD pulsed-wave Doppler; VTI velocity time integral

variations in FHR (within the normal range) alone do not result in any significant change in PSV or CO [36], and only in cases with heart rates far from the normal physiologic range do the volume blood flow diminish due to the effects of the Frank–Starling mechanism [37].

The present study has several limitations. Firstly, we only provide information limited to the second and third trimester. Although the range of 18th week of pregnancy to term covers most potential applications of PWD echocardiographic examination, it would also be necessary to ascertain reference values at lower GAs so that early detection of abnormal hemodynamics could be possible. Another limitation involved the fact that the time intervals were affected by FHR. Nevertheless, these measurements have been shown to be reproducible. Additionally, the present study only demonstrated a normal pattern of PWD indices without testing their accuracy in identifying abnormal cases or predicting neonatal outcomes. Therefore, further studies should be conducted to explore their clinical usefulness.

In summary, we propose Z-score reference ranges for PWD indices against GA and EFW at the level of aortic and pulmonary valve in normal fetuses. These Z-scores allow exact quantification of how much a measurement deviate from normal and may help researchers to more accurately identify fetuses with abnormal blood flow, particularly in cases of cardiac outflow tract malformation and IUGR. Prospective serial studies are warranted to validate the effectiveness of these parameters.

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Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

Ethical approval All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional committee and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards.

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